The Effect of Maternal Vitamin D Intake on the Incidence of Nonspecific Respiratory Distress in Infants: A Randomized Clinical Trial

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Research Article

Keywords: Non-specific respiratory distress syndrome (NRDS), vitamin D, infant, vitamin D deficiency

Posted Date: January 10th, 2022

DOI: <https://doi.org/10.21203/rs.3.rs-1101288/v1>

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Abstract

Background

vitamin D deficiency is associated with respiratory problems in neonates. The late preterm or near-term neonates who have been admitted for tachypnea and fully recovered before 12 h, we called Non-specific respiratory distress syndrome (NRDS). The present study aimed to evaluate the effect of 25(OH) D administration in pregnant women at risk of preterm delivery on the incidence of NRDS in their infants.

Methods

This single-blind clinical trial was performed on mothers and neonates with a gestational age of 32-37 weeks who were referred with labor pains from February 20th 2021 to June 29th 2021 in the obstetrics and gynecology department and intensive treatment unit of Ghaem Hospital, Mashhad University of Medical Sciences, Iran. Within 72 h of preterm delivery, a single dose of 50,000 units of intramuscular 25-hydroxy vitamin D was injected into pregnant women in the intervention group. Also a sample containing 1.5 ml of whole blood was taken from the umbilical cord of the infant and mother to assess the level of vitamin D.

Results

In the present study, there was a significant relationship between the two groups of control and intervention in terms of weight ($P=0.001$), first ($P=0.027$) and fifth minute Apgar score ($P=0.001$) in infant, incidence of NRDS ($P=0.001$) and maternal age ($P=0.004$). The results showed no statistically significant difference between the two groups in terms of gender ($p = 0.673$), type of delivery ($p = 0.299$), level of vitamin D of the mothers ($P=0.053$) and infants ($P=0.805$).

Conclusions

The single injection of vitamin D into the mother prone to preterm birth over 31 weeks of gestation reduces transient respiratory problems in these infants.

Trial registration:

IRCT20110807007244N7 (19/02/2021)

Background

In my 20 years of neonatal practice, I have seen many cases of late preterm or near-term neonates who have been admitted for tachypnea and fully recovered in a few hours. Initial tests, blood gas, radiography, and blood culture of these infants were normal. Although most of these neonates have a good prognosis, separation from the mother in the first few hours imposes much stress on the mother and deprives the mother and the newborn of the indescribable benefits of mother-infant contact in the first hours, wasting much time of medical and nursing staff. This condition is common in elective cesarean section and non-academic centers. We have used non-specific respiratory distress (NRDS) to describe this condition. Undoubtedly, the most crucial cause of this problem is the disorder in the passage of intra-uterine to extra-uterine life in the respiratory system. Probably, the most important predisposing factors for NRDS are elective cesarean section and premature births.

During pregnancy, the fetal lung secretes fluid into the alveolar space through the Chloride-driven liquid secretion mechanism. Na-K-2Cl channel inhibitors block this secretion. This fluid plays an important role in developing the lungs, providing a framework for preventing airway collapse and promoting its growth. The fetus faces a challenge at birth because empty its airways quickly of fluid (which is secreted throughout pregnancy) with an initial warning (labor) or even without warning (elective cesarean section). A significant component of this process is the lung epithelium, which switches placental to pulmonary gas exchange. Effective gas exchange is based on clearing the airways of fluid and increasing pulmonary blood circulation to match ventilation and perfusion (1). Any disturbance in the above process can lead to respiratory problems for the infant. If this respiratory problem lasts for more than 12 h and improves within 24 to 48 h, transient tachypnea of the newborn (TTN) is suggested (2). However, it often improves before 12 h, which we called Non-specific respiratory distress syndrome (NRDS). One way to prevent this problem is to allow normal labor. Nevertheless, despite knowing the countless benefits of normal labor, elective cesarean section is still very high without a valid reason. Therefore, before birth, the fetal lung must be prepared for extra-uterine life.

