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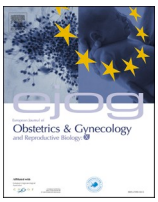

Evaluation of amniotic fluid neutrophil gelatinase-associated lipocalin and L-type fatty acid-binding protein levels during pregnancy

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ABSTRACT

Objective: We aimed to examine amniotic fluid neutrophil gelatinase-associated lipocalin (NGAL) and L-type fatty acid-binding protein (L-FABP) levels during pregnancy.
Study design: This study included singleton pregnancies. Amniotic fluid samples were collected at the time of vaginal delivery, cesarean section, amniocentesis, amnioreduction, and amnioinfusion. We analyzed changes of the NGAL and L-FABP levels during pregnancy and the factors affecting these values and their association with clinical outcomes.
Results: Three hundred and one pregnancies were analyzed. Respective Pearson correlation coefficients for the NGAL and L-FABP levels and gestational age at inspection were − 0.351 and − 0.819 ($p < 0.001$ and $p < 0.001$, respectively); weak and strong negative correlation were observed. The NGAL level was significantly higher in the intra-amniotic infection group than in the control group ($p < 0.001$). The L-FABP level was significantly higher in the fetal blood flow abnormalities group than in the control group ($p < 0.001$). The NGAL and L-FABP levels were significantly higher in the adverse outcomes group than in the control group ($p = 0.019$ and $p < 0.001$, respectively), and the respective areas under the concentration-time curve, with optimal cutoff values, for the NGAL and L-FABP levels were 0.693 (14,800 μg/gCr) and 0.864 (378 μg/gCr).
Conclusions: Amniotic fluid NGAL and L-FABP levels reflect fetal and neonatal immaturity. Additionally, the NGAL level is a useful predictive factor of intra-amniotic infection, and the L-FABP level is a useful predictive factor of fetal condition and short- and long-term prognoses.

1. Introduction

Amniotic fluid biomarkers are expected to reflect the fetal condition and intrauterine environment because amniotic fluid contains fetal urine. Various amniotic fluid biomarkers have been reported, such as neutrophil elastase, lactate dehydrogenase, white blood cell, glucose, [1] interleukin-6, [2] and matrix metalloproteinase-9 levels, [3] for predicting intra-amniotic infection. We previously reported novel amniotic fluid biomarkers: the amniotic fluid neutrophil gelatinase-associated lipocalin (NGAL) level as a useful predictive factor of fetal inflammatory response syndrome (FIRS) [4] and L-type fatty acid-binding protein (L-FABP) as a useful predictive factor of fetal hypoxia. [5] Urinary NGAL and L-FABP levels are useful in detecting acute kidney injury caused by septic shock. [6] NGAL levels correlate with inflammatory markers such as the white blood cell count and C-reactive protein level, whereas L-FABP levels correlate with organ hypoperfusion or oxidative stress. [6] These amniotic fluid biomarkers might have the potential to predict fetal short- and long-term prognoses and are expected to be applied clinically.

However, amniotic fluid NGAL and L-FABP levels during pregnancy have not been systematically examined. In this study, we aimed to examine the NGAL and L-FABP levels during pregnancy.

Abbreviations: NGAL, neutrophil gelatinase-associated lipocalin; FIRS, fetal inflammatory response syndrome; L-FABP, L-type fatty acid-binding protein; GA, gestational age; UmA, umbilical arterial; RDS, respiratory distress syndrome; CLD, chronic lung disease; NEC, necrotizing enterocolitis; IVH, intraventricular hemorrhage; PVL, periventricular leukomalacia; LGA, large for gestational age; FGR, fetal growth restriction; HDP, hypertensive disorders of pregnancy; PE, pre-eclampsia; ROC, receiver operating characteristic; PPV, positive predictive value; NPV, negative predictive value; OR, odds ratio; CI, confidence interval.

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2. Material and methods

2.1. Ethics statements

Informed consent was obtained from all patients, and the study protocol was approved by the Institutional Review Board of Shiga University of Medical Science Hospital.

2.2. Study design and patients

This study included singleton pregnancies managed and delivered at the Department of Obstetrics and Gynecology, Shiga University of Medical Science Hospital, Shiga, Japan, between August 2020 and February 2023.

