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Therapeutic Effects of the *Phenomenon* CCl4-Induced Experimental Liver Cirrhosis in Laboratory Rats

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(Presented by Academy Member Ramaz Khetsuriani)

Liver cirrhosis is a chronic, progressive liver disease and a global health concern, with high prevalence across the world. The study is aimed at investigating the therapeutic effects of the agents "phenomenon" and S-ademetionine in white laboratory rats with CCl4-induced liver cirrhosis. Experiments were carried out on the white laboratory rats with the weight 200-250g. Rats were injected intraperitoneally with 0.1 ml of CCl4, twice a week, for 2 months to induce liver fibrosis. S-ademetionine (5 mg/kg) was injected intraperitoneally once a day. The "phenomenon" (12 mg/kg) was administered orally once a day and phenomenon dissolved in 1.5 ml of honey – orally, once-daily for 20 days, respectively. Treatment efficacy was evaluated according to biochemical parameters: Aspartate aminotransferase (AST), Alanine aminotransferase (ALT), Sodium phosphatase (ALP), total bilirubin (TBIL), cholesterol, serum creatinine (CREA), Blood glucose, superoxide dismutase (SOD). The investigation results in the control group revealed notable elevations in AST, ALT, ALP, TBIL, cholesterol, and triglycerides, while there were concurrent reductions in creatinine and SOD levels in comparison to the data observed in healthy rats. Massive hepatic necrosis and accompanying inflammatory processes have led to impaired liver functions. The liver detoxification functions were impaired, resulting in disrupted metabolism of cholesterol, bilirubin, and triglycerides. Additionally, there was a decrease in protein synthesis, inhibition of glycogen synthesis, and an increase in blood glucose concentration. The better therapeutic effects were observed in animals treated with Phenomenon, especially at combination of Phenomenon+honey compared to the animals treated with S-ademetionine. Consequently, „phenomenon" demonstrates antioxidant, membrane-stabilizing and hepatoprotective properties, reduces lipid peroxidation, inhibits hepatocyte damage, and enhances liver functions in white laboratory rats with CCl4-induced liver cirrhosis. *© 2023 Bull. Georg. Natl. Acad. Sci.*

antioxidant, hepatoprotective, CCl4-induced liver cirrhosis, "phenomenon"

Liver cirrhosis is a chronic, progressive liver disease and a global health concern, with a high prevalence worldwide. The replacement of healthy liver tissue with fibrous tissue disrupts the normal functioning of the liver. As the disease advances, ability of the liver to perform detoxification,

metabolism, and protein synthesis is severely compromised. The complications associated with liver cirrhosis, such as portal hypertension, ascites and hepatic encephalopathy increased the risk of developing liver cancer, posing significant challenges in terms of treatment [1-4]. The primary goal of treatment is to slow down the progression of liver damage, manage complications, and improve the patient's quality of life. Despite significant advancements in medical research, the development of effective pharmacological therapies for liver cirrhosis has remained limited. The treatment outcome depends on the individual's response and the extent of liver damage. Currently, complete reversal of cirrhosis is not possible, and liver transplantation remains the only curative option for end-stage cirrhosis. While there are medications available to manage these complications, it is important to note that they may not ensure complete resolution. Although, several clinical trials are underway to explore potential treatments targeting fibrosis, inflammation, and other pathways involved in the progression of cirrhosis, consequently, more research is required to develop safe and effective therapies that have the potential to limit, or even reverse liver fibrosis. In the light of the above, the study is aimed to investigate the therapeutic efficacy of the pharmaceutical products – "phenomenon" and S-ademetionine in laboratory rats with CCl4-induced liver cirrhosis. Pharmaceutical product – "phenomenon" comprises plant-derived components, such as: Frogera, Birkava, Chamomile, Chinese limonur, Manchurian Aralias, Ginchara, Pumpkin pulp and also, Sulphur and Iron sulfate [5].

