DOI: 10.4149/BLL_2012_035

Bratisl Lek Listy 2012; 113 (3)

145 – 151

EXPERIMENTAL STUDY

The comparison of the effects of hepatic regeneration after partial hepatectomy, silybum marinaum, propofol, N-acetylcysteine and vitamin E on liver

Yormaz S[1], Bulbuloglu E[1], Kurutas EB[2], Ciralik H[3], Yuzbasioglu MF[1], Yildiz H[4], Coskuner I[4], Silay E[4], Kantarceken B[5], Goksu M[1], Senoglu N[1], Kale IT[1]

Department of General Surgery, KSU, Faculty of Medicine, Kahramanmaras, Turkey.
Ertanbulbuloglu@yahoo.com.t

Abstract: *Aim:* We investigated the comparison of the effects of N-acetylcysteine, silybum marinaum, propofol, and vitamin E on liver hepatic regeneration after partial hepatectomy.
Method: Forty-eight rats were randomized into 6 different groups of the same age and weight. After partial hepatectomy, all animals were resuscitated with 5 ml of isotonic sodium chloride solution administered subcutaneously while group 1 (sham) did not receive any injection, group 2 (control) received serum physiologic intraperitoneally, group 3 received 25 mg /kg of propofol intraperitoneally, group 4 received 20 mg/kg of N-acetylcysteine intraperitoneally, group 5 received 400 mg/kg of vitamin E intraperitoneally, and group 6 received 10 mg/kg of silybum intraperitoneally. None of these groups were given antibitotics. On the third day, a half of the rats, and on the seventh day, the other half of rats were reoperated and sacrificed.
Results: Blood samples were used for biochemical parameters (AST, ALT). Ki-67 proliferation index was used for histopathologic parameters. A statistically meaningful difference was detected in silybum, vitamin E, N-acetylcysteine, and propofol groups for AST, ALT levels when compared to control and sham groups ($p<0.05$). Ki-67 regeneration proliferation index of all groups, which were given agents on the third and seventh days were statistically higher than the control and sham groups ($p<0.05$). During the evaluation, AST, ALT, Ki-67, Ro (regeneration value) levels of silybum group displayed a statistically significant difference according to other groups ($p<0.05$).
Conclusion: Our experimental study indicates that hepatic regeneration after partial hepatectomy was meaningful and significant in groups with intraperitoneal administration of silybum marinaum,vitamin E, N-acetylcysteine and propofol. Hepatic regeneration rate was particularly higher in silybum group compared to other groups *(Fig. 16, Ref. 26).* Full Text in PDF *www.elis.sk.*
Key words: partial hepatectomy, silybum marinaum, liver regeneration.

The liver is the largest internal organ of the human body. It has important roles in regulating the metabolic functions, and in the immune system. After resection or traumatic injury, the liver is a single organ capable of fast tissue regeneration. In order to maintain the existing liver functions after liver resection, the period and effect of hepatic ischemia before and during the resection should be reduced to minimum,. Besides, after the resection, the increase in hepatic blood flow and the reduction of inflammatory events are currently getting increasingly important.

The postoperative mortality rate in liver resection depends directly on preoperative liver functions and liver resection volume. After tissue injuries and hepatocellular necrosis, regeneration of the liver tissue remaining in normal parenchyma to replace the functional hepatic mass in a short time is a very important supportive mechanism. In hepatocytes, mitosis is very rare, yet within 24 hours after partial hepatectomy, active cell replication begins and continues until it reaches the initial weight of the organ. Regeneration occurs rather in a way of growth of residual lobules and formation of new lobules. Although the regeneration mechanism has become better understood based on studies conducted in 21st century, none of various chemical, physical or biological agents studied in association with liver regeneration have been brought to common clinical use. Propofol is an intravenous anesthetic agent in animal model of partial hepatectomy to reduce lipid peroxidation during liver regeneration, tissue antioxidant defense systems to strengthen the protective effect against oxidative damage.

After partial hepatectomy, an abnormal increase in free oxygen radicals revealed during liver regeneration is met by mitochondrial antioxidant enzymes, which become reduced depending on their use (1, 2). By bestowing cysteine, N-acetylcysteine enhances the glutathione synthesis in the lung and liver. However, it binds free oxygen radicals and thus prevents the cell damage (3, 4).

