EFFECT OF MATERNAL OBESITY AND PASSIVE SMOKING ON NEONATAL NUCLEATED RED BLOOD CELLS

Abeer Mohamed Nour ElDin Abd ElBaky* **and Tarek Mohamad Farid El Shaer**

National Research Centre, Egypt

Amany Ezat Elwan†

Cairo University, Egypt

Hanaa Rasmy‡

National Research Centre, Egypt

Abstract: Passive cigarette smoking and obesity during pregnancy are risk factors for adverse outcome in infant. Elevated umbilical cord neonatal nucleated red blood cells (NRBCs) have been suggested as a marker of intrauterine foetal hypoxia. *Aim to* demonstrate whether maternal risk factors during pregnancy are capable of elevating circulating NRBCs measured at birth. We compared the count of NRBCs in the cord blood in three groups. Group I neonates born to obese mothers, group I) neonates born to mothers exposed to tobacco smoke during pregnancy and control group III. The results reveal that maternal body mass index and infant birth weight were significantly higher in group I ($p = < 0.001$ and 0.037, respectively). The absolute NRBC count was higher in groups I and II compared to control group ($p = 0.02$ and 0.01, respectively). In conclusion, the neonates of obese mothers and passive maternal smoking have increased NRBCs at birth.

Keywords: NRBCs; nucleated red blood cells; cigarette smoking; obesity; pregnancy.

INTRODUCTION

Nucleated red blood cells (NRBCs) are immature erythrocytes that are found in the peripheral blood of healthy newborn infants (Miller and Baehner, 1995). Immediately after birth, a rapid decline in the number of haematopoietic progenitor cells is observed commonly in the healthy neonate (Sills and Hadley, 1983). Elevated umbilical cord NRBC counts have been suggested both as a marker of acute and chronic intrauterine foetal hypoxia and as a predictor of adverse

*Corresponding author: Department of Pediatric, National Research Centre, Cairo, Egypt; e-mail: abeernour66@yahoo.com
†Department of Pediatric, Faculty of Medicine, Cairo University, Cairo, Egypt; e-mail: amanyelwan@hotmail.com
‡Department of Clinical Pathology, National Research Centre, Cairo, Egypt; e-mail: hanarasmy2000@yahoo.com

neonatal outcomes (Blackwell et al., 2000; Buonocore et al., 1999; Hanlon-Lundberg and Kirby, 1999; Phelan et al., 1995).

The identification of prognostic markers for adverse perinatal outcome is a major focus of modern foetal and neonatal medicine. NRBCs are among the markers that have been studied, those that reflect compromised metabolic status during the transition from foetal to neonatal life. The perinatal and long-term liabilities of foetal growth restriction are due to the combination of adverse intrauterine environment, peripartum events and post-delivery complications that are manifested in many organ systems (Baschat et al., 2007).

The prevalence of obesity in pregnant women, a known risk factor of the reproductive cycle, has increased significantly over the last few years (Atalah and Castro, 2004). Maternal obesity at conception alters gestational metabolic adjustments and affects placental, embryonic, foetal growth and development. Neural tube defects and other developmental anomalies are more common in infants born to obese women; these defects have been linked to poor glycemic control. Pre-eclampsia, a gestational disorder which occurs more frequently in obese women appears to be due to a subclinical inflammatory state that impairs early placentation and development of its blood supply. Foetal growth and development during the last half of pregnancy depends on maternal metabolic adjustments detected by placental hormones and the subsequent oxygen and nutrient supply (King, 2006).

Cigarette smoking during pregnancy is a known risk factor for adverse outcome in the human foetus and infant; there is a warning on cigarette smoking effects during pregnancy. Maternal smoking significantly increases the risks of spontaneous abortion and of pre-term or low birth weight delivery (Windham et al., 1992). Also, it is capable of causing foetal growth restriction in the third trimester (Lieberman et al., 1994) and is associated with increased neonatal morbidity, such as neonatal asphyxia (Wright and Catz, 1998), intraventricular haemorrhage (Spinillo et al., 1991), reduced lung function and increased incidence of perinatal death as well as sudden infant death syndrome (Hoffman et al., 1988). Maternal cigarette smoking during pregnancy also increases the risk of neurodevelopmental impairment later in childhood (Olds et al., 1994). Although, the mechanism of foetal injury seems to involve many factors, it is likely that chronic foetal hypoxia affects the process (Windham et al., 1992).

