

Neurological Development and Iron Supplementation in Healthy Late-Preterm Neonates: A Randomized Double-Blind Controlled Trial

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Abstract

Objective

Late-preterm infants (LPT) are at increased risk for long-term neurodevelopmental sequelae and iron deficiency. Aim of the study is to assess the positive effect of iron supplementation on neurological development in healthy LPT.

Design

We designed a perspective, randomized placebo-controlled double-blind trial. The newborns were randomized in two groups: thirty-three patients received martial prophylaxis, thirty-three placebo. Every patient was assessed using the Griffith Mental Development Scales (GMDS)-II edition at 12 months of post-conceptual age.

Setting

The study was performed at the Neonatology Unit of Fondazione Policlinico Gemelli IRCCS.

Patients

Sixty-six healthy LPT infants born between 34^{0/7} and 36^{6/7} weeks of Gestational Age were enrolled in the study.

Interventions

One group received martial prophylaxis from the third week of life to six months of post-conceptual age (2 mg/kg/day of iron pidolate), the other received placebo.

Main outcome measures

Fifty-two of the enrolled infants were assessed using the GMDS at 12-month of post-conceptual age. Statistical analysis of the mean scores of the Griffith subscales was performed.

Results

There was a difference in the mean Developmental Quotient (DQ) ($p < 0.01$) between the two groups: Iron Group mean DQ 121.45 ± 10.53 vs Placebo Group mean DQ 113.25 ± 9.70 . Moreover, mean scores of the Griffith subscales A, B and D showed significant differences between the two Groups (scale A $p < 0.05$, scale B $p < 0.02$, scale D $p < 0.01$ respectively).

Conclusions

Our data show that newborns who received iron supplementation during the first six months of life achieved significantly better neurological outcomes at GMDS than Placebo group.

What Is Known

- Late-preterm infants (LPT) are at increased risk for long-term neurodevelopmental sequelae and iron deficiency.
- Iron deficiency is an independent risk factor for adverse neurological outcomes.

What Is New

- Healthy late-preterm who received iron supplementation during the first six months of life achieved better neurological outcomes at twelve months than LPT who received placebo.
- Our study strongly supports the need for the implementation of maternal prophylaxis in healthy late-preterm.

Introduction

The term 'Late-preterm infants' (LPT), is used to define infants born at 34^{0/7} through 36^{6/7} weeks of gestation. They account for more than 70% of all preterm births¹.

Several papers have reported that, when compared to infants born at term, LPT infants are at increased risk for neonatal morbidities and long-term neurodevelopmental sequelae^{2,3,4}. It has also been reported that LPT newborns have an increased risk of developing iron deficiency (ID), due to both limited reserves and increased iron requirements^{5,6}. As extensively demonstrated by preclinical studies in rodents^{7,8} and human trials^{9,10}, iron deficiency is an independent risk factor for adverse neurological outcomes.

Despite the European Society for Pediatric Gastroenterology Hepatology and Nutrition (ESPGHAN) Committee on Nutrition and American Academy of Pediatrics (AAP)^{11,12} recommends iron supplementation in all preterms and low-birthweight infants (birthweight <2500 g), iron supplementation in LPT population is not a current clinical practice¹³ as it is in neonates born at lower gestational ages.

Aim of our study is to assess the effects of iron supplementation on neurological development in healthy late-preterm infants evaluated at 12 months of corrected age.

Material And Methods

We designed a perspective, randomized placebo-controlled double-blind trial (RCT) in order to assess the possible effect of iron supplementation on neurological development (primary outcome of the study) in healthy LPT. The RCT was sponsored by Pediatrix Specialist[®], that provided both iron and placebo preparation.

Sixty-six healthy LPT infants born between 34^{0/7} and 36^{6/7} weeks of Gestational Age (GA) at Fondazione Policlinico Gemelli IRCCS - Catholic University of Rome and admitted to the Neonatology Unit (Rooming-

in ward) were enrolled in the study, once informed consent was obtained from the parents. GA was evaluated according to the first trimester ultrasound scans or, when not available, with the last menstrual period confirmed by Ballard's score¹⁴.

All enrolled neonates were submitted to cerebral ultrasound assessment at birth.

