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EXTRACELLULAR MATRIX DEGRADATION PRODUCTS IN THE HEART OF RATS WITH METABOLIC SYNDROME UNDER CHROMIUM PICOLINATE ADMINISTRATION

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The populace of Ukraine shows a trend of increasing percentage of persons with obesity, complicated by metabolic syndrome (MetS), which causes damage to the heart extracellular matrix. According to recent studies chromium, picolinate (CrPIC) has the potential to attenuate lipid metabolism disorders and protect the extracellular matrix from degradation. The aim of this research was to estimate the blood lipid profile and the content of glycosaminoglycans, L-hydroxyproline and sialic acids in the heart of rats with simulated metabolic syndrome under Chromium picolinate administration. Mature male Wistar rats were divided into 4 groups of 6 animals each – control; metabolic syndrome induction; CrPIC administration; metabolic syndrome + CrPIC administration. Metabolic syndrome was reproduced by using a 20% fructose solution as the only source of water for 60 days. CrPIC was administered orally at a dose of 80 µg/kg daily for 60 days. The concentration of the heart extracellular matrix degradation proteins was determined spectrophotometrically in a 10% heart homogenate. CrPIC administration to healthy animals stimulated the accumulation of glycosaminoglycans chondroitin fraction in the rat heart. Metabolic syndrome modeling resulted in an increase in TG, TC and LDL-C blood levels, intensification of collagenolysis, degradation of glycoproteins and glycosaminoglycans with a predominance of the keratan-dermatan fraction. CrPIC administration to animals with metabolic syndrome reduced collagenolysis and glycoproteins degradation, changed the dominating fraction of glycosaminoglycans from keratan-dermatan to chondroitin in rat heart connective tissue indicating its potential to prevent cardiac tissue remodeling during metabolic syndrome.

Keywords: metabolic syndrome, chromium picolinate, heart extracellular matrix, glycosaminoglycans, L-hydroxyproline, sialic acids, lipid profile.

Modern populace of developed countries, such as USA, countries of European Union, Japan etc., are prone to suffer from obesity and obesity-induced diseases. According to Chong B. et al. obesity-related disability-adjusted life years rose 0.48% annually from 2000 to 2019, predicted to increase by 39.8% from 2020 to 2030, with highest rates in Eastern Mediterranean [1]. In 2021, an estimated 15.1 million children and young adolescents, 21.4 million older adolescents (aged 15-24 years), and 172 million adults had overweight or obesity in the USA [2]. The populace of Ukraine shows a trend of an increasing percentage of overweight and obese persons, especially in children [3].

Obesity can be complicated by metabolic syndrome (MetS) and /or diabetes type II (T2D) development [4]. Obesity by itself or its complications may lead to heart injury [5, 6]. Leading role of obesity-induced cardiac injury may be attributed to metaflammation and extracellular matrix (ECM) disorders. During metaflammation several matrix metalloproteinases (MMP) can be activated, which, in turn, will lead to organ remodeling [7]. This leads to development of cardiac fibrosis often observed during MetS and T2D [8]. Cardiac fibrosis induced by MetS development is caused by influence of advanced glycation end products (AGEs), which activate their respected receptors (RAGEs), which trough activation of extracellular-signal-regulated kinase (ERK) pathway induce transcriptional factor NF-κB [9]. NF-κB being a redox sensitive transcriptional factor causes oxidative stress development in cardiac tissues [10]. Oxidative stress activates ECM and myofibroblasts aggravating cardiac remodeling [11]. Besides structural proteins of heart ECM development of MetS can influence cell surface proteoglycans such as syndecan and glypican [12].

Therefore, the search for prophylaxis and treatment of obesity-induced changes in ECM of heart is an actual problem of modern medicine. Chromium picolinate (CrPIC) is a mineral chelate that consists of three picolinate molecules with Cr^{3+} at its center. CrPIC has a potent influence on lipid metabolism and on inflammation, so it can be considered a possible candidate for medicinal treatment and prophylaxis of high calorie intake-induced obesity and metabolic syndrome [13]. Influence of CrPIC supplementation on oxidative stress is a subject to a debate in scientific community, since some works suggest CrPIC having an antioxidant enhancing properties, while others show its inefficiency in altering redox balance in cell [14, 15]. Some authors show, that prolonged CrPIC supplementation to healthy animals may induce oxidative damage to heart tissues [16]. CrPIC supplementation in complex with myo-inositol, glycine, *Coprinus comatus*, α-lipoic acid, phlorizin, zinc, and vitamin B_6 to humans with MetS significantly ameliorated a number of metabolic parameters (body weight, body-mass index, total cholesterol and triglyceride content) without measurable side effects [17]. Scientific literature provides limited amount of data regarding influence of CrPIC on ECM metabolism in heart during metabolic syndrome.

