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REVIEW ARTICLE

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Is chlorine and sodium levels in the amniotic fluid a new marker for fetal lung maturation and RDS severity?

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ABSTRACT

Background: Amniotic fluid (AF) is a dynamic liquid whose contents vary according to the needs of the fetus. Levels of the amniotic components have been used in numerous studies as potential biomarkers to screen pregnancy-related abnormalities. As a reflection of Na^+ and Cl^- levels of fetal lung fluid, amniotic fluid's Na^+ and Cl^- levels can be used as an indicator of lung maturation in the newborn period. This study aimed to investigate whether Na^+ and Cl^- levels in the amniotic fluid would be a new marker to determine the severity of respiratory distress and pulmonary maturation in the newborn.

Methods: This prospective cohort study was conducted at Hacettepe University Neonatal Intensive Care Unit. One hundred twenty single infants who were delivered with the cesarean section between January 2015 and March 2016 were included. Na^+ and Cl^- levels were measured from AF.

Results: There were 46 of 120 infants (33.3%) in Group-1 and 74 infants (66.7%) in Group-2. Na + and Cl^- levels of the AF of Group-1 were higher than Group 2 and this was statistically significant ($p < .001$/p: .01, respectively). Na^+ and Cl^- levels of the AF were significantly higher in infants who needed surfactant ($p < .001$/p: .001, respectively).

Conclusion: Our results showed that Na^+ and Cl^- levels of the AF can be used as an indicator of infant lung maturation.

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KEYWORDS
Amnion fluid; lung maturation; newborn; respiratory distress syndrome

Introduction

Amniotic fluid is a vital liquid that is essential for supporting the fetus and protecting it from the external environment. During pregnancy, dynamic regulatory processes maintain the volume and chemical composition of the AF within certain limits based on fetal requirements [1–3].

AF consists of 98% water and 2% electrolytes, proteins/peptides, carbohydrates, and fetal epithelial cells and hormones. Levels of these components have been studied in numerous studies as potential biomarkers for screening pregnancy-related abnormalities [4–6]. For example, since AF interacts directly with fetal airways, parameters related to biologic substances produced or secreted by the fetal lungs, such as lecithin/sphingomyelin ratio, phosphatidylglycerol, and lamellar body count, are used as indicators of fetal lung maturity [7].

Early in pregnancy, AF is essentially a dialysate of maternal serum. In the later stages of pregnancy, the composition of AF changes due to the presence of fetal urine, fetal lung fluid (FLF) and gastric secretions, and fluids filtered through the umbilical cord and placenta [7]. FLF, which has a key role in fetal lung development, is secreted at volumes 100 times greater than necessary (250–300 ml/24 h). Excess FLF exits the lungs through the trachea during fetal respiratory movements. About 50% of the lung secretions are swallowed, and 50% mixes with AF. This FLF constitutes one-third of the AF [8]. While FLF is secreted into the alveolar space throughout gestation, its reabsorption begins at the onset of labor [9,10]. Changes in the synthesis of FLF, which is hypertonic relative to AF, also cause changes in AF composition (Table 1) [8,11]. However, the relationship between changes in Na^+ and Cl^- levels in the AF and lung maturation is

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Table 1. Demographic features and amniotic fluid Na^+ and Cl^- levels of the all groups.

	Group-1 (*n*: 46)	Group-2 (*n*: 74)	*p* Value*
Gestational age (week)	30.4 (28.2–33.0)	38.1 (37.1–38.4)	<.001*
Birth weight (g)	1300 (840–1870)	3140 (2770–3400)	<.001*
Male (%)	23 (50)	42 (56.8)	.47
Amniotic fluid Na^+ (meq/dl)	128.5 (125.4–132.5)	125.1 (119.3–127.5)	<.001*
Amniotic fluid Cl^- (meq/dl)	109.1 (107.3–112.3)	106.7 (105.0–110.8)	.01*
Amniotic fluid pH	7.3 (7.2–7.6)	7.5 (7.2–7.5)	.641

Median (IQR), *Mann Whitney *U* test (test $p < .05$ significant).

