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Incidence of metabolic bone disease in neonates under 32 weeks at the Hospital Universitario de Santander in Colombia

Incidencia de enfermedad metabólica ósea en neonatos <32 semanas en el Hospital Universitario de Santander en Colombia

Metabolic bone disease in neonates <32 weeks

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Introduction. Metabolic bone disease of premature infants is a rare complication, established due to lower mineral content in bone tissue.

Objective. To establish the incidence of metabolic bone disease in premature infants and associated risk factors.

Materials and method. A descriptive prospective cohort study was carried out for one year in all newborns under 32 weeks or 1500 g at the Hospital Universitario de Santander, to determine the incidence of metabolic bone disease.

In the selected patients, demographic data and prenatal history were established, later a measurement of serum alkaline phosphatase and serum phosphorus was performed at the third week of birth, having reference values for diagnosis <5.6mg/dl and alkaline phosphatase >500IU/ L.

For the analysis of the information, statistical tools were applied such as average proportions, dispersion measures, distribution measures, association measures, and binomial regression.

Results. A total of 58 patients were, of which 7 had a diagnosis of metabolic bone disease, with an incidence of 12%. The weight was reported as an independent variable for the development of the disease, being significant in children under 1160g, as well as prolonged parenteral nutrition >24 days. When performing the multivariate analysis, weight and time of parenteral nutrition are described as risk factors, but also maternal age below <22 years and birth weight under 1160g are characterized by having a higher relative risk than weight <1160g.

Conclusion. The importance of early intervention in patients with metabolic bone disease-enhancing risk factors, such as weight and prolonged parenteral nutrition, is established to prevent severe complications.

Key words: Bone diseases, metabolic; infant, premature; alkaline phosphatase; phosphorus, vitamin D.

Introducción. La enfermedad metabólica ósea del prematuro es una complicación poco común, establecida por menor contenido mineral en el hueso.

Objetivo. Establecer la incidencia de la enfermedad metabólica ósea en prematuros y factores de riesgo asociados.

Materiales y métodos. Se realizó un estudio de cohorte prospectivo descriptivo durante un año a todos los recién nacidos menores de 32 semanas o <1500 g en el Hospital Universitario de Santander.

En los pacientes seleccionados se establecieron datos demográficos y antecedentes prenatales, posteriormente se realizó una medición de fosfatasa alcalina y fósforo séricos a la tercera semana de nacimiento, con valores de referencia diagnóstica < 5,6mg/dl y fosfatasa alcalina > 500UI/L.

Para el análisis de la información se aplicaron herramientas estadísticas como proporciones de promedio, medidas de dispersión, distribución, y asociación; y regresión binomial.

Resultados. Se obtuvieron un total de 58 pacientes, de los cuales 7 tuvieron diagnóstico de enfermedad metabólica ósea, con una incidencia del 12%. De las variables estudiadas, el peso se reportó como variable independiente para el desarrollo de la enfermedad, siendo significativa en menores de 1160g, igualmente la nutrición parenteral prolongada >24 días. Al realizar el análisis multivariado se describen el peso y tiempo de nutrición parenteral como factores de riesgo, además la edad materna <22 años presentó un riesgo relativo mayor, a comparación del peso <1160 g.

Conclusión. Se establece la importancia de intervención temprana de pacientes con factores de riesgo potenciadores de enfermedad metabólica ósea, como el

peso y nutrición parenteral prolongada mayor a 24 días, con el fin de prevenir complicaciones severas.

Palabras clave: enfermedades óseas metabólicas; recién nacido prematuro; fosfatasa alcalina; fósforo, vitamina D.

Metabolic Bone Disease (MBD) of preterm infants is a medical condition preceded by inadequate aggregation of the bone mineral component with biochemical down regulation deregulation of Calcium (Ca) and Phosphorus (P), which is insufficient in neonates of younger gestational age (1,2).

Although the incidence remains unknown, a 60% occurrence has been reported in preterm infants weighing less than 1000g, with fracture rates of 2% to 10% and 23% of newborns weighing less than 1500 g may be affected (3).

In fetal life, 80% of the micronutrients elements are transported to the fetus at 24 weeks. At birth, the passage of Ca is interrupted and levels drop suddenly, in neonates weighing between 500 and 2000g, Ca requirements vary from 99 to 173 mg/kg/day and those of P from 63 to 126 mg/kg/day (2,4).

