Original Article

Antioxidative Effects of Isoflurane and Propofol on Serum Levels of Catalase, Glutathione and Superoxide Dismutase in Craniotomy for Supratentorial Tumor

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Abstract

Background: Surgical stresses can reduce the activity level of antioxidant enzymes. The brain is one of the vital organs of the body and yet vulnerable to oxidative damage. Considering the antioxidative effects of anesthetics, this study aimed to compare the effect of isoflurane and propofol on the serum levels of catalase, glutathione peroxidase and superoxide dismutase in patients who experienced craniotomy due to supratentorial tumor.

Methods and materials: This randomized clinical trial was performed on 40 patients with supratentorial tumor craniotomy in Loghman Hakim hospital in Tehran (capital city of Iran), who were randomly assigned to receive propofol and isoflurane. Data were collected using a questionnaire including age, sex, serum glutathione peroxidase level, serum superoxide dismutase level, serum catalase levels, duration of surgery, and fentanyl. Data were analyzed using SPSS-21 software, Chi-Square, Paired T-test, ANOVA, independent t-test, Kruskal-Wallis and Wilcoxon tests.

Results: Increasing in serum catalase level after surgery was significant in propofol group (p=0.002), but not significant in isoflurane group. Reduction of serum glutathione peroxidase level after surgery was significant in the isoflurane group (p=0.003), but not significant in propofol group. Reduction of serum level of superoxide dismutase after surgery was not significant in both groups. Serum levels of catalase, glutathione and superoxide dismutase were not significantly correlated with age, sex, fentanyl and duration of surgery.

Conclusion: The study showed that serum catalase levels increased in the propofol group after surgery. However, serum glutathione peroxidase levels decreased in the isoflurane group after surgery. Changes in serum level of superoxide dismutase after surgery were not related to the type of anesthetic drug.

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Keywords: Isoflurane, Propofol, Catalase, Glutathione peroxidaase, Superoxide dismutase, Craniotomy

Please cite this article as: Nashibi M, Sezari P, Mottaghi K, Yeganeh-Khah S, Safari F. Comparison of the Antioxidative Effects of Isoflurane and Propofol on Serum Levels of Catalase, Glutathione and Superoxide Dismutase in Patients Undergoing Craniotomy for Supratentorial Tumor. J Cell Mol Anesth. 2019;4(2):33-40.

Introduction

Surgeries usually result in some unwanted reactions and consequently disrupt patient's hemostasis, which is known as surgical stress. Several studies claimed that surgical stress would decrease the activity of antioxidant enzymes. Since brain is the most vulnerable organ in this regard (1, 2) and regarding anti-oxidative role of anesthetic medications, the current study assessed and compared the effects of two medications namely propofol and isoflurane on serum levels of antioxidants catalase (CAT), superoxide dismutase (SOD) and glutathione peroxidase (GPX) among patients who underwent craniotomy due to supratentorial brain tumors. Recent studies found direct correlation between increased oxidative stress and poor prognosis among ill patients, which could increase the risk of apoptosis, inflammation and finally death (3-7). Herbert et al. showed that low levels of antioxidants in addition to high O_2 consumption in brain, imposesstress damage to the brain following craniotomy (8).

Propofol is an anesthetic medication, which may have key role in decreasing reactive oxygen species (ROS) due to similarity of its molecular structure to phenol-based antioxidants (9, 10). In this way, de la Cruz et al, believed that propofol resulted in increased GPX serum level in rats as well as in platelets in patients who had surgery (11). Likely, Ucar and Braz found improvement in body antioxidative capacity by propofol and isoflurane (12, 13).

Antioxidative system in the body divides into enzymatic and non-enzymatic categories among which the former includes superoxide dismutase (SOD), glutathione peroxidase (GPX), glutathione (GSH), Catalase (CAT), and other variables such as bilirubin, sexual hormones, melatonin, co-enzyme Q, uric acid and nutritional antioxidants (vitamins C, E, beta-carotene and flavonoids) (14, 15).