[1]Department of General Surgery, KSU, Faculty of Medicine, Kahramanmaras, Turkey, [2]Department of Pathology, Faculty of Medicine, Kahramanmaras Sutcuimam University, [3]Department of Biochemistry, KSU, Faculty of Medicine, Kahramanmaras, Turkey, [4]Department of Anesthesiology, KSU, Faculty of Medicine, Kahramanmaras, Turkey, and [5]Department of Gastroenterology, General Surgery, KSU, Faculty of Medicine, Kahramanmaras, Turkey

Address for correspondence: E. Bulbuloglu, MD, Department of General surgery, Faculty of Medicine, Kahramanmaras Sutcuimam University, Kahramanmaras, Turkey 46050.
Phone: +90.344.2212337, Fax: +90.344.2212371

The type of tocopherol that is most widely distributed in nature as a vitamin and found to have the greatest biological activity is α-tocopherol. α- Tocopherol (vitamin E) is a potent antioxidant, and especially affects the membrane lipid peroxidation. In monocytes and / or kupfer cells, tumor necrosis suppresses the production of factor-α, interleukin-1, interleukin-6, interleukin -8, and inhibits the gene expression of α1, collagen in the liver (5). Silymarin consisting of flavonolignans found in the composition of silybum marinaum plant, toxophylin, quercetin, albumin, substances such as mucilages demonstrate liver protecting features. Also, because this plant contains 70–80 % of silymarin, by showing an antioxidant effect it protects the liver from damage caused by free radicals. At the same time it is responsible for detoxification of liver hormones, drugs and chemicals (6).

Today, there is still a need of new treatment strategies for regeneration; therefore we aim to investigate the comparative effects of N-acetylcysteine, propofol, vitamin E (α-tocopherol) and silybum marianum we consider to be effective in liver regeneration after experimental partial hepatectomy due to its effects on the liver.

Materials and methods

We used 48 wistar albino male rats weighing 200–250 g in this study. Animals were housed at 24.5 °C and given standard rat chow diet and water ad libitum. The study protocol was approved by the animal ethic review committee of the Faculty of Medicine University of Kahramanmaras Sütcü Imam.

All rats were kept in standard cages in 6 groups for 7 days creating 12-hour light/dark cycles (on the seventh day after partial hepatectomy that's why regeneration was completed by 75 % that day was chosen). The rats chosen by randomization were divided into 6 groups consisting of 8 rats in each group. Operations were performed in the morning between 08:00 and 12:00. In this way the results were obtained without being affected by diurnal changes. The rats were left hungry 12 hours before the operation. Rats to be used in our study were divided into 6 groups of 8 animals in each group. No rat was given antibiotics.

Groups: Group 1 (sham): After partial hepatectomy had been applied on animals, hemorrhage control was done and the abdomen was closed. Group 2 (control): After partial hepatectomy had been applied on animals, hemorrhage control was done; 5 ml intraperitoneal injection of saline was applied, the abdomen was closed. Group 3: After partial hepatectomy had been applied on animals, hemorrhage control was done; 25 mg / kg intraperitoneal injection of propofol was applied, the abdomen was closed. Group 4: After partial hepatectomy had been applied on animals, hemorrhage control was done; 20 mg / kg intraperitoneal injection of N-acetylcysteine was applied, the abdomen was closed. Group 5: After partial hepatectomy had been applied on animals, hemorrhage control was done; after 400 mg / kg intraperitoneal injection of vitamin E was applied, the abdomen was closed. Group 6: After partial hepatectomy had been applied on animals, hemorrhage control was done; 10 mg / kg intraperitoneal injection of silybum was applied, the abdomen was closed.

Surgical procedures

All rats were anesthetized with 50 mg/kg ketamin hydrochloride (Ketalar, Eczacibasi.Türkiye) The abdomen was shaved and swabbed with a povidone iodine solution preoperatively. A 4-cm midline incision was made and the abdomen was opened under clean surgical conditions, after laparatomy as described, the left lateral and median lobe peduncles of the liver (consisting approximately 75 % of liver in rat) was resected from the vena cava conjunction, peduncles were tied with a 3/0 silk, the extracted liver tissues were weighed and recorded (7). Abdominal laparotomy incision lines of all subjects were closed with 4/0 silk and continuous double row sutures, and cleaned with baticon (mean operation time was approximately 15 minutes), all animals were resuscitated with 5 ml of isotonic sodium chloride solution administered subcutaneously. Postoperative needs of all rats were met with normal rat chow diet and water. Because 50 % and 75 % of the liver regeneration after partial hepatectomy occur on 3rd and 7th postoperative days, respectively, on these days 4 subjects from each group were sacrificed by forming hypovolemia from vena cava inferior, ensuring the sterile conditions. Then the liver tissue remaining for regeneration was removed completely and weighed on a sensitive scale, the extracted tissues were retained for pathological examination, saved in 10 % neutral buffered and formulated Petri containers. Blood drawn while subjects were sacrificed was taken to the Eppendorf tubes, centrifuged for 10 minutes at 1500 rpm and stored at –80 °C until the day of study. On the day of study, plasma in dissolved serum samples was separated and AST and ALT parameters were measured.

Morphological parameters were calculated by subtracting the remaining liver weight after resection from the liver weight at autopsy (estimated 30 %), then by dividing the liver weight removed in the resection (8). The rate of regeneration was found by multiplying 100 to the value obtained. The whole weight of the liver was considered to be 3.4 % of the rat weight (9).

Biochemical assay: Liver Function tests: Blood samples were centrifuged at 1500 rpm for 10 minutes for taking plasma. Plasma was measured by Cobas Integra 800 model biochemistry autoanalyzer. Biochemistry parameters of AST and ALT (IU / LT) were ROCHE brand animal kits.