2.3. Measurement data

To measure the amniotic fluid NGAL and L-FABP levels, amniotic fluid samples were collected transvaginally following the rupture of membranes in vaginal delivery or by using an 18-gauge needle to puncture the membrane before rupturing in cesarean section. Additionally, amniotic fluid samples were collected during amniocentesis for chromosomal examination, amnioreduction for polyhydramnios, and amnioinfusion for relieving umbilical cord compression due to oligohydramnios in cases of fetal blood flow abnormalities. [7,8] The samples were refrigerated at − 4 ℃ until the analysis was performed. The NGAL and L-FABP levels were measured using the two-step sandwich chemiluminescent enzyme immunoassay kit (SRL, Tokyo, Japan) following the manufacturer's protocol. The influence of variation in the urine flow rate should be considered for adequate evaluation of urinary biomarkers. Creatinine correction reportedly has a higher diagnostic power for acute kidney injury than the urine flow rate correction. [9] Therefore, the creatinine-corrected value was used for the amniotic fluid NGAL and L-FABP levels since it is difficult to evaluate the amount of amniotic fluid and fetal urine flow rate accurately.

We collected the following data on the maternal characteristics and clinical outcomes: maternal age, parity, body mass index, gestational ages (GAs) at examination and delivery, mode of delivery, birth weight, and umbilical arterial (UmA) pH, neonatal outcome including hospitalization in the neonatal intensive care unit, transient tachypnea of the newborn, respiratory distress syndrome (RDS), the need for respiratory support, such as nasal directional positive airway pressure and conventional mechanical ventilation, chronic lung disease (CLD), necrotizing enterocolitis (NEC), intraventricular hemorrhage (IVH), periventricular leukomalacia (PVL), retinopathy of prematurity, and neonatal and infant death.

Regarding factors influencing the NGAL and L-FABP levels, we used the amniotic-fluid collection method, blood contamination, turbidity, GA at examination, chromosomal or genetic abnormalities, major structural anomalies, large for gestational age (LGA), fetal growth restriction (FGR), fetal blood flow abnormalities, gestational diabetes mellitus, hypertensive disorders of pregnancy (HDP), preeclampsia (PE), intra-amniotic infection, UmA pH < 7.1, and Apgar score at 5 min < 7. LGA and FGR were diagnosed as > 1.5 and < 1.5 standard deviation based on the Japanese Society of Ultrasound in Medicine standards. [10] Fetal blood flow abnormalities were defined as the presence of an umbilical artery with absent or reversed end-diastolic velocity and/or ductus venosus with an absent or reversed A-wave that were associated with short- and long-term prognoses. [11,12] Intra-amniotic infection was defined as histological chorioamnionitis and funisitis because the presence of them is diagnostic criteria for FIRS, which is associated with neonatal adverse events. [13] Qualified pathologists histologically diagnosed the placenta and umbilical cord. A UmA pH < 7.1 and Apgar score at 5 min < 7 due to fetal hypoxia have been reported as prognostic factors for adverse neurological outcomes. [14,15] We defined UmA pH < 7.1, an Apgar score at 5 min < 7, CLD, NEC, IVH, and PVL related to adverse neurological outcomes and neonatal and infant death as adverse outcomes. [14–18].

2.4. Statistical analyses

Amniotic fluid NGAL and L-FABP levels were log-transformed to meet the assumption of normality of errors as much as possible. We used multiple regression analysis to analyze the relationship between factors and the NGAL and L-FABP levels. To clarify the change of the NGAL and L-FABP levels during pregnancy, the correlation between the NGAL and L-FABP levels and GA was assessed using the Pearson correlation coefficient. We also analyzed the relationship between the NGAL and L-FABP levels and clinical outcomes, and we calculated the receiver operating characteristic (ROC) curves of the adverse outcomes to demonstrate the relationship between the sensitivity and false positive rate (1–specificity) for the NGAL and L-FABP levels. The point corresponding to the highest sensitivity in relation to the highest specificity was the optimal cutoff point.

A p-value < 0.05 was considered statistically significant. Statistical analyses were performed using Easy R (EZR, R Foundation for Statistical Computing, Vienna, Austria) for Windows. [19].

