Assessment of fetal antioxidant and oxidant status during different anesthesia techniques for elective cesarean sections

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Background: We aimed to investigate the effects of general, spinal and epidural anesthesia on fetal total antioxidant status (TAS) and total oxidant status (TOS), and oxidative stress index (OSI) during elective cesarean section in this study. Materials and Methods: Forty-seven parturients scheduled for elective cesarean section were randomly allocated into three groups: Group spinal (n = 15), group epidural (n = 17), and group general (n = 15). This prospective randomized study was performed in Faculty of Medicine, Turgut Ozal University, Turkey. After the baby was delivered; TAS, TOS levels, and arterial blood gases parameters were analyzed in an umbilical arterial blood sample. OSI values are calculated by a ratio of TOS to the TAS. Results: The levels of TAS and TOS in umbilical arterial blood sample were not statistically different among three. However, OSI values were significantly different among the three groups (P = 0.042). Median OSI values is 24 (interquartile range [IQR], 2-37) in group spinal, 19 (IQR, 4-44) in group epidural, and 8 (IQR, 4-36) in group general. There was no significant difference in OSI values in the comparison of group spinal with group general and group epidural, but it was significantly lower in group general when compared with group epidural with Bonferroni correction (P = 0.017). Umbilical cord arterial blood gas values (pH, PaCO2, PaO2, SaO2, HCO3, and CtO2), glucose, lactate, and hemoglobin levels were similar in three groups. Conclusion: General anesthesia may be more favorable than epidural in those undergoing cesarean section when fetal oxidative status gains importance.

Key words: Anesthesia, cesarean section, oxidative stress

INTRODUCTION

A lot of factors may which might affect mother and fetus in intrauterine and peripartum period increase reactive oxygen radicals.11 Reactive oxygen radicals and imbalance between the oxidant and body’s antioxidant defenses were associated with several serious diseases at newborn such as bronchopulmonary dysplasia, respiratory distress syndrome, necrotizing enterocolitis, periventricular leukomalacia, hypoxic-ischemic encephalopathy, and sudden infant syndrome.2-3 Administration of regional anesthesia has become more and more popular because of the development of anesthesia techniques and materials besides patients can remain conscious. In the case of cesarean section surgery, for the determination of the method of anesthesia, not only patient and doctors preferences but also fetal status is important. Although general anesthesia is known to have negative effects on the baby’s Apgar scores,4 it is unavoidable for some patients who refused regional anesthesia due to fear of the needless, emergency conditions, and medical contraindication such as coagulation disorders, aortic stenosis, and infection.5 Although oxidative and anti-oxidative stress marker levels were shown to be higher in cesarean section...
operations than vaginal deliveries in previous studies, different anesthesia techniques and conditions were not evaluated in cesarean section. The aim of the present study was to investigate the effects of general anesthesia that was induced with propofol, spinal and epidural anesthesia on fetal total antioxidant status (TAS) and total oxidant status (TOS), and oxidative stress index (OSI) during elective cesarean section.

MATERIALS AND METHODS

Study design and participants
This prospective randomized study was performed in Faculty of Medicine, Turgut Ozal University, Turkey. After obtaining the approval of the local Clinical Research Ethics Committee of the Fatih University, written informed consent was received from American Sociological Association I, II, and a singleton normal pregnant at term (38-40 weeks) who were scheduled for elective cesarean section in 6-month period. Patients who undergo emergency cesarean section, those who used drugs except iron supplements and those who smoked during pregnancy, those with complications of pregnancy, those who diagnosed with intrauterine growth retardation and fetal malformation were excluded from the study. The number of patients enrolled in this study was determined based on our preliminary study and a power of 80% and significant difference for OSI value amount three groups with a significance level of 5%. The sample size was calculated as 14 patients for each group. Participants were randomly allocated to three groups using a computer: spinal anesthesia (Group S), epidural anesthesia (Group E), and general anesthesia (Group G) were performed.

