GEORGIAN MEDICAL NEWS

ISSN 1512-0112

No 7-8 (280-281) Июль-Август 2018

ТБИЛИСИ - NEW YORK

ЕЖЕМЕСЯЧНЫЙ НАУЧНЫЙ ЖУРНАЛ

Медицинские новости Грузии

საქართველოს სამედიცინო სიახლენი

GEORGIAN MEDICAL NEWS

No 7-8 (280-281) 2018

Published in cooperation with and under the patronage
of the Tbilisi State Medical University

Издается в сотрудничестве и под патронажем
Тбилисского государственного медицинского университета

გამოიცემა თბილისის სახელმწიფო სამედიცინო უნივერსიტეტთან
თანამშრომლობითა და მისი პატრონაჟით

ЕЖЕМЕСЯЧНЫЙ НАУЧНЫЙ ЖУРНАЛ

ТБИЛИСИ - НЬЮ-ЙОРК

GMN: Georgian Medical News is peer-reviewed, published monthly journal committed to promoting the science and art of medicine and the betterment of public health, published by the GMN Editorial Board and The International Academy of Sciences, Education, Industry and Arts (U.S.A.) since 1994. **GMN** carries original scientific articles on medicine, biology and pharmacy, which are of experimental, theoretical and practical character; publishes original research, reviews, commentaries, editorials, essays, medical news, and correspondence in English and Russian.

GMN is indexed in MEDLINE, SCOPUS, PubMed and VINITI Russian Academy of Sciences. The full text content is available through EBSCO databases.

GMN: Медицинские новости Грузии - ежемесячный рецензируемый научный журнал, издаётся Редакционной коллегией и Международной академией наук, образования, искусств и естествознания (IASEIA) США с 1994 года на русском и английском языках в целях поддержки медицинской науки и улучшения здравоохранения. В журнале публикуются оригинальные научные статьи в области медицины, биологии и фармации, статьи обзорного характера, научные сообщения, новости медицины и здравоохранения.

Журнал индексируется в MEDLINE, отражён в базе данных SCOPUS, PubMed и ВИНИТИ РАН. Полнотекстовые статьи журнала доступны через БД EBSCO.

GMN: Georgian Medical News – საქართველოს სამედიცინო სიახლენი – არის ყოველთვიური სამეცნიერო სამედიცინო რეცენზირებადი ჟურნალი, გამოიცემა 1994 წლიდან, წარმოადგენს სარედაქციო კოლეგიისა და აშშ-ის მეცნიერების, განათლების, ინდუსტრიის, ხელოვნებისა და ბუნებისმეტყველების საერთაშორისო აკადემიის ერთობლივ გამოცემას. GMN-ში რუსულ და ინგლისურ ენებზე ქვეყნდება ექსპერიმენტული, თეორიული და პრაქტიკული ხასიათის ორიგინალური სამეცნიერო სტატიები მედიცინის, ბიოლოგიისა და ფარმაციის სფეროში, მიმოხილვითი ხასიათის სტატიები.

ჟურნალი ინდექსირებულია MEDLINE-ის საერთაშორისო სისტემაში, ასახულია SCOPUS-ის, PubMed-ის და ВИНИТИ РАН-ის მონაცემთა ბაზებში. სტატიების სრული ტექსტი ხელმისაწვდომია EBSCO-ს მონაცემთა ბაზებიდან.

МЕДИЦИНСКИЕ НОВОСТИ ГРУЗИИ

Ежемесячный совместный грузино-американский научный электронно-печатный журнал
Агентства медицинской информации Ассоциации деловой прессы Грузии,
Академии медицинских наук Грузии, Международной академии наук, индустрии,
образования и искусств США.
Издается с 1994 г., распространяется в СНГ, ЕС и США

Website:
www.geomednews.org

The International Academy of Sciences, Education, Industry & Arts. P.O.Box 390177,
Mountain View, CA, 94039-0177, USA. Tel/Fax: (650) 967-4733

Версия: печатная. **Цена:** свободная.
Условия подписки: подписка принимается на 6 и 12 месяцев.
По вопросам подписки обращаться по тел.: 293 66 78.
Контактный адрес: Грузия, 0177, Тбилиси, ул. Асатиани 7, III этаж, комната 313
тел.: 995(32) 254 24 91, 995(32) 222 54 18, 995(32) 253 70 58
Fax: +995(32) 253 70 58, e-mail: ninomikaber@hotmail.com; nikopir@dgmholding.com

По вопросам размещения рекламы обращаться по тел.: 5(99) 97 95 93

GEORGIAN MEDICAL NEWS
Monthly Georgia-US joint scientific journal published both in electronic and paper formats of the Agency of Medical Information of the Georgian Association of Business Press; Georgian Academy of Medical Sciences; International Academy of Sciences, Education, Industry and Arts (USA).
Published since 1994. Distributed in NIS, EU and USA.

CONTACT ADDRESS IN TBILISI

GMN Editorial Board
7 Asatiani Street, 3th Floor
Tbilisi, Georgia 0177

Phone: 995 (32) 254-24-91
995 (32) 222-54-18
995 (32) 253-70-58
Fax: 995 (32) 253-70-58

CONTACT ADDRESS IN NEW YORK

NINITEX INTERNATIONAL, INC.
3 PINE DRIVE SOUTH
ROSLYN, NY 11576 U.S.A.

Phone: +1 (917) 327-7732

WEBSITE
www.geomednews.org

К СВЕДЕНИЮ АВТОРОВ!

При направлении статьи в редакцию необходимо соблюдать следующие правила:

1. Статья должна быть представлена в двух экземплярах, на русском или английском языках, напечатанная через **полтора интервала на одной стороне стандартного листа с шириной левого поля в три сантиметра**. Используемый компьютерный шрифт для текста на русском и английском языках - **Times New Roman** (**Кириллица**), для текста на грузинском языке следует использовать **AcadNusx.** Размер шрифта - **12**. К рукописи, напечатанной на компьютере, должен быть приложен CD со статьей.

2. Размер статьи должен быть не менее десяти и не более двадцати страниц машинописи, включая указатель литературы и резюме на английском, русском и грузинском языках.

3. В статье должны быть освещены актуальность данного материала, методы и результаты исследования и их обсуждение.

При представлении в печать научных экспериментальных работ авторы должны указывать вид и количество экспериментальных животных, применявшиеся методы обезболивания и усыпления (в ходе острых опытов).

4. К статье должны быть приложены краткое (на полстраницы) резюме на английском, русском и грузинском языках (включающее следующие разделы: цель исследования, материал и методы, результаты и заключение) и список ключевых слов (key words).

5. Таблицы необходимо представлять в печатной форме. Фотокопии не принимаются. **Все цифровые, итоговые и процентные данные в таблицах должны соответствовать таковым в тексте статьи**. Таблицы и графики должны быть озаглавлены.

6. Фотографии должны быть контрастными, фотокопии с рентгенограмм - в позитивном изображении. Рисунки, чертежи и диаграммы следует озаглавить, пронумеровать и вставить в соответствующее место текста **в tiff формате**.

