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Prematurity and growth retardation: different causes of fetal coagulation disorders

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Abstract: *prematurity and fetal growth retardation complicates in general one in five pregnancies. The combination of prematurity with severe growth retardation and hypoxia worsens the prognosis due to increase of periventricular hemorrhage incidence, and later coronary heart disease, hypertension, stroke, miscarriage and fetal growth retardation in own pregnancies in adulthood. The causes of impaired blood coagulation and fibrinolysis in retardated and premature newborns remain poorly understood. The goal of the study – to compare the parameters of blood acid-base and gases balance, coagulation and fibrinolysis in prematurity and growth retardation. Material and methods. Newborns of three clinical groups were examined. 24 full-term newborns with a postnatal diagnosis of I-II growth retardation stage constituted group I. 18 newborns of group II were born prematurely, corresponding to pregnancy term of 28-34 weeks. Control group III consisted of 20 full-term healthy infants. The birth took place without emergency interventions and without perinatal losses. Blood samples of the umbilical cord artery were obtained immediately after it was clamped. The parameters of blood acid-base and gas compositionof mothers and newborns blood and the fetal kinetic characteristics of blood coagulation and fibrinolysis were compared. Results. Newborns of group I had a pH of 7.27±0.012, in group II – 7.33±0.022, in group III – 7.30±0.024, which corresponded to the difference between maternal and fetal values of 0.10; 0.07 and 0.13. The pH difference in growth retarded fetuses is higher than in premature fetuses, closely to that of full-term fetuses, despite the lowest pH. Parameters of pO_2 and pCO_2 in labor do not have an indicative difference in newborns of all groups. Thromboelastometric parameters of the premature infants blood indicate low density and accelerated clot lysis, but while growth retarded fetuses accelerated formation and incomplete clot lysis. That is because of notable predominance of endothelial origin procoagulant factors in growth retarded fetuses. Indicators of acid-base and gas balance in newborns with growth retardation are shifted towards acidosis, in contrast to anthropometrically similar premature infants, whose indicators are higher than full-term fetuses ones.*

Key words: Premature Birth; Fetal Development; Umbilical Artery; Acidosis; Blood Coagulation; Fibrinolysis.

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Inroduction

According to the WHO, approximately 10% of children are born prematurely. (Chawanpaiboon, 2019). Complications arising from preterm birth (PB) are the leading cause of death among children under five years of age (Perin, 2022, WHO, 2023, Bila, 2024). A similar trend is observed with fetal growth restriction (FGR). Based on local or national standards of fetal growth, the prevalence of fetal growth 15 years ago ranged from 3% to 7% of the total population, an average of 5.13%, but with a noticeable increase (Romo, 2009), and the rate has reached 10% of all pregnancies up to present (Leal, 2023). That is, prematurity and fetal growth retardation complicates one in five pregnancies in general, and often in combination.

Yet few studies address the development of preterm infants with FGR in the literature (Ortigosa Rocha, 2010; Barreto, 2020). Studies have shown a paradoxical increase in the incidence of respiratory distress syndrome (RDS) with advancing gestational age among preterm infants with FGR. A similar paradoxical relationship was observed for periventricular haemorrhage (PVH) and necrotizing enterocolitis (NEC). A critical threshold appears around the 34th week of gestation, after which the mutual burden of prematurity and FGR increased (Gilbert & Danielsen, 2003). Two decades have passed since then, but no data have been found to supplement or deny the published results. And if researchers provided data were differed, then most often the difference was associated with the authors' use of another definitions of pathology (Ortigosa Rocha, 2010).

Neurological complications of varying severity are generally characteristic of prematurely born in further life. The mildest forms are impaired neuroadaptation, behavioral and cognitive disorders (van den Broek, 2010, Benítez-Marín, 2021). Prognosis worsens in cases of prematurity combined with severe FGR, when the incidence of hemorrhages in the lateral cerebral ventricles increases (Hernandez-Andrade, 2013, Klim, 2019). Prevalence of coronary heart disease, hypertension, stroke, and metabolic syndrome increases in adulthood. Impaired growth of cardiomyocytes and architectonics of heart muscle fibers leads to lifelong cardiac remodeling. Against the background of such disorders, FGR can complicate even own future pregnancy (Su, 2023).

There is a difference in the incidence of obstructive respiratory and cardiovascular diseases (Chan, 2010, Vidal, 2022). Respiratory morbidity is characteristic of prematurity, it is twofold high at birth less than 32 weeks without combination with FGR, the threshold is considered to be 30 weeks (Gutvirtz, 2022).

