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ЕЖЕМЕСЯЧНЫЙ НАУЧНЫЙ ЖУРНАЛ

Медицинские новости Грузии

საქართველოს სამედიცინო სიახლენი

GEORGIAN MEDICAL NEWS

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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 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 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.

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WEBSITE

www.geomednews.com

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 compu-ter-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.

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Содержание:

Yu-Ri Choi, Su-Bin Yu, Seoul-Hee Nam.
ANTIBACTERIAL EFFECT OF CRATAEGUS PINNATIFIDA EXTRACT AGAINST ENTROCOCCUS FAECALIS A ROOT CANAL DISEASE-CAUSINGBACTERIA……6-10

Larisa Melia, Revaz Sulukhia, Lali Pkhaladze, Nino Davidova, Archil Khomasuridze.
MIFEPRISTON IN OBSTETRICS – WHY NOT?.....11-14

Maryna Stoliarchuk.
CORRELATION BETWEEN TRANSVERSE CEPHALOMETRIC PARAMETERS AND THE SEVERITY OF SKELETAL MALOCCLUSIONS…….15-18

Deepak, Prashant Rao, Archana, Sowmya M, Sandeep. S, Suma S.
A CROSS-SECTIONAL STUDY ON COVID-19 VACCINATION HESITATION AMONG UNIVERSITY STUDENTS……..……19-23

Tchernev G, Broshtilova V, Ivanov L, Alexandrov A, Smilov N, Kordeva S.
DRUG RELATED NITROSOGENESIS, PHOTOCARCINOGENESIS AND ONCOPHARMACOGENESIS OF NODULAR MELANOMA: A CASE RELATED ANALYSIS CONCERNING THE POLYCONTAMINATION OF THE POLYMEDICATION WITH VALSARTAN/ HYDROCHLOROTHIAZIDE AND BISOPROLOL…….24-27

Rawaa J. Matloob, Zeina A. Althanoon, Saad A. Algburi, Mudheher I. Salih, Marwan M. Merkhan.
UPDATE ON THE USE OF METHOTREXATE IN THE MANAGEMENT OF RHEUMATOID ARTHRITIS…….28-33

Georgi Tchernev.
(N-NITROSO) PROPAFENONE INDUCED ADVANCED NODULAR MELANOMA-FIRST REPORTED CASE IN THE WORLD LITERATURE: THE INEXTRICABLE LINKS BETWEEN THE PHOTOCARCINOGENESIS, DRUG RELATED NITROSOGENESIS AND PHARMACO-ONCOGENESIS…….34-37

Elham M. Mahmood, Entedhar R. Sarhat, Maryam T. Tawfeq, Siham A. Wadee.
HISTOLOGICAL AND BIOCHEMICAL STUDY OF THE EFFECT OF FEXOFENADINE ON SALIVARY GLAND IN RATS…….38-40

Valerii Vovk, Igor Duda, Alla Vovk.
THE EFFECT OF A MULTIMODAL APPROACH ON THE RESULTS OF TREATMENT IN SURGERY: INTEGRATION OF CHEMOTHERAPY, SURGERY, AND RADIOTHERAPY…….41-46

Haitham Alhussain, Deepak, Bharath Chandra V, Lakshmi. R, Sumana A, Jishamol KR.
EXAMINATION OF THE INCIDENCE OF POOR SLEEP QUALITY AND FACTORS ASSOCIATED FOR POOR SLEEP DURING THE VARIOUS PHASES OF PREGNANCIES…….47-53

N. Ksajikyan, H. Aghababyan, M. Sargsyan.
ASSESSMENT OF REACTIVITY TO THE BODY UNDER CONDITIONS OF PHYSICAL ACTIVITY IN STUDENTS AGED 17-20 YEARS….54-58

Abinaya Srinivasa Rangan, Dhanush Balaji.S, Utham Chand, Raghunathan E.G, Deepthi.N, Prasanna Karthik.S.
TRIGLYCERIDE – GLUCOSE INDEX, REMNANT CHOLESTEROL AND COMMON CAROTID ARTERY INTIMA-MEDIA THICKNESS AS AN ATHEROSCLEROTIC MARKER IN ISCHEMIC STROKE PATIENTS.....…….59-65