В подписях к микрофотографиям следует указывать степень увеличения через окуляр или объектив и метод окраски или импрегнации срезов.

7. Фамилии отечественных авторов приводятся в оригинальной транскрипции.

8. При оформлении и направлении статей в журнал МНГ просим авторов соблюдать правила, изложенные в «Единых требованиях к рукописям, представляемым в биомедицинские журналы», принятых Международным комитетом редакторов медицинских журналов - http://www.spinesurgery.ru/files/publish.pdf и http://www.nlm.nih.gov/bsd/uniform_requirements.html
В конце каждой оригинальной статьи приводится библиографический список. В список литературы включаются все материалы, на которые имеются ссылки в тексте. Список составляется в алфавитном порядке и нумеруется. Литературный источник приводится на языке оригинала. В списке литературы сначала приводятся работы, написанные знаками грузинского алфавита, затем кириллицей и латиницей. Ссылки на цитируемые работы в тексте статьи даются в квадратных скобках в виде номера, соответствующего номеру данной работы в списке литературы. Большинство цитированных источников должны быть за последние 5-7 лет.

9. Для получения права на публикацию статья должна иметь от руководителя работы или учреждения визу и сопроводительное отношение, написанные или напечатанные на бланке и заверенные подписью и печатью.

10. В конце статьи должны быть подписи всех авторов, полностью приведены их фамилии, имена и отчества, указаны служебный и домашний номера телефонов и адреса или иные координаты. Количество авторов (соавторов) не должно превышать пяти человек.

11. Редакция оставляет за собой право сокращать и исправлять статьи. Корректура авторам не высылается, вся работа и сверка проводится по авторскому оригиналу.

12. Недопустимо направление в редакцию работ, представленных к печати в иных издательствах или опубликованных в других изданиях.

При нарушении указанных правил статьи не рассматриваются.

REQUIREMENTS

Please note, materials submitted to the Editorial Office Staff are supposed to meet the following requirements:

1. Articles must be provided with a double copy, in English or Russian languages and typed or computer-printed on a single side of standard typing paper, with the left margin of **3** centimeters width, and **1.5** spacing between the lines, typeface - **Times New Roman** (**Cyrillic**), print size - **12** (referring to Georgian and Russian materials). With computer-printed texts please enclose a CD carrying the same file titled with Latin symbols.

2. Size of the article, including index and resume in English, Russian and Georgian languages must be at least 10 pages and not exceed the limit of 20 pages of typed or computer-printed text.

3. Submitted material must include a coverage of a topical subject, research methods, results, and review.

Authors of the scientific-research works must indicate the number of experimental biological species drawn in, list the employed methods of anesthetization and soporific means used during acute tests.

4. Articles must have a short (half page) abstract in English, Russian and Georgian (including the following sections: aim of study, material and methods, results and conclusions) and a list of key words.

5. Tables must be presented in an original typed or computer-printed form, instead of a photocopied version. **Numbers, totals, percentile data on the tables must coincide with those in the texts of the articles.** Tables and graphs must be headed.

6. Photographs are required to be contrasted and must be submitted with doubles. Please number each photograph with a pencil on its back, indicate author's name, title of the article (short version), and mark out its top and bottom parts. Drawings must be accurate, drafts and diagrams drawn in Indian ink (or black ink). Photocopies of the X-ray photographs must be presented in a positive image in **tiff format**.

Accurately numbered subtitles for each illustration must be listed on a separate sheet of paper. In the subtitles for the microphotographs please indicate the ocular and objective lens magnification power, method of coloring or impregnation of the microscopic sections (preparations).

7. Please indicate last names, first and middle initials of the native authors, present names and initials of the foreign authors in the transcription of the original language, enclose in parenthesis corresponding number under which the author is listed in the reference materials.

8. Please follow guidance offered to authors by The International Committee of Medical Journal Editors guidance in its Uniform Requirements for Manuscripts Submitted to Biomedical Journals publication available online at: http://www.nlm.nih.gov/bsd/uniform_requirements.html
http://www.icmje.org/urm_full.pdf
In GMN style for each work cited in the text, a bibliographic reference is given, and this is located at the end of the article under the title "References". All references cited in the text must be listed. The list of references should be arranged alphabetically and then numbered. References are numbered in the text [numbers in square brackets] and in the reference list and numbers are repeated throughout the text as needed. The bibliographic description is given in the language of publication (citations in Georgian script are followed by Cyrillic and Latin).

9. To obtain the rights of publication articles must be accompanied by a visa from the project instructor or the establishment, where the work has been performed, and a reference letter, both written or typed on a special signed form, certified by a stamp or a seal.

10. Articles must be signed by all of the authors at the end, and they must be provided with a list of full names, office and home phone numbers and addresses or other non-office locations where the authors could be reached. The number of the authors (co-authors) must not exceed the limit of 5 people.

11. Editorial Staff reserves the rights to cut down in size and correct the articles. Proof-sheets are not sent out to the authors. The entire editorial and collation work is performed according to the author's original text.

12. Sending in the works that have already been assigned to the press by other Editorial Staffs or have been printed by other publishers is not permissible.

Articles that Fail to Meet the Aforementioned Requirements are not Assigned to be Reviewed.

ავტორთა საყურადღებოდ!

რედაქციაში სტატიის წარმოდგენისას საჭიროა დავიცვათ შემდეგი წესები:

1. სტატია უნდა წარმოადგინოთ 2 ცალად, რუსულ ან ინგლისურ ენებზე,დაბეჭდილი სტანდარტული ფურცლის 1 გვერდზე, 3 სმ სიგანის მარცხენა ველისა და სტრიქონებს შორის 1,5 ინტერვალის დაცვით. გამოყენებული კომპიუტერული შრიფტი რუსულ და ინგლისურენოვან ტექსტებში - **Times New Roman (Кириллица)**, ხოლო ქართულენოვან ტექსტში საჭიროა გამოვიყენოთ **AcadNusx.** შრიფტის ზომა – 12. სტატიას თან უნდა ახლდეს CD სტატიით.

2. სტატიის მოცულობა არ უნდა შეადგენდეს 10 გვერდზე ნაკლებს და 20 გვერდზე მეტს ლიტერატურის სიის და რეზიუმეების (ინგლისურ, რუსულ და ქართულ ენებზე) ჩათვლით.

3. სტატიაში საჭიროა გაშუქდეს: საკითხის აქტუალობა; კვლევის მიზანი; საკვლევი მასალა და გამოყენებული მეთოდები; მიღებული შედეგები და მათი განსჯა. ექსპერიმენტული ხასიათის სტატიების წარმოდგენისას ავტორებმა უნდა მიუთითონ საექსპერიმენტო ცხოველების სახეობა და რაოდენობა; გაუტკივარებისა და დაძინების მეთოდები (მწვავე ცდების პირობებში).

4. სტატიას თან უნდა ახლდეს რეზიუმე ინგლისურ, რუსულ და ქართულ ენებზე არანაკლებ ნახევარი გვერდის მოცულობისა (სათაურის, ავტორების, დაწესებულების მითითებით და უნდა შეიცავდეს შემდეგ განყოფილებებს: მიზანი, მასალა და მეთოდები, შედეგები და დასკვნები; ტექსტუალური ნაწილი არ უნდა იყოს 15 სტრიქონზე ნაკლები) და საკვანძო სიტყვების ჩამონათვალი (key words).

