Research Article

Oxidative stress during spontaneous vaginal delivery: comparison between maternal and neonatal oxidative status

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Abstract

Introduction: Spontaneous vaginal delivery inflicts a change in the redox status of the mother and the neonate. Hypoxia followed by reperfusion during powerful labour contractions causes increased generation of free radicals affecting the maternal oxidative status. The difference that exists between the extra uterine and intra uterine environment in the partial pressures of oxygen can induce free radical formation which affects the antioxidant mechanism of the neonate during vaginal delivery. The present study was aimed at quantifying the changes with respect to oxidative stress that occurs in uncomplicated laborin both the mother and newborn. **Method:** 20 singlet on term pregnant women in labour without any complications, delivered by spontaneous vaginal delivery were included in our study. Estimation of plasma MDA (Malondialdehyde) as a marker of lipid peroxidation and antioxidants like SOD (Superoxide dismutase), Total reduced Glutathione (GSH), and Glutathione Peroxidase (GPX) in hemolysate were performed using spectrophotometric method from cord blood and women in labour respectively. **Result:** The results showed significantly elevated plasma MDA in cord blood compared to maternal plasma MDA, before ($P<0.001$) and after delivery ($P<0.001$). Statistically significant higher GSH levels in the cord blood was observed compared to maternal GSH levels both before ($P <0.05$) and after delivery ($P< 0.05$). Maternal GPX level was significantly elevated when compared to cord ($P< 0.05$). However, SOD levels did not show any significant change. **Conclusion:** Vaginal delivery predisposes the neonate to increased oxidative stress when compared to maternal system which is reflected as an alteration in their antioxidant levels.

Keywords: Antioxidants, Lipid peroxidation, Oxidative stress, Labour

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Introduction

The role of oxidative stress is quite an important aspect in female reproduction. Pregnancy is a state of physiological stress due to increased energy demand by the growing fetus. As pregnancy advances the pattern of lipid peroxidation induced by oxidative stress increases and antioxidant enzymes also exhibits adaptation. At the time of delivery enhanced production of free radicals occurs which promotes oxidative stress in the maternal as well as neonatal system [1,2]. The presence of antioxidant system to counteract the deleterious effect produced by free radicals induced lipid peroxidation is beneficial to some extent. The extent of damage is determined by the balance between pro-oxidant and antioxidant system. Imbalance can create oxidative stress in both the maternal as well as fetal compartment, compromising the fetal outcome.

Antioxidants are substances capable of curtailing either the initiation or propagation of injury induced by free radicals. Enzymatic antioxidants include Superoxide dismutase (SOD), Catalase (CAT) and Glutathione Peroxidase (GPX) which are endogenous enzymes. Vitamin C and Vitamin E belong to non-enzymatic antioxidants which are obtained from exogenous dietary sources [3].

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Pregnancy can create a state of oxidative imbalance owing to higher metabolic demand. The pregnancy out come due to oxidative imbalance can pose serious complications such as PIH, Eclampsia, miscarriages, pre-term labour and IUGR [4, 5, 6]. Since pregnancy imposes high energy demand, evolution of maternal antioxidant system to combat any impending complications calls for specific attention. Accelerated production of reactive oxygen species occurs even in normal pregnancies due to higher metabolic demand as per previous reports [7]. There are also altered antioxidant defence mechanisms during normal uncomplicated pregnancies due to augmented oxidative stress [8].

Placenta acts as a connecting link between mother and foetus and also serves as a source for lipid peroxides. Placental antioxidant enzymes are capable of conferring protection to some extent [9]. Complications of pregnancy like PIHarise due to diminished placental antioxidant reserve [10].

