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The concentration of pregnancy-associated plasma protein-a in the blood serum of tobacco smoking pregnant women in the first trimester of pregnancy

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Exposition to tobacco smoke during pregnancy is an important risk factor for health of both mother and fetus. Pregnancy-associated plasma protein-A (PAPP-A) plays role in immunosuppression, where it causes trophoblasts to be not recognized by the mother's organism as a foreign body. It initiates growth and vitality of fetus through influence on structure and functioning of placenta.

Purpose — to estimate levels of PAPP-A in serum blood of smoking and non-smoking women in the first trimester of pregnancy.

Methods. In total 4473 patients aged 18 to 47 years with singleton pregnancy were examined and performed the non-invasive maternal screening test in the first trimester. Biochemical measurements were fully automated by means of immunofluorescence method, using Delfia Xpress analyzers (Perkin Elmer). The software «Statistica v.10.0» was used for statistical analysis.

Results. We observed that PAPP-A MoM value of the smoking woman was on average 16.0 % lower in comparison with the results of the non-smoking woman of the same weight with the standard error of 3.5 %. While 1kg weight gain decreased PAPP-A MoM value in both groups by 1.8 % with the standard error of 0.1 %.

Conclusions. Smoking during pregnancy decreases PAPP-A MoM level by 16.0% on average in comparison with the results of a non-smoking woman of the same weight. Considering the relations between PAPP-A level and smoking, body weight and gestational age, all these factors should be taken into account for the correction of PAPP-A level and genetic risk assessment.

Key words: PAPP-A, pregnancy, first trimester, tobacco smoking.

Рівень асоційованого з вагітністю протеїну-А плазми (PAPP-A) в сироватці крові жінок у першому триместрі вагітності

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Експозиція на тютюновий дим під час вагітності є важливим фактором ризику для здоров'я матері і плода. Протеїн плазми, асоційований з вагітністю (PAPP-A), відіграє роль в імуносупресії, в результаті організм матері сприймає трофобласт як чужорідне тіло, стимулює ріст і життєздатність зародка через вплив на структуру і функцію плаценти.

Мета — провести оцінку рівня PAPP-A в сироватці крові жінок, які палять, і жінок, які не мають такої звички, у першому триместрі вагітності.

Методи. Обстежено 4473 вагітні пацієнтки віком від 18 до 47 років з одноплідною вагітністю. Зроблено пренатальний тест із сироватки крові, неінвазивний тест першого триместру. Проведено вимірювання концентрації біохімічних показників повністю автоматизованим методом імунофлюоресценції за допомогою апарату Delfia Xpress (Perkin Elmer). Для статистичного аналізу використано програму Statistic версії 10.0.

Результати. Значення МоМ PAPP-A для жінки-курця в середньому на 16,0% ((1-10-0,076)?100%) нижче порівняно з результатами жінки, яка не палить, при однаковій вазі і стандартній похибці 3,5%. У той час як збільшення ваги на 1 кг знижує рівень МоМ PAPP-A в обох групах на 1,8% зі стандартною похибкою 0,1%.

Висновки. Куріння вагітними впливає на зниження рівня PAPP-A в середньому на 16% при однаковій вазі жінок. Оскільки паління, вага пацієнтки і термін гестації впливають на рівень PAPP-A, то всі ці фактори мають бути враховані для корекції значення PAPP-A і оцінки генетичного ризику.

Ключові слова: PAPP-A, вагітність, перший триместр, куріння.

Уровень ассоциированного с беременностью протеина-А плазмы в сыворотке крови курящих женщин в первом триместре беременности

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Экспозиция на табачный дым во время беременности является важным фактором риска для здоровья матери и плода. Протеин плазмы, ассоциированный с беременностью (PAPP-A), играет роль в иммуносупрессии, в результате организм матери воспринимает трофобласт как чужеродное тело, стимулирует рост и жизнеспособность зародыша через влияние на структуру и функцию плаценты.

Цель — провести оценку уровня PAPP-A плазмы в сыворотке крови курящих и некурящих в первом триместре беременности.

Пациенты и методы. Обследованы 4473 беременные пациентки в возрасте от 18 до 47 лет с одноплодной беременностью. Сделан пренатальный тест из сыворотки крови, неинвазивный тест первого триместра. Измерение концентрации биохимических показателей проведено полностью автоматизированным методом иммунофлюоресценции с помощью аппарата Delfia Xpress (Perkin Elmer). Для статистического анализа использована программа Statistic версии 10.0.

Результаты. Значение МоМ PAPP-A для женщины-курильщика в среднем на 16,0% ((1-10-0,076)х100%) ниже по сравнению с результатами женщины, которая не курит, при одинаковом весе и стандартной погрешности 3,5%. В то время как увеличение веса на 1 кг снижает значение МоМ PAPP-A в обеих группах на 1,8% со стандартной погрешностью 0,1%.

