EFFECTS OF PROPOFOL ON EXPRESSION ICAM-1 IN RABBIT GASTRIC ENDOTHELIAL CELLS

Sennur Ketani*, Berna Ersoz Kanay2, Hakan Sagsoz3

1. Department of Biology Education, Education Faculty, University of Dicle, 21280 Diyarbakır-Turkey.
2. Department of Surgery Veterinary Medicine Faculty, University of Dicle, 21280 Diyarbakır-Turkey.
3. Department of Histology & Embriyology, Veterinary Medicine Faculty, University of Dicle, 21280 Diyarbakır-Turkey.

Abstract

It was examined the dose-dependent effects of propofol on expression intracellular adhesion molecule (ICAM-1) in rabbit gastric endothelial cells.

Twenty adult New Zeland albino rabbits were used in this study. One control and three experimental groups designed. In experimental groups 0.5, 4.0, 8.0 mg/kg propofol were applied to rabbits by marginal ear vein. One hour after applying propofol, control and experimental group rabbits were sacrificed and their gaster were removed.

The sections were stained with APAAP immunohistochemical staining for evaluation using a light microscope. No inflammatoric reactions were seen in sections of gastric endothelial cells of control and experimental groups.

Keywords: ICAM-1; rabbit; gastric endothelium; propofol.

Received date: 20 February 2010

Introduction

Propofol is widely used for the induction and maintenance of anesthesia and a sedative in intensive care units, where it is given as a constant intravenous infusion for periods of many days1,2.

Gastrointestinal mucosa is one of the most rapidly changing tissues in the body and the balance between cell regeneration and cell loss may lead to mucosal lesion and ulceration3.

Leucocytes are pivotal component of the inflammatory cascade that results in tissue injury in a large group of disorders such as ischemia, non-steroidal antiinflammatory drugs (NSAIDs) and ethanol1,4,5.

The major lines of evidence that implicate leucocytes in the tissue injury associated with them include; leucocytes accumulate in the gastric mucosa prior to or during the development of tissue injury and that deplation of leucocytes decreases the degree of injury. Free radical production and endothelial activation promote leucocyte-endothelium interaction via endothelial expression of vascular cell adhesion molecule 1 (VCAM-1) and intracellular adhesion molecule 1 (ICAM-1)1,4,5.

The aim of present study was to investigate the effects of different doses of propofol on expression intracellular adhesion molecule (ICAM-1) in rabbit gastric endothelial cells.

Material and Methods

Twenty adult New Zeland albino rabbits were used in this study and the sex of them wasn’t remarkable, 2000-2500 g in weight were obtained from the Department of Medical Science Application and Research Centre of Dicle University (DUSAM).

They were housed in individual cages in temperature-controlled environment (22°C) with a 12:12 h light-dark cycle. All rabbits were fed standard pellet food and adlibitum tap water, which were performed according to the Declaration of Helsinki with the permission of the Governmental Animal Protection Committee.

*Corresponding author:
Dr. Sennur KETANI
Department of Biology Education, Education Faculty, University of Dicle, 21280 Diyarbakır-Turkey
E-mail: ketani.sennur@gmail.com
Group I (Control group): In this group the interval of the study nothing was done to the rabbits (n:5).

Group II (0.5 mg/kg IV propofol applied): In this group 0.5 mg/kg propofol was applied to rabbits by marginal ear vein (n: 5).

Group III: (4.0 mg/kg IV propofol applied): In this group 4 mg/kg propofol was applied by marginal ear vein (n: 5).

Group IV: (8.0 mg/kg IV propofol applied). In this group 8 mg/kg propofol was applied by marginal ear vein (n: 5).

After 1 hour applying of propofol, control and experimental group rabbits were sacrificed and their gaster were removed.

Tissues were fixed for 6-8 hours in Bouin's solution at 4 ºC. They were dehydrated though increasing concentrations of the ethanol series and the tissues were embedded in paraffin and cut into 4-5µm transversal, dewaxed in xylene, and incubated for 20 minutes in 0.3% H2O2 to block endogenous peroxidase activity. Section then were microwaved for 4 minutes in 20 % goat serum in PBS in order to avoid undesired background staining, put into 20 minutes.

Monoclonal mouse anti-Human ICAM-1 (BioGenex San Ramon USA) primary antibody (dilution: 1/200) was applied to the sections for 3 hours at 37 ºC in a humidified staining chamber. Sections were then incubated in anti-mouse IgG secondary antibody (Lab Vision, dilution: 1/1000) for 1 hour, and they were put into the APAAP complex for an hour. Sections were mounted with a glycerol-PBS mixture (1:1 glycerol: PBS).