Histopathological parameters: Immunohistochemical applications: Histopathological assessment was performed at University Faculty of Medicine Department of Pathology, University Hospital Laboratory. Tissue samples were identified in 10 % buffered formalin for histopathologic examination. We used 5-micrometer thick sections prepared from paraffin blocks for slides with poly-L-Lysine for determining the Ki-67 expression. Ki-67 (SP6) (Neomarkers, USA), ready to use immunohistochemical staining procedure was performed with rabbit-rat monoclonal antibody. We used Wintzer et al technique for ki-67 staining pattern. The number of cells showing nuclear staining with Ki-67 % of the total cell number was calculated (10–14).

Statistical analysis

All variables were expressed as mean and standard error. Differences between groups were evaluated by Kruskal–Wallis

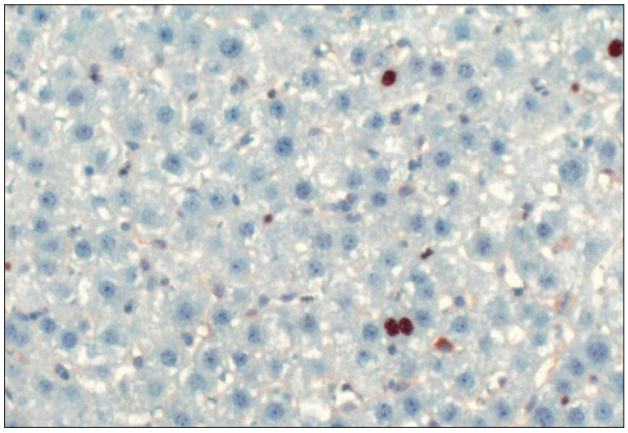


Fig. 1. 3rd day: Control Group, the percentage of nuclear stained cells slightly increased.

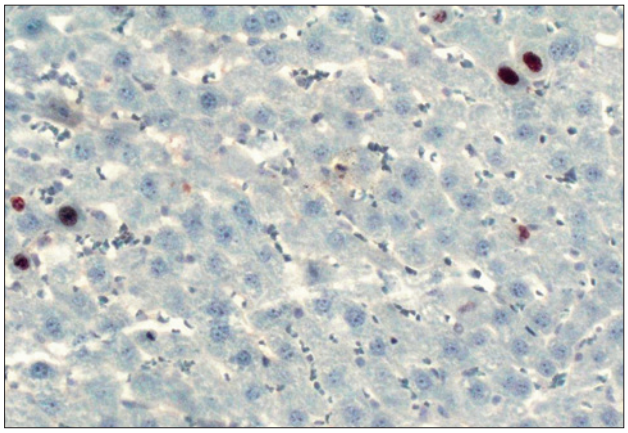


Fig. 2. 7th day: Control Group, the percentage of nuclear stained cells slightly increased.

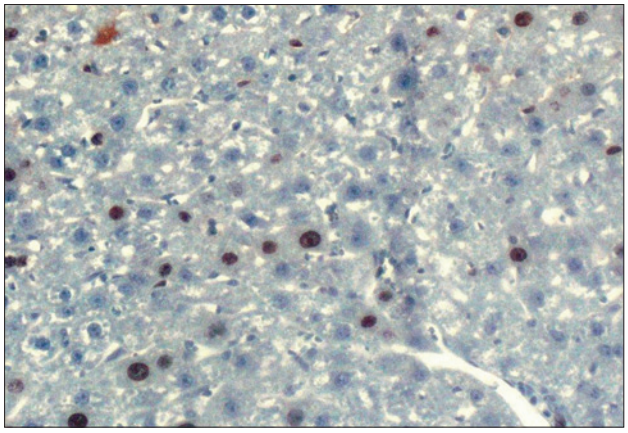


Fig. 3. 3rd day: Sham Group, the percentage of nuclear stained cells slightly increased.

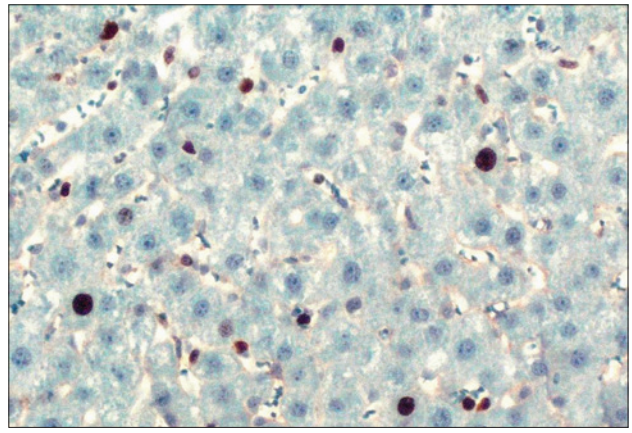


Fig. 4. 7th day: Sham Group, the percentage of nuclear stained cells slightly increased.