The aim of our study was to demonstrate whether maternal obesity and passive exposure to cigarette smoking during pregnancy are capable of elevating the absolute number of circulating NRBCs measured at birth.

MATERIAL AND METHODS

Our study is a case control study of full term born infants (38–41 weeks of gestation by last menstrual period, confirmed by early ultrasound), appropriate for gestational age. All were born vaginally at three private hospitals in the period between January 1, 2008 and March 31, 2008. They were divided into three groups; group I consisted of 29 infants of obese mothers; body mass index was calculated according to equation (BMI = Wt/Ht^2), weight and height of the mothers taken retrospectively from files in antenatal care clinics, only data of the first visit in the first trimester was taken and we excluded infant of mothers visit the clinic for the first time after the first trimester.

Group II consisted of 21 infants of non-smoking mothers who were exposed to passive smoking during pregnancy (at home or

at the workplace or both locations, more than 10 cigarettes/day for a period of more than three months) and Group III consisted of 15 control infants of mothers neither exposed to passive smoking nor obese mothers. We excluded infants born to women with gestational or insulin-dependent diabetes; pregnancy-induced hypertension; placental abnormalities; any maternal heart, kidney, lung or other chronic condition and perinatal infections (e.g. fever, leukocytosis and signs of chorioamnionitis). Informed consents were taken from the parents of neonates according to the guidelines of the ethical committee of National Research Center, Dokki, Egypt. Venous blood samples for complete blood cell counts were collected from the infant within 12 hr of birth and analysed according to laboratory routine using Cell-Dyn 1600 Abbott counter. Differential cell counts were performed manually and NRBC counts were expressed per 100 white blood cells (WBCs). We calculated the number of NRBCs as an absolute number rather than per 100 leukocytes, and the WBCs' count was expressed as corrected for the presence of NRBCs.

Table 1 Demographic data of the different groups

Parameters	Obese group (I) (n = 29)	Passive smoker group (II) (n = 21)	Control group (III) (n = 15)
Birth weight (gm)	3395 ± 534*	3195 ± 493	3137 ± 258
Maternal BMI	32.73 ± 5.12	27.24 ± 3	25.22 ± 1.92
Gender(males : females)	15 : 14	11 : 10	7 : 8
1-min Apgar score	7–9	7–9	7–9
5-min Apgar score	8–10	9–10	9–10

*p = 0.037.

Table 2 Haematological data of the different groups

Parameters	Obese group (I) (n = 29)	Passive smoker group (II) (n = 21)	Control group (III) (n = 15)
Haematocrit (%)	40.6 ± 5	39.4 ± 6.7	37.5 ± 9
RBCs (X109/L)	4.21 ± 0.57	4.05 ± 0.71	3.92 ± 0.97
White blood cells(corrected) (X109/L)	11.26 ± 4.39	10.33 ± 3.64	12.14 ± 5.24
Lymphocytes (X109/L)	38.07 ± 11	43.95 ± 12.34	37.48 ± 9.04
Platelets (X109/L)	185 ± 69	182 ± 71	171 ± 97
Absolute nucleated RBCs(X109/L)	(0–2220) 742*	(0–1548) 723**	(0–1312) 374

*p = 0. 02; **p = 0.01.

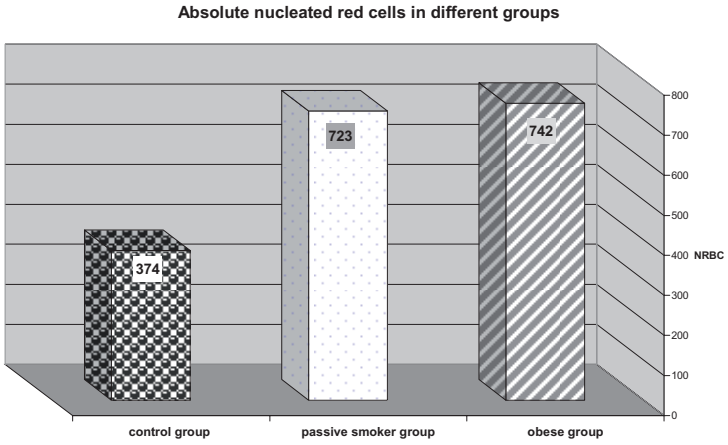

Figure 1 Absolute neonatal nucleated red blood cells in different groups

STATISTICAL ANALYSIS

SPSS for Windows, version 10.0 computer program was used for statistical analysis. Data were represented as frequency, percent, range and mean ± SD. The *t*-test was used to compare between two independent means.