3. Results

In this study, 301 single pregnancies were analyzed. Overall, 26, 1, 10, and 276 pregnancies had amniotic fluid samples collected at amniocentesis, amnioreduction, amnioinfusion, and delivery, respectively. Twenty-four (8.0%), 269 (89.4%), and 8 (2.6%) pregnancies had amniotic fluid samples collected only during pregnancy, only during delivery, and during both pregnancy and delivery, respectively. The patient characteristics and clinical outcomes are shown in Tables 1 and 2, respectively. Pathological examinations of the placenta and umbilical cord were performed in 114 pregnancies. A UmA pH < 7.1 and Apgar

Table 1
Patient characteristics.

Characteristic	Value
Number of patients	301
Number of amniotic fluid samples	314
Age (years)[a]	33 (18–47)
Primipara women (%)	155 (51.5)
In vitro fertilization (%)	62 (20.6)
Body mass index (kg/m^2)[a]	21.7 (14.3–45.2)
Vaginal collection (%)	121 (38.5)
Blood contamination (%)	87 (27.7)
Turbidity (%)	12 (3.8)
Chromosomal abnormalities (%)	30 (9.6)
Major structural anomalies (%)	76 (24.2)
Large for gestational age (%)	30 (9.6)
Fetal growth restriction (%)	84 (26.8)
Fetal blood flow abnormalities (%)	35 (11.1)
Gestational diabetes mellitus (%)	19 (6.1)
Hypertensive disorders of pregnancy (%)	48 (15.3)
Preeclampsia (%)	28 (8.9)

[a] Data are presented as median (range).

Table 2
Patient clinical outcomes.

Characteristic	Value
Number of patients	276
Gestational age at delivery (weeks)[a]	37.7 (25–41.4)
Cesarean section (%)	154 (55.8)
Birth weight (g)[a]	2690 (372–3788)
Umbilical arterial pH[a]	7.287 (7.027–7.474)
Intra-amniotic infection (%)	28 (24.6)
Neonatal intensive care unit admission (%)	147 (53.6)
transient tachypnoea of the newborn (%)	44 (16.1)
Respiratory distress syndrome (%)	30 (10.9)
Respiratory support (%)	87 (31.8)
Chronic lung disease (%)	12 (4.4)
Necrotizing enterocolitis (%)	3 (1.1)
Intraventricular hemorrhage (%)	4 (1.5)
Periventricular leukomalacia (%)	2 (0.7)
Retinopathy of prematurity (%)	9 (3.3)
Fetal death (%)	2 (0.7)
Neonatal death (%)	6 (2.1)
Infant death (%)	8 (3.0)

[a] Data are presented as median (range).

score at 5 min < 7 were observed in 8 and 9 (2.9% and 3.2%) pregnancies, respectively. Two pregnancies with fetal death had chromosomal or genetic abnormalities, 2 and 3 pregnancies of neonatal death had fetal major structural anomalies and chromosomal or genetic abnormalities, and 2 and 3 pregnancies of infant death had fetal major structural anomalies and chromosomal or genetic abnormalities, respectively.

Regarding the relationship between factors and amniotic fluid NGAL and L-FABP levels, vaginal collection, blood contamination, GA at examination, and intra-amniotic infection influenced the NGAL levels, whereas turbidity, GA at examination, and fetal blood flow abnormalities influenced the L-FABP levels (Table 3).

Respective Pearson correlation coefficients for the NGAL and L-FABP levels and GA at examination were − 0.351 and − 0.819 ($p < 0.001$ and $p < 0.001$, respectively); weak and strong negative correlation were observed between them (Figs. 1 and 2). To analyze the change of the NGAL and L-FABP levels during pregnancy, we used values without strong influencing factors such as intra-amniotic infection for the NGAL level and turbidity and fetal blood flow abnormalities for the L-FABP level.