Studies showed that craniotomy and other surgeries or any kind of anesthesia would temporarily decrease antioxidant enzymes in the body, which is obvious after two to four hours of the surgery, which returns to normal level after 24 hours (16). Meanwhile, propofol and dexmedetomidine seem to be more active in antioxidation than midazolam during anesthesia (14). However, brain sensitivity to oxidative stress may cause serious complications after surgery including neuron damage and early Alzheimer that could be prevented through some suitable acts (17). In this regard, Osier et al, showed short-term effects of melatonin in 2018 but there is no idea for long-term management in this matter (18).

The current study tried to assess the changes in enzymatic antioxidant system following craniotomy for supratentorial brain tumors by measuring three fundamental enzymes in this regard to compare two anesthetic medications to be used safely.

Methods

Through a randomized controlled trial, patients with supratentorial tumors enrolled into the study. The participants were divided into two groups who received isoflurane or propofol as anesthetics during their surgery using random numbers. All the participants were 18-65 years old with American society of anesthesiologists (ASA) class I and II, who had neither history of rheumatologic diseases nor history of taking anti-inflammatory drugs, β-blockers, calcium channel blockers and vitamin supplements. Patients with high intracranial pressure (ICP), drug allergy, requirement to more than four units of packed red blood cells and individuals who were transferred to intensive care unit (ICU) while trachea was intubated, were quitted from the study as well.

Disregarding the groups, all the participants were thoroughly monitored using noninvasive blood pressure (NIBP), invasive blood pressure (IBP), continuous electrocardiography and peripheral arterial O_2 saturation (SpO2) during surgery. The first blood sample (5 mL) was gathered before induction of anesthesia, and then patients' were oxygenated with 100% O_2 for three minutes and 0.03 mg/kg midazolam and 3 µg/kg fentanyl were injected via intravenous access line as premedication. Anesthesia was induced using sodium thiopental (5 mg/kg) and cisatracurium 0.2 mg/kg and maintained by using cisatracurium intermittent injections (0.1 mg/kg) with the guide of train of four (TOF) monitoring and 50 mcg of fentanyl

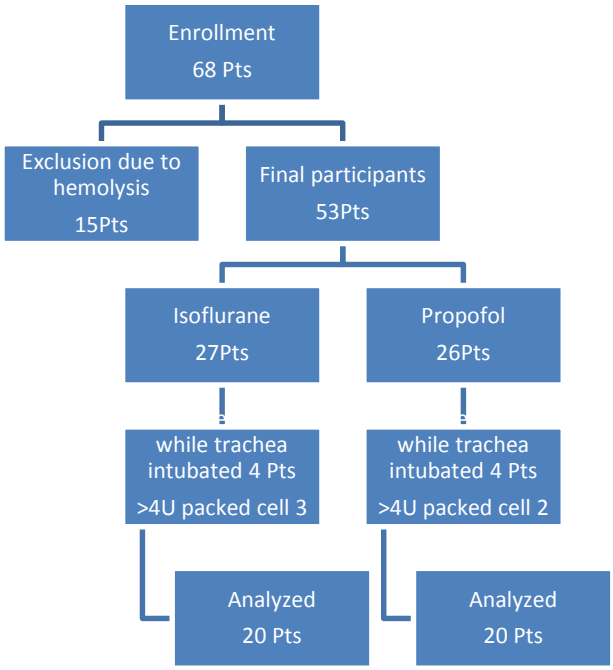

Figure 1. Flowchart of participation of the current study.

every 45 minutes. In propofol group, infusion rate of 100-300 mcg/kg/min and in isoflurane group1.2 to 1.5% of isoflurane were used to keep the level of anesthesia using cerebral state index (CSI) between 40 and 60.

All the patients were in supine position and benefited forced air warmer to fix central body temperature (nasopharyngeal) at 36-37°c while fluid intake and output were recorded. Intermittent pneumatic compression (IPC) was used to prevent deep venous thrombosis (DVT). During anesthesia, patients' blood pressure and heart rate were kept by 20% and heart rate <45 beat/min was considered as bradycardia to use 0.5 mg atropine each time. Crystalloid fluid was used in case of 20% reduction in blood pressure before using 5mg of ephedrine if needed. The second blood sample (5mL) was obtained postoperatively in post anesthesia care unit (PACU) for analyzing antioxidants serum levels. Measured CAT, SOD and GPX were then compared between the groups as well as in each group alone. Blood samples were kept in EDTA tubes and centrifuged for 10 minutes at 448 relative centrifugal forces, and then, the gathered supernatant, which was plasma, was kept at -20°c until the time of biochemical measurement.