Riyam AH. Al-Barwani, Entedar R. sarhat.
BREAST CANCER-MODULATED OMENTIN AND VASPIN PLASMA LEVELS…….66-69

Tchernev G, Dimova D.
PERIOCULAR HIGH RISK BCCS AFTER ADDITIONAL/PARALLEL INTAKE OF TORASEMIDE, MOXONIDINE AND MIRABEGRON: IMPORTANT LINKS TO SKIN CANCER RELATED (PHOTO-) NITROSOGENESIS IN THE CONTEXT OF PHARMACO-ONCOGENESIS…….70-76

Abinaya Srinivasa Rangan, Dhanush Balaji.S, Saranya.C, Raghunathan E.G, Deepthi.N, Prasanna Karthik.S.
ASSOCIATION OF MPV AND RDW WITH DISEASE ACTIVITY IN PATIENT WITH RHEUMATOID ARTHRITIS…….77-81

Julieta Nino Gulua, Lela Sturua, Maia Khubua, Lela Shengelia.
THYROID CANCER AS A PUBLIC HEALTH CHALLENGE IN GEORGIA…….82-86

Rahma S. Almallah, Hani M. Almukhtar.
MIRABEGRON INDUCED RELAXATION OF ISOLATED BOVINE CORONARY SEGMENTS: ROLE OF NO AND K+ CHANNEL…….87-92

Gogotishvili Mariam, Gogebashvili Nino, Bakradze Mzia, Gorgiladze Tinatin, Japaridze Fridon.
MANIFESTATIONS OF DISEASES OF THE ORAL MUCOSA OF PATIENTS IN THE ADJARA REGION DURING THE COVID-19 PANDEMIC…….93-95

Nithesh Babu R, Fathima S Nilofar, Saranya Palanisamy, Gnanadeepan T, Mahendra Kumar K.
EXPLORING THE INCIDENCE AND PREVALENCE OF NEW-ONSET AUTOIMMUNE DISEASE FOLLOWING COVID-19 PANDEMIC: A SYSTEMATIC REVIEW…….96-103

E. Mosidze, A. Chikovani, M. Giorgobiani.
ADVANCES IN MINIMALLY INVASIVE SURGERY FOR PECTUS EXCAVATUM: ENHANCING OUTCOMES AND PATIENT CARE……..……..104-107

Nithesh Babu R, Fathima S Nilofar, Saranya Palanisamy, Gnanadeepan T, Mahendra Kumar K.
SIGNIFICANCE OF NEUTROPHIL-LYMPHOCYTE RATIO AND PLATELETLYMPHOCYTE RATIO AS PROGNOSTIC MARKERS OF DISEASE SEVERITY IN SYSTEMIC LUPUS ERYTHEMATOSUS…….....…108-112

Athraa E. Ahmed, Nibras H. Hameed.
PREVALENCE OF FETAL CONGENITAL ANOMALIES IN PATIENTS ATTENDING TIKRIT TEACHING HOSPITAL…..……113-116

Kazantcev A.D, Kazantceva E.P, Sarkisyan I.P, Avakova A.E, Shumakova A.O, Dyachenko Y.E, Mezhenko D.V, Kustov Y.O, Makarov Daniil Andreevich, Guliev M.T, Babaeva M.M.
COMPARATIVE ANALYSIS OF POSITIVE AND NEGATIVE EXPECTATIONS WITH CONTROL OF VOLITIONAL EFFORT IN YOUNG AND OLD AGES AS RISK FACTORS OF SOCIAL AGING….....……117-121

Arnab Sain, Sarah Arif, Hoosai Manyar, Nauman Manzoor, Kanishka Wattage, Michele Halasa, Arsany Metry, Jack Song Chia, Emily Prendergast, Ahmed Elkilany, Odiamehi Aisabokhale, Fahad Hussain, Zain Sohail.
CURRENT CONCEPTS IN THE MANAGEMENT OF BOXER'S FRACTURE…….....……122-124