Procedures and variables
Before the surgery, all patients were administered 1000 ml Ringer’s lactate infusion, standard monitoring was performed with electrocardiography, pulse oximetry, and noninvasive blood pressure. In Group G, general anesthesia was induced with 2 mg/kg propofol, 0.5 μg/kg remifentanil, and 0.6 mg/kg rocuronium. Patients were intubated after 1 min and surgery was started, ventilation was continued with 1% sevoflurane in 50% O2/air mixture. After delivery of the baby, anesthesia was maintained with 1% sevoflurane in 50% O2/N2O mixture, 0.05 mg/kg midazolam intravenously (IV), and 0.25 μg/kg/min remifentanil continuous IV infusion. In Group E, 20 gauge epidural catheter (Perifix®, Brown, Germany) was placed at L3-4 or L4-5 interspinous space using 18 Gauge Tuohy needle (Perican®, Brown, Germany). The patients were positioned at supine position with a 15° left tilt and 30° head up, 15 ml 0.5% levobupivacaine and 2 ml (100 μg) fentanyl were injected to epidural space after test dose lidocaine. In Group S, 2 ml 0.5% levobupivacaine and 20 μg fentanyl were injected to intrathecal space with 27 gauge pencil point spinal needle (Espocan®, Brown, Germany). Mean arterial pressure (MAP), heart rate (HR), and oxygen saturation in all patients were recorded intraoperatively. A decrease in systolic pressure of >20% from baseline was considered as hypotension treated with 5-10 mg ephedrine, and this application was noted. Oxygen with nasal cannula was not given to patients in group spinal and group epidural unless desaturation or severe hypotension. After the baby was delivered, and umbilical cord clamped, umbilical artery were isolated and two blood samples were obtained. The first blood sample of umbilical artery was assessed for arterial blood gases analyses and measured hemoglobin, glucose, lactate. The second blood samples were centrifuged at 3000 rpm for 10 min and remaining plasma samples were stored at −80°C until analysis of plasma TAS and plasma TOS levels.

Apgar scores were assessed at 1st and 5th min by a pediatrician who never enters the operating room and was blinded to anesthesia techniques. The cases in which meconium was observed in amniotic fluid, a blood sample was taken from umbilical vein instead of the umbilical artery and birth weight was >3000 or >3500 g were excluded from the study.

Measurement of TAS was evaluated using Erel’s automated method,[6] which depends on the production of hydroxyl radical, the most potent of biological radicals. The results are expressed in mmol Trolox equivalents/L.

Measurement of TOS was performed by Erel’s method,[7] in which oxidants present in the sample oxidize the ferrous-ion-o-dianisidine complex to ferric ion. The results were expressed in terms of micromolar hydrogen peroxide equivalent per liter (μmol H2O2 equivalent/L).

For calculation of OSI, the resulting unit of TAS was converted to μmol/L and the OSI value was calculated according to the following formula:

\[ \text{OSI (arbitrary unit)} = \frac{\text{TOS (μmol H}_2\text{O}_2 \text{equivalent/L)}}{\text{TAS (μmol Trolox equivalent/L)}} \]

Statistical analysis
All statistical calculations were performed using Statistical Package for the Social Sciences for Windows software, version 17.0 (SPSS Inc., Chicago, IL, USA). Normal distribution of the collected data was tested by the Shapiro-Wilk test. Normally distributed data were presented as mean ± standard deviation (SD) and were tested with analysis of variance. Nonnormally distributed data were analyzed with Kruskal–Wallis test and presented as median (interquartile range [IQR]) and were tested with Kruskal-Wallis test. Post-hoc analysis for ANOVA was performed with Tukey test. Kruskal-Wallis test with Mann-Whitney U-test using Bonferroni adjustment were
used if there is a statistically significant difference among the three groups. Values were presented as mean ± SD or median (IQR), \( P < 0.05 \) was considered as statistically significant.

**RESULTS**

Overall, 79 patients were included in this study and patients were divided into three groups randomly. Overall, 32 patients in all groups were excluded because of birth weight, meconium-stained amniotic fluid, taking blood samples from umbilical vein and hemolysis and data of 47 patients were analyzed (15 in the spinal group, 17 in the epidural group, and 15 in the general anesthesia group) [Figure 1].

