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OH&RM ONE HEALTH & RISK MANAGEMENT

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The Moldovan Association for Biosafety and Biosecurity (MDBBA) is a scientific and practical, instructive and educational, non-governmental, apolitical and non-profit professional organization, created in 2017.

The main objective of the association is the development of good practices and culture in the field of biosafety and biosecurity and the promotion of knowledge within professional and research-innovation groups.

Biosafety – includes security principles, technologies and rules to be followed to prevent unintended exposure to pathogens and toxins or their accidental release/leakage.

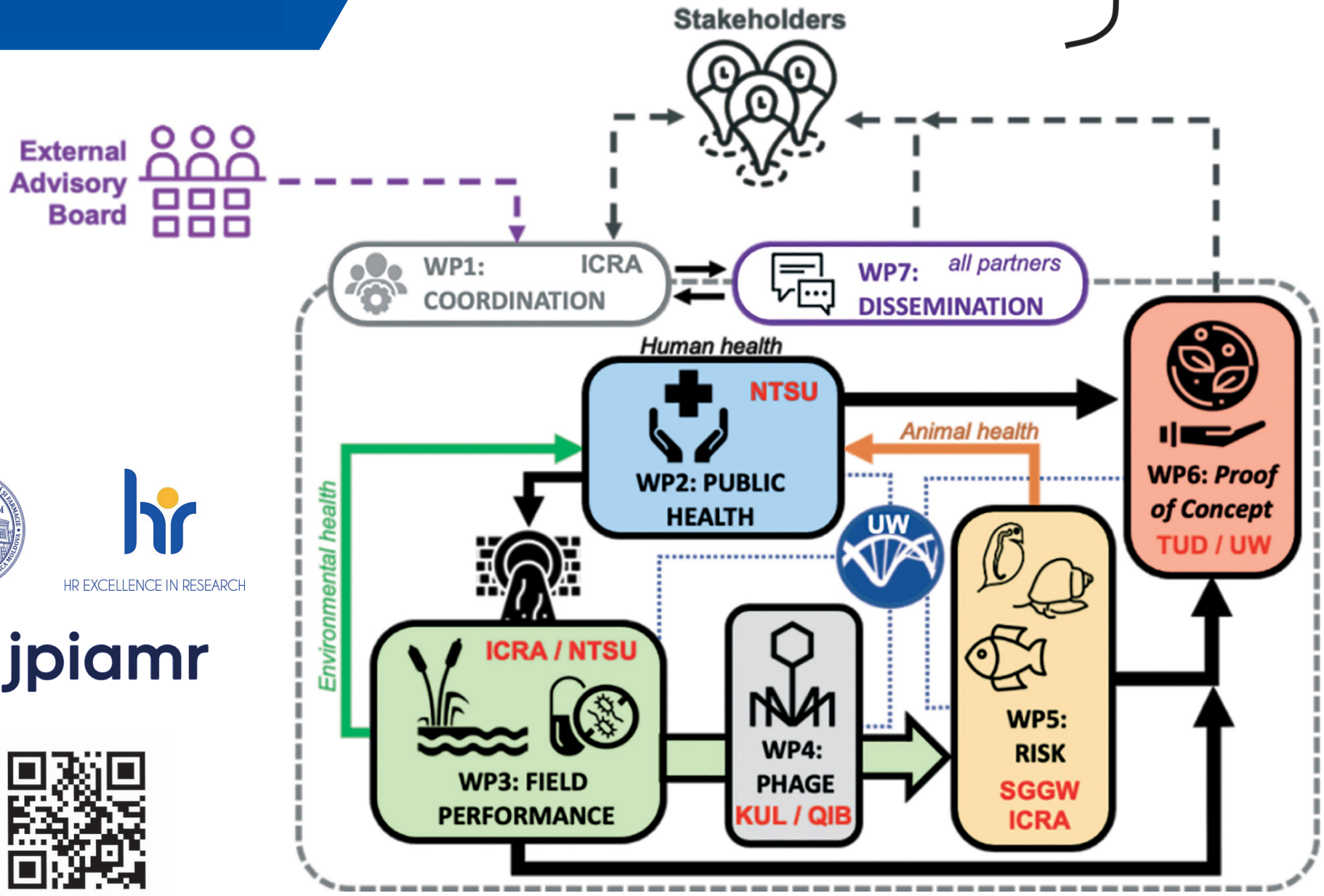
"Protection of personnel, population from unintended exposure to pathogens/biohazardous material".

