Identification of neurodevelopmental trajectories in infancy and of risk factors affecting deviant development: a longitudinal birth cohort study

（幼児期の言語・運動・認知発達軌跡と、発達の遅れに影響する危険因子の特定：
縦断的出生コホート研究）

大阪大学大学院
大阪大学・金沢大学・浜松医科大学・千葉大学・福井大学
連合小児発達学研究科
小児発達学専攻

西　村　倫　子

２０１６年３月　博士学位論文

Identification of neurodevelopmental trajectories in infancy and of risk factors affecting deviant development: a longitudinal birth cohort study

Tomoko Nishimura[1], Nori Takei[1,2,3,*], Kenji J. Tsuchiya[1,2], Ryosuke Asano[1,2], Norio Mori[1,2,4], for HBC Study Team

Author Affiliations:
[1]United Graduate School of Child Development, Hamamatsu University School of Medicine, Japan
[2]Research Centre for Child Mental Development, Hamamatsu University School of Medicine, Japan
[3]Department of Psychosis studies, Institute of Psychiatry, King's College of London, United Kingdom
[4]Department of Psychiatry and Neurology, Hamamatsu University School of Medicine, Japan
*****Corresponding author: Nori Takei, PhD, MD, MSc

Abstract

Background:

We investigate patterns of neurodevelopmental trajectories in infants using a representative population, and identify risk factors that predict delayed growth.

Methods:

Participating infants (n = 952; 82.8% of the total sample) were assessed by Mullen Scales of Early Learning at 7 time-points from 1 month to 24 months of age. Mothers were recruited in early pregnancy and data on demographic characteristics were collected during pregnancy. Trajectory patterns were investigated using latent class growth analysis and risk factors for the derived trajectory classes were investigated by multinomial logistic regression.

Results:

Participants were found to be a fairly representative sample with respect to their demographic characteristics. Five classes of high normal (11.5%), normal (49.2%), low normal (21.2%), delayed (14.1%), and markedly delayed (4.0%) were identified. The markedly delayed class was characterised by overall delay from the early developmental stages; notably, such delay first became salient in motor domains and was then exceeded by language domains, especially receptive language. This class was predicted by male sex (odds ratio 4.0; 95% confidence interval 1.7-9.1), small for gestational age (2.8; 1.0-7.5), low placenta-to-birthweight ratio (2.8; 1.2-6.4), and low maternal education (4.7; 1.2-19.0). The delayed class was characterised by gradual downward deviation after first birthday, and was predicted by male sex (2.5; 1.5-4.2), preterm birth (4.4; 1.6-12.6), and advanced paternal age (1.9; 1.0-3.5).

Conclusions:

Our results demonstrate that about 1 out of 5 infants exhibits delayed neurodevelopment. Infants with distinct patterns of delayed trajectories and varying risk factors are considered to have different pathophysiological mechanisms.

Keywords

neurodevelopment; trajectory; delayed growth; birth cohort; risk factor, small placenta

Introduction

Many studies have investigated the early emergence and features of neurodevelopmental problems, and have found that many of early signs were evident in the first and second years of life in individuals with neuropsychiatric conditions such as autism spectrum disorder (ASD).[1,2] It is, however, critically important to comprehend neurodevelopmental trajectories, defined as longitudinal developmental patterns captured by behavioural features such as motor and language, in the general population as a whole. In regard to overall cognitive ability, Brian et al.[3] identified three patterns of neurodevelopmental trajectories as measured by the Mullen Scales of Early Learning (MSEL)[4] in the population of siblings of ASD and their controls. However, MSEL consists of five neurodevelopmental sub-domains, and it has been reported that the pattern of development is different among sub-domains and types of neurodevelopmental disorders.[5] In siblings of children with ASD, Landa *et al*.[6] examined neurodevelopmental trajectories longitudinally across multiple developmental domains as measured by MSEL. They depicted four different types of trajectories consisting of two normal and two delayed trajectories. These trajectories derived from the population of siblings of children with ASD may be affected by the hereditary attributes of ASD, and thus the results obtained from this population cannot be generalised.

Our aims in this study were twofold. First, we sought to investigate patterns in the neurodevelopmental trajectories in a representative population of infants in Japan. We anticipated that the percentage of infants with delayed growth in our study would be smaller than that reported in the previous study of the sample of siblings of children with ASD.[6] Second, we attempted to identify factors that predict and characterise infants in each of the assigned classes derived from the latent class growth analysis, especially infants allocated to the delayed class (es). The prenatal[7,8,9] and postnatal factors[10] have been associated with neurodevelopmental disorders. The neurodevelopment of infants with these disorders has been reported to differ from that of infants with typical development.[11] We postulated that such risk factors would be identified as risk factors in the delayed class (es) in the general population in the present study. These factors would also have important implications for predicting neurodevelopmental trajectories.

Methods

This study was conducted as part of an ongoing cohort study, the Hamamatsu Birth Cohort Study for Mothers and Children (HBC Study), which is described elsewhere.[12,13]

Participants

Participants included a consecutive series of the mothers (n = 1065) and their infants (n = 1152) born between 24 December 2007 and 30 June 2011. All women who visited in the first or second trimester of pregnancy at either of our two research sites, the Hamamatsu University Hospital and

Kato Maternity Clinic, were invited to participate in the study. In Japan, pregnant women can freely choose any maternity clinic, from a private clinic to a large general hospital. There was no between-site difference in demographic characteristics of the participants included in the analyses; the only one exception to this was age of mothers. Mothers who visited Kato Maternity Clinic first were younger than those who visited the Hamamatsu University Hospital first ($t = -2.93$, $p = 0.003$). All of the mothers who agreed to participate in the study, including mothers from Kato Maternity Clinic, gave birth at the same facility, i.e., Hamamatsu University Hospital. The assessment after birth was also performed at the same facility (Hamamatsu University Hospital). By referring to the reports from the Department of Health, Labour and Welfare, Japan,[14] we found that the enrolled mothers in this study were representative of Japanese mothers with respect to age, socioeconomic status and parity, and their offspring were representative of Japanese offspring with respect to birthweight and gestational age at birth. Therefore, participants in this cohort are considered to be a fairly representative sample of the general population.[12,13]

We excluded 183 participating mothers and 198 infants who missed 5 or more of the total 7 follow-up evaluations after birth (Fig. 1). The major reason for missing was a Japanese traditional support system for childbirth, called "*satogaeri bunben*".[15] We also excluded two mother-infant dyads as the infants were diagnosed with Down syndrome. Thus, 952 infants (82.6%) and 880 mothers (82.6%) were included in the analyses. Table 1 displays demographic characteristics of the sample included in the analysis.

Measures

Developmental assessment

We used the Mullen Scales of Early Learning (MSEL) to evaluate neurodevelopmental progress. MSEL is a composite scale for assessing child development and is made up of five subscales: gross motor, visual reception, fine motor, receptive language, and expressive language. Measurement was performed when the infants reached the ages of 1, 4, 6, 10, 14, 18 and 24 months. Prior to follow-up assessments of the birth cohort, two experienced clinicians performed 3-month video training sessions, through which agreement of their scoring of each item on the MSEL scale was attained. Subsequently, separate 3-month video training sessions were set up, including additional 5 assessors (child health professionals), who engaged in actual ascertainment. Because the assessment criteria change according to the development (i.e., ageing), similar training and quality-maintaining sessions using video recorded assessments were repeated prior to each of the 7 time-point follow-ups. Developmental assessments with MSEL were conducted without referring to previously evaluated data. The assessors for the development were masked to information about demographic variables (Background assessment), which was collected by independent and separate researchers.