A *p*-value of less than 0.05 was considered statistically significant.

RESULTS

Tables 1 and 2 showed some descriptive and haematological data of the studied cases and controls, there were no significant differences between group I and controls in terms of gender, RBCs, haemoglobin, haematocrit, platelets, corrected WBCs and lymphocytes. There was statistically significant increase in baby birth weight in obese group I compared to controls ($p = 0.037$). The absolute NRBCs count was statistically significant higher in obese group than in controls ($p = 0.02$).

Regarding passive smoker group (group II), there were no significant differences between them and controls in gender, RBCs, haemoglobin, haematocrit, platelets, corrected WBCs, lymphocytes and baby birth weight. The absolute NRBC count was statistically significantly higher in the passive smoker group than in controls ($p = 0.01$).

Figure 1 showed the absolute NRBCs count in different groups.

DISCUSSION

Foetal hypoxemia can trigger erythropoietin release that causes stimulation of RBCs, both at intramedullary and extramedullary sites. In the human foetus, erythropoiesis typically progresses from erythroid commitment of colony forming stem cells to extrusion of nuclear material with concomitant reduction in cell size (Maier et al., 1994). This process yields a mature RBC without a nucleus that contains the highest concentration of haemoglobin. Typically, early stages of mature erythropoiesis are confined to the bone marrow, where capillary fenestrations limit the passage of large NRBC precursors into the peripheral circulation. Conversely, extramedullary sites are believed to have larger capillary fenestration that permits the release of large NRBCs. During periods of high extramedullary production, NRBC

counts of up to 30/100 WBCs are physiologic at 30 weeks of gestation, although levels of 5–10/100 are normal thereafter (Moritz et al., 1997).

The increasing prevalence of obesity in young women is a major public health concern. These trends have a major impact on pregnancy outcomes in these women, which have been documented by several researchers (Bhattacharya et al., 2007). In our study, the baby birth weight of obese mothers was statically, significantly, higher than controls ($p = 0.037$), this is in agreement with Ducarme et al. (2007), who reported that birth weight of children was significantly higher in obese patients (3, 305 vs. 3,181 g; $p < 0.01$) with no impact on Apgar score. The absolute NRBCs were statically, significantly, higher in infants of obese women than in controls ($p = 0.02$), which was the same result of infants of Sheffer-Mimouni et al. (2007), who found that overweight and obese mothers have increased NRBCs at birth compared with controls ($p = 0.01$). So they speculated that even apparently healthy foetuses of overweight and obese mothers are exposed to a subtle hypoxemic environment. The perinatal outcome of the infant of obese mother is adversely affected (Sheffer-Mimouni et al., 2007).

Maternal obesity affects these metabolic adjustments as well. Basal metabolic rates are significantly higher in obese women, and maternal fat gain is lower, possibly in response to altered leptin function. The usual increase in insulin resistance seen in late pregnancy is enhanced in obese mothers, causing marked post-prandial increases in glucose, lipids and amino acids and excessive foetal exposure to fuel sources, which in turn increases foetal size, fat stores and risk for disease post-natally (King, 2006).

The absolute NRBCs in infants of mothers exposed to passive smoking in our study was statically significantly higher than controls ($p = 0.01$), which was the same result reported by Dollberg et al. (2000). Also other study showed that infants born to smoking mothers have increased circulating absolute NRBC counts compared to controls (Yeruhimovich et al., 1999).

The mechanism by which maternal smoking increases circulating neonatal absolute NRBC counts is unknown. A likely explanation is relative foetal hypoxia, but the mechanisms that may be involved are multiple. In theory, there may be nicotine-induced placental vasoconstriction (Morrow et al., 1988), decreased foetal tissue oxygenation attributable to production of foetal carboxyhaemoglobin and placental vascular disease (Gupta et al., 1993). Other indicators of foetal hypoxia during maternal smoking include foetal growth restriction, increased risk of spontaneous abortion and neurodevelopmental anomalies that occur more frequently among infants of smoking mothers (Lieberman et al., 1994). Dollberg et al. (2000) said that passive smoking affects NRBC in a manner similar to active smoking.