Biosecurity - includes a wide spectrum of measures (biosecurity policies, regulatory regime, scientific and technical measures) applied in an organized framework, necessary to minimize risks (prevention of actions, terrorist attacks by the intentional release of pathogens or toxins as well as loss, their theft or misuse).

"Protection and prevention of theft, intentional misuse of pathologies/biohazardous material".

Risk management – is a decision-making process in which the results of risk assessment (the process of estimating workplace hazards) are integrated with economic, technical, social and political principles to generate strategies for risk reduction.

PhageLand



HR EXCELLENCE IN RESEARCH

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REZUMAT

PhageLand își propune să dezvolte o nouă strategie de intervenție combinând capacitatea de purificare ieftină și ecologică a sistemelor pasive de tratare, și anume a zonelor umede construite (ZUC), cu un tratament dedicat pe bază de fagi pentru a preveni transmiterea rezistenței la antibiotice (RA) din apele uzate în apele de suprafață.

PhageLand include investigații de sănătate publică care vizează agenți patogeni bacterieni multirezistenți la medicamente (RAM) în țările cu venituri mici și medii (low-middle income countries - LMIC) din Europa de Est. Tratamentul pe bază de fagi va fi adaptat în mod special pentru a elimina acești agenți patogeni multirezistenți la medicamente din apele uzate. În paralel, PhageLand își propune să evalueze: a) capacitatea de auto-purificare a modelelor de ZUC la scară largă care operează în Spania și Moldova în îndepărtarea reziduurilor de antibiotice, ARB și a genelor de rezistență la antibiotice (RAG); și b) riscul potențial asociat cu diseminarea RAM și RAG în cadrul comunităților bacteriene indigene și printre animalele care locuiesc în ZUC.

Tipărit din resurse financiare ale proiectului transnațional multilateral **„Phage treatment and wetland technology as intervention strategy to prevent dissemination of antibiotic resistance in surface waters (*PhageLand*)”**, 22.80013.8007.1M.

USMF „NICOLAE TESTEMIȚANU”

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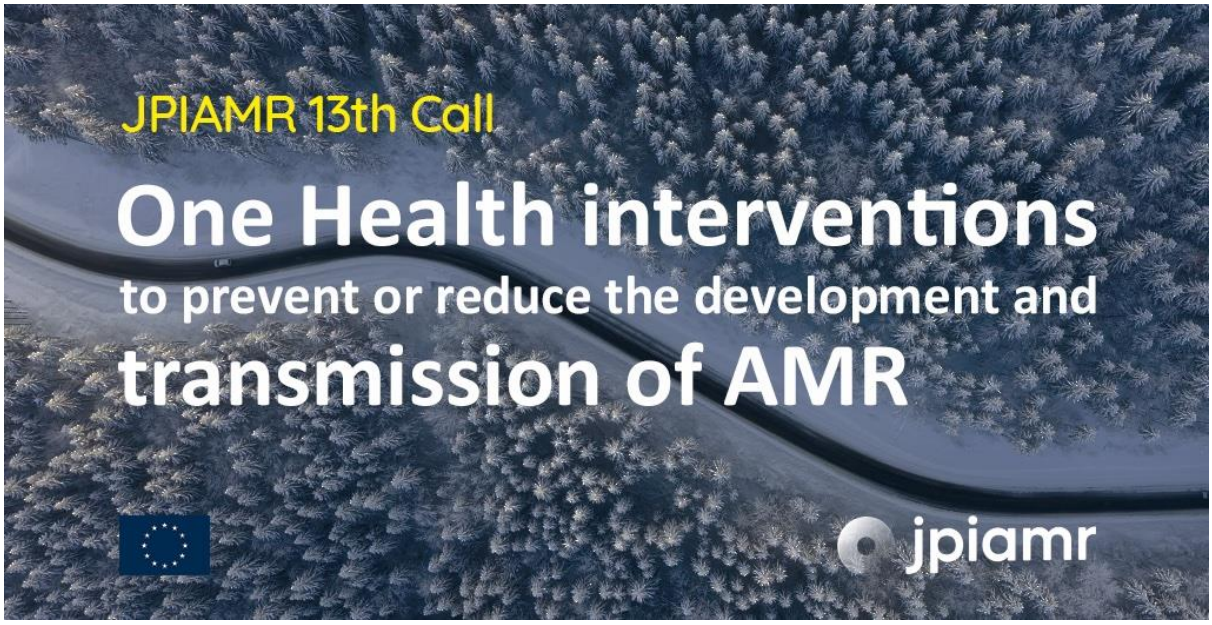
P R O G R A M