The aim of this research is to establish influence of chromium picolinate administration on total glycosaminoglycans content, individual glycosaminoglycans fractions, concentration of L-hydroxyproline and sialic acids in rat heart under metabolic syndrome conditions.

Materials and Methods

The study was conducted on 24 mature male Wistar rats weighing 200-260 g. The animals were divided into 4 groups of 6 animals each. The first group was a control group, the animals of this group received manipulations similar to those of the other groups, but instead of the active substances, they received a 0.9% solution of sodium chloride. The second group was the experimental metabolic syndrome group (MetS group). Experimental MetS was reproduced by using a 20% fructose solution as the only source of water for 60 days [18]. The third group was Chromium picolinate (Sigma-Aldrich, CAS number 14639-25-9) administration group (CrPIC group). CrPIC was administered daily orally in a dose 80 μg/kg for 60 days [19]. The fourth group was the group of the combined effect of administration of CrPIC on the background of MetS modelling (MetS+CrPIC group). Animals from this group received CrPIC daily orally in a dose 80 μg/kg for 60 days and had 20% fructose solution as the only source of water for 60 days.

The animals were kept in the vivarium of the Poltava State Medical University under standard conditions. We worked with laboratory animals according to European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes (Strasbourg, 1986). In the evening (8 p.m.) before withdrawal of animals from the experiment they were deprived from food and 20% fructose solution (was replaced by water *ad libitum*). The withdrawal of animals from the experiment was carried out in the morning (8 a.m.) under thiopental anesthesia by taking blood from the right ventricle of the heart. All manipulations with laboratory animals were approved by the Bioethics Commission of the Poltava State Medical University (Record No. 206 from 24.06.2022).

In the blood we studied concentration of following metabolic substances: glucose (Assay HP009.02), triglycerides (TG, Assay HP022.03), total cholesterol (TC, Assay HP026.07), cholesterol from low-density lipoproteins (LDL-C, Assay HP026.05), cholesterol from high-density lipoproteins (HDL-C, Assay HP026.03). All abovementioned substances were evaluated by assays produced by Filicit-Diagnostics (Ukraine). We also calculated body-mass index (BMI) according to recommendations [20].

In order to evaluate development of insulin resistance we calculated following indexes:

Triglyceride glucose-body mass index (TyG-BMI). $\text{TyG-BMI} = \text{Ln}\,[\text{TG (mg/dl)} \times \text{Glucose (mg/dl)} \div 2] \times \text{BMI (kg/m}^2)$ [21].

Metabolic score for insulin resistance (METS-IR) index. $\text{METS-IR} = \text{Ln}\,[(2 \times \text{Glucose (mg/dl)}) + \text{TG (mg/dl)}] \times \text{BMI (kg/m}^2) \div \text{Ln}\,[\text{HDL-C (mg/dl)}]$ [21].

Before biochemical studies we prepared a 10% rat heart homogenate, which was made using 0.2 M Tris buffer solution (pH 7.4). For tissue homogenate exactly 1 g of heart ventricle tissue was used, it was cut with scissors into small pieces, then it was grinded in porcelain mortar with porcelain pestle. Afterwards 9 ml of 0.2 M Tris buffer solution (pH 7.4) were added. Obtained mixture of tissue and 0.2 M Tris buffer solution (pH 7.4) was centrifugated at 3000 g for 10 min. Upper layer of obtained centrifugate was moved to new centrifuge tube and then used for biochemical analysis of concentration of heart ECM degradation molecules.

The total concentration of glycosaminoglycans (GAG), the concentration of heparin-heparan, keratan-dermatan and chondroitin fractions of GAG, the concentration of free L-hydroxyproline and the concentration of sialic acids were determined spectrophotometrically in 10% rat heart homogenate [22].

Total concentration of GAG was determined by content of hexuronic acids formed after GAG hydrolysis by concentrated sulfuric acid. Content of hexuronic acids was determined spectrophotometrically by estimation of content of colored products formed in reaction of hexuronic acids with carbazole in 10 mm cuvette on wavelength 530 nm using Ulab 101 spectrophotometer (China) [22].

GAG concentration determination protocol:

1. We poured 0.5 ml of 10% heart tissue homogenate into a centrifuge tube.

2. Then we added 2 ml of working 16.1 M (94%) ethanol solution (contains in 100 ml of 96% ethanol solution 0.98 g of potassium acetate and 9.75 ml of acetic acid) cooled to 1-5°C to the sample for precipitation of glycosaminoglycans.

3. The sample was mixed and after 5-6 min centrifuged at 3000 g for 15 min.

4. The resulting precipitate was emulsified in 3 ml of 6% trichloroacetic acid. The resulting mixture was hydrolyzed in a boiling water bath for 30 min. Then the sample cooled to 10-22°C, it was centrifuged at 3000 g for 10 min.