Some studies have suggested the role of vitamin D in type-II pneumocyte cell maturation, fibroblast proliferation, surfactant synthesis, and Alveolarization and Vitamin D deficiency may be a risk factor for respiratory problems (3, 4). Prematurity increases the risk of vitamin D deficiency. In this respect, the prevalence of vitamin D deficiency in premature infants has varied in different parts of the world. For instance, vitamin D deficiency has been reported in 64%, 83%, and 82% of preterm infants in the United States, India, and Iran, respectively (5). Numerous studies have been performed on the relationship between vitamin D deficiency and respiratory diseases such as bronchopulmonary dysplasia (BPD), TTN, and respiratory distress syndrome (RDS), and the deficiency of this vitamin has been considered among the factors influential in the occurrence of these diseases in infants (4, 6–9). We hypothesize that a deficiency of this vitamin may also be effective in NRDS. Therefore, the present research aimed to assess the relationship between non-specific respiratory distress at birth and maternal vitamin D intake.

Method

Study design: This single-blind clinical trial was performed on mothers and neonates with a gestational age of 32-37 weeks who were referred with labor pains from February 20th 2021 to June 29th 2021 in the obstetrics and gynecology department and intensive treatment unit of Ghaem Hospital, Mashhad University of Medical Sciences, Iran. Once the Ethics Committee of Mashhad University of Medical Sciences (IR.MUMS.MEDICAL.REC.1399.613 and number 990699) approved the study, the researcher received informed consent from the participating parents. The study was registered in the Iranian Clinical Trial Registration Center (IRCT20110807007244N7) before starting the project.

Inclusion and exclusion criteria: Inclusion criteria included pregnant women with a gestational age of 32-37 weeks, having a risk of preterm delivery, normal fetal prenatal ultrasound, lack of significant medical and obstetric problems for the mother (such as cardiovascular problems, diabetes mellitus, epilepsy, and asthma).

Exclusion criteria, on the other hand, included meconium aspiration syndrome, congenital anomaly, congenital infection, RDS, TTN, intubation in delivery room, death in the delivery room, severe asphyxia, addicted mothers, pneumonia, and persistent respiratory distress after the first 12 h of life.(Figure 1)

Randomization: In this study, blocking is used as a reliable randomization method as it leads to a balanced allocation of individuals in treatment groups at the end of each block. For this purpose, a web-based online method is used to determine the volume and number of random blocks. This online software can create random blocks with desired variable sizes. Twenty blocks with variable sizes of 6, 8, and 10 were introduced for a total of 160 samples using this website. The blocks were marked with numbers 1 to 20. Each block includes an equal number of intervention and control groups according to the size of that block. Following the reference of the first mother, one of the blocks was randomly selected. Then, it was assigned according to the order of the contents of the block in one of the groups. By referring to the next mother, the next item of the same block was selected. The remaining blocks were processed, and these steps were repeated until sampling was completed. This process continued until the completion of the first block. The next block was selected randomly from the remaining blocks, and the above process was repeated until sampling was completed. The neonates were followed by the principal researcher, who did not know the relevant, control, or intervention group regarding the occurrence or non-occurrence of respiratory distress and the time of recovery and diagnosis of respiratory distress. Data were classified as A and B in the SPSS software, and the statistical analyst interpreted the data without knowing the type of grouping.

Intervention: Within 72 h of preterm delivery, a single dose of 50,000 units of intramuscular 25-hydroxy vitamin D was injected into pregnant women in the intervention group who had not taken a 25-hydroxy vitamin D in the last three months.

Laboratory evaluation: A sample containing 1.5 ml of whole blood was taken from the umbilical cord of the infant and mother to assess the level of vitamin D. The prepared samples were centrifuged, and its serum was kept at -20°C and sent to the central laboratory of Ghaem Hospital for evaluation. The level of vitamin D was measured in ng/mL by ELISA Reader device model RT2100c (Germany) and ELISA

Washing device using the ELISA method. The 25-hydroxy vitamin D lower and higher than 30 ng/mL was considered 25-hydroxy vitamin D deficiency and sufficient values. Deficiency values were divided into three groups of severe deficiency (≤ 10 ng/mL), moderate deficiency (10.1-20 ng/mL), and mild deficiency (20.1-30 ng/mL).

