Oxidative stress features in patients with combined course of type 2 diabetes mellitus and osteoporosis residing in anthropogenic pressure conditions

V. I. Kryvenko, O. I. Borodavko*
Zaporizhzhia State Medical University, Ukraine

Key words: oxidative stress, osteoporosis, type 2 diabetes mellitus.

The purpose of the study is to compare indicators of oxidative stress (OS) and bone mineral density in patients with combined course of type 2 diabetes mellitus (DM) and osteoporosis (OP), who are permanently resident in industrial regions and in environmentally friendly conditions.

Materials and methods. A total of 74 patients with combined course of type 2 DM and OP have been examined. The main group included 50 patients who had permanent residence in Zaporizhzhia city (industrial region). The comparison group included 24 patients with combined course of type 2 DM and OP who were born and lived in Mykhailivka town of the Zaporizhzhia region (ecological region). All the patients underwent ultrasound densitometry using the Omnisense 7000 ultrasound densitometer (Beam Medical Ltd, Israel). Osteoporosis risk index (ORI) was calculated for each patient as an average index for 2 bones. The OS intensity was assessed by nitrotyrosine and 8-hydroxyguanine serum levels. Serum glucose and glycated hemoglobin (HbA1c) were also measured.

Results. The main group patients did not significantly differ from the comparison group patients in terms of OP risk factors, age, gender, duration and course of type 2 DM. The mean values of nitrotyrosine and 8-hydroxyguanine levels were significantly 2.5 times (P < 0.05) and 1.4 times (P < 0.05) higher, respectively, in the main group as compared to similar patients from the environmentally friendly region. The bone mineral density in the industrial region group was significantly lower. Thus, the Rad-Tib ORI was 12 % higher in the main group, Rad-Ph ORI – by 19 %, and Tib-Ph ORI – by 16 % compared with the comparison group (P < 0.05).

Conclusions. The processes of OS are intensified and severity of osteoporosis is increased in the patients with combined course of type 2 diabetes mellitus and OP who have been permanently living in the large industrial city compared to the residents of the environmentally friendly region.

Особливості оксидативного стресу у хворих із поєднаним перебігом цукрового діабету 2 типу та остеопорозу, які постійно проживав в умовах антропогенного навантаження

В. І. Кривенко, О. І. Бородавко

Мета роботи – здійснити порівняльний аналіз показників оксидативного стресу (ОС) і мінеральної щільності кісткової тканини у хворих із поєднаним перебігом цукрового діабету (ЦД) 2 типу та остеопорозу (ОП), які постійно проживають у промисловому регіоні та в екологічно сприятливих умовах.

Матеріали та методи. Обстежено 74 пацієнтів з поєднаним перебігом ЦД 2 типу та ОП. До основної групи увійшли 50 пацієнтів, які постійно проживали у м. Запоріжжі (промисловий регіон). До групи порівняння увійшли 24 пацієнти з поєднаним перебігом ЦД 2 типу та ОП, які народилися й проживали у смт Михайлівка Запорізької області (условно сприятливий регіон).

Результати. Пацієнти основної групи більші за факторами ризику ОП, віком, статтю, тривалістю та перебігом ЦД 2 типу від осіб групи порівняння. Середнє значення рівня нітротирозину та 8-гідроксигуаніну в основній групі складало 2.5 рази (P < 0.05) та 1.4 рази (P < 0.05) відповідно порівняно з аналогічними хворими з екологічно сприятливим регіоном. Основна група мала значно нижчу мінеральну щільність кісткової тканини в порівнянні з контрольною групою. Так, IRO Rad-Tib був на 17 % вищим в основній групі, IRO Rad-Ph – на 19 %, а IRO Tib-Ph – на 27 % щодо групи порівняння.

Висновки. У хворих із поєднаним перебігом ЦД 2 типу та ОП, які постійно проживали в умовах великої індустріального регіону, виявлено стійкість OKO та гідроспіральний регіон.
Introduction

People living in conditions of technogenic civilization are subjected to development of pathologic changes in organs and systems. The negative influence of environmental factors (air pollution by emissions from transport and industrial enterprises, xenobiotics, ultraviolet radiation, etc.) is accompanied by the increased formation of free radicals [1]. According to the literature, oxidative stress (OS) caused by adverse environmental conditions is one of the determining factors in the pathogenesis of a range of diseases, including diabetes mellitus (DM) and osteoporosis (OP) [1,2].

