UDC 617.7-07.681-092

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OXIDATIVE STRESS CORRECTION WITH METABOLIC PREPARATIONS, NEW POSSIBILITIES OF ANTIOXIDANT PROTECTION IN PRIMARY GLAUCOMA

There were studied antioxidant action of water-soluble form of quercetin – corvitin and melatonin in rabbits with glaucoma model. It was shown intravenous injections of corvitin essentially decreased oxidative stress in animals with advanced stage of experimental glaucoma. Prophylactic-protective apply of melatonin on the basis of model induction in significant degree protected oxidative stress development.

Key words: quercetin; melatonin; glaucoma; oxidative stress

INTRODUCTION

Today it is known that mechanisms of free radical damage and oxidative stress play an important role in the pathogenesis of primary glaucoma, contributing primarily formation of glaucomatous optic neuropathy [15, 22]. The formation of active atoms or molecules with unpaired electron in an outer orbit, so-called free radicals, in the body takes place continuously. However, under physiological conditions, the amount and rate of their formation is compensated by the speed of the elimination with antioxidants. Imbalance in the amount and activity of pro-oxidants and antioxidants leads to oxidative stress. This mechanism, in turn, can lead to damage of cell macromolecules, membrane degradation and change functions that contribute to making cell death.

According to some authors [17, 12] the formation of oxidative stress caused by unstable ocular blood flow is one of the main risk factors for glaucomatous optic neuropathy. As it states in repeated mild reperfusion, as a consequence of unstable blood flow, a large amount of free radicals is formed. Reperfusion injury occurs when the blood supply returns to the tissue after a period of ischemia. This mechanism is probably the most traumatic for the eye tissues, especially the nervous structures of the eye. Oxidative stress, which occurs in the conditions of reperfusion, the most severely damages the mitochondria. These organelles are present in large numbers in the optic nerve, as a structure with high energy consumption. The damage of the mitochondria disrupts the production of energy, leading to malnutrition and neural cell degeneration [11].

Besides effects on the nervous tissue, glaucoma oxidative stress may cause degeneration of the trabecular meshwork and be thus trigger mechanism for raising intraocular pressure (IOP), changing the paths of outflow of intraocular fluid [21, 10]. The idea of a pathogenic role of oxidative stress in raising IOP is supported by a number of researchers. Thus, it was shown that patients with primary open angle glaucoma (POAG) had have a decrease in the total antioxidant activity of intraocular fluid, reduced glutathione levels [10, 13]. The use of antioxidants in an attempt to treat glaucoma was started by S.C. Evans. He used of large doses of vitamins A and C [9, 16]. It was assumed that vitamin A supports dehydrated status of collagen in the eye drainage system, and hydration of collagen structures in the outflow tract leading to increased IOP. High doses of vitamin C reduce IOP by osmotic effect, although this method is hardly practical use for the control of glaucoma in the present conditions.

In this regard, it is important that the treatment strategy of glaucoma gradually changes. It is receding into the past approach to doctoring it as a disease, not related to other organs and the organism as a whole. In search of strategies to treat this pathology as the suffering of "a sick organ in a sick body" the use antioxidants with systemic effects is reasonable. The use of the natural origin medication with minimal side effects, in our view, is the most appropriate. In this direction, carried out a study on the effectiveness of glaucoma treatment with the antioxidant effect drugs such as lipoic acid [6], pyridine derivative (an analogue of vitamin B6) emoksipin [1], carotenoids of ginkgo biloba extract and lutein [4, 20].

In the study of the possibility of increasing the effectiveness of antioxidant effects in primary glaucoma our focus attracted such biologically active substances with a very powerful antioxidant potential as quercetin and melatonin. The antioxidant properties of bioflavonoids quercetin are determined by its chemical structure, which

allows it to counteract the damaging effects of reactive oxygen species and free radicals, i. e., are natural scavengers. In addition to the ability to be "free radical scavengers", quercetin prevents lipid peroxidation and chelating metal ions [14]. It's proved for Quercetin neuroprotective possibility in neurodegenerative diseases (Alzheimer's, Parkinson's, epilepsy, stroke) and cardioprotective effects in cardiovascular diseases. The mechanism of this protection is linked with the antioxidant activity of quercetin [3, 7, 23].

