NEURODEGENERATIVE CHANGES IN THE RAT BRAIN INDUCED BY ISOFLURANE AND IRON-DEXTRAN

UNIVERSITY OF ZAGREB FACULTY OF SCIENCE Department of Biology

Nikola Lesar, Dyana Odeh, Gordana Brozović, Nada Oršolić

Department of Animal Physiology, Faculty of Science, University of Zagreb, Croatia

SVEUČILIŠTE U ZAGREBU 1946 PMF 1876 PRIRODOSLOVNO - MATEMATIČKI FAKULTET

Abstract

Isoflurane, inhalation anesthetic, is commonly used to rapidly induce unconsciousness as well as analgesia. In CNS, Isoflurane can have neurotoxic effects by promoting oxidative stress and DNA damage. Same effects can be induced by iron, which is heavy metal that plays an important role in the production of myelin, metabolism of monoamine transmitters and GABA synthesis in brain but the accumulation of iron in the brain changes brain cell metabolism and leads to increased oxidative stress and neurodegeneration.

Aim of this study was to research the possible antioxidant/prooxidative effect of Isoflurane and Fe-dextran, as well as their combined interaction on the rat brain tissue samples also investigate the oxidative status of brain tissue which indicate clear neurotoxic effect and consequent neurodegeneration.

Methodology

Neurodegenerative changes in rats were induced by intraperitoneal subchronical doses (50 mg/kg animal weight) of iron-dextran solution,and by Isoflurane (1.5 %) anaesthesia during the 28 day period . Experimental animals (three months-old rats of both sexes from highly fertile Y59 strain) were divided into four groups with six animals in each.

Neurodegeneration was evaluated by:

a) changes in the oxido-reduction status by measuring the level of malondialdehyde (MDA) (Fig. 1.) and glutathione (GSH) (Fig. 2.), activity of catalase (CAT) (Fig. 3.) and superoxide dismutase (SOD) (Fig. 4.) in brain tissue

b) evaluation of the neuro-inflammation, by measuring the relative weight of the brain compared to a healthy control group (Fig. 5.)

c) analyzing osmotic fragility of erythrocytes (Fig. 6.)

Experimental group	Experimental chemicals	Dose	Administration	Treatment period (days)
0. Control	0.9 % NaCl	0.5 mL every other day	ip (intraperitoneal)	28
1. Isoflurane	Isoflurane,	1.5 % every other day	inhalation	28
2. Fe-dextran 50 mg/kg	Fe-dextran 50 mg/kg in 0.5 mL pro aqua	0.5 mL every other day	ip (intraperitoneal)	28
3. Fe-dextran 50 mg/kg + Isoflurane	Fe-dextran 50 mg/kg in 0.5 mL pro aqua + Isoflurane	0.5 mL of experimental solution + Isoflurane every other day	ip + inhalation	28

Results

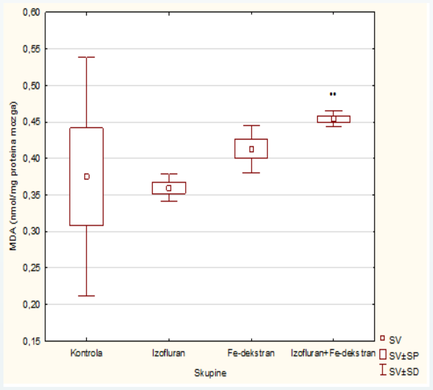

Fig. 1. Increased concentration of MDA was observed in the group treated with a combination of Isoflurane and Fe-dextran compared to the Isoflurane group, where only a statistically significant difference was recorded (P <0.01).

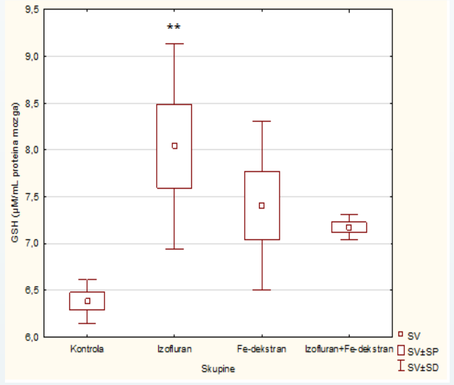

Fig. 2. The results of measurement of total GSH concentration shows an increase in GSH concentration in all treated groups compared to the control group, but only a statistically significant difference was observed in the Isoflurane-treated group (P <0.01). The results indicate that the body tries to defend itself by creating increased concentrations of GSH, as one of the most important mechanisms of antioxidant defense whose high concentration protects against ROS (reactive oxygen species).