Both categories of newborns are characterized by disturbances in coagulation balance. Among the neonates who had PVH premature infants predominate (72.3%), mainly at 29-32 weeks' gestation, with hypoxia diagnosed in labor. Almost all of them required oxygen support, a third of them required strict respiratory parameters (Klim, 2019).

The fetus (its brain especially) normally consumes more oxygen than an adult. Although the pO_2 is 70% lower compared to adults, saturation is reduced by only 35%. Because hyperdynamic blood circulation with a high fetal heart rate, a high concentration of unique fetal hemoglobin (HbF) with an improved O_2 binding and transport capacity, ensure sufficient tissue oxygenation (Olofsson, 2023). That is, the hyperdynamic type of blood circulation is a condition for maintaining normal blood circulation with a high concentration of hemoglobin. The second important component of maintaining rheology is the balance in the blood coagulation system, which in the fetus, newborn and adult differs in development, as well as in the level of study. The small volume of fetal blood samples that can be obtained, the ethical aspects associated with this, complicate the vast majority of full-fledged laboratory tests (Chan, 2010). The synthesis of coagulation proteins begins at 20-22 weeks of gestation, gradually increasing until the parturition time. Their quantity is expected to be lower in premature infants (Strauss, 2011). However, the coagulation system of preterm fetuses is functionally balanced at a lower level than that in adults. Therefore, the actual risk of hemorrhage or thrombosis in healthy full-term and preterm infants is equal (Monagle & Massicotte, 2011, Katsaras, 2021). Where then, do the prerequisites for the PVH and NEC come from?

Aim

The goal of the study was to compare the parameters of blood acid-base and gases balance,

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coagulation and fibrinolysis in fetuses with prematurity and growth retardation.

Material and Methods

The retrospective cohort study was conducted from November 2022 to May 2023 on the basis of the Obstetrics and Gynecology Dept. No. 1 of the Bogomolets National Medical University in the Communal Non-Profit Enterprise "Perinatal Center of Kyiv". Approved by the protocol of the bioethics commission No. 163 dated 07.11.2022. Newborns from mothers aged 18 to 35 years with spontaneous labor onset were included. Women diagnosed with diabetes, preeclampsia or perinatal infection, long-term use of antiplatelet and tocolytic drugs were not included in the study. Bleeding during childbirth and fetal distress recorded by fetal monitoring were also excluded. All hospitalizations to the maternity unit took place in the active phase of the first stage of labor. The birth took place without emergency interventions. Also, there were no perinatal losses among them. Blood acid-base balance, gas composition, and coagulation and fibrinolysis parameters of newborns in three clinical groups and the pH of maternal cubital veins blood were compared.

24 full-term newborns with body weight close to 2000.0 and less than 10-th percentile of expected to gestation term constituted group I. All of them had the FGR I-II grades as main diagnosis after birth. The 18 of similar weight premature newborns of group II corresponded to 28-34 weeks of gestation. Control group III consisted of 20 healthy full-term infants. The composition of blood gases was examined using the biochemical analyzer of blood gases and electrolytes "Easy Stat" (Medica Corp., USA) and the pH-meter "LAURA Smart" (Erba Lachema, Czech Republic).

The kinetic characteristics of blood clot formation and lysis obtained from the umbilical cord arteries immediately after its clamping were measured automatically using the ROTEM® delta device (Instrumentation Laboratory, Germany) with the fib-tem® system reagent (Tem Innovations GmbH, Germany) which includes cytochalazin D, platelets blocker, excluding them from the coagulation process. The advantage of such a study of the hemostatic system is the minimal amount of blood for the study (340 µl vs the usual 3.5 ml) and obtaining the first values of the indicators within 10-20 minutes. The parameters of rotational thromboelastometry were used: CT – clotting time, angle α – coagulation polymerization rate, A5, A10 – amplitude at 5 and 10 minutes, MCF/MA – coagulation density / max. amplitude (maximal clot firmness/maximal amplitude), LI 30 – lysis for 30 min. (%), ML – maximal lysis. To assess statistical significance, the Student's t-test was used for comparing prevalence, the Mann-Whitney U-test for quantitative data, and the Kruskal-Wallis test for rank-based analysis.

Results

The average body weight in group I was 1908±110.4 g, in group II – 2016±85.8 g ($p>0.05$). Gestational age in days, respectively, 263±9.6 and 229±12.1 ($p<0.05$), in group III – 272±11.2 days ($p<0.05$). Acid-base and gas balance were determined and compared in three newborns groups formed depending on these values (table 1).