Labour is a process during which over whelming oxidative imbalance can occur in the mother as a result of ischemia reperfusion injury produced by powerful phasic uterine contractions [11]. The fluctuations of oxygen tension also affect the neonate during the process of labour predisposing to increased oxidative stress [12]. The effectiveness of antioxidant mechanism in the neonate largely depends upon prevailing maternal antioxidant system. There are reports indicating transplacental transfer of antioxidants to help the neonate to tackle the situation at the end of gestation [13]. The maturation of antioxidant mechanism takes place at term in the neonate. Most of the preterm neonatal complications arise due to immature antioxidant defense mechanism. Bronchopulmonary dysplasias, hypoxic ischemic encephalopathy, IUGR in preterm neonates are caused due to weak and immature antioxidant system [14]. Administration of antioxidants in high risk mothers has established increased efficiency of antioxidant potential in the neonate [15]. As labour involves complex interplay of various factors, the fetal outcome remains questionable in case ofany intrapartum complications which can augment the oxidative stress. The efficiency of antioxidant mechanisms to curtail the increased generation of free radicals during labour in turn can be influenced by maternal factors like age, parity, nutritional status of the mother even in case of uncomplicated vaginal deliveries. There are also other parameters like environmental and genetic factors which can influence the oxidative imbalance in newborn in addition to fetal maturity. There are earlier reports regarding the imbalance between the antioxidant defence and oxidative stress in complications of pregnancy and during labour [16]. Scanty information and reports prevail pertaining to the antioxidant mechanisms and its adaptation during normal vaginal uncomplicated deliveries in neonate and mother. Hence, this study was planned in order to evaluate and compare the oxidative stress and antioxidant mechanisms in maternal and the neonatal system in uncomplicated spontaneous vaginal delivery.

Materials and Methods

This study was conducted after approval by institutional ethical committee on singleton term pregnant women in labour attending Sri Ramachandra Medical College and Research Institute. 20 pregnant women between 21-35 age group in term labour without any complications were included in our study (n= 20). They were delivered by spontaneous vaginal delivery. Pregnancy with maternal and fetal complications such as DM, HT, PIH, anemia, infections, PROM, abnormal lie, multiple gestations etc. were excluded. Patients delivered by caesarean section were also excluded.

Two samples containing 3 CC of blood were taken under strict aseptic conditions in EDTA containing sterile vacutainer tubes from women in active labour. Timely assessment of progress of labour was done. First sample was taken at the onset of active labour, with at least 4-5 cm dilatation of cervix. Second sample was taken from the mother after delivery of placenta. Cord blood was obtained after clamping immediately after delivery. Detailed confinement history with parameters like duration of labour, birth weight, sex of the baby, APGAR at 1 and 5 minutes, colour of the amniotic fluid were also taken.

RBCs were separated from plasma by centrifugation at 3000 r.p.m at 10^0 C for 30 minutes. Supernatant plasma was used for MDA estimation immediately. Malondialdhyde (MDA), a metabolite of lipid peroxidation detectable in plasma was used as the indicator. Plasma MDA concentrations were estimated as thiobarbituric acid reacting substances (TBARS) by the method of Cynamon et al, [17]. The lipid peroxidation activity was expressed as n mol/ 100 ml of plasma RBCs were washed 3 times with 0.9% ice cold sterile saline and centrifuged at same speed for 5 min

after each wash. Cells were lysed in 4 times volume of distilled water, allowed 1 hour for complete hemolysis. It was centrifuged and supernatant was stored in – 80^0 C until analysis was done. Antioxidant enzymes such as superoxide dismutase (SOD), glutathione peroxidase (GPX) and total reduced glutathione (GSH) were estimated in hemolysate at 37^0 c using ELICO SL 150 UV-VIS spectrophotometer.

The involvement of superoxide anion radical in the auto-oxidation of pyrogallol was used as a convenient assay of superoxide dismutase (Marklund and Marklund,) [18]. Total Reduced Glulathione (GSH) estimation was done by the method given by Moron et al [19]. Glutathione peroxidase activity was measured by its ability to utilize the standard glutathione in the presence of specific amount of hydrogen peroxide (1mM) (Rotrucket al.)[20]. Antioxidant activity was expressed as U/L.