MSEL T-scores, which are equivalent to Z-scores but instead with a mean of 50 and a standard

deviation (SD) of 10, are commonly used and a useful index which enables one to discern deviations from normative development. As anticipated, however, the US version of the normative data was found not to be correspondent, particularly in the language domains, with the Japanese sample in this study. We therefore developed the Japanese version of the T-scores, using our HBC sample (Supplement 2), in accordance with the original procedure described by Mullen.[4]

Background assessment

Data on the demographic characteristics of mothers were collected during pregnancy. They included the age of the mother and partner, their educational level and their annual household income. Perinatal variables were obtained from medical records, including gestational age, birthweight and placental weight.

Statistical Analysis

Latent class growth analysis

Latent class growth analysis enables us to distinguish distinct subgroups of individuals, called "a latent class", following distinct patterns of change over time.[16,17] To investigate neurodevelopmental trajectories, we applied a parallel process latent class growth analysis, which allowed simultaneous processing of 5-domain data and contained combinations of continuous latent growth variables including the intercept (I), slope (S), the quadratic (Q) term for each of the five sub-domains of outcome and a latent categorical variable (Fig. 2). Individual membership was assigned on the basis of the most likely posterior probabilities (maximum-probability assignment rule).[17] We set the Japanese version of T-scores as the dependent variables. Because T-scores were found to follow *nearly* normal distributions at each time point, but slightly skewed, we used an MLR estimator.[18] As missingness related to attrition was at least not associated with outcome measures, as evaluated by MSEL (Supplement 1), the full information maximum likelihood algorithm we employed under assumption of missing at random (MAR) was deemed to be sustained.

Since the appropriate number of latent classes was initially unknown, the model was run from one class solution to increasing number of classes solution. To compare models with the different numbers of classes and determine the optimum model, we employed several fit indices[19]: the smallest Bayesian information criterion (BIC), consistent Akaike's information criterion (CAIC) and approximate weight of evidence criterion (AWE); the adjusted Lo-Mendell-Rubin likelihood ratio test (adjusted LMR-LRT)[20] and the bootstrap likelihood ratio test (BLRT)[21]; and entropy. In addition, the number of classes was ultimately determined by integrating other considerations, including theoretical justification and interpretability.[22]

Multinomial logistic regression

To explore relationships between the latent class assignment and predictor variables, we used a

three-step approach developed by Vermunt.[23] In this approach, the latent class model was estimated in a first step using only latent class indicator variables. In the second step, assignment of the most likely class was made for each child using the latent class posterior distribution obtained during the first step. In the third step, a factor of the most likely class was regressed on predictor variables, taking into account the misclassification in the second step.[24] As potential risk factors, gender, preterm birth before 37 weeks, small for gestational age (SGA), low placenta-to-birthweight ratio, parental age, parental educational level and parental income were investigated. SGA infants were defined as having a birthweight less than the 10th percentile for the gestational age.[25] The placenta-to-birthweight ratio was calculated by dividing placental weight by birthweight, and a low ratio less than 10th percentile indicated a small placenta relative to offspring birthweight.[26] Nineteen mother-infant dyads who had missing values in mother's placental weight were excluded from the analysis. In addition, since the 10th percentile cannot be simply applied to twin deliveries, we excluded twin deliveries (n = 28). Because some offspring were born from the same mother, such clustering (i.e., family clustering) was allowed for in the analyses using a Huber sandwich estimator. All statistical analysis was performed with Mplus version 7.11.

Ethical Issues

The study protocol was approved by the Hamamatsu University School of Medicine and University Hospital Ethics Committee. Written informed consent was obtained from each mother for her own and her infant participation.

Results

Latent class growth analysis

We ran the model from the simplest solution to more complex solutions. The value of BIC and CAIC continued to decrease, but the AWE had the smallest value for the 5-class solution (see Supplement 3). The p-values of the adjusted LMR-LRT were less than 0.05 up to the 2-class solution, whereas the p-values of BLRT was less than 0.001 up to the 7-class solution. Entropy was high enough for all the solutions, with 2- and 5-class solutions closer to 1.0 indicating better classification quality. We counted chiefly on the results of these fit indices, but the prominent and exclusive solution did not emerge; two solutions (2-class and 5-class) were judged to be equally optimal model fit on the grounds of the results of AWE, adjusted LMR-LRT, and entropy. Among these optimal two solutions, we ultimately selected the 5-class solution on the basis of our a priori postulation that the number of growth trajectory classes of infants in our study of a representative sample of the general population would be equal to or larger than the 4-class solution reported in a sample of siblings of children with ASD who are considered to be more homogenous than the general population and share hereditary attributes of ASD.[6]

Figure 3 shows developmental trajectories by the MSEL domain and class assignment. The first

class was taken as “high normal” with a proportion of 11.5% of the sample (n = 110). This class was characterised by the relatively accelerated development in all five domains. The second class was “normal”, and approximately half of the total sample was allocated to this class (n = 468; 49.2%). The progress of growth in this class was nearly linear and parallel along the mean value of 50 over the period and thus closely matched the norm trajectory. The third class was “low normal”; these infants showed slight delay in the early developmental stages, but caught up by 24 months (n = 202; 21.2%). These three classes were considered normal because the trajectories were in the range of the mean ± 1 SD. The fourth “delayed” class showed a downward deviation in T-scores over time that became marked after around 12 months of age (n = 134; 14.1%). This downward pattern (i.e., slower rates of developmental gain) was distinct, particularly in receptive language. The motor function of infants in this class gradually diverged from that of their peers in the normative group over 24 months, although the estimated mean remained above -1 SD. The fifth class was designated as “markedly delayed”, and was characterised by an overall delay from the early developmental stages (n = 38; 4.0%). The estimated mean in motor domains deviated from the norm steeply within 10 months of age after birth, whereas the deviation became conspicuous in the language domains after 12 months. Furthermore, the estimated means for the language domains dropped down below -2 SD at around 20 months of age. In addition, an overview reveals that the degree of divergence across 5 classes appears to become larger over time, and it was most pronounced in the receptive language domain at 24 months of age.

Multinomial logistic regression

In Table 2, the reference was the “normal” class. The distributions of all variables in this class were coincident with the norm figures in Japan, and in fact did not deviate from the grand value for the total sample.

Male infants were more likely to be assigned to the delayed (odds ratio, OR: 2.5; 95% confidence interval, CI: 1.5-4.2; $p < 0.001$) and markedly delayed classes (OR: 3.8; 95%CI: 1.4-10.4; $p = 0.01$) than to the normal class. Infants born before 37 weeks were more likely to be assigned to the delayed class (OR: 4.4; 95%CI: 1.6-12.6; $p = 0.005$) than to the normal class. SGA infants were more likely to be allocated to the markedly delayed class (OR: 2.8; 95%CI: 1.0-7.5; $p = 0.04$) than to the normal class. When the placental weight was small relative to offspring birthweight, the probability of assignment to the markedly delayed class was elevated (OR: 2.8; 95% CI: 1.2-6.4; $p = 0.02$) compared with the normal class. With every 10-year increase in paternal age, the probability of assignment to the delayed class increased (OR: 1.9; 95% CI: 1.0-3.5; $p = 0.04$) compared with the normal class. When the educational history of the mothers was less than 12 years, the probability of assignment to the markedly delayed class was elevated (OR: 4.7; 95% CI: 1.2-19.0; $p = 0.03$) compared with the normal class. When the clustering within the recruited site was further allowed for in the analysis, the results remained almost identical (data not shown).

Attrition

We compared neurodevelopmental growth patterns, as assessed by MSEL, between a group of infants excluded from analysis and the infants included in the analysis. To examine whether there is any association between missingness and growth patterns of neurodevelopment, we examined the interaction of group (excluded vs. included) x slope using a linear mixed model. There were no differences in the MSEL T-scores between the infants excluded and included in the analysis, although some of background characteristics differed between them; in addition, loss to follow-up was kept minimal (see Supplement 1 and 5). In multinomial logistic regression, attenuation due to missing data was almost negligible (4.9%). In effect, the results remained identical when we applied multiple imputation.