Gonashvili Meri, Kilasonia Besarion, Chikhladze Ramaz, Merabishvili Gela, Beriashvili Rusudan.
MEDICO-LEGAL APPLICATIONS OF FRACTURE HEMATOMA: REVIEW…….....……125-130

Zynab J. Jarjees, Entedhar R. Sarhat.
ASSESSMENT OF OSTEOPONTIN, SCLEROSTIN, AND OSTEOCALCIN LEVELS IN PATIENTS WITH HYPOTHYROIDISM ON MEDICAL THERAPY…….....……131-135

Tchernev G, Dimova D.
EDUCATION FROM DERMATOLOGISTS: THE SIMULTANEOUSLY DEVELOPMENT OF 16 KERATINOCYTIC CANCERS AFTER USE OF METFORMIN IN COMBINATION WITH LOSARTAN/ HYDROCHLOROTHIAZIDE, METOPROLOL AND NIFEDIPINE-IMPORTANT LINKS TO DRUG RELATED (PHOTO)-NITROSO-CARCINOGENESIS AND ONCOPHARMACOGENESIS…….....……136-141

Ismayilov M.U, Polukhov R.Sh, Poddubny I.V, Magammedov V.A.
COMPARATIVE ASSESSMENT OF SURGICAL TREATMENT OF COMPLICATIONS OF ULCERATIVE COLITIS IN CHILDREN…….....……..142-148

Arnab Sain, Arsany Metry, Nauman Manzoor, Kanishka Wattage, Ahmed Elkilany, Michele Halasa, Jack Song Chia, Sarah Arif, Fahad Hussain, Odiamehi Aisabokhale, Zain Sohail.
THE ROLE OF DISTAL LOCKING IN INTRAMEDULLARY NAILS FOR HIP FRACTURE FIXATION: A REVIEW OF CURRENT LITERATURE…….....……149-150

Buba Chachkhiani, Manana Kalandadze, Shalva Parulava, Vladimer Margvelashvili.
EFFECT OF SURFACE ABRASION AND TEMPERATURE TREATMENT ON METASTABLE TETRAGONAL ZIRCONIUM DIOXIDE (EXPERIMENTAL STUDY)…….....……..151-155

Abdulrahman A Abdulhamed, Luma W Khaleel.
CARDIOPROTECTIVE EFFECT OF GLYCYRRHIZA GLABRA EXTRACT AND GLYCYRRHIZA GLABRA SILVER NANOPARTICLE AGAINST ALLOXAN AND NICOTINAMIDE INDUCED DIABETIC CARDIAC INJURY IN RATS……...…156-159

Larysa Pentiuk, Tetiana Niushko, Emiliia Osiadla.
FEATURES OF BLOOD PRESSURE DAILY MONITORING INDICATORS, STRUCTURAL AND FUNCTIONAL CHANGES OF THE LEFT VENTRICLE AND VESSELS IN WOMEN WITH HYPERTENSION II STAGE OF DIFFERENT REPRODUCTIVE AGE AND THEIR RELATIONSHIP WITH SEX HORMONES LEVEL…….....……160-167

Rana dawood Salman Al-kamil, Thamir F. Alkhiat, H. N. K. AL-Saman, H. H. Hussein, Dawood Chaloob Hilyail, Falah Hassan Shari.
THE EFFECT OF NUTRITIONAL GENOMICS ON CARDIOVASCULAR SYSTEM…….....……168-176

Sopiko Kvaratsthelia.
PREVALENCE OF DENTITION, DENTAL ARCHES AND DENTAL ANOMALIES…….....……177-180

Dorosh D, Liadova T, Popov M, Volobuieva O, Pavlikova K, Tsivenko O, Chernuskiy V, Hrek I, Kushnir V, Volobuiev D.
THE EFFECT OF MELATONIN ON THE SERUM LEVEL OF INTERLEUKIN 31 IN HERPESVIRUS SKIN DISEASES ON THE BACKGROUND OF HIV…….....……181-184

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ANTIBACTERIAL EFFECT OF CRATAEGUS PINNATIFIDA EXTRACT AGAINST ENTROCOCCUS FAECALIS A ROOT CANAL DISEASE-CAUSING BACTERIA

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Abstract.