5. ცხრილები საჭიროა წარმოადგინოთ ნაბეჭდი სახით. ყველა ციფრული, შემაჯამებელი და პროცენტული მონაცემები უნდა შეესაბამებოდეს ტექსტში მოყვანილს.

6. ფოტოსურათები უნდა იყოს კონტრასტული; სურათები, ნახაზები, დიაგრამები - დასათაურებული, დანომრილი და სათანადო ადგილას ჩასმული. რენტგენოგრამების ფოტოასლები წარმოადგინეთ პოზიტიური გამოსახულებით **tiff** ფორმატში. მიკროფოტოსურათების წარწერებში საჭიროა მიუთითოთ ოკულარის ან ობიექტივის საშუალებით გადიდების ხარისხი, ანათალების შეღებვის ან იმპრეგნაციის მეთოდი და აღნიშნოთ სურათის ზედა და ქვედა ნაწილები.

7. სამამულო ავტორების გვარები სტატიაში აღინიშნება ინიციალების თანდართვით, უცხოურისა – უცხოური ტრანსკრიპციით.

8. სტატიას თან უნდა ახლდეს ავტორის მიერ გამოყენებული სამამულო და უცხოური შრომების ბიბლიოგრაფიული სია (ბოლო 5-8 წლის სიღრმით). ანბანური წყობით წარმოდგენილ ბიბლიოგრაფიულ სიაში მიუთითეთ ჯერ სამამულო, შემდეგ უცხოელი ავტორები (გვარი, ინიციალები, სტატიის სათაური, ჟურნალის დასახელება, გამოცემის ადგილი, წელი, ჟურნალის №, პირველი და ბოლო გვერდები). მონოგრაფიის შემთხვევაში მიუთითეთ გამოცემის წელი, ადგილი და გვერდების საერთო რაოდენობა. ტექსტში კვადრატულ ფჩხილებში უნდა მიუთითოთ ავტორის შესაბამისი N ლიტერატურის სიის მიხედვით. მიზანშეწონილია, რომ ციტირებული წყაროების უმეტესი ნაწილი იყოს 5-6 წლის სიღრმის.

9. სტატიას თან უნდა ახლდეს: ა) დაწესებულების ან სამეცნიერო ხელმძღვანელის წარდგინება, დამოწმებული ხელმოწერითა და ბეჭდით; ბ) დარგის სპეციალისტის დამოწმებული რეცენზია, რომელშიც მითითებული იქნება საკითხის აქტუალობა, მასალის საკმაობა, მეთოდის სანდოობა, შედეგების სამეცნიერო-პრაქტიკული მნიშვნელობა.

10. სტატიის ბოლოს საჭიროა ყველა ავტორის ხელმოწერა, რომელთა რაოდენობა არ უნდა აღემატებოდეს 5-ს.

11. რედაქცია იტოვებს უფლებას შეასწოროს სტატია. ტექსტზე მუშაობა და შეჯერება ხდება საავტორო ორიგინალის მიხედვით.

12. დაუშვებელია რედაქციაში ისეთი სტატიის წარდგენა, რომელიც დასაბეჭდად წარდგენილი იყო სხვა რედაქციაში ან გამოქვეყნებული იყო სხვა გამოცემებში.

აღნიშნული წესების დარღვევის შემთხვევაში სტატიები არ განიხილება.

Содержание:

ცენტრი) რეანიმაციულ განყოფილებებში ჩატარდა კვლევა შემთხვევა-კონტროლის მეთოდით. შესწავლილია *K. pneumoniae*-ს გამოყოფილი კულტურების ანტიბიოტიკებისადმი მგრძნობელობა დისკ-დიფუზიის მეთოდისა და E-ტესტის საშუალებით. პირველად საქართველოში გამოკვლეულია CR *K. pneumoniae*-ს კულტურები პოლიმერაზული ჯაჭვური რეაქციით KPC, VIM, IMP, NDM და OXA კარბაპენემაზების შემცველობაზე.

იზოლირებული იყო *K. pneumoniae*-ს 46 შტამი, მათ შორის 20 (43.5%) იყო კარბაპენემ-რეზისტენტული. ყველაზე ხშირად CR *K. pneumoniae*-ს შტამები გამოიყოფოდა ვენტილატორულ-ასოცირებული პნევმონიით დაავადებული პაციენტებისაგან. CR *K. pneumoniae*-ს ძირითად რისკ-ფაქტორებს წარმოადგენენ ფილტვების ხელოვნური ვენტილაცია (OR 30.4, 95% CI 3.504-263.752, p=0.0003), ნაზოგასტრალური ზონდი (OR 17.0, 95% CI 3.202-90.257, p=0.0002), ცენტრალური ვენური კათეტერი (OR 10.06, 95% CI 1.152-87.849, p=0.028) და ანტიბიოტიკების მიღება (OR 10.059, 95% CI 1.152-87.849, p=0.028), განსაკუთრებით კარბაპენემის და მესამე თაობის ცეფალოსპორინის. CR *K. pneumoniae*-ს იზოლატებს შორის ყველაზე გავრცელებულია OXA-მაპროდუცირებელი, შედარებით იშვიათია NDM-მაპროდუცირებელი.

CR *K. pneumoniae*-ს გავრცელების კონტროლისთვის აუცილებელია სამედიცინო დაწესებულებებში ინფექციების კონტროლის გაძლიერება და პაციენტების სკრინინგი CR *K. pneumoniae*-ს ადრეულ გამოსავლენად.

INTERACTIONS OF RANKL, OSTEOPROTEGERIN AND ADIPOKINES IN REGULATION OF BONE REMODELING IN EXPERIMENTAL CHRONIC KIDNEY FUNCTION DISORDER

[1]Goncharova A., [2]Pavlov S., [2]Kumetchko M., [3]Berezniakova M., [3]Yeriomenko R.

[1]Kharkov National Medical University; [2]Kharkov Medical Academy of Postgraduate Education; [3]National University of Pharmacy, Ukraine

Bone and kidneys have close physiological relations. So, if initial pathological changes are in one of them, other is being involved in the pathogenesis. Kidney impairment causes bone remodeling disorders and osteoporosis development.

RANK-RANKL-osteoprotegerin (OPG) axis is considered as a key direction in the cytokine network of regulatory signaling pathways of bone remodeling. Receptor activator of nuclear factor-kB ligand (RANKL) enhances osteoclast resorptive activity in bone. It is expressed in osteoblasts, stromal cells, T-lymphocytes and other cells. OPG is RANKL receptor trap and its function in bone remodeling is to inhibit RANKL activity by blocking its binding with RANK thus preventing osteoclast differentiation [26]. RANKL increase usually occurs in bone loss [33].

