



THE EFFECTS OF HYPERBARIC OXYGEN APPLIED FOR ONE AND FIVE DAYS ON BLOOD BRAIN BARRIER PERMEABILITY IN RATS

Selçuk Tatar¹, Nadir Arıcan², Nurcan Orhan³, Canan Ugur Yılmaz⁴, Bulent Ahishali⁵, Mehmet Kaya⁷, Akin Savas Toklu¹

Departments of Underwater and Hyperbaric Medicine¹, Forensic Medicine², Physiology³, Histology and Embryology⁴ of Istanbul Faculty of Medicine, and Departments of Neuroscience⁵, Department of Laboratory Animals Biyology and Biomedical Application Techniques⁴ of Institute of Experimental Medicine, ISTANBUL UNIVERSITY, ISTANBUL, TURKEY



INTRODUCTION

The blood-brain barrier (BBB) composed predominantly by the brain capillary endothelial cells plays a fundamental role in maintaining brain homeostasis under physiological conditions. The studies about the effect of hyperbaric oxygen (HBO) on BBB permeability are limited and these studies have different results. In these studies the tracers used to check BBB permeability and HBO protocols are varied. There are still questions about HBO and BBB to be answered. In this experimental study, the effects of HBO applied for one and five days, on BBB permeability and oxidant-antioxidant systems were investigated in rats.

Questions About HBO & BBB;

- Is there any side effects because of changes in BBB integrity?
- Does HBO increase the side effects of other treatments?
- Can HBO be used to increase BBB permeability?
- Can HBO be used to repair disrupted BBB?
- Is it the pressure or oxygen that has effects on BBB?
- In which doses HBO has effects on BBB?
- For which molecules HBO increase the permeability? Which kind of transport is effected in BBB by HBO?

MATERIAL and METHODS

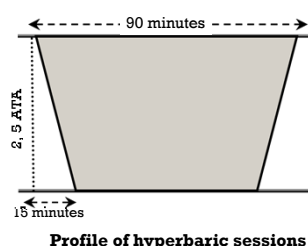
Healthy young female Wistar albino rats were divided into three groups as, control, HBO administered for 1 day (HBO-1) and HBO administered for 5 days (HBO-5) groups. HBO-1 and HBO-5 groups were divided into three subgroups, each consisted of eight rats, to investigate oxidant-antioxidant levels, immunohistochemical and electron microscopic changes. Hyperbaric oxygen sessions were set as; compression in 15 min, 100% oxygen for 60 min at 2,5 ATA and decompression in 15 min. Three HBO sessions were applied to HBO-1 group with 6 hours intervals in 24 hours. Twelve HBO sessions were applied to the HBO-5 group as; 3 sessions a day in first two days and twice a day during the last three days. The rats were catheterized about 2 hours after the last hyperbaric sessions to inject horseradish peroxidase (HRP), to make the fixation for immunohistochemical evaluation, to take tissue samples for measurement of the oxidant-antioxidant level and decapitation. BBB permeability was examined electron microscopically by using HRP tracers.

What we looked for;

- **Oxidant and Antioxidant Levels**
Malondialdehyde (MDA), Glutathione (GSH), Superoxide dismutase (SOD)
- **Immunohistochemical Changes**
Glial fibrillary acidic protein (GFAP)
- **BBB Permeability by Electron Microscopy**
HRP as tracer

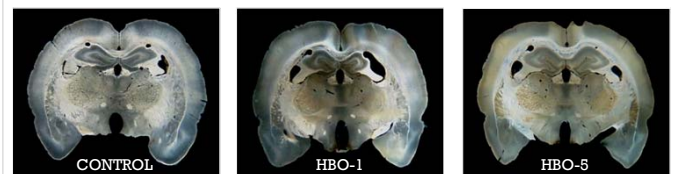


Experimental hyperbaric chamber

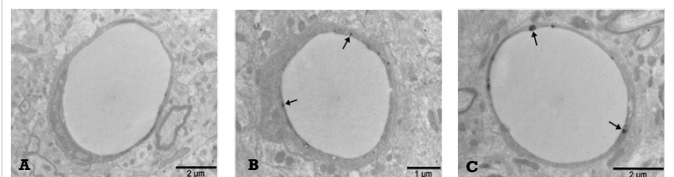


RESULTS

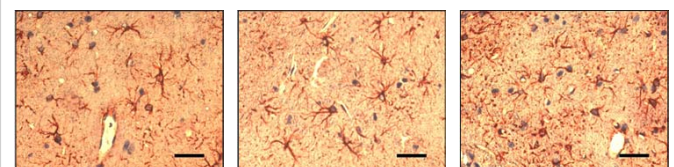
When examined by electron microscopy, we did not detect any HRP reaction product in brain section of BBB control group, but it was detected in animals in HBO-1 and HBO-5 groups. Immunohistochemically, no significant difference was detected in hippocampal regions of the rats in experimental groups. GSH levels in cerebral cortex and hippocampal tissues in HBO-1 and HBO-5 were measured significantly lower comparing to control group. SOD activities in both experimental groups were higher than control group. MDA levels in cerebral cortex and hippocampal tissues were lower in both HBO-5 and HBO-1 groups.



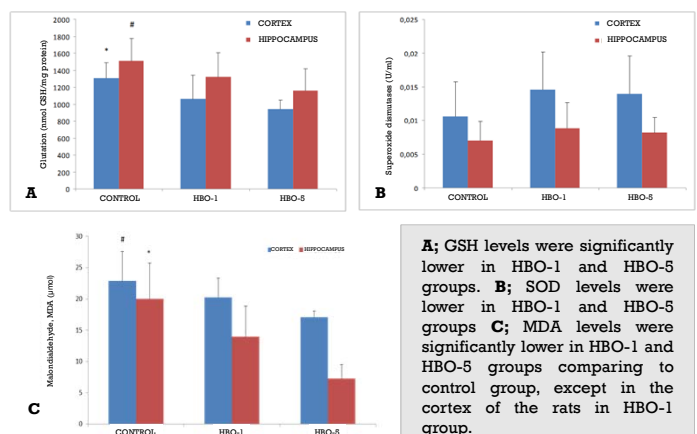
Photomicrographs showing the pattern of HRP extravasation in Vibratome sections



Electron micrographs of capillaries in hippocampus region.
A: No HRP in endothelial cells, B and C: HRP vesicles in endothelial cells (Arrows)



Immunohistochemically, no significant difference in GFAP was detected between groups



A; GSH levels were significantly lower in HBO-1 and HBO-5 groups. **B;** SOD levels were lower in HBO-1 and HBO-5 groups **C;** MDA levels were significantly lower in HBO-1 and HBO-5 groups comparing to control group, except in the cortex of the rats in HBO-1 group.

CONCLUSION

HBO administered to healthy rats for 1 and 5 days caused an increase in BBB permeability to HRP, decrease in MDA and GSH level in brain tissue and slight increase in SOD activity.