Studies have substantiated the anti-inflammatory and anti-thrombotic effects of (*C. pinnatifida*); however, research on its antibacterial activity using organic solvent remains limited. Therefore, in this study, we aimed to validate the antibacterial activity of *C. pinnatifida* as a natural extract against *Enterococcus faecalis (E. faecalis)*, a multidrug-resistant bacterium.

E. faecalis was treated with different concentrations of *C. pinnatifida* to determine the optimal concentration for the most effective antibacterial effect. Fifteen different concentrations were applied for 6 and 24 h. The experimental method centered on confirming antibacterial activity using colony-forming units.

The experimental results demonstrated a proportional increase in antibacterial activity with elevated *C. pinnatifida* concentration. Notably, 99.99% and 100% antibacterial activity were observed at 10 mg/mL and 40 mg/mL concentrations, respectively.

Our results suggest that *C. pinnatifida* holds potential as an antibacterial agent against the multidrug-resistant *E. faecalis*.

Key words. *Crataegus pinnatifida, Enterococcus faecalis*, Antibacterial activity.

Introduction.

Recently, there has been a growing interest in health, with an increased focus on natural products with nutritional and functional properties. This rise parallels the increasing identification of natural bioactive substances. Among these, *Crataegus pinnatifida* (*C. pinnatifida*), a fruit belonging to the Rosaceae family, stands out for its unique aroma and taste, serving medicinal and culinary purposes, such as anti-obesity, digestive disease management, and high blood pressure regulation effects. *C. pinnatifida* also exhibits antioxidant, antibacterial, anti-thrombotic, hyperlipidemia improvement, weight control, and liver damage prevention effects [1].

Various studies are underway, focusing on natural products as a preliminary research area. However, limited studies have explored certain physiological activities, such as antioxidant effects encompassing reducing free radicals, mitigating radical reactions, and enhancing reducing power [2]. The mature fruit of the hawthorn tree within the Rosaceae family is represented by *C. pinnatifida*. Its anti-inflammatory activity has been substantiated, and the anti-thrombotic effects of specific fractions have been validated using various solvent extracts. However, research on the antibacterial activity of the organic solvent fraction of *C. pinnatifida* extract remains limited.

Previous studies on *C. pinnatifida* have highlighted the antibacterial activity of its methanol extract fractions and residues against various pathogenic and food-poisoning microorganisms, such as *Pseudomonas aeruginosa* and *Candida albicans*, which are multidrug-resistant bacteria [3].

Enterococcus faecalis (E. faecalis) is a gram-positive bacterium that exists in humans and animals and is recognized for inducing life-threatening infections through antibiotic-resistant nosocomial infections. Notably, this bacterium exhibits a high prevalence in teeth subjected to root canal treatment, demonstrating an affinity for adhering to dentin and adeptly penetrating dentinal tubules [4]. Following root canal treatment, *E. faecalis* can re-enter the root canal through openings that remain unclosed despite root canal filling during treatment. The surviving bacteria, along with those invading the root canal, can proliferate, leading to the occurrence of root canal disease [5].

E. faecalis bacteria occur in a wide range of areas in the oral cavity, ranging from 0 to 50% [6]. It has been confirmed in previous studies that it appears in a variety of areas such as saliva, root canals, and plaque. According to a clinical experiment involving 21 patients, *E. faecalis* bacteria were observed in 43 gingival sulcus, 14 saliva, 10 root canals, and 10 percent of the tongue, and were frequently identified in patients with gingivitis and periodontitis [7].

E. faecalis is highly significant in the field of dental treatment, particularly during root canal treatment; therefore, sodium hypochlorite is used to clean root canals. However, the effectiveness of this chemical compound is limited, resulting in the persistence of numerous cases of post-root canal treatment infections by *E. faecalis* [8]. Various drugs are currently under application for their antibacterial properties against *E. faecalis*. However, this bacterium has demonstrated resistance to vancomycin, prompting extensive research endeavors aimed at the development of alternative antibiotics [9].