Infants with uncertain GA, major congenital anomalies, congenital or neonatal infections, neurological and/or neurosensory disorders, cerebral ultrasound anomalies, severe intrauterine growth restriction (SGA <3° centile), neonatal asphyxia, respiratory distress, hematologic disorders were excluded from the study, as well as newborns admitted to Neonatal Pathology Unit or Intensive Care Unit.

All neonates had a complete blood count check to exclude anemia at two weeks of age according to Christensen reference values¹⁵.

The newborns enrolled in the study were randomized in two equal groups (thirty-three patients each group); one group received martial prophylaxis from the third week of life to six months of post-conceptual age (Iron group), the other received placebo (Placebo group), similar in shape and flavor and administered in the same way. Iron supplementation consisted of a dosage of 2 mg/kg/day orally in two administrations of iron pidolate in drops (Pediafer plus® drops 15 ml).

A clinical examination was performed at 6 months of age to ascertain the compliance in treatment regimen and to assess the infants' general wellness. Every patient was assessed using the Griffith Mental Development Scales (GMDS)-II edition at 12 months of post-conceptual age. The GMDS includes five subscales: A (locomotor), B (personal and social development), C (hearing and speech), D (hand and eye coordination) and E (performance). Each scale provides a mental age and a developmental quotient (DQ). The total DQ was calculated from the mean of the developmental quotients obtained in each of the five subscales.

The DQ was considered "normal" if greater than 85, "borderline" between 85 and 70 and suggestive of development delay if less than 70.

The RCT was approved by the Ethical Committee of our institution, Protocol N° 11218/13.

STATISTICAL ANALYSIS

We calculated the sample size on the hypothesis that iron supplementation would be able to increase neurodevelopmental GMDS scores by approximately 8%. We decided to fix an 8% improvement in supplemented group based on Morag et al.¹⁶ study results. The Authors compared the scores obtained by 124 late preterm and 33 term infants and found significantly lower scores in the preterm group in all subscales, being 8% the greatest difference obtained in the Performance scale. According to the study by Morag et al.¹⁶, where the mean Developmental Quotient for Performance Subscale was 84, with a Standard Deviation (SD) of 10, the expected mean Developmental Quotient for Performance Subscale

was estimated as 91 with a SD of 10. Setting a significance level of 0.05 and a power of 0.80, the calculated sample size required was determined to be 33 for each study group.

Statistical analysis was performed using Graphpad software 2018[®]. Student t test for independent data was used to evaluate differences in neurological assessment between the two groups of our study. For all analyses a p-value <0.05 was considered significant.

Results

Thirty-three newborns were randomized to the Iron group, thirty-three to the Placebo group.

Five neonates did not comply with the drops assumption protocol and were excluded from the study, nine neonates were lost to follow-up before the twelve-month neurologic evaluation.

Fifty-two of the initially enrolled infants (twenty-seven in the Iron group and twenty-five in the Placebo group) were assessed using the GMDS at 12-month of post-conceptual age (*Figure 1*). The two groups had similar baseline clinical features (*Table 1*). All the infants assessed at 12 months of age were found in good clinical conditions when evaluated at 6 months of age.

There was a difference in the mean Developmental Quotient (DQ) ($p < 0.01$) between the Iron (mean: 121.45; SD: 10.53) and the Placebo group (mean: 113.25; SD: 9.70) (*Graphic 1*).

Treated infants had higher scores in all Griffith's scales.

The analysis of the mean scores of the Griffith subscales showed differences in scale A (motor scale) ($p < 0.05$), B (behavior) ($p < 0.02$) and D (hand and eye coordination) ($p < 0.01$). The differences found in the C (hearing and speech) and E (performance) scales did not reach statistical significance (*Graphic 2, Table 2*).

None of the patients recruited in the study had DQ scores suggestive of developmental delay. Three infants in the Placebo group had borderline scores compared to none in the Iron group (*Table 3*).