Melatonin is a synthesis product of a pineal gland (epiphysis) and other organs, including the retina, has been known mainly as a regulator of circadian rhythms in the body. Recently it became known antioxidant properties of melatonin. It has been identified as a direct scavenger of free radicals [19] and indirect antioxidant – regulator of antioxidant enzymes activity [18]. Melatonin is highly active endogenous antioxidant and its lipophilic properties allows it to diffuse into the various cell compartments and provide a protective effect against cytotoxic free radicals both in the cytoplasm and in the nucleus. Clinical trials have shown that the antioxidant properties of melatonin may be effective in the treatment of acute medical conditions (sepsis, asphyxia) or chronic diseases (metabolic and neurodegenerative diseases, cancers, inflammation) [8].

The purpose of our study was to investigate the antioxidant activity of melatonin and quercetin preparations on glaucoma model in rabbits.

MATERIALS AND METHODS

As a quercetin-containing preparation we used domestic product Corvitin for injection, which is a freeze-dried substance of quercetin with polyvinylpyrrolidone as a modifier of solubility. We also used domestic product Vita-melatonin. It is a synthetic analog of neuropeptide melatonin produced by the pineal gland.

The investigations were carried out on rabbits, which glaucoma was caused by systemic administration of adrenaline according to the developed scheme [5]. The study includes the following groups: control group – healthy animals; advanced glaucoma model (GM), a glaucoma model, treated with Corvitin (GMC); a reproducible of glaucoma model with introduction of melatonin (GMM).

In GMC group rabbits with developed experimental glaucoma obtained Corvitin as 10 daily intravenous infusions (2 % solution of the preparation in isotonic saline) at a dose of 10 mg/0.5 ml.

In GMM group rabbits obtained Melatonin in the form of an aqueous suspension per os at a dose of 0.3 mg for 4 months: the entire period of the modeling, as well as before and 2 weeks after the induction of the model.

Biochemical studies were conducted in animal blood plasma. Malonic dialdehyde (MDA) was determined by reaction with thiobarbituric acid, the total antioxidant activity of blood plasma (TAOA) – using a model system with linolenic acid. The statistical data processing was carried out using t-test.

Table

OXIDATIVE STRESS LEVELS IN RABBITS WITH GLAUCOMA MODEL UNDER ADMINISTRATION OF QUERCETIN- AND MELATONIN-CONTAINING PREPARATIONS

	Groups	Statistic Indices	Metabolic Indices	
			MDA (нМ/ml)	TAOA (%)
I	Control (n = 20)	M ± m	0.58 ± 0.02	52.3 ± 0.72
II	GM (glaucoma) (n = 23)	M ± m p_1 <	1.18 ± 0.03 0.001	34.0 ± 0.87 0.001
III	GMC (glaucoma + corvitin) (n = 15)	M ± m p_1 < p_2 <	0.71 ± 0.02 0.01 0.001	43.9 ± 0.92 0.001 0.001
IV	GMM (glaucoma + melatonin) (n = 21)	M ± m p_1 p_3 <	0.66 ± 0.03 > 0.05 0.001	45.4 ± 1.03 <0.01 0.001

Note: p_1 – comparing with control; p_2 – comparing III c II; p_3 – comparing IV c II.

RESULTS AND DISCUSSION

Our study of pro- and antioxidant status of animals with the induced glaucoma have confirmed our previous results [2] and found a high level of oxidative stress in animals of group GM. It has been manifested by an increase in the content of secondary product of lipid peroxidation MDA by 103 % (compared to control), and a decrease in antioxidant status, reflected in a reduction of 40 % TAOA in blood plasma (Table).

