ECRONICON OPEN ACCESS

EC PAEDIATRICS
Research Article

Endothelial Dysfunction during Pregnancy: Prediction of Child Morbidity

Mironov Alexey Valentinovich[1,2]*, Thapar N[3], Torchinov AM[1], Umahanova MM[1] and Galachiev OV[1]

[1]*A.I. Evdokimov Moscow State University of Medicine and Dentistry, Moscow, Russia*

[2]*City Clinical Hospital №13, Moscow, Russia*

[3]*Kamlesh Medical Center, Delhi, India*

***Corresponding Author**: Mironov Alexey Valentinovich, Assistant, A.I. Evdokimov Moscow State University of Medicine and Dentistry, Moscow, Russia.

Received: July 10, 2018; **Published:** September 28, 2018

Abstract

Objective: To study the influence of endothelial dysfunction, during the first trimester of pregnancy, on the health of neonates and infants by using cytometric studies of desquamated endothelial cells in the peripheral blood.

Material and Methods: The study included 163 pregnant women with single fetuses. Endothelial dysfunction was assessed in mothers in their first trimester, on the basis of cytometric studies of desquamated endothelial cells in the peripheral blood. Physical health of patients (i.e. mothers) was assessed. The status of 163 newborns and their catamnesis in the first year was also assessed.

Results: In patients with endothelial dysfunction detected in 1st trimester of pregnancy there was reduction in average diameter of desquamated endothelial cells. The children of these mothers more frequently encountered disorders of the Central nervous system and higher frequency of psychomotor developmental disorders. There was also a significant increase incidence of infections, inflammatory diseases and iron deficiency anemia in these kids.

Discussion: Endothelial dysfunction in patients with early 1st trimester pregnancy causes formation of an impaired utero-fetal relationship, which reflects in the formation of organ and systems in the fetus and leads to disorders in neonates and children of early pediatric age group.

Conclusion: Endothelial dysfunction is of huge importance and is consistent in defining somatic health of a newborn child. The cytometric analysis of endotheliocytes of peripheral blood of patients in their first trimester of pregnancy can be used as an informative test in early diagnosis of an increase in incidence of neonatal and early pediatric diseases.

Keywords: *Endothelial Dysfunction; Pregnancy; Infant Morbidity*

Introduction

Complete implantation and transformation of spiral arteries with formation of adequate utero-placental flow are the main conditions for successful development of pregnancy [1,2]. In first weeks of pregnancy, chorionic villi form a system of utero-placental vessels which help in forming a basis of circulation of blood to the fetus. It is important that this hemodynamic system provides sufficient amount of placenta-fetal flow and thus adequate nutrition for the fetus throughout the pregnancy [3,4].

It has been proved that any disturbance in the process of trophoblast invasion in the first trimester of pregnancy can lead to late gestational complications: preeclampsia, premature birth and may result in perinatal death of fetus and/or increased infantile and maternal mortality. The complications in a pregnancy have direct effect on formation of the organs and systems in a fetus.

In last few years a lot of research has been done in the field of endothelium as a main factor responsible in maintaining homeostasis and fetoplacental micro and macro circulation [1,5]. Endothelial dysfunction is followed by failure of compensatory opportunities of endotheliocytes and initiation of pathological processes in vascular system [6]. Many authors studied a role of markers of violation of endotheliocytes at cardiovascular diseases, a diabetes mellitus, cerebral infarction, a metabolic syndrome, renal pathology [7-13]. Early detection of vascular pathology is decisive in prophylaxis of obstetric and perinatal pathology [14]. In literature we found a lot of material about a role of endothelial dysfunction in pathogenesis of miscarriage [15,16] and eclampsia [12,14,17-21]. It has been proved that there is an increase in frequency of intrauterine growth restriction in the fetuses of mothers having polymorphism of NO-synthase (NOS) [22]. Complicated gestations lead to preeclampsia, chronic placental insufficiency further leading to developmental delays in fetuses, reduced growth and decreased synthesis of endogenous nitrogen oxide with activation of angiogenic growth factors (PlGF, VEGF etc.) [23]. Change in the level of NO and endothelin-1 is associated with development of cerebral ischemia and cardiovascular diseases in neonates [9]. Amount of total nitrite and endothelin-1 in umbilical cord blood in neonates of patients having arterial hypertension and positive endothelial dysfunction directly correlates with features of chronic hypoxia in a fetus, development of intrauterine growth restriction, intraventricular hemorrhages and patent ductus arteriosus in newborns at 3-month age [24]. The analysis of endothelial dysfunction in mothers is useful in prognosticating the health of newborns, to study possible risk of perinatal complications and postnatal morbidity and mortality.