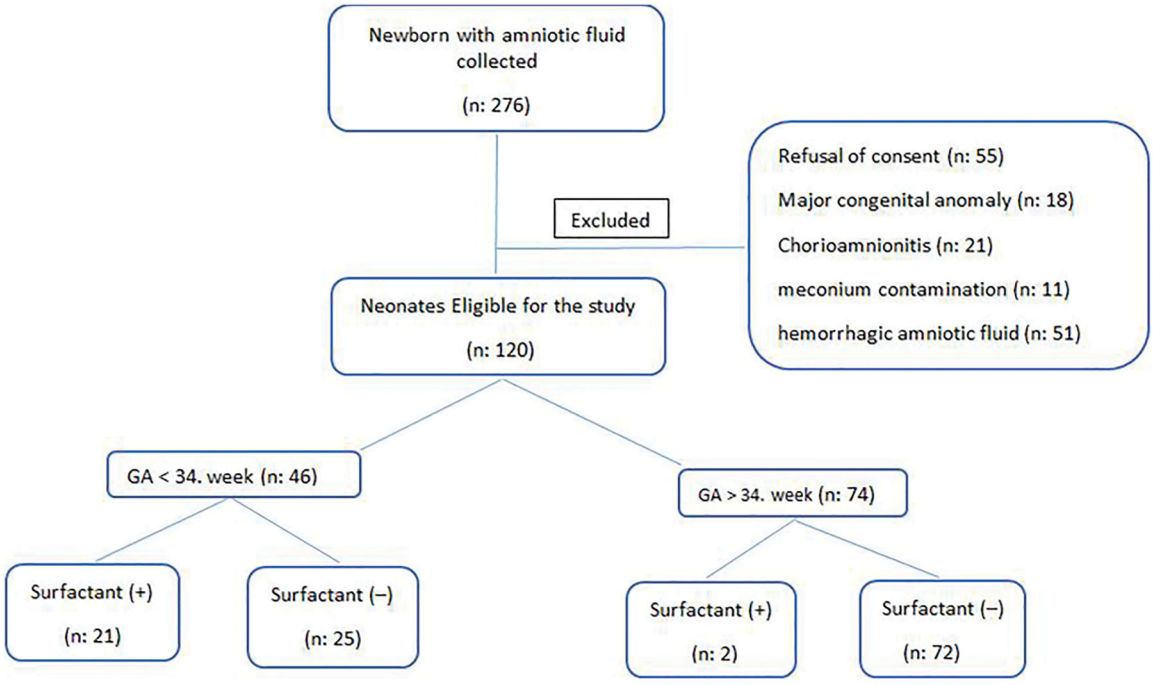


Figure 1. Flow chart.

unknown. We believe that changes in AF Na^+ and Cl^- levels may be used as indicators of lung maturity.

In this study, we aimed to investigate the utility of AF Na^+ and Cl^- levels as new markers in determining fetal lung maturity.

Materials and methods

This prospective cohort was conducted at Hacettepe University Neonatal Intensive Care Unit. The research procedure was approved by the local ethical committee (IRB Number: GO 15/292). Single infants who were born between September 2015 and March 2016 and whose mothers were admitted to the hospital with birth actions and were decided to deliver by cesarean section were included. Approximately 10 ml of sterile AF was collected during a cesarean. Sodium and Cl^- levels in the AF were measured by biochemistry analyzer (Beckman Coulter, Atlanta/US) and pH levels in the AF were analyzed with Pen Type pH Meter- pH-009 (I) (Technology Place – BCIT Campus 4475 Wayburne/British Columbia). Infants with major congenital malformations and chromosomal anomalies were excluded from the study. Contaminated AF with meconium and/or blood and suspected of chorioamnionitis were excluded (Figure 1).

One hundred twenty infants were included in the study. Infants were divided into two groups: Group-1: <34 gestational weeks and younger, Group-2: 37 gestational weeks and elder.

All infants were evaluated by a neonatologist after birth. The evaluating neonatologist was unaware of the amniotic fluid result. Infants were administered surfactant with respiratory distress syndrome (RDS) according to European Consensus Guidelines [12]. The demographic data of all infants were obtained from

patient files. Gestational weeks of all infants were determined according to their mothers' last menstrual date. The demographic information of their mothers were obtained from the hospital patient's files.