Discussion

To our knowledge, this is the first longitudinal study to comprehensively examine the neurodevelopment of infants in the general population and to investigate risk factors that are associated with deviated growth trajectories in early life. The existence of aberrant trajectories may be known empirically, but we directly identified them from a representative sample of infants. In addition, the measurements were prospective and fairly comprehensive, with each infant being directly evaluated at multiple time points (i.e., 7 times) and the *same* developmental scale was employed throughout.

We discovered five neurodevelopmental classes consisting of three normal (81.9%) and two delayed (18.1%) trajectories. Law *et al.*[27] reported that the prevalence of speech and language delay is 5.0% and the prevalence of language delay is 16.0% at 2 years of age. The prevalence of any developmental disability in children aged 3 to 17 years in the United States was reported 15.0%.[28] It is not possible to compare these reports with our results because the examined developmental domains and the targeted ages were different, but the proportion of the identified delayed classes was similar, albeit slightly higher, to the prevalence reported in these earlier studies.

In the markedly delayed class, the deviation became marked in motor domains earlier, and somewhat later in language domains. Interestingly, in their population-based cohort study in the Netherlands, van Batenburg-Eddes *et al*.[29] reported that minor deviation from normal neuromotor development at 9-15 weeks of age is associated with receptive and expressive language delay at 1.5 and 2.5 years of age. The deviation of motor function may precede in early developmental stages and serve as a useful indicator for the subsequent emergence of widespread developmental delays, including delayed language.

The “delayed” class, another type of infants with delayed development, in this study showed trajectories with the same convex curve and downward turn with ageing in all five domains.

Disturbed developments as demonstrated in this delayed class as well as the markedly delayed class are reminiscent of developmental disorders such as ASD. The diverse patterns of developmental trajectories observed in this study may pertain to variability of manifestation of developmental disorders such as ASD: e.g., distinctive patterns of onset.[30] However, comparatively poor directions of development, especially in receptive language, highlight the need for further study of the subsequent developments in this population, in relation to identification of emerging patterns of developmental disorders.

We investigated the risk factors that determine class assignment. The finding of delayed neurodevelopment in males in early life is compatible with the notion that males are more vulnerable at early developmental stages.[31] Recent studies have shown that the sexually dimorphic brain is formed by gender-varying mechanisms such as the hormone,[32] gene expression[33] and immune systems,[34] which in turn affect the development of the central nervous system (CNS) processes such as myelination, migration and synaptogenesis.[35] The fact that developmental disorders such as ASD occur predominantly in males[36] suggests that the infants in delayed classes identified in this study may share common mechanisms with children with neurodevelopmental disorders.

We also found that SGA infants were more likely to be assigned to the markedly delayed class, and premature infants were more likely to be assigned to the delayed class. It has been reported that intrauterine growth restriction (IUGR), which is closely related to SGA,[37,38] causes CNS structural and functional abnormalities,[39] and is associated with sensory, motor and intellectual impairments.[40] The brain is especially susceptible to the sequelae of preterm birth, resulting in high rates of long-term neurological and health problems.[41] However, two delayed classes identified in this study were predicted by different risk factors. This result suggests that these classes may have different pathophysiological mechanisms in which deviated growth patterns are determined.

Of particular interest is a finding of the relationship between small placenta relative to the birthweight and marked neurodevelopmental delay in this study. It has been reported that placental weight was lower in SGA infants than in appropriate-for-gestational-age infants of the same birthweight.[42] Our finding of an increased risk of the developmental delay associated with small placenta, independent of SGA, suggests that small placenta *per se* incurs neurodevelopment. Although the association between lower placental weight and schizotypal traits in adult women,[43] and placental size and mental health problems such as inattention-hyperactivity in boys[44] has been demonstrated, this study is the first to our knowledge to demonstrate that small placenta may play an important role in predisposition to delayed neurodevelopment. In parallel to emergence of studies showing the association between small placenta and neuropsychiatric disorders, research interest has been increasingly growing in understanding the mechanisms underlying the possible

relationship between placental insufficiency in fetal life and abnormal brain development.[45,46] Our current finding further encourages this direction.

Advanced paternal age, which has been related to developmental disabilities,[47,48] was identified as a risk factor of the delayed class. Kong *et al*.[49] reported that a one-year increase of paternal age leads to approximately 2 *de novo* mutations of single-nucleotide polymorphisms. A portion of infants with the delayed growth identified in this study could be interpreted as having a precursor of neurodevelopmental disorders linked with *de novo* mutations.

A poor history of maternal education was identified as a risk factor only in the markedly delayed class. However, inspection of the results in Table 3 reveals an increasing gradient in odds ratio (OR) from the high normal (0.8) to markedly delayed classes (4.7). In fact, there was a linear trend for ORs across five classes ($\chi^2 = 8.8$, df = 1, $p < 0.005$). This indicates that poor history of maternal education exerts a “one-class lowering effect" in terms of class assignment. In other words, lower maternal education may generally affect the performance of offspring and lead to a lower level of growth trajectory.

In the present analysis, there was a clear separation and divergence of growth patterns, even at an early stage, among different trajectory classes over the follow-up period. Given this fact, one may ask whether outcome groups created using only the data on developmental assessments obtained at the end of the observation period (i.e., 24 months of age) would show risk factor characteristics identical to those found in the present study. To answer this question, we examined the relationships between antenatal risk factors and outcome groups as defined using the sum of scores of the MSEL 5 domains at 24 months of age (the group proportions were the same as in the classes identified in this study). The results for male gender, paternal age, low placental-to-birthweight ratio, and maternal education were similar to those identified in the trajectory analysis (Table S4 and Table 2). While advanced maternal age was not identified as a predictor in the trajectory analysis, its seemingly protective effect emerged in the fourth cross-sectional outcome group. This result was unexpected, since it has been reported that advanced maternal age has a risk-increasing effect,[9] suggesting that this estimate based on the cross-sectional outcome assignments may be unreliable. Further, low maternal education was a predictor for poor growth in the offspring of the fifth class (OR 4.7; 95% CI 1.2-19.0; p = 0.03), but the detrimental effect became more prominent (a 1.5-fold increase in OR) when the cross-sectional measurements at 24 months of age alone were used for determining outcomes: an OR of 7.0 (95% CI: 1.9-26.4; p = 0.004) was estimated for the fifth group. Although low income was not identified as a predictor for any classes in the trajectory analysis, it was found to be a risk factor for the third and fourth cross-sectional outcome groups. In addition, there was a discrepancy in two other predictors; that is, preterm birth and small-for-gestational-age, both of which were found to serve as predictors of delayed growth in the latent class trajectory analysis, were not identified as risk factors in the cross-

sectional outcome groups. Low income as well as poor maternal education can be regarded as social and environmental factors, whereas the latter two factors (preterm birth and small-for-gestational-age) allude to biological involvement with disturbed growth that may exist immediately after birth or even before it. It is reasonable to assume, therefore, that the cross-sectional outcome measurements at 24 months of life may represent environmental influences on growth that accumulate towards the end of the follow-up period, and that such environmental effects may, to some extent, mask impaired growth that originates in early life. Latent class growth analysis is a powerful modality well-suited to our sample. For example, although there was no linear trend for ORs associated with poor maternal education across the five cross-sectional outcome groups, a linear trend was evident in the trajectory analysis, which facilitates a more plausible interpretation. Taken together, the these results demonstrate that latent class growth analysis is a useful tool to provide optimum information by capturing unique traces extending throughout the observation period.

