Growth and Neurodevelopment in Very Preterm Infants receiving a High Enteral Volume-Feeding Regimen -a Population-Based Cohort Study

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ABSTRACT

Aim: To evaluate a feeding regimen routinely providing > 180 ml/kg/d fortified human milk to very preterm infants and impact on in-hospital growth, osteopenia and neurodevelopment.

Method: Retrospective population-based descriptive study of infants < 30 weeks gestation admitted within 24 h of birth and discharged during the six-year period 2005 to 2010. Growth and neurodevelopment was assessed until two years corrected age, and cerebral palsy up to 4 years corrected age

Results: Ninety-nine infants below 30 weeks gestation were admitted within 24 h of birth during the six-year period, of which 84 (85%) survived to discharge. Two infants had surgical necrotizing eneterocolitis, both survived to two years follow up. Seventy-eight infants (mean 27 weeks) had complete growth data until discharge. Full enteral feeds were tolerated after mean 10 days. Average milk volumes were 193 ml/kg/d from 15-42 days of life. Rates of weight below 10^{th} centile were 10% at birth and 14% at discharge. Head circumference Z-scores were stable from birth to discharge. Blood values did not indicate osteopenia. Increasing head circumference Z-scores were associated with improved language development.

Conclusion: This high enteral feeding volume regimen was associated with low rates of in-hospital growth restriction and good head growth. High enteral volume intake seems safe and may improve nutritional status of very preterm infants.

Key words: Infant, newborn, preterm, enteral nutrition, breast milk, feeding volume

INTRODUCTION

In very preterm infants in-hospital growth restriction is still common (1, 2). Early life malnutrition and poor in-hospital growth is associated with increased risk of retinopathy, osteopenia and later impaired long-term growth and neurodevelopment (3-5). It is therefore essential to improve nutrition of preterm infants in order to reduce the negative impact of poor nutrition on growth and later neurodevelopment (6).

It is rarely possible to provide enough enteral nutrition to cover nutritional needs for very preterm infants during the first seven to fourteen days of life. Recent guidelines and expert recommendations thus strongly emphasize the importance of early adequate parenteral nutrition (7-9). There has been less focus on how to optimize the enteral nutrition after these infants have achieved full enteral tolerance and entered a stable growing phase. Guidelines recommend an enteral protein intake of 4.0-4.5 g/kg/d for the healthy growing preterm infant. It is suggested that enteral feeding volumes of 150-180 ml/kg/d, using formula or fortified breast milk, are likely to meet these requirements (10). In a recent large international survey on enteral feeding practice in very preterm infants the majority of neonatal units aimed for enteral feeding volumes of 160-180 ml/kg/d using fortified human milk or preterm formula, and only 9 % of surveyed units aimed for higher feeding volumes (11).

The main objective of this study was to evaluate a feeding regimen routinely targeting a daily enteral feeding volume > 180 ml/kg/d to very preterm infants and its effect on in-hospital growth. Secondly, we evaluated possible biochemical markers of osteopenia of prematurity during hospitalization and the association between in-hospital growth and neurodevelopment at two years corrected age.

PATIENT AND METHODS

Setting and study design

All infants with gestational age (GA) < 30 weeks born in the two northernmost counties in Norway are treated in the tertiary neonatal unit at the University Hospital of North Norway (12). In this population-based, retrospective and descriptive study, we included all very preterm infants born before 30 weeks gestation, admitted within 24 h of birth and discharged during the six-year period January 2005 to December 2010.

Feeding policy

The feeding policy in the neonatal unit remained unchanged during the six-year period. Intravenous amino acids were commenced from first day of life (DOL 1) with 0.5-1.0 g/kg/d and gradually increased to 3.5 g/kg/d on DOL 3-5. Intravenous lipids were increased to 2 g/kg/d on DOL 4-5. Concomitantly enteral nutrition was initiated early, usually on DOL 1. Enteral feeds were gradually increased using continuous gastric drip for the first seven to fourteen days (13). The infants were primarily fed mothers own raw milk. If this was not available, we used unpasteurized donor human milk from the unit's milk bank. Formula was rarely used except under special circumstances when there was a lack of donor milk. For infants older than seven to ten days of age tolerating full feeds (≥ 150 ml/kg/d) we added a multinutrient human milk fortifier (Nutriprem®, Nutricia, Schipol, The Netherlands) providing 0.8 g extra protein per 100 ml breast milk. We routinely continued fortification until the infant reached a body weight of around 2500 g, and we did not adjust the amount of breast milk fortification. The prescribed enteral feeding volume during the "stable growth phase" was adjusted between 170-220 ml/kg/d aiming to achieve optimal weight gain (15-20 g/kg/d) without signs of overhydration.