5. Then we determined the content of hexuronic acids in the supernatant. To do this, we added 5 ml of a working solution of concentrated sulfuric acid (contains 0.95 g of sodium tetraborate in 100 ml of concentrated sulfuric acid) to 1 ml of the supernatant. The sample was heated for 10 min in a boiling water bath and cooled to 10-22°C. Then 0.1 ml of a working solution of carbazole (0.167 g of carbazole (corresponding to 0.01 M) dissolved in 100 ml of working 16.1 M (94%) ethanol solution) was added. Then the test tube was shaken and heated for 120-130 sec in a boiling water bath until a violet-pink color appeared.

6. Absorbance of a sample was measured on in 10 mm cuvette on wavelength 530 nm using Ulab 101 spectrophotometer against working solution of sulfuric acid.

Before estimation of different fractions of GAG we performed differential precipitation of corresponding GAG fraction according to N. Volpi [23].

GAG differential precipitation protocol:

1. We poured 0.5 ml of 10% heart tissue homogenate into a centrifuge tube.

2. 0.4 ml of propanol-1 (pure) was added to the sample to precipitate the heparin-heparan fraction of glycosaminoglycans.

3. The sample was mixed and after 5-6 min centrifuged at 3000 g for 15 min. The supernatant was poured into another test tube for further analysis. In precipitate we determined heparin-heparan GAG content according to GAG determination protocol starting at step 4.

4. 0.2 ml of propanol-1 (pure) was added to the supernatant to precipitate the keratan-dermatan fraction of glycosaminoglycans.

5. The sample was mixed and after 5-6 min centrifuged at 3000 g for 15 min. The supernatant was poured into another test tube for further analysis. In precipitate we determined keratan-dermatan GAG content according to GAG determination protocol starting at step 4.

6. 0.2 ml of propanol-1 (pure) was added to the supernatant to precipitate the chondroitin fraction of glycosaminoglycans.

7. The sample was mixed and after 5-6 min centrifuged at 3000 g for 15 min. The supernatant was poured into another test tube for further analysis. In precipitate we determined chondroitin GAG content according to GAG determination protocol starting at step 4.

L-hydroxyproline content was determined by reaction of pyrrole-2-carboxylic acid, which is formed during the oxidation of L-hydroxyproline, with paradimethylaminobenzaldehyde [22].

L-hydroxyproline content determination protocol:

1. 0.1 ml of 10% heart tissue homogenate was added to centrifugation tube.

2 0.5 ml of chilled to 5°C absolute (100%) alcohol was added.

3. Test sample was centrifugated the for 10 min at 3000 g.

4. Supernatant was transferred to new centrifugation tube.

5. 0.1 ml of oxidant solution (1 part of 7% chloramine with 4 parts of acetate-citrate (pH 6.0) buffer solution, which contains on 100 ml 5.7 g of sodium acetate trihydrate, 3.75 g of trisubstituted sodium citrate 5.5-hydrate, 0.85 g of citric acid monohydrate, and 38.5 ml of propanol-2) was added to centrifugation tube and was left to stand for 4.5 min.

6. 0.5 ml of propanol-2 was added and solution incubated for 4 min under room temperature.

7. 0.5 ml of Ehrlich reagent was added.

8. 1 ml of propanol-2 was added.

9. Sample was placed in a water bath and incubated for 30 min. at a temperature of 60°C.

10. Sample was cooled to room temperature.

11. Absorbance was measured on Ulab 101 at a wavelength of 540 nm in a 10 mm cuvette against the control sample (same procedure but contains distilled water instead of tissue homogenate).

Sialic acids, which are acylated derivatives of neuraminic acid, were determined after their hydrolysis by formation of a colored compound between heated sialic acids and acetic acid-sulfuric acid reagent (Hess reaction) [24].

Sialic acids determination protocol:

1. 0.2 ml of 10% heart tissue homogenate was added to 0.8 ml of distilled water and transferred to centrifuge tube. Then 1 ml of 10% trichloroacetic acid solution was added.

2. Centrifuge tube were placed in a boiling water bath and held for 5 min.

3. Then tube was placed in cold ice water for 5 min.

4. Then Sample was centrifugated at 3000 g for 15 min.

5. 0.4 ml of supernatant was removed and transferred to a new centrifuge tube.

6. 5 ml of acetic-sulfuric acid reagent (5 ml of concentrated sulfuric acid was added to 95 ml of glacial acetic acid) was added.

7. Centrifuge tube were placed in a boiling water bath and held for 30 min.

8. Tube was cooled under running water to room temperature.

9. Absorbance was measured on a spectrophotometer at a wavelength of $\lambda = 540$ nm against a blank sample (containing only acetic acid-sulfuric acid reagent).