Harmful substances can affect bone tissue, both directly and indirectly. Therefore, cadmium, in particular, inhibits the formation of a hormonally active form of vitamin D3 (calcitriol) in proximal renal tubules, which leads to a decrease in the calcium absorption from the digestive tract [3,4]. Lead is accumulated in bone tissue, replacing calcium and thereby decreasing bone mineral density (BMD) [5].

Indirect impact is caused by OS development. As a result of environmental contamination, active oxygen forms are accumulated in large quantities in the body, which stimulates the processes of protein and lipid peroxidation and contributes to a reduction in antioxidant defense system activity [1].

In the available literature, we have not found any reports on the OS study in patients with combined course of type 2 DM and OP with regard to the impact of anthropogenic stress, which requires further research.

The purpose

The purpose of the study is to compare indicators of oxidative stress (OS) and bone mineral density in patients with combined course of type 2 diabetes mellitus (DM) and osteoporosis (OP), who are permanently resident in industrial regions and in environmentally friendly conditions.

Materials and methods

We have examined 74 patients with the combined course of type 2 DM and OP. The main group included 50 patients who lived permanently in the city of Zaporizhzhia (industrial region). According to the data collected in the Institute of Medical and Environmental Problems of Zaporizhzhia State Medical University, the content of industrial pollutants in the environment exceeds the maximum permissible concentrations, in particular, heavy metal compounds (lead, copper, manganese, etc.) – 4.7 times, benz[a]pyrene – 6.4 times, ammonia – 1.6 times, phenol – 1.3 times, fluorides – 2 times, sulfur dioxide – 2.5 times, hydrogen sulfide – 1.5 times [6].

The comparison group included 24 patients with combined course of type 2 DM and OP who were born and lived in Mykhalivka town of the Zaporizhzhia region. According to the Main Department of Statistics in the Zaporizhzhia region, this urban-type settlement is located in the region with a relatively favorable ecological situation. For example, in 2014, 52 kg of harmful substances were emitted per one resident of the Mykhalivka district per year, while this rate was 163 kg per inhabitant in Zaporizhzhia per year. In addition, the main specific pollutants content, according to the official statistics, is much lower in the atmospheric air of the Mikhailivsky district than in Zaporizhzhia city [6].

The following inclusion criteria were applied: a written informed consent obtained from each patient to participate in the study, verified diagnosis of type 2 DM (according to the Health Ministry of Ukraine Order No. 1118 dated 21.12.2012), verified diagnosis of osteoporosis (according to the Health Ministry of Ukraine Order No. 676 dated 12 October, 2006, and the recommendations of the International Society for Clinical Densitometry, 2015), age from 50 to 70 years old.

Exclusion criteria included diagnosis of type 1 diabetes, secondary insulin-dependent DM, glucocorticosteroids intake for more than 3 months, thyroid disorders, chronic heart failure, chronic kidney disease, systemic connective tissue diseases, women with premature (up to 45 years of age) or artificial menopause.

All the patients underwent ultrasound densitometry of the three bones, namely, proximal phalanx of the III finger, the distal radius, the midshaft tibia using the Omnisense 7000 ultrasound densitometer (Beam Medical Ltd., Israel). The bone tissue assessment was performed according to the WHO criteria: normal condition – T-index > -1.0 SD, osteopenia – T-index – from -1.0 to -2.5 SD, osteoporosis – T-index < -2.5 SD [7].
Table 1. Characteristics of the examined patients with combined course of type 2 DM and OP depending on the region of residence