To develop the disease, rapid depletion of substrates is required, especially phosphorus; which falls due to urinary loss caused by PTH. Also exclusive breastfeeding, poor supplementation results in lower P stores, while vitamin D increases intestinal absorption. Nutrient supplementation in preterm is close to 60% of Ca and 80-90% of phosphorus (5).

The clinical manifestations appear between the 3rd and 12th week of birth, but they can be asymptomatic for several weeks. The severity of MBD is directly related to the increase in Alkaline Phosphatase (AP) levels and the decrease in serum P, which are referent serum markers to establish a diagnosis. It has been described that AP levels below 500 IU/L are highly specific for determining normal bone density. The impact on Ca levels is expressed in changes in bone densitometry, being the method of choice to assess bone mineralization, since the results are

independent of anthropometry and gestational age; however, its availability is scarce (6).

With regards to the clinical presentation, it can be as serious as pathological rib fractures in up to 32% of patients, respiratory distress, myopia in the newborn, and long-term low bone mineral content during growth, which implies an increased risk of osteoporosis.

Management is based on the supplementation of calcium carbonate, vitamin D and phosphorus to meet the requirements of neonates with MBD, from doses established in international consensus as phosphorus 10-20 mg/kg/day with a maximum dose of 40-50 mg/kg/day, calcium carbonate 20 mg/kg/day with a maximum dose of 40-50 mg/kg/day and vitamin D of 400-1000 IU/day, being a major factor to reduce the incidence of MBD (7,8).

Follow-up focused on biochemical monitoring will be based on the severity of the disease and the clinical context. Mineral supplements can be gradually decreased as the markers approach their normal state to avoid adverse reactions. It is recommended to measure AP 2 to 4 weeks after discharge in exclusively breastfed very low birth weight infants with direct mineral supplements if AP >800 to 1000 IU/L. Closer follow-up is required in those patients with a moderate to severe condition or with constant risk of deficient bone mineralization since they may require prolonged fortification or direct mineral supplementation, the latter should be monitored with biochemical markers (8).

It is essential to study MBD to characterize the population and determine diagnostic cut-off points through serum markers, since MBD has an unfavorable impact in childhood, such as a reduction in bone mass, determining low height

compared to the reference population (9). For this reason, a study was carried out to establish the incidence of MBD and associated risk factors. For this reason, a study was carried out to establish the incidence of MBD and associated risk factors in our unit, including medications like caffeine and steroids.

Materials and methods

A descriptive prospective cohort study of all preterm newborns hospitalized at the Hospital Universitario de Santander and who were born with a gestational age < 32 weeks during one year.

All newborns from the University Hospital of Santander ≤32 weeks were included, and only newborns who remained hospitalized for less than 3 weeks in the institution due to discharge, transfer or death were excluded.

All newborns from the University Hospital of Santander ≤32 weeks without discrimination of underlying pathologies or neonatal evolution during hospitalization were included, and only newborns who remained hospitalized for less than 3 weeks in the institution due to discharge, transfer or death were excluded.

Demographic and perinatal data as well as clinical evolution including the presence of IntraUterine Growth Restriction (IUGR) and the use of methylxanthines were collected to describe the associated risk factors (table 1). This data was obtained in a format established and accepted by the ethics committee, after signing the informed consent by the mother or legal representative of the admitted newborn, with subsequent migration to the database created in the REDCap system, complying with all safety criteria according to current ethical standards.

The diagnosis of MBD was established with reference values of AP and P serum (table 2) through serum measurements at 3 weeks after birth or earlier if there was

clinical suspicion (tenderness on bone palpation or bone deformities on physical examination) or incidentally found fractures.

Patients diagnosed with MBD will start ionic supplementation with calcium carbonate at doses of 10 -20 mg/k/day to maximum of 40-50 mg/kg/day; and vitamin D at doses of 400-1000 IU/day, according to international consensus. Phosphorus supplementation was met in the clinical practice guideline for parenteral and enteral feeding at the University Hospital of Santander, so it was not necessary to perform supplementation.

Diagnosed patients were kept in weekly follow-up for AP and P laboratories until they reached normal values, and thus discontinued supplementation. In addition, follow-up by endocrinology was indicated at hospital discharge.

Statistical analysis

Nominal and ordinal proportions were established; such as mean and standard deviation or median and interquartile range for discrete or continuous variables, whether these are normal or not. Then, the incidence of MBD was compared between patients with a history of IUGR vs. non-IUGR, as well as between those who received or not methylxanthines, differences that will be evaluated by means of the chi-square test, accepting differences with $p<0.05$ as significant. Finally, these potential associations will be evaluated by means of binomial regression to adjust for potential confounding biases due to the incidence of infections, among others.