RANKL and OPG are the regulators not only of bone turnover, but also of bone-renal and bone-immune cross talks [31, 32], vascular calcification and even development of metastases in cancer [14, 15, 24, 34]. There is growing evidence that these molecular markers are suggested to be the triggers in cytokine networks, which perform interrelations between bone, kidneys and vasculature [23, 37]. It is possible that in certain circumstances they are able to switch the systems mentioned above into the mode which can result in the development of pathology.

Increased RANKL level in blood and its expression in cells have been revealed in kidney disorders [36]. These three systems – bone, kidney and vasculature – interact with each other via the regulatory triade RANK-RANKL-OPG axis and the character of RANKL participation is inflammatory. RANKL is the TNF family member and its activity has the same direction as TNFα, IL-1α and other proinflammatory cytokines [4, 7]. The role of inflammation in the bone-vascular axis was shown in chronic kidney disease (CKD) [30]. It is one of the risk factors of bone disorders and osteoporosis development and is closely connected with acute kidney injury and renal insufficiency in its pathogenesis. These are two interconnected syndromes, which mechanisms of transition are under consideration [9, 11, 22]. Experimental acute kidney injury initiates CKD in rats [1].

Studying of the role of RANKL and OPG and their relationship in regulation of different parameters of homeostasis is of great importance for understanding of their biological role and for new possibilities of correction of many pathological states, especially such as CKD and acute kidney injury.

Besides this, RANKL is a link of a signaling pathway, which is connected with adipokines. Adipokines are also involved in the bone remodeling regulation. Now their role is studied in connection with metabolic syndrome, obesity and diabetes mellitus, which also are etiologic factors of osteoporosis development. Adiponectin is the adipokine, which provides insulin sensitivity. The data on the nature of its influence on bone remodeling are controversial, insulin and insulin growth-factor receptors are implicated in the activation of Akt and extracellular-signal-regulated kinases which may be the mechanism of RANKL function [35]. Visfatin has insulin-mimetic properties, correlating with markers of systemic inflammation and is connected with vascular damage [25].

The major goal of this study was to investigate the role of RANKL, OPG and adipokines adiponectin and visfatin and their relationships in experimental chronic kidney function disorder.

Material and methods. Two groups of 9-month-old female rats (control (20 animals) and experimental (20 animals)) weighing 210 ± 30 g were used in this study. The maintenance, use, and treatment of all animals in this study were in accordance with the principles of the European Convention on the Protection of Vertebrate Animals (Strasbourg, 1986). The animals were given standard laboratory feed and water and kept in a room with the temperature of 22 ± 4 °C and the light on from 7:00 to 19:00.

It was created an acute kidney injury with a single intramuscular 50% glycerol injection in dose of 1.0 ml/100 g weight of the animal as described [1]. In this experimental model chronic impairment of renal excretory function develops in 8 weeks after glycerol injection.

The control group consisted of intact animals. Blood samples were collected through heart puncture of the narcotized with

chloroform rats before being sacrificed with excess amounts of anesthetic after 12 weeks on glycerol injection.

Chronic kidney function disorder was verified morphologically by hematoxylin–eosin and Van Hizon staining. The impairment of bone remodeling was controlled by direct measurements of the bone density which was calculated as a ratio between the bone mass (g) and the bone volume (cm^3). The bone volume was measured by the liquid replaced [2].

Serum RANKL was measured using an ampli-sRANKL ELISA (Biomedica (Austria)). Serum osteoprotegerin was measured using an osteoprotegerin ELISA (eBioscience (Austria)). Serum adiponectin was measured using an adiponectin ELISA (Bio-Vendor (Brno, Czech Republic)), and serum visfatin was measured using a visfatin ELISA (RayBio set (the USA)).

All the experimental data are presented as the mean±SEM. The statistical analyses were performed by Statistica 6.0 programs. The significance was considered at $p<0.05$.

Results and their discussion. Though CKD is a result of many underlying risk factors its common features include renal interstitial fibrosis, tubular epithelial cell loss through apoptosis, glomerular damage, and renal inflammation [21]. The basis of acute kidney injury are dystrophic and necrotic processes in the proximal and distal tubules epithelium, and the transition to the stage of chronic kidney disease is characterized by the formation of interstitial nephritis and nephrosclerosis [1]. CKD is characterized by reduction in glomerular filtration rate (GFR), albuminuria, and structural or functional abnormalities of the kidney [17]. In glycerol injection kidney injury model such structural and functional abnormalities of the kidney (reduction in GFR, blood and urine creatinin increase) were described [1]. Administration of glycerol results at first in dystrophic changes in tubules and glomerular hypertrophy and reduction of the glomerular sizes, tubulointerstitial fibrosis and glomerulosclerosis at a stage of chronic kidney disease which develops in 8 weeks after glycerol injection as it was said before.

Lesion of bone remodeling was demonstrated in rats with chronic kidney disorder. We observed decreasing of hip bone density (1,43±0,07 g / cm^3 in rats at 12 weeks after 50 % glycerol injection, comparing with rats of control group with 1,62±0.04 g / cm^3 ($p<0,05$)).

Kidney impairment results in changes of cytokines profile in experimental animals which are expressed both in cytokines levels increase and the changes of the correlations between them and the directions of their effects.

Chronic kidney function disorder in rats increased RANKL, OPG and adipokines levels comparing with control group: RANKL level was increased on 38,1 %, OPG – 31 %, adiponectin – on 15 % and visfatin in 1.98 times (Table).

Studying of dependence of these cytokines revealed existence of the weak positive correlation of RANKL with adiponectin levels (r=0.30), weak positive correlation of RANKL with visfatin levels (r=0.36) in the intact animals of the control group. This fact demonstrates that the effects of these cytokines are unidirectional. It was no correlation of OPG with RANKL and both adipokines in the intact animals.

Medium strength correlation of RANKL level with adiponectin one and change of its direction (r=-0.63), slight increase of correlation strength (r=0.48) in RANKL with visfatin level was found in rats with chronic kidney function disorder. Also, it was revealed appearance of medium strength positive correlation of RANKL with OPG level (r=0.59) and medium strength negative correlation (r=-0.79) of adiponectin with visfatin level in rats with chronic kidney function disorder model.

These changes can reflect engagement of certain links of adaptation mechanisms in the functional system of maintaining balance of bone remodeling.

Mechanism balancing bone formation and resorption has both sides regulating levers. RANKL is a stimulator of the latter one and the opposite side is presented not by a certain cytokine but the totality of them. Their net effect on the bone formation should be expressed in changes of osteoblast activity. RANKL regulates the process that underlies the control loop and can be enhanced or reduced by the work of other regulatory links probably connected by feedback mechanisms with each other. They are OPG and adipokines. OPG represents an independent direction unlike the system of adipokines.

Under physiological conditions OPG contribute compensatory effects into bone turnover, opposite to RANKL effects. It also affects the normal state of the vascular wall by inhibiting vascular calcification and modulating inflammation. In situation of pathology especially CKD it takes part in functional adaptations leading to the progression of disease. Circulating OPG in humans is associated with the presence of CKD. It was increased in such patients and has the potential of inducing kidney damage. Here the inflammation may be the underlying mechanism in OPG increase in CKD as OPG production is under the regulation by proinflammatory cytokines [6, 18].