Chlorhexidine (CHX) has demonstrated efficacy in eliminating *E. faecalis* during experiments. However, despite its broad spectrum of antibacterial properties, CHX exerts long-term effects attributed to its binding to dentin [10]. An essential consideration regarding the effectiveness of a drug lies in the fact that bacteria within the root canal form a biofilm, developing resistance to the drug. The hypothesis posits that resistant polysaccharide substrates act as physical and chemical barriers, preventing the spread of antibiotics. Additionally, the bacteria become resistant to antibiotics owing to reduced metabolic activity as they grow slowly in response to limited nutrients [11].

Therefore, in this study, we aimed to validate the antibacterial effects of *C. pinnatifida*, as a natural extract, against the antibiotic-resistant bacterium *E. faecalis*.

Materials and Methods.

***E. faecalis* culture.**

E. faecalis was obtained from the Korean Collection for Type Cultures (KCTC, Daejeon, Korea) and subjected to passaging in brain heart infusion medium (BHI; Sigma-Aldrich, St. Louis, MO, USA). Following incubation, culture was performed in an environment maintained at 37°C for 24 h.

***C. pinnatifida* extract.**

C. pinnatifida was purchased from Hwarim Pharmaceutical, ground into small pieces, mixed with 70% ethanol (ethyl alcohol, sigma, saint louis, USA) and shaken at 60°C for 12 h. Subsequently, only the supernatant was filtered, concentrated using filter paper (fiter disc, whatman, saint louis, USA), lyophilized, and diluted for further use.

Colony forming unit assay.

E. faecalis was treated with various concentrations of *C. pinnatifida* extract (0.01mg/ml, 0.05mg/ml, 0.1mg/ml, 0.3 mg/ml, 0.5 mg/ml, 1 mg/ml, 3 mg/ml, 5 mg/ml, 10 mg/ml, 15 mg / ml, 20 mg/ml, 25 mg/ml, 30 mg/ml, 35 mg/ml, 40 mg/ml) for 6 and 24 h. Subsequently, antibacterial activity was measured in colony-forming units (CFUs).

Statistical analysis.

Antibacterial effects were statistically analyzed using IBM SPSS Statistics 24.0 (SPSS Inc., Chicago, IL, USA) at a 95% significance level, followed by one-way analysis of variance and Tukey test as post hoc analysis ($P<0.05$).

Results.

Treatment with *C.pinnatifida* extracts for 6 h resulted in a 99.99% bacterial growth inhibition at 10 mg/mL and complete bacterial death (100%) at 40 mg/mL. The inhibition rates at lower concentrations were 1.07%, 2.85%, and 40.81% at 0 mg/mL, 0.05 mg/mL, and 0.1 mg/mL, respectively. Increasing *C. pinnatifida* extract concentration correlated with a higher bacteria inhibition rate.

E. faecalis was treated with *C. pinnatifida* extracts for 24 h. At 10 mg/mL and 15 mg/mL, 96.79% and 99% bacterial growth inhibition was observed. However, at 0 mg/mL, 0.05 mg/mL, and 0.1 mg/mL, 14.10%, 18.11%, and 22.26% bacterial growth inhibition was observed. The bacteria inhibition rate increased with higher *C. pinnatifida* concentration (Figure 1).