One of methods of diagnosing endothelial dysfunction is by counting the circulating endotheliocytes of peripheral blood. For the first time, existence of the circulating desquamated endotheliocytes was described in 1978 by J Hlodavec. He observed a phenomenon of endotheliopathy in rats after infusing an endotoxin hyaluronidase, streptokinase and some vasoactive medicines [25]. Cytological analysis of endotheliocytes shows damage of vascular system. The author connected this increase in quantity of the circulating desquamated endotheliocytes with increased tendency of thrombosis and hyperpermeability of the vascular wall. Endothelial cells measuring 1 - 2 micrometers thick and a diameter of 10 - 50 microns have a flat surface, an extended core and centrally located nucleus and are characterized by marked morphological heterogeneity [26]. Endothelial cells get detached from the wall of a vessel either completely or in smaller cell fragments. The circulating endotheliocytes can be present in peripheral blood as apoptotic cells which can be distinguished by staining, as in apoptosis the plasma membrane is damaged [8]. Death of endotheliocytes is mediated by apoptosis [27]. The morphological picture of apoptosis includes blebbing, cell shrinkage, nuclear fragmentation, chromatin condensation, chromosomal DNA fragmentation and mRNA decay into membranous vesicles. Apoptosis produces cell fragments called apoptotic bodies which are later engulfed by phagocytic cells [28]. We can study these apoptotic cells using simple light microscopy after staining. Studying the optical properties of circulating endotheliocytes opens new windows to early diagnosis of diseases of the endothelial system.

Objective

To study the influence of endothelial dysfunction, during the first trimester of pregnancy, on neonatal health using cytometric studies of desquamated endothelial cells in peripheral blood of mothers.

Material and Methods

The study included 163 patients with single fetus. Peripheral blood of pregnant women in their first trimester was analyzed and cytometric assessment was done for endothelial dysfunction in desquamated endotheliocytes (DEC). Cytometry of the apoptotic cells revealed change in geometric parameters of DEC. when there is a rise in endothelial dysfunction majority of cells are noted in the blebbing stage and apoptotic cell stage which have a reduced diameter than endotheliocytes. The same technique as used by J Hladovec [25] in 1978, was used to selectively separate DEC. The principle of this method is based on selective separation of DEC along with thrombocytes and later on removal of thrombocytes with the help of adenosine diphosphate. 4 - 5 ml peripheral venous blood is taken and stabilized with 3.8% sodium citrate. Thrombocytes are separated using ADP solution and then centrifuged. The plasma that has separated from thrombocytes is then carefully removed and centrifuged again. The sediments are then separated again and 0.1 ml 0.9% NaCl was added. Then computerized cytometry of DEC was performed. Leika DM 1000 microscope was used with computer video system for processing and then

images were analysed using Leika Application Suite LAZ EZ Version 2.1.0. (2012). Then mean diameter of circulating DEC was calculated.

According to the final data, the contingent was divided into 3 groups. The first group was of 74 women where in the first trimester of pregnancy mean diameter of DEC of peripheral blood was more than 40 microns (Figure 1).

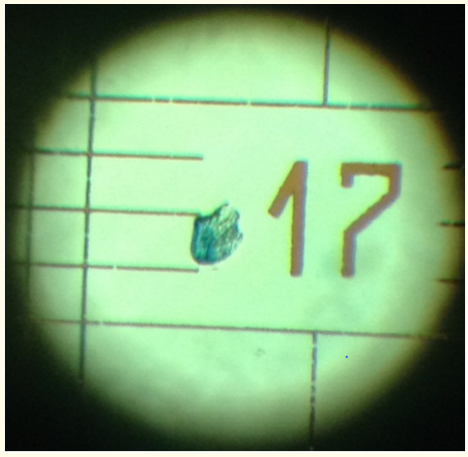

Figure 1: *Desquamated endotheliocyte (mean diameter of 48 microns). Coloring the methylene blue (x600).*

45 patients had a mean diameter of 30 - 40 microns (Figure 2), which formed the second group.