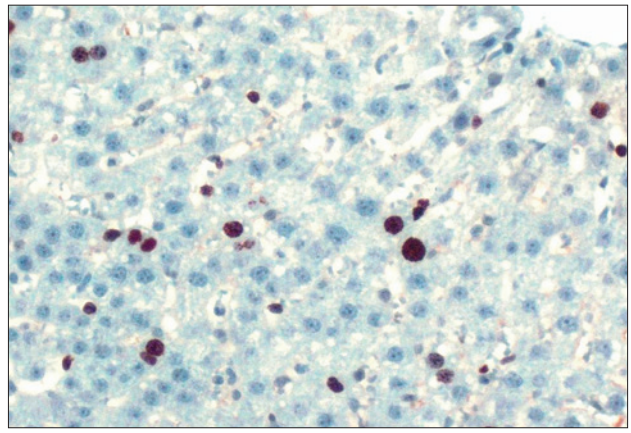


Fig. 5. 3rd day: NAC Group, the percentage of nuclear stained cells slightly increased.

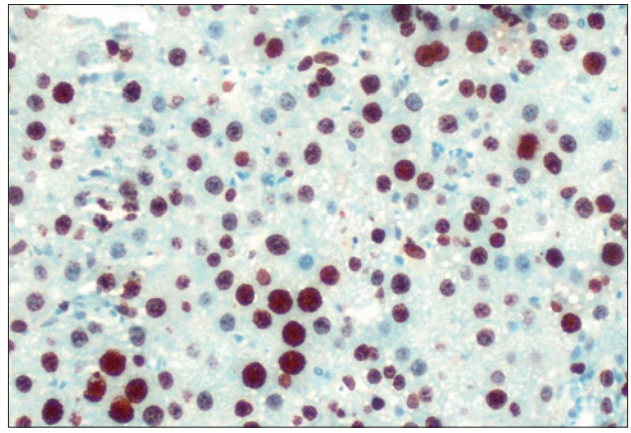


Fig. 6. 7th day: NAC Group, the percentage of nuclear stained cells increased middle degree.

variance analysis followed by a post hoc Mann Whitney U test. p values <0.05 were considered statistically significant. All data were entered into and processed by SPSS Inc 18.00 for Windows statistical package.

Results

The results obtained in this study in which the effects of N-acetylcysteine, propofol, vitamin E, Silybum marianum on liver

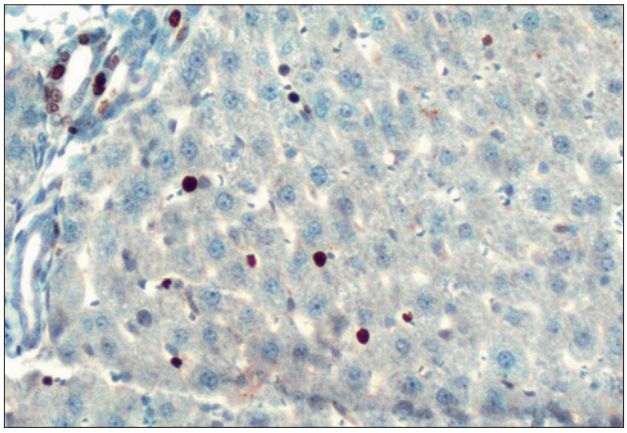


Fig. 7. 3rd day: Propofol Group, the percentage of nuclear stained cells slightly increased.

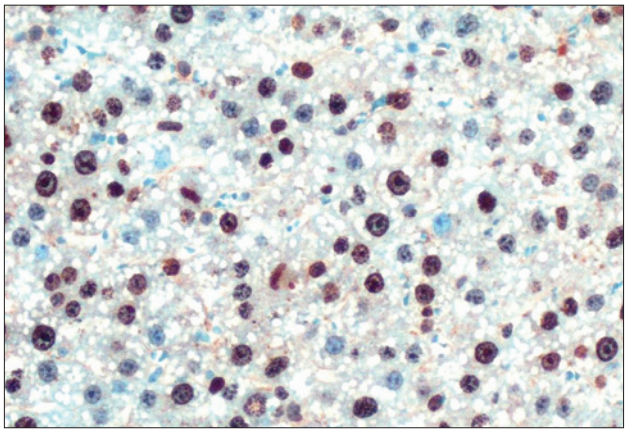


Fig. 8. 7th day: Propofol Group, the percentage of nuclear stained cells increased middle degree.

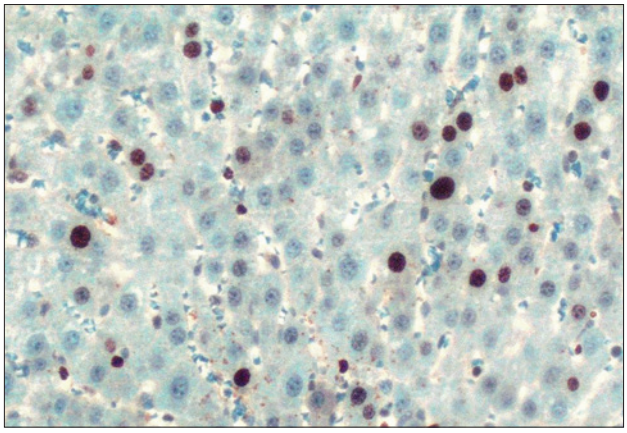


Fig. 9. 3rd day: Silybum Group, the percentage of nuclear stained cells increased middle degree.