Table 1. Laboratory parameters of preterm and grow restricted newborns (M±m)

Indicators, units	Group I (n=24)	Group II (n=18)	Group III (n=20)
pH (mother), U.	7,37±0,010)*	7,40±0,021*	7,43±0,015*
pH (newborn), U.	7,27±0,012	7,33±0,022	7,30±0,024
Δ pH (mother-newborn), U.	0,10 [a b]	0,07 [b]	0,13
pO_2 mmHg	18±4,4	19±3,1	21±2,1
pCO_2 mmHg	40,8±1,88	44,8±2,65	44,3±3,47
HCO_3^- mmol/l	15,4±3,32 [b]	20,5±5,62 [b]	24,2±1,48
BE mmol/l	-(10,4±1,12)	-(6,7±1,96)	-(6,4±1,53)
Ht %	53,9±11,28 [b]	49,2±9,47	45,8±5,18

[a] – probable comparison with group II; [b]- probable comparison with the group III;
* – probable mother/fetus difference in the group.

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Despite the fact that there was no significant difference in weight of newborns, 17 of them (70.8%) in group I had a birth weight below the approximate average, while in group II there were 10 of them (55.5%), according to this indicator, the difference between the groups is probable ($p<0.05$). The probability of a difference in gestational age and a lower average body weight confirms fetal growth retardation in group I. Also in this group, 16 (66.7%) hematocrit values exceeded the average value, while in group II there were 8 (44.4%). Group I newborns had the lowest pH in the umbilical cord artery (7.27±0.012), the highest – in group II (7.33±0.022) with an average value in the control – 7.30±0.024 ($p>0.05$). Such values at the time of birth are not considered abnormal, but the difference between maternal and fetal indicators is clearly visible: the smallest difference is in group II (0.07), the largest in control group III (0.13), and 0.10 in group I. That is, in newborns with FGR the difference (Δ pH) with their mothers despite the low pH is higher than in premature infants. And this means better conditions for the elimination of metabolic substances toward maternal blood flow than those ones in preterm fetuses.

Of the water balance indicators in this study, we focused on the hematocrit value. It is highest in group I – 53.9±11.28% compared to groups II and III (49.2±9.47% and 45.8±5.18% respectively, $p<0.05$). The carbonate ion concentration differed significantly between groups; however, this difference may be attributed to automatic calculation by the diagnostic device, based on the entered patient temperature and inhaled oxygen percentage (FiO_2). In addition, at the completion of the second stage of labor (the time of obtaining the studied indicators), the respiratory component of the fetal blood gas balance uninformative. All of the above parameters of liquid and gas homeostasis are directly related to the rheological properties of blood, so the study of coagulation balance, which is quickly and graphically demonstrated by rotational thromboelastometry, is of great interest. By means of kinetic elastometric values in the fib-tem® test given in table 2, the parameters of blood fibrinogen concentration, fibrin coagulation formation and its lysis were investigated.

It does not matter that the blood clotting in group I in the fib-tem® test is probably faster, since all subsequent stages of blood clot formation are determined from the very beginning of its formation and are the same in their sequence. The difference is only quantitative. Parameters A5 and A10, as well as MCF/MA, reflect the formation and density of blood clots. According to these parameters, group I lags far behind groups II and III (A5 24.6±2.57, 13.2±3.34 and 16.2±3.38 (sec), $p<0.05$, respectively). Similarly, other amplitude measures show that fibrin clot formation in group I was 30-40% slower than in premature infants in group II, where clotting processes occurred 12-15% faster than in the control group. Clot lysis in newborns of groups I and II is slower than in group III, with this deceleration more pronounced in FGR cases—maximum lysis values were 9.6±5.36%, 8.1±2.50%, and 17.1±2.34% for groups I, II, and III, respectively ($p<0.05$). Such kinetic parameters of clot formation and destruction indicate a noticeable predominance of procoagulant factors in fetuses with growth restriction.

Table 2. Thromboelastometric illustration of the difference between preterm and grow restricted newborns

Parameters, units	Group I (n=24)	Group II (n = 18)	Group III (n = 20)
CT, sec.	21,9±5,31 [a b]	52,5±25,03 [b]	43,3±21,48
α angle, deg.	80,9±4,64	76,7±4,84	79,9±4,62
A5, mm	24,6±2,57 [a b]	13,2±3,34	16,2±3,38
A10, mm	27,3±9,62 [a b]	15,5±4,34	16,0±7,39
MCF/MA, mm	28,4±9,61 [a b]	15,2±11,37	17,4±7,04
LI 30, %	97,6±4,50 [b]	91,7±15,28 [b]	85,4±22,81
ML, %	9,6±5,36 [b]	8,1±2,50 [b]	17,1±2,34

[a] – probable comparison with group II; [b] – probable comparison with the group III.

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Discussion. This phenomenon requires further careful study, since the cellular component is excluded from the coagulation process. However, fetuses with all signs of FGR have the same features of the kinetic parameters of the elastogram as in native blood, from which platelet elements seem not to have been removed.