Growth-restricted neonates have significantly higher NRBC counts than adequately grown counterparts, but elevated NRBC counts and NRBC persistence also correlated with short- and long-term outcomes (Baschat et al., 2003). Baschat et al. (2007) said that the NRBCs were needed further investigations to define the dynamics of NRBCs regulations as neonatal life progresses.

A limitation of our study is that we cannot exclude the possibility that some infants in the control group were exposed to nicotine or to other sources of tobacco smoke or carbon monoxide not on a daily routine base. Despite these limitations, the absolute NRBC count of infants exposed to maternal passive smoking was significantly higher than that of non-exposed controls.

We believe that our data give further support for health hazards associated with some maternal risk factors like maternal obesity and exposure to passive smoking; we speculate that even apparently healthy neonates of obese mothers and passive smoker mothers were exposed to a subtle hypoxemic environment. So, we recommended effort to control maternal obesity and trying prevention of exposure of pregnant mothers to passive smoking whenever possible and to insist on strict enforcement of smoking regulations at the workplace and at home for well-being of our infants, also we recommend close observation for those neonates as regard their neurodevelopmental progress.

BIOGRAPHY

Dr Abeer Mohamed Nour Eldin AbdElBaky is a Research Associate Professor of Pediatrics at Pediatrics Department-NRC. She focused her research activities on different Pediatrics Fields. She had more than 14 local and international publications in hepatology, gasteroentrology, endocrinology and renal fields of Pediatrics.

Tarek Mohamad Farid El Shaer is a Researcher of Pediatrics at Pediatrics Department-NRC. He focused his research activities on different Pediatrics Fields. He had more than 6 local and international publications in neurology, neonatology, endocrinology and renal fields of Pediatrics. He participate in many national and international Congresses.

Dr Hanaa Rasmy Mohamed Attia is a Research Professor of Clinical and Chemical Pathology at the Clinical and Chemical pathology Department-NRC. Editor for training program book in Clinical and Chemical Pathology. She focused her research activities on Immunological determinants of different diseases.

REFERENCES

Atalah, E. and Castro, R. (2004) 'Maternal obesity and reproductive risk', *Revista médica de Chile*, Vol. 132, No. 8, pp.923–930.

Baschat, A.A., Gembruch, U., Reiss, I., Gortner, L. and Harman, C.R. (2003) 'Neonatal nucleated red blood cell count and postpartum complications in growth restricted fetuses', *Journal of Perinatal Medicine*, Vol. 31, pp.323–329.

Baschat, A.A., Gungor, S., Kush, M.L., Berg, C., Gembruch, U. and Harman, C.R. (2007) 'Nucleated red blood cell counts in the first week of life: a critical appraisal of relationships with perinatal outcome in preterm growth-restricted neonates', *American Journal of Obstetrics and Gynecology*, Vol. 197, pp.286.e1–286.e8.

Bhattacharya, S., Campbell, D.M., Liston, W.A. and Bhattacharya, S. (2007) 'Effect of Body Mass Index on pregnancy outcomes in nulliparous women delivering singleton babies', *BMC Public Health*, Vol. 7, No. 147, p.168.

Blackwell, S.C., et al., (2000) 'The relationship between nucleated red blood cell counts and early-onset neonatal seizures', *American Journal of Obstetrics and Gynecology*, Vol. 182, pp.1452–1457.

Buonocore, G., et al., (1999) 'Nucleated red blood cell count at birth as an index of perinatal brain damage', *American Journal of Obstetrics and Gynecology*, Vol. 181, pp.1500–1505.

Dollberg, S., Fainaru, O., Mimouni, F.B. and Shenhav, M. (2000) 'Effect of passive smoking in pregnancy on neonatal nucleated red blood cells', *Pediatrics*, Vol. 106, p.e34.

Ducarme, G., Rodrigues, A., Aissaoui, F., Davitian, C., Pharisien, I. and Uzan, M. (2007) 'Pregnancy in obese patients: which risks is it necessary to fear'? *Gynécologie, Obstétrique and Fertilité*, Vol. 35, No. 1, pp.19–24.