Results

Description of the clinical characteristics of the study groups is shown in Table1. The mean age of the study group in years was 23.95 ± 3.83. The mean gestational age in weeks was 38.1± 1.27. The study group involved pregnant females from low socioeconomic status who underwent complete antenatal care. The statistical package used was SPSS for windows (version 11.5). Level of significance was kept at P<0.05.

Table-1: Clinical characteristics of maternal and neonatal parameters

Parameters	Mean ± SD
Age (years)	23.95 ±3.83
Parity	1.95 ± 0.94
Gestational age (weeks)	38.1± 1.27
Duration of labour (minutes)	289.6 ± 240.09
Birth weight (Kg)	2.979 ± 0.38
APGAR 1’	7.4 ± 1.39
APGAR 5’	8.8 ± 0.523

Table-2: Comparison between maternal and cord Plasma MDA and antioxidant levels

	Cord	Maternal	
MDA(n Mol/ 100 ml of Plasma)	0.815±0.06*†	Before	0.572± 0.07
		After	0.588± 0.09
GSH (U /L)	95.85 ± 12.6*†	Before	50.04 ± 7.85
		After	45.67 ± 6.26
SOD (U/L)	32.963±2.46	Before	35.45 ±2.26
		After	35.03 ± 2.4
GPX (U/L)	26.55 ± 3.34*†	Before	65.51± 11.16
		After	77.78 ± 8.94

Values are given as Mean ± SEM, *significant compared with Before delivery,† Significant compared with After delivery

Cord Plasma MDA concentration was significantly higher when compared to maternal plasma MDA from both pre delivery (Cord Vs Before, P< 0.001) and immediately post –delivery (Cord VsAfter,P< 0.001). However, there was no significant difference between plasma MDA of the mother before and after delivery (Table 2). We observed statistically significant higher GSH levels in the cord blood compared to maternal GSH levels both before (P <0.05) and after delivery (P< 0.05). There was no significant difference in SOD levels between maternal and cord blood. Cord GPX level was reduced when compared to maternal GPX levels before (P <0.05) and after delivery (P <0.05), which was statistically significant. Not much of significant statistical difference existed between maternal antioxidant enzymes before and after delivery.

Correlation of parameters like gestational age, parity, birth weight, APGAR, duration of labour and sex of the baby with antioxidants and MDA was not statistically significant. However, maternal GPX levels were positively correlated with maternal age (r= 0.016, P<0.05).

Discussion

Oxidative stress occurs during pregnancy which gets over whelmed during parturition. In uncomplicated pregnancy, the antioxidant system was reported to be more powerful than the peroxidation [21, 22]. The balance of pro-oxidant and antioxidant status can be disrupted due to complications of pregnancies and labour resulting in poor pregnancy outcome [23, 24]. The efficiency of antioxidant system is the key determinant of the level of oxidative stress.

We studied the effect of oxidative stress during labour in uncomplicated pregnancies by estimating lipid peroxidation product, MDA in maternal plasma and compared it with cord plasma MDA. Labour predisposes the neonate to increased oxidative stress as evidenced by higher plasma MDA than maternal plasma MDA levels which was statistically significant. Oxidative status of both mother and the neonate during spontaneous vaginal delivery is highly altered. Physiologically, it is an adaptation as the neonate is exposed to hyperoxic challenge during labour which can generate ROS [25]. Strong and phasic uterine contractions produce fluctuations in uterine blood flow inducing ischemia which is followed by reperfusion [26].This leads to generation of ROS in the mother which is detrimental if not removed by counter balancing antioxidant system. During parturition pro-inflammatory mediators like PGE_2, TNFα, IL6 evoke the production of ROS in a vicious manner [27]. Mode of delivery influences oxidative status of neonate according to studies which compared vaginal delivery with that of caesarean section [28]. Our observations clearly indicate perinatal oxidative stress as reflected by higher MDA in cord blood in addition to maternal oxidative stress which also affects the neonate as observed by other studies [29].