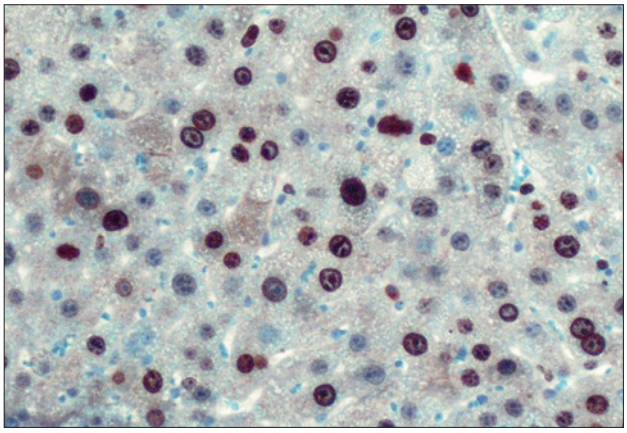


Fig. 10. 7th day: Silybum Group, the percentage of nuclear stained cells increased high degree.

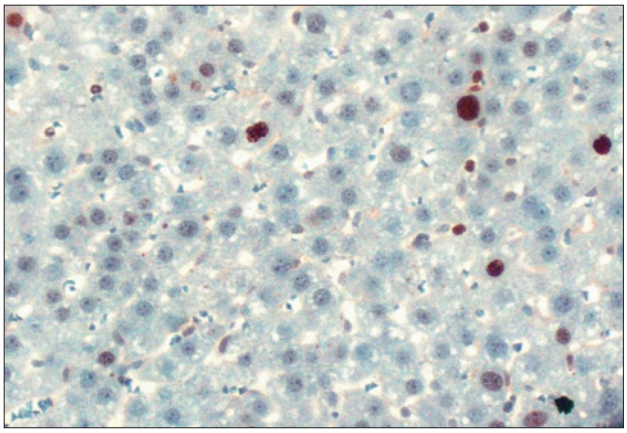


Fig. 11. 3rd day: Vitamin E Group, the percentage of nuclear stained cells slightly increased.

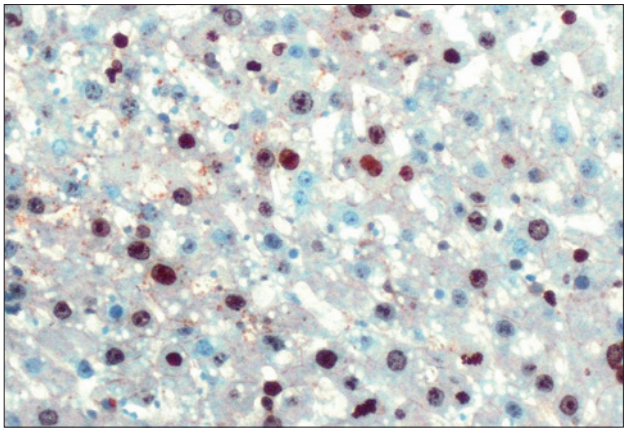
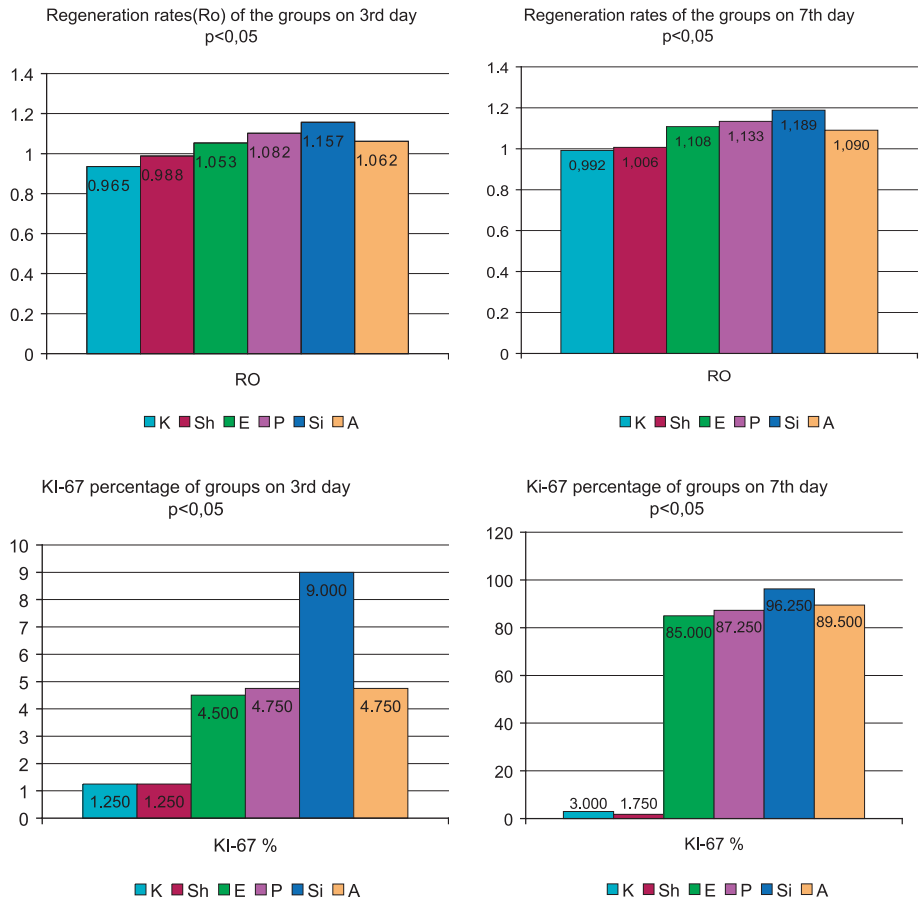