Statistical analysis was performed in Microsoft Office Excel with usage of extension Real Statistics 2019 (by Charles Zaiontz, license CC BY 4.0). The statistical significance of the difference between groups was determined using the non-parametric analysis of variance by Kruskal-Wallis method, followed by pairwise comparisons using the Mann-Whitney U-test. The difference was considered statistically significant at $P < 0.05$. In the tables, data were presented in the form of mean and standard error (M±SE).

Results

Modelling of MetS disrupted lipid metabolism in rat organism leading to increase in TG, TC and LDL-C levels, simultaneously, HDL-C level was decreased (Fig. 1, *A*, *B*, *C*, *D*). Such changes in lipid metabolism may lead to atherosclerosis development. MetS also increased fasting blood glucose levels and insulin resistance indexes, which can lead to type II diabetes (Fig. 2, *A*, *B*, *C*).

Administration of chromium picolinate to healthy animals did not change glucose content in rat blood and insulin resistance indexes remained on the level of control group. In blood lipid profile the only thing that changed was LDL-C level, which decreased by 23.5% compared to control group.

Administration of chromium picolinate to animals on which we modeled MetS led to decrease in blood glucose level by 42.8% compared to MetS group. Insulin resistance indexes TyG-BMI and METS-IR decreased by 23.3 and 13.3% respectively, compared to MetS group. Content of TG, TC and LDL-C decreased by 55.6, 22.0 and 35.7% respectively, while HDL-C content increased by 84.2% compared to MetS group.

ECM of the heart contains structural proteins like collagens and elastins. Free L-hydroxyproline can serve as a fine marker of collagen degradation and possible sig of heart remodeling. Besides structural proteins ECM of the heart contains proteoglycans and glycoproteins, which are located in basement membrane, on cell surface or form amorphous substance of connective tissue. The extent of damage to proteoglycans can be assessed by evaluation of GAG content in tissue, while damage to glycoproteins can be evaluated by excessive accumulation of free sialic acids.

Metabolic syndrome modeling led to increased proteoglycan degradation in rat heart as evidenced by increase in GAG content with predominance of keratan-dermatan fraction of GAG. At the same time, MetS modeling led to increase collagenolysis and elevated degradation of glycoproteins in rat heart (Table).