GSH level in cord was significantly elevated compared to maternal GSH in our study. Glutathione is a powerful antioxidant capable of rendering adequate antioxidant protection and regenerating other antioxidants like Vitamin E and Vitamin C. It has a substantial role in maintenance of pregnancy and prevention of oxidative stress in birth process [30]. Upregulation of GSH is

advantageous in situations of increased oxidative stress. The process of labour itself induces upregulation of GSH levels according to previous reports [31].

Precursors of glutathione like NAC (N-Acetyl Cyestine), have been used in animal models to increase antioxidant potential [32].

Maternal GPX levels exhibited significantly higher activity than cord GPX levels. This finding is in accordance with previous studies which showed higher GPX levels towards term in maternal system which is reported to be a protective mechanism against harmful effects of oxidizing agents like hydrogen peroxide [33]. SOD level compared between maternal and cord did not have any statistical significance in accordance with previous reports [34].

The process of labour can be influenced by certain maternal and fetal variables. Maternal and fetal parameters like maternal age, parity, weight of the baby, sex of the baby, duration of labour were studied with respect to oxidative status. Maternal age was positively correlated with GPX levels of the mother ($r=0.016$, $P<0.05$). Hence, maternal age at parity can be considered as an important determinant of oxidative stress [35].
Maternal nutrition is yet another determining factor for fetal outcome. The enzymatic antioxidant mechanisms rely on micronutrients for its effective functioning against free radicals induced lipid peroxidation. Manganese, copper, zinc and selenium are co factors for functioning of antioxidant enzymes. Deficiency of these micro nutrients can largely influence the enzymatic activity of antioxidants. Vitamin C, Vitamin E and glutathione which are non-enzymatic antioxidants are also diet based. Antioxidant supplementations to high risk mothers are beneficial according to some studies[36].

Hence, nutrition during antenatal period can influence oxidative status of the mother which indirectly can affect the antioxidant reserve of the neonate. Our study group which involved subjects from low socio economic status also would possibly have had some micro nutrient deficiency of co factors involved in antioxidant protection. In addition to nutrition, oxidative status can be modified by environmental factors like air pollution, chemicals in drinking water, genetic polymorphism of oxidative related enzymes [37, 38]. Taking these facts into consideration, our study enables us to understand clearly the role of efficient antioxidant systems during labour in uncomplicated pregnancies.

Conclusion

Oxidative imbalance does occur during labour in uncomplicated pregnancies which affects both the neonate and mother exhibiting alterations in their antioxidant mechanisms. The neonate exhibited higher oxidative stress than the mother. In order to enrich the antioxidant mechanisms during pregnancy and labour, supplementation of antioxidants may be advocated and its efficacy has to be eludedby further research and trials.

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References

1. Fainaru O, Almog B, Pinchuk I, Kupferminc MJ, Lichtenberg D, Many A. Active labour is associated with increased oxidisibility of serum lipids ex vivo. BJOG. 2002 Aug;109(8):938-41.

2. Rogers MS, Mongelli JM, Tsang KH, Wang CC, Law KP. Lipid peroxidation in cord blood at birth: the effect of labour. Br J Obstet Gynaecol. 1998 Jul;105(7):739-44.

3. Halliwell B, Gutteridge JM. Role of free radicals and catalytic metal ions in human disease: an overview. Methods Enzymol. 1990;186:1-85.

4. Davidge ST, Hubel CA, Brayden RD, Capeless EC, McLaughlin MK. Sera antioxidant activity in uncomplicated and preeclamptic pregnancies. Obstet Gynecol. 1992 Jun;79(6):897-901.

5. Carone D, Loverro G, Greco P, Capuano F, Selvaggi L. Lipid peroxidation products and antioxidant enzymes in red blood cells during normal and diabetic pregnancy. Eur J Obstet Gynecol Reprod Biol. 1993 Oct;51(2):103-9.

6. Łagód L, Paszkowski T, Sikorski R, Rola R. [The antioxidant-prooxidant balance in pregnancy complicated by spontaneous abortion]. Ginekol Pol. 2001 Dec;72(12):1073-8.