Radiological evaluation: Infants having more than 2 h of respiratory distress and being admitted to the neonatal ward underwent a chest radiographic examination. Next, the cause of neonatal respiratory distress was determined based on radiographic findings and clinical course.

Outcomes: The primary outcome of the neonates in the study group included the incidence of non-specific respiratory distress. The diagnosis criteria included respiratory distress (tachypnea, intercostal muscle retraction, grunting, and nasal flaring) more than 2 h and less than 12 h, normal chest radiograph, and negative blood culture.

Data collection: Complete characteristics of infants (birth weight, age, sex, gestational age, Apgar score, and clinical signs), maternal history (age, pregnancy, and delivery problems, and delivery method), and laboratory results were collected and recorded in a questionnaire. The need for resuscitation, duration of respiratory distress, duration of oxygen therapy, duration of hospitalization in the NICU, and mortality were also recorded in the checklist. The levels of calcium and vitamin D of the infant were also recorded.

Patients and Methods

A single-blinded randomized controlled clinical trial was conducted on pregnant mothers with a gestational age (GA) of 32-37 weeks at risk of preterm labor. The mothers were randomly assigned into two groups, including control group (with no injection) and intervention. A single dose of 50,000 units of intramuscular 25-hydroxy vitamin D was injected into pregnant women in the intervention group within 72 h of preterm delivery. A sample containing 1.5 ml of whole blood was taken from the mothers and infants' umbilical cord to assess the level of vitamin D. Short-term outcomes, including neonatal respiratory findings, and laboratory results of mothers and infants were recorded in checklist.

This was a clinical trial on 169 infants with gestational age of 32-37 weeks at birth. Neonates with more than 2 h and less than 12 h respiratory distress, normal chest radiograph, and negative blood culture met the criteria for the study. The infants were placed under oxyhood with closed monitoring for worsening respiratory symptoms. Umbilical catheter was inserted for infants and ABG according to the patient status was obtained. All infants were monitored continuously; pulse oximetry was performed by a probe at right upper extremity, and respiratory symptoms were examined repeatedly. Blood gas analysis and oxygen saturation were gold standard to detect infants at risk of respiratory failure.

Data analysis: Statistical analysis was performed by the SPSS software (IBM SPSS Statistics, Version 23). The mean of quantitative variables between groups was compared using an independent t-test or Mann-Whitney U test. Chi-square test and Fisher's exact test were used to compare qualitative variables

between the studied groups. Data were expressed as mean $\pm$ standard deviation (SD). $P < 0.05$ was considered as the statistically significant level of the data.

Results

In this study, of 169 mothers, 14 ones were excluded from the study, and 154 mothers were randomly divided into intervention and control groups. Of 77 infants, 40 infants in the control group and 31 infants in the intervention group were excluded from the study for the reasons mentioned in Fig. 1. Eventually, 37 infants in the control group and 46 infants in the intervention group were included in the study.

In the control group, 51.4% were boys and 48.6% girls. Also, in the intervention group, 53.3% were boys and 46.7% were girls. The results showed no statistically significant difference between the two groups in terms of gender ($p = 0.673$) and type of delivery (cesarean section and normal vaginal delivery) ($p = 0.299$). However, in both groups, the rate of cesarean section was higher.

This study showed that the mean weight of neonates in the control and intervention groups was 2017.64 ± 651.11 and 2624.56 ± 639.70 gr, respectively, and the mean gestational age was 34.81 ± 1.88 and 35.60 ± 1.87 weeks, respectively.

The mean level of vitamin D of the mother in both control and intervention groups was less than 30 and was 18.81 ± 11.40 and 23.68 ± 9.78 , respectively, but there was no significant difference between the two groups.

In the present study, there was a significant relationship between the two groups of control and intervention in terms of weight, first and fifth minute Apgar score in infant and maternal age. ($p < 0.05$) (Table 1).