Methodical approach. The experiment was carried out on white male laboratory rats with a body weight 200-250 g. The animals were provided with optimal conditions of care and housing (free access to food and water) that adhered to internationally recognized standards. The research protocol was approved by the TSMU Ethics Committee for the Biomedical Research. After one-week adaptation period, the animals were randomly divided into 5 study groups: Group I – healthy rats; Group II – control (rats with CCl4-induced liver cirrhosis, untreated); Group III – rats with CCl4-induced liver cirrhosis, treated with S-ademetionine; Group IV – rats with CCl4-induced liver cirrhosis treated with the drug – Phenomenon; Group V – rats with CCl4-induced liver cirrhosis, treated with "phenomenon" + honey. Each of study groups consisted of 10 animals. For the purpose of modeling the disease, 0.1 ml of CCl4 was intraperitoneally injected into the study animals, 2 times a week for 2 months.

The commencement of treatment coincided with the morphological confirmation of liver cirrhosis in animals. Group III rats were intraperitoneally injected with S-ademetionine (5 mg/kg), once a day. In the group IV, "phenomenon" (12 mg/kg) was administered orally once a day, and in the group V, "phenomenon" dissolved in 1.5 ml of honey was administered orally once a day. All study group animals were treated for 20 days.

Aspartate aminotransferase (AST), alanine aminotransferase (ALT), sodium phosphatase (ALP), total bilirubin (TBIL), direct bilirubin (DBIL), cholesterol, triglycerides, creatinine (CREA), blood glucose have been measured by spectrophotocolorimetric method (closed system) with fully automated Roche cobas e111 analyzer, and antioxidant superoxide dismutase (SOD) was measured by ELISA method, respectively.

Results

ALT, AST, ALP, TBIL, cholesterol and triglycerides are highly sensitive diagnostic markers of liver function and their elevated levels serve as reliable indicators of liver damage. Determining serum creatinine levels provides valuable information about both protein synthesis in the liver and the kidney function status. Superoxide dismutase is an enzyme that acts as a natural antioxidant and its decreased level indicates lipid peroxidation [6-9]. The results of biochemical

investigations revealed that in group II animals (control, CCl4-induced liver cirrhosis), there were significant increases in AST, ALT, ALP, TBIL, cholesterol and triglyceride levels while, serum creatinine and superoxide dismutase were decreased in comparison to the data observed in healthy rats.

Based on the abovementioned, it is evident that hepatocytes at CCl4-induced liver cirrhosis were damaged by oxidative stress. Massive necrosis of hepatocytes and accompanied inflammatory processes alters/decreases the major functions of the liver. In particular, the liver detoxification functions were impaired, resulting in disrupted metabolism of cholesterol, bilirubin, and triglycerides. Additionally, there was a decrease in protein synthesis, inhibition of glycogen synthesis, and an increase in blood glucose concentrations.

As a result of the treatment, there was notable improvement in the studied parameters. In particular, the functional parameters of the liver (ALT, AST, ALP, TBIL), as well as serum creatinine and superoxide dismutase indicators were improved. Statistical analysis of the data obtained after the treatment showed that better therapeutic effects were obtained after treatment with "phenomenon", especially at combination of "phenomenon"+honey compared to the animals treated with S-ademethionine. "Phenomenon" decreases lipid peroxidation, inhibits the damage of hepatocytes and improves liver functions. Although the treatment with "phenomenon"+honey showed the best therapeutic effect, one important fact must be mentioned, namely, when treated with this regimen higher levels of blood glucose were observed, compared to the control group animals. Taking into account group V animals, taking honey-containing preparations for 20 days, the high glucose level was natural. With high probability, it can be assumed that in case of clinical treatment, the human glucose level remained within the normal range. However, when treating patients with diabetes mellitus, it is possible that the treatment may have some impact on blood sugar levels. To address this issue, it is essential to establish the optimal dilution ratio of "phenomenon" with honey.