We compared and analyzed the relationship between the NGAL and L-FABP levels at delivery and clinical outcomes for 264 pregnancies except for 12 pregnancies with fetal, neonatal, and infant deaths due to fetal major structural anomalies and chromosomal or genetic abnormalities, 144 pregnancies with vaginal collection and blood

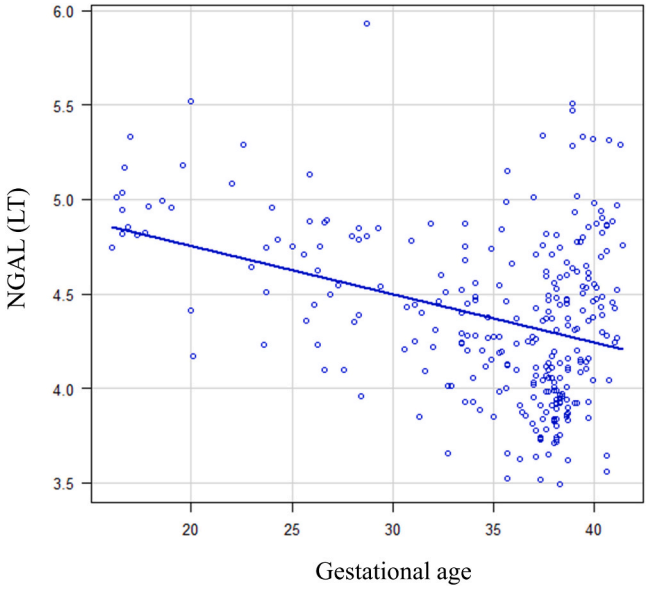

Fig. 1. Relationship between the amniotic fluid NGAL level and gestational age (r = −0.351). NGAL, neutrophil gelatinase-associated lipocalin; LT, log-transformed.

Table 3
Relationship between factors and the NGAL and L-FABP levels in multiple regression analysis.

	NGAL			L-FABP		
	Estimated value	95% CI	p-value	Estimated value	95% CI	p-value
Vaginal collection	1.8	(1.2–2.7)	0.002	0.65	(0.47–0.92)	0.016
Blood contamination	1.5	(1.0–2.4)	0.040	0.94	(0.65–1.3)	0.771
Turbidity	1.3	(0.53–3.4)	0.518	23	(10–50)	< 0.001
GA at examination	0.88	(0.84–0.92)	< 0.001	0.83	(0.80–0.87)	< 0.001
Chromosomal or genetic abnormalities	0.69	(0.30–1.5)	0.387	1.7	(0.85–3.3)	0.128
Structural anomalies	1.3	(0.80–2.3)	0.237	1.4	(0.93–2.2)	0.091
LGA	0.99	(0.54–1.8)	0.983	0.8	(0.52–1.4)	0.546
FGR	0.95	(0.60–1.5)	0.837	1.1	(0.79–1.7)	0.433
Fetal blood flow abnormalities	0.89	(0.48–1.6)	0.736	3.3	(2.0–5.7)	< 0.001
GDM	0.99	(0.47–2.0)	0.991	1.25	(0.67–2.3)	0.477
HDP	0.72	(0.37–1.4)	0.349	1.3	(0.78–2.3)	0.261
PE	1.2	(0.51–3.2)	0.592	2.1	(0.98–4.5)	0.055
Intra-amniotic infection	4.3	(2.7–6.8)	< 0.001	0.85	(0.58–1.2)	0.430
UmA pH < 7.1	0.56	(0.21–1.5)	0.258	0.56	(0.24–1.2)	0.176
Apgar score at 5 min < 7	1.0	(0.82–1.2)	0.887	0.92	(0.77–1.0)	0.347

The values are converted to the scale before logarithmic transformation.
NGAL, neutrophil gelatinase-associated lipocalin; L-FABP, L-type fatty acid-binding protein; GA, gestational age; LGA, large for gestational age; FGR, fetal growth restriction; GDM, gestational diabetes mellitus; HDP, hypertensive disorders of pregnancy; PE, preeclampsia; UmA pH, umbilical arterial pH; CI, confidence interval.