Clinical data, measurements and follow-up

We collected clinical data from medical records. Initial illness severity was assessed using Clinical risk index babies 2nd version score (CRIB2). Verified necrotizing enterocolitis (NEC) was diagnosed according to Bell stage 2-3 (14). Bronchopulmonary dysplasia (BPD) was defined as need for oxygen or respiratory support at postmenstrual age (PMA) 36 weeks. We collected data on nutrition including duration of parenteral nutrition, onset of enteral nutrition and age (in days) at full enteral tolerance, defined as 150 ml/kg/d. A previous audit in our neonatal unit showed that most very preterm infants tolerated full enteral feeds within ten to fourteen days of age, in line with findings from other Norwegian units (11). We therefore defined DOL 15-49 (week 3-7) as a "stable growth phase". During this period we calculated the average daily enteral volume intake for infants tolerating full enteral feeds (≥ 150 ml/kg/d) on at least 6 out of 7 days, and present these data for DOL 15-49 (week 3-7). We excluded weeks where infants had not yet achieved full feeds or weeks with intercurrent illness episodes making them too sick to tolerate full enteral feeds. Thus, the presented intake values are data from infants on full feeds in a stable growth phase, reflecting enteral volume tolerance. Weight was assessed daily for the first two to three weeks and then every second to third day until discharge. Weight and height was also recorded on follow-up until two years corrected age.

Data on weight and head circumference (HC) from birth to discharge were converted to standard deviation (SD; Z) scores using the 2013 Fenton growth chart (15). The Fenton chart does not estimate HC Z-score for infants < 24.0 weeks gestation and we decided to use the Z-scores for 24.0 weeks gestation for all five infants with GA between 23.0-23.6 weeks. Data on length from birth to discharge was not reported due to the poor reliability of length measurement in the routine neonatal intensive care and many missing values. Small for gestational age (SGA) was defined as weight < 10th centile for age. To illustrate in-hospital

growth we calculated difference in Z-score for weight and HC from birth to discharge. Weight and body mass index at one and two year corrected age was assessed using the WHO growth chart as reference for Z-scores (16).

Biochemical markers suggested indicating osteopenia of prematurity (alkaline phosphatase, phosphate and ionized calcium) were measured from around two weeks of age and thereafter weekly during the study period. We report these results for three separate periods; DOL 15-28, DOL 29-42 and DOL 43-56. We averaged the values if several blood tests were taken within these periods. Hyperphosphatasia, hypophosphatemia or hypocalcemia were defined as alkaline phosphatase (ALP) > 800 U/L, serum phosphate <1.4 mmol/L and serum ionized calcium < 1.10 mmol/L, respectively.

Most infants were included in our routine follow-up program at one and two years corrected age with assessment of growth, neurological examination by a pediatrician and motor assessment by a physiotherapist. Bayley III (BSID-III) neurodevelopmental assessment including total cognitive and language scores, but not motor score, was performed at two years corrected age.

Statistical analysis

Data were analysed using IBM-SPSS statistical software version 23 (IBM, New York, USA). Normally distributed interval data are displayed using mean and standard deviation (SD), and non-normally distributed data are displayed using median and interquartile range (IQR). Categorical data are displayed as frequency (%). We used the chi square test for categorical data and the T-test or Mann-Whitney U test for interval data. A multiple regression model was used to analyze associations between in-hospital growth and neurodevelopment. We included five possible predictors for neurodevelopment; sex, BPD, SGA at birth and Z-score changes for weight and HC from birth to discharge. We performed unadjusted analyses and a stepwise

logistic regression analysis including all five predictors. We defined p values < 0.05 as significant.

Ethics

The regional ethical committee of Northern-Norway approved this study.