Our findings showed five distinct types of neurodevelopmental trajectories in the first two years of life in the representative sample of the general population, and an apparent downward deviation in growth was seen in two of them. Although the probabilities for assignment of the class membership were not perfect, it was satisfactorily high (0.82 to 0.93). Furthermore, these two classes were predicted by different sets of risk factors. The results suggest that these classes may have different pathophysiological mechanisms in which deviated growth patterns are determined. The strength of this study is that our sample comprised a representative sample of infants and thus the findings are generalisable. This study has extensively examined the neurodevelopment of infants using the *same* developmental scale throughout the follow-up period. Another strength is the low attrition rate; 91% of the initially enrolled mother-infant dyads, after eliminating "*satogaeri bunben*", was retained in the analysis. As one of the limitations of this study, it is uncertain whether infants with missing data were allocated to each of 5 classes with equal probabilities, although attrition was minimal. As some background characteristics differed between infants between with and without missing data, there is a possibility that infants with deviant growth may have been excluded from the analysis. Another limitation is that the trajectory patterns we examined were limited to the first two years of life, and it is possible that these patterns may change after further follow-ups. In addition, there is little measurable variation and much difficulty in performing overt assessments in the very early months of life. T-scores we employed tend not to be secured until infants can pass certain developmental milestones. Hence, data obtained in the very early stages may not contribute greatly to the latent class assignments. It will be necessary to follow-up with a study of subsequent trajectories, including outcome measures such as certain diagnostic entities, and to confirm the relationships between trajectory patterns and predictors found in this study.

KEY MESSAGES

- The best model in the latent class growth analysis identified five distinct neurodevelopmental trajectories.
- The 18.1% of infants in either the delayed or markedly delayed classes made measurable neurodevelopmental gains more slowly than their peers in the normative classes.
- The markedly delayed class characterised by overall delay from the early developmental stages was predicted by male sex, small for gestational age, low placenta-to-birthweight ratio, and low maternal education, and the delayed class characterised by a gradual downward deviation after first birthday was predicted by male sex, preterm birth, and advanced paternal age.
- Small placenta relative to birthweight may play an important role in predisposition to delayed neurodevelopment.

Funding
This work was supported by the Strategic Research Programme for Brain Sciences ("Integrated research on neuropsychiatric disorders") (NM and KJT) and Grant-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science & Technology in Japan (grant numbers 26670540, 24659542: NT, 26380878: NM, and 25461758: KJT).

Acknowledgements
The authors would like to thank Dr. Tetsuo Kato of the Kato Maternity Clinic for promoting recruitment of potential participants. The authors are also grateful to Drs. N. Kanayama, H. Itoh, K. Sugihara, M. Sugimura, K. Takeuchi, K. Suzuki, Y. Murakami, Y. Koumura, Y. Miyabe, K. Hirai, Y. Nakamura, R. Koizumi, H. Murakami, Y. Kobayashi-Koumura, and K. Muramatsu-Kato, and all the attending obstetricians. The authors also thank Ms. Kiyomi Hinoki, and all the midwives and staff at the maternity clinic of the Hamamatsu University School of Medicine, for enrolling participants.
The HBC study team includes Ms. Y. Kugizaki, C. Nakayasu, A. Okumura, Y. Suzuki, N. Kodera, E. Higashimoto, A. Nakamura, R. Takabayashi, T. Mori, H. Muraki, M. Narumiya, M. Honda, Y. Seno, E. Sato, C. Nishizawa, Mr. R. Nakahara, Drs. T. Harada, Y. Kameno, T. Wakuda, D. Kurita, K. Takebayashi, Y. Iwata, S. Takagai, T. Sugiyama, M. Tsujii, A.A. Pillai, T. Ismail, K. Matsumoto, K. Iwata, C. Shimmura, Y. Yoshihara, S. Yamamoto, M. Kawai, K. Nakamura, H. Matsuzaki, G. Sugihara, K. Hirano, Y. Endoh, and T. Suzuki. Finally, we thank all the participating families and infants.

Conflict of interest: None declared.

References

1. Bolton PF, Golding J, Emond A, Steer CD. Autism spectrum disorder and autistic traits in the Avon Longitudinal Study of Parents and Children: precursors and early signs. *J Am Acad Child Adolesc Psychiatry* 2012;**51**:249-260.
2. Lemcke S, Juul S, Parner ET, Lauritsen MB, Thorsen P. Early signs of autism in toddlers: a follow-up study in the Danish National Birth Cohort. *J Autism Dev Disord* 2013;**43**:2366-2375.
3. Brian AJ, Roncadin C, Duku E et al. Emerging cognitive profiles in high-risk infants with and without autism spectrum disorder. *Res Autism Spectr Disord* 2014;**8**:1557–1566.
4. Mullen EM. Mullen Scales of Early Learning: AGS Edition. Minneapolis, MN: Pearson Assessments. 1995
5. Barbaro J, Dissanayake C. Developmental Profiles of Infants and Toddlers with Autism Spectrum Disorders Identified Prospectively in a Community-Based Setting. *J Autism Dev Disord* 2012;**42**:1939–1948.
6. Landa RJ, Gross AL, Stuart EA, Bauman M. Latent class analysis of early developmental trajectory in baby siblings of children with autism. *J Child Psychol Psychiatry* 2012;**53**:986–996.
7. Savchev S, Sanz-Cortes M, Cruz-Martinez R et al. Neurodevelopmental outcome of full-term small-for-gestational-age infants with normal placental function. *Ultrasound Obstet Gynecol* 2013;**42**:201-206.
8. Sutton PS, Darmstadt GL. Preterm birth and neurodevelopment: a review of outcomes and recommendations for early identification and cost-effective interventions. *J Trop Pediatr* 2013;**59**:258–265.
9. Idring S, Magnusson C, Lundberg M et al. Parental age and the risk of autism spectrum disorders: findings from a Swedish population-based cohort. *Int. J Epidemiol* 2014;**43**:107-115.
10. Hanscombe KB, Trzaskowski M, Haworth CMA, Davis OSP, Dale PS, Plomin R. Socioeconomic status (SES) and children's intelligence (IQ): in a UK-representative sample SES moderates the environmental, not genetic, effect on IQ. *PLoS One* 2012;**7**:e30320.
11. Burns TG, King TZ, Spencer KS. Mullen Scales of Early Learning: The utility in assessing children diagnosed with autism spectrum disorders, cerebral palsy, and epilepsy. *Appl Neuropsychol Child* 2013;2:33-42.
12. Tsuchiya KJ, Matsumoto K, Suda S et al. Searching for very early precursors of autism spectrum disorders: the Hamamatsu Birth Cohort for Mothers and Children (HBC). *J Dev Orig Health Dis* 2010;**1**:158–173.
13. Takagai S, Tsuchiya KJ, Itoh H et al. Cohort profile: Hamamatsu Birth Cohort for Mothers and Children (HBC Study). *Int. J Epidemiol* 2015; doi: 10.1093/ije/dyv290
14. MHLW. *Ministry of Health, Labour and Welfare*. Vital , Health and Social Statistics Division, Statistics and Information Department, Minister's Secretariat, Ministry of Health , Labour and Welfare; 2013, Retrieved 14 February 2015 from http://www.mhlw.go.jp/english/database/db-hw/dl/81-1a2en.pdf
15. Yoshida K, Yamashita H, Ueda M, Tashiro N. Postnatal depression in Japanese mothers and the