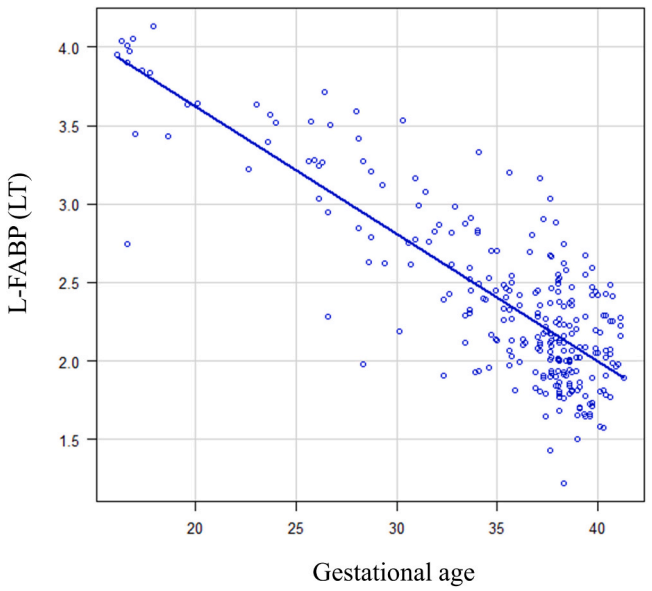

Fig. 2. Relationship between the amniotic fluid L-FABP level and gestational age (r = −0.819). L-FABP, L-type fatty acid-binding protein; LT, log-transformed.

contamination in the NGAL group, and 12 pregnancies with turbidity in the L-FABP group. In multiple regression analysis using significant factors in univariate analysis, the NGAL level tended to be higher in the RDS group than in the control group ($p = 0.092$). The L-FABP level was significantly higher in the UmA pH < 7.1, RDS, respiratory support, CLD, NEC, and PVL groups than in the control group ($p < 0.001$, $p = 0.044$, $p = 0.024$, $p < 0.001$, $p < 0.001$, and $p < 0.001$, respectively)., And we compared and analyzed the relationship of the NGAL and L-FABP levels at delivery with adverse outcomes. The NGAL and L-FABP levels were significantly higher in the adverse outcomes group than in the control group ($p = 0.019$ and $p < 0.001$, respectively) (Table 4).

We calculated the ROC curve of NGAL and L-FABP for predicting adverse outcomes. The respective AUC and optimal cutoff values of NGAL were 0.693 and 14,800 μg/gCr (sensitivity: 71.4%; specificity: 68.9%; positive predictive value [PPV]: 69.7%; negative predictive value [NPV]: 70.7%; odds ratio [OR]: 5.53; 95% confidence interval [CI]: 4.56–6.69), and those of L-FABP were 0.864 and 378 μg/gCr (sensitivity: 87.5%; specificity: 82.9%; PPV: 83.7%; NPV: 86.9%; OR: 33.93; 95% CI: 26.44–43.54).

4. Discussion

Amniotic fluid NGAL and L-FABP levels might have the potential to be applied clinically as a predictor of fetal condition and the intrauterine environment. However, knowledge regarding amniotic fluid NGAL and L-FABP levels are insufficient. Therefore, we examined these biomarkers during pregnancy systematically. Here, we established that the amniotic fluid NGAL and L-FABP levels were negatively correlated with GA. Additionally, the NGAL levels are useful predictive factors of FIRS, whereas the L-FABP levels are useful predictive factors of the fetal condition.

To analyze the change of amniotic fluid NGAL and L-FABP levels during pregnancy accurately, we excluded intra-amniotic infection for the NGAL level and turbidity and fetal blood flow abnormalities for the L-FABP level, as these influencing factors had three-fold or more influence. This study also showed a negative correlation between the NGAL and L-FABP levels and GA. A decrease in the urinary NGAL level from prematurity to childhood illustrates the age dependency, and the

Table 4
Relationship between amniotic fluid NGAL and L-FABP levels and neonatal clinical outcomes in univariate analysis.