Выводы. Курение беременными влияет на снижение уровня PAPP-A в среднем на 16% при одинаковом весе женщин. Поскольку курение, вес пациентки и срок гестации влияют на уровень PAPP-A, то все эти факторы должны быть учтены для коррекции уровня PAPP-A и оценки генетического риска.

Ключевые слова: PAPP-A, беременность, первый триместр, курение.

Introduction

Exposition to tobacco smoke during pregnancy is an important risk factor for the health of both the mother and the fetus. It has a significant effect on a higher rate of miscarriages, sudden fetal deaths, lower birth weight, and respiratory insufficiency after the child's birth [8].

Correlation between smoking by mothers and chilognathopalatoshisis occurrence in children has also been proven. Substances contained in tobacco smoke, such as nicotine, carbon oxide and cyanides, have toxic properties. Their presence in an organism can influence immunological mechanisms which compose an important element determining equilibrium between the bodies of the mother and the fetus [6].

Pregnancy Associated Plasma Protein A (PAPP-A) is a multicellular glycoprotein, containing zinc, built from heterodimer with molecular weight of 500 kDa. This protein is created in syncytiotrophoblasts and plays a role in immunosuppression, causing trophoblasts to be not recognized by the mother's organism as a foreign body [9, 10]. It stimulates growth and development of the fetus' cells through IGF *(insulin-like growth factor)*, released from a complex created with IGFBP *(insulin-like growth factor binding protein)*. In early pregnancy, IGF-II *(insulin-like growth factor II)* stimulates growth and vitality of the fetus, while IGF-I *(insulin-like growth factor I)* regulates flow of nutrients in the maternal-fetal unit. Impaired expression of IGF in placenta in initial stages of pregnancy may be a cause of placental insufficiency, which in turn may lead to impaired fetal growth [9, 10].

Purpose — to estimate levels of pregnancy-associated plasma protein-A (PAPP-A) in blood serum of smoking and non-smoking women in the first trimester of pregnancy, taking into account pregnancy duration, and also age and body weight of pregnant women.

Materials and methods

Study group consisted of 4473 women aged 18 to 47 with unifetal pregnancy who underwent first trimester non-invasive screening through blood serum tests referred to Prenatal Genetic Clinic of University Hospital in Bydgoszcz. General characteristics of the group are shown in Table 1. Average age was 33.1 with standard deviation of 5.3 years. Gestational age, obtained from CRL *(crown rump length)* measurements conducted by ultrasonography screening, was between 72 to 97 days with the mean of 89 days.

Biochemical parameter levels measurement was conducted through a fully automated immunofluorescence method used in Delfia Xpress (Perkin Elmer) analyzer in Center of Medical Diagnostics «Lipowa» in Bydgoszcz. The blood serum was obtained between 11th and 13.6th week of pregnancy.

Statistical analysis was conducted using program Statistica v.10.0. For comparison of distribution of continuous variables Kruskal—Wallis test was used, together with a multiple comparison test. Since PAPP-A variable has an approximately log-normal distribution, we analyze differences in distribution of $\log_{10}$PAPP-A. Prior to conducting correlation and regression analysis, outliers for $\log_{10}$PAPP-A were removed by making use of the Grubbs test. In order to adjust difference of $\log_{10}$PAPP-A values for smoking and non-smoking women for potential confounding factors multiple regression. Differences were considered as significant for p value lower than 0.05 [2].

Results

In the analyzed group there were 4119 (92.1%) non-smoking patients, 288 (6.4%) smokers and 66 (1.5%) patients who quit smoking during pregnancy. There were no significant differences between these groups in distribution of age,

Table 1

Characteristics of the study group (n=4473)

	Average	Standard deviation	Median	Range
Age at the time of delivery (years)	33.1	5.3	34.1	(18-47)
Gestational age (days)	89	4	89	(72-97)
Weight (kg)	66.0	12.4	64.0	(40.0-145.0)
PAPP-A	2961	1946	2484	(1-18121)
$\log_{10}$ PAPP-A	3.381	0.297	3.395	(-0.009-4.260)
MoM PAPP-A	1.175	0.727	1.000	(0.006-7.600)

Table 2

Comparison of smoking, non-smoking, and the group that stopped smoking during pregnancy*