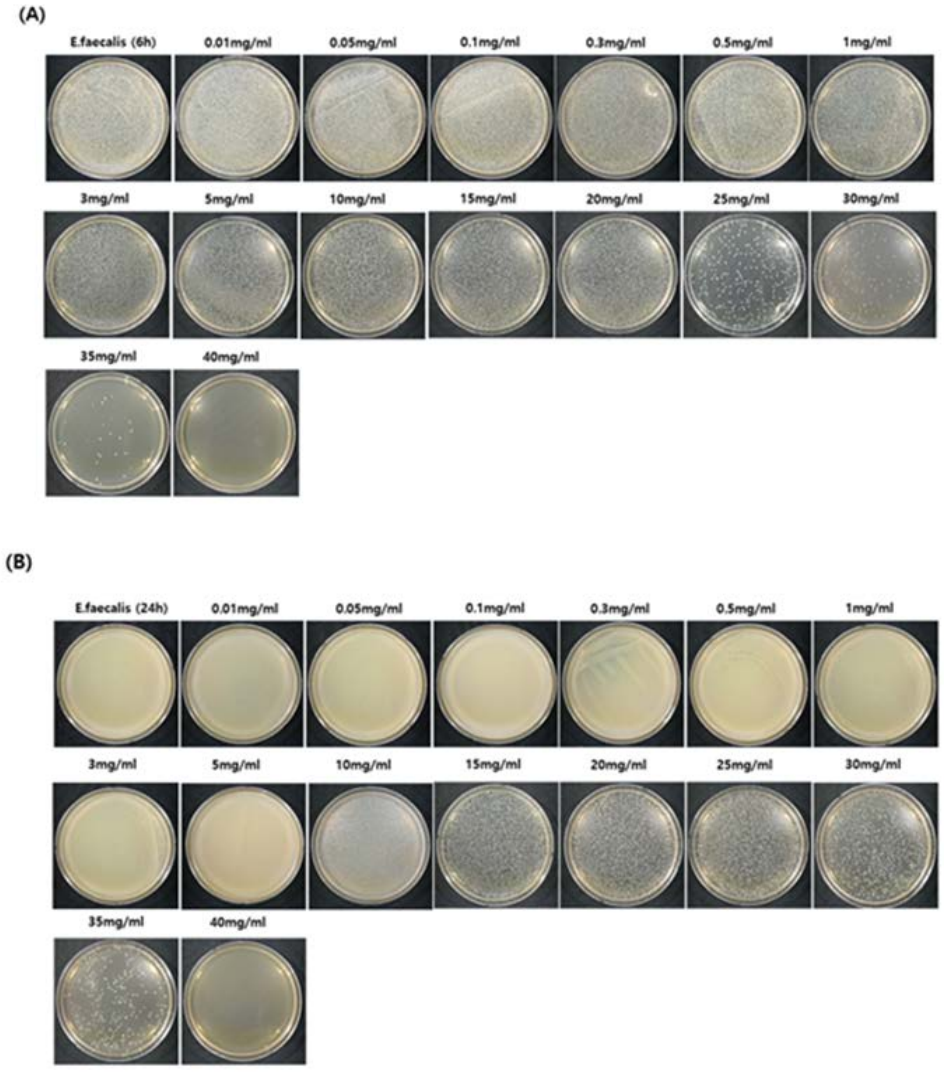

Figure 1. *CFU results from applying C. Pinnatifida (0, 0.01,0.05, 0.1, 0.3, 0.5, 1, 3, 5, 10, 15, 20, 25, 30, 35 and 40 mg/mL) to E. faecalis bacteria for 6 hours(A) and 24(B).*