reconsideration of ‘Satogaeri bunben’. *Pediatr Int* 2001;**43**:189–193.
16. Muthén B, Muthén L. Integrating person-centered and variable-centered analysis: Growth mixture modeling with latent trajectory classes. *Alcohol Clin Exp Res* 2000;**24**:882–891.
17. Andruff H, Carraro N, Thompson A, Gaudreau P, Louvet B. Latent class growth modelling: a tutorial. *Tutor Quant Methods Psychol* 2009;**5**:11–24.
18. Asparouhov T, Muthén B. Multivariate statistical modeling with survey data. Proceedings of the Federal Committee on Statistical Methodology (FCSM) Research Conference. 2005
19. Masyn KE. Latent Class Analysis and Finite Mixture Modeling. In: Nathan P, Little T, editors. The Oxford Handbook of Quantitative Methods, Vol. 2: Statistical Analysis. New York, NY: Oxford University Press; 2013. p. 551–610.
20. Lo Y, Mendell NR, Rubin DB. Testing the number of components in a normal mixture. *Biometrika* 2001;**88**:767–778.
21. Nylund KL, Asparouhov T, Muthén B. Deciding on the number of classes in latent class analysis and growth mixture modeling: A Monte Carlo simulation study. *Struct Equ Modeling* 2007;**14**:535–569.
22. Jung T, Wickrama KAS. An introduction to Latent Class Growth Analysis and Growth Mixture Modeling. *Soc Pers Psychol Compass* 2008;**2**:302–317.
23. Vermunt JK. Latent class modeling with covariates: Two improved three-step approaches. *Polit Anal*, 2010;**18**:450–469.
24. Asparouhov T, Muthén B. Auxiliary variables in mixture modeling: Three-step approaches using Mplus. *Struct Equ Modeling* 2014:**21**;329-341.
25. Ogawa Y, Iwamura T, Kuriya N et al. Birth size standards by gestational age for japanese neonates. *Acta Neonat Jpn* 1998;**34**:624-632.
26. Almog B, Shehata F, Aljabri S, Levin I, Shalom-Paz E, Shrim A. Placenta weight percentile curves for singleton and twins deliveries. *Placenta* 2011;**32**:58-62.
27. Law J, Boyle J, Harris F, Harkness A Nye C. Screening for Speech and Language Delay: A Systematic Review of the Literature. *Health Technol Assess* 1998;**2**:1–184.
28. Boyle CA, Boulet S, Schieve LA et al. Trends in the Prevalence of Developmental Disabilities in US Children, 1997-2008. *Pediatrics* 2011;**127**:2010–2989.
29. van Batenburg-Eddes T, Henrichs J, Schenk JJ et al. Early infant neuromotor assessment is associated with language and nonverbal cognitive function in toddlers: the Generation R Study. *J Dev Behav Pediatr* 2013;**34**:326–334.
30. Landa RJ, Gross AL, Stuart EA, Faherty A. Developmental trajectories in children with and without autism spectrum disorders: the first 3 years. *Child Dev* 2013;**84**:429–442.
31. Kraemer S. The fragile male. *BMJ* 2000;**321**:1609–1612.
32. Collaer ML, Hines M. Human behavioral sex differences: a role for gonadal hormones during early development? *Psychol Bull* 1995;**118**:55–107.
33. Kang HJ, Kawasawa YI, Cheng F et al. Spatio-temporal transcriptome of the human brain. *Nature* 2011;**478**;483–489.
34. Lenz KM, Nugent BM, Haliyur R, McCarthy MM. Microglia are essential to masculinization of brain

and behavior. *J Neurosci* 2013;**33**:2761–2772.

35. Bale TL, Baram TZ, Brown AS et al. Early life programming and neurodevelopmental disorders. *Bio Psychiatry* 2010;**68**:314–319.
36. Ruigrok AN, Salimi-Khorshidi G, Lai MC et al. A meta-analysis of sex differences in human brain structure. *Neurosci Biobehav Rev* 2014;**39**:34–50.
37. Walker SP, Wachs TD, Grantham-McGregor S et al. Inequality in early childhood: risk and protective factors for early child development. *Lancet* 2011;**378**:1325–1338.
38. Suda S, Takei N. Disturbed growth in early life and later neurocognitive development related especially to psychiatric disorders. In: Preedy VR, Watson RR, Martin CR, editors. Handbook of Behavior, Diet and Nutrition. London: Springer; 2011. p. 1541–1554.
39. Mallard C, Loeliger M, Copolov D, Rees S. Reduced number of neurons in the hippocampus and the cerebellum in the postnatal guinea-pig following intrauterine growth-restriction. *Neuroscience* 2000;**100**:327–333.
40. Mackay DF, Smith GC, Dobbie R, Cooper SA, Pell JP. Obstetric factors and different causes of special educational need: retrospective cohort study of 407,503 schoolchildren. *Int J Gynaecol Obstet* 2013;**120**:297–308.
41. Saigal S, Doyle LW. An overview of mortality and sequelae of preterm birth from infancy to adulthood. *Lancet* 2008;**371**:261–269.
42. Heinonen S, Taipale P, Saarikoski S. Weights of placentae from small-for-gestational age infants revisited. *Placenta* 2001;**22**:399–404.
43. Lahti J, Raïkkönen K, Sovio U et al. Early-life origins of schizotypal traits in adulthood. *Br J Psychiatry* 2009;**195**:132-137.
44. Khalife N, Glover V, Hartikainen AL et al. Placental size is associated with mental health in children and adolescents. *PLoS One* 2012;7:e40534.
45. Girardi G, Fraser J, Lennen R, Vontell R, Jansen M3, Hutchison G. Imaging of activated complement using ultrasmall superparamagnetic iron oxide particles (USPIO) – conjugated vectors: an in vivo in utero non-invasive method to predict placental insufficiency and abnormal fetal brain development. *Mol Psychiatry* 2014;110:1–10.
46. Zeltser LM, Leibel RL. Roles of the placenta in fetal brain development. *Proc Natl Acad Sci USA* 2011;**108**:15667–15668.
47. D'Onofrio BM, Rickert ME, Frans E et al. Paternal age at childbearing and offspring psychiatric and academic morbidity. *JAMA Psychiatry* 2014;**71**:432–438.
48. Idring S, Magnusson C, Lundberg M et al. Parental age and the risk of autism spectrum disorders—findings from a Swedish population-based cohort. *Int J Epidemiol* 2014;43:107–15.
49. Kong A, Frigge ML, Masson G et al. Rate of de novo mutations and the importance of father's age to disease risk. *Nature* 2012;**488**:471–475.