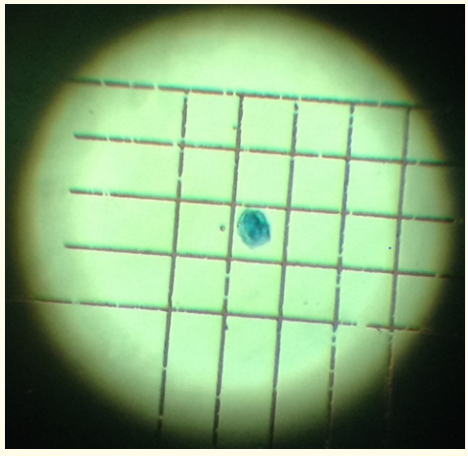

Figure 2: *Desquamated endotheliocyte (effective diameter of 36 microns). Coloring the methylene blue (x600).*

44 patients that had diameter less than 30 microns (Figure 3) entered the third group.

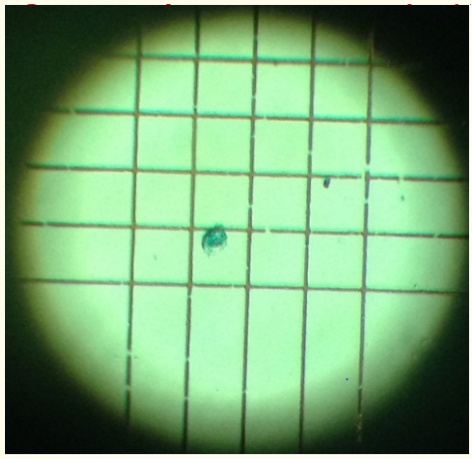

Figure 3: *Desquamated endotheliocyte (effective diameter less than 22 microns). Coloring the methylene blue (x600).*

The following parameters were considered: age, profession related adversities and addictions, extragenital, obstetrical and gynecological anamnesis, physical condition of the pregnant women. Women at whom chronic diseases in an aggravation stage are revealed were excluded. Diabetes mellitus and hypertension are themselves responsible for endothelial dysfunction, so they were exclusion criteria.

163 patients with single fetuses in whom pregnancy reached full term were studied. Considering higher incidence of perinatal pathology and neonatal morbidity, born before 37 weeks of gestation was exclusion criteria. Then the condition of 163 newborn children and their follow up till one year was assessed.

Results were statistically analyzed. Statistical processing was carried out using Excel (Microsoft Office of Excel 2003) and Statistica (for Windows release 6.0 of the StatSoft.Inc company., 2002). Calculations were carried out by Student's t-tests.

Results and Discussion

Average age of women of the first group was 26.02 ± 5.2 years, the second group - 28.24 ± 5.3 years and the third group was - 26.3 ± 4.6 years. In the studied groups, addictions and professional adversities were not significantly different, statistically. Obesity was most common in the second group - 26.7%, in the first group it was 13.5% and in the third group - 13.6%. Cardiovascular diseases were found prenatal assessment in 28.4% of women in the first group, 37.8% of women in the second group, and in 29.5% - in the third group. Diabetes mellitus and hypertension are themselves responsible for endothelial dysfunction, so they were excluded. All main cardiovascular risk factors are associated with dysfunction of an endothelium: obesity, hyperlipidemia, essential hypertension, diabetes mellitus [29,30]. At the diseases interfaced to atherosclerotic defeat of vessels, the endothelium represents a target organ as endotheliocytes regulate a vascular tone and a hemostasis. Increase in a vasoconstriction, decrease in an elastance of vessels, progressing of an atherothrombosis form a chronic ischemia of tissue which is an inductor of an apoptosis of endotheliocytes, initiating their destruction [31]. Thus, on major factors of risk of development of endothelial dysfunction the studied groups statistically were comparable. Diseases of the urinary system were more common in the third group - 25%, in the first and second - 14.9% and 17.8% respectively. Gastro intestinal diseases: 5.4%

in the first group, 11.1% in the second group, 2.3% pregnant women in the third group. Thyroid gland pathologies were found in the frequency of 18.9% - in the first group, 20% in the second group and 13.6% in the third group. Somatic health of pregnant women in the studied groups was insignificant.