Patients’ mean age was 30 ± 0.5 years old, mean gestational age was 38.3 ± 0.5 week, mean weight was 78 ± 10 kg, mean height was, 163 ± 4 cm. There was not any significant difference between groups according to demographic findings \( (P > 0.05) \) [Table 1]. Apgar scores at 1 and 5 min were similar among the three groups [Table 1].

At the intraoperative 30th min, HR was statistically lower in Group G than Group S and Group E \( (P = 0.002) \) (93.6 ± 11.7 in Group S, 97.1 ± 12.8 in Group E, and 78.1 ± 13.9 in Group G). MAP was found to be statistically significantly lower in Group S than the other groups at the 5th min intraoperatively \( (P = 0.011) \) (65.6 ± 17.0 in Group S, 79.4 ± 13.4 in Group E, and 83.7 ± 15.7 in Group G). The hemodynamic data at other follow-up times were similar in all groups.

**Table 1: Patient’s characteristics and intraoperative properties**

| Characteristic                     | Group spinal \( (n = 15) \) | Group epidural \( (n = 17) \) | Group general \( (n = 15) \) | \( P \) |
|-----------------------------------|-----------------------------|-----------------------------|-----------------------------|-------|
| Demographic properties            |                             |                             |                             |       |
| Height (cm)                       | 163±4                       | 163±5                       | 164±4                       | 0.827 |
| Weight (kg)                       | 79±8                        | 76±11                       | 79±10                       | 0.635 |
| Age (year)                        | 31±3                        | 30±3                        | 29±6                        | 0.322 |
| Gestational age (week)            | 38.2±0.4                    | 38.2±0.6                    | 38.3±0.6                    | 0.787 |
| Birth weight (g)                  | 3285±3                      | 3225±3                      | 3246±2                      | 0.914 |
| Operative properties              |                             |                             |                             |       |
| Duration of surgery (min)         | 31±6                        | 31±7                        | 37±10                       | 0.115 |
| Apgar score 1st min               | 9 (8-10)                    | 9 (8-9)                     | 9 (6-9)                     | 0.445 |
| Apgar score 5th min               | 10 (10-10)                  | 10 (10-10)                  | 10 (9-10)                   | 0.315 |

Data presented as mean ± SD or median (IQR); *\( P < 0.05 \) was accepted statistically significant; SD = Standard deviation; IQR = Interquartile range.
Umbilical cord arterial blood gas values were similar in three groups [Table 2]. The levels of TAS, TOS in umbilical cord arterial blood sample were not statistically different among three groups. However, the OSI values calculated by a ratio of the TOS to the TAS were significantly different among the three groups ($P = 0.042$). Median OSI values is $24$ (IQR, $2-37$) in group spinal, $19$ (IQR, $4-44$) in group epidural, and $8$ (IQR, $4-36$) in group general [Table 3]. There was no significant difference in the comparison of Group S with Group G and E, but OSI values were significantly higher in Group E when compared with Group G with Bonferroni correction ($P = 0.017$). There was no significant correlation among TAS, TOS, OSI values, and the evaluation of umbilical arterial blood gas parameters.