	N	NGAL (10^3 μg/gCr)			N	L-FABP (10^2 μg/gCr)		
			Control	p-value			Control	p-value
UmA pH < 7.1	6	15 (3.3–56)	11 (3.6–851)	0.634	6	57 (5.0–395)	1.5 (0.16–137)	< 0.001
Apgar score at 5 min < 7	5	24 (6.8–803)	11 (3.3–851)	0.117	7	35 (0.63–39)	1.5 (0.16–139)	0.001
NICU admission	82	13 (3.3–851)	8.3 (4.4–252)	< 0.001	133	2.8 (0.44–395)	1.1 (0.16–8.0)	< 0.001
TTN	27	15 (3.3–721)	11 (4.3–851)	0.242	41	2.4 (0.58–79)	1.5 (0.16–395)	0.009
RDS	21	27 (7.1–851)	10 (3.3–721)	< 0.001	28	11 (1.5–139)	1.3 (0.16–395)	< 0.001
Respiratory support	52	17 (3.3–851)	8.9 (4.2–252)	< 0.001	78	6.2 (0.58–395)	1.2 (0.16–19.5)	< 0.001
CLD	6	43 (9.1–803)	11 (3.3–851)	0.012	10	56.9 (4.2–395)	1.5 (0.16–79.2)	< 0.001
NEC	3	31 (24–136)	11 (3.3–851)	0.041	3	97 (79–137)	1.5 (0.16–395)	0.003
IVH	3	17 (11–47)	11 (3.3–851)	0.25	4	8.7 (3.9–79.2)	1.5 (0.16–395)	0.010
PVL	1	25	11 (3.3–851)	0.341	2	201 (7.3–395)	1.5 (0.16–139)	0.037
ROP	5	64 (9.1–851)	11 (3.3–803)	0.015	9	25 (6.1–139)	1.5 (0.16–395)	< 0.001
Neonatal death	0	-	-	-	1	395	1.5 (0.16–139)	0.085
Infant death	3	31 (24–136)	15 (3.3–851)	0.203	3	97 (79–137)	2.8 (0.44–395)	0.005
Adverse outcomes	14	21 (6.8–803)	10 (3.3–851)	0.019	24	14 (0.58–395)	1.4 (0.16–46.2)	< 0.001

NGAL, neutrophil gelatinase-associated lipocalin; L-FABP, L-type fatty acid-binding protein; UmA pH, umbilical arterial pH; NICU, neonatal intensive care unit; TTN, transient tachypnoea of the newborn; RDS, respiratory distress syndrome; CLD, chronic lung disease; PVL, periventricular leukomalacia; NEC, necrotizing enterocolitis; IVH, intraventricular hemorrhage.; ROP, retinopathy of prematurity.
The Mann–Whitney U test was used for between-group comparisons, and data are presented as median (range).

urinary NGAL level is higher in preterm neonates, reflecting immature tubular transport mechanisms. [20] Immature tubular transport explains the changes of the NGAL and L-FABP levels during pregnancy. There is a decreasing trend observable during pregnancy, especially in the L-FABP level; therefore, the L-FABP level may reflect immaturity more than the NGAL level.

NGAL, which is expressed in the granules of human neutrophils and in several tissues, such as the lung, liver, and kidney, has renal protective and infection-protective effects. [6] Systemic inflammation stimulates NGAL synthesis in renal and nonrenal tissues and reduces tubular reabsorption, leading to an increased urinary NGAL level. [21] This study showed that the amniotic fluid NGAL level was strongly associated with intra-amniotic infection without being affected by other factors, and it was a useful predictor of FIRS, which is similar to the finding of a previous report. [4] There was also a higher NGAL level in the vaginally collection and blood contamination groups, and this may have been due to more inflammatory cells present in the cervix, vagina, and maternal blood.

L-FABP, expressed in the liver and proximal epithelial tubules, [6,22] reflects hypoxia induced by organ hypoperfusion. It serves as a target for highly cytotoxic aldehydes that are inevitably generated from lipid peroxidation reactions during reperfusion, thereby reducing lipid peroxidative stress. [22,23] Expression of renal L-FABP was suggested to protect the kidney tissue from renal ischemic stress. [22] This study showed that the amniotic fluid L-FABP level was strongly associated with fetal blood flow abnormalities without being affected by other factors and was a useful predictor of the fetal condition, specifically fetal hypoxia, which is similar to the finding of a previous report. [5] Additionally, the L-FABP level was not associated with HDP, but it tended to be associated with PE, which is the more severe category of HDP, causing maternal hypoperfusion. [24] L-FABP is expressed in human placenta, [25] and we speculated that increased expression of L-FABP in placenta caused by maternal hypoperfusion due to PE might lead to an elevated amniotic fluid L-FABP level. This could be the subject of a future study. There also was a higher L-FABP level in the vaginally collection and turbidity groups. L-FABP may be expressed in the cervix and vagina; however, this is unknown. The reason why vaginal collection influences the L-FABP level is unclear. L-FABP is expressed in the intestine, [22] and turbidity influences the L-FABP level since turbidity means the presence of meconium. [26] The precise etiology of amniotic fluid turbidity is unclear and potentially multifactorial. [26].