Ethics

This research work meets the requirements of national and international regulations is governed by national regulations according to Resolution 008430 of

October 4, 1993, under which it is considered as research with minimal risk as soon as a routine common diagnostic procedure was will be carried out through venous puncture in neonates corresponding to less than 2% of its blood volume and that it is a condition typical of the neonatal age with high scientific importance and great value in the health of the person involved, without diagnostic and screening mechanisms other than those described here. According to chapter 15, 16 of the same resolution, it will have the duly completed informed consent, which will be signed by the mother or whoever confers the responsibility of the patient, given prior information of risks and benefits.

The participants will benefit from the screening carried out worldwide for the disease, as well as its appropriate management as appropriate and the decrease in consequences in the medium and long term that it generates, in the same way the field will be opened for the routine determination of a disease until today not studied in our environment.

The project was reviewed by the ethics and research committee of the Industrial University - CEINCI and the ethics committee of the University Hospital of Santander.

Results

A sample of 78 newborns was obtained between 2020 and 2021, who were admitted to the neonatal unit of the University Hospital of Santander. Of the total, 58 patients met the inclusion criteria and 20 patients were excluded (figure 1). Exclusions were made as follow: 18 patients didn’t have informed consent, 1 deceased and 1 had a Ballard score over 32 weeks.

The demographic characteristics of the global sample are described, contemplating the maternal and neonatal population (table 3). A predominance of the female sex was found in 55.1% compared to the male sex with 44%.

The variables studied are described in table 4. Within the representative data, supplemental oxygen was used in all patients as the first step in neonatal resuscitation and subsequent INSURE procedure maneuver in 98.2%.

The main neonatal morbidity was sepsis in 56.9%, followed by bronchopulmonary dysplasia in 39.6% and necrotizing enterocolitis in 20.6%. 84.4% received caffeine citrate for their premature condition.

1. Univariate analysis of factors associated with the presence of MBD.

The incidence of MBD was 12.0%, the majority with a gestational age of less than 28 weeks with a statistically significant value, with a predominance of the female sex in 85.7% of the cases.

Within the maternal morbidities preeclampsia prevailed, followed by chorioamnionitis and gestational diabetes; however, there was no statistically significant relationship.

All patients with MBD had neonatal sepsis in common and necrotizing enterocolitis in 57.4%, both with statistical significance.

All patients diagnosed with MBD received parenteral nutrition for an average of 37 days. Of the total, 4 patients received parenteral nutrition between 15 and 30 days and 3 of them received it for more than 30 days with a statistically significant value.

No association was found with the use of caffeine citrate, diuretics, postnatal steroids, or oral nutrition. No relationship was found with the requirement of neonatal resuscitation or admission to intensive care.

1. Multivariate analysis.

When performing the multivariate analysis of the significant variables in the development of MBD (table 5); It is evident that parenteral nutrition time greater than 24 days, maternal age less than 22 years, birth weight, comorbidities such as enterocolitis and neonatal sepsis appear as association factors. The analysis indicates that parenteral nutrition >24 days as an independent variable related to the development of MBD (RR 18.85 CI 95%, 2.47-143.5 p=0.005). Birth weight less than 1160g also has an independent relationship with MBD (RR 9.58 CI 95%; 2.11-43.45 p=0.003).

Although necrotizing enterocolitis had a significant relationship, by including parenteral nutrition this relationship no longer exists.

Steroid use had no independent relationship, but by including NPT, maternal age, and birth weight less than 1160g, steroids show a protective factor with a significant p value.

Discussion

Currently, the care of preterm newborns has been improved, as well as the prevention, early detection and progression of age-related diseases. Similarly, a door has been opened to describe other diseases with less prevalence such as MBD, which is the objective of our study.

The global reported incidence of MBD is estimated to be between 16-40%, and a recent study established an incidence of 12.3%, being similar to our study with a value of 12% (n=7)(3,10).