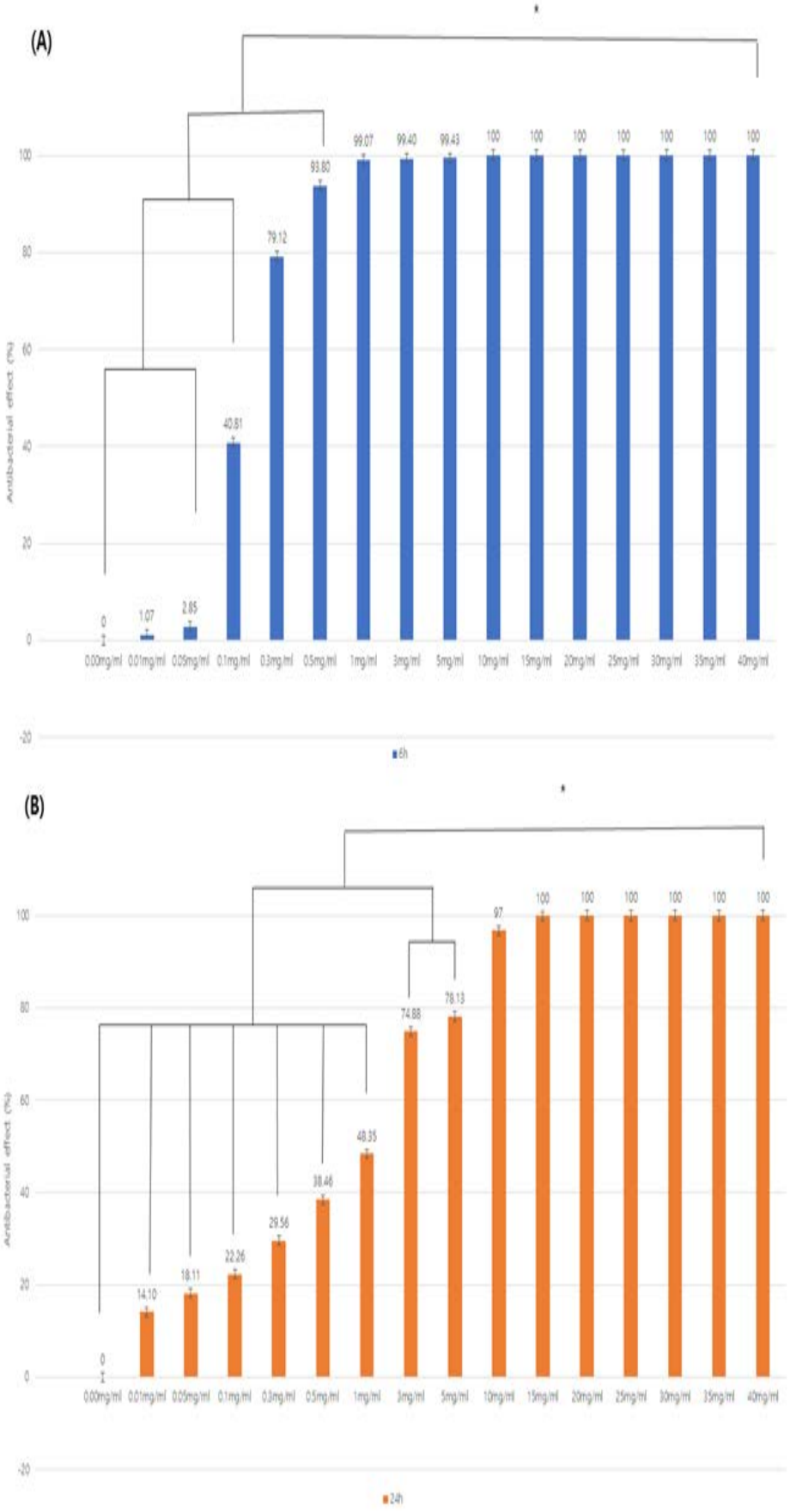

Figure 2. *Antibacterial effects(%) of C. pinnatifida CFUs with 0, 0.01,0.05, 0.1, 0.3, 0.5, 1, 3, 5, 10, 15, 20, 25, 30, 35 and 40 mg/mL (P<0.05) (A): Antibacterial effect applied to E.faecalis for 6 hours, (B): Antibacterial effect applied to E.faecalis for 24 hours.*

The results of *E. faecalis* exposure to *C. pinnatifida* extracts for 6 and 24 h are presented graphically in Figure 2. Significant antibacterial activity was observed from 10 mg/mL for both durations, indicating an increase in antibacterial effectiveness with higher concentrations of *C. pinnatifida* extract (Figure 2).

Discussion.

Achieving successful root canal treatment necessitates the complete eradication of bacteria within the root canal. This process involves not only mechanical removal using a file but also chemical removal through drug application [12]. Various clinical studies have utilized drugs, such as calcium hydroxide; however, resistance, specifically from bacteria, such as *E. faecalis*, has been confirmed. Notably, resistant bacteria remain within the root canal and are considered significant contributors to root canal treatment failures [13].