	Non-smoking (n=4119)	Smoking (n=288)	Stopped smoking during pregnancy (n=66)	p value for Kruskal-Wallis test
Age at the time of delivery (years)	34.0 (24.0–40.6)	34.6 (20.8–42.0)	33.5 (24.7–40.9)	0.77
Gestational age (days)	89 (81–96)	89 (81–96)	88 (81–95)	0.35
Weight (kg)	64.0 (50.0–90.0)	63.0 (50.5–92.0)	63.0 (52.7–92.8)	0.84
PAPP-A	2518 (812–6683)	2069 (684–5718)	2383 (677–5609)	<0.001
$\log_{10}$ PAPP-A	3.401 (2.910–3.825)	3.316 (2.835–3.757)	3 377 (2 831–3 749)	<0.001
MoM PAPP-A	1.013 (0.352–2.580)	0.823 (0.280–2.343)	0.878 (0.321–2.020)	<0.001

* — table contains median and percentile values (P5-P95)

weight, and gestational age. Results are presented in Table 2. Nevertheless, we observed significant differences for PAPP-A levels, $\log_{10}$PAPP-A, and MoM PAPP-A. Multiple comparison test proved the results of smoking and non-smoking groups to be substantially different. Median values for variables were substantially lower in the group of smoking women (p<0.001).

It was found that log10 PAPP-A variable was significantly correlated with gestational age (R=0.38, p<0.001), patient's weight (R=-0.33, p<0.001), and age at the moment of delivery (R=-0.07, p<0.001). A positive correlation between log10 PAPP-A and gestational age means that when gestational age increases, $\log_{10}$ PAPP-A value tents to increase. A negative correlation between log10 PAPP-A and weight indicates that the women who had higher weight had a tendency to obtain low $\log_{10}$ PAPP-A value, and vice versa. Correlation of log10 PAPP-A with age was very low, it was however still statistically significant.

Differences between average $\log_{10}$ PAPP-A values for groups of smoking and non-smoking women with adjustment for gestation age, weight, and patients' age were studied using multiple regression. Smoking was considered as a dichotomous variable, with 0 value being assigned to non-smokers and 1 to smokers. Results of multiple regression are shown in Table 3. The simultaneous adjustment for weight and age was not allowed, since the correlation between age of patients and weight was much higher than the correlation between age of patients and log10 PAPP-A. Not to compromise the assumptions of a multiple regression, two models were considered. In both models log10 PAPP-A difference between smoking and non-smoking women were statistically significant and higher than original difference. Partial correlations were higher than correlations in models with one variable. Coefficient of determination allows to assess the goodness of fit of the regression equations. It indicates that the model which with body weight, gestational age, and smoking gives better fit than the model with patients' age, gestational age and smoking. The first model explains 28% of the total variation in log10 PAPP-A, while the second one explains only 17%.

The results shown that PAPP-A level was on average 16.0% ($(1-10^{-0.076})\times100\%$) lower in smoking group than in non-smoking group with same values for body mass and gestational age with the standard error of 3.5%. Moreover, we observed that 1 kg difference of weight with the same level of the other variables gave on average 1.8% ($(1-10^{-0.008})\times100\%$) lower PAPP-A level with the standard error of 0.1%. We also

Table 3

Multiple regression analysis for log10 PAPP-A and selected independent variables (n=4401)

	Model 1			Model 2		
	Regression coefficients	Partial correlations	p value	Regression coefficients	Partial correlations	p value
Age at the time of delivery (years)				-0.004	-0.08	<0.001
Gestational age (days)	0.026	0.41	<0.001	0 026	0.39	<0.001
Smoking (0-smoking 1-not smoking)	-0.078	-0.08	<0.001	-0.080	-0.07	<0.001
Weight (kg)	-0.008	-0.37	<0.001			
Intercept	1.611		<0.001	1.237		<0.001
Coefficient of determination R^2	0.27			0.16		
p value for model	<0.001			<0.001		

Table 4

Multiple regression analysis for log10 PAPP-A MoM and selected variables (n = 4400)

	Regression coefficients	Standard error	Partial correlations	P value
Smoking (1-smoking 0-not smoking)	-0.076	0.015	-0.37	<0.001
Weight (kg)	-0.008	0.0003	-0.07	<0.001
Intercept	0.518	0.02		<0.001
Coefficient of determination R^2	0.14			
p value for the model	<0.001			

verified that PAPP-A level rise on average 6.3% $((10^{0.027-1})\times100\%)$ in every day of pregnancy. In this case the standard deviation was 0.2%. It should be stressed that cited estimates can only be referred to the studied range of variable values.

Evaluation of the influence of smoking on PAPP-A MoM value with adjustment for patients' weight is shown in Table 4. Results allow us to make the following prediction. PAPP-A MoM value for a smoking woman was on average 16.0% $((1-10^{-0.076})\times100\%)$ lower than for a non-smoking woman with the same weight, whereas 1kg increase in weight caused on average 1.8% $((1-10^{-0.008})\times100\%)$ decrease of PAPP-A MoM value with the standard error of 0.1%.

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