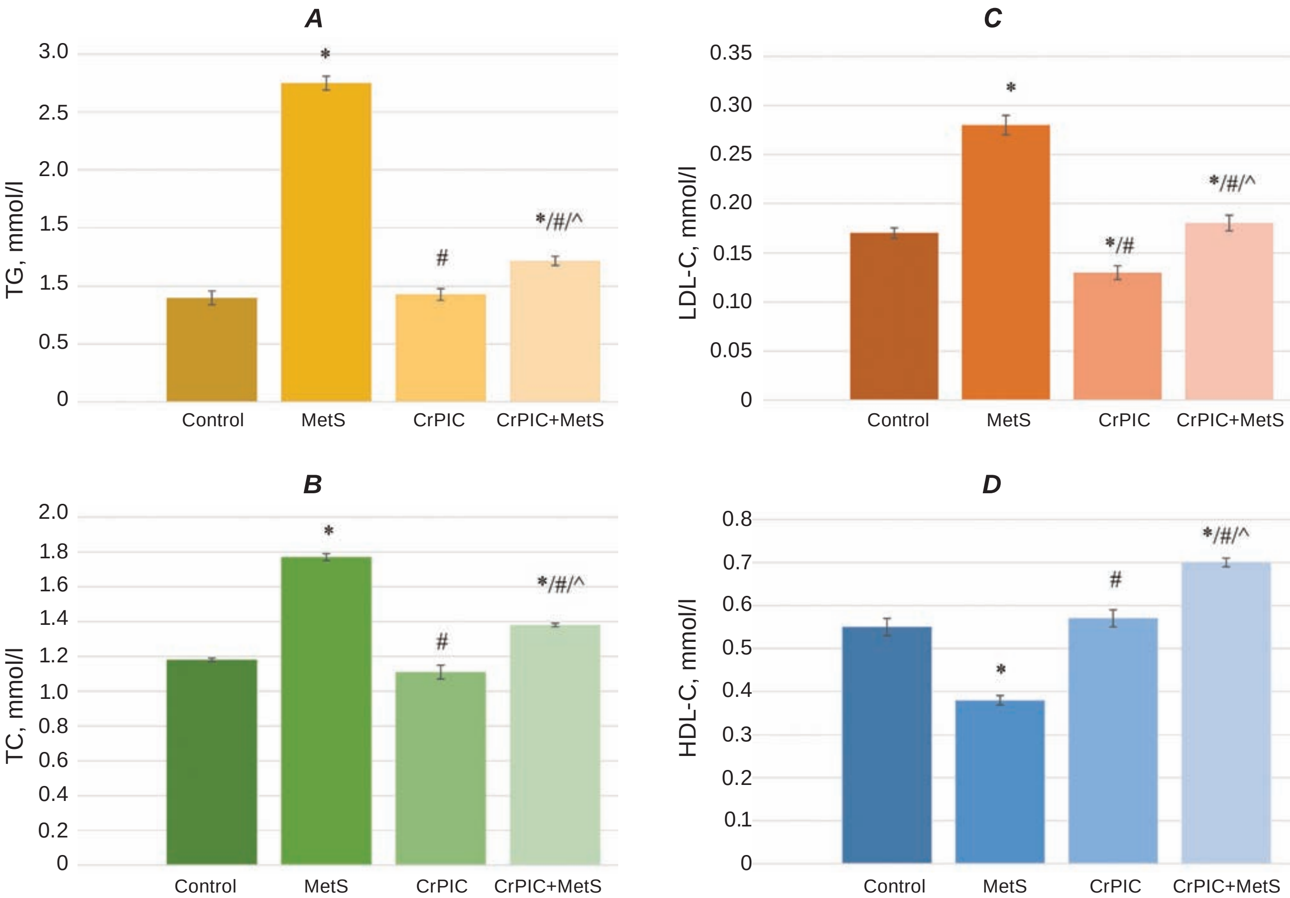

Fig. 1. ***A*** *– Triglycerides (TG) content in rat blood under chromium picolinate administration and metabolic syndrome modeling (M ± SE).* ***B*** *– Total cholesterol (TC) content in rat blood under conditions of chromium picolinate administration and metabolic syndrome modeling (M ± SE).* ***C*** *– Low density lipoprotein cholesterol (LDL-C) content in rat blood under chromium picolinate administration and metabolic syndrome modeling (M ± SE).* ***D*** *– High density lipoprotein cholesterol (HDL-C) content in rat blood under chromium picolinate administration and metabolic syndrome modeling (M ± SE). *The data are statistically significantly different from the control group (P < 0.05); #the data are statistically significantly different from the experimental metabolic syndrome group (P < 0.05); ^the data are statistically significantly different from the CrPIC group (P < 0.05)*

Administration of CrPIC to healthy animals increased total GAG content by 13.6%, heparin-heparan fraction by 29.9%, and chondroitin fraction by 16.3%, but did not change the content of keratan-dermatan fraction, free L-hydroxyproline and concentration of sialic acids compared to control group.

Comparing results in CrPIC group with data from MetS group we established, that total GAG content was decreased by 27.3%, heparin-heparan fraction by 18.7% keratan-dermatan fraction by 69.8% free L-hydroxyproline by 30.3% and concentration of sialic acids by 22.9%, but increased content of chondroitin fraction by 78.6% compared to MetS group.

Administration of CrPIC to animals on which we modeled MetS increased total GAG content in rat heart by 31.5%, content of heparin-heparan fraction by 55.7%, keratan-dermatan fraction by 10.4%, chondroitin fraction by 33.2%, concentration of sialic acids by 14.8%, but did not change concentration of free L-hydroxyproline compared to control group.

Combination of CrPIC introduction and MetS modeling decreased total GAG content in rat heart by 15.8%, content of keratan-dermatan fraction by 65.3%, concentration of free L-hydroxyproline by 30.8%, concentration of sialic acids by 12.2%, increased content of chondroitin fraction by 104.6%,