Statistical analysis

Data were analyzed using the Statistical Package for Social Sciences 21.0 for Windows (SPSS Inc., Chicago, IL). The frequencies of categorical variables were expressed as percentages. The variables were investigated using visual (histogram, probability plots) and analytical methods (Kolmogorov-Siminov/Shapiro-Wilk's test) to determine whether or not they are normally distributed. Descriptive analyses were presented using medians and interquartile range (IQR) for the non-normally disturbed and ordinally variables. The Mann-Whitney *U* test was used to compare Na^+ and Cl^- levels of the AF between the groups. For the multivariate analysis, the possible factors identified with univariate analyses were further entered into the logistic regression to determine independent predictors of infants' outcomes. Predicting the presence of lung maturity of Na^+ and Cl^- levels of the AF were analyzed using ROC (Receiver Operating Characteristics) curve analysis. When a significant cut-off value was observed, the sensitivity, specificity, positive and negative predictive values were presented. $p < .05$ was considered statistically significant.

Result

One hundred twenty infants were included in this study. Forty six of 120 infants (33.3%) were Group-1, 74 of 120 infants (66.7%) were Group-2. The demographic characteristics of the groups and Na^+ and Cl^- levels of the AF were summarized in Table 1. Na + and Cl − levels of the AF of the infants in Group-1 were higher than Group 2 and this was statistically significant ($p < .001$/*p*: .01, respectively).

We found a moderate, negative correlation between Na^+ levels of the AF and gestational week (*r*: −0.440, *p*: .001). On the other hand, there was a low negative correlation between Cl^- levels of the AF and gestational week (*r*: −0.243, *p*: .007).

When subgroup analysis was performed, Na^+ levels of the AF of infants between 24th and 28th gestation weeks were significantly higher than the levels of infants with 28–34th gestation weeks ($p < .001$), but there was no difference between the Cl^- levels in the AF according to gestation weeks (Table 2).

Table 2. Subgroup analysis of Amniotic Na^+ and Cl^- levels in under 34 weeks of infants.

	24–28 (*n*: 19)	GH:28–34 (*n*: 27)	*p* Value*
Amniotic fluid Na^+ (meq/dl)	132.1 (128.5.1–132.7)	127.0 (124.9–130.2)	<.001*
Amniotic fluid Cl^- (meq/dl)	111.1 (107.2–112.5)	108.3 (106.5–111.6)	<.24
Amniotic fluid pH	7.3 (7.2–7.5)	7.5 (7.3–7.6)	.730

GA: gestation age, Median (IQR). *Mann Whitney *U* test (test $p < .05$ significant).

Table 3. Subgroup analysis of Amniotic Na^+ and Cl^- levels in under 34 weeks of infants who use and do not use surfactantn.

	Surfactant (−) (*n*: 21)	Surfactant (+) (*n*: 25)	*p* Value*
Amniotic fluid Na^+ (meq/dl)	126.6 (124.8–127.2)	132.3 (130.1–132.7)	<.001*
Amniotic fluid Cl^- (meq/dl)	107.4 (105.7–108.7)	111.5 (107.8–113.0)	<.001*
Amniotic fluid pH	7.5 (7.4–7.6)	7.4 (7.5–7.5)	.763

Median (IQR), *Mann Whitney *U* test (test $p < .05$ significant).

Sodium and Cl^- levels of the AF of infants between 28th and 34th gestation weeks were compared according to their surfactant need. Na^+ and Cl^- levels of the AF were significantly higher in infants who needed surfactant ($p < .001$/*p*: .001, respectively) (Table 3).

Figure 2 shows the probability chart for surfactant requirement under the 34th gestation weeks according to AF Na^+ and Cl^- levels. We divided surfaktant requirement probability into three goups of low ($125 < AF\ Na^+ > 127.5$; $101 < ACl^- > 107$), moderate ($127.5 < AF\ Na^+ > 130$; $107 < ACl^- > 111$) and high ($130 < AF\ Na^+$; $111 < ACl^-$) probability. If aNa^+ levels are 125 meq/dl, Cl^- levels are 101 meq/dl and lower, it can be said that lung maturation is completed.Sodium levels of the AF an area under the curve of 0.91 (95% confidence interval: 0.84–0.97, $p < .001$) for the ability to predict infants needed for surfactant and for Cl^- levels in the AF an area under the curve of 0.79 (95% confidence interval: 0.71–0.88, $p < .001$) for the ability to predict infants needed of surfactant (Table 4 and Figure 3).

Discussion

Our results suggest that Na^+ and Cl^- levels of the AF reflect those in the FLF and can be used as indicators for lung maturity in the neonatal period.

