

Research Article

**Analysis of levels of antioxidants and Catalases in the blood
with the use of Ropivacaine for Anesthesia**

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ABSTRACT

Introduction: Anaesthesia is a loss of sensation or a loss of consciousness and its mechanisms by which drugs can produce this state yet undetermined. Previous observations described how anaesthetic agents work at the molecular level and cause a loss of consciousness. **Objectives of the study:** The objective of this study was to determine if ropivacaine has any effects on the antioxidant defense system and in lipid peroxidation as Malondialdehyde (MDA) of spinal fluid. **Material and method:** To measure the levels of lipid peroxidation, glutathione (GSH), superoxide dismutase (SOD) and the levels of catalases (CAT), samples were taken from the vein before anesthesia (0 min) and at 5, 15, and 60 min. after anesthesia. **Results:** The levels of MDA, SOD and GSH slightly increased at 5 min after spinal injection of ropivacaine and this increase continued throughout anesthesia ($P < 0.001$). Catalases levels were decreased at 5 min after anesthesia ($P < 0.001$) and this level not recovered after 60 min of anaesthesia. **Conclusion:** In conclusion, we found that spinal injection of ropivacaine increased free radical levels in spinal fluid which may be supported antioxidant environment of spinal fluid during anesthesia.

Key words: Anaesthesia, Antioxidants, Radical

INTRODUCTION

General anesthetics agents' effect on the ion channels and neurotransmitter receptor subtypes¹. They even affect actin-based motility in dendritic spines. Anesthesiologists frequently discuss the actions of many drugs counting opioids, muscle relaxants and anticoagulants. However, emergence of general anesthesia is still treated as a passive process, dictated by the pharmacokinetics of anesthetic drug clearance².

Even though there has been active research onto the mechanisms of general anesthesia for over a

century but still there is no commonly accepted or fully suitable definition of general anesthesia itself³. The critical effectors sites that cause general anesthesia may be moderately different from those primary target sites where anesthetic molecules actually bind. For example, it is generally thought that changes in the properties of neuronal ion channels cause general anaesthesia, but this could result when anaesthetic molecules interact with any of the following primary sites: a) the channel proteins, b) channel regulatory

proteins (e.g., by phosphorylating them), or c) the surrounding lipid bilayer⁴. Anaesthetic agents are mostly hydrophobic in nature and usually behave like polar ions. Intravenous anaesthetic agents used for induction and for short surgical procedure and they can produce apnoea and hypotension. They are contraindicated if the anaesthetist is not confident of being able to maintain an airway. Any muscle relaxant must be given before intubation and the requirement for Individual varies considerably⁵.

There are many agents used as a local anaesthetic for the treatment and surgery. Ropivacaine is a commonly used anaesthetic drug which belongs to the amino amide group⁶. Ropivacaine is the (S)-enantiomer of 1-propyl-2', 6'- pipercoloxylidide, an amide local anesthetic with a structure similar to that of mepivacaine and bupivacaine. Animal studies have indicated that ropivacaine is less toxic than bupivacaine. Both drugs have a comparatively long duration of action, but their relative potency has still to be determined in humans. As Ropivacaine used as a long acting local anesthetic drug and it has a same clinical profile as bupivacaine⁷.

Objectives of the study

The objective of this study was to determine if ropivacaine has any effects on the antioxidant defense system and in lipid peroxidation as Melanodialdehyde (MDA) of spinal fluid.

Materials and methods

The whole experimental work was conducted at Services Institute of medical sciences Lahore, Pakistan.

Experimental design

This study was conducted according to the rules and regulations of authority of services Institute of medical sciences Lahore. 50 patients were selected to study the effect of ropivacaine. 5.0 ml blood sample was taken from vein to measure the levels of lipid peroxidation (MDA), glutathione (GSH), SOD and Catalases before anaesthesia (0 min), and at 5, 30 and 60 minute after anaesthesia. Commercially available enzymatic kits of Randox were used

Statistical Analysis

Statistical analysis (Anova Test and Post Hoc) was performed using the SPSS software program (17.0). All results were expressed as the mean \pm standard deviation (SD). As P value <0.05 was considered to be statistically significant.