In the tenase complex an important (starting) role belongs to the tissue factor, which was previously associated with platelet membranes, and nowaday it is already clear that these substances are nothing more than acidic or negatively charged phospholipids (PhL) of cell membranes, mainly phosphatidylserine and phosphatidylethanolamine (Tsuda, 2006, Sakuragi & Nagata, 2023). Coagulation and anticoagulation reactions are coordinated and controlled by changes in the phospholipid composition of the cell membrane. Under normal conditions, acidic, or anionic PhL are always found on the internal surface of the cell membrane. The process of maintaining such lipid membrane asymmetry is ATP-dependent, therefore, under conditions of hypoxia, the maintenance of membrane asymmetry of PhL is inhibited (Daleke, 2003, Wang & Kinoshita, 2023). Hypoxic damage to the cell membrane (here we are talking about endothelial cells) is realized by externalization of acidic PhL, or their exposition on the external cell surface. This is where the cascade of blood clotting reactions begins (Bevers & Williamson, 2016).

The difficulty of differential diagnosis of fetuses with growth retardation, preterm and small to term ones begins with the position that provides for FGR when fetal weight is below the tenth percentile for gestational age. This criterion does not take into account constitutionally small fetuses, and can also be a factor in underestimating the FGR or prematurity in fetuses with an average normal weight before term, when they do not reach their growth potential. Although such underestimation is less problematic, each additional opportunity to accurately distinguish between normal and restricted growth can aid in timely and accurate diagnosis (Su, 2023).

Conclusions

Indicators of acid-base and gas balance in newborns with growth retardation are shifted towards acidosis, in contrast to anthropometrically similar premature infants, whose indicators are higher than full-term fetuses ones.

Kinetic signs of fibrin clot formation and lysis processes in premature infants are accelerated by 12-15% compared to full-term infants, and slowed down by 30-40% in fetuses with growth retardation.

The fib-tem® thromboelastometric test can be an important addition to the differential diagnosis of fetal growth restriction and prematurity due to the speed of obtaining the results of coagulation and fibrinolytic parameters of newborns, which are fundamentally different in prematurity and in growth retardation.

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Conflict of interests

Authors declare the absence of conflict of interests

Consent to publication

All authors have got consent to publication of this article.

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Недоношеність і затримка росту: різні причини порушень коагуляції у плода

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Абстракт: *недоношеність та затримка росту плода спільно ускладнюють кожну п'яту вагітність. Поєднання недоношеності із затримкою росту важких ступенів та гіпоксією погіршує прогноз через зростання частоти перивентрикулярних крововиливів, а в дорослому віці – через ішемічну хворобу серця, гіпертонію, інсульт, невиношування та затримку росту плода у власних вагітностях. Причини порушення коагуляції та фібринолізу крові новонароджених з дефіцитом маси та недоношених залишаються недостатньо вивченими. Мета дослідження – порівняти параметри кислотно-лужної та газової рівноваги, коагуляції та фібринолізу при недоношеності та затримці росту. Матеріал та методи. Обстежено новонароджених трьох клінічних груп. Групу I склали 24 доношених новонароджених з постнатальним діагнозом затримки росту I-II ст. 18 дітей II групи народилися передчасно в терміні вагітності 28-34 тижні. Контрольну групу III склали 20 здорових доношених дітей. Екстрених втручань і перинатальних втрат не було. Зразки крові артерії пуповини отримували одразу після її перетискання. Порівняли параметри кислотно-лужного і газового складу крові матерів і новонароджених та плодові кінетичні характеристики зсідання крові та фібринолізу. Результати. Новонароджені групи I мали показник pH 7,27±0,012, в групі II – 7,33±0,022, в групі III – 7,30±0,024, що відповідало різниці між материнськими і плодовими показниками 0,10; 0,07 і 0,13. Різниця pH у плодів із затримкою росту вища, ніж у недоношених, наближена до доношених, попри найнижчий pH. Параметри pO_2 та pCO_2 в процесі пологів не мають показової відмінності у новонароджених всіх груп. Тромбоеластометричні параметри крові недоношених вказують на невисоку щільність та прискорений лізис згортка, а при затримці росту – на прискорене утворення та неповний лізис згортка. Тобто, у плодів із затримкою росту помітне переважання прокоагулянтних чинників ендотеліального походження. Показники кислотно-лужної та газової рівноваги у новонароджених із затримкою росту зрушені в бік ацидозу, на відміну від антропометрично подібних недоношених, чиї показники вищі, ніж у доношених плодів.*

Ключові слова: передчасні пологи, розвиток плода, пупкова артерія, ацидоз, коагуляція крові, фібриноліз.