Fig. 12. 7th day: Vitamin E Group, the percentage of nuclear stained cells increased middle degree.

regeneration in the experimental partial hepatectomy were investigated are as follows (Figs 1–12):

1) Serum AST and ALT levels representing the liver functional biochemical reserve in the study and control groups were investigated. To summarize AST and ALT values on the third and seventh days, whereas the difference between the control group and the sham group is not statistically significant within this time period ($p>0.05$), the difference between the control group and each of vitamin E, propofol and NAC groups was found to be statistically significant ($p<0.05$). The difference between vitamin



Figures 13–16. Comparison of regeneration rates and Ki-67 percentages of the groups on the 3rd and 7th days. Our results showed that the highest regeneration rate and Ki-67 % group is the silybum marinaum group. Silybum group is the best group compared to other groups (K: Controlgroup, Sh: Sham group, E: Vitamin E group, P: Propofol group, Si: Silybum Marinaum group, A: N-acetylcysteine (NAC) group).

E, propofol and N-acetylcysteine groups were not found to be statistically significant (p>0.05). In addition, the difference between the control, sham, vitamin E, propofol and NAC groups and the group in which we applied silybum was found to be statistically significant (p<0.05). If we compare the values on the third and seventh days with each other, although there is a numerical difference between each AST value of the control and sham groups, on the 3rd and 7th days, this difference is not statistically significant (p>0.05). It is confirmed that there is a numerical difference between the measured values of each group of vitamin E, propofol, Nac on the 3rd and 7th days, and this difference is statistically significant (p<0.05). It is determined that AST values of the silybum group decrease by 43 % from the 3rd day to the 7th day and this difference is statistically significant (p<0.05). It has been observed that agents used in line with the biochemical results in varying proportions have positive effects on the protection and regeneration of the existing liver reserve.

2) In this study where the rates of liver regeneration of each group are compared with each other for the 3rd and 7th days, even though there is numerical difference among RO values of each control and sham groups for the 3rd and 7th days, this difference is not statistically significant (p>0.05). Numerical values for the 3rd and 7th days of each vitamin E, NAC, propofol groups are different, this difference is not statistically significant (p>0.05). However, the 3rd and 7th day values of the silybum group are numerically different and this difference is statistically significant (p<0.05). At the same time the highest RO value has been observed in the silybum group again. If we compare the 3rd and 7th day values with each other, there is numerical difference between the 3rd and 7th day RO values of each control and sham groups, but this difference is not statistically significant (p>0.05). While the 3rd and 7th day value of each vitamin E, NAC, propofol groups are numerically different, this difference is statistically significant (p<0.05). However, the 3rd and 7th day values of the silybum group are numerically different and this difference is statistically significant, also the highest RO value has been observed in this group (p<0.05).

3) In this study where rates of the Ki-67 staining pattern as histopathological indicators of liver regeneration are compared with that of marking pattern, if we look at the third and seventh day values among groups, no statistically significant difference between control and sham groups for both days has been observed (p>0.05). When the third day and seventh day values of vitamin

E, N-acetylcysteine, propofol groups from these groups, other working groups are evaluated separately, the difference is statistically significant ($p<0.05$). Both the 3rd and 7th day values of the silybum group are different from all the other groups, this difference is statistically significant ($p<0.05$). If we compare the third and seventh day values with each other, although Ki-67 index values of the control and sham groups on the 3rd and 7th days are numerically different, they are not statistically significant ($p>0.05$). The 3rd and the 7th day values of each NAC, propofol, vitamin E groups are numerically different. This difference is statistically significant. The difference between the 3rd and the 7th day values of the silybum group is 87.25 %, while this difference is statistically significant. The highest index value among the groups has been found in the silybum group again.