To analyze the data, chi-square test, paired t-test, ANOVA, independent t-test, Kruskal–Wallis, and Wilcoxon were used. The confidence interval was %95

Table 1: Preoperative and postoperative serum levels of the studied enzymes in both groups.

Serum Level		**Isoflurane (mean±SD)**	**Propofol (mean±SD)**
Preoperative	**Catalase**	8.1±3.8	7.8±2.6
	Glutathione	207.7±54.9	186.8±68
	Superoxide dismutase	7.6±3.4	5.8±0.9
Postoperative	**Catalase**	10.6±4.1	11.6±4.2
	Glutathione	159.9±56.4	162.5±69.8
	Superoxide dismutase	7.5±2.8	5.7±1.9

Table 2: Changes in serum levels of the enzymes in both groups after surgery.

Enzymes and groups		Mean±SD	t	df	P value
CAT	Isoflurane	-2.5±5.3	-2.1	19	0.05
	Propofol	-3.8±4.8	-3.6	19	0.002
GPX	Isoflurane	47.8±64	3.3	19	0.003
	Propofol	24.2±82.9	1.3	19	0.208
SOD	Isoflurane	0.099±2.3	Z=-0.299		0.77
	Propofol	0.096±1.4	Z=0.635		0.53

with 0.05 as type-1 error to get 80% study power. Kolmogrov-Smirnov and Shapiro-Wilk were the tests for normality assessment.

All the private information was confidential. All the patients gave their written informed consents before enrolling into the study after clear explanation about the aim and trend of the study by the investigators. Helsinki declaration was considered throughout the performance. There were changes in neither the routine treatment nor extra charge or procedure for the participants during the study.

Results

The current study enrolled 40 participants for two groups of isoflurane and propofol (Figure1). The former group included 40% females and 60% males which were 35% and 65% in the latter group, respectively (p=0.74). The mean age of the patients was 47.6±11 years in the group of isoflurane and 47.1±12.2 years in the other with no significant difference.

Table 3: Comparison of serum level of the studied enzymes regarding fentanyl administration in two groups via ANOVA.

			Sum of squares	df	Square mean	F	P value
Isoflurane	**CAT**	**Between groups**	198.1	4	49.5	2.2	0.12
		Into groups	337.8	15	22.5		
		Overall	535.9	19	-		
	GPX	**Between groups**	19120.6	4	4780.2	1.2	0.34
		Into groups	58765.4	15	3917.7		
		Overall	77885.9	19	-		
Propofol	**CAT**	**Between groups**	59.4	3	19.8	0.858	0.48
		Into groups	369.1	16	23.1		
		Overall	428.5	-	-		
	GPX	**Between groups**	2378.4	3	792.8	0.099	0.96
		Into groups	128461.5	16	8028.8		
		Overall	130839.9	19	-		
Isoflurane	**SOD**	Chi square=6.7		4	-	-	0.2
Propofol	**SOD**	Chi square=2.8		3	-	-	0.4

Table 4: Comparison of serum levels of the studied enzymes regarding the duration of surgery in two groups.

			Sum of squares	df	Squares mean	F	P value
Isoflurane	**CAT**	**Between groups**	246	8	30.8		
		Into groups	289.9	11	26.4	1.2	0.39
		Overall	535.9	19	-		
	GPX	**Between groups**	40657.6	8	5082.2		
		Into groups	37228.4	11	3384.4	1.5	0.26
		Overall	77885.9	19	-		
	SOD	Chi square=12.2		8	-	-	0.14
Propofol	**CAT**	**Between groups**	284.2	7	40.6		
		Into groups	44.4	12	12	3.4	0.03
		Overall	428.5	19	-		
	GPX	**Between groups**	31262.3	7	4466		
		Into groups	99577.5	12	8298.1	0.53	0.79
		Overall	130839.9	19	-		
	SOD	Chi square=8.5		7	-	-	0.29

Serum levels of CAT, GPX and SOD were measured before and after surgery in both groups as can be seen in table 1. To assess the normal distribution of the variables, Kolmogorov-Smirnov test and Shapiro-Wilk test were used to show that serum SOD had no normal distribution ($p<0.05$) while the others were normally distributed variables.

