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Србије и Црне Горе

04-06. октобар 2023. године

Будва, Црна Гора

ДРУШТВО ЗА ЗАШТИТУ ОД ЗРАЧЕЊА
СРБИЈЕ И ЦРНЕ ГОРЕ

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Овај Зборник је збирка радова саопштених на XXXII Симпозијуму Друштва за заштиту од зрачења Србије и Црне Горе који је одржан у Будви, Црна Гора, 04-06.10.2023. године. Радови су према обрађеној проблематици груписани у једанаест секција. Сви радови у Зборнику су рецензирани од стране Научног одбора, а за све приказане резултате и тврдње одговорни су сами аутори.

Југословенско друштво за заштиту од зрачења основано је 1963. године у Порторожу, а од 2005. носи име "Друштво за заштиту од зрачења Србије и Црне Горе". На XXXII Симпозијуму, ове године обележавамо веома значајан јубилеј - ***60 година организоване заштите од зрачења на нашим просторима****.*

Од оснивања, Симпозијуми Друштва за заштиту од зрачења представљају прилику да се кроз стручни програм прикажу резултати истраживања у области заштите од зрачења, представе различите области примене извора и генератора зрачења, анализирају актуелна дешавања, размене искуства са колегама из региона, дефинишу проблеми и правци даљег унапређивања наше професионалне заједнице.

Поред тога, Симпозијуми друштва представљају и прилику да у мање формалном маниру сретнемо старе и упознамо нове пријатеље и колеге, обновимо старе и започнемо нове професионалне сарадње.

Ауторима и коауторима научних и стручних радова саопштеним на XXXII Симпозијуму се захваљујемо на уложеном труду и настојању да квалитетним радовима заједно допринесемо остваривању циљева и задатака Друштва и наставимо традицију дугу импозантних 60 година.

Посебно се захваљујемо свима који су подржали одржавање овог Симпозијума.

Свим члановима Друштва, сарадницима и колегама честитамо овај значајан јубилеј!

Организациони одбор XXXII Симпозијума ДЗЗСЦГ

THE EFFECT OF HONEY ON MALONDIALDEHYDE LEVEL IN PLASMA EXPOSED TO A THERAPEUTIC DOSE OF RADIATION

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ABSTRACT

Malondialdehyde (MDA) is the end product of lipid peroxidation and biomarker of free radicals. The aim of this study is to investigate the effect of honey on concentrations of MDA in plasma exposed to a therapeutic dose of radiation. In this experiment animals were divided into two groups: in CONTROL group the animals were given distilled water orally for 28 days, and HONEY group consisted of animals given 1,5 ml/kg honey orally for 28 days. Blood samples of both groups were collected from a rat's tail. All blood samples were exposed to radiation of 2 Gy. Malondialdehyde concentration in plasma was determined using *spectrophotometric method.* Honey treatment in the blood plasma exposed to a therapeutic dose of 2 Gy significantly decreased MDA level *by 30 %* compared with control group. The present results indicated that honey had an effect on the levels of MDA in plasma exposed to radiation.

Introduction

Radiotherapy is one of the main therapies in the treatment of cancer. During radiotherapy, ionizing radiations generate reactive oxygen species (ROS) in the irradiated tissue. Literature data have confirmed that ROS are responsible for structural and functional damage to membrane lipids [1]. Namely, Tabarraei and colleges [2] have confirmed that cell membrane permeability in the irradiated tissue is hampered. Free radical metabolites play an important role in the pathogenesis of radiation induced tissue injury [3,4]. Malondialdehyde (MDA) is the end product of lipid peroxidation and biomarker of free radicals. Numerous animal studies have shown that endogenous antioxidants reduce the cellular damage induced by ionizing radiation, and provide some degree of protection [5]. In addition, literature data imply that natural radioprotectors such as melatonin, curcumin, sheng-mai-san, dan-hong and metformin protect against ionizing-radiation toxicities to several organs [5]. Many studies have shown that honey plays a role in reducing cell death resulting from oxidative stresss. For example, Gharzouli and coworkers [6] have confirmed that honey is considered good natural dietary source of antioxidants. Additionally, earlier studies showed that honey significantly increased total antioxidant status [7,8]. However, very little is known about the ability of honey to inhibit the development of lipid peroxidation and neutralize ROS during radiotherapy. Because radiation induces oxidative damage, detecting the changes of lipid peroxidation level in irradiated plasma of rats treated with honey may by very important in the research of the antioxidant capacity of honey.

Determination of MDA levels is the usually applied assay for lipid peroxidation in biomedical sciences [9]. For this reason, in this study we examined the effect of linden honey on malondialdehyde (MDA) level in rat plasma exposed to a therapeutic dose of radiation of 2 Gy. It is a dose that can be given in one session, without damaging the healthy tissues beyond repair [10].