RESULTS

Ninety-nine infants below 30 weeks gestation were admitted within 24 h of birth during the six-year period, of which 84 (85%) survived to discharge. The fifteen infants who died were significantly more immature (mean GA 24.9 vs 27.3 weeks), had lower Apgar values (mean 5.1 vs 7.5) and higher initial illness severity (mean CRIB2 score 13.3 vs 9.2) than those who survived. Both infants with NEC were surgically treated and both survived to follow-up at two years corrected age. Six infants with mean GA 28 weeks and mean birth weight 1100 g were transferred to other neonatal units prior to 34 weeks PMA, and were not included in our follow-up.

The core population of this study was therefore 78 infants with mean GA 27 weeks (Table 1). The average enteral daily volume intake from third to seventh week of life is shown in Figure 1. From the third to the sixth week of life most infants received between 190-200 ml/kg/d, provided full enteral tolerance. Thereafter the enteral volume was gradually reduced, but on average kept around 180 ml/kg/d until the seventh week of life. Data on growth and nutrition, from birth and up to two years corrected age for all infants in this population based-study, are shown in Table 1 and Figure 2a-b. The proportion of infants classified as SGA was: at birth 10%, at PMA 34 weeks 17% and at discharge (median 39 weeks) 14%. The mean Z-score for weight decreased from -0.16 at birth to -0.48 at discharge. The mean Z-score for HC was stable from birth to discharge.

Compared with infants born at 28-29 weeks gestation, infants born before 28 weeks gestation tolerated full feeds later, received longer parenteral nutrition and had higher rates of BPD (Table 1). The more immature infants were discharged later and had correspondingly higher weight at discharge. The immature infants (< 28 weeks gestation) seemed to have more catch-up growth during the last weeks of admission, reflected by a decrease in the proportion of infants with weight < 10^{th} centile from 18% at 34 weeks to 12% at discharge.

Growth data were missing for six and fifteen children at one and two years corrected age, respectively. The Z-scores for weight at discharge and the Z-score changes between birth and discharge were similar between those missing data and those with complete follow-up weight data until two years corrected age. Overall, the Z-scores for weight were stable for the whole study population from discharge and until two years corrected age (Table 3, Figure 2a).

The mean serum values for ALP, phosphate and ionized calcium were within normal limits for the majority of all infants (Table 2). There were five, three and no infant with ALP > 800 U/L during first (DOL 15-28), second (DOL 29-42) and third (DOL 43-56) period, respectively. No infants had ALP > 900 U/L. There were six infants in the first period with hypophosphatemia (1.07-1.35 mmol/L). During the second and third period, no infants had hypophosphatemia. Infants with phosphate < 1.8 mmol/L had significantly higher ALP levels in the first period compared to those with phosphate ≥ 1.8 mmol/L (mean 560 vs 380 U/L, p 0.029). During the second and third period, there was no correlation between ALP and phosphate levels. One patient in the first period, one in the second period and no patients in the third period had hypocalcemia.

Seven of 78 (9.0%) infants developed cerebral palsy up to four years of age. Four out of five infants diagnosed with mild cerebral palsy (gross motor function classification scale-GMFCS-1-3) were diagnosed after two years of age. BSID-III cognitive and language scores were available for 54 of 78 infants. The Z-scores for weight and HC at discharge, and the

respective Z-score changes between birth and discharge, were similar between those missing (n=24) and those with BSID-III data (n= 54) available. For all 54 infants the mean (SD) cognitive index was 95.7 (8.8) and the mean (SD) language index was 94.4 (12.1). Three children (5.5 %) had cognitive score index < 85 and nine children (17%) had language score index < 85. In the unadjusted linear regression analysis, Z-score changes in HC and female sex were both associated with improved language score (Figure 3, Table 3). In the stepwise multiple regression models only Z-score changes in HC remained significant (Table 3). We did not find any other associations between Z-score for birth weight and HC or in-hospital growth and total language or cognitive scores.

DISCUSSION

In this population-based observational study of very preterm infants born before 30 weeks gestation we found that the proportion of infants growth restricted at discharge (14%) was similar to those who were SGA at birth (10%). Median age at discharge was 39 weeks with a body weight around 3 kg, and the Z-scores for weight at discharge were within normal limit for most infants. Follow-up at one and two years corrected age showed a stable growth pattern along the same Z-scores for weight and body mass index.