Following this step, sections were incubated in the fast red/TR naphtol mixture until the specific regions were stained red, and then the sections were either briefly put into Mayer’s hematoxilen in order to visualize the nuclei, or were not subjected to counterstaining. Sections were mounted with a glycerol-PBS mixture (1:1 glycerol: PBS).

The control staining of some sections was performed without the primary antibody, and no ICAM-1 positive immunostaining was observed in these sections (Figure 1).

Results

Immunohistochemical Examination

The aim of present study was to investigate the effects of propofol on expression intracellular adhesion molecule (ICAM-1) in rabbit gastric endothelial cells. There was no significance changes in immunoreactivity among section evenwithin groups, the difference between control and experimental groups was not clear (Table 1).

<table>
<thead>
<tr>
<th>Groups</th>
<th>Propofol Applied (IV)</th>
<th>ICAM-1 Expression</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group I Control group</td>
<td></td>
<td>No staining</td>
</tr>
<tr>
<td>Group II Experimental group</td>
<td>0.5 mg/kg</td>
<td>Weak staining</td>
</tr>
<tr>
<td>Group III Experimental group</td>
<td>4.0 mg/kg</td>
<td>Weak staining</td>
</tr>
<tr>
<td>Group IV Experimental group</td>
<td>8.0 mg/kg</td>
<td>Moderate staining</td>
</tr>
</tbody>
</table>

The control staining of some sections was performed without the primary antibody, and no ICAM-1 positive immunostaining was observed in these sections (Figure 1).

Figure 1 Immunohistological appearance of Control Group. The control staining was performed without the primary antibody. Immunogens show no positive staining, Lumen (L), Lamina epithelialis (le), lamina propria (lp), gastric gland (g), blood vessel (bv), (Original magnification X40, Scale Bar: 25µm).

Tunica mucosa of the stomachs of the rabbits in control and experimental groups were normal in histological examination (Figure 2,3,4,5).
In control group immunohistochemistry of ICAM-1 expression showed weak staining in gastric endothelial cells (Figure 2).

In group II (which were given 0.5 mg/kg propofol) showed weak staining with ICAM-1 (Figure 3).

In group III (which were given 4 mg/kg propofol) showed weak staining with ICAM-1 (Figure 4).

In group IV (which were given 8 mg/kg propofol) showed moderate staining with ICAM-1 (Figure 5).
Discussion

Nuclear factor kappa B can be activated by lesion induced oxidative stress, bacterial endotoxin or cytokines and subsequently increases transcriptionally the expression of the genes for many cytokines, enzymes and adhesion molecules, which have been believed to be involved in the acute inflammatory response. Adhesion molecules can recruit inflammatory cells, such as neutrophils, eosinophils and T lymphocytes from the circulation to the site of inflammation to release inflammatory mediators responsible for the gastric mucosal damage.

Antioxidants within cell membranes protect the phospholipids from free radical mediated lipid peroxidation and oxidative stress. α-Tocopherol (Vitamine E) is used to protect lipid from oxidation. This compound contains a phenol group that donates hydrogen to free radicals, thus terminating lipid peroxidation. Propofol is an i ntavenous anesthetic with a chemical structure similar to phenol-based free radical scavengers such as Vitamine E.

Reduction of free radical may improve outcomes of patients undergoing on surgeries. Common antioxidants such as vitamine E and buthylated hydroxytoluene can not be used routinely. Propofol may be the first candidate because of its anesthetic properties, rapid acting and recovering. Therefore it may have a protective role in gastric disorders and surgeries where free radical mediated injury promates leucocytes-endothelium adhesive interactions. ICAM-1 expression is strongly inducible by inflammatory cytokines. Here no immunohistochemical staining seen in groups shows that propofol had not caused inflammation. Ketamine is commonly used as an anesthetic agent in veterinary medicine. Kenneth et al., (2003) had mentioned that ketamine inhibits gastric injury. Because ketamine interacts with a number of inflammatory pathways and may be useful in inflammatory models of tissue injury.

Conclusions

In this study it has been shown that propofol do the same inhibition just as ketamine in gastric mucosa by the nonexpression of ICAM-1. Here it also shown that the anti-inflammatory effects of antioxidants and molecular mechanisms involved may help the understanding of the inflammatory cascade and may lead to future development for the propofol in the treatment of acute abdomen surgery and pathologic inflammation in animals.

Declaration of Interest

The authors report no conflict of interest and the article is not funded or supported by any research grant.

References