Paired t-test showed a significant increase in serum CAT level in the group of propofol ($p=0.002$) but isoflurane group showed no significant change before and after surgery. On the other hand, serum GPX decreased significantly after surgery in the isoflurane group ($p=0.003$) but not in the other group. Concerning SOD, Wilcoxon test disclosed that no group had significant reduction postoperatively. Table 2 summarizes all the relevant findings in this regard.

ANOVA failed to find significant correlation between the consumption of fentanyl during surgery and perioperative serum levels of CAT and GPX. Kruskal-Wallis test showed similar finding for serum SOD and the value of the used fentanyl as table 3 shows. ANOVA showed a raise in serum level of CAT in the group of propofol as the only factor which was affected by the duration of surgery ($p=0.03$) but the duration of surgery correlated neither with serum CAT and GPX in the group of isoflurane nor with GPX in the group of propofol as Kruskal-Wallis test represented (table4).

Age did not correlate with serum CAT and GPX changes using ANOVA. Likely, Kruskal-Wallis test showed no correlation between age and SOD changes (Table 5). Sex correlated with no enzyme changes, as table six shows, by the relevant statistical test as well (independent t-test and Kruskal-Wallis test).

Discussion

The current study found significant increase in serum level of CAT in the group of propofol ($p=0.002$) but not in the other group who used isoflurane. On the contrary, there was obvious decrease in serum level of GPX after surgery in isoflurane users ($p=0.003$) which did not occur in the other group. On the other hand, no significant reduction was found in the serum level of SOD after surgery in both groups. Changes in serum levels of the studied enzymes were independent from age, sex, fentanyl dosage, or duration of surgery

Table 5: Table5: Comparison of serum levels of the studied enzymes regarding patients' age in two groups.

			Sum of squares	df	Squares mean	F	P value
Isoflurane	**CAT**	**Between groups**	370.7	13	28.5		
		Into groups	165.2	6	27.5	1.03	0.5
		Overall	535.9	19	-		
	G	**Between groups**	60873.8	13	4682.6		
		Into groups	17012.2	6	2835.4	1.7	0.28
		Overall	77885.9	19	-		
	SOD	Chi square=12.5		13	-	-	0.49
Propofol	**CAT**	**Between groups**	263.7	15	17.6		
		Into groups	164.8	4	41.2	0.43	0.89
		Overall	428.5	19	-		
	G	**Between groups**	84265.1	15	5617.7		
		Into groups	46574.7	4	11643.7	0.48	0.87
		Overall	130839.9	19	-		
	SOD	Chi square=14.9		15	-	-	0.46

Table 6: Comparison of serum levels of the studied enzymes regarding patients' sex in two groups.

		Females (mean±SD)	**Males (mean±SD)**	**df**	**t**	**P value**
Isoflurane	**CAT**	0.85±6.5	3.5±4.3	18	-1.1	0.28
	GPX	68.3±63.6	34.1±63.2	18	-1.2	0.25
	SOD	Chi=1.5	-	1	-	0.21
Propofol	**CAT**	4.4±6.8	3.5±3.5	18	0.43	0.67
	GPX	38.7±57.9	16.4±95.1	18	-0.56	0.58
	SOD	Chi=0.07	-	1	-	0.78

(unless for catalase in propofol group).

Increased serum levels of antioxidant enzymes were reported before, particularly during major surgeries. Li et al, found obvious decrease in the levels of SOD, GPX and CAT after four hours of craniotomy that were corrected simultaneously after 24 hours. This showed the fact that change in enzyme levels in serum are temporary after craniotomy that is generally consistent with the current study (16). Propofol is known as an antioxidant medication and de la Cruz et al, showed its effect on GPX stimulation by platelets in rats during surgery. This would be resulted by limited lipid peroxidase to change GPX antioxidative system (11).