Many recent studies from the USA and Europe still report high prevalence of in-hospital growth restriction in very preterm infants (1, 2, 17). There are some exceptions. Graziano and co-workers introduced an improved “feeding bundle” in their unit, and reported in a cohort of very preterm infants that 19% were growth restricted at discharge and Z-scores for weight at discharge were similar to the findings in our study (18). In their study, with slightly more mature infants, time to full enteral feeds was much longer (18-21 days), milk fortification started earlier, but target maximum daily fluid intake of 160 ml/kg/d was lower than in our

study. A retrospective study from Germany, with a similar population of preterm infants than in our study, reported only eight days to full enteral feeds and a low rate of in-hospital growth restriction (19). However, in both the German study (19) and a recent study from the USA (20) the use of fortified human milk compared to preterm formula was associated with lower weight gain among preterm infants. In our study, fortified human milk was exclusively used up to 34 weeks gestation and for most infants until discharge, and growth was still good.

The main difference between our study and many other reports on enteral nutrition to preterm infants is the high enteral volume intake (18, 21). For our cohort of infants with a mean birth weight < 1000 g the recommend enteral protein intake is 4.0-4.5 g/kg/d (10). After the first four weeks of lactation, the protein content of expressed preterm milk declines to around 1.2 g/100 ml (22). During the study period we used a human milk fortifier increasing protein content by 0.8 g/100 ml breast milk, but more current commercial human milk fortifiers increases protein content by 1.0-1.4 g/100 ml (23). Still, an enteral volume of 180 (160-200) ml/kg/d is required to achieve a protein intake of 4.0-4.5 g/kg/d. It is also a clinical perception that not all preterm infants tolerate full strength fortification. Increasing enteral fluid volume is then an alternative in order to increase protein and energy delivery. The parenteral nutrition regimen in our unit has been slightly modified since 2011. However, an "aggressive approach" with early very high amino acid and lipid delivery may have early and prolonged side effects (24-26). We believe it is safer to focus on achieving early full enteral tolerance and optimize enteral nutrition in the stable growth phase. An adequate protein intake is the major driving force of growth in preterm infants (27). Early enteral protein intake is positively associated with neurodevelopmental outcome (21). However, recent data indicate that there may be a ceiling effect of enteral protein supply (23). We still believe that low daily enteral volumes often leads to an intake of protein below the recommended range which may increase the risk of in-hospital growth restriction.

In Scandinavia, enteral breast milk feeding is usually commenced early and increased relatively fast, and the prevalence of NEC among extremely low birth weight infants is around 5-6% (28, 29). In our study, infants < 30 weeks gestation tolerated 150 ml/kg/d at a median age of ten days and only two children developed NEC, both surviving to discharge. Systematic review data suggest that advancing enteral feed volumes at daily increments of 30-40 ml/kg (compared to 15-24 ml/kg) does not increase the risk of NEC or death in very low birth weight infants (30). Large fluid volumes may be difficult to handle for some preterm infants, in particular those with BPD. However, fluid restriction may lead to energy deficiency and can negatively impact postnatal growth (31). A careful balance of fluid and energy supply is crucial.

Infants provided high enteral feeding volumes of fortified breast milk experienced a modest decrease in Z-scores for weight (-0.3), but no changes in Z-score for HC from birth to discharge. Better head growth than overall weight gain in human milk fed very preterm infants is consistent with the previously reported “breastfeeding paradox” that also describes better neurodevelopmental outcome in human milk fed infants in spite of suboptimal initial weight gain (20).

In our study most blood values suggested as “biochemical markers of osteopenia of prematurity” were normal or normalised without any specific intervention. In one study the combination of ALP > 900 U/L and phosphate < 1.8 mmol/L detected lower bone mineral density with a sensitivity of 100% and a specificity of 70% (32). No infants had ALP > 900 U/L and there was no correlation between phosphate < 1.8 mmol/l and ALP values after the fourth week of life. Osteopenia of prematurity is a radiological diagnosis, but a major loss of bone mineralization has to occur before radiological changes are visible. We did not diagnose any infant with osteopenia of prematurity during the study period, but no routine screening with radiology or bone density measurements were performed. The reassuring values for ALP

and phosphate combined with low rates of growth restriction may thus indicate that infants received sufficient minerals and nutrition for bone growth. Currently, there is insufficient evidence that any blood values are valid as early biochemical markers of osteopenia in preterm infants (4).