Because FLF constitutes one-third of the AF and has a more hypertonic composition compared to AF, its secretion into the alveolar space during gestation

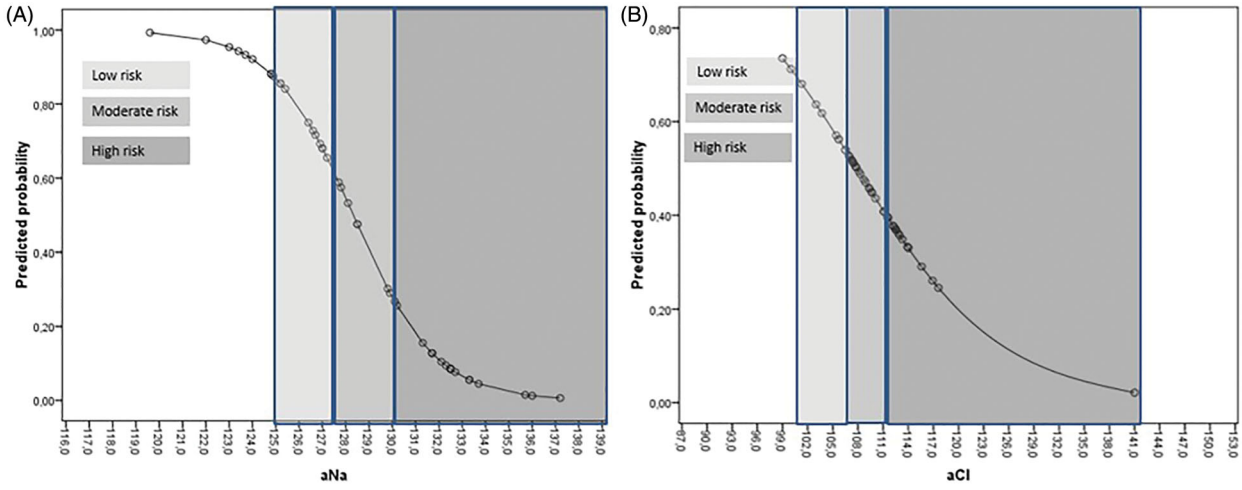


Figure 2. The probability chart for surfactant requirement under the 34th gestation weeks according to AF Na^+ (A) and Cl^- (B) levels.

Table 4. Predictive values and of amniotic fluid Na^+ and Cl^- level for the diagnosis of lung maturation.

	Cutoff value (meq/dl)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV(%)
Amniotic fluid Na^+ (meq/dl)	128.4	88.9	86.0	60	96.9
Amniotic fluid Cl^- (meq/dl)	111.0	66.7	81.7	60	91.3

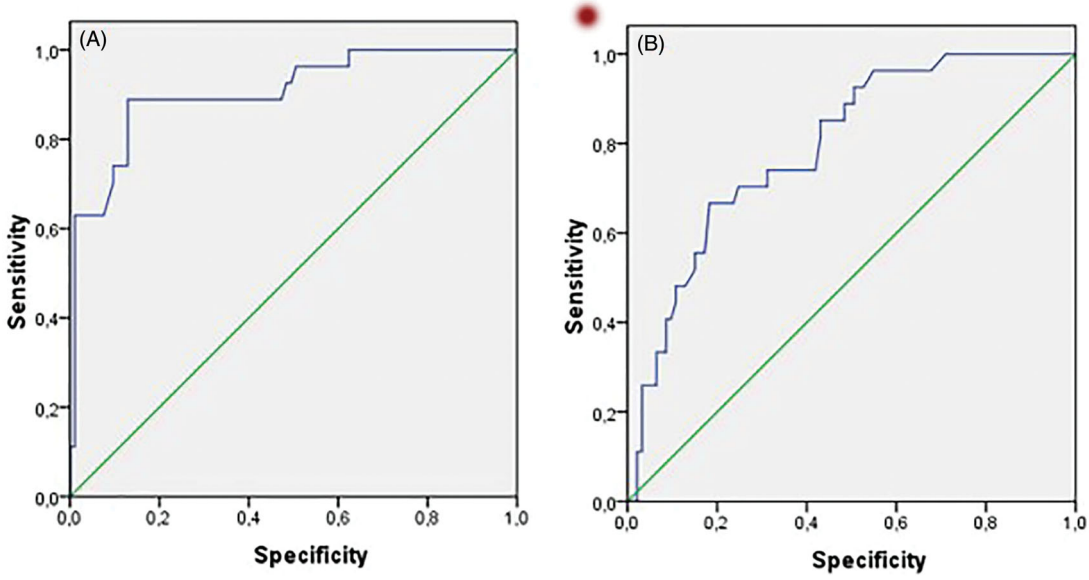


Figure 3. (A) Comparison of The Receiver Operating Characteristic (ROC) Curves of Amniotic fluid Na^+ Level. The area under the curve was 0.906 and $p < .001$ (95% CI: 0.838–0.974. The difference between areas was significant (95% confidence interval, CI; 0.74–0.89) ($p = .001$). (B) Comparison of The Receiver Operating Characteristic (ROC) Curves of Amniotic fluid Cl^-. The area under the curve was 0.793 and $p < .001$ (95% CI: 0.705–0.882).