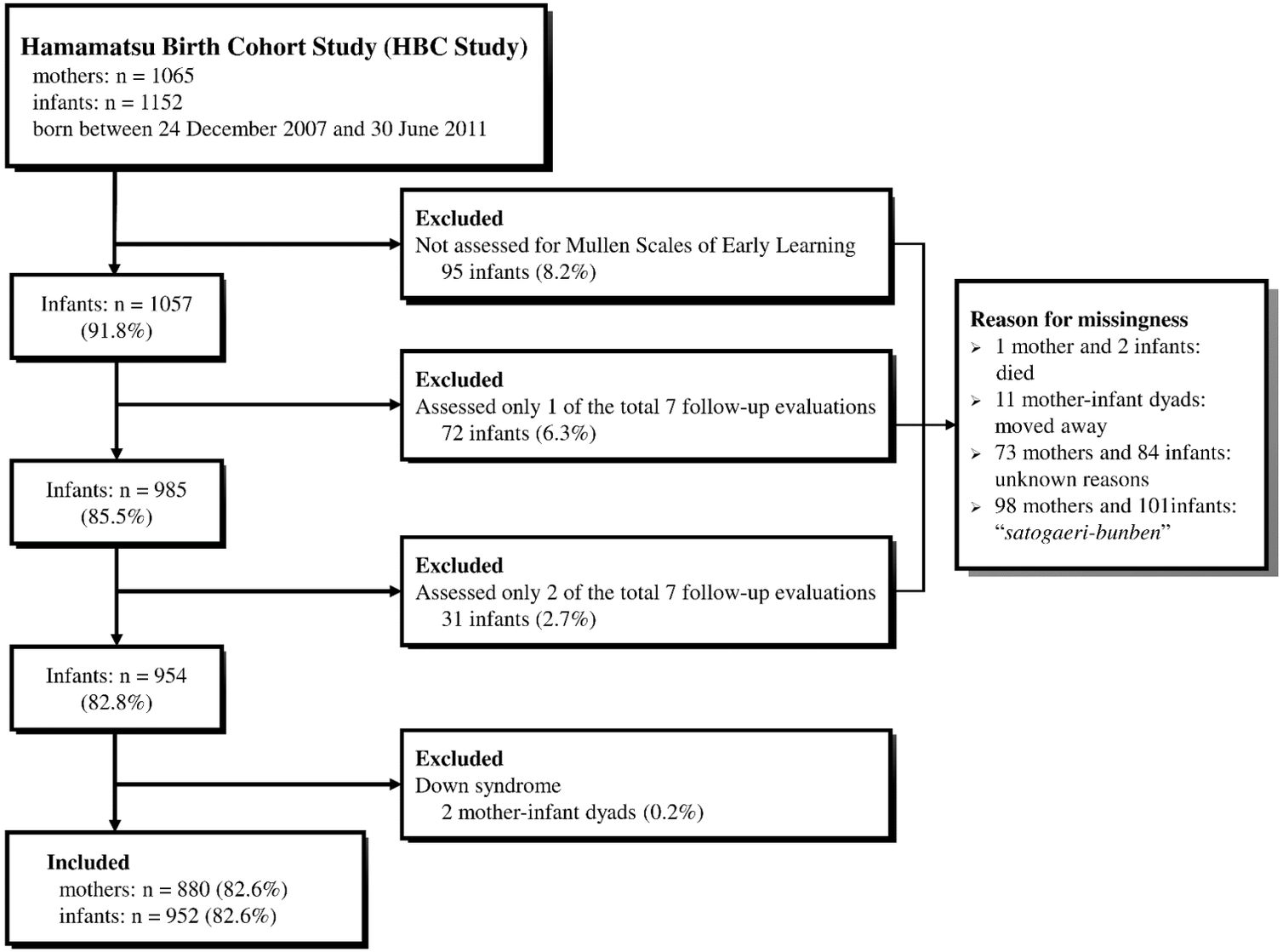

Figure 1. Flow chart of participants who met inclusion/exclusion criteria

Table 1. Characteristics of participating infants and their parents

	Mean (SD)
Birth Weight [g]	2951.0 (429.0)
Gestational Age at birth [weeks]	39.0 (1.5)
Placental weight [g]	561.6 (131.9)
Paternal Age at birth [yrs]	33.0 (6.0)
Maternal Age at birth [yrs]	31.2 (5.0)
Parental income at 2nd trimester of the index pregnancy [million JPY]	6.1 (3.2)
	n (%)
Gender	
Male	470 (49.4)
Female	482 (50.6)
Small for gestational age	
<10th percentile	853 (89.6)
10th - 100th percentile	99 (10.4)
Prematurity	
<37 weeks	56 (5.9)
37weeks and longer	896 (94.1)
Placental-to-birthweight ratio (twin excluded)	
<10th percentile	169 (18.7)
10th - 100th percentile	736 (81.3)
Paternal education	
<12 years	70 (7.4)
12 years and longer	882 (92.6)
Maternal education	
<12 years	41 (4.3)
12 years and longer	911 (95.7)

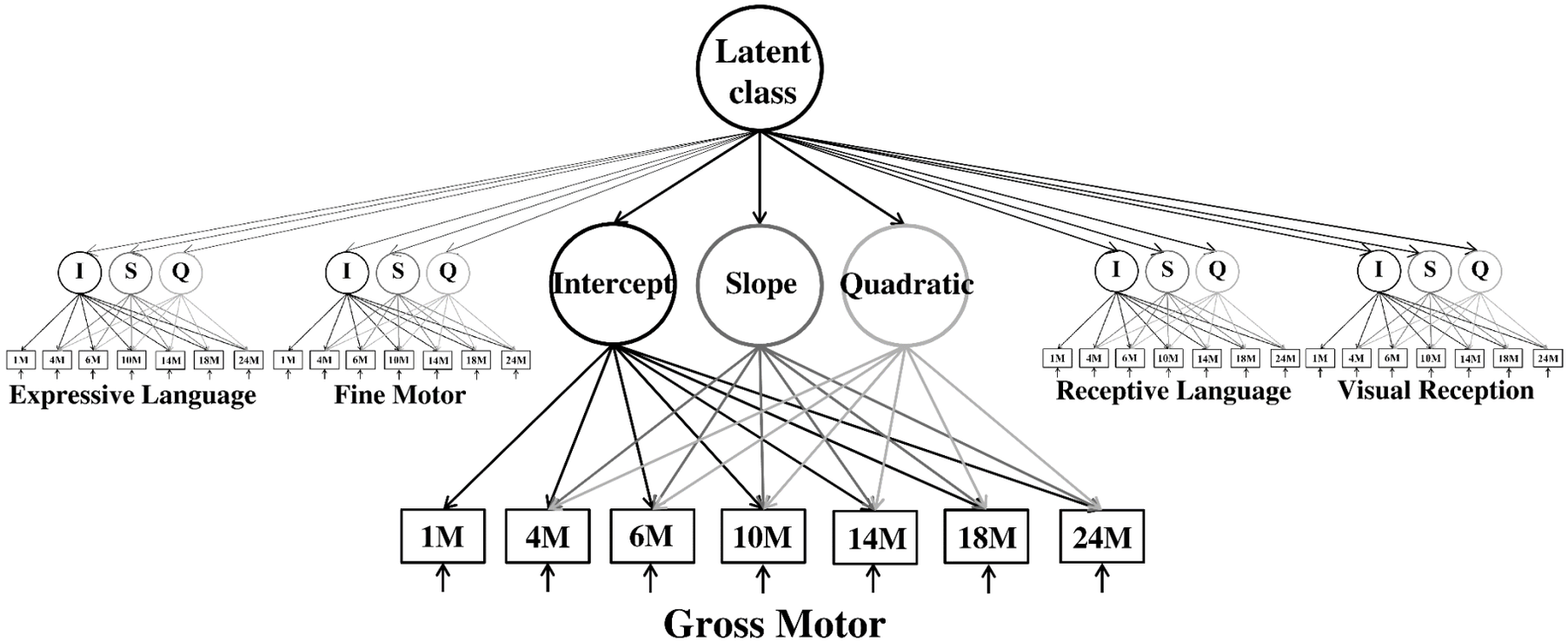

Figure 2. Parallel process latent class growth analysis model. Here we spread only the Gross Motor domain, but this model is specified for all 5 domains.