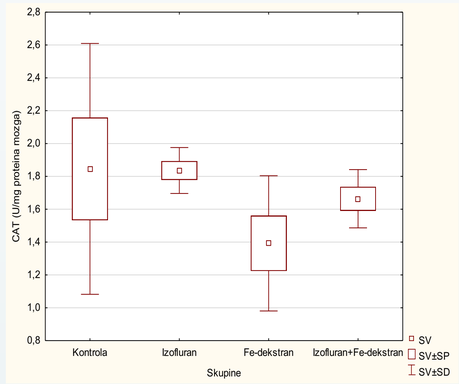

Fig. 3. The results of measuring the enzyme activity of catalase in rat brain tissue samples do not show a statistically significant difference in any of the treated groups compared to the control.

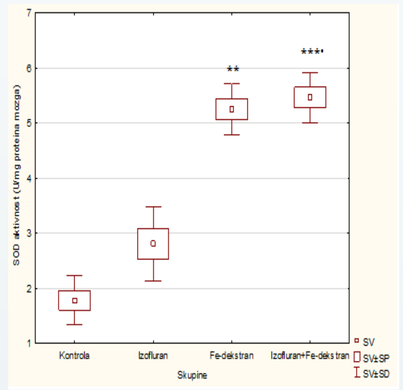

Fig.4. A statistically significant difference in the measurement of SOD enzyme activity was observed in the group treated with Fe-dextran (P <0.01) and the group treated with a combination of Isoflurane and Fe-dextran (P <0.001) compared to the control group, and in the group treated with Isoflurane <0.05) relative to the group treated with a combination of Isoflurane and Fe-dextran.

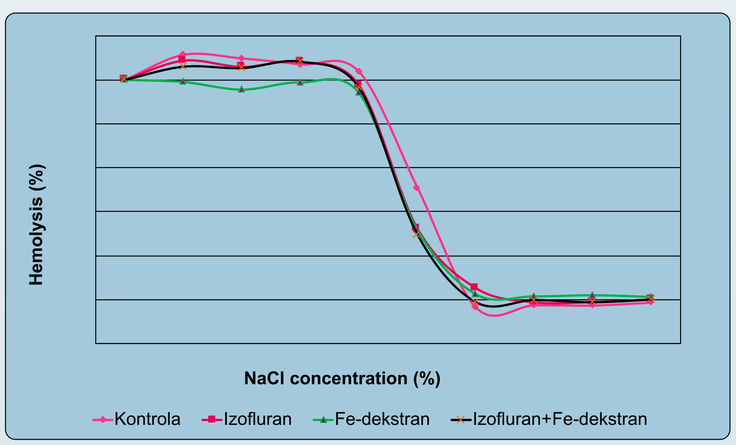

Fig. 5. The control group had slightly higher hemolysis compared to the other treated groups. Analysis of the results showed that significant hemolysis occurred at 0.5% NaCl concentration in the control group, while in all treated groups it was observed that 50% of hemolysis occurred at a slightly lower NaCl concentration.

We can assume that in the treated groups by the action of Isoflurane and Fe-dextran there was a faster hemolysis due to increased oxidative stress, which consequently leads to the deterioration of mature erythrocytes during the treatment period of 28 days.

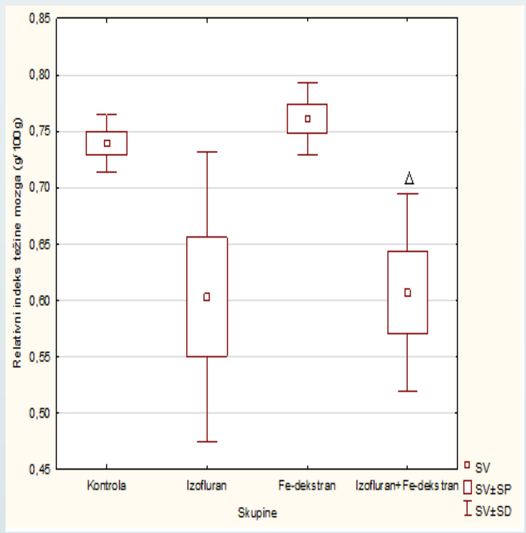

Fig. 6. The relative rat brain index is decreased in the Isoflurane- reated group and the Isoflurane + Fe-dextran treated group compared to the control group.

Conclusion

We can make conclusion that the used dose of both compounds in combination was sufficient for the occurrence of oxidative stress which led to increased lipid peroxidation and an increase in the concentration of MDA.

Based on the research above which shows decreased relative brain index, it is obvious that repeated anesthesia with Isoflurane or in combination with Fe-dextran leads to increased deterioration of brain cells.

This research suggest that different conditions and lengths of Isoflurane exposure in the presence of iron can certainly be different considering iron accumulation in individual regions as well as the complex mechanisms that take place during iron loading in brain tissue.

A better understanding of all these important points of view will significantly improve our knowledge of iron metabolism in the brain and the use of inhalation anesthetics as well as their role in the development of oxidative stress and neurodegenerative disorders.