In the population studied with MBD, statistical significance was found in relation to gestational age, birth weight and time of parenteral nutrition. This is supported by

prospective studies and meta-analysis, which describe birth weight, parenteral nutrition time and prolonged use of diuretics in relation to the development of MBD (10-12). On the other hand, Chen et al. recently examined MBD risk factors in an observational cohort study of 16 newborns diagnosed with MBD, using logistic regression analysis, showed that gestational age <30 weeks and achievement of full enteral nutrition beyond 28 days risk factors independent (13). Although there are important differences between these studies, we can establish as common risk factors, low birth weight and duration of parenteral nutrition greater than 24 days (14). In agreement with other studies, it was found that weight is an independent variable in the development of MBD; explained by the pathophysiological mechanism due to mineral deficiency in bone metabolism, increasing the risk by 9.5 times (P=0.003- CI 95% 2.11 - 43.45) in children under 1160g.

In addition, maternal age less than 22 years predisposes to preterm birth and intrauterine growth restriction, increasing the risk of developing MBD.

Studies reported in relation to the use of caffeine and MBD describe the average duration of treatment with caffeine citrate as 60 days ± 45.8 days and an accumulated dose of 425.33 ± 235.2 mg; however, in our study, the median treatment time was 30 days and the cumulative dose was 255 mg, values below the average reported in the literature, and our study was a non-significant variable. It could be inferred that the relationship between caffeine citrate and the development of MBD depends on the time of use, but it is necessary to carry out more multicenter studies with a larger population to reach this conclusion.

The use of diuretics such as furosemide has been related to the pathophysiology of MBD, explained by the renal excretion of calcium, however, in our study no

significant relationship was observed, which could be attributed to the time of use of this medication being less than 5 days, establishing a lower accumulated dose (15).

When evaluating prolonged parenteral nutrition >24 days, the impact on delaying the start of fortified enteral nutrition was identified, being of vital importance to prevent the development of MBD. For this reason, it is essential to stabilize the other comorbidities associated with prematurity, seeking a balance between the start of early enteral nutrition and the progressive withdrawal of parenteral nutrition, allowing an adequate supply of P and Ca (16).

Although a significant individual relationship with necrotizing enterocolitis was found, when analyzed with parenteral nutrition, this relationship disappeared. This is explained by the limitation of early initiation of enteral feeding.

Neonatal sepsis is described as a risk factor for MBD, however, it has been described that there are no significant changes that correspond to a biochemistry of reduced bone turnover (17).

The results obtained show the risk factors associated with the development of MBD, with low birth weight and parenteral nutrition having the greatest impact. Therefore, it is important to detect this disease in its early stages to offer timely treatment and avoid late complications. Despite not having a consensus for the diagnosis, the use of serum biomarkers is described according to the pathophysiology; being useful for the screening and diagnosis of the disease. Weight and prolonged parenteral nutrition are the most important risk factors to intervene in this population, being able to set an alarm to screen for this disease in

neonatal care centers, in addition to implementing early nutritional interventions in diagnosed patients, and thus prevent severe complications.

The limitations of our study are the size of the population group, the non-measurement of PTH, the absence of interpretation of diagnostic images in MBD; however, an adequate correlation has been found between P values less than 5.6 mg/dl and AP values greater than 500 IU/L used as cut-off points in the study.

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Conflicts of interest

The authors declare that they have no conflict of interest regarding the current study.

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Table 1. Study variables

Prenatal history	Immediate neonatal	Neonatal evolution
a) Preeclampsia b) Intrauterine Growth restriction (IGR) c) Corioamnionitis d) Gestational diabetes or pre-diabetes e)Thyroid disease f) use of prenatal corticosteroids (2 or more doses) g) Unique or multiple pregnancy	- Sex - Weight - Mass body index - Gestational age (obstetric, Last menstrual period, physical exam) - Basic or advanced life support - Oxygen requirement - continuous positive airway pressure - compressions - use of vasopressors	a) Airway support b) Haemodynamic support c) Use of: - Diuretics - methylxanthines d) Use of parenteral nutrition over 4 weeks e) Presence of: -Bronchopulmonary dysplasia -Sepsis -Necrotizing enterocolitis

Table 2. Reference values for phosphorus and alkaline phosphatase

	Reference	Metabolic Bone Disease
phosphorus (P)	> 1.81 mmol / L o 5.6 mg/dL to 2.91 mmol/L or 9.01 mg/dL	< 1.81 mmol / L or 5.6 mg/dL
Alkaline phosphatase (AP)	< 500 UI / L	> 500 UI / L