7. Little RE, Gladen BC. Levels of lipid peroxides in uncomplicated pregnancy: a review of the literature. Reprod Toxicol. 1999 Sep-Oct;13(5):347-52.

8. Nakai A, Oya A, Kobe H, Asakura H, Yokota A, Koshino T, Araki T. Changes in maternal lipid peroxidation levels and antioxidant enzymatic activities before and after delivery. J Nippon Med Sch. 2000 Dec;67(6):434-9.

9. Walsh SW, Wang Y, Jesse R. Peroxide induces vasoconstriction in the human placenta by stimulating thromboxane. Am J Obstet Gynecol. 1993 Oct;169(4):1007-12.

10. Takagi, Y., Nikaido, T., Toki, T., Kita, N., Kanai, M., Ashida, T., Ohira, S. and Konishi, I.Levels of oxidative stress and redox-related molecules in the placenta in preeclampsia and fetal growth restriction. Virchows Arch.2004 Jan; 444(1):49-55. Epub 2003 Oct 22.

11. Woods JR Jr, Cavanaugh JL, Norkus EP, Plessinger MA, Miller RK. The effect of labor on maternal and fetal vitamins C and E. Am J Obstet Gynecol. 2002 Nov;187(5):1179-83.

12. Maximo V, Miguel A, Juan S, Anna L, Fernando G, Jose V. Oxidative stress in a asphyxiated term infants resuscitatead with 100% Oxygen. J Pediatr. 2003 Jun; 142(6):616.

13. Georgeson GD, Szony BJ, Streitman K, Varga IS, Kovács A, Kovács L, László A. Antioxidant enzyme activities are decreased in preterm infants and in neonates born via caesarean section. Eur J Obstet Gynecol Reprod Biol. 2002 Jul 10;103(2):136-9.

14. Saugstad OD. Mechanisms of tissue injury by oxygen radicals: implications for neonatal disease. Acta Paediatr. 1996 Jan;85(1):1-4.

15. Bolisetty S, Naidoo D, Lui K, Koh TH, Watson D, Whitehall J. Antenatal supplementation of antioxidant vitamins to reduce the oxidative stress at delivery--a pilot study. Early Hum Dev. 2002 Apr;67(1-2):47-53.

16. Al-Shebly MM, Mansour MA. Evaluation of oxidative stress and antioxidant status in diabetic and hypertensive women during labor. Oxid Med Cell Longev. 2012;2012:329743. doi: 10.1155/2012/329743. Epub 2012 Jul 24.

17. Cynamon HA, Isenberg JN, Nguyen CH. Erythrocyte malondialdehyde release in vitro: a functional measure of vitamin E status. Clin Chim Acta. 1985 Sep 30;151(2):169-76.

18. Marklund S, Marklund G. Involvement of the superoxide anion radical in the autoxidation of pyrogallol and a convenient assay for superoxide dismutase. Eur J Biochem. 1974 Sep 16;47(3):469-74.

19. Moron MS. Estimation of glutathione, glutathione reductase, glutathione S transferase in rat lung and liver. Biochemistry Biophysics Acta1979; 582: 67 – 8.

20. Rotruck JT, Pope AL, Ganther HE, Swanson AB, Hafeman DG, Hoekstra WG. Selenium: biochemical role as a component of glutathione peroxidase. Science. 1973 Feb 9;179(4073):588-90.

21. Stípek S, Měchurová A, Crkovská J, Zima T, Plášteník J. Lipid peroxidation and superoxide dismutase activity in umbilical and maternal blood. Biochem Mol Biol Int. 1995 Apr;35(4):705-11.

22. Uotila J, Tuimala R, Aarnio T, Pyykkö K, Ahotupa M. Lipid peroxidation products, selenium-dependent glutathione peroxidase and vitamin E in normal pregnancy. Eur J Obstet Gynecol Reprod Biol. 1991 Nov 26;42(2):95-100.