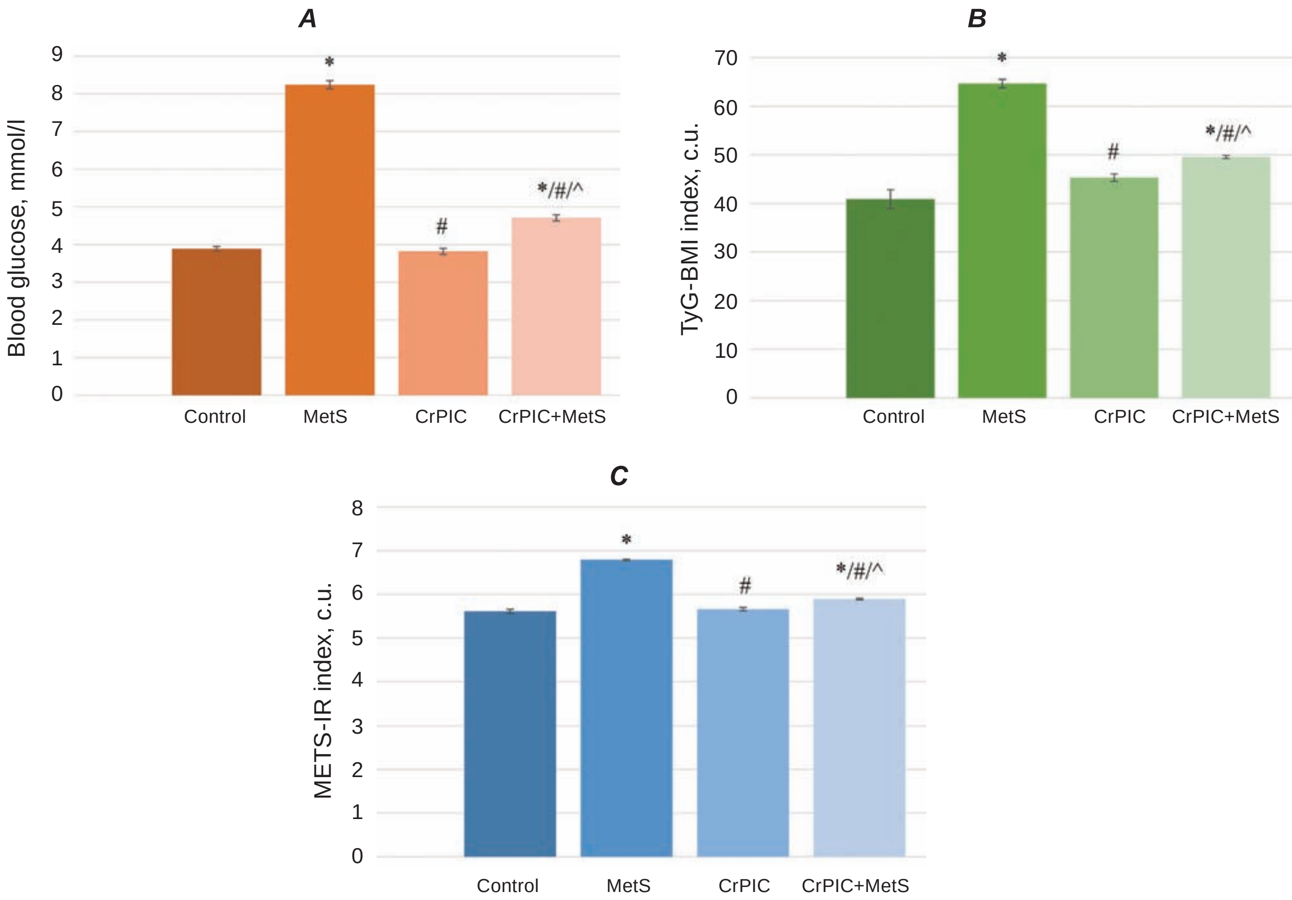

Fig. 2. ***A*** *– Changes of blood glucose levels under conditions of chromium picolinate administration and metabolic syndrome modeling (M ±S E).* ***B*** *– Changes of TyG-BMI index under conditions of chromium picolinate administration and metabolic syndrome modeling (M ± SE).* ***C*** *– Changes of METS-IR index under conditions of chromium picolinate administration and metabolic syndrome modeling (M ± SE). *The data are statistically significantly different from the control group (P < 0.05); # the data are statistically significantly different from the experimental metabolic syndrome group (P < 0.05); ^ the data are statistically significantly different from the CrPIC group (P < 0.05)*

but did not change concentration of heparin-heparan fraction compared to MetS group.

Modeling of MetS with simultaneous administration of CrPIC increased total GAG content in rat heart by 15.8%, content of heparin-heparan fraction by 19.8%, keratan-dermatan fraction by 15.1%, chondroitin fraction by 14.5%, but did not change concentration of free L-hydroxyproline and concentration of sialic acids by compared to CrPIC group.

Discussion

In the blood of rats on which we modeled MetS we observed development of dyslipidemia, evidenced by increased TG and TC levels, increased LDL-C and decreased HDL-C content (Fig. 1, *A*, *B*, *C*, *D*). We also observed hyperglycemia, evidenced by increased blood glucose level, and increased insulin resistance, evidenced by increased in TyG-BMI and METS-IR indexes (Fig. 2, *A*, *B*, *C*,). These metabolic changes are typic for MetS.

Administration of chromium picolinate to healthy animals did not show any adverse effects on metabolic parameters studied in our research. On contrary, CrPIC decreased LDL-C level in healthy animals, which may become beneficial for weight control and atherosclerosis prophylaxis (Fig. 1, *C*).