E. faecalis has been identified in over 30% of endodontically treated teeth with periapical lesions. Furthermore, bacteria subjected to root canal treatment have been demonstrated to exhibit resistance to a wide range of antibacterial and antibiotic drugs, rendering them a primary cause of recurrent chronic apical periodontitis [14]. Bacteria within the root canal form biofilms and exhibit resistance to drugs, which increases the likelihood of treatment failure [14]. Therefore, this study was undertaken to assess the resistance of *E. faecalis*, a bacterium known to exhibit resistance to existing antibiotics, to the extract of *C. pinnatifida*, a natural substance [15].

We assessed the antibacterial activity of *C. pinnatifida* against *E. faecalis* at various concentrations (0.01 mg/ml to 40 mg/ml). Following its application for 6 and 24 h, antibacterial activity was validated through CFUs, with a notable increase in antibacterial activity observed as the concentration of *C. pinnatifida* rose.

Considering the resistance of *E. faecalis* to drugs and the resistance of biofilm-state bacteria, it is necessary to acknowledge the possibility of bacteria-induced root treatment failure and adopt a strategic elimination approach [16]. In addition, the antibacterial effects of natural products, specifically *C. pinnatifida*, observed in this study confirm the presence of substances capable of combating existing antibiotics, as reported regarding antibiotic resistance in *E. faecalis* [17].

C. pinnatifida contains known pharmacologically active substances, such as caffeic acid, ursolic acid, hyperoside, pyrogallol, corosolic acid, salicylic acid, ferulic acid, catechin, protocatechuic acid, isoquercitrin, and chlorogenic acid [18]. Additionally, 1% *C. pinnatifida* extract, added to pork seasoning, was demonstrated to reduce the total number of bacteria [19]. In this study, a 99% bacterial inhibition rate was observed at 1 mg/ml after 6 h, with complete inhibition (100%) observed at 40 mg/ml. After 24 h, the bacterial growth inhibition rate was 99% at 15 mg/ml. These results indicate the efficacy of *C. pinnatifida* at both high and low concentrations, particularly in short-term use.

There was a difference in antibacterial activity by concentration between the group applied for 6 hours and the group applied for 24 hours. When applying the extract, the weakening antibacterial effect over time means that the medicinal effect of the extract is reduced, so application time and concentration must be taken into consideration.

However, the antibacterial test results showed a bacterial inhibition rate of 99% from 1mg/ml under 6-hour conditions. Therefore, the appropriate antibacterial concentration can be confirmed as 1mg/ml.

C. pinnatifida is considered biologically active, given its polyphenol and flavonoid content. Previous studies have demonstrated the excellent antibacterial activity of *C. pinnatifida* against various pathogenic and foodborne microorganisms, including multidrug-resistant *Pseudomonas aeruginosa* and *Candida sp*. [20]. In our study, antibacterial activity was also confirmed against the *E. faecalis*, with the observed antibacterial effect increasing alongside elevated concentration of *C. pinnatifida*. Confirmed in *C. pinnatifida* extract are components with antibacterial activity, particularly phenolic compounds, and flavonoids, exhibiting radical scavenging activity and demonstrating antibacterial activity against *E. faecalis*.

Although this study did not conduct research on multi-resistant bacteria, *E. Faecalis* is known to have resistance to vancomycin. Based on the results of this study, it was confirmed that the development of antibiotics against *E. Faecalis* is possible, and it can be applied to various bacteria in future studies. Therefore, we plan to apply various antibacterial tests.

E. Faecalis, which is known to have a high prevalence in dentistry and is known to be good at penetrating the dentinal tubules, re-enters the root canal using gaps in the root canal that are not closed even after root canal filling during root canal treatment, and the surviving bacteria and bacteria that invade the root canal Since it can proliferate again and cause root canal disease, it can be used as an alternative drug in dental clinics during root canal treatment by using *C. pinnatifida*, whose antibacterial activity was confirmed in this study.

Conclusion.

The antibacterial activity of *C. pinnatifida* extract against *E. faecalis* was assessed using the CFU method for 6 and 24 h, confirming 99.99% antibacterial activity at 10 mg/mL. Given its antibacterial activity against bacteria prevalent in root canals, *C. pinnatifida* extract holds promise for future development as a root canal antibacterial drug.

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