| Patients characteristics                        | Main group n = 50 | Comparison group n = 24 | P  |
|------------------------------------------------|-------------------|--------------------------|----|
| Age, years                                     | 61.0 (56.0; 65.0) | 62.5 (75.5; 64.0)        | 0.71|
| Gender (m/f)                                    | 20/31             | 19/14                    | 0.89|
| DM duration, years                             | 5.0 (2.0; 8.0)    | 4.0 (2.0; 6.5)           | 0.38|
| Glucose, mmol/l                                | 10.9±2.4          | 10.1±1.9                 | 0.39|
| HbA1c, %                                       | 8.7 (7.9; 10.6)   | 8.4 (7.6; 10.1)          | 0.45|
| Reduced sensitivity in the lower limbs (yes/no), % | 58/42             | 11/13                    | 0.32|
| Pain in the lower limbs (yes/no), %             | 22/78             | 16/84                    | 0.90|
| Heredity for OP (yes/no), %                    | 12/88             | 8/92                     | 0.95|
| Back pain (yes/no), %                          | 44/56             | 54/46                    | 0.13|
| Fractures with minimal trauma, (yes/no), %     | 10/90             | 25/75                    | 0.81|
| Height loss 3 cm or more (yes/no), %           | 6/94              | 2/22                     | 0.80|
| Daily alcohol intake more than 2 doses, (yes/no), % | 16/84             | 8/92                     | 0.70|
| Smoking (yes/no), %                            | 16/84             | 21/79                    | 0.61|
| Daily physical activity less than 30 minutes, (yes/no), % | 8/92              | 8/92                     | 0.94|
| Food intolerance (yes/no), %                   | 16/84             | 12/88                    | 0.40|

Table 2. The levels of nitrotyrosine and 8-hydroxyguanine in patients with type 2 diabetes and osteoporosis, depending on the region of residence

| Indicator                                  | Main group n = 50 | Comparison group n = 24 | P  |
|--------------------------------------------|-------------------|--------------------------|----|
| Nitrotyrosine, nmol/ml                     | 41.4 (11.7; 56.5) | 16.6 (10.0; 32.8)        | 0.006|
| 8-hydroxyguanine, nmol/ml                  | 13.0 (9.9; 15.4)  | 9.0 (7.3; 11.5)          | 0.008|

Table 3. Ultrasonic densitometry indicators in patients with a combined course of type 2 DM and OP depending on the region of residence

| Indicators                  | Main group n = 50 | Comparison group n = 24 | P  |
|----------------------------|-------------------|--------------------------|----|
| T-index for the radius     | -2.7 ± 0.9        | -2.5 ± 1.3               | 0.76|
| T-index for the tibia       | -2.4 ± 0.9        | -2.1 ± 1.2               | 0.30|
| T-index for the phalanx     | -1.4 ± 1.1        | -1.2 ± 0.9               | 0.36|
| Rad-Tib ORI                | -2.5 ± 0.6        | -2.2 ± 0.1               | 0.04|
| Rad-Ph ORI                 | -2.1 ± 0.4        | -1.7 ± 0.5               | 0.03|
| Tib-Ph ORI                 | -1.9 ± 0.7        | -16 ± 0.8                | 0.04|

Osteoporosis risk index (ORI) was calculated for each patient as an average index for 2 bones. When conducting ultrasound densitometry of the three bones, the following combinations were possible: radius bone – tibia (Rad-Tib), radius bone – phalanx (Rad-Ph), tibia– phalanx (Tib-Ph).

Results

Table 1 shows the characteristics of the examined patients. The main group patients did not significantly differ from the comparison group patients in terms of the OP risk factors such as age, gender, duration and course of type 2 DM.

The mean values of nitrotyrosine and 8-hydroxyguanine levels were significantly 2.5 times (P < 0.05) and 1.4 times (P < 0.05) higher, respectively, in the main group as compared to similar patients from the environmentally friendly region (Table 2).

Osteoporosis risk index (ORI) was calculated for each patient as an average index for 2 bones. When conducting ultrasound densitometry of the three bones, the following combinations were possible: radius bone – tibia (Rad-Tib), radius bone – phalanx (Rad-Ph), tibia– phalanx (Tib-Ph).

Thus, the Rad-Tib ORI was 12 % higher in the main group, Rad-Ph ORI – by 19 %, and Tib-Ph ORI – by 16 % compared with the comparison group (P < 0.05).

It was specified by the correlation analysis that the increase in nitrotyrosine level was significantly associated with a decrease in T-index for the tibia, radius and Rad-Tib ORI (r = -0.42, P < 0.05; r = -0.37, P = 0.05; r = -0.34, P < 0.05).