Discussion

During the first 5 to 6 months of life, the need for iron expressed by term infants is filled primarily from iron stores created during pregnancy, corresponding to approximately 25% of total body iron¹¹. The magnitude of iron stores, however, depends on gestational age at birth. Because the iron supply from the placenta is abruptly interrupted with preterm birth, iron storage at birth is lower in late preterm than term infants. In contrast to minor storage at birth, a higher supply of iron is required during the first months of life after a preterm birth because growth velocity is maximally increased between 28 and 38 weeks of gestation. The iron stores of late preterm infants may, consequently, be depleted earlier than 5-6 months of life when iron rich complementary foods are added to the diet. Thus, iron supplementation is needed in

preterm infants to meet the sustained high demands for hematopoiesis, tissue accretion and brain development¹⁷.

Iron is important for neurological development due to its determinant role in dendritic arborization, myelination, neurotransmitters' metabolism, glucose homeostasis and metabolites utilization in the hippocampus^{7,8,18}. Several studies have also shown the negative effects of iron deficiency in cognitive and motor functions, socio-emotional behavior, auditory and visual function^{9,10}. Berglund et al., reported a lower rate of behavioral problems on the Child Behavior Checklist (CBCL) at 3.5 and 7 years in moderately low birth weight infants (MLBW = BW 2000-2500 g) supplemented with iron when compared to a placebo group. They demonstrated in favor of supplemented children¹⁹. Following these studies the ESPGHAN Committee on Nutrition included MLBW infants in the list of children who should receive iron supplementation at a dose of 1-2 mg/kg/day¹¹.

A recent survey by Parodi et al. however showed that less than a quarter of neonatology units of seventy-five Italian hospitals routinely prescribed iron supplementation in healthy late-preterm newborns¹³.

Our data show that newborns who received iron supplementation had significantly higher scores at 1 year of post-conceptual age compared to untreated placebo patients on both mean total DQ and on the subscales assessing locomotor, personal social and eye and hand coordination. The quotients in the other subscales were also higher than in the placebo group but did not reach statistical significance.

Analyzing the whole recruited population, we found three scores indicative of a "borderline" neurological development, all in the Placebo group, while none of the newborns in the Iron group presented borderline or pathological scores. The results in the untreated placebo group are consistent with the natural history of LPT infants^{3,4}. These results should not be driven by possible bias in the selection of the groups as they had similar clinical variables at birth (sex, gestational age, weight and cardiorespiratory adaptation at birth).

Our data, obtained in a relatively small cohort, provide preliminary information that could be used to power larger studies that would allow a better stratification by sex, gestational age and other variables. Another limitation is the short follow-up and our preliminary data should be confirmed by a longer follow-up. Further studies will be needed to evaluate the neurological outcome of these infants in preschool and scholar age.

Conclusions

Our data show that newborns who received iron supplementation during the first six months of life achieved significantly better neurological outcomes at GMDS than Placebo group in terms of total DQ and A, B and D subscales. Therefore, our study strongly supports the need for the implementation of martial prophylaxis in this population.

The main limit of the study is the timing of neurodevelopmental assessment, which does not have the same predictive value of a preschool evaluations.

Reevaluation of children at pre-scholar age would be of interest as well as collecting data on larger populations with the aim to perform assessments for subgroups based on sex, gestational age and breast milk assumption.

Abbreviations

AAP: American Academy of Pediatrics

DQ: Developmental Quotient

ESPGHAN: European Society for Pediatric Gastroenterology Hepatology and Nutrition

GA: Gestational Age

GMDS: Griffith Mental Development Scales

ID: Iron Deficiency

LPT: late-preterm

RCT: Randomized Controlled Trial

SD: Standard Deviation

SGA: Small for Gestational Age

Declarations

Funding: the research was funded by Pediatrix Specialist[®].

Conflicts of interest: the Authors have nothing to declare.

Availability of data and material (data transparency): data are available at Fondazione Policlinico Gemelli IRCCS

Code availability: N/A

Authors' contributions: RL, GV, CR and EM contributed to conception and design of the study. RL, GV, CR, EM and DR wrote the protocol. DR, SS, CV, RL, CS, GM performed the assessments. CD and GM contributed to acquisition and analysis of data for the work. RL, EM, DR and ATC wrote the first draft of the manuscript. All authors contributed to manuscript revision, read, approved and verified the accuracy of the submitted version.

Ethics approval: The RCT was approved by the Ethical Committee of our institution, Protocol N° 11218/13

Consent to participate: all the parents of the children gave consent to participate in the trial

Consent for publication: all the parents of the children gave consent for publication of the resulting data.