Administration of chromium picolinate to animals, on which we modeled MetS attenuated hyperlipidemia, evidenced by decreased TG, TC and LDL-C levels and increased HDL-C content (Fig. 1, *A*, *B*, *C*, *D*). Chromium picolinate also attenuated hyperglycemia, evidenced by decreased

T a b l e. Parameters of the extracellular matrix degradation in the heart of rats under the condition of chromium picolinate administration and metabolic syndrome modeling (M ± SE, n = 6)

Parameters	Groups			
	Control	MetS	CrPIC	CrPIC+MetS
Total GAG concentration, µmol/l	1.62 ± 0.01	2.53 ± 0.01*	1.84 ± 0.01*/#	2.13 ± 0.02*/#/^
Heparin-heparan fraction, µmol/l	0.361 ± 0.005	0.577 ± 0.009*	0.469 ± 0.010*/#	0.562 ± 0.004*/^
Keratan-dermatan fraction, µmol/l	0.442 ± 0.006	1.405 ± 0.010*	0.424 ± 0.011#	0.488 ± 0.010*/#/^
Chondroitin fraction, µmol/l	0.834 ± 0.012	0.543 ± 0.005*	0.970 ± 0.008*/#	1.111 ± 0.008*/#/^
Concentration of free L-hydroxyproline, µmol/g	0.473 ± 0.008	0.757 ± 0.046*	0.528 ± 0.023#	0.524 ± 0.036#
Concentration of sialic acids, mg/g	5.81 ± 0.03	7.60 ± 0.33*	5.86 ± 0.28#	6.67 ± 0.10*/#

Note. *The data are statistically significantly different from the control group (P < 0.05). #The data are statistically significantly different from the experimental metabolic syndrome group (P <0.05). ^The data are statistically significantly different from the CrPIC group (P <0.05)

blood glucose level (Fig. 2, *A*), and decreased insulin resistance (Fig. 2, *B*, *C*).

Increase in free L-hydroxyproline content in rat heart in MetS group evidences about increase in destruction of collagen fibers (collagenolysis), which in turn may be considered as the sign of heart remodeling, which may lead to cardiac fibrosis. The reason for increased L-hydroxyproline content in rat heart in MetS group may be activation of MMPs, namely MMP-9, which one of the key agents in heart remodeling [25]. Increase in heparin-heparan fraction of GAGs may be the indicator of cell injury and subsequent regeneration and may be seen as adoptive response in MetS group [26]. However, taking into account, that heparin-heparan fraction of GAGs stimulates lipid deposition in various tissues, such increase may be considered as a response to increased LDL-C and TC content in blood [27]. From one point of view, sharp increase in keratan-dermatan fraction of GAGs in rat heart may be also viewed as adoptive response to oxidative damage to heart tissues, since it has ability to stimulate antioxidant effects [28]. However, Large concentrations of keratan-dermatan fraction of GAGs, especially dermatan, may cause heart remodeling, with excessive deposition of collagen fibers [29]. We also should take into consideration, that keratan sulfate is a potent controller of inflammatory process, and increase in keratan-dermatan fraction may correspond to severity if metaflammation in the heart [30]. Chondroitin sulfate can increase endothelialization of blood vessels and diminish pro-inflammatory response [31]. Increase in content of this fraction of GAGs may indicate increased growth of the blood vessels, while its prolonged decrease may lead to atrophy of blood vessels [32]. Chondroitin sulfate can reduce short-chain fatty acid synthesis of intestinal flora, slow down inflammatory response, inhibit osteoclast differentiation, promote calcium absorption and antioxidant mechanism, and alleviate osteoporosis in low-calcium feeding rats [33]. Thus, a decrease in content of chondroitin fraction in MetS group may be a sign of exhaustion of adaptive mechanisms of heart. Increased sialic acids content in rat heart may be a sign of excessive LDL desialylation, which plays a significant role in the initiation and development of atherosclerosis. Low content of sialic acids in LDL is associated with increased cholesterol accumulation in human aortic smooth muscle cells and macrophages [34].

Increase of heparin-heparan and chondroitin fractions in rat heart on the background of administration of chromium picolinate to healthy animals shows potential beneficial role of chromium picolinate in heart ECM metabolism during physiological conditions. These fractions of GAG will ensure anti-inflammatory effect, increase amount of blood vessels and tissue regeneration [26, 32]. Absence of increase in free L-hydroxyproline and sialic acids content indicates low toxicity of chromium picolinate towards heart ECM.

Administration of chromium picolinate on the background of metabolic syndrome modeling alleviated negative effects of MetS on collagenolysis and desialization. As was mentioned before intensity of collagenolysis depends on activation of MMPs and inhibition of tissue inhibitors of matrix metalloproteinases (TIMPs). Most of MMPs are under direct transcriptional control of NF-κB, which is one of the key factors activated during MetS [35]. According to Gossa Al-Saadde DL et al. CrPIC administration is able to inhibit NF-κB activation and increase heme oxygenase 1 (HO-1) activity, subsequently leading to lower MMP activity and collagenolysis intensity [36]. The ability of CrPIC influence Nrf-2/HO-1 axis can potentially attribute to increase of antioxidant defense in heart tissues due to CrPIC-induced Nrf-2 activation [37]. Increase in sialic acids content during MetS is usually attributed to enhanced activity of sialidases, namely neuraminidase1, which are induced by NF-κB activation and increased cytokine production (especially TNF-alpha) [38, 39]. CrPIC can inhibit NF-κB through AMPK activation, which can cause decrease in cytokine production and systemic inflammation, leading to lower content of sialic acids in heart tissues observed in our study [36]. Another hypothetical mechanism by which CrPIC can decrease sialic acids content in rat heart is direct inhibition of sialidases in heart tissues. Such possibility is not excluded due to research done by Karhadkar TR et al., who showed, that picolinic acid can directly inhibit sialidase [40]. CrPIC contains three picolinic acid residues, therefore it theoretically can inhibit sialidase directly, however, further research is necessary to evaluate such possibility. Our previous studies also showed effectiveness of CrPIC supplementation on alleviation of negative effects of MetS modelling on degradation of ECM in skeletal muscles [41].