RESULTS

Mean values of investigated parameters and differences in the values between, before and after anaesthesia are represented in the table 01. The values are expressed in terms of mean \pm SD. According to the analysis, the level of SOD, MDA and GSH increase as compared to normal level. But the level of catalases decreased with the value of 0.43 ± 0.39 (after 5 minutes of anaesthesia). All the data are explained in table 01.

Table 01: Analysis of parameters

No.of Observation	Analysis of blood	Normal $\mu\text{g/mL}$	Before treatment $\mu\text{g/mL}$	After treatment(5min) $\mu\text{g/mL}$	After treatment(15min) $\mu\text{g/mL}$	After treatment(60min) $\mu\text{g/mL}$
01	SOD	0.32 \pm 0.00	0.33 \pm 0.23	0.39 \pm 0.00	0.45 \pm 0.19	0.51 \pm 0.21
02	CAT	4.16 \pm 0.00	0.90 \pm 0.00	0.43 \pm 0.39	0.30 \pm 0.24	0.19 \pm 0.18
03	GSH	1.89 \pm 0.00	2.48 \pm 1.29	3.23 \pm 0.03	4.92 \pm 0.57	5.64 \pm 0.55
04	MDA	2.35 \pm 0.00	4.26 \pm 0.00	4.95 \pm 0.97	5.13 \pm 1.06	6.58 \pm 0.00

Effects of spinal injection of Ropivacaine on the levels of MDA in spinal fluid indicated in Fig. 1. The levels of MDA slightly increased at 5 min after spinal injection of ropivacaine and this increase continued throughout anaesthesia ($P < 0.001$).

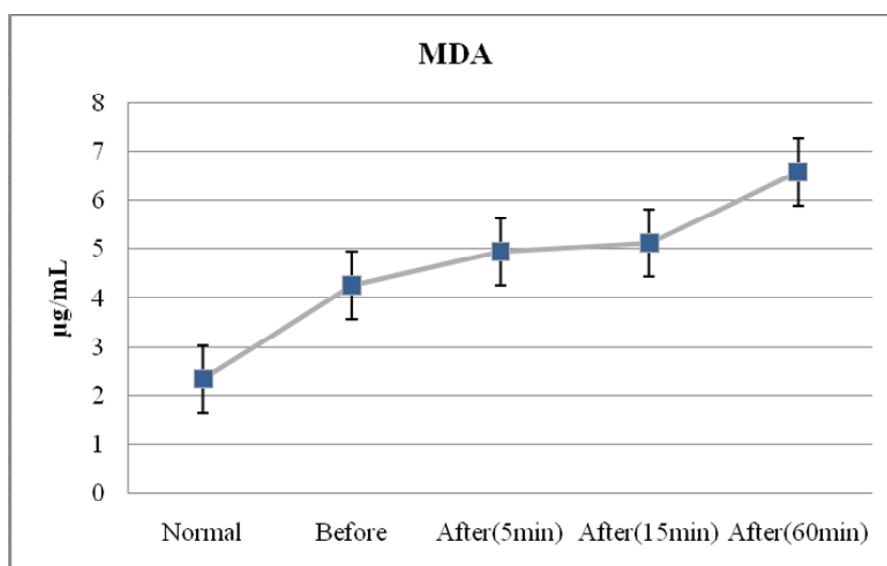


Fig 01: Effects of spinal injection of ropivacaine on the levels of MDA in blood. $P < 0.001$ in comparison to the 0th point of the time. Values are expressed as mean \pm SD.