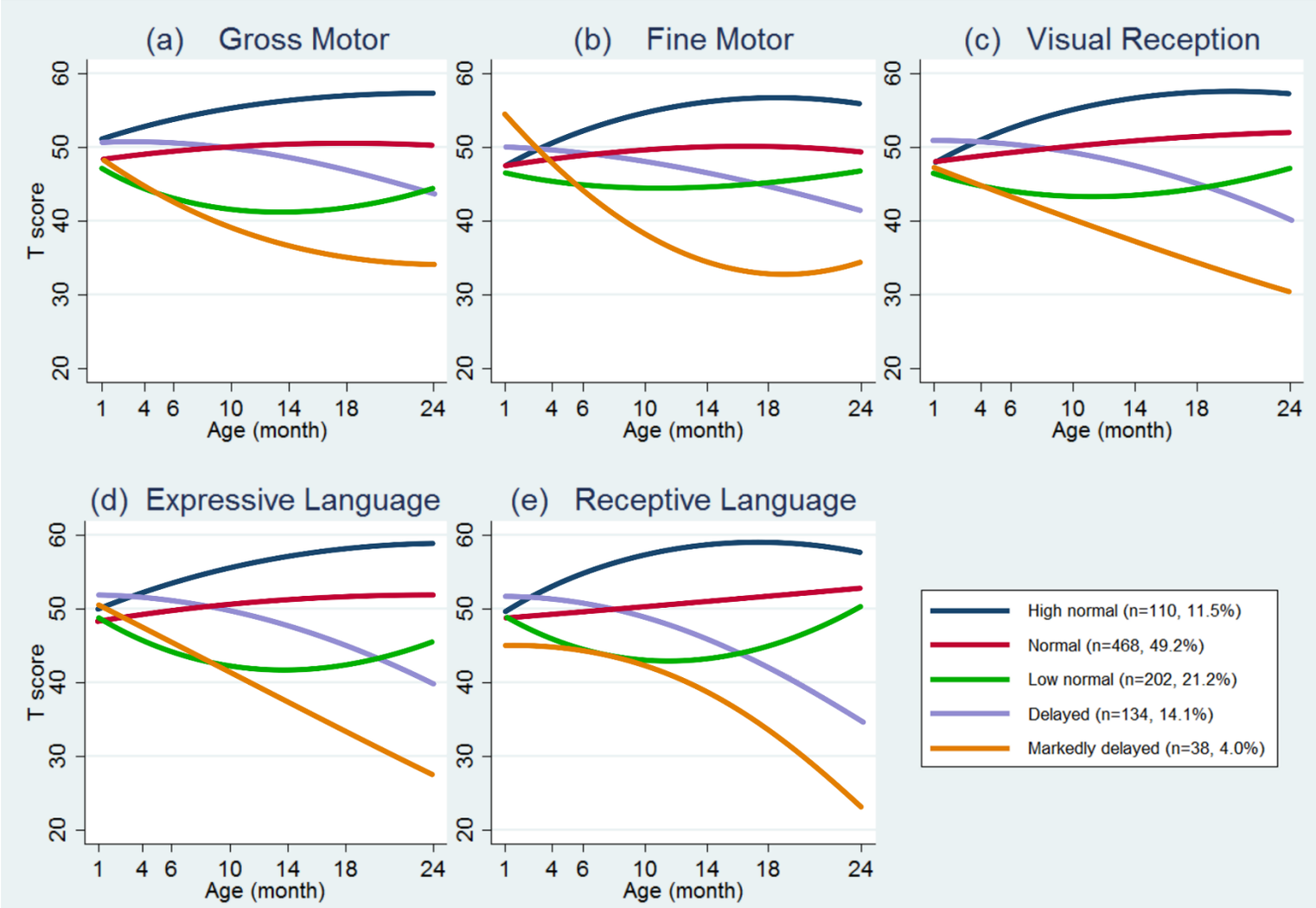

Figure 3. Five trajectory classes of MSEL T score from age 1 to 24 months.

The estimated neurodevelopmental trajectories of each class in (a) gross motor, (b) fine motor, (c) visual reception, (d) receptive language, and (e) expressive language domain. "T-scores = 50" indicates normative development (mean = 50, SD = 10), and a downward pattern reflects slower rates of developmental gain relative to peers in the normative group.

Table 2. Risk factors for each trajectory class: Odds ratio and 95% confidence intervals in the analysis using multinomial logistic regression.

Risk factors	Latent class				
	High Normal (n = 110, 11.5%)	Normal (n = 468, 49.2%)	Low Normal (n = 202, 21.2%)	Delayed (n = 134, 14.1%)	Markedly Delayed (n = 38, 4.0%)
	OR (95% CI)	(Base Outcome)	OR (95% CI)	OR (95% CI)	OR (95% CI)
Male gender	0.6 (0.3-1.0)	-	1.0 (0.7-1.5)	**2.5 (1.5-4.2)******	**3.8 (1.4-10.4)****
Premature birth before 37 weeks	2.7 (0.8-8.6)	-	1.7 (0.5-5.0)	**4.4 (1.6-12.6)*****	0.6 (0.0-13.2)
Small for gestational age	0.6 (0.2-1.8)	-	1.5 (0.8-2.8)	0.8 (0.3-1.9)	**2.8 (1.0-7.5)***
Low placental-to-birthweight ratio (<10th percentile)	0.9 (0.4-1.9)	-	1.1 (0.7-1.9)	1.0 (0.5-1.9)	**2.8 (1.2-6.4)***
10-year increase of paternal age at birth	1.4 (0.7-3.0)	-	1.6 (1.0-2.7)	**1.9 (1.0-3.5)***	1.6 (0.5-5.0)
10-year increase of maternal age at birth	0.8 (0.3-2.0)	-	1.0 (0.5-1.8)	0.7 (0.3-1.4)	0.7 (0.2-3.1)
Lower paternal education (<12yrs)	0.7 (0.2-2.2)		0.9 (0.4-2.1)	1.0 (0.4-2.5)	1.6 (0.5-5.1)
Lower maternal education (<12yrs)	0.8 (0.2-3.9)	-	1.6 (0.5-5.0)	2.1 (0.7-6.6)	**4.7 (1.2-19.0)***
1-million-JPY decrease of parental income	1.1 (1.0-1.1)	-	1.0 (1.0-1.1)	1.1 (1.0-1.2)	1.1 (0.9-1.2)

OR: odds ratio; CI: confidence interval

*p<0.05, **p<0.01, ***p<0.005, ****p<0.001

Online supplementary information for "Identification of neurodevelopmental trajectories in infancy and of risk factors affecting deviant development: a longitudinal birth cohort study"

Supplement 1: missingness

We compared neurodevelopmental growth patterns, as assessed by MSEL, between a group of infants excluded from analysis (excluded group) and the remaining infants included in the analysis (included group) (outcome was MSEL T-score). To examine whether there is any association between missingness and growth patterns of neurodevelopment, we examined the interaction of group (excluded vs. included) x slope using a linear mixed model. Because there was no significant interaction or intercepts in any of the MSEL five domains (**Table S1**), we can conclude that missingness is not related to growth patterns as assessed by MSEL. Because we were unable to draw the slope in the case of infants who underwent measurement on one occasion only, we compared the mean scores of MSEL using one-way ANOVA. In the excluded group, almost all of the infants who participated in measurement only once at 1 month of age were found to be births associated with *satogaeri bunben*. We compared the mean scores of MSEL at 1 month of age between the *satogaeri bunben* group and the *non-satogaeri bunben* group (data not shown). Because there was no significant difference in the mean scores between these groups, infants who were born as in the context of *satogaeri bunben* were considered not to be a specific, selected subsample in terms of the neurodevelopment as ascertained by MSEL. We further examined whether, in the included infants, there were any differences in mean scores on each subscale of MSEL between the group of infants who underwent only 3 measurements and the group of infants who underwent all measurements at all 7 time points. Infants who infrequently attended assessment sessions in our cohort may have had a unique profile that is related to neurodevelopmental problems. However, we found no significant interaction of group x slope or intercept in any of the five domains (data not shown). These results suggest that missingness is deemed to be random with respect to growth patterns as measured by MSEL. Therefore, the assumption of "missing at random" (MAR) was not violated when the full information maximum likelihood algorithm was employed in the procedure of latent class growth analysis.

Table S1. Comparison between the group included in the analysis and the group excluded from the analysis by linear mixed model for each domain of Mullen scales.

Domain		Coefficient	SE	P value	95% CI
Expressive Language	age	-0.25	0.13	0.06	-0.50 to 0.01
	intercept (group)	0.24	0.84	0.77	-1.41 to 1.89
	age x group	0.23	0.13	0.08	-0.02 to 0.49
Fine Motor	age	-0.21	0.12	0.07	-0.44 to 0.02
	intercept (group)	-0.56	0.85	0.51	-2.23 to 1.11
	age x group	0.21	0.12	0.08	-0.02 to 0.44
Gross Motor	age	-0.19	0.12	0.11	-0.42 to 0.04
	intercept (group)	0.32	0.85	0.71	-1.35 to 1.98
	age x group	0.16	0.12	0.16	-0.07 to 0.40
Receptive Language	age	-0.10	0.14	0.47	-0.37 to 0.17
	intercept (group)	0.72	1.04	0.49	-1.32 to 2.77
	age x group	0.10	0.14	0.49	-0.18 to 0.37
Visual Reception	age	-0.08	0.12	0.51	-0.32 to 0.16
	intercept (group)	0.16	0.96	0.87	-1.72 to 2.03
	age x group	0.12	0.12	0.32	-0.12 to 0.36

SE: standard error; CI: confidence interval

Supplement 2: development of the Japanese version of T-score norms

To make up T-score norms, we set the standardization sample from our HBC sample. The following were excluded from the procedures: infants with known congenital diseases (Down syndrome; n = 2); pre-term births, defined as birth occurring before 37 weeks (n = 67); twins (n = 34); and parents whose first language was not Japanese (n = 9). Thus, the total size of the standardization sample used for this purpose was 1052. Children included in this sample underwent the Mullen scale assessment on more than one of the seven time points: 1, 4, 6, 10, 14, 18, and 24 months of age.