Food supplementation of patients suffering from MetS and T2D with well known trace metals like chromium is considered as alternative way to medicinal treatment of these states [42]. Chinese scientists established a connection between low chromium plasma levels and MetS [43]. Therefore, chromium supplementation is a theoretically sound approach to MetS treatment. There are evidences, that combination of chromium preparations with omega-3 fatty acids, vitamins and other supplements are effective in treatment of some metabolic changes in people with already developed metabolic syndrome [18, 44]. In perspective, CrPIC may be used as means for prevention of cardiac tissue remodeling during MetS due to its ability to decrease MetS-induced collagenolysis.

The limitations of our study are:

1. Effects of CrPIC were studied on animal model.

2. Small group size.

3. Limited number of biochemical parameters chosen for ECM degradation evaluation.

Taking into consideration the limitations of our study, we consider that further research regarding safe dosage and adverse effects is necessary before we can consider application of CrPIC as medicine for treatment and prophylaxis of MetS in humans.

Conclusions. Chromium picolinate administration attenuates insulin resistance and lipid metabolism disruption caused by metabolic syndrome modelling.

Administration of chromium picolinate to healthy animals stimulates predominant accumulation of chondroitin fraction of glycosaminoglycans in rat heart.

Chromium picolinate reduces metabolic syndrome induced collagenolysis and degradation of glycoproteins and proteoglycans of rat heart extracellular matrix.

Chromium picolinate changes the dominating fraction of glycosaminoglycans in rat heart during metabolic syndrome from keratan-dermatan to chondroitin fraction.

Conflct of interest. Authors have completed the Unified Conflicts of Interest form at http://ukrbiochemjournal.org/wp-content/uploads/2018/12/coi_disclosure.pdf and declare no conflict of interest.

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ПРОДУКТИ ДЕГРАДАЦІЇ ПОЗАКЛІТИННОГО МАТРИКСУ В СЕРЦІ ЩУРІВ З МЕТАБОЛІЧНИМ СИНДРОМОМ ЗА УМОВ ВВЕДЕННЯ ПІКОЛІНАТУ ХРОМУ

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Серед населення України спостерігається тенденція до зростання відсотка осіб з ожирінням, ускладненим метаболічним синдромом (MetS), який спричиняє пошкодження позаклітинного матриксу серця. Згідно з нещодавніми дослідженнями, піколінат хрому (CrPIC) має потенціал для послаблення порушень ліпідного обміну та захисту позаклітинного матриксу від деградації. Метою цього дослідження було оцінити ліпідний профіль крові та вміст глікозаміногліканів, L-гідроксипроліну та сіалової кислоти в серці щурів із змодельованим метаболічним синдромом за введення піколінату хрому. Статево зрілих щурів лінії Вістар розділили на 4 групи по 6 тварин у кожній – контрольна група; індукція метаболічного синдрому; введення CrPIC; метаболічний синдром + введення CrPIC. Метаболічний синдром відтворювали, використовуючи 20% розчин фруктози як єдине джерело води протягом 60 днів. CrPIC вводили перорально в дозі 80 мкг/кг щодня протягом 60 днів. Концентрацію протеїнів деградації позаклітинного матриксу серця визначали спектрофотометрично у 10% гомогенаті серця. Введення CrPIC здоровим тваринам стимулювало накопичення глікозаміногліканів хондроїтинової фракції в серці щурів. У тварин з метаболічним синдромом спостерігали підвищення рівня тригліцеридів, загального холестерину та холестерину ЛПНЩ у крові, посилення колагенолізу, деградації глікопротеїнів та глікозаміногліканів з переважанням кератан-дерматанової фракції. Проте, введення цим тваринам CrPIC зменшило колагеноліз та деградацію глікопротеїнів, змінило домінуючу фракцію глікозаміногліканів із кератан-дерматану на хондроїтин у сполучній тканині серця, що вказує на його потенціал для запобігання ремоделюванню серцевої тканини у разі метаболічного синдрому.

Ключові слова: метаболічний синдром, серце, екстрацелюлярний матрикс, L-гідроксипролін, глікозаміноглікани, піколінат хрому.

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