Through an in vitro study it was determined that propofol could inhibit oxidative stress-induced lipid peroxidation in liver, microsomes, mitochondria and brain synaptosomes (19) although it seems that propofol is not active in this regard in low doses (<10 mg/ml) (20).

Superoxide dismutase is a vital enzyme in charge for scavenging superoxide radicals to play its

antioxidant role in aerobic cells. Despite no changes in SOD during the current study and likely in Ucar's, Turkan et al, indicated that anesthetics but isoflurane decreased GPX and SOD in stress oxidative conditions like surgery (12, 21, 22). Lin et al, concluded in 2019 that metformin could improve anesthetic effects of propofol in lower dosage as well as increasing in SOD antioxidant activity to inhibit free radicals in mice (23).

Another study by Yang et al. determined that both sevoflurane and isoflurane could attenuate oxidative stress in rat model during surgery via reduced malondialdehyde (MDA) and nitric oxide (NO) and elevated SOD serum levels (24). In 2016, a group led by Luo showed significant increase in SOD release during craniotomy resection after using dexmedetomidine in a group of patients with brain glioma in comparison with another group who received normal saline instead (25).

In spite of controversy in terms of the antioxidant effects of anesthetics through changes in antioxidant enzymes, it seems that the majority of studies believe in positive effects in this regard or at least they offered some protocols to boost antioxidant activity such as additional medications like metformin.

This study performed in limited number of patients in just one hospital, authors recommend extended number of patients and multicenter approach for future similar studies.

Conclusion

The current study showed increased and decreased levels of CAT and GPX, respectively in propofol and isoflurane maintained anesthesia, which reflects the better antioxidant ability of propofol, while SOD changes had lower sensitivity to both medications. More studies are needed with bigger sample size focusing on all kinds of surgery using the evaluated medications to get better results, especially for SOD response.

Acknowledgment

Authors are grateful to anesthesia research center of SBMU for their cooperation.

Conflicts of Interest

The authors declare that they have no conflict of interest.

References

1. Endo H, Nito C, Kamada H, Nishi T, Chan PH. Activation of the Akt/GSK3beta signaling pathway mediates survival of vulnerable hippocampal neurons after transient global cerebral ischemia in rats. J Cereb Blood Flow Metab. 2006;26:1479-89.
2. Wang JK, Yu LN, Zhang FJ, Yang MJ, Yu J, Yan M. Postconditioning with sevoflurane protects against focal cerebral ischemia and reperfusion injury via PI3K/Akt pathway. Brain Res. 2010;1357:142-51.
3. Calabrese V, Giuffrida Stella AM, Calvani M, Butterfield DA. Acetylcarnitine and cellular stress response: roles in nutritional redox homeostasis and regulation of longevity genes. J Nutr Biochem. 2006;17(2):73-88.
4. Pignatelli P, Tellan G, Marandola M. Effect of L-carnitine on oxidative stress and platelet activation after major surgery. ActaAnaesthesiol Scand. 2011;55(8):1022-8.
5. Cui Ke, Luo X, Xu K, Murthy MRV. Role of oxidative stress in neurodegeneration: recent developments in assay methods for oxidative stress and Nutraceuticalantioxidants. Prog Neuro-Psychopharmacol Biol Psychiatry. 2004;28:771-99.
6. Cao Y, Wang YX, Liu CJ, Wang LX, Han ZW, Wang CB. Comparison of pharmacokinetics of L-carnitine, acetyl-L-carnitine and propionyl-Lcarnitine after single oral administration of L-carnitine in healthy volunteers. Clin Invest Med. 2009;32(1):E13-E19.
7. Ye J, Li J, Yu Y, Wei Q, Deng W, Yu L. L-Carnitine attenuates oxidant injury in HK-2 cells via ROS–mitochondria pathway. Regul Pept. 2010;161:58-66.
8. Herbert V, Shaw S, Jayatilleke E, Stopler-Kasdan T. Most free-radical injury is iron-related: it is promoted by iron, hemin, holoferritin and vitamin C, and inhibited by desferoxamine and apoferritin. Stem Cells. 1994;12**:**289-303.
9. Lee JY. Oxidative stress due to anesthesia and surgical trauma and comparison of the effects of propofol and thiopental in dogs. J Vet Med Sci. 2011;74:663e5.
10. Tsuchiya H, Ueno T, Tanaka T. Comparative study on determination of antioxidant and membrane activities of propofol and its related compounds. Eur J Pharm Sci. 2010;39:97–102.
11. De La Cruz JP, Zanca A, Carmona JA. The effect of propofol on oxidative stress in platelets from surgical patients. Anaesth Analg. 1999;89:1050e5.
12. Ucar M, Ozgül U, Polat A, Toprak HI, Erdogan MA, Aydogan MS, Durmus M, Ersoy MO. Comparison of antioxidant effects of isoflurane and propofol in patients undergoing donor hepatectomy. Transplant Proc. 2015;47(2):469-72.
13. Braz MG, Braz LG, Freire CM, Lucio LM, Braz JR, Tang G, Set al. Isoflurane and propofol contribute to increasing the antioxidant status of patients during minor elective surgery: A randomized clinical study. Medicine (Baltimore). 2015;94(31):e1266.
14. Han C, Ding W, Jiang W. A comparison of the effects of

