Disorders in the Lungs-Placenta System under Conditions of Experimental Gestosis in Rats

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Study of the arteriovenous difference in hormone levels and hemostasis parameters in rats with experimental gestosis induced by hypersodium diet showed decreased production of progesterone, increased level of hydrocortisone (resultant from its increased production and additional release of the hormone by the lungs), hypercoagulation, and retarded fetal development. Involvement of the lungs into the maintenance of optimum rheological parameters of arterial blood and a relationship between the level of fetoplacental hormones and the function of pulmonary fibrinolytic filter were detected.

Key Words: *fetoplacental complex; lungs; hemostasis; hormones; experimental gestosis*

The function of the fetoplacental complex (FPC) during gestation induced physiological changes in the maternal organism, specifically changes in the endocrine and hemostasis system: adaptation mechanisms promoting the maintenance of pregnancy are triggered with the beginning of gestation [2,3,5,6].

We studied the relationships between FPC status, hormone-producing function of the placenta, effects of FPC on maternal hemostasis and adaptive mechanisms, including the hemostasis-regulating and hormone-regulating functions of the lungs, on the model of gestosis [4].

MATERIALS AND METHODS

Experiments were carried out on female albino rats (170-190 g). Day 1 of pregnancy was determined by the presence of spermatozoa in vaginal smears after caging with a fertile male. After 7-day quarantine the animals were randomly divided into 2 groups (7 per group). Control group consisted of females with uncomplicated pregnancy, experimental group included rats with experimental gestosis induced by daily NaCl-enriched diet (1.8% NaCl in drinking water) starting from day 7 of gestation [4]. This ration led to changes similar to the development of gestosis (hypertension, activation of the renin-angiotensin system, reduction of uteroplacental bloodflow, intrauterine growth retardation).

On day 21 of pregnancy narcotized (60 mg/kg hexenal) rats were fixed by the limbs and tail and hysterotomy with removal of fetuses and euthanasia were carried out. Blood was collected from the umbilical artery and vein during hysterotomy before removal of the fetuses and placenta and placental and fetal weights at birth were measured. Biochemical studies included measurement of plasma Na level by plasma spectrophotometry (Vital Diagnostics kit), and hemostasis parameters: blood clotting time (BCT) and contents of fibrinogen [1] and antithrombin III by immunodiffusion in Hoechst dishes (Antithrombin Antigen test system, Nascola Ecat Foundation). Studies of the hormone status included enzyme immunoassay of progesterone (Immunotech reagents) and radioimmunoassay of hydrocortisone (Cea-Ire-Sorin test system) in arterial and venous blood.

The data were statistically processed using Statistica A software. The significance of differences and

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correlations were evaluated using parametrical (Student's *t* test) and nonparametrical (Spearman's and Wilcoxon's) tests.

RESULTS

The increase in blood Na concentrations in experimental animals was paralleled by physiological effects characteristic of gestosis: intrauterine growth retardation (10% decreased body weight, $p<0.05$), decreased weight of the placenta at birth, hemostasis disorders (Fig. 1).

Hypercoagulation developed in rats with experimental gestosis: BCT decreased by 12% in arterial and by 5% in venous blood compared to the control group ($p<0.05$). Blood fibrinogen content increased by 13% in the artery ($p<0.05$) and by 10% in the vein compared to uneventful pregnancy. Moreover, gestosis provoked a decrease in plasma anticoagulant activity: antithrombin III content decreased by 12.5% in the artery ($p<0.05$) and by 13.6% in the vein. This was paralleled by a decrease in plasma hydrocortisone concentrations (by 32%) characteristic of gestosis and a decrease in progesterone content (by 20%), indicating strained adaptation mechanisms and suppression of the placental hormone-producing function (Table 1).

In control rats physiological hypercoagulation, developing in pregnancy at the expense of more intensive formation of fibrin in the uteroplacental basin, is compensated by the function of the pulmonary fibrinolytic filter. This can be seen from positive arteriovenous difference (AVD), BCT ($p<0.01$), and increased consumption of antithrombin III in the pulmonary vessels (negative AVD for this parameter, Table 1). Hence, the defense (compensatory) systems, maintaining the rheology of arterial blood flowing to the placenta, normally prevent increase of blood clotting in chorionic villi under conditions of slow bloodflow.

