CHANGE OF RHEOLOGICAL STATUS AND FIBRINOGEN, AS MARKERS OF THE BLOOD CIRCULATORY SYSTEM, IN DIFFERENT PREGNANCY TRIMESTERS

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ABSTRACT

We studied the rheological profile and fibrinogen in healthy pregnant women in the I, II, and III trimesters, and a control group of women in the 2nd phase of the menstrual cycle. It turned out that rheological changes in different trimesters are heterogeneous and do not always correlate with the percentage changes in fibrinogen. Having discussed the data, we concluded that studying the full spectrum of rheological status is advisable to determine blood fluidity.

Keywords: aggregation; deformation; plasma viscosity; healthy pregnant; fibrinogen

Introductio

The regular operation of the circulatory system involves the interaction of the heart, arterial and venous vascular systems, blood volume, and viscosity. The pumping function of the heart depends on complex cardiomechanical processes, such as the inflow and outflow of blood to the heart. Acute circulatory failure is based on cardiac and vascular heart failure, which are the subject of study in clinical biomedicine, particularly cardiology. Clinical rheology, an integral part of this field, studies the fluidity of blood in micro and macro vessels. A change in the work of the heart entails a reaction from the microcirculation and microcirculation systems and the formation of compensatory mechanisms. The macrocirculation system includes: 1) a Cardiac pump, 2) Buffer vessels (arteries), and 3) Capacitor vessels (veins).

The microcirculation system includes 1) Resistance vessels (arterioles, venules), 2) Exchange vessels (capillaries), and 3) Shunt vessels (arteriole-venous anastomoses).

Blood flow must be adequate. This definition was introduced into world literature by Giorgi

Mchedlishvili.[1-3] He is the founder of clinical rheology in the world.
Characteristics of adequate blood have mathematical approaches.[4]
Heart performance is assessed by stroke volume and cardiac output.

$$SV = EDV - ESV$$

$$CO = SV \times HR$$

The cardiac output depends on the preload, determined by the venous return required for the stroke volume, and afterload, which is made up of the pressure in the aorta and the total vascular resistance.
PVR is calculated using the Poiseuille formula: $PVR = (1333 \times MDP \times 60)/MV)$. Cardiac output: $MDP = \frac{PP}{3} + DP\ CO = \frac{PP}{3} + DD$ If you have data on PVR and know the SI (cardiac index), you can determine the patient's blood circulation type. In obstetrics, where the postural syndrome acts in the same direction, a critical decrease in venous return may occur. Also, the types of blood circulation correlate with the uteroplacental blood flow (the most favorable is eukinetic).
Pregnancy places increased demands on the circulatory system, but at the same time includes mechanisms to meet them. The first of these mechanisms is an increase in the circulating blood volume. The average increase in water during pregnancy is from 6 to 8 liters, of which 4-6 liters are in the extracellular sector.
Most critical conditions that determine not only maternal but also perinatal mortality are accompanied by severe disturbances in water balance. There are three types of capillaries. In the arterial part, hydrostatic pressure predominates; in the middle, it is balanced with colloid-osmotic pressure; and in the venous part, colloid-osmotic pressure predominates. If blood pressure decreases or (and) colloid-osmotic pressure increases, then filtration decreases and reabsorption increases. The body compensates for the deficit of circulating blood volume (from the interstitium).[5]
If systemic blood pressure increases and colloid-osmotic pressure decreases, then filtration increases and reabsorption decreases, i.e., "the capillary drains excess fluid into the interstitial space.
Pregnancy brings about profound changes in the woman's cardiovascular system to meet the increased oxygen and nutrient needs of the mother and developing fetus.[6]
From the very beginning of pregnancy, the woman's blood flow begins to increase, peaking by the third trimester. This increase in blood flow provides enough oxygen and nutrients to support the growing fetus and the changing mother.
The heart begins to pump more blood with each beat, increasing cardiac output. This change helps provide the fetus with the resources it needs to thrive.
Vasculines dilate, which reduces total peripheral resistance and allows blood to flow freely to essential organs, including the placenta.
The mother's body adapts to cope with the increased demands on the cardiovascular system. The volume of circulating blood increases, which supports circulatory efficiency and the delivery of oxygen and nutrients.
The changes in blood flow and the vascular system during pregnancy are important adaptations that help support the viability and development of the fetus. Adequacy of blood circulation occurs at the level of rheological status of blood, which is provided by red blood cells, especially erythrocytes. Therefore, our study aimed to investigate the rheological status in pregnant women in different trimesters.

Materials and Methods

We investigate pregnant women (24-32years old). We investigate them in 4-13 weeks of gestation (first point of study), and next in 13-26 gestation weeks (second point of study), and next 37-38 gestation weeks (tried point of study). The control group consisted of 14 practically healthy women in the 2nd phase of the menstrual cycle. The Ethics Committee of the Society of Rheology approved the research design.

Inclusion parameters: First pregnancy.

Exception parameter: Hematological diseases in history, cancer, and anemia of pregnant women.