Relatively few studies report neurodevelopmental outcome of feeding regimens (3, 6), but recent recommendations suggest this to be mandatory when reporting neonatal nutrition and growth outcomes (22). In our study, the majority of infants had cognitive and language score above -1 SD. In an explorative statistical analysis, we found an association between increasing Z-scores of HC and positive language scores. This finding need cautious interpretation due to small numbers and no prospective predefined selection of statistical analyses.

This study has strengths, but also inherent limitations associated with a retrospective single-center observational study. First, we have analyzed an unselected population-based cohort but the small sample size limits statistical power to detect differences in outcomes after subgroup comparisons. Second, body weight is only a proxy of growth and protein accretion. The infants in our cohort had low rates of postnatal growth restriction, but investigations with air displacement plethysmography or dual-energy X-ray absorptiometry scan would have revealed better data on body composition (22). Third, we did not have consistent data on head circumference during follow-up after discharge and no data on parental socioeconomic status, both parameters that may affect neurodevelopmental outcome. Fourth, we did not have a control group and other factors than the feeding regimen may contribute to the good growth observed. Finally, the incomplete data on neurodevelopment assessment limits the power to detect differences depending on growth. However, our data shows that it is possible to achieve satisfactory in-hospital growth in a population of very preterm infants with corresponding good neurodevelopment up to 2 years corrected age.

CONCLUSIONS

In this retrospective study a feeding regimen routinely providing > 180 ml/kg/d fortified human milk to very preterm infants was associated with low rates of in-hospital growth restriction and good head growth. Potential benefits or harm of providing high enteral feeding volumes to clinically stable preterm infants after 2 weeks of life should be tested in a large randomized trial.

List of abbreviations:

BPD; Bronchopulmonary dysplasia

BSID-III, Bayley Scales of Infant and Toddler Development, Third Edition

DOL; day of life

GA; gestational age

HC; head circumference

NEC; necrotizing enterocolitis

IQR; interquartile range

PMA; postmenstrual age

SD; standard deviation

SGA; Small for gestational age

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Authors' contributions

CK designed the study, performed the analyses, drafted the initial manuscript and revised the manuscript. FKM collected and analyzed data on growth and neurodevelopment and performed the analyses. CEI collected and analyzed data on growth and feeding. TN and MT collected and analyzed data on bone health and growth. CM-H performed all neurodevelopmental tests. All authors made important intellectual contributions, read and approved the final manuscript.

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Table 1: Core study population – nutrition and growth data

	All	**GA 28-29 weeks**	**GA < 28 weeks**	**p**
	n=78	**n=29**	**n=49**	
Gestational age (weeks), mean (SD)	27.2 (1.5)	28.7 (0.5)	26.3 (1.1)	
Birth weight (gram), mean (SD)	943 (231)	1089 (248)	857 (170)	
Z-score, birth weight, mean (SD)	-0.16 (0.79)	-0.29 (0.82)	-0.09 (0.77)	0.30
SGA at birth (%)	8 (10)	4 (14)	4 (8)	0.50
Z-score, HC at birth, mean (SD)	0.32 (1.02)	0.13 (1.14)	0.44 (0.93)	0.21
Male (%)	47 (60)	17 (59)	30 (61)	0.82
Onset enteral nutrition 0-24 h (%)	63 (81)	24 (83)	39 (80)	0.85
Day of life full feeds (150 ml/kg), median (IQR)	10 (7-14)	7.5 (6-12)	11 (8-15)	**0.024**
Days parenteral nutrition, median (IQR)	8 (5-13)	6 (2-11.5)	9 (6-14)	**0.013**
Bronchopulmonary dysplasia (%)	31 (40)	6 (21)	25 (51)	**0.009**
Necrotising enterocolitis (%)	2 (2.6)	0 (0)	2 (4.1)	0.27
Weight (gram), 34 weeks PMA, mean (SD)	1999 (319)	1973 (343)	2015 (307)	0.58
Z-score, weight 34 weeks PMA, mean (SD)	-0.50 (0.79)	-0.57 (0.86)	-0.47 (0.75)	0.60
SGA at 34 weeks PMA (%)	13 (17)	4 (14)	9 (18)	0.60
PMA in weeks at discharge, median (IQR)	39 (37-41)	37 (36-40)	39 (37-42)	**0.01**
Weight at discharge (gram), median (IQR)	2994 (2707-3420)	2755 (2543-2994)	3180 (2778-3580)	**0.001**
Z-score, weight at discharge, mean (SD)	-0.48 (0.79)	-0.61 (0.84)	-0.40 (0.77)	0.24
SGA at discharge (%)	11 (14)	5 (17)	6 (12)	0.63
Z-score, HC at discharge, mean (SD)	0.35 (0.90)	0.48 (0.81)	0.27 (0.95)	0.34
Z-score, weight 1 year CA, mean (SD)	-0.47 (1.19)	-0.52 (1.50)	-0.44 (0.96)	0.78
Z-score, BMI 1 year CA, mean (SD)	-0.20 (1.16)	-0.11 (1.38)	-0.26 (1.02)	0.61
Z-score, weight 2 year CA, mean (SD)	-0.41 (1.13)	-0.47 (1.26)	-0.38 (1.07)	0.76
Z-score, BMI 2 year CA, mean (SD)	0.13 (1.17)	0.34 (1.40)	0.03 (1.03)	0.34