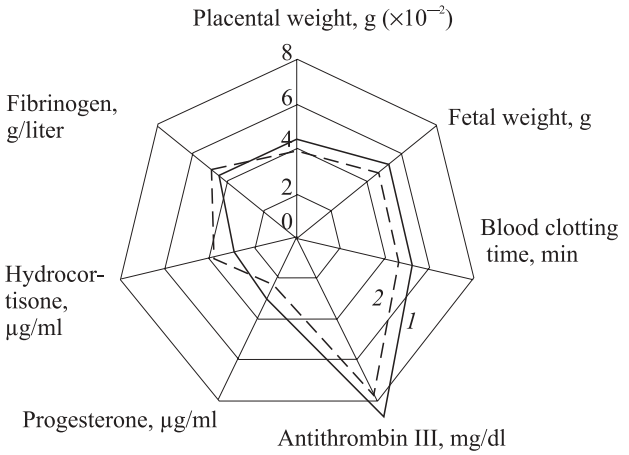

Fig. 1. Parameters of fetal development, coagulation and hormonal homeostasis in rats with uneventful pregnancy (*1*) and experimental gestosis (*2*).

TABLE 1. Fetal and Placental Weights, Hemostasis Parameters, and Hormone Levels in Pregnant Rats under Conditions of Hypersodium Diet ($M\pm m$; $n=7$)

Parameter		Control group	Experimental group
Plasma Na concentration, mmol/liter		140.00±1.08	154.50±1.93^{+}
Placental weight, g		0.44±0.02	0.39±0.01
Fetal weight, g		5.25±0.11	4.72±0.15^{+}
Plasma progesterone, μg/ml	artery	3.05±0.16	2.47±0.05^{+}
	vein	2.84±0.24	2.27±0.11^{+}
	AVD	0.21±0.22	0.19±0.06
Plasma hydrocortisone, μg/ml	artery	2.82±0.15	3.77±0.23^{+}
	vein	2.75±0.14	3.40±0.08*
	AVD	0.07±0.04	0.37±0.23
BCT, sec	artery	322.45±6.51	285.75±7.98^{+}
	vein	290.82±10.39*	276.00±5.59^{+}
	AVD	17.28±5.27	9.75±9.61^{+}
Fibrinogen, g/liter	artery	4.38±0.03	4.95±0.06^{+}
	vein	4.39±0.05	4.85±0.15
	AVD	-0.01±0.05	0.15±0.18
Antithrombin III, mg/dl	artery	8.87±0.37	7.76±0.47^{+}
	vein	9.62±0.39*	8.37±0.31*
	AVD	-0.75±0.15	-0.67±0.25

Note. AVD: arteriovenous difference. $p<0.05$ compared to *values in the artery; $^{+}$control group.

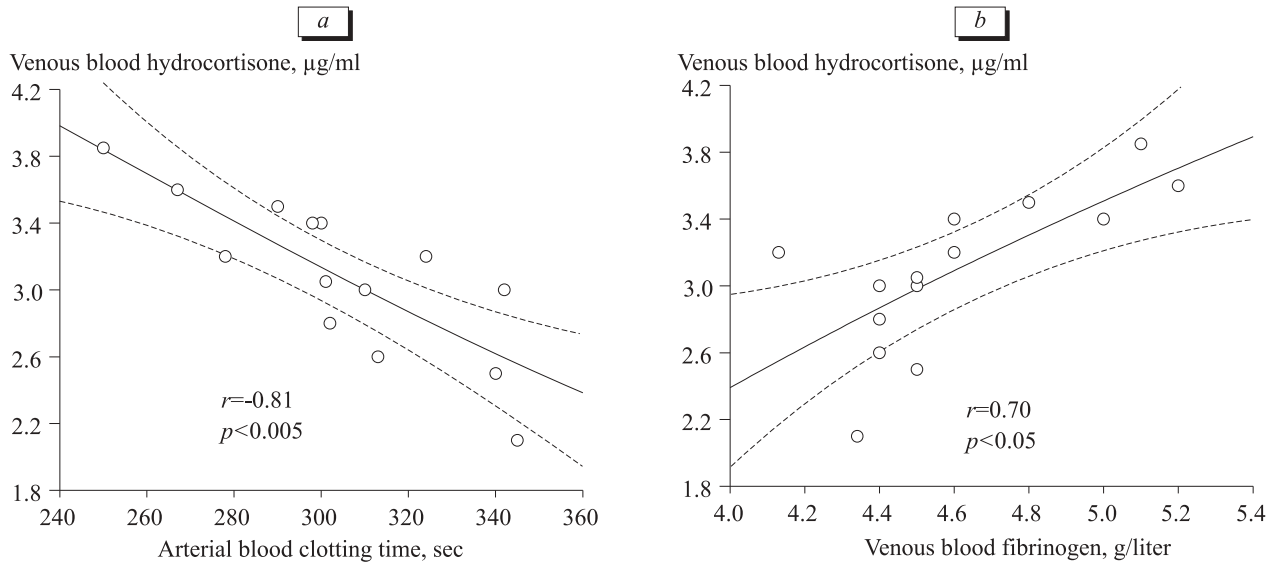

Fig. 2. Correlation between arterial blood clotting time and hydrocortisone level in venous blood (*a*) and fibrinogen and hydrocortisone levels in venous blood (*b*).