Materials and methods

Experiments were performed on 11-week-old Wistar male rats weighing (300-350) g. The animals were maintained under standard vivarium conditions in a temperature-controlled room (22±1.0) $^{\circ}$C and 12 h/12 h light/dark cycle, with water and food *ad libitum* and kept three to four per cage. The care was taken to minimize the pain and discomfort of the animals according to the recommendations of the Ethical Committee of the Vinča Institute of Nuclear Sciences (03/2023).

In this experiment linden honey was used, collected in 2022, in the farm by dr Vojislav Stanić (region Vojvodina-Fruška Gora) and stored at room temperature in the dark until the experiment.

The honey was mixed with distilled water in a ratio of 1:1. The doses of honey used in this study have been reported by Abdulmajeed and colleges [7] and Halawa and coworkers (1.5 ml/kg honey orally for 28 days) [8]. Ten animals were randomly divided into two groups: CONTROL group (n=5) and HONEY group (n=5). Due to the administration of honey, the rats were housed individually (one rat per cage) (Photo 1). While the HONEY group animals were given the solution of honey in distilled water (1.5 ml/kg), the CONTROL group animals were given distilled water in the same period (1 hour a day, 5 days a week). The experiment lasted 28 days [7, 8].

Photo 1. Animals take honey orally

Photo 2. Blood samples collection.

After the described treatment, blood samples were collected from both groups. Blood was sampled from a rat's tail (Photo 2). After blood collection, the animals remained alive.

All blood samples (about 1 ml blood in a test tube) placed on the radiation bench at a position where the absorbed dose was 6 Gy/h, in Department of Radiation and Environmental Protection, Institute of Nuclear Sciences "Vinča" (Photo 3). The exposure to gamma radiation lasted for 20 minutes, so the samples received an absorbed dose of 2 Gy. It is a dose that can be given in one session, without damaging the healthy tissues beyond repair [10].

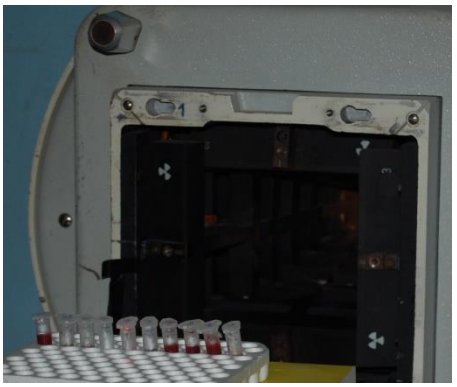

Photo 3. Blood irradiation.

The blood samples were centrifuged at 2400g for 10 minutes to obtain plasma. Aliquots were then prepared and stored at -70^0 C until the time of biochemical analysis.

Determination of MDA level was performed using spectrophotometric method previously described by Siddique and coworkers [11]. All samples were read on spectrophotometer at 586 nm. Malondialdehyde concentration was expressed as μM of plasma.

The data are presented as means ± S.E.M. The differences of plasma concentration of MDA between CONTROL group and HONEY group were analyzed by t-test. Statistical analysis was carried out using the SigmaPlot 10.0 with SigmaStat integration. The statistical significance was accepted at $p < 0.05$.

Results

Honey treatment in the blood plasma exposed to a therapeutic dose of 2 Gy significantly decreased MDA level by 30 % ($p < 0.05$, t-test) compared with control group.

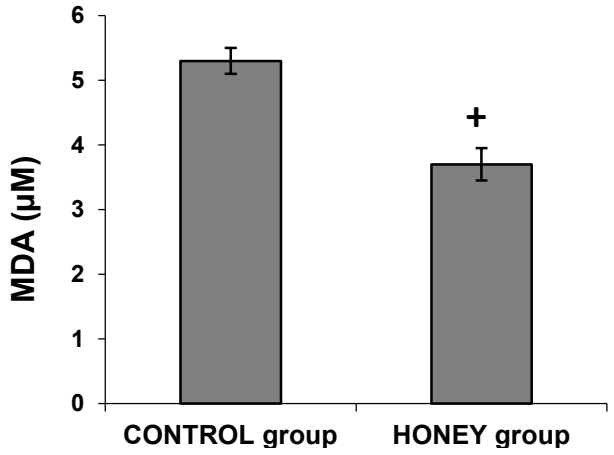

Figure 1. Effects of honey on malondialdehyde (MDA) level in plasma. The values are means ± S.E.M. of 10 rats. Statistical significance: +p < 0.05 HONEY group *vs*. CONTROL group (t-test). Concentration of MDA was expressed as μM of plasma.