For a detailed description of the rheological status of blood, we studied erythrocyte aggregation, erythrocyte deformability, plasma viscosity, rheological index of red blood, and fibrinogen in pregnant women in the first, second, and third trimesters and compared them with control values.

The index of red blood cell aggregability (EAI)

The index of red blood cell aggregability represents the aggregated red blood cells' area ratio against the whole area of the red blood cells. Red blood cell aggregation was evaluated using the recently developed "Georgian technique", which provided us with direct and quantitative data. Blood samples (4ml) from the cubital veins were centrifuged, and about 0.1 ml of blood was diluted 1:200 in its own plasma in the Thoma pipettes, and then preliminary rinsed with 5% sodium citrate solution without the addition of any other anticoagulants to the blood under study. The diluted blood was placed into a glass chamber 0.1 mm high after standard mixing. The quantitative index of red blood cell aggregation, which was assessed with a special program at the Texture Analysis System (TAS-plus, Leitz, Germany), represented the relationship between the aggregated and unaggregated red cells.[7-9]

Red blood cell deformability index (EDI)

Evaluation of red blood cell deformability was performed with the aid of the nucleopore membrane filter method, which is based on assessing the velocity of the red blood cells' passage through the tiny pores (5 µm, which is a diameter of the smallest capillary) of the filter, at constant pressure (10 cm of water column) and temperature (37°C). Obtaining the pure red blood cells was performed by centrifuging the blood sample at 3000 rpm for 15 min. The resulting plasma was aspirated with a micropipette, and the remaining blood cells were added with bovine serum albumin (0.2 mg per 5 ml) dissolved in a phosphate buffer. Then, the blood was centrifuged a second time at 1000 rpm for 5 min. The precipitated red blood cells and a thin layer of leukocytes and thrombocytes were separated from the phosphate buffer. This procedure was repeated three times. Purified red blood cell mass was diluted in the phosphate buffer with a hematocrit of 10%. The evaluation of the deformability index involved measuring the velocity of red blood cell passage through the filter (mm/min). The high-quality polycarbonate filters (with 5 µm diameter pores) were used in measuring procedures.[7-9]

Plasma viscosity

Blood plasma viscosity was examined in a capillary viscometer at 37 °C. The Diameter of the capillary was about 1.8 mm. The displacement of plasma samples was induced by the gravitational force related to the difference in the levels of the plasma under study, about 65. (without ap-

plication of additional pressure) For the evaluation of the plasma viscosity in centipoises (cP), we determined the calibration factor (F). Blood plasma viscosity was calculated by multiplying the time for plasma displacement through the capillary by the instrument calibration factor.[7-9]

Blood rheological index in silico (BRI)

We used a new theologically significant parameter - the Blood Rheological Coefficient (BRI) to study blood rheology. The BRI is a complex indicator that mathematically reflects such indicators as the number of erythrocytes, their overall size, volume, and the amount of hemoglobin in each of them. The calculation of the BRI gives the clinician a comprehensive view of all the independent parameters of the red blood cells involved in the formation of laminar or turbulent flow, depending on blood viscosity.

Thus, if we assume that the viscosity of the plasma is constant, then the BRI is responsible for haematology.

If we analyze each of the parameters we are investigating from the point of view of its physical significance, we get that.

$$RDW = \frac{\max(OS)}{\min(OS)} \qquad (1)$$

The parameter of red blood cell distribution is the ratio of the largest red blood cell to the smallest red blood cell. It follows from (1) that

$$\max(OS) = RDW \times \min(OS) \qquad (2)$$

On the other hand, if we multiply the total number of red blood cells by the average haemoglobin, we get the numerical value of haemoglobin

$$\max(OS) + \min(GS) = RBC \times MCH \qquad (3)$$

It follows from (3) that

$$\max(OS) = RBC \times MCH - \min(OS) \qquad (4)$$

On the other hand, the volume of a red blood cell is equal to the sum of all red blood cells with large dimensions and all red blood cells with smaller dimensions divided by “2”, if n is the number of red blood cells with large dimensions, and (RBC - n) is the number of red blood cells with smaller dimensions

$$MCV = \frac{((\max(os) \times n + \min(OS) \times (RBC - n))}{2} \qquad (5)$$

Hence, it follows that

$$\max(GS) \times n + \min(GS) \times (RBC - n) = 2MCV \qquad (6)$$

Thus, if we group formulas (2), (4), and (6) into a system of equations, we have three unknowns and three easily solvable equations in the system

$$\begin{cases} \max(OS) = RDW \times \min(OS) \\ \max(OS) = RBC \times MCH - \min(OS) \\ \max(OS) \times n + \times (RBC - n) = 2MCV \end{cases}$$

where max(OS) × n is the coefficient of red blood cells Fibrinogen.

We study the determination of fibrinogen using the Rutberg method. The analysis is a gravimetric method; that is, to determine the fibrinogen level, the weight of the formed clot is measured. The method of studying fibrinogen, according to Rutberg, is as follows. First, it is necessary to obtain a fibrin clot. Thromboplastin (the third factor of the coagulation system) with calcium is added to the obtained biological material (blood plasma). Then, the resulting clot is dried, removing the remaining plasma with filter paper, and washed. Only then is the mass of the precipitated fibrin estimated, and the fibrinogen content is determined from it.