Neonatal clinical outcomes were analyzed excluding 12 pregnancies in which fetal major structural anomalies and chromosomal or genetic abnormalities were the direct cause of death. The elevated amniotic fluid NGAL level tended to be associated with RDS and may reflect immaturity since RDS is likely to develop in preterm neonates. [27] The elevated amniotic fluid L-FABP level was associated with UmA pH < 7.1, RDS, respiratory support, CLD, NEC and PVL. Fetal hypoxia leads to UmA pH < 7.1; [14] CLD is associated with FGR, intra-amniotic infection, and prematurity; [16] and NEC is associated with mesenteric ischemia, hypoxia, prematurity, and intra-amniotic infection. [17] Therefore, these results were reasonable because this study showed that the L-FABP level reflects immaturity more than the NGAL level as well as fetal hypoxia. We suggested that the amniotic fluid L-FABP level is a useful predictor of not only short-term but also long-term prognoses because UmA pH < 7.1, CLD, NEC, and PVL are associated with adverse neurological outcomes. [14,16–18] Actually, the NGAL and L-FABP levels were associated with adverse outcomes including UmA pH < 7.1, Apgar score at 5 min < 7, CLD, NEC, IVH, and PVL related to adverse neurological outcomes and neonatal and infant death; Furthermore, although the NGAL level had insufficient accuracy in predicting adverse outcomes, the L-FABP level had high accuracy.

This study has a couple of limitations. First, this study had a small number of cases with severe adverse neonatal outcomes, so further studies are needed before amniotic fluid NGAL and L-FABP levels can be applied clinically as predictors of adverse neonatal outcomes. Moreover, the precise etiology of fetal and neonatal adverse outcomes is multifactorial. Therefore, we suggest that studies use multiple amniotic fluid biomarkers to predict the prognosis more accurately. Second, knowledge of NGAL and L-FABP related to pregnancy is insufficient yet. This could also be the subject of future study.

5. Conclusions

Amniotic fluid NGAL and L-FABP levels reflect fetal and neonatal immaturity; the NGAL level is a useful predictive factor of FIRS, whereas the L-FABP level is a useful predictive factor of the fetal condition and short- and long-term prognoses. Amniotic fluid samples should be obtained by amniocentesis because the NGAL and L-FABP values are affected by the collection method.

Author contributions

DK conceived and designed the study, collected the data, performed the analysis, and wrote the manuscript. STs, AI, STo, TH, and NK collected the data. TM discussed the results and contributed to the final manuscript.

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None.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The datasets used in the current study are available from the corresponding author upon reasonable request.

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References

[1] Kidokoro K, Furuhashi M, Kuno N, Ishikawa K. Amniotic fluid neutrophil elastase and lactate dehydrogenase: association with histologic chorioamnionitis. Acta Obstet Gynecol Scand 2006;85:669–74.
[2] Romero R, Yoon BH, Mazor M, et al. The diagnostic and prognostic value of amniotic fluid white blood cell count, glucose, interleukin-6, and Gram stain in patients with preterm labor and intact membranes. Am J Obstet Gynecol 1993;169:805–16.
[3] Harirah H, Donia SE, Hsu CD. Amniotic fluid matrix metalloproteinase-9 and interleukin-6 in predicting intra-amniotic infection. Obstet Gynecol 2002;99:80–4.
[4] Katsura D, Tsuji S, Hayashi K, et al. Amniotic fluid neutrophil gelatinase-associated lipocalin and L-type fatty acid-binding protein in predicting fetal inflammatory response syndrome. J Obstet Gynaecol Res 2021;47:2616–22.
[5] Katsura D, Tsuji S, Hayashi K, et al. Amniotic fluid L-type fatty acid-binding protein in predicting fetal condition. Tohoku J Exp Med 2021;254:267–73.
[6] Asada T, Isshiki R, Hayase N, et al. Impact of clinical context on acute kidney injury biomarker performances: differences between neutrophil gelatinase-associated lipocalin and L-type fatty acid-binding protein. Sci Rep 2016;6:33077.
[7] Takahashi Y, Iwagaki S, Chiaki R, et al. Amnioinfusion before 26 weeks' gestation for severe fetal growth restriction with oligohydramnios: preliminary pilot study. J Obstet Gynaecol Res 2013;40:677–85.
[8] Katsura D, Takahashi Y, Iwagaki S, et al. Amnioinfusion for variable decelerations caused by umbilical cord compression without oligohydramnios but with the sandwich sign as an early marker of deterioration. J Obstet Gynaecol 2018;22:1–5.
[9] Tonomura Y, Uehara T, Yamamoto E, Torii M, Matsubara M. Decrease in urinary creatinine in acute kidney injury influences diagnostic value of urinary biomarker-to-creatinine ratio in rats. Toxicology 2011;290:241–8.