These facts are consistent with our results showing increase of OPG in CKD rats. It may be interpreted as compensatory OPG effect to damaging of vessels by inflammation if latter is primary process in the sequence of regulatory events (this issue is not yet clear) and to vascular calcification.

Also, in chronic kidney disorder increase of OPG level may be interpreted as compensatory effect of RANKL increase because OPG is a decoy receptor for RANKL and prevents bone resorption strengthening. In control group resorption processes are balanced with bone formation, so this mechanism of homeostasis restoring is not necessary.

Thus, the modeling of chronic kidney disorder causes inflammatory processes in the kidneys, which in turn can activate intracellular integrating mechanisms which are the triggers of the majority of the signaling pathways regulating many metabolic

Table. Levels of cytokines in RANKL-RANK-osteoprotegerin signaling pathway and adipokines in rats with the chronic kidney function disorder model

Cytokines	Group	
	Control	Chronic kidney function disorder
RANKL, pmol/L	0.131±0.033	0.184±0.071*
Osteoprotegerin, pg/mL	21.588±0.763	28.338±1.223*
Adiponectin, mkg/mL	0.663±0.008	0.767±0.013*
Visfatin, ng/mL	141.606±8.69	279.935±10.153*

** - statistically different ($p<0,05$) compared to control group*

processes. In particular, we can talk about the NF-kB signaling pathway activated by RANKL, which is involved in the processes of inflammation and bone resorption. Fact of revealed appearance of positive correlation of RANKL with OPG level can reflect turning on of certain may be additional or indirect regulating pathways where these cytokines are in the central controlling positions.

Complex interrelations of bone and vascular system don't allow to define what is the cause and what is the consequence: whether RANKL and OPG are independent of vascular calcification processes or they are the reason of these processes and what is the role of inflammation. RANKL can directly stimulate vascular calcification and OPG inhibits it [20]. RANK- RANKL – OPG axis can regulate bone turnover, inflammatory processes and vascular calcification in CKD by activation of simultaneous work of certain signaling pathways and this mechanism is staying unclear yet.

The nature of the established relationship between the RANKL and adipokines in chronic kidney disorder model depends, probably, on various reasons. In this case, inflammatory reactions initiated by renal insufficiency became the cause of the activation of bone resorptive processes. The signaling NF-kB pathway is the integrating mechanism connecting inflammatory and bone resorptive processes. Inflammatory reactions possibly deepen the background physiological signaling activity of the NF-kB. Thus, inflammatory osteolysis is induced [3]. In their turn, osteoblasts increase the RANKL production under the positive feedback principle. That is proved by the results obtained. Apparently, the increased level of visfatin is connected with inflammatory reactions. As described above, the inflammatory agents increase the visfatin production by the macrophages [8].

Direct physiological correlation between adiponectin and RANKL levels discovered in the intact rats changes its direction under the conditions of inflammation.

There are the cross-links between adiponectin and RANKL / RANK signaling pathways in the osteoclasts. It is well known that adiponectin inhibits RANKL-induced osteoclastogenesis and suppresses enhanced by RANKL expression of osteoclast regulators including NFAT2, TRAF6, cathepsin K and tartrate resistant acid phosphatase, and increases osteoclasts differentiation and activity. Adiponectin inhibitory effect on osteoclasts is induced by APPL1- mediated suppression of RANKL-dependent Akt 1 activity. APPL1 is the key adapter protein that interacts with adiponectin receptors and mediates in adiponectin signaling and coordination of various signaling pathways. Akt 1 is a protein kinase. Its increased activity supports osteoclast survival and differentiation. A decrease in Akt 1 activity is the main cause of adiponectin mediated inhibition of osteoclast formation [28].

According to [19], adiponectin may normally provide a regenerating effect on the bone. It has a positive modulatory effect on osteoblasts, inhibits differentiation of macrophage precursors of osteoclasts and suppresses the resorptive activity of osteoclasts.

Thus, adiponectin normally has the multidirectional effects on the bone metabolism. At the same time the realization of these effects takes place at the different levels: from the cellular to the system-level. Adiponectin is able to regulate the same function in two opposite ways depending on the site of its action. It has partially opposite to leptin effect on the sympathetic nervous system, that is adiponectin antagonizes leptin regulation of the sympathetic tone. Adiponectin decreases the sympathetic tone, thereby increasing bone mass and decreasing the energy expenditure. It takes part in signaling in neurons of the locus coeruleus through FoxO1. Locally adiponectin signals to the osteoblasts, thereby preventing their proliferation and promotes their apoptosis, thus reducing bone mass and circulating osteocalcin level. Adiponectin performs this function regardless of known receptors and signaling pathways due to decreasing of FoxO1 activity in the PI3 kinase - FoxO1 way [13].

Most likely, RANKL may realize its effect partially regardless of adiponectin and to some extent they act cooperatively, as RANKL is produced by osteoblasts, and adiponectin may downregulate RANKL because its place is above the RANKL in the signaling cascade.

In the intact animals the relationship between adiponectin and RANKL were unidirectional, that is adiponectin probably stimulated the expression of RANKL in osteoblasts, but the process was balanced with the stimulation of osteoblast proliferation and differentiation. In the chronic kidney function disorder model opposite effects of these cytokines may indicate the predominance of another adiponectin effect on osteoblasts, when it decreases FoxO1 activity in PI3 kinase - FoxO1 signaling pathway. Thus, osteoblast proliferation is inhibited and apoptosis is activated.

The impairments caused by kidney disease can change the fate of specific signaling pathways that are involved in the regulation of the bone cells activity. We can suggest that the mode of the RANKL-adiponectin bone regulation mechanisms functioning is changed in chronic kidney function disorder model. Adiponectin can have both direct and indirect effects on bone remodeling regulation in this pathology. But possibly adiponectin has rather opposite to RANKL effect here.

In chronic kidney function disorder model the appearance of a negative correlation between the levels of adiponectin and visfatin and change of correlation direction between RANKL level and adiponectin one can evidence visfatin effect has the same direction as RANKL. Visfatin is involved in regulatory processes of tissue remodeling through the development of inflammatory response likely via the mechanisms of regulation of central transcription factor – NF-kB [16]. It can directly impair vascular reactivity and is connected with endothelial dysfunction [27, 29]. Increased visfatin level is associated with GFR decline rate and future progression of kidney disease in nondiabetic hypertensive patients with preserved renal function [10]. Thus, it is possible to suggest that visfatin in conditions of chronic kidney disorder may have rather damaging effect as on bone remodeling so on the endothelium. Although some data state that it inhibits the early stages of osteoclastogenesis by downregulating RANKL-dependent early signaling pathways JNK, Akt and its downstream target GSK3β, Btk and PLCγ2 [5]. But the data are not connected with visfatin role in CKD.

So, inflammatory responses in the chronic kidney disorder model induce the increase in resorptive activity which is stimulated by an increased RANKL level. Also, the increase in adiponectin and visfatin takes place. The character of the relationship between adiponectin level and osteoclasts activity changes. Osteoprotegerin and RANKL levels, as well as adiponectin and visfatin levels become bounded together in this disorder.