In line with these results, silybum marinaum, N-acetylcysteine, propofol and vitamin E seem to increase the regeneration of the liver after partial hepatectomy. In our study, especially silybum marinaum among the agents we use, compared to other agents we use have been determined to be more effective in liver regeneration (Figs 13–16).

Discussion

The main purpose of liver regeneration is to repair and regenerate tissues that have suffered a loss after resection as soon as possible and to restore the hepatic functions (15, 16). In experimental studies it is pointed out that partial hepatectomy is an ideal model for liver regeneration because tissue damage in methods using hepatic toxins or viruses might not always start enough inflammation (17).

The first scientific studies on liver regeneration were conducted by Higgins and Anderson in 1900s. Higgins and Anderson removed the middle and left lobes of liver in rats (70–80 %) and explained the model used in partial hepatectomy studies (18). Easy implementation and undamaged remaining liver tissue are among significant advantages of this model (19). Further studies were done by taking the example of this model.

In studies on effects of olive oil, fish oil and vitamin E on cellular proliferation after partial hepatectomy conducted by Kırımlioglu and his colleagues, vitamin E and fish oil were reported to be more effective on regeneration. This is in compliance with our study in terms of positive effects of vitamin E on regeneration (20). In a study on damaged rat liver with acetaminophen conducted by Kostopanagiotou and his colleagues, the triggering effect of propofol on the rate of regeneration was found statistically insignificant (21). In our experimental study, after hepatectomy, propofol's positive contribution to the regeneration has been confirmed. This outcome is in contrast with the Kostopanagiotou's study. In the study of Uzun and his colleagues after partial hepatectomy in nonalcoholic fatty liver, the effects of N-acetylcysteine on regeneration were investigated and a positive effect on regeneration was reported (22). In studies conducted by Das and his colleagues, the comparative effects of silybum and ascorbic acid on regeneration in the liver damaged by ethanol and ascorbic acid were investigated, while the regeneration in rats given ascorbic acid was higher than that of the group given silybum, and this difference was reported as statistically significant (23). Since our views differ from the above-mentioned studies, the difference between the results of silybum on regeneration in liver damaged by ethanol in the study of Das and the results of propofol on regeneration in rat liver damaged by acetaminophen considered to be statistically insignificant in the study of Kostopanagiotou and his colleagues would be referred to the degree of toxicity applied to the liver.

In the study of Yildiz and his colleagues, in rats subjected to partial hepatectomy, no statistically significant difference was confirmed in terms of AST and ALT values on days 0 and 7 in a relevant study of different forms of enteral and parenteral nutrition on liver function and regeneration in all subject groups, ($p>0.05$). When regeneration rates were evaluated on the 7th day of the study, they reported that statistically significant difference was confirmed between the subjects fed by either enteral or parenteral nutrition ($p<0.05$) (24).The results of study conducted by Yildiz and his colleagues about the impact of different forms of enteral and parenteral nutrition on AST, ALT, and regeneration in rats subjected to partial hepatectomy support the results of AST and ALT values in our study. We refer this significant outcome to the proximity of their finishing time of the studies. After 70 % partial hepatectomy applied to rats, the impact of pentagastrin on liver regeneration was investigated by Berberoglu et al. At the end of the study, regeneration rates of the group given pentagastrin and control group had no difference on postoperative day 2, but had a statistically significant difference on the 4th day and they suggested that the regeneration increased in proportion to the process ($p<0.05$) (25).

Ozalpan and Altun have found that as a result of partial hepatectomy by 35 % in rats, regeneration was rapidly increasing until the third day, reached 40 %, while after the third day it slowly continued (26). The increase in regeneration rates after partial hepatectomy on the 3rd day in our study is supported by the result of regeneration on the 4th day at a 70 % rate after partial hepatectomy in the study of Berberoglu and by the result of regeneration on the 3rd day at a 35 % rate after partial hepatectomy in the study of Altun and Ozalpan. Only one among these studies that lasted to the 7th day is the Sasanuma's study. But he reviewed only the rates of regeneration on the 7th day; he could not evaluate the clear difference of regeneration rates between the 3rd and 7th days. Based on information we have obtained, we conclude that regeneration after partial hepatectomy increases on the 3rd day and continues to increase by 75 % in the same way as described in literature.

When we compared all our findings after partial hepatectomy, the vitamin E, propofol, N-acetylcysteine and silybum marinaum were confirmed to increase liver regeneration, while silybum marinaum was found to have the greatest triggering effect on regeneration. It is concluded that vitamin E and propofol and N-acetylcysteine groups are not as effective as silybum in liver regeneration. After serious operations with high mortality and morbidity such as liver resection, the use of silybum marinaum may be preferred to ensure the integrity of tissue in regeneration. However, extensive clinical and experimental studies are needed for this. Our study shows that this plant is worth to be researched and developed from experimental aspect.

References

1. Trevor AJ, Miller RD. General Anesthetics. Basic and Clinical Pharmacology. Ed. Katzung Bg. 7 th Ed. Connecticut, Appleton & Lange, 1998: 409–423.