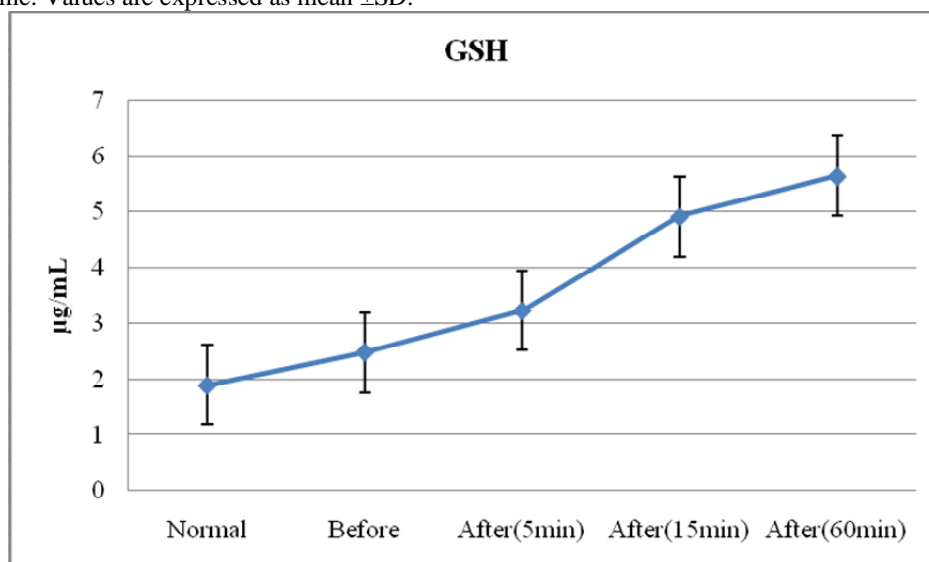


Fig 02: Effects of spinal injection of ropivacaine on the levels of GSH in blood. Values are expressed as mean \pm SD. $P < 0.001$ in comparison to the 0th point of the time.

As shown in Fig. 2 the levels of GSH in the blood regularly increased throughout anesthesia ($P < 0.001$). Effects of spinal injection of ropivacaine on the activity of SOD in blood introduced in Fig 3.

DISCUSSION

The results of this study show that spinally injected ropivacaine has an effect on change the lipid peroxidation and antioxidant enzymes in the blood. Lipid peroxidation is one of the prominent

revealed of oxidative stress. Reactive oxygen species are induced oxidation and peroxidation of membrane phospholipids, thereby causing damage to the phospholipid molecule as well as to other molecules in the cells⁸. Polyunsaturated fatty acids are found in abundance in mammalian membrane lipids and are the most likely targets of Reactive Oxygen Species (ROS). Activities of enzymatic antioxidants like catalase, superoxide dismutase and glutathione peroxidase significantly decrease in prostate cancer patients compared to normal

subjects⁹. Oxidative stress plays an important role for the initiation of DNA damage. In the present study, we observed an increase in MDA levels and increase in SOD and GSH levels and decrease in catalases level. Increased levels of lipid hydroxyl peroxides and hydroxyl phospholipids have been associated with oxidative stress and membrane injury that occur in pathological conditions such as spinal cord injury¹⁰. Lenfant et al found that bupivacaine provide a protective effect against induced free radical increase with 2,2-azobis di hydrochloride¹¹.

Increased levels of anti-oxidants and decreased activities of catalases can be correlated to enhanced lipid peroxidation and subsequent neoplastic transformation¹². Antioxidant enzymes which catalyze the conversion of reactive oxygen species to water include catalase (CAT), manganese containing superoxide dismutase (Mn-SOD) and copper and zinc containing superoxide dismutase, a mitochondrial enzyme that plays a key role in protecting the cell from oxidative damage¹³.

Free radical-mediated damage occurs as a consequence of GSH depletion, depressed antioxidant enzyme activities and enhanced lipid peroxidation. On the other hand, marked increase in oxidative stress may have been potentiated by increased antioxidant enzyme activities¹⁴. Nevertheless, antioxidants can scavenge ROS before they can cause damage to various biomolecules or prevent oxidative damage from spreading out by interrupting the radical chain reaction of lipid peroxidation. The disappearance of the endogenous antioxidants in plasma was measured, in relation to the formation of lipid hydroperoxides formed from endogenous lipids. GSH is a major non-protein thiol in mammals and is essential for structural and metabolic integrity of cells¹⁴. GSH plays an important role in maintaining the stability of the membrane as a direct free radical scavenger and in the protection of intracellular components such as sulfhydryl enzymes against oxidative denaturation. Also, it has been demonstrated to scavenge superoxide

Radical in a dose dependent manner¹⁵.

CONCLUSION

In conclusion, we found that while spinal injection of ropivacaine increased free radical levels in the blood. According to our study levels of free radicals will increase in the blood and it may be supported antioxidant environment of spinal fluid during anesthesia.

Conflict of interest

There is no conflict of interest.

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