The raw scores obtained by the standardization sample were divided into the following seven age groups: 0.50–1.49, 3.50–4.49, 5.50–6.49, 9.50–10.49, 13.50–14.49, 17.50–18.49, and 23.50–24.49 months of age. We repeated re-sampling 5,000 times using the bootstrap method and obtained means and SDs of each age group. Then, using the age mid-points of these groups, cubic spline interpolation (1.0–24.0 months of age) or linear extrapolation (0.1–0.9 and 24.1–30.0 months of age) was performed to produce the final set of norm tables for the five Mullen scales. Norms are provided at 0.1-month intervals for ages from 0.1 to 30.0 months. As an example, **Table S2** shows a part of the normalized T-scores for the expressive language domain in a Japanese representative population. Complete T-score tables covering 5 domains are available on request. The data were computed by converting the raw scores obtained for each child into T-scores from the norm tables. The T-score means for each age group on each Mullen scale are close to 50, with SDs close to 10. Good achievement of normalization in T-scores was confirmed (the data not shown).

Table S2. A part of norm table of Expressive Language T-scores corresponding to scale raw scores, by age

Raw score/ Month	0	1	2	3	4	5	…..
…..	…..	…..	…..	…..	…..	…..	…
0.6	17.161076	36.559490	55.957919	75.356322	94.754751	114.15315	
0.7	17.401547	36.363060	55.324585	74.286086	93.247611	112.20911	
0.8	17.642018	36.166630	54.691249	73.215850	91.740470	110.26507	
0.9	17.882488	35.970200	54.057915	72.145615	90.233330	108.32104	…
1.0	18.122959	35.773769	53.424580	71.075378	88.726189	106.37700	…
1.1	18.363430	35.577339	52.791245	70.005142	87.219048	104.43296	…
1.2	18.602648	35.380386	52.158127	68.935860	85.713600	102.49134	…
1.3	18.839357	35.182404	51.525448	67.868484	84.211533	100.55458	…
1.4	19.072308	34.982864	50.893425	66.803970	82.714531	98.625092	…
1.5	19.300243	34.781258	50.262272	65.743279	81.224297	96.705307	…
1.6	19.521912	34.577061	49.632210	64.687347	79.742500	94.797653	…
1.7	19.736061	34.369759	49.003456	63.637146	78.270851	92.904549	…
1.8	19.941435	34.158833	48.376232	62.593624	76.811028	91.028427	…
1.9	20.136782	33.943768	47.750751	61.557732	75.364723	89.171707	…
2.0	20.320848	33.724045	47.127235	60.530426	73.933624	87.336823	…
2.1	20.492380	33.499142	46.505901	59.512661	72.519424	85.526192	…
2.2	20.650124	33.268547	45.886967	58.505386	71.123817	83.742241	…
2.3	20.792828	33.031742	45.270653	57.509563	69.748482	81.987396	…
2.4	20.919235	32.788204	44.657173	56.526138	68.395111	80.264084	…
…..	…..	…..	…..	…..	…..	…..	…

The Japanese version of T-score tables covering 5 domains is available from the corresponding author.

Supplement 3: Fit indices in the latent class growth analysis

Table S3. Fit indices, classification qualities, and class proportions in the latent class growth analysis

Number of classes	1 class	2 classes	3 classes	4 classes	5 classes	6 classes	7 classes
Number of free parameters	50	66	82	98	114	130	146
BIC	217718.3	215239.7	214678.6	214258.9	213962.8	213805.9	213722.0
CAIC	217768.3	215305.7	214760.6	214356.9	214076.8	213935.9	213868.0
AWE	218211.2	215890.4	215487.0	215225.1	**215086.7**	215087.6	215161.3
Adjusted LMR-LRT p-value	-	**<0.001**	0.588	0.257	0.264	0.214	0.725
BLRT p-value	-	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Entropy	-	**0.846**	0.815	0.785	**0.835**	0.805	0.805
Class proportions (%)	100.0	62.6, 37.4	49.8, 36.4, 13.8	39.5, 30.9, 19.4, 10.2	49.2, 21.2, 14.1, 11.5, 4.0	34.9, 22.3, 20.2, 12.8, 8.5, 1.4	36.6, 19.4, 13.9, 12.1, 10.0, 6.7, 1.4

BIC: Bayesian information criterion

$BIC = -2LL + d\log(n)$,

where *LL* is the maximized log likelihood function value to which the EM algorithm converges during the model estimation;

d is the number of parameters estimated in the model;

n is the number of subjects or cases, in the analysis sample.

CAIC: Consistent Akaike information criteria

$CAIC = -2LL + d[\log(n) + 1]$

AWE: Approximate weight of evidence criterion

$AWE = -2LL + 2d[\log(n) + 1.5]$

Supplement 4: Risk factors for the outcome classes measured at 24 months of age

Table S4. Risk factors for the outcome groups based on an MSEL total score at only 24 months of age: Odds ratio and 95% confidence intervals in the analysis of multinomial logistic regression.

24 outcome groups	1. high score 11.5%	2. average score 49.2%	3. average-to-low score 21.2%	4. low score 14.1%	5. lowest score 4.0%
	OR (95%CI)	Reference	OR (95%CI)	OR (95%CI)	OR (95%CI)
Male gender	0.8 (0.5-1.2)		1.1 (0.8-1.6)	**2.2 (1.4-3.4)*****	**3.3 (1.3-8.4)***
Preterm birth before 37 weeks	2.5 (1.0-6.2)		1.3 (0.5-3.1)	1.9 (0.7-5.0)	0.7 (0.1-4.1)
Small for gestational age	0.3 (0.1-1.1)		1.2 (0.7-2.3)	0.8 (0.4-1.8)	2.6 (1.0-6.9)
Low placental- to-birthweight ratio (<10th percentile)	0.9 (0.5-1.7)		1.1 (0.7-1.8)	1.2 (0.7-2.2)	**2.4 (1.0-5.7)***
10-year increase of paternal age at birth	1.5 (0.8-2.9)		1.1 (0.7-1.8)	**1.9 (1.1-3.3)***	0.9 (0.3-2.7)
10-year increase of maternal age at birth	0.6 (0.3-1.2)		1.1 (0.6-1.8)	**0.5 (0.2-1.0)***	1.9 (0.6-6.1)
Lower paternal education (<12yrs)	1.5 (0.7-3.5)		1.2 (0.6-2.6)	1.2 (0.5-2.8)	1.7 (0.5-6.6)
Lower maternal education (<12yrs)	0.7 (0.2-2.6)		1.5 (0.6-3.8)	0.6 (0.2-2.3)	**7.0 (1.9-26.4)*****
1-million-JPY decrease of parental income	1.0 (1.0-1.1)		**1.1 (1.0-1.2)***	**1.1 (1.0-1.2)****	1.0 (0.9-1.1)

OR: odds ratio; CI: confidence interval

*p<0.05, **p<0.01, ***p<0.005, ****p<0.001

Supplement 5: low attrition rates for MSEL measures

In the analysis of subsequent developments after birth, as assessed with the MSEL, for each of 7 time-points (1 month to 24 months), all of the measures achieved high rates of data collection, ranging from 81.9 to 95.7% with the exception of only two measurements on one occasion (47% for receptive language and 60% for visual reception, both at one month of age). Taking into account the nature of difficulties in maintaining high rates of follow-up in the longitudinal studies,[1] these figures can be viewed as very satisfactory. In the analyses, all data available were employed, with the use of full information maximum likelihood methods.

Reference

1. Gilding J, Birmingham K. Enrolment and response rates in a longitudinal birth cohort. *Paediatr Perinat Epidemiol* 2009;23(Suppl 1):73-85.