At a research of health of children in the considered groups the following results were received (Figure 4). It should be noted that in literature influence of endothelial dysfunction at mother on a condition of newborns is described. Identification of markers of endothelial dysfunction at mother correlates with a chronic hypoxia of a fruit, violation of a fetation [22,24]. Endothelial involvement is associated with the development of cerebral ischemia in newborns, cardiovascular disorders [9].

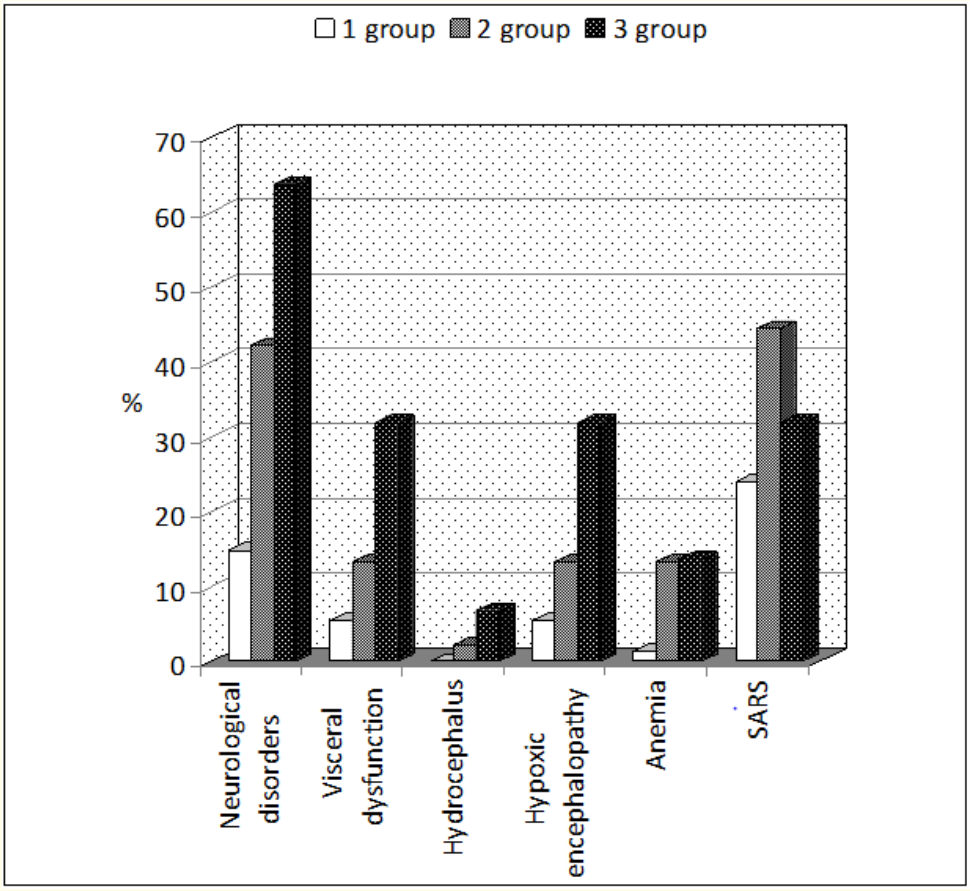

Figure 4: *The chart of frequency of infantile and children's incidence in the studied groups.*

In our research in children of the second and third group, diseases of the central nervous system (CNS) were 1.5 - 3 times more common in the first year. Residual phenomena of neurological disorders was found in the second and third group, only in the second year. Neurological disorders, till 1 year of age were diagnosed in 14.9% of kids in the first group. In group with mild degree of endothelial dysfunction in the first trimester - in 42.2% ($p \leq 0.05$; $t = 3.2$) and in the third group - in 63.6% kids (after full term birth) ($p \leq 0.001$; $t = 6.3$). Neurological disorders were not found in first group after one year, in the second group - found in 6.7% of children, in the third - in 15.9% of children ($p \leq 0.05$; $t = 2.9$). Hydrocephalus was not found in a single child in the first group, 2.2% in the second group and in 6.8% of chil-