Table 3. Neonatal and maternal demographic characteristics

NEONATAL AND MATERNAL DEMOGRAPHIC CHARACTERISTICS	
VARIABLES	**TOTAL**
Sex - n[%] Male Female	 26 [44,8] 32 [55,1]
Median of gestational age - n [IQ]	30 [28-31]
Way of delivery - n [%] cesarean section Vaginal delivery	 46 [79,3] 12
Birth weight [g]	1334.7 [600-2005]
Average birth weight - g [IQ]	1350 [1185-1540]
Use of prenatal steroids - n [%]	40 [71,4]
Neonatal life support - n (%) Oxygen INSURE maneuver CPAP thoracic compressions Use of vasopressors	 58(100) 57(98,2) 56(96,5) 7(12,0) 7(12,0)
Neonatal Intensive Care Unit requirement	39
Morbidities- n [%] Bronchopulmonary dysplasia Necrotizing enterocolitis Neonatal sepsis	 23 [39,6] 12 [20,6] 33 [56,9]
Pharmacological exposure - n [%] Caffeine citrate Furosemide Corticosteroids Parenteral nutrition	 49 (84,4) 12 (20,7) 21 (36,2) 51 (87,9)
Metabolic Bone Disease diagnosis- n(%) Yes No	 7(12,0) 51(88,0)
Maternal characteristics Median of maternal age - n [IQ]	 26 [22-31]

Comorbidities- n [%]	
Preeclampsia	21 [37,5]
Chorioamnionitis	18 [32,1]
Intrauterine growth restriction	4 [7,1]
hypothyroidis	0[0,0]

INSURE= Intubation-Surfactant-Extubation
CPAP= Continuous positive airway pressure
NICU = Neonatal intensive care unit
MBD= Metabolic Bone Disease
IQ= Interquartile range

Table 4. Factors associated with MBD

	MBD n=7	Without MBD n=51	Pr
Maternal age < 21 years >22 years	 4 3	 7 44	 0.006
Maternal comorbidities Preeclampsia Gestational diabetes Chorioamnionitis	 3 (42.86%) 1(20% 2(28,5%	 18(35,2%) 5(13,1%) 16(31,3%)	 0,696 0,678 0,881
Prenatal corticosteroids	4(66,6%)	37(78,7%)	0,506
Gestational age <28 weeks >28 weeks	 3 4	 9 49	 0,010
Sex Female Male	 6(85,7%) 1(14,3%)	 32 26	 0,080
Average birth weight (g) <1000 1000-1500 >1500	980(674 – 1455) 4 3 0	1375(600-2005) 7 28 16	 0,014
APGAR score less 3 points	3(20,6%)	9(17,65&)	0,123
Neonatal reanimation Oxygen INSURE CPAP Thoracic compressions Mechanical ventilation NICU	 7(100%) 7(100%) 7(100%) 2(28,5%) 2(28,5) 4(57,1%)	 (100%) 50(98%) 49(96%) 5(9,8% 5(9,8%) 35(68,6%)	 - 0,789 0,594 0,153 0,153 0,544
Neonatal comorbidities			
Bronchopulmonary dysplasia	4(57.4%)	19(37,2%)	0,313

Necrotizing enterocolitis	4(57.4%)	8(15,6%)	0,011
Neonatal Sepsis	7(100%)	26(50,9%	0,014
Pharmacological intervention			
Caffeine citrate	7(100%)	42(82,3%)	0,227
Days of use <28 days >28 days	 0 7	 3 39	0,275
Furosemide	2(28,5%)	11(21,5%)	0,677
Days of use < 5 days 5-10 days >10 days	 1 0 1	 8 3 0	0,338
Postnatal corticosteroids	4(57,1%)	17(33,3%)	0,219
Days of use(d) <7 days 7-14 days >15 days	 3 0 1	 15 0 2	0,195
Nutrition			
Parenteral nutrition	7(100%)	44(86,2%)	0,296
Days of use (median) (IQ) <7 days 7-14 days 15-30 days >30 days	37 (24-62) 0 0 4 3	13 (0-60) 9 18 14 3	0,002
Exclusive breastfeeding	0	51(100%)	-
Mixed breastfeeding	7 (100%)	47 (92,1%)	0,443
Fortified milk	2 (28,5%)	6 (11,7%)	0,227

Table 5. Variables representative associated with MBD

Variable	Relative Risk	Standard error	Pr	IC 95%
Parenteral nutrition >24 days	7,95	8,25	0,04	1.04 -60.72
Maternal age <22 years	4,18	2,15	0,01	1.52 – 11.48
Corticosteroids	0,92	0,00	0,00	0.91 -0.91
Birth Weight <1160 gr	3,19	0,01	0,08	0.85 – 11.91

Figure 1. Inclusion and exclusion criteria