2. Rang HP, Dale MM, Ritter JM, Gardner P. General Anesthetic Agents. Pharmacology. 3 th Ed. Churchil Livingstone, New York. Inc. 1995: 532–547.

3. Ventresca GP, Cicchetti V, Ferrari V. Thiols, Medical Department, Zambon group, Italy, 1989, 77–102.

4. Zafarullah M, Li WQ, Sylvester J, Ahmad M. Molecular mechanisms of N acetyl cysteine actions. Cell Mol Life Sci 2003; 60: 6–20.

5. Kayaalp O. Tibbi farmakoloji 2002; 91: 1484–1489.

6. Zeybek N, Zeybek U. Farmasötik Botanik/ Ege Üniversitesi Eczacilik Fakültesi, Izmir, 1994.

7. Higgins G, Anderson R. Experimental Pathology of The Liver. Exp Surg Pathol 1931; 186–202 .

8. Hashimoto M, Kothary BC, Raper S. The effects of transforming growth factor alpha and somatostatin on regenerating hepatocytes in the rat. Regulatory peptides 1993; 44: 49–59.

9. Fan Y, Praet M, Huysse JV, Lelie B, Hemptinne B. Effects of portal vein arterialization on liver regeneration after partial hepatectomy in the rat. Liver Transplantation 2002; 8 (2): 146–152.

10. Maruyama H, Harada A, Kurokawa T, Kobayashi H, Nonami T, Nakao A. Duration of liver ischemia and hepatic regeneration after hepatectomy in rats. J Surg Res 1995; 58: 290–294.

11. Hopf NJ, Brem J, Bohl J, Perneczky A. Image analysis of proliferating cells in tumors of the human nervous system:an immunohistological study with the monoclonal antibody Ki-67. Neurosurgery 1994; 35 (5): 917–923.

12. Wintzer HO, Zipfel I, Schulte-Mönting J, Hellerich U, von Kleist S. Ki-67 immunostaining in human breast tumors and its relationship to prognosis. Cancer 1991; 67 (2): 421–428.

13. Navarro-Gonzalves JA, Garcia-Benayas C, Arenas J. Semiautomated measurement of nitrate in biological fluids. Clin Chem 1998; 44: 679–681.

14. Gwatsuki S, Shaw BW Jr, Starzi TE. Experience with 150 liver resections. Ann Surg 1983; 197: 247–252.

15. Wheatley J, Rosenfield NS, Berger L et al. Liver regeneration in children after major hepatectomy for malignancy**.** J Surg Res 1996; 61: 183–189.

16. Watanabe M, Yamaguchi K, Chijiiwa K, Tanaka M. FR167653 Improves survival and pulmonary injury after partial hepatectomy under ischemia/reperfusion in rats. J Surg Res 2001; 101, 146–151.

17. Michalopoulos GK, DeFrances MC. Liver regeneration. Science 1997; 276: 60–66.

18. Hamanoue M, Kawaida K, Takao S et al. Rapid and marked induction of hepatocyte growth factor during liver regeneration after ischemic or chrush injury. Hepatology 1992; 16: 1485–1492.

19. Kirimlioglu V, Kirimlioglu H, Yilmaz S, Ozgor D. Effect of fish oil, olive oil, and vitamin E on liver pathology, cell proliferation, and antioxidant defense system in rats subjected to partial hepatectomy. Transplant Proc 2006; 38 (2): 564–567.

20. Kostopanagiotou GG, Grypioti AD, Matsota P, Mykoniatis MG et al. Acetaminophen-induced liver injury and oxidative stress: protective effect of propofol. Eur J Anaesthesiol 2009; 26 (7): 548–553.

21. Uzun MA, Koksal N, Kadioglu H, Gunerhan Y. Effects of N-acetylcysteine on regeneration following partial hepatectomy in rats with nonalcoholic fatty liver disease. Surg Today 2009; 39 (7): 592–597.

22. Das SK, Vasudevan DM. Protective effects of silymarin, a milk thistle (Silybium marianum) derivative on ethanol-induced oxidative stress in liver. Indian J Biochem Biophys 2006; 43 (5): 306–311.

23. Yildiz et al. Parsiyel Hepatektomi Yapilan Ratlarda Enteral Ve Parenteral Beslenmenin Farkli Formlarinin Karaciger Fonksiyonlari Ve Rejenerasyonu Üzerine Etkisi.Ulusal Cerrahi Dergisi 2001;17 (2): 75–81.

24. Ugur Berberoglu. Kaptan gülben, hakan mersin, cigdem irkkan deneysel parsiyel hepatektomi modelinde pentagastrinin karaciger rejenerasyonuna etkisi. Ulusal cerrahi dergisi 2002; 18 (3): 134–139.

25. Altun S, Özalpan A. Hepatektomi Orani Ile Rejenerasyon Arasindaki Iliski. Itf Dergisi 1998; 61 (4): 485–491.

Received July 26, 2011.
Accepted January 9, 2012.