Introduction of Corvitin intravenous injections to GMC group rabbits with the ocular and metabolic functions disorders has largely hindered severity of oxidative stress in them. In this group MDA levels compared to the control was increased by 22 % and TAOA reduced by 16.1 %. Comparing the severity of oxidative stress in the GMC and GM groups it has been revealed that under the influence of Corvitin high MDA levels have been decreased by 39.2 % and TAOA has been increased by 40.3 % in blood of rabbits with developed glaucoma.

Under long-term administration of melatonin in GMM group has been largely prevented the development of oxidative stress in the simulation of glaucoma. Thus, the content of MDA in a month after the 4-months administration of melatonin has been increased by 13.7 % and TAOA – reduced by 13.2 % compared with the control. When comparing these results with the untreated GM group, MDA levels has been lower by 44.1 % and TAOA – above by 33.5 % under the action of melatonin.

Thus, the study of the activity of quercetin-containing Corvitin and melatonin in the experiment showed that both of these compounds with known antioxidant properties proved to be effective in the inducing of glaucoma in rabbits by means of chronic stressing adrenaline. Both of these substances expressed direct antioxidant property as scavengers of free radicals, which are indirectly judged by the decrease the level of secondary lipid peroxidation product in the blood of experimental animals. Furthermore, as quercetin and melatonin in our experi-

ment acted also as indirect antioxidants, activating antioxidant system, which we concluded to increase TAOA of blood. Wherein melatonin has been used long in prophylactically – tread mode, namely, during induction of the adrenaline glaucoma model, before and after the cessation of administration of epinephrine. It is possible to largely stop the development of oxidative stress, preventing a substantial increase in free-radical processes and activating the antioxidant defense system.

CONCLUSIONS

1. Introduction of 10 intravenous injections of quercetin-containing Corvitin to rabbits with advanced stage of glaucoma model significantly reduced the severity of oxidative stress in them.
2. Preventive and treat administration of the stress protective hormone melatonin during 4 months in rabbits with reproducible adrenaline glaucoma model has largely prevented the formation of oxidative stress in them.
3. The experimental data, which showed significant antioxidant effects of Corvitin and melatonin in experimental glaucoma, can serve as a basis for recommending the use of these preparations in the clinic of primary glaucoma.

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УДК 617.7-07.681-092

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КОРЕКЦІЯ ОКСИДАТИВНОГО СТРЕСУ МЕТАБОЛІЧНИМИ ЗАСОБАМИ, НОВІ МОЖЛИВОСТІ АНТИОКСИДАНТНОГО ЗАХИСТУ ПРИ ПЕРВИННІЙ ГЛАУКОМІ

Вивчено на моделі глаукоми у кролів антиоксидантну дію водорозчинної форми кверцетину-корвітину та препарату мелатоніну. Показано, що курс внутрішньовенних введень корвітину суттєво знизив оксидативний стрес у тварин з розвитою стадією експериментальної глаукоми. Профілактично-протекторне введення мелатоніну на тлі індукції моделі значною мірою завадило розвитку оксидативного стресу.

Ключові слова: кверцетин; мелатонін; глаукома; оксидативний стрес

УДК 617.7-07.681-092

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КОРРЕКЦИЯ ОКСИДАТИВНОГО СТРЕССА МЕТАБОЛИЧЕСКИМИ СРЕДСТВАМИ, НОВЫЕ ВОЗМОЖНОСТИ АНТИОКСИДАНТНОЙ ЗАЩИТЫ ПРИ ПЕРВИЧНОЙ ГЛАУКОМЕ

На модели глаукомы у кроликов изучено антиоксидантное действие водорастворимой формы кверцетина – корвитина и препарата мелатонина. Показано, что курс внутривенных инъекций корвитина существенно снизил оксидативный стресс у животных с развитой стадией экспериментальной глаукомы. Профилактически-протекторное длительное введение мелатонина на фоне индукции модели в значительной степени предотвратило развитие оксидативного стресса.

Ключевые слова: кверцетин; мелатонин; глаукома; оксидативный стресс

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Надійшла до редакції 11.04.2016 р.