23. Arikan S, Konukoğlu D, Arikan C, Akçay T, Davas I. Lipid peroxidation and antioxidant status in maternal and cord blood. Gynecol Obstet Invest. 2001;51(3):145-9.

24. Rao G, Kamath U, Raghothama C, Pradeep KS, Rao P. Maternal and fetal indicators of oxidative stress in various obstetric complications. Indian J Clin Biochem. 2003 Jul;18(2):80-6. doi: 10.1007/BF02867371.

25. Many A, Roberts JM. Increased xanthine oxidase during labour--implications for oxidative stress .Placenta. 1997 Nov;18(8):725-6.

26. Lees MH, Hill JD, Ochsner AJ 3rd, Thomas CL, Novy MJ. Maternal placental and myometrial blood flow of the rhesus monkey during uterine contractions. Am J Obstet Gynecol. 1971 May 1;110(1):68-81.

27. Kelly RW. Inflammatory mediators and parturition. Rev Reprod. 1996 May;1(2):89-96.

28. Calderon TC, Wu W, Rawson RA, Sakala EP, Sowers LC, Boskovic DS, Angeles DM. Effect of mode of birth on purine and malondialdehyde in umbilical arterial plasma in normal term newborns. J Perinatol. 2008 Jul;28(7):475-81. doi: 10.1038/jp.2008.29. Epub 2008 Mar 27.

29. Argüelles S, Machado MJ, Ayala A, Machado A, Hervías B. Correlation between circulating biomarkers of oxidative stress of maternal and umbilical cord blood at birth. Free Radic Res. 2006 Jun;40(6):565-70.

30. Kharb S. Low whole blood glutathione levels in pregnancies complicated by preeclampsia and diabetes. Clin Chim Acta. 2000 Apr;294(1-2):179-83.

31. Buhimschi IA, Buhimschi CS, Pupkin M, Weiner CP. Beneficial impact of term labor: nonenzymatic antioxidant reserve in the human fetus. Am J Obstet Gynecol. 2003 Jul;189(1):181-8.

32. Buhimschi IA, Buhimschi CS, Weiner CP. Protective effect of N-acetylcysteine against fetal death and preterm labor induced by maternal inflammation. Am J Obstet Gynecol. 2003 Jan;188(1):203-8.

33. Mihailović M, Cvetković M, Ljubić A, Kosanović M, Nedeljković S, Jovanović I, Pesut O. Selenium and malondialdehyde content and glutathione peroxidase activity in maternal and umbilical cord blood and amniotic fluid. Biol Trace Elem Res. 2000 Jan;73(1):47-54.

34. Telfer JF, Thomson AJ, Cameron IT, Greer IA, Norman JE. Expression of superoxide dismutase and xanthine oxidase in myometrium, fetal membranes and placenta during normal human pregnancy and parturition. Hum Reprod. 1997 Oct;12(10):2306-12.

35. Noor N, Islam N, Moin S, Mahdi AA, Jaiswal S, Bano F. Normal delivery induced stress alters glutathione peroxidase and TNF-α in elderly primigravidas mononuclear cells. Indian J Clin Biochem. 2008 Jul;23(3):227-32. doi: 10.1007/s12291-008-0051-1. Epub 2008 Oct 1.

36. Chappell LC, Seed PT, Kelly FJ, Briley A, Hunt BJ, Charnock-Jones DS, Mallet A, Poston L. Vitamin C and E supplementation in women at risk of preeclampsia is associated with changes in indices of oxidative stress and placental function. Am J Obstet Gynecol. 2002 Sep;187(3):777-84.

37. Mistry HD, Williams PJ. The importance of antioxidant micronutrients in pregnancy. Oxid Med Cell Longev. 2011;2011:841749. doi: 10.1155/2011/841749. Epub 2011 Sep 13.

38. Da Costa LA, Badawi A, El-Sohemy A. Nutrigenetics and modulation of oxidative stress. Ann Nutr Metab. 2012;60 Suppl 3:27-36. doi: 10.1159/000337311. Epub 2012 May 15.

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