and its reabsorption with the onset of labor also cause changes in AF electrolyte levels [4]. Epithelial sodium channels (ENaC) in the apical membranes of epithelial cells play an active role in the clearance of FLF from the airways. ENaC can be demonstrated in fetal airways from the early stages of pregnancy [13,14]. However, ENaC gene expression is correlated with gestational age. Therefore, preterm infants have lower ENaC expression compared to term infants [15]. Low ENaC expression leads to reduced absorption of FLF Na^+ and Cl^-, resulting in higher than expected levels of Na^+ and Cl^- in the AF. Tong et al. [16] found that AF Na^+ and Cl^- levels at 27 weeks of gestation were 129.9 meq/l and 106.18 meq/l, respectively. Oliveira et al. [10] showed that AF Na^+ level was 128.35 meq/l in infants at 27–34 weeks of gestation and decreased as gestation continued. Semerci et al. [17] showed that AF Na^+ and K^+ levels may be beneficial for the

prediction of respiratory morbidities in neonates. In our study, AF Na^+ and Cl^- levels in infants under 34 weeks' gestation were similar to those reported in the literature. We detected a negative correlation between gestational age and AF Na^+ and Cl^- levels. We found that infants under 34 weeks of gestation had significantly higher AF Na^+ and Cl^- levels than infants at gestational ages of 37 weeks or elder.A subanalysiss of preterm infants showed that AF Na^+ and Cl^- levels were significantly higher at 24–28 weeks' gestation compared to 28–34 weeks. We attribute the higher levels of AF Na^+ and Cl^- in preterm infants to insufficient FLF reabsorption.

The effect of delayed FLF clearance on respiratory distress syndrome (RDS) has not been determined [15]. However, in a study conducted in mice carrying ENaC mutation, Hummler et al. [18] reported that the mutant mice developed an RDS-like clinical presentation and died after birth. O'Brodovich [19] claimed that, in addition to surfactant deficiency, delayed FLF clearance due to ENaC deficiency played a mechanical role in the development of RDS in premature infants. Pitkänen et al. [20] also stated that inadequate reabsorption of FLF is an important factor contributing to respiratory distress in premature infants.

The need for neonatal surfactant replacement therapy is an indicator of incomplete lung development [21–23]. We observed significantly higher AF Na^+ and Cl^- levels in infants at gestational ages of 34 weeks or younger who needed surfactant compared to those who did not. This finding suggests that AF Na^+ and Cl^- levels may be used as an indicator of lung maturity.

Our results showed that Na^+ level over 128.4 mg/dl and Cl^- level over 111.0 mg/dl of the AF have high positive and negative predictive values and can be used as markers of lung maturity in neonates.

Conclusion

Our study shows that AF Na^+ and Cl^- levels can be used to screen lung maturation in newborn. In addition, this study showed that AF Na^+ and Cl^- levels can be used as early warning markers in determining the need for surfactant in premature infants. However, we think there is a need for multicenter large series for the routine use of AF Na^+ and Cl^- as an indicator of lung maturation.

Ethical approval

The research procedure was approved by the local ethical committee (IRB Number: GO 15/292).

Disclosure statement

None of the authors have any conflicts of interest in respect of the work presented in this paper.

Author contributions

All authors designed, supervised, and analyzed the study, and prepared the manuscript. All authors read and approved the final manuscript.

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Data availability statement

All data generated or analyzed during this study are included in this published article.