Thus, chronic kidney disorder model causes some changes in regulatory cytokines interrelationships. RANKL and visfatin acts unidirectional, more likely as damaging factors because of their inflammatory character. OPG has the same direction of action and may also impair vascular wall state but compensate the RANKL effect of bone resorption. Adiponectin in chronic kidney disorder model has complex role opposite to the abovementioned cytokines, that is in accordance with literature [12].

Conclusion. Chronic kidney disorder model results in the in-

crease in RANKL / RANK / osteoprotegerin signaling pathway cytokines and adipokines and in certain changes of their relationships and effects direction. Regulatory mechanisms of bone remodeling and vascular wall state are disturbed by proinflammatory directed effects of RANKL and visfatin, controversial effects of osteoprotegerin and adiponectin bone preserving effects.

REFERENCES

1. Кондаков І.І., Топчій І.І., Кірієнко О.М. (2013) Вплив гліцеролу на функціонально-морфологічні показники нирок при моделюванні гострої та хронічної ниркової недостатності у щурів // Український журнал нефрології та діалізу. № 3 (39). 14–20.
2. Подковкин В.Г., Иванов Д.Г., Иванов Г.А. (2008) Влияние постоянного магнитного поля на состояние костной ткани крыс с повышенным уровнем резорбции // Успехи современного естествознания. № 7. 13–16.
3. Abu-Amer Y (2013). NF-kappaB signaling and bone resorption // Osteoporos Int 24 (9) 2377–2386.
4. Amarasekara DS, Yun H, Kim S, Lee N, Kim H, Rho J. (2018) Regulation of Osteoclast Differentiation by Cytokine Networks // Immune Netw. 7;18 (1):e8.
5. Baek J. M., Ahn S.-J., Cheon Y.-H., Lee M. S., Oh J., Kim J.-Y. (2017). Nicotinamide phosphoribosyltransferase inhibits receptor activator of nuclear factor-κB ligand-induced osteoclast differentiation in vitro // Molecular Medicine Reports, 15(2), 784–792.
6. Bernardi S., Toffoli B., Bossi F., Candido R., Stenner E., Carretta R., Barbone F., Fabris B. (2017). Circulating osteoprotegerin is associated with chronic kidney disease in hypertensive patients //BMC Nephrology, 18, 219.
7. Cafiero C, Gigante M, Brunetti G, Simone S, Chaoul N, Oranger A, Ranieri E, Colucci S, Pertosa GB, Grano M, Gesualdo L. (2018) Inflammation induces osteoclast differentiation from peripheral mononuclear cells in chronic kidney disease patients: crosstalk between the immune and bone systems // Nephrol Dial Transplant. 1; 33 (1), 65-75.
8. Dahl TB, Yndestad A, Skjelland M, **Øie E**, Dahl A, Michelsen A, Damås JK, Tunheim SH, Ueland T, Smith C, et al. (2007). Increased expression of visfatin in macrophages of human unstable carotid and coronary atherosclerosis: possible role in inflammation and plaque destabilization // Circulation 115 (8):972-80.
9. Fiorentino M, Grandaliano G, Gesualdo L, Castellano G. (2018) Acute Kidney Injury to Chronic Kidney Disease Transition. Contrib Nephrol.193:45-54.
10. Hsu C.-Y., Huang P.-H., Chen T.-H., Chiang C.-H., Leu H.-B., Huang C.-C., Chen J.-W., Lin S.-J. (2016). Increased Circulating Visfatin Is Associated With Progression of Kidney Disease in Non-Diabetic Hypertensive Patients // American Journal of Hypertension, 29(4), 528–536.
11. Hsu RK, Hsu CY. (2016) The Role of Acute Kidney Injury in Chronic Kidney Disease // Semin Nephrol. 36(4): 283-92.
12. Jia T, Carrero JJ, Lindholm B, Stenvinkel P. (2012) The complex role of adiponectin in chronic kidney disease // Biochimie. 94(10):2150-6.
13. Kajimura D, Lee HW, Arteaga-Solis E, Ferron M, Zhou B, Clarke CJ, Hannun YA, DePinho RA, Guo XE, et al. (2013). Adiponectin regulates bone mass via opposite central and peripheral mechanisms through FoxO1 // Cell Metab 17 (6): 901-15.
14. Kawakami R, Nakagami H, Noma T, Ohmori K, Kohno M, Morishita R. (2016) RANKL system in vascular and valve calcification with aging // Inflamm Regen. 1;36:10.
15. Kurabayashi M. (2016) Interaction between bone and artery // Clin Calcium. Aug;26(8):1119-26. [Article in Japanese].
16. Laiguillon MC, Houard X, Bougault C, Gosset M, Nourissat G, Sautet A, Jacques C, Berenbaum F, Sellam J. (2014) Expression and function of visfatin (Nampt), an adipokine-enzyme involved in inflammatory pathways of osteoarthritis // Arthritis Res Ther. 31;16 (1):R38.
17. Levey AS, Eckardt K-U, Tsukamoto Y et al (2005). Definition and classification of chronic kidney disease: a position statement from kidney disease: improving global outcomes (KDIGO). Kidney International 67 (6): 2089-2100.
18. Lewis JR, Lim WH, Zhu K, Wong G, Dhaliwal SS, Lim EM, Ueland T, Bollerslev J, Prince RL. Elevated osteoprotegerin predicts declining renal function in elderly women: a 10-year prospective cohort study // Am J Nephrol. 2014;39(1):66-74.
19. Liu Y, Song CY, Wu SS, Liang QH, Yuan LQ, Liao EY (2013). Novel adipokines and bone metabolism // Int J Endocrinol 2013: 895045.
20. Lu, K.-C., Wu, C.-C., Yen, J.-F., & Liu, W.-C. (2014). Vascular Calcification and Renal Bone Disorders // The Scientific World Journal, 2014, 637065.
21. Mohammed-Ali Z, Cruz GL, Lu Ch, et al (2015). Development of a model of chronic kidney disease in the C57BL/6 mouse with properties of progressive human CKD // BioMed Research International vol 2015, Article ID 172302, 10 pages, doi:10.1155/2015/172302.
22. Nangaku M, Hirakawa Y, Mimura I, Inagi R, Tanaka T. (2017) Epigenetic Changes in the Acute Kidney Injury-to-Chronic Kidney Disease Transition // Nephron. 137 (4):256-259.
23. Peres LA, Pércio PP. (2014) Mineral and bone disorder and vascular calcification in patients with chronic kidney disease //J Bras Nefrol. Apr-Jun; 36(2):201-7.
24. Renema, N., Navet, B., Heymann, M.-F., Lezot, F., & Heymann, D. (2016). RANK–RANKL signalling in cancer // Bioscience Reports, 36(4), e00366.
25. Romacho T, Sánchez-Ferrer CF, Peiró C. (2013) Visfatin/Nampt: an adipokine with cardiovascular impact // Mediators Inflamm.; 2013:946427. doi: 10.1155/2013/946427.
26. Sagalovsky S, Schonert M (2011). RANKL-RANK-OPG system and bone remodeling: a new approach on the treatment of osteoporosis // Clin Exptl Pathol 10 (2): 146–153.
27. Sun, L., Chen, S., Gao, H., Ren, L., & Song, G. (2017). Visfatin induces the apoptosis of endothelial progenitor cells via the induction of pro-inflammatory mediators through the NF-κB pathway // International Journal of Molecular Medicine, 40(3), 637–646.
28. Tu Q, Zhang J, Dong LQ, Saunders E, Luo E, Tang J, Chen J (2011). Adiponectin inhibits osteoclastogenesis and bone resorption via APPL1-mediated suppression of Akt1// J Biol Chem; 286 (14): 12542–12553.
29. Vallejo, S., Romacho, T., Angulo, J., Villalobos, L. A., Cercas, E., Leivas, A., Bermejo, E., Carraro, R., Sánchez-Ferrer C. F., Peiró, C. (2011). Visfatin Impairs Endothelium-Dependent Relaxation in Rat and Human Mesenteric Microvessels through Nicotinamide Phosphoribosyltransferase Activity // PLoS ONE, 6(11), e27299.
30. Viaene L, Behets GJ, Heye S, Claes K, Monbaliu D, Pirenne J, D'Haese PC,Evenepoel P. (2016) Inflammation and the bone-vascular axis in end-stage renal disease // Osteoporos Int. 27(2):489-97.
31. Walsh M. C., Choi Y. (2014). Biology of the RANKL–RANK–OPG System in Immunity, Bone, and Beyond // Frontiers in Immunology, 5, p. 511.
32. Weitzmann M. N., Ofotokun I. (2016). Physiological and pathophysiological bone turnover — role of the immune system. Nature Reviews. Endocrinology, 12(9), 518–532.
33. Xiong J., Piemontese M., Thostenson J. D., Weinstein R. S., Manolagas S. C., O'Brien C. A. (2014). Osteocyte-derived RANKL is a critical mediator of the increased bone resorption caused by dietary calcium deficiency //Bone, 66, 146–154.