Conferința națională

„Sănătatea și fenomenul rezistenței la antimicrobiene în țările cu venituri mici și medii din Europa de Est”

(Rezultatele intermediare ale proiectului *PhageLand*)

27 ianuarie 2024



în cadrul

Proiectului transnațional multilateral „Phage treatment and wetland technology as intervention strategy to prevent dissemination of antibiotic resistance in surface waters (*PhageLand*)”, 22.80013.8007.1M.

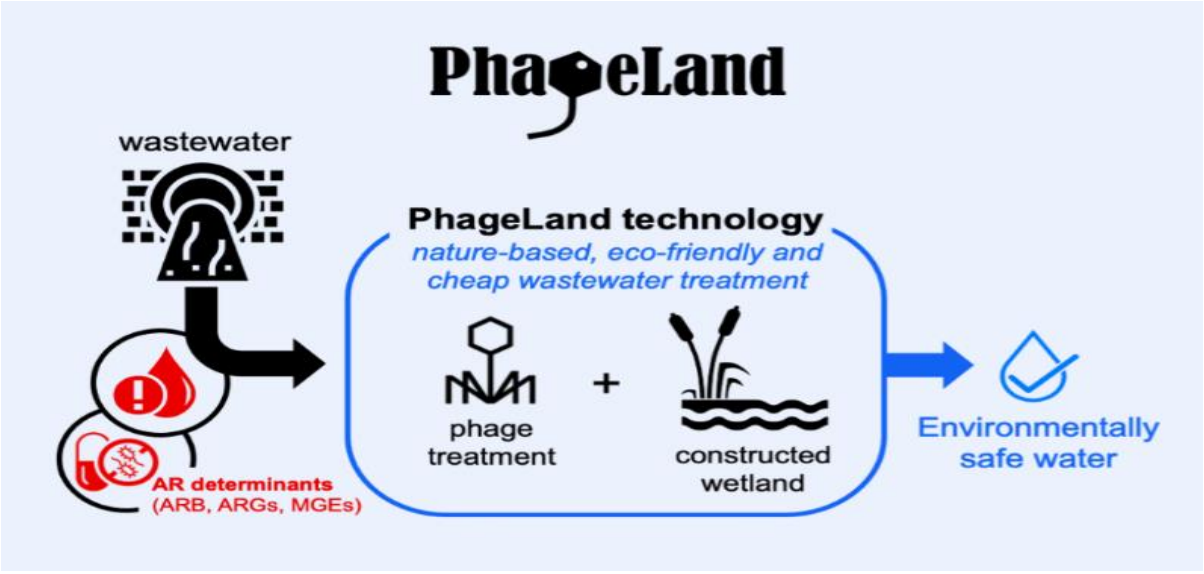
Descrierea conferinței

Conferința națională ***„Sănătatea și fenomenul rezistenței la antimicrobiene în țările cu venituri mici și medii din Europa de Est”*** este organizată de Universitatea de Stat de Medicină și Farmacie „Nicolae Testemițanu”, Institutul Național de Cercetare în Medicină și Sănătate (INCMS), în cadrul Apelului multilateral **JPIAMR HARISSA/ Joint Program Initiative (JPI) Antimicrobial Resistance** „One Health Interventions to prevent or reduce the development and transmission of Antimicrobial Resistance, AMR", Proiectului transnațional multilateral „Phage treatment and wetland technology as intervention strategy to prevent dissemination of antibiotic resistance in surface waters (***PhageLand***)”, 22.80013.8007.1M.