midazolam, propofol and dexmedetomidine on the antioxidant system: A randomized trial. ExpTher Med. 2015; 9(6):2293-8.
15. Zhang Y, Tian SY, Li YW, Zhang L, Yu JB, Li J, et al. Sevoflurane preconditioning improving cerebral focal ischemia-reperfusion damage in a rat model via PI3K/Akt signaling pathway. Gene. 2015;569(1):60-5.
16. Li HT, Zhao ZH, Ding HY, Wang LX, Cao Y. Effect of craniotomy on oxidative stress and its effect on plasma L-carnitine levels. Can J PhysiolPharmacol. 2014;92(11):913-6.
17. Cobley JN, Fiorello ML, Bailey DM. 13 reasons why the brain is susceptible to oxidative stress. Redox Biol. 2018;15:490-503.
18. Osier N, McGreevy E, Pham L, Puccio A, Ren D, Conley YP, Alexander S, Dixon CE. Melatonin as a Therapy for Traumatic Brain Injury: A Review of Published Evidence. Int J Mol Sci. 2018;19(5).
19. Musacchio E, Rizzoli V, Bianchi M, Bindoli A, Galzigna L. Antioxidant action of propofol on liver microsomes, mitochondria and brain synaptosomes in the rat. Pharmacol Toxicol. 1991;69: 75e7.
20. Green TR, Bennett SR, Nelson VM. Specificity and properties of propofol as an antioxidant free radical scavenger. ToxicolAppl Pharmacol. 1994;129:163e9.
21. Turkan H, Bukan N, Sayal A, Aydin A, Bukan MH. Effects of halothane, enflurane, and isoflurane on plasma and erythrocyte antioxidant enzymes and trace elements. Biol Trace Elem Res. 2004;102:105e12.
22. Sudakshina Gh, Willard B, Comhair S, Dibello P, Xu W, Sruti Sh, et al. Disulfide Bond as a Switch for Copper-Zinc Superoxide Dismutase Activity in Asthma. Erzurum Antioxid Redox Signal. 2013;18(4):412-23.
23. Liu NH, Zhu L, Zhang XB, Chen Y. Metformin with propofol enhances the scavenging ability of free radicals and inhibits lipid peroxidation in mice. Eur Rev Med Pharmacol Sci. 2019;23(11):4980-7.
24. Yang P, Du Y, Zeng H, Xing H, Tian C, Zou X. Comparison of inflammatory markers between the sevoflurane and isoflurane anesthesia in a rat model of liver ischemia/reperfusion injury. Transplant Proc. 2019;5(19)30290-8.
25. Luo X, Zheng X, Huang H. Protective effects of dexmedetomidine on brain function of glioma patients undergoing craniotomy resection and its underlying mechanism. Clin Neurol Neurosurg. 2016;146:105-8.