dren in the third group. Visceral dysfunction was diagnosed in 5,4% of children of the first group, 13.3% of children of second, and 27.3% of children in group with mothers having average to heavy degree of endotheliopathy ($p \leq 0.05$, $t = 3.04$). Hyperactivity disorder is diagnosed in 2.7% of children of the first group, 13.3% of children in the second group, and 27.3% of children of the third group ($p \leq 0.001$; $t = 3.5$). Frequency of hypoxic encephalopathy was 5.4% - in the first group, 13.3% - in the second group, and 31.8% - in the third group ($p \leq 0.001$; $t = 3.5$). Psychomotor lag in development was seen in 13.3% - in the second group, and 11.4% - in the third group and only in 2.7% in the first group. Frequency of hypochromic anemia in children till 1 year of age was also more in the second and third groups than in the first group. Hypochromic anemia in the first year of life is diagnosed in the first group in 1.4% of children, the second and third groups: 13.3% and 13.6% respectively. The incidence of SARS in the first year of life, in groups having endotheliopathy in mothers, was 1.5 times higher than in control group: in the first group was 1.9 times a year and 3.4 times a year - in the second group and 1.6 times - in the third group. The children who contracted SARS, 4 and more times in a year were- 24% in the first group, 44.4% - in the second group ($p < 0.05$; $t = 2.2$), and 31.8% - in the third group.

In the group of children, where mothers in the first trimester of pregnancy expressed endotheliopathy and thus also decrease in diameter of DEC had higher frequency of neuropathy and higher frequency of delayed psychomotor development. In these groups infectious and inflammatory diseases were also more often diagnosed. The incidence of hypochromic anemia increased. Endothelial dysfunction in early stages of gestation causes disruption of utero placental circulation and this affects the formation of organ systems in a fetus. This leads to development of pathologies in neonates and infants. Cytometry of DEC of peripheral blood opens new opportunities in early diagnosis of pathology in endothelial system. Identification of endotheliopathy in early antenatal periods is an important marker in determining possible perinatal complications [32].

Conclusion

Endothelial dysfunction, being a pathogenetic link of many diseases, is of key value in development of pathology in the fetoplacental complex, defining the physical health of a newborn. The cytometric research of DEC in peripheral blood of a mother in the first trimester of pregnancy can be used in medicine as an informative test in early diagnosis of neonatal and infant health.

Bibliography

1. Bicadze VO., *et al*. "Thrombophilia as a key link in the pathogenesis of pregnancy complications". *Practical Medicine* 60.5 (2012): 22-29.

2. Serov VN. "Manual of outpatient care in obstetrics and gynecology". M.: GEOTAR-Media (2016).

3. Bashiri A., *et al*. "Oligohydramnios associated with a long umbilical cord". *International Journal of Case Reports and Images* 2.1 (2011): 15-17.

4. Nevzorova IA., *et al*. "Markers of endothelial damage in pregnant women with threat of miscarriage". V Russian Congress "Outpatient care at the heart of women's health". Moscow, Russia (2013): 53-55.

5. Koshuhov RH., *et al*. "Diagnostic and prognostic significance of microalbuminuria in pregnant women at risk for preeclampsia". *Vestnik RGMU* 4 (2009): 51-56.

6. Volkova EV and Kopylova YV. "The role of vascular growth factors in the pathogenesis of placental insufficiency". *Obstetrics, Gynecology and Reproduction* 7.2 (2013): 29-33.

7. Sushkina IF., *et al*. "Comparative assessment of a condition of a microhemodynamic and vasculomotor function of an endothelium at patients of an ischemic heart disease and almost healthy faces of a different age". *Vestnik VolgGMU* 2.50 (2014): 63-65.

8. Feoktistova VS., *et al*. "New approach to endothelium dysfunction assessment: determination of quantity of the circulating endothelial cells by method of flowing cytometry". *Clinical Laboratory Diagnostics* 60.4 (2015): 23-27.

9. Chistyakova GN., *et al*. "Assessment of a condition of cardiovascular system of the children who were born from women with chronic arterial hypertension". *Pediatrics* 94.2 (2015): 8-12.

10. Fadini GP and Avogaro A. "Cell-based methods for ex vivo evaluation of human endothelial biology". *Cardiovascular Research* 87.1 (2010): 12-21.

11. Jim B., *et al*. "Hypertension in pregnancy: a comprehensive update". *Cardiology in Review* 18.4 (2010): 178-189.

12. Karthikeyan VJ and Lip GY. "Endothelial damage/dysfunction and hypertension in pregnancy". *Frontiers in Bioscience* 3 (2011): 1100-1108.