Finanțator: Comisia Europeană
Acțiune de cofinanțare: JPIAMR-Action
Acronim: PhageLand
Coordonator: VETENSKAPSRADET (SRC), Suedia
Perioada de implementare: 2021- 2025

Partenerii internaționali aprobați prin Consorțium:

1) Catalan Institute for Water Research, Department of Water Quality, Spain
2) University of Warsaw, Faculty of Biology, Institute of Microbiology, Dept. of Environmental Microbiology and Biotechnology, Poland
3) Warsaw University of Life Sciences (SGGW), Department of Biochemistry and Microbiology, Poland
4) KU Leuven, Laboratory of Gene Technology, Leuven, Belgium
5) Quadram Institute Bioscience, Norwich Research Park, Rosalind Franklin Road, Norwich, the United Kingdom
6) Delft University of Technology, Department of Biotechnology, the Netherlands
7) *Nicolae Testemitanu* State University of Medicine and Pharmacy, National Institute for Health and Medical Research, Republic of Moldova



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9.00-9.15	**ÎNREGISTRAREA PARTICIPANȚILOR (online)**	
9.15-9.30	**Cuvânt de deschidere** Angela PARASCHIV Liliana CONDRATICOVA Aurelia HANGANU Emil CEBAN Stanislav GROPPA	***Moderatori:*** **Mihail TODIRAŞ** **Alina FERDOHLEB**
9.30-10.00	**Prezentare plenară** Alina FERDOHLEB ***Prezentarea generală a proiectului transnațional multilateral PhageLand***	***Moderator:*** **Mihail TODIRAŞ**

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10.00-11.30 **Sesiunea 1**

ABORDAREA MULTISECTORIALĂ ÎN FENOMENUL DE REZISTENȚĂ LA ANTIMICROBIENE (RAM)

Moderatori:
Greta BĂLAN
Olga BURDUNIUC

- Maria ANTON
  Sistemul de supraveghere epidemiologică a rezistenței microorganismelor la antimicrobiene în Republica Moldova
- Oana-Simina IACONI, Greta BĂLAN, Alina FERDOHLEB
  Terapia cu bacteriofagi: metodă de combatere a rezistenței la antimicrobiene
- Olga SOFRONIE
  Diagnosticul de laborator în infecția cu virusul encefalitei transmise de căpușe
- Svetlana COLAC, Mariana ULINICI, Olga BURDUNIUC
  Analiza diversității genetice a virusului SARS-CoV-2
- Lucia GALBEN, Oana-Simina IACONI, Greta BĂLAN
  Utilizarea potențială a bacteriofagilor la bioaugmentarea proceselor biologice de epurare a apelor uzate
- Lucia TURCAN, Svetlana LATUS, Anastasia CARACAS
  Resistance of pathogens causing urinary tract infections in children
- Nicolae BACINSCHI, Anastasia CARACAS, Svetlana LATUS, Eugenia VASILACHE
  Rational use of antibacterials in pleural empyema
- Nicolae PUȘCAȘ
  Procedee de determinare rapidă a microorganismelor în medicamente
- Dmitri IUNAC, Greta BĂLAN
  Acțiunea sinergică a compușilor chimici și extractelor din spirulină

- Valentina CRETU, Olga BURDUNIUC, NICOLAE STARCIUC
  A review on prevalence and antimicrobial resistance of Salmonella spp. and Campylobacter spp: one health perspective