Statistical analysis.

Statistical significance was tested using one-way ANOVA and a two-sample test. Relationships yielding P-values less than 0,05 were considered significant. All values were expressed as the mean±standard error.

Results

Our studies showed that all rheological parameters and fibrinogen changed in the first trimester compared to the norm, as well as in comparison with the second and third trimesters of the same patients. Heterogeneous changes were observed between the second and third trimesters. You can see the details in Table 1.

Discussion

Changes in blood rheology in the first trimester of pregnancy are associated with complex hormonal, vascular, and metabolic changes to ensure normal fetal development and adaptation of the mother's body. Increased levels of estrogens and progesterone lead to changes in vascular tone and increased vascular permeability. Active production of human chorionic gonadotropin stimulates changes in the hemostasis system. In the first trimester, an increase in plasma volume begins, leading to hemodilution and a decrease in hematocrit, which leads to changes in rheological status. Our studies confirmed this. There is an increase in the level of fibrinogen, coagulation factors, and aggregation capacity of platelets, which subsequently affects the change in the rheology of red blood, as shown by our studies. Due to the increase in plasma volume, the blood becomes less viscous, which improves its fluidity and microcirculation. This is important for the normal functioning of the placenta and the blood supply to the fetus. Their deformability increases to improve permeability through capillaries. This is one of the adaptation moments.[10]

In the second trimester of pregnancy, changes in blood rheology continue and intensify, adapting the mother's body to the growing needs of the fetus. The main mechanisms of these changes are associated with increased blood volume, changes in plasma composition, and strengthening of the hemostasis system. By mid-pregnancy, plasma volume increases by 30-40%, and the number of red blood cells by only 20%. This leads to physiological hemodilution (blood thinning) and a relative decrease in hematocrit. Due to hemodilution, blood viscosity decreases, which improves microcirculation and blood supply to the placenta. However, the balance between fluid and formed elements must be maintained to avoid blood stagnation

and hypoxia; the fibrinogen level increases. The activity of fibrinolysis decreases, which makes the blood more prone to thrombosis. This is an important mechanism for protecting against blood loss in case of possible complications of pregnancy and childbirth. Increased platelet activation compensates for blood thinning and helps maintain optimal hemostasis. An environment that increases platelet aggregation becomes favorable for increased red blood cell aggregation. This is confirmed by an increase in the rheology coefficient in our studies. High estrogen levels promote vasodilation, improving blood flow. Progesterone maintains vascular tone, preventing excessive blood viscosity. In response to increased oxygen demand, erythropoietin levels increase, stimulating the formation of new red blood cells. Our studies confirm this.[11]

In the third trimester of pregnancy, blood rheology continues to change. The main changes include increased hypercoagulability, further increase in circulating blood volume, and adaptation of the vascular system. By the end of pregnancy, the circulating blood volume increases by 40-50% compared to the control. This ensures placental blood flow. Due to the imbalance between plasma and erythrocytes, physiological anemia of pregnancy persists. Fibrinogen levels increase sharply. Functional activity and the tendency to aggregation increase. [12,13] Our studies confirm this.

The parallelism of rheological status and blood circulation is a reliable physiological effect. Naturally, this also occurs during physiological pregnancy. For this purpose, the study of rheological status in pregnant women is one of the informative analyses.

Table 1. Parameters of rheological status and fibrinogen in the control group women and healthy pregnant women in different trimesters (M ± m)

Parameter	Unit	Control (N = 20)	I trimester (N = 20)	II trimester (N = 25)	III trimester (N = 25)
EAI	%	24 ± 2.3	31 ± 4.5*	26 ± 2.9	33 ± 4.5*
EDI	%	2.1 ± 0.01	2.2 ± 0.03	2.1 ± 0.04	2.2 ± 0.01
PV	sP	1.13 ± 0.05	1.15 ± 0.05	1.14 ± 0.05	1.13 ± 0.05
RI	unit	1.00 ± 0.05	1.15 ± 0.05*	1.10 ± 0.05	1.10 ± 0.05
Fibrinogen	g/L	2.6 ± 0.5	3.5 ± 0.7*	4.6 ± 0.9*	5.2 ± 0.6*

Abbreviations (by order of use in the text)

SV - stroke volume
CO - cardiac output
HR - heart rate
MDP - mean dynamic pressure
EDV - end diastolic volume
ESV - end systolic volume
TVR - total vascular resistance
PVR -peripheral vessels resistance
PP - pulse pressure
DP - diastolic pressure
PV - plasma viscosity

IR - index resistance
EAI - erythrocytes aggregation index
EDI - erythrocytes deformation index
BRI - blood rheological coefficient
MCH - mean corpuscular volume
MCV - mean corpuscular hemoglobin
RDW - red cell distribution width
OS - overall size of red cell distribution width
MCH - mean corpuscular hemoglobin

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