**DISCUSSION**

In the present study, we compared the effects of different anesthetic techniques for elective cesarean section operations on TAS, TOS, and OSI values in umbilical artery blood sample and fetal umbilical cord arterial blood gas parameters. We observed that OSI values in umbilical cord arterial blood sample at the cesarean section under general anesthesia were lower than epidural. In the majority of prior studies evaluating fetal oxidative stress, normal spontaneous vaginal delivery, and elective cesarean section were compared. According to some studies, mode of delivery was not effective on fetal oxidative status. There is not known the previous study which evaluates only the effect of different anesthesia methods on fetal oxidative stress parameters in elective cesarean sections. However, there are two similar studies. In one of these studies, Compagnoni et al. observed that the antioxidant levels of coenzyme Q were evaluated in maternal and umbilical blood samples in vaginal delivery and elective cesarean section. In this study, only one antioxidant level was evaluated, and it was found to be higher in the spinal anesthesia-cesarean section and normal vaginal delivery group than general anesthesia-cesarean section. In another study, in which normal vaginal delivery, cesarean section undergoing epidural and general anesthesia were compared, although umbilical artery PO$_2$ level was found to be higher in the general anesthesia group than others, malondialdehyde and glutathione levels were found lower in the epidural-cesarean group. It was concluded that cesarean delivery with epidural anesthesia might be safer considering lipid peroxidation. As mentioned in both studies, regional anesthesia is superior to general anesthesia in terms of oxidative stress. These results are not congruent with the results of our study. Pence et al. observed a decrease in both oxidant and antioxidant marker levels in the cesarean section with epidural anesthesia. Similar to our study, in the study of Compagnoni et al., supplemental O$_2$ was not applied in spinal anesthesia group and they have suggested that coenzyme Q levels were found lower in general anesthesia group because of oxidative stress enhancing effect of 50% oxygen administration. However, in a study 30%, 50%, or 100% O$_2$ in equivalent minimum alveolar concentration of sevoflurane and nitrous oxide were used for elective cesarean section under general anesthesia, although higher PaO$_2$ levels was observed in 100% O$_2$ group, there were no differences in the maternal and fetal lipid peroxidation. In another study, two groups of patients undergoing regional anesthesia for emergency cesarean section were compared, higher umbilical artery PaO$_2$ and CiO$_2$ were observed in 60% O$_2$ with the venturi mask group than room air group but, similar 8-isoprostane levels in maternal and umbilical artery blood samples have been reported in both groups. As a result, authors concluded that there is no need routine oxygen support in elective cesarean sections. In the present study, 50% air/50% oxygen were used in the general anesthesia group until the delivery of the baby, O$_2$ support was not used in the regional anesthesia groups. Despite the use of 50% O$_2$ in general anesthesia group, the difference was not observed at

**Table 2: Umbilical arterial blood gas, Hb, glucose, and lactate values**

| Variable                  | Group spinal (n = 15) | Group epidural (n = 17) | Group general (n = 15) | P       |
|---------------------------|----------------------|------------------------|------------------------|---------|
| Umbilical arterial blood gas |                      |                        |                        |         |
| pH                        | 7.35±0.03            | 7.35±0.01              | 7.33±0.02              | 0.080   |
| PaCO$_2$ mmHg             | 42 (33-47)           | 41 (33-48)             | 45 (35-55)             | 0.138   |
| HCO$_3$ mmol/L            | 21 (19-22)           | 21 (19-22)             | 21 (19-22)             | 0.504   |
| PaO$_2$ mmHg              | 15 (10-31)           | 17 (12-27)             | 18 (12-30)             | 0.359   |
| SaO$_2$ %                 | 29 (9-68)            | 34 (15-68)             | 35 (15-66)             | 0.755   |
| O$_2$ content (ml/dl)     | 5.8 (1.2-13.2)       | 7.2 (3.4-13.9)         | 7.1 (3.9-12.5)         | 0.581   |
| Hb g/dl                   | 13.7±0.8             | 14.4±1.2               | 14.9±2.1               | 0.153   |
| Glucose mg/dl             | 61±9                 | 63±6                   | 63±5                   | 0.518   |
| Lactate mmol/dl           | 1.4 (1.1-2.4)        | 1.5 (1.2)              | 1.4 (1.2-1.9)          | 0.801   |

Data presented as mean ± SD or median (IQR); SD = Standard deviation; IQR = Interquartile range; Hb = Hemoglobin

| Table 3: Total antioxidant and oxidant status, OSI values in umbilical arterial blood sample |
|---------------------------------------------|---------------------------------------------|---------------------------------------------|---------|
| Variable                     | Group spinal (n = 15) | Group epidural (n = 17) | Group general (n = 15) | P       |
| TAS (mmol Trolox equivalents/L)w         | 2.1 (1.6-2.5)        | 2.2 (1.4-2.8)           | 2.1 (1.8-2.4)          | 0.448   |
| TOS (μmol H$_2$O$_2$ equivalent/L)       | 5.8 (0.4-8.6)        | 4.4 (0.8-10.5)          | 1.6 (0.8-9.3)          | 0.050   |
| OSI (arbitrary unit)                 | 24 (2-37)            | 19 (4-44)               | 8 (4-36)**             | 0.042   |