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11.30-12.30 **Sesiunea 2**

ABORDAREA O SINGURĂ SĂNĂTATE şi ACTUALITĂŢII ÎN SĂNĂTATEA PUBLICĂ

Moderatori:
Cătălina CROITORU
Elena CIOBANU

- Doina MACARI, Corina SCUTARI, Simona NEGREŞ, Mihail TODIRAŞ ***Mapping the antibiotic consumption in the Republic of Moldova***
- Alina FERDOHLEB, Elena BUCATA, Vladimir BERNIC
  Benefits and challenges in using wetlands for wastewater treatment
- Lilia ANDRONACHE, Valeriana PANTEA, Aurelian GULEA, Olga TAGADIUC, Valerii MATCOVSCHI, Valentin GUDUMAC
  Use of micromolecular copper complexes of thiosemicarbazidates as an endogenous catalase inducer/activator
- Ana VÎLCOVA
  Rezistența la antibiotice: factor de risc în mediul ocupațional al lucrătorilor medicali
- Ioana CALIGA
  Maladii bacteriene cu manifstări neurologice influențate de schimbarea climei
- Cătălina CROITORU
  Microorganisme, boli infecţioase şi relaţia cu schimbările climatice
- Elena CIOBANU
  Efectele schimbărilor climatice asupra calităţii apei potabile

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12.30-14.00 **Sesiunea 3**

PROMOVAREA CALITĂŢII VIEŢII POPULAŢIEI ÎN ŢĂRILE CU VENITURI MICI ŞI MEDII (LMIC)

Moderatori:
Larisa SPINEI
Alina FERDOHLEB

- Livia ŢAPU, Alina FERDOHLEB, Larisa SPINEI, Carles M. BORREGO
  Cunoştinţe, atitudini şi practici privind rezistenţa la antimicrobiene în ţările cu venit mic şi mediu
- Raisa PUIA, Galina BUTA, Alina FERDOHLEB, Mihail TODIRAS
  The concept of the biobank information system of Nicolae Testemitanu State University of Medicine and Pharmacy of the Republic of Moldova
- Lyubov VLASYK, Natalia RYNHACH, Anastasiya SUKHOLOTYUK, Svitlana MARINOVA, Leonid VLASYK
  Changes in young people's attitudes to the problem of COVID-19, flu and other upper respiratory infections during the war in Ukraine

- Iulianna LUPASCO, Tatiana GHELIMICI, Ludmila GOLOVATIUC
  Investigation of pancreatic activity parameters in patients during COVID-infection in Republic of Moldova
- Elina ȘOR
  Assessment of the life quality after surgery for perforated gastroduodenal ulcer
- Elena BEREZOVSCAIA
  Indicii calității vieții la bolnavii cu hepatopatii cronice din Republica Moldova
- Ludmila GOMA
  Strategii de promovare în marketingul cabinetului stomatologic
- Mariana CERNIȚANU
  Methods for stimulating the cognitive abilities used by students
- Ecaterina GÎNCOTA, Ludmila MURZAC, Larisa SPINEI
  Case presentation - unilateral dyskinetic cerebral palsy: time for a change in cerebral palsy classification?
- Stela RACOVITA, Veaceslav MOȘIN , Mariana SPRÎNCEAN
  Cytogenetic study in men from the population of the Republic of Moldova
- Anastasia BENDELIC, Valentin BENDELIC, Ilia CATERENIUC
  Riscul varicelor recidivante și variantele anatomice ale venei safene mari

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13.00-13.30	**Închiderea lucrărilor conferinței**	***Moderatori:*** **Alina FERDOHLEB** **Greta BĂLAN** **Olga BURDUNIUC** **Cătălina CROITORU** **Elena CIOBANU** **Larisa SPINEI**

CUPRINS

CUVÂNT ÎNAINTE

ARTICOLE

REZUMATE

The joint transnational call for proposals 2021, JPIAMR HARISSA

Olga TAGADIUC,
PhD in medical science, Professor,
Chair of Biochemistry and Clinical Biochemistry discipline,