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Tables

Table 1: study population: characteristics

	IRON Group (n=27)	PLACEBO Group (n=25)
Sex (M)	13/27	15/25
GA (weeks)	35,33 ± 0,73	35,2 ± 0,76
Birth weight	2465 ± 410,22	2522,17 ± 392,72
Apgar Score 1'	8,48 ± 0,77	8,57 ± 0,93
Apgar Score 5'	9,4 ± 0,5	9,67 ± 0,48

Table 2: Mean Developmental Quotient of the Griffith's Subscales in the two groups and SD

	Iron Group (n=27)		Placebo Group (n=25)		P value
	Mean	SD	Mean	SD	
DQ (A)	127,73	18,30	117,37	15,53	p <0,05
DQ (B)	123,76	11,50	116,29	10,60	p <0,02
DQ (C)	117,90	13,04	112,86	13,33	Non-significant
DQ (D)	120,67	14,25	109,06	14,42	p <0,01
DQ (E)	117,19	15,01	110,66	11,70	Non-significant

Table 3. Borderline and Pathological DQ results in the two groups

	Iron Group	Placebo Group	P value
Borderline DQ	0/27	3/25	Non-significant
Pathological DQ	0/27	0/25	Non-significant

Figures

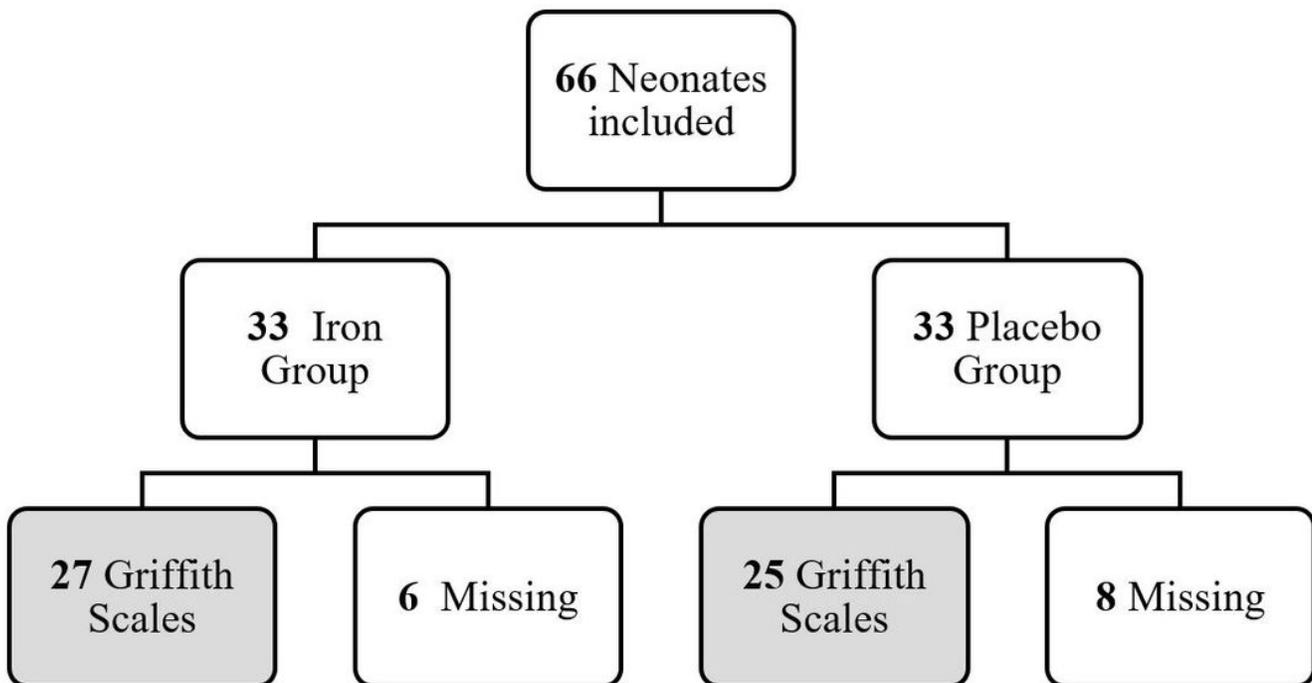


Figure 1

Study population

Supplementary Files

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