13. Lampka M., *et al*. "Circulating endothelial cells in coronary artery disease". *Kardiologia Polska* 68.10 (2010): 1100-1105.

14. Moiseeva IV and Mansur-Hassan SH. "The functional indexes of an endothelium in dynamics of physiological pregnancy at women (according to jet test of a humeral artery)". *Aspirantskij vestnik Povolzh'ya* 3-4 (2015): 78-82 .

15. Ajeni Daniel' Olusola and Ture Pengo Ehl'vira DF. "Feature of ednotelialny function of vessels of a uterus at women with early reproductive losses". IV international scientific confenertion "Science 4 health". Moscow, Russia (2012): 20.

16. Kovalenko AE., *et al*. "Biochemical markers of the menacing premature births". *Obstetrics and Gynecology* 3 (2013): 43-47.

17. Medvedev BI., *et al*. "Predictors of development of a preeclampsia". *Obstetrics and Gynecology* 5 (2013): 30-35.

18. Sidorova IS and Nikitina NA. "Features of a pathogenesis of an endotheliosis at a preeclampsia". *Obstetrics and Gynecology* 1 (2015): 72-78.

19. Kenny LC., *et al*. "Screening for Pregnancy Endpoints Consortium. Robust early pregnancy prediction of later preeclampsia using metabolomic biomarkers". *Hypertension* 56.4 (2010): 741-749.

20. Saleem Alkasiy K., *et al*. "Electron microscopic study of the effect of preeclampsia on the placental endothelial cells ultra structures during pregnancy". *Al-Kindy College Medical Journal* 6.1 (2010): 39-44.

21. Ziganshina MM., *et al*. "Autoantibodies to endothelium antigens at a preeclampsia". *Obstetrics and Gynecology* 3 (2016): 24-31.

22. Lanir N., *et al*. "Haemostatic mechanisms in human placenta". *Best Practice and Research: Clinical Haematology* 16.2 (2003): 183-195.

23. Bashmakova NV., *et al*. "Markers of endothelial dysfunction abnormal uterine bleeding of puberty in girls born with intrauterine growth restriction". *Rossijskij Vestnik Akushera-Ginekologa* 15.2 (2015): 19-24.

24. Chistyakova GN., *et al*. "Assessment of the cardiovascular system of children born to women with chronic hypertension". Pediatriya. ZHurnal imeni G.N. Speranskogo 94.2 (2015): 8-12.

25. Mihalenko IV and Mihalev EV. "Humoral indicators of the system state of the vascular endothelium and hemostasis in premature infants with hypoxic lesions of the central nervous system". *Rossijskij Vestnik Perinatologii i Pediatrii* 2 (2014): 61-65.

26. Hladovec J. "Circulating endothelial cells in acute myocardial infarction and angina pectoris". *Wiener klinische Wochenschrift* 56.20 (1978): 1033-1036.

27. Rowand JL., *et al*. "Endothelial Cells in Peripheral Blood of Healthy Subjects and Patients with Metastatic Carcinomas". *Cytometrya* 71.2 (2007): 105-113.

28. Gerilovich LA., *et al*. "Endothelin-1 in patients with various forms of infertility in programs of auxiliary reproductive technologies". *Medicina i obrazovanie v Sibiri* 6 (2013): 19.

29. Mnihovich MV. "Evaluation of apoptosis in breast cancer according to study polutorka sections and electron microscopy". *Morfologiya* 5.2 (2011): 45-48.

30. Astashev PE and Gudymovich VG. "Markers of endothelial dysfunction as prognostic sign of a course of endocardiac infectious process". Vestnik Nacional'nogo mediko-hirurgicheskogo Centra im. N.I. Pirogova 6.3 (2011): 3-8.

31. Zagorodskih EB., *et al*. "Markers of endothelial dysfunction and their prognostic value at sharp pancreatitis of a heavy current". *Fundametal'nye issledovaniya* 9 (2013): 355-361.

32. Barsuk AL., *et al*. "Regulatory role of an endothelium and some aspects of influence of a pharmacotherapy on its function". *The Modern Technologies in Medicine* 3 (2011): 142-146.

Volume 7 Issue 8 August 2018