Discussion

The results in this study suggest a lower level of lipid peroxidation and free radical generation after the ingestion of honey, which points out the beneficial effects of honey in preventing oxidative damage. We assume that honey increases antioxidant activity and in that way can protect tissue against oxidative stress. Our result is in accordance with the reports of Steinberg and Witztum [12] who found that antioxidant supplementation gave therapeutic effects in subjects with high oxidative stress. Oxidative stress is the condition in which the cellular antioxidant defence system is insufficient to keep the levels of ROS low in the body. It is known that antioxidants are molecules that can safely interact with ROS and terminate the chain reaction before vital molecules are damaged. The first line of defense includes antioxidant enzymes such as copper, zinc superoxide dismutase, manganes superoxide dismutase and catalase which directly remove ROS. In addition, glutathione peroxidase is one of the most important enzymes for removing peroxides from cells. Shabeeb and coworkers [1] have showed that irradiation of the skin tissues led to a significant reduction in superoxide dismutase and catalase activities as well as increased MDA levels. The ability of honey to prevent oxidative damage might be due to its phenolic and non phenolic antioxidant content or indirectly through the action of antioxidant enzymes activity in reducing hydrogen peroxide [13]. Lodovici and colleges [14] have confirmed that polyphenols have the capacity to decrease lipid peroxidation, and prevent DNA oxidative damage. Also, protective effect of honey may be attributed to the biologically active compounds flavonoids that work to scavenge ROS [15]. Kečkeš et al. [16] identified a total of 43 polyphenols in Serbian unifloral honey. Numerous flavonoids (such as apigenin, pinocembrin, pinobanksin, kaempferol, quercetin, galangin, chrysin, and luteolin) and phenolic acids (caffeic, gallic, cinnamic, protocatechuic, p-coumaric, and chlorogenic acids) were identified in samples of Serbian unifloral honey [17]. Literature data imply that honey also can contain vitamins A, C, E as antioxidants that can capture ROS [18]. Additionally, the antioxidant trace elements iron, zinc and selenium which are essential cofactors for the enzymatic antioxidant defense system represented by catalase, superoxide dismutase and glutathione peroxidase [19]. Costa-Silva and coworkers [20] explained that honey induced increase in glutathione peroxidase activity might be due to the presence of selenium in honey. In this experiment we used linden honey because it is a source of more vitamins and minerals then other honey varieties, including more than 400 substances and compounds. These include calcium, magnesium, sulfur, copper, potassium,

zinc, iodine, aluminum, nickel, phosphorus, manganese and cobalt, vitamins (B1, B2, B5, B6, C, biotin, tocopherol and niacin) and organic and inorganic acids (gluconic, citric, lactic malic, tartaric, linolenic, oxalic, succinic, hydrochloric and phosphoric). In addition, the results of Gašić and coworkers [17] have showed that samples of honey from Vojvodina and Zlatibor region were distinguished from the honey varieties from the rest of Serbia due to the presence of dicaffeoylquinic acid, ellagic acid, caffeic acid phenethyl ester, and chlorogenic acid. Halawa and colleges [8] have suggested that honey exert its protective role through its antioxidant mechanism and through restoring glutathione activity. Namely, honey is a good natural source of sulfhydryl (SH) group. Also, it is known that the most important endogenous antioxidant is glutathione, as well as that SH group of glutathione is important as a direct scavenger of ROS [21].

It can be concluded that honey had an effect on the levels of MDA in the blood plasma exposed to a therapeutic dose of radiation. Findings from the present study have shown that linden honey has the potential to protect against radiotherapy-induced oxidative stress. However, in order to better understand this mechanism, future studies should investigate the activity of antioxidant enzymes, as well as the content of phenolic and non phenolic antioxidants in linden honey.

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DELOVANJE MEDA NA NIVO MALONDIALDEHIDA U PLAZMI IZLOŽENOJ TERAPIJSKOJ DOZI ZRAČENJA

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SAŽETAK

Malondialdehid (MDA) je krajnji proizvod peroksidacije lipida i biomarker slobodnih radikala. Cilj ovog istraživanja je bio da se ispita uticaj meda na koncentraciju MDA u krvnoj plazmi izloženoj terapijskoj dozi zračenja. U ovom eksperimentu životinje su podeljene u dve grupe: u kontrolnoj grupi životinje su dobijale destilovanu vodu oralno tokom 28 dana, a drugu grupu činile su životinje koje su tokom 28 dana dobijale oralno 1.5 ml/kg meda. Uzorci krvi obe grupe su sakupljeni iz repa pacova. Svi uzorci krvi bili su izloženi zračenju od 2 Gy. Koncentracija MDA u plazmi određena je spektrofotometrijskom metodom. Tretman medom u krvnoj plazmi izložen terapijskoj dozi od 2 Gy značajno je smanjio nivo MDA (30 %) u poređenju sa kontrolnom grupom.

САДРЖАЈ