BMI; Body mass index, CA; corrected age, GA; gestational age, HC; head circumference, IQR; interquartile range, PMA; postmenstrual age, SD; standard deviation, SGA; small for gestational age < 10^{th} centile. Growth data at one and two years CA for 72 and 63 infants, respectively

Table 2: Metabolic markers suggested indicating osteopenia

	DOL 15-28	**DOL 29-42**	**DOL 43-56**
Serum alkaline phosphatase, U/L	452 (159)	405 (154)	362 (129)
Serum phosphate, mmol/L	2.07 (0.36)	2.11 (0.25)	2.09 (0.23)
Serum ionized calcium, mmol/L	1.37 (0.08)	1.38 (0.07)	1.38 (0.05)

DOL; day of life.
All values are mean and standard deviation

Table 3: Neurodevelopment at 2 years corrected age - growth and clinical variables

Linear regression analysis - Language index BSID-III

Unadjusted analyses	Regression coeffecient	95% CI	Adjusted analyses	Regression coeffecient	95% CI
Female sex	7.00	0.39 to 13.63	Female sex	0.21	-2.20 to 11.91
Δ Z-score HC - birth to discharge	3.61	0.03 to 7.19	Δ Z-score HC - birth to discharge	3.61	0.03 to 7.19
Δ Z-score weight - birth to discharge	-1.11	-5.51 to 3.28	Δ Z-score weight - birth to discharge	-0.19	-9.63 to 1.79
BPD	2.00	-4.68 to 8.68	BPD	0.14	-3.61 to 10.18
SGA	-1.76	-11.67 to 8.15	SGA	0.03	-10.48 to 10.39

Linear regression analysis - Cognitive index BSID-III

Unadjusted analyses	Regression coeffecient	95% CI	Adjusted analyses	Regression coeffecient	95% CI
Female sex	2.00	-3.00 to 7.00	Female sex	1.03	-6.20 to 13.25
Δ Z-score HC - birth to discharge	0.59	-2.04 to 3.23	Δ Z-score HC - birth to discharge	1.30	-4.61 to 7.21
Δ Z-score weight - birth to discharge	-1.46	-4.65 to 1.73	Δ Z-score weight - birth to discharge	-1.65	-9.53 to 6.22
BPD	2.42	-2.43 to 7.26	BPD	5.97	-3.54 to 15.48
SGA	0.79	-6.44 to 8.02	SGA	-0.22	-12.75 to 16.12

BPD; bronchopulmonary dysplasia, BSID-III, Bayley Scales of Infant and Toddler Development, Third Edition; HC; head circumference, SGA; small for gestational age < 10th centile.