The study of the time course of hormones produced by the fetoplacental complex showed a positive, though statistically negligible, AVD for progesterone and hydrocortisone in 72% cases.

In experimental gestosis AVD for the studied parameters also attests to a drop of the fibrinolytic effect of the lungs. Despite increased consumption of antithrombin III in pulmonary vessels, the characteristics of venous blood with increased coagulation potential, flowing from the placenta, virtually did not change after passing the pulmonary circle. In contrast to the controls, arterial blood BCT did not change in experimental animals and a high level of fibrinogen persisted (Table 1). Moreover, progesterone content increased in gestosis, indicating suppressed synthetic function of the placenta and hydrocortisone level, which usually indicates fetal hypoxia. Plasma hydrocortisone and progesterone levels were in negative correlation, both in arterial (r=-0.79, p<0.005) and venous blood (r=-0.60, p<0.05).

In contrast to the control group, hydrocortisone concentration in arterial blood of experimental rats was significantly higher than in venous blood. This indicates predominance of hormone release from the lungs. Positive AVD of progesterone content was observed in 100% cases, which also attested to hormone release by the lungs under conditions of experimental gestosis. BCT and hydrocortisone level were in negative correlation (r=-0.70, p<0.05 in arterial blood; r=-0.81, p<0.005 in venous blood). Analysis of correlations also showed a direct significant relationship between the levels of fibrinogen and hydrocortisone: r=0.64, p<0.05 in arterial blood and r=0.70, p<0.05 in venous blood (Fig. 2).

Hypercoagulation shifts in the clotting system corresponded to the maximum hydrocortisone levels, detected in experimental gestosis: shorter BCT and higher level of fibrinogen (Fig. 2). In addition to relationship between hydrocortisone level and hemostasis parameters, we detected an inverse correlation between hydrocortisone level and placental and fetal weights at birth. Coefficient of correlation between hydrocortisone content and placental weight was -0.69 (p<0.05), between hydrocortisone content and fetal weight -0.73 (p<0.05).

By contrast, analysis of the dynamics of progesterone content showed an inverse correlation between its levels and fibrinogen content in the venous blood (r=-0.61, p<0.05 for progesterone level in arterial blood and r=-0.57, p<0.05 in venous blood). Decreased production of progesterone by FPC in experimental gestosis was paralleled by an increase in blood fibrinogen content. Vasodilating and endothelium-protective effects of progesterone suggest that its sufficient level was due to the lung capacity to reduce the coagulation potential of the blood flowing from the placenta.

Hence, not only the lungs regulate the rheology of the uteroplacental bloodflow, but the function of pulmonary fibrinolytic filter also depends on the humoral stimuli of the placenta. These interactions suggest the existence of a lungs-placental functional system. The placenta, which forms a sort of barrier providing immunological tolerance and secretes hormones with endothelium-protective effects, and the lungs regulating the coagulation potential and hormonal homeostasis, are components of the adaptation system providing normal course of gestation.

However, the presence of pathological factors, induced by 1.8% NaCl (under conditions of experimental gestosis) activates the renin-angiotensin mechanism, and disseminated vasospasm promotes disorders in the coagulation homeostasis. Thrombogenic potential of the blood flowing from the placenta notably increases, surpassing the barrier potentialities of the placental thrombolytic system and of the pulmonary fibrinolytic filter. Generalized vascular spasm, induced by electrolyte and hormone imbalance (decreased synthesis of progesterone and estrogen leads to predominance of vasoconstrictive effects), in turn, stimulates aggregation and coagulation activities of the plasma. Together with these disorders, imbalance in hormone production by the FPC maintains endothelial damage, specifically, in the pulmonary circulation basin. Increase in Na concentration promoting the development of arterial hypertension [4] seems to trigger hydrocortisone release in experimental rats. Hyperproduction of hydrocortisone by maternal adrenals and FPC and additional release of the hormone by the lungs in experimental rats serve as the defense mechanism providing prolongation of pregnancy under conditions of stress and decreased defense potential of the immune system.

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