Data presented median (IQR); *Statistically significant among the three groups; **Statistically significant between Group G and E with Bonferroni correction ($P = 0.017$); TAS = Total antioxidant status; TOS = Total oxidant status; OSI = Oxidative stress index
umbilical arterial blood gas values in all three groups, and OSI was significantly lower in the general anesthesia group. The results obtained in this study support the hypothesis that application of 50% oxygen in the general anesthesia group did not increase the level of fetal oxidants.

Evaluation of umbilical artery blood gas can yield important information about the status of the baby before and during delivery. According to meta-analysis results, umbilical artery pH values were found to be lower in spinal anesthesia than general anesthesia and epidural anesthesia. These results were in association with the development of maternal hypotension and using vasopressor agents for the treatment of hypotension in spinal anesthesia. In our study, the decrease in MAP was observed at 5th min in the spinal group when compared with other groups. This might explain why OSI is found to be higher in a group of spinal anesthesia.

The positive effect of propofol on oxidative stress has been reported in both in vivo and in vitro studies. This protective effect of oxidative status was connected with scavenging of oxygen derived free radicals in a tocopherol-like manner. Administration of propofol was reported to exert a scavenging effect on peroxynitrite in later studies. Continuous infusion of propofol for general anesthesia or sedation was demonstrated to attenuate oxidative damage. In our study, propofol was used for anesthesia induction in general anesthesia group. Lower OSI values in general anesthesia could be attributed to the antioxidant effect of propofol because the efficacy of propofol continued from anesthesia until the induction to the baby delivery.

As in previous studies, since different oxidant and antioxidant species may have additive effects, serum concentrations of different oxidant and antioxidant species may be measured separately but this method is not practical and can achieve false results. Nowadays, total antioxidant and oxidant levels can be measured by a simple, stable, reliable, and sensitive method. We preferred assessment of fetal oxidative stress by measurements of the TOS, TAS, and calculating the value of the OSI instead of measurement of several oxidant and antioxidant molecules in our study. Indeed, we have seen the importance of the calculation of the OSI when we look at the results of our study. Although we did not observe the statistically significant difference in umbilical cord TAS values between all groups, TAS values were found to be lower in Group G than other groups but this was not statistically significant. OSI values which are the ratio of TAS/TOS were observed to be significantly lower in favor of the general anesthesia group.

In summary, we observed that the application of 50% oxygen under general anesthesia by elective cesarean operations had no effect on oxidative stress and different anesthesia techniques used did not affect Apgar scores and blood gas parameters. Regional anesthesia for the elective cesarean section is often implemented for perioperative patient comfort. However, it is our conclusion that general anesthesia with induced propofol for elective cesarean section may be more favorable than epidural in term fetal oxidative status due to the fact that OSI values were lower in general anesthesia group than regional anesthesia groups.

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Conflicts of interest
There are no conflicts of interest.

AUTHOR’S CONTRIBUTION
SK contributed in the conception and design of the work, conducting the study, drafting and revising the draft, approval of the final version of the manuscript, agreed for all aspects of the work. EAK contributed in the conception of the work, conducting the study, revising the draft, approval of the final version of the manuscript, and agreed for all aspects of the work. AK contributed in the conception of the work, conducting the study, revising the draft, approval of the final version of the manuscript, and agreed for all aspects of the work. CK contributed in the conception of the work, conducting the study, revising the draft, approval of the final version of the manuscript, and agreed for all aspects of the work. OE contributed in the conception and design of the work, conducting the study, revising the draft, approval of the final version of the manuscript, and agreed for all aspects of the work. 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