NS; not significant

Figure 1

Average enteral daily volume intake from third to seventh week of life in infants tolerating full enteral feeds. Core study population (n=78)

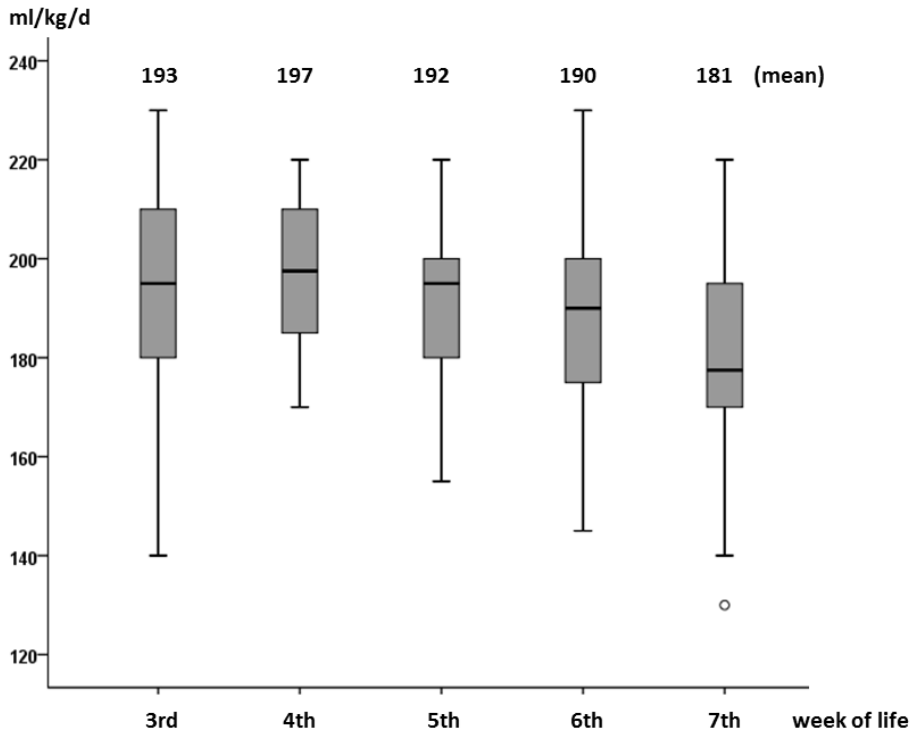

Figure 2 a

Z-score weight and body mass index (BMI). From birth to discharge (n=78) and at 1 year (n = 72) and 2 year corrected age (n=63)

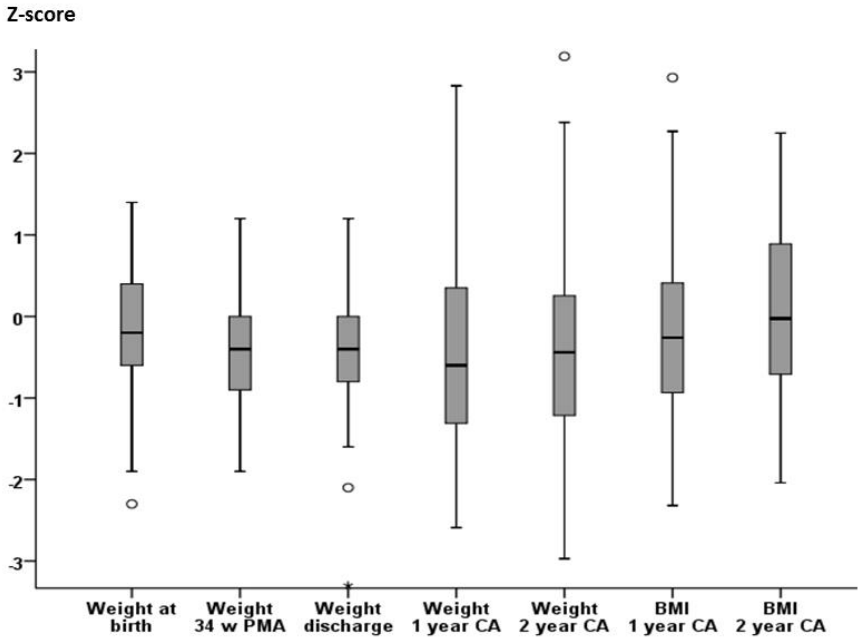

Figure 2A

Figure 2 b

Z-score head circumference from birth to discharge (n=74)