Table 1
Comparison of neonatal and maternal characteristics between the control group and the group of mothers receiving vitamin D

variable	Control Mean ± SD	Intervention Mean ± SD	P value
Gestational age	34.81±1.88	35.60±1.87	0.058
weight	2017.64±651.11	2624.56±639.70	0.000*
maternal age	29.56±4.55	33.02±6.19	0.004*
parity	1.81±0.93	2.24±1.33	0.089
Apgar 1minute	7.48±1.87	8.32±1.63	0.027*
Apgar 5minute	8.98±0.92	9.64±0.81	0.001*
Mother vitamin D	18.81±11.40	23.68±9.78	0.053
Neonatal vitamin D	20.10±13.58	20.95±14.24	0.805
*t-test (p<0.05)			

In the present study, no significant relationship was observed between groups of neonates without respiratory distress and neonates with non-specific respiratory distress in terms of gestational age, weight, parity, maternal age, and serum levels of vitamin D in neonates and mothers ($P > 0.05$). The results showed a significant difference between the two groups of mothers receiving vitamin D and the control group in terms of non-specific respiratory distress and improvement within less than 12 h in the infant. The results also indicated that maternal intake of vitamin D significantly reduced non-specific respiratory distress in the infant. (Table 2)

Table 2
Comparison of neonates in terms of non-specific respiratory distress between the control group and the group of mothers receiving vitamin D

Vitamin D use mother	No respiratory distress	Nonspecific respiratory distress	P value
control	21(56.8%)	16(43.2%)	0.001*
intervention	41(89.1%)	5(10.9%)	
*chi- square(p<0.05)			

Discussion

Our results showed that injecting 50,000 units of vitamin D to a mother at risk of preterm delivery reduces the transient respiratory problems of their infants fourfold. This result indicates the significant importance and function of vitamin D in accelerating the maturation of the lungs of infants. Also, it

seems that vitamin D during the delivery process causes the correct development of the cardiorespiratory system and the successful transition from fetal to neonatal life. In the present study, for the first time, we investigated the effect of vitamin D on non-specific neonatal respiratory problems. We hope this study is a prelude to efforts to reduce the respiratory problems of preterm infants. To the best of our knowledge, there is no study similar to ours in the literature. However, several animal articles have suggested the role of vitamin D in fetal lung maturity (1, 7). A systematic review conducted in 2015 based on animal and laboratory data reported that vitamin D stimulates fetal lung maturation. These data support the hypothesis of vitamin D deficiency as a risk factor, as well as modifiable RDS and BPD (3). The present study results are consistent with those of this article in terms of improved lung function and faster adaptation to the extra-uterine environment. Vitamin D may help transform fetal lungs into neonates by protecting the integrity of pulmonary epithelial barrier in the premature or near-term infant. Lack of vitamin D receptors in the pulmonary epithelial barrier leads to acute lung injury caused by lipopolysaccharides. Vitamin D treatment reduces lipopolysaccharide-induced lung damage and maintains alveolar barrier function. Thus, vitamin D treatment has been suggested as a potential treatment strategy in acute lung injury and RDS (10).

Hansdottir et al. reported that the effects of vitamin D are very broad, including increased secretion of cathelicidin antimicrobial peptide, decreased chemokine production, inhibition of dendritic cell activation, and altered T-cell activation (11). All these cases effectively reduce inflammation and lung damage and are more important in preterm and near-term infants who are more prone to inflammatory processes and lung damage. The results of our study also show the effect of vitamin D in reducing lung damage and inflammatory processes. Numerous studies have suggested vitamin deficiency as a risk factor for respiratory problems in preterm infants, including RDSs and TTNs (4, 12).

Successful transition from fetal to neonatal life at delivery requires a series of extensive physiological changes in the cardiorespiratory system, leading to a change in the gas exchange pathway from the placenta to the lungs. Replacement of intra-alveolar fluid with air, initiation of regular respiration, and increased pulmonary blood circulation are the most critical respiratory changes at birth. In this respect, normal and term labor are two fundamental factors that facilitate this important process. Numerous mechanisms are involved in alveolar fluid clearance, including the delivery process, initial respiration, and thoracic pressure. Secretion of chloride during pregnancy is vital in lung growth and development. In late pregnancy, the lung epithelium changes from the active secretion of chloride and fluid into the airways to the active absorption of sodium and fluid in response to increased concentrations of Catecholamines and other hormones. Increased oxygen pressure at birth increases sodium transfer and expression of the epithelial sodium channel gene and causes a high absorption of alveolar fluid (1, 13).