34. Yamamoto M. (2015) Vascular Calcification - Pathological Mechanism and Clinical Application - . Vascular calcification as a clinical manifestation of bone-vascular axis //Clin Calcium. May;25(5):655-60. [Article in Japanese]
35. Yuan LQ, Zhu JH, Wang HW, Liang QH, Xie H, Wu XP, Zhou H, Cui RR, Sheng ZF, Zhou HD, Zhu X, Liu GY, Liu YS, Liao EY. (2011) RANKL is a downstream mediator for insulin-induced osteoblastic differentiation of vascular smooth muscle cells // PLoS One. 6 (12):e29037.
36. Zhou YX, Shi LX, Yang H, Long YG, Meng LU, Lv LS, Zhang Y, Yao H, Li L, Yu YN. (2016) Effects of a GSK-3β inhibitor on the renal expression levels of RANK, RANKL and NF-κB in a rat model of diabetic nephropathy // Exp Ther Med. 11 (6):2495-2502.
37. Znorko B, Oksztulska-Kolaneka E, Michałowska M, Kamiński T, Pawla K (2017) Does the OPG/RANKL system contribute to the bone-vascular axis in chronic kidney disease? // Adv. Med. Sci. 62 (1), 52-64.

SUMMARY

INTERACTIONS OF RANKL, OSTEOPROTEGERIN AND ADIPOKINES IN REGULATION OF BONE REMODELING IN EXPERIMENTAL CHRONIC KIDNEY FUNCTION DISORDER

[1]Goncharova A., [2]Pavlov S., [2]Kumetchko M., [3]Berezniakova M., [3]Yeriomenko R.

[1]Kharkov National Medical University; [2]Kharkov Medical Academy of Postgraduate Education; [3]National University of Pharmacy, Ukraine

RANKL and osteoprotegerin are the key cytokines of the axis regulating not only bone remodeling but also bone-kidney interrelations. They are connected with adipokines in the regulatory actions. The aim of the article was to investigate the role of the main parts of RANK-RANKL-osteoprotegerin axis and cytokines of additional link of regulatory network – adiponectin and visfatin and the interrelationship of these cytokines in chronic kidney disorder model in rats. Chronic kidney disorder model was created in the rats 12 weeks after intramuscular 50 % glycerol injection-induced acute kidney injury. RANKL, osteoprotegerin, adiponectin and visfatin levels was measured using ELISA. Increase of all cytokines and changes of their interrelations has been revealed in rats with chronic kidney disorder model. Conclusion was made that effects of the cytokine axis where RANKL is a trigger in the regulation network of interconnections «bone – kidney» has inflammatory character, so as visfatin effect, and adiponectin has opposite preserving bone effect.

Keywords: bone remodeling, chronic kidney function disorder, cytokines, adipokines.

РЕЗЮМЕ

ВЗАИМОДЕЙСТВИЯ RANKL, ОСТЕОПРОТЕГЕРИНА И АДИПОКИНОВ В РЕГУЛЯЦИИ КОСТНОГО РЕМОДЕЛИРОВАНИЯ ПРИ ЭКСПЕРИМЕНТАЛЬНОМ ХРОНИЧЕСКОМ НАРУШЕНИИ ФУНКЦИИ ПОЧЕК

[1]Гончарова А.В., [2]Павлов С.Б., [2]Кумечко М.В., [3]Березнякова М.Е., [3]Еременко Р.Ф.

[1]Харьковский национальный медицинский университет; [2]Харьковская медицинская академия последипломного образования; [3]Национальный фармацевтический университет, Украина

RANKL и остеопротегерин являются ключевыми цитокинами оси, регулирующей не только ремоделирование костной ткани, но также и взаимосвязи костной ткани с почками. В своих регуляторных действиях они связаны с адипокинами.

Целью данного исследования явилось определить роль основных участников оси RANK-RANKL-остеопротегерин и цитокинов дополнительного звена регуляторной сети - адипонектина и висфатина и взаимосвязь этих цитокинов на модели хронического нарушения функции почек у крыс. Модель хронического нарушения функции почек у крыс создана спустя 12 недель после острой почечной недостаточности, вызванной внутримышечной инъекцией 50% глицерина. Уровни RANKL, остеопротегерина, адипонектина и висфатина измеряли методом ИФА. Увеличение количества цитокинов и изменения в их взаимосвязи выявлены у крыс с моделью хронического нарушения функции почек. Сделан вывод о том, что эффекты оси цитокинов, где RANKL является триггером, в сети регуляции межклеточных связей «костная ткань - почки», имеют воспалительный характер, так же, как и влияние висфатина, а адипонектин оказывает противоположный сохраняющий костную ткань эффект.

რეზიუმე

RANKL, ოსტეოპროტეგერინისა და ადიპოკინის ურთიერთქმედება ძვლის რემოდელინგის რეგულაციაში თირკმლის ფუნქციის ქრონიკული უკმარისობის დროს ექსპერიმენტში.