Figure 2B

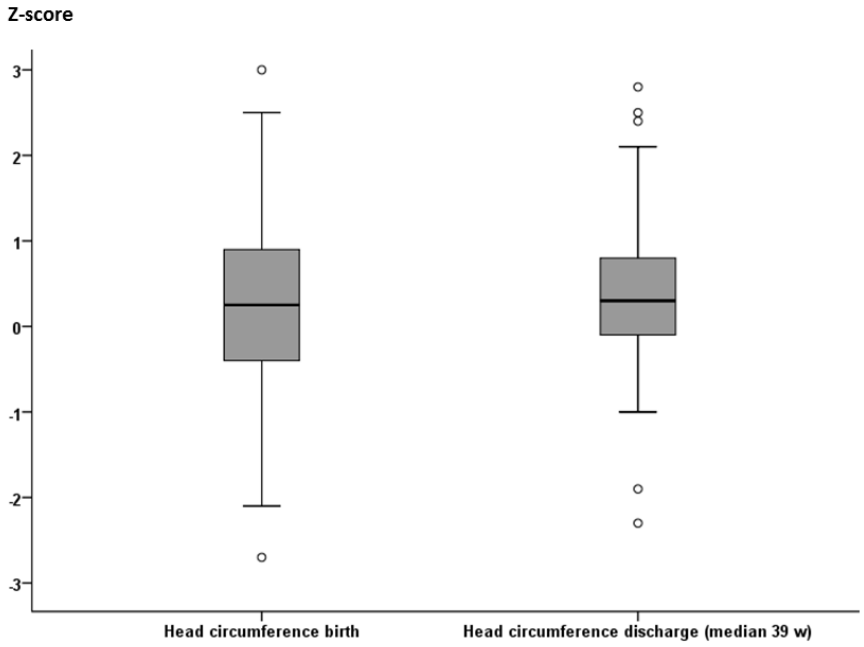

Figure 3

Correlation between change in Z-score head circumference from birth to discharge and total language score

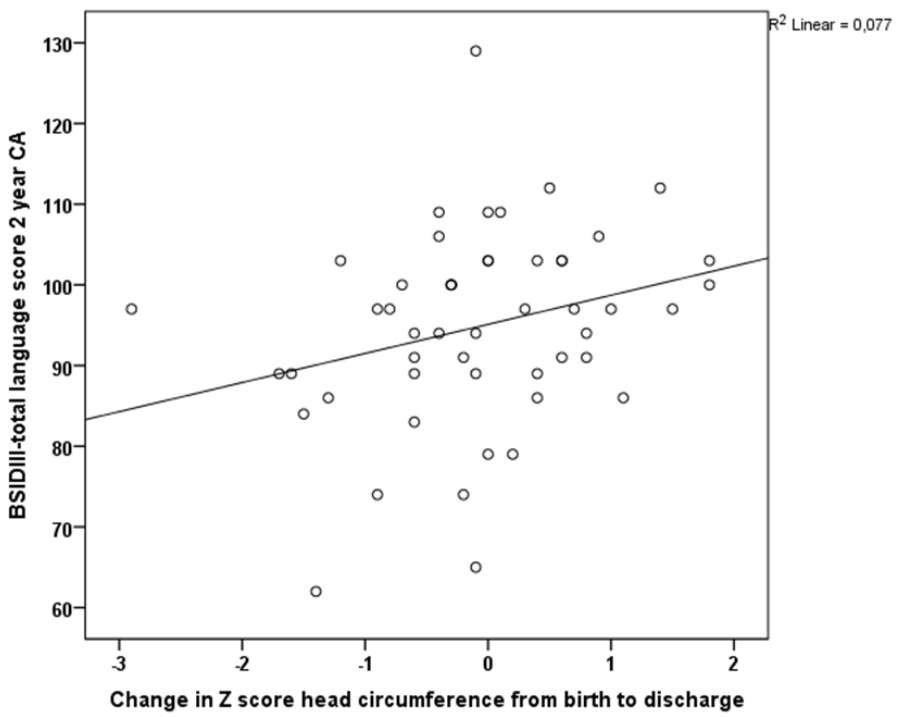

Figure 3