An increase in 8-hydroxyguanine level had a significant negative correlation with the T-index for the tibia, Rad-Tib ORI and Tib-Ph ORI (r = -0.38, P < 0.05; r = -0.41, P < 0.05; r = -0.35, P < 0.05).
Discussion

The groups of examined patients were comparable, which is important to exclude the factors that may affect the OS markers. In individuals who have been living in the industrial region, the levels of nitrotyrosine and 8-hydroxyguanine were significantly higher compared to the similar patients from the environmentally friendly region. These data indicate the OS intensification and point to a high oxidative modification of proteins and DNA in patients with combined course of type 2 DM and OP who have been constantly exposed to anthropogenic pressure (Table 2).

It should be noted that endogenous production of active oxygen forms occurs during normal physiological processes and such factors as industrial pollutants, ultraviolet radiation or air pollution contribute to their formation and accumulation [11]. It is believed that the body of an individual who resides in an industrial region can be affected by several hundred thousand of chemicals, combined effect of which can lead to an increase in their toxicity, despite their small amounts [12]. According to scientists, the workers exposed to carbon dioxide demonstrated an increase in malonic aldehyde level and a decrease in antioxidant system markers – superoxide dismutase, catalase and peroxidase, which indicates the OS development [13].

The literature data show that active oxygen forms can affect bone tissue [10]. In our study, the groups did not differ significantly by T-indexes for phalanx, tibia and radius bones, but a significant difference was found by ORI, which is a generalized indicator of ultrasound densitometry and indicates the severity of OP, which may be explained by higher OS rates. The obtained data are confirmed by the presence of a possible reverse correlation of the OS markers levels with ORI and T-index.

There are limited literature data that city residents have a higher incidence of OP and osteopenia compared to rural residents. So, according to V. V. Povorozniuk, V. M. Vajda, N. I. Dzerovych (2010), a significantly higher incidence of OP was registered among urban compared to rural residents [14].

The results of the experimental studies testify the development of structural changes in bone tissue, as well as an imbalance in the bone turnover under the influence of combined effects of toxicants [4]. Industrial pollutants promote activation of free radical processes in case of antioxidant deficiency, which adversely affects the BMD, bone turnover causing the development of OP. It was revealed that the workers who have harmful working conditions exhibit changes in the bone turnover and OP development more often and at younger ages [15].

The results obtained indicate that the OS processes are intensified in patients with a combined course of type 2 diabetes and OP who have been permanently living in the large industrial city compared with the residents of the environmentally friendly region. It is logical to assume that in the conditions of chronic oxidative stress, there is a dysregulation of adaptive processes and a decrease in the reserve capacity of adaptive responses. It is advised that the patients with combined course of type 2 DM and OP permanently residing under anthropogenic pressure should use pharmacological agents with antioxidant properties in the complex treatment to reduce the effect OS.

Conclusions

1. There is an increase in nitrotyrosine and 8-hydroxyguanine levels in patients with combined course of type 2 DM and OP who permanently reside in the industrial region, which indicates the development of OS in this category of patients.

2. Individuals who permanently reside under the anthropogenic stress have an increase in the severity of osteoporosis according to ORI, compared to the similar patients from the ecological region.

Funding

The study was carried out within the framework of the research work Zaporizhzhia State Medical University: “Diagnosis, treatment and prophylaxis of comorbid pathology of internal organs in the conditions of the industrial region” (state registration number 0115U001765).

Conflicts of interest: authors have no conflict of interest to declare.

References

[1] Nagornaya, N. V., & Chetverik, N. A. (2010). Oksidativnyj stress: vliyaniye na organizm cheloveka, metody ocenki [Oxidative Stress: Its Influence on a Human Body, Estimation Methods]. Zdorov'ye rebenka, 2(23), 140–145. [in Russian].

[2] Sendur, O. F., Turan, Y., Tastaban, E., & Sertler, M. (2009). Antioxidant status in patients with osteoporosis: a controlled study. Joint Bone Spine, 76(5), 514–8. doi: 10.1016/j.jbspin.2009.02.005

[3] Antonyak, H. L., Babych, N. O., Biletska, L. P., Panas, N. E., & Zhylyshchych, Y. V. (2010). Kadmii v orhanizmi liudyny i tvaryn. Vplyv na organizm cheloveka, metody ocenki [Oxidative Stress: Its Influence on a Human Body, Estimation Methods]. Zdorov'ye rebenka, 2(23), 140–145. [in Russian].