ა. გონჩაროვა, ს. პავლოვი, მ. კუმეჩკო, მ. ბერეზნიაკოვა, რ. ერემენკო

ხარკოვის ეროვნული სამედიცინო უნივერსიტეტი, ხარკოვის დიპლომისშემდგომო განათლების სამედიცინო აკადემია, ერობნული ფარმაცევტული უნივერსიტეტი, უკრაინა.

ოსტეოპროტეგერინი და RANKL წარმოადგენენ საკვანძო ციტოკინებს იმ ღერძისა, რომელიც არეგულირებს ძვლოვანი ქსოვილის არამხოლოდ რემოდელირებას, არამედ მის ურთიერთქმედებას თირკ-

მლებთან. ამ აქტივობაში ისინი უკავშირდებიან ადიპოკინებს.

ნაშრომის მიზანია განისაზღვროს RANKL-RANKL-ის ოსტეოპროტეგერინის ღერძის ძირითადი რგოლებისა, და ციტოკინების დამატებითი სარეგულაციო ქსელის კომპონენტების - ადიპონექტინისა და ვისფატინის როლი და ამ ციტოკინების ურთიერთქმედება ვირთაგვის თირკმლის ფუნქციის ქრონოკულტი უკმარისობის მოდელზე. მოდელი განხორციელდა მწვავე უკმარისობიდან მე-12 კვირას, რაც გამოწვეული იყო გლიცერინის 50% ხსნარის ინიექციით კუნთებში. ოსტეოპროტეგერინის, RANKL, ადიპონექტინისა და ვისფატინის დონე განისაზღვრა იმუნოფერმენტული ანალიზით.

დადგენილია, რომ ციტოკინების ღერძის ეფექტი, რომელშიც RANKL ტრიგერს წარმოადგენს, „ძვლოვანი ქსოვილი - თირკმელი" ურთიერთრეგულაციაში ვლინდება ანთებითი პროცესით, ისევე როგორც ვისფატინისა, ხოლო ადიპონექტინი ძვლოვან ქსოვილზე ავლენს საპირისპირო დამცველობით ზემოქმედებას.

SYNTHESIS OF THE NEW 2-(3,4-dihydro-3-oxo-2*H*-[1,2,4]triazino[4,3-*c*]quinazolin-4-yl) ACETIC ACID DERIVATIVES AND ANALYSIS OF THEIR ANTIOXIDANT ACTIVITY IN NITROSATIVE STRESS MODELS

[1]Belenichev I., [3]Gorchakova N., [4]Puzyrenko A., [2]Kovalenko S., [1]Bukhtiayrova N.

Zaporozhye State Medical University, [1]Department of the Pharmacology; [2]Department of the Organic and Bioorganic Chemistry, [3]Bogomolets National Medical University, Department of the Pharmacology, Kiev, Ukraine; [4]All Saints University School of Medicine Department of the Pharmacology, Dominika

Neuroprotection and antioxidant treatment today are a perspective and proved direction because neurodestruction due to activation free radical oxidation can provoke the permanent loss of motor or mental function. Nitric oxide ($NO^{\cdot}$) can play the role of a modulator as active free radical in the central nervous system. In CNS pathology nitric oxide can be a pathogenic factor of changes inside the cells during cerebral ischemia [1,2]. In the acute phase of ischemia $NO^{\cdot}$ plays a neuroprotective role but then especially in reperfusion (recirculation) period $NO^{\cdot}$ effect becomes neurotoxic [3,4]. Especially at that moment in the nerve cells oxidative-nitrosative stress develops where reactive nitrogen species act together with reactive oxygen species. It results in nitration of the metal groups in enzymes and causes their dysfunction. It also causes nucleic acid fragmentation, decreasing NAD^+ and ATP level in the cells, inhibition of mitochondrial enzymes activities [5].

Major aggressive factor during this oxidative-nitrosative stress is peroxynitrite ($ONOO^-$) due to extra production $NO^{\cdot}$. As a strong oxidant peroxynitrite damages different biologically active molecules and cell organelles. Also $ONOO^-$ reacts with superoxide dismutase's active center and produces toxic nitrosonium (NO^+) which nitrosizes phenol groups of amino acids (including tyrosine). These impulses in the cells can start their apoptosis [6,7]. So investigation of the antioxidants which will be able to decrease $NO^{\cdot}$ and $ONOO^-$ toxicity (especially towards superoxide dismutase) seems to be very of current interest.

[1,2,4]triazino[4,3-*c*]quinazoline represents a scantly explored group of biologically active substances. It was established that derivatives of 3,4,6,7-tetrahydro- and 3,4-dihydro-2*H*-[1,2,4] triazino[4,3-*c*]quinazolines could potentiate barbiturate action and also showed antidepressant, anti-inflammatory and analgesic activity.

So we conducted investigation antioxidant activity of the nine new original derivatives of 2-(3,4-dihydro-3-oxo-2*H*-[1,2,4]triazino[4,3-*c*]quinazolin-4-yl)acetic acid in vitro and in vivo by using two nitrosative stress models. We studied protective effects of these acid derivatives on superoxide dismutase (SOD) activity under conditions of excessive $NO^{\cdot}$ and $ONOO^-$ production.

Materials and methods. *In vitro study*

In the first experimental model, the antioxidant activity of compounds was estimated in vitro with $NO^{\cdot}$ induction [8]. $NO^{\cdot}$ induction was performed under the action of light on sodium nitroprusside sample (the light source was 300W, wavelength > 425nm). The light ray was focused on the sample with the help of a lens. To remove the thermal effect, the ray was directed through the water filter. We used a water solution of sodium nitroprusside $Na_2[Fe(NO)(CN)_5]\times 2H_2O$ (1.0 mM) which was radiated in quartz flask lasting 30min. The efficiency of $NO^{\cdot}$ generation (control) and antioxidant activity of the investigated substances were measured by oxidation rate of ascorbic acid (40 mM, wavelength = 265 nm). The investigated substances were added to the samples before radiation at a concentration 10^{-6}M. Antioxidant activity was expressed in percent of inhibition of ascorbic acid oxidation.

In vivo study. In second experimental model, investigation was carried out in the brain supernatant obtained from the white Wistar rats (male, 200-250g, 4.5 months old). The rats were supplied by the Institute of Pharmacology and Toxicology of the Academy of Medical Sciences of Ukraine, Kyiv. All procedures were conducted accordingly with the Directive 2010/63EU of European Parliament and Council regarding the protection of animals used for experimental and other scientific purposes (Zaporozhye State Medical University Ethics Committee permit No. 62 from 05.IV.2017). The rats were assigned to individual housing in stainless steel, wire-bottomed cages. The quarantine period for all animals was fourteen days. The animals were examined every day regarding the general state, health and death rate. Cages with animals were placed in separate rooms. Lighting was maintained at the 12h light and 12h dark cycles. The temperature was within 19-25C°, humidity – 50-70%. The ventilation was set at 15 air volumes per hour and provided a concentration of CO_2 no more than 0.15%, ammonia – no more than 0.001 mg/l. Water (processed by reverse osmosis) and food were available ad libitum from individual bottles and feeders.

In the day of the experiment after reaching the deep anesthesia with sodium pentobarbital (40.0 mg/kg) rat's cranium was