[10] Shinozuka N. Fetal biometry and fetal weight estimation: JSUM standardization. Ultrasound Rev Obstet Gynecol 2002;2:156–61.
[11] Vossbeck S, de Camargo OK, Grab D, Bode H, Pohlandt F. Neonatal and neurodevelopmental outcome in infants born before 30 weeks of gestation with absent or reversed end-diastolic flow velocities in the umbilical artery. Eur J Pedia 2001;160:128–34.
[12] Baschat AA. Neurodevelopment following fetal growth restriction and its relationship with antepartum parameters of placental dysfunction. Ultrasound Obstet Gynecol 2011;37:501–14.
[13] Gomez R, Romero R, Ghezzi F, Yoon BH, Mazor M, Berry SM. The fetal inflammatory response syndrome. Am J Obstet Gynecol 1998;179:194–202.
[14] Yeh P, Emary K, Impey L. The relationship between umbilical cord arterial pH and serious adverse neonatal outcome: analysis of 51,519 consecutive validated samples. BJOG 2012;119:824–31.
[15] Lau SL, Lok ZLZ, Hui SYA, Fung GPG, Lam HS, Leung TY. Neonatal outcome of infants with umbilical cord arterial pH less than 7. Acta Obstet Gynecol Scand 2023;102:174–80.
[16] Thébaud B, Goss KN, Laughon M, Whitsett JA, Abman SH, Steinhorn RH, et al. Bronchopulmonary dysplasia. Nat Rev Dis Prim 2019;5:78.
[17] Hackam D, Caplan M. Necrotizing enterocolitis: pathophysiology from a historical context. Semin Pediatr Surg 2018;27:11–8.
[18] Choi EK, Park D, Kim TK, et al. Animal models of periventricular leukomalacia. Lab Anim Res 2011;27:77–84.
[19] Kanda Y. Investigation of the freely available easy-to-use software 'EZR' for medical statistics. Bone Marrow Transpl 2013;48:452–8.
[20] van Donge T, Welzel T, Atkinson A, van den Anker J, Pfister M. Age-dependent changes of kidney injury biomarkers in pediatrics. J Clin Pharm 2019;59 (Supplement 1):S21–32.
[21] Mårtensson J, Bellomo R. The rise and fall of NGAL in acute kidney injury. Blood Purif 2014;37:304–10.
[22] Yamamoto T, Noiri E, Ono Y, et al. Renal L-type fatty acid–binding protein in acute ischemic injury. J Am Soc Nephrol 2007;18:2894–902.
[23] Derikx JP, Poeze M, van Bijnen AA, Buurman WA, Heineman E. Evidence for intestinal and liver epithelial cell injury in the early phase of sepsis. Shock 2007;28: 544–8.
[24] Rana S, Lemoine E, Granger JP, Karumanchi SA. Preeclampsia: pathophysiology, challenges, and perspectives. Circ Res 2019;124:1094–112.
[25] Campbell FM, Bush PG, Veerkamp JH, Dutta-Roy AK. Detection and cellular localization of plasma membrane-associated and cytoplasmic fatty acid-binding proteins in human placenta. Placenta 1998;19:409–15.
[26] Narendran V, Wickett RR, Pickens WL, Hoath SB. Interaction between pulmonary surfactant and vernix: a potential mechanism for induction of amniotic fluid turbidity. Pediatr Res 2000;48:120–4.
[27] McPherson C, Wambach JA. Prevention and treatment of respiratory distress syndrome in preterm neonates. Neonatal Netw 2018;37:169–77.