References

[1] Underwood MA, Gilbert WM, Sherman MP. Amniotic fluid: not just fetal urine anymore. J Perinatol. 2005; 25(5):341–348.
[2] Phelan JP, Smith CV, Broussard P, et al. Amniotic fluid volume assessment with the four-quadrant technique at 36–42 weeks' gestation. J Reprod Med. 1987;32(7): 540–542.
[3] Moore TR, Cayle JE. The amniotic fluid index in normal human pregnancy. Am J Obstet Gynecol. 1990; 162(5):1168–1173.
[4] Brace RA. Physiology of amniotic fluid volume regulation. Clin Obstet Gynecol. 1997;40(2):280–289.
[5] Ghidini A, Poggi SH, Spong CY, et al. Role of lamellar body count for the prediction of neonatal respiratory distress syndrome in non-diabetic pregnant women. Arch Gynecol Obstet. 2005;271(4):325–328.
[6] Russell JC. A calibrated fluorescence polarization assay for assessment of fetal lung maturity. Clin Chem. 1987;33(7):1177–1184.
[7] Neerhof MG, Haney EI, Silver RK, Ashwood ER, et al. Lamellar body counts compared with traditional phospholipid analysis as an assay for evaluating fetal lung maturity. Obstet Gynecol. 2001;97(2):305–309.
[8] Bhutani VK. Development of the respiratory system. In: Donn SM, Sinha SK, editor. Manual of neonatal respiratory care. 4th ed. Springer International Publishing Switzerland; 2017. p. 37–40.
[9] Benzie RJ, Doran TA, Harkins JL, et al. Composition of the amniotic fluid and maternal serum in pregnancy. Am J Obstet Gynecol. 1974;119(6):798–810.
[10] Oliveira FR, Barros EG, Magalhaes JA. Biochemical profile of amniotic fluid for the assessment of fetal and renal development. Braz J Med Biol Res. 2002;35(2):215–222.

[11] Johnson M, Allen L, Dobbs L. Characteristics of Cl-uptake in rat alveolar type I cells. Am J Physiol Lung Cell Mol Physiol. 2009;297(5):L816–L827.
[12] Sweet DG, Carnielli V, Greisen G, et al. European consensus guidelines on the management of respiratory distress syndrome – 2016 update. Neonatology. 2017; 111(2):107–125.
[13] Smith DE, Otulakowski G, Yeger H, et al. Epithelial Na(+) channel (ENaC) expression in the developing normal and abnormal human perinatal lung. Am J Respir Crit Care Med. 2000;161(4 Pt 1):1322–1331.
[14] Gaillard D, Hinnrasky J, Coscoy S, et al. Early expression of beta- and gamma-subunits of epithelial sodium channel during human airway development. Am J Physiol Lung Cell Mol Physiol. 2000;278(1): L177–L184.
[15] Helve O, Janer C, Pitkanen O, et al. Expression of the epithelial sodium channel in airway epithelium of newborn infants depends on gestational age. Pediatrics. 2007;120(6):1311–1316.
[16] Tong XL, Wang L, Gao TB, et al. Potential function of amniotic fluid in fetal development novel insights by comparing the composition of human amniotic fluid with umbilical cord and maternal serum at mid and late gestation. J Chin Med Assoc. 2009;72(7):368–373.
[17] Yılmaz Semerci S, Yücel B, Erbas IM, et al. The utility of amniotic fluid pH and electrolytes for prediction of neonatal respiratory disorders. J Matern Fetal Neonatal Med. 2018;22:1–5.
[18] Hummler E, Barker P, Gatzy J, et al. Early death due to defective neonatal lung liquid clearance in alpha-ENaC-deficient mice. Nat Genet. 1996;12(3):325–328.
[19] O'Brodovich HM. Immature epithelial Na + channel expression is one of the pathogenetic mechanisms leading to human neonatal respiratory distress syndrome. Proc Assoc Am Physicians. 1996;108(5): 345–355.
[20] Pitkanen OM, O'Brodovich HM. Significance of ion transport during lung development and in respiratory disease of the newborn. Ann Med. 1998;30(2): 134–142.
[21] Samtani MN, Pyszczynski NA, Dubois DC, et al. Modeling glucocorticoid-mediated fetal lung maturation: II. Temporal patterns of gene expression in fetal rat lung. J Pharmacol Exp Ther. 2006;317(1):127–138.
[22] Shi F, Liao Y, Dong Y, et al. Claudin18 associated with corticosteroid-induced expression of surfactant proteins in pulmonary epithelial cells. J Matern Fetal Neonatal Med. 2019;32(5):1–6.
[23] Cimato A, Facorro G, Martinez Sarrasague M. Developing an exogenous pulmonary surfactant-glucocorticoids association: effect of corticoid concentration on the biophysical properties of the surfactant. Respir Physiol Neurobiol. 2018;247:80–86.