Early effective breaths cause high transpulmonary pressures. The mean esophageal pressure is estimated to be -52 cmH₂O during inhalation and 71 cmH₂O during exhalation in the term infants. Increased pulmonary blood circulation occurs due to increased systemic vascular resistance and decreased pulmonary vascular resistance (13). In one study, 49, 78, and 100% of infants over 35 weeks had complete airway clearance at 2, 4, and 24 h postpartum, respectively (14). Prematurity and Vitamin D

deficiency may be two predisposing factors that delay this process, leading to respiratory distress in the early hours of life in preterm and near-term infants, although it lasts less than 12 h and all laboratory and radiological evaluations are normal. This phenomenon, which occurs probably due to the prolongation of the transition phase from the fetal stage to the neonatal, was called “non-specific respiratory distress.” Expression of Vitamin D receptors in the lungs in late pregnancy in animals highlights the role of vitamin D in alveolar maturation (3). Also, vitamin D may be involved in postpartum lung growth and Alveolarization, but the evidence is limited in this regard. Premature infants have lower levels of 25-hydroxy vitamin D at birth and are at greater risk than term infants for postpartum vitamin-D deficiency. This problem, which is due to intestinal failure and comorbidities, theoretically puts them at risk for acute and chronic lung disease (15).

According to our results, vitamin D injection into the mother improved the first and fifth accurate Apgar scores in infants. Vitamin D affects the airways through several mechanisms, including increased resistance and airway smooth muscle mass, pulmonary parenchyma density, and alveolar septa (16). Its effects on surfactant production have also been supported in human studies (17). All of these can be considered effective causes in improving the neonatal Apgar score. A significant correlation was observed in another study between vitamin D levels and the first and fifth minute Apgar scores (4). In a study in Pakistan, maternal vitamin D intake improved the first and fifth minutes Apgar score in the neonates (18). However, Amegah et al. reported that the relationship between vitamin D and Apgar score is contradictory (19).

Nevertheless, attributing the incidence of these diseases in the infant to vitamin D deficiency strongly needs more research on the role of vitamin D in the incidence of neonatal respiratory diseases. Also, more research should be done on the pathophysiology and cause of NRDS such that to consider it a transient respiratory disorder in the infant and thus prevent its occurrence.

The most significant limitation of the present study was the lack of measurement of vitamin D before injection of vitamin D into the mother. The second important limitation is not controlling the injection distance until delivery, which can explain the lack of differences in serum vitamin D levels in infants of the two groups.

Conclusion

The present study results showed that the single injection of vitamin D into the mother prone to preterm birth over 31 weeks of gestation facilitates intrauterine breathing and reduces transient respiratory problems in these infants. Therefore, proper vitamin D supplementation during pregnancy may reduce the respiratory problems of these infants, although more studies are needed in this field.

Declarations

Ethics approval and consent to participate: This study was approved by the Ethics Committee of the Mashhad University of Medical Science (Ethics code: IR.MUMS.MEDICAL.REC.1399.613). The researcher received informed consent from the participating parents. The study was registered in the Iranian Clinical Trial Registration Center (IRCT20110807007244N7) before starting the project.

Consent for publication: not applicable.

Availability of data and materials: All data generated or analysed during this study are included in this published article.

Competing interests: The authors declare that they have no competing interests.

Funding: his work was supported by Mashhad University of Medical Sciences (grant number of 990699).

Authors' contributions: HB and NP participated in the design of the study. Statistical analyses were conducted by AD and MRS. All authors contributed to interpretation of the results. NP and MA contributed to the drafting of the manuscript. All authors contributed to the critical revision of the manuscript for important intellectual content, approved the final version and are accountable for the integrity of its content.

Acknowledgements: This research was supported by the Vice Chancellor for Research of Mashhad University of Medical Sciences under grant number 990699.

Statement :All methods were performed in accordance with the relevant guidelines and regulations

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Figures

Figure 1

CONSORT 2010 Flow Diagram