It could be concluded that "phenomenon" demonstrated antioxidant, membrane-stabilizing, and hepatoprotective properties. The "phenomenon"+honey showed the best therapeutic effect improving liver functional parameters (ALT, AST, ALP) and normalizing creatinine, TBIL and SOD concentrations. The pharmaceutical product – "phenomenon" could be suggested as hepatoprotective and antioxidant agent for liver pathologies such as hepatitis, fatty dystrophy, and cirrhosis.

ადამიანისა და ცხოველთა ფიზიოლოგია

"ფენომენონის" სამკურნალო ეფექტები CCl4-ით გამოწვეული ექსპერიმენტული ღვიძლის ციროზის დროს ლაბორატორიულ ვირთაგვებში

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ღვიძლის ციროზი არის ქრონიკული, პროგრესირებადი დაავადება და გლობალური ჯანმრთელობის პრობლემა, რომელიც ფართოდაა გავრცელებული მთელ მსოფლიოში. კვლევის მიზანი იყო პრეპარატების – "ფენომენის" და S-ადემეთიონინის თერაპიული ეფექტების შესწავლა ლაბორატორიულ ვირთაგვებში CCl4-ით გამოწვეული ღვიძლის ციროზის დროს. ფარმაცევტული პროდუქტის-ფენომენონის შემადგენლობაში შედის: ბაყაყურა, ბირკავა, გვირილა, ჩინური ლიმონურა, მანჯურიული არალია, ჯინჭარა, გოგრის რბილობი, გოგირდი, რკინის სულფატი. სხვადასხვა საცდელი ჯგუფის ცხოველები იღებდნენ S-ადემეთიონინს (5 მგ/კგ/დღე) ინტრაპერიტონეალურად, ფენომენონს (12 მგ/კგ/დღე) – პერორულად, ხოლო ფენომენონს 1,5 მლ თაფლთან ერთად პერორულად დღეში ერთხელ 20 დღის განმავლობაში. მკურნალობის ეფექტიანობა შეფასდა ბიოქიმიური პარამეტრების: ასპარტატამინოტრანსფერაზა (AST), ალანინამინოტრანსფერაზა (ALT), ნატრიუმისფოსფატაზა (ALP), მთლიანი ბილირუბინი (TBIL), ქოლესტერინი, შრატის კრეატინინი (CREA), სისხლში გლუკოზა, სუპეროქსიდისდისმუტაზა (SOD) და ღვიძლის ქსოვილის მორფოლოგიის მიხედვით. დადგინდა, რომ საკონტროლო ჯგუფში AST, ALT, ALP, TBIL, ქოლესტერინი და ტრიგლიცერიდები მნიშვნელოვნად გაიზარდა, ხოლო კრეატინინი და SOD შემცირდა ჯანმრთელი ვირთაგვების მონაცემებთან შედარებით. ჰეპატოციტების მასიურმა ნეკროზმა და თანმხლებმა ანთებითმა პროცესებმა დააქვეითა ღვიძლის დეტოქსიკაციური ფუნქციები, ქოლესტერინის, ბილირუბინის და ტრიგლიცერიდების მეტაბოლიზმი, შემცირდა ცილების სინთეზი, შეფერხდა გლიკოგენის სინთეზი და გაიზარდა სისხლში გლუკოზის დონე. უკეთესი თერაპიული ეფექტები გამოვლინდა ფენომენონით მკურნალობისას, განსაკუთრებით, ფენომენონი+თაფლის კომბინაციის შემთხვევაში, ვიდრე S-ადემეთიონინით მკურნალობისას. ამრიგად, ფენომენონი ავლენს ანტიოქსიდანტურ, მემბრანომასტაბილიზებელ და ჰეპატოპროტექტორულ თვისებებს. CCl4-ით გამოწვეული ღვიძლის ციროზის დროს ლაბორატორიულ ვირთაგვებში

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