Gupta, I., Hillier, V.F. and Edwards, J.M. (1993) 'Multiple vascular profiles in the umbilical cord; an indication of maternal smoking habits and intrauterine distress', *Placenta*, Vol. 14, pp.117–123.

Hanlon-Lundberg, K.M. and Kirby, R.S. (1999) 'Nucleated red blood cells as a marker of acidemia in term neonates', *American Journal of Obstetrics and Gynecology*, Vol. 181, pp.196–201.

Hoffman, H.J., Damus, K., Hillman, L. and Krongrad, E. (1988) 'Risk factor for SIDS: results of the National Institute of Child Health and Human Development Cooperative epidemiological study', *Annals of the New York Academy of Sciences*, Vol. 533, pp.13–30.

King, J.C.(2006) 'Maternal obesity, metabolism and pregnancy outcomes', *Annual Review of Nutrition*, Vol. 26, pp.271–291.

Lieberman, E., Gremy, I., Lang, J.M. and Cohen, A.P. (1994) 'Low birth weight at term and the timing of fetal exposure to maternal smoking', *American Journal of Public Health*, Vol. 84, pp.1127–1131.

Maier, R.F., Gunther. A., Vogel, M., Dudenhausen, J.W. and Obladen, M. (1994) 'Umbilical venous erythropoietin and umbilical arterial pH in relation to morphologic placental abnormalities', *Obstetrics and Gynecology*, Vol. 84, pp.81–87.

Miller, D.R. and Baehner, R.L. (1995) *Blood Diseases of Infancy and Childhood*, 7th edition, St. Louis, MO: Mosby; pp.39–40.

Moritz, K.M, Lim, G.B. and Wintour, E.M. (1997) 'Developmental regulation of erythropoietin and erythropoiesis', *The American Journal of Physiology*, Vol. 273, pp.1829–1944.

Morrow, R.J., Ritchie, J.W. and Bull, S.B. (1988) 'Maternal cigarette smoking: the effects on umbilical and uterine blood flow velocity', *American Journal of Obstetrics and Gynecology*, Vol. 159, pp.1069–1071.

Olds, D.L., Henderson, C.R. and Tatelbaum, R. (1994) 'Intellectual impairment in children of women who smoke cigarettes during pregnancy', *Pediatrics*, Vol. 93, pp.221–227.

Phelan, J.P., Ahn, M.Y., Korst, L.M. and Martin, G.I. (1995) 'Nucleated red blood cells: a marker for fetal asphyxia?' *American Journal of Obstetrics and Gynecology*, Vol. 173, pp.1380–1384.

Sheffer-Mimouni, G., Mimouni, F.B., Dollberg, S., Mandel, D., Deutsch, V. and Littner Y. (2007) 'Neonatal nucleated red blood cells in infants of overweight and obese mothers', *Journal of the American College of Nutrition*, June, Vol. 26, No. 3, pp.259–263.

Sills, R.H. and Hadley, R.A.R. (1983) 'The significance of nucleated red blood cells in the peripheral blood of children', *The American Journal of Pediatric Hematology/Oncology*, Vol. 5, pp.173–177.

Spinillo, A., Ometto, A., Stronati, M., Piazzi, G., Iasci, A. and Rondini, G. (199l) 'Epidemiologic association between maternal smoking during pregnancy and intracranial hemorrhage in preterm infants', *The Journal of Pediatrics*, Vol. 127, pp.472–478.

Windham, G.C., Swan, S.H. and Fenster, L. (1992) 'Parental cigarette smoking and the risk of spontaneous abortion', *American Journal of Epidemiology*, Vol. 135, pp.1394–1403.

Wright, L.L. and Catz, C.S. (1998) 'Drug distribution during fetal life', In R.A. Polin and W.W. Fox (Eds.), *Fetal and Neonatal Physiology* (2nd Edn.). Philadelphia, PA: WB Saunders Co, pp.169–175.

Yeruhimovich, M., Dollberg, S., Green, D.W. and Mimouni, F.B. (1999) 'Nucleated red blood cells in term appropriate for gestational age infants of smoking mothers', *Obstetrics and Gynecology*, Vol. 93, pp.403–406.