[4] Fryer, S. G., Dye, A., & Farooq, S. (2009). Oxidative stress in diabetes mellitus [Review]. European Journal of Preventive Medicine, 27(S1), S8–S10. doi: 10.1016/j.ejpm.2009.03.004

Zaporozhye medical journal. Volume 21. No. 2, March – April 2019
ISSN 2306-4145  http://zmj.zsmu.edu.ua 185
II. Effect on functional activity of organs and systems. Biologichni studii, Studia Biologica, 4(3), 125–136. [in Ukrainian]. doi: https://doi.org/10.30970/sbi.0403.110

[4] Khopta, N. S. (2015). Vplyv solei kadmiiu ta nitrytiv na metabolizm u kistkovoi tkanyni [Influence of cadmium and nitrite salts on bone tissue metabolism]. [Extended abstract of candidate’s thesis]. Lviv. [in Ukrainian].

[5] Biletska, E. M., Onul, N. M., & Kalinchcheva, V. V. (2016). Porivnialna otsinka bioprotektornoi dii tsynku v orhanichnii ta neorhanichnii formi na osteotropnist svyntciu v eksperimentalnykh umovakh [Comparative evaluation of bioprotective action of zinc in organic and inorganic form on osteotropic lead in experimental conditions]. Medychnyi perspektivy, 21(4), 123–129. [in Ukrainian].

[6] Holoveshka, V. P. (2015). Statystychnyi zhoporizkoi oblasti za 2014 rok [Statistical Yearbook of Zaporizhzhia region for 2014]. Zaporizhzhia: Holovne upravlinnia statystyky u Zaporizkii oblasti. [in Ukrainian].

[7] (2015) Official Positions of The International Society for Clinical Densitometry – 2015. Retrieved from https://iscd.app.box.com/v/OP-ISCD-2015-Adul

[8] Makurina, G. I. (2015). Nitroziruyushchij stress pri psoriaticheskoi bolezni v sochetanii s e'ksperimental'noj gipertennej [Nitrosative Stress in Psoriatic Diseases Combined with Essential Hypertension]. Vestnik problem biologii i medcyny, 2, 3(123), 159–165. [in Russian].

[9] Pesta, D., & Roden, M. (2017). The Janus Head of Oxidative Stress in Metabolic Diseases and During Physical Exercise. Curr Diab Rep., 17(6), 41. doi: 10.1007/s11992-017-0867-2

[10] Pastushikova, E. V. (2016). Analiz syvayi vozniknoveniya ekologicheskikh obstanevok v donshceiskoj oblasti [Analysis of the occurrence of ecological conditions in the example of Sverdlovsk region]. Nauchno obozrenie. Biologicheskie nauki, 6, 53–59. [in Russian].

[11] Da Costa, L. A., Garcia-Bailo, B., Badawi, A., & El-Sohemy, A. (2012). Genetic determinants of dietary antioxidant status. Mol. Biol. Transl. Sci., 108, 170–203. doi: 10.1016/B978-0-12-398397-8.00008-3

[12] Povorozniuk, V. V., Vajda, V. M., & Dzerovych, N. I. (2010). Vikovi ta statovi osoblyvosti chastoty perelomiv stehnovoi kistky u naselennia Zakarpatskoi oblasti. Problemy stareniya i dolgoletiya, 1, 99–106. [in Ukrainian].

[13] Bikmetova, E’. R., Ramazanova, L. M., Men’shikova, I. A., & Kulagina, I. G. (2010). Osobennosti obmena kostnoj tkani u rabotnikov khimicheskogo predpriyatiya, imeyushchih proizvodstvennyj kontakt s klorproizvodnymi nizkomolekulyarnymi alifaticheskimi uglavodorodov [Features of bone tissue exchange in workers of a chemical enterprise who have industrial contact with chlorine derivatives of low molecular weight aliphatic hydrocarbons]. Klinicheskaya biokhimiya: edinstvo fundamental’noj nauki i laboratornoj diagnostiki. Proceedings of the Regional Scientific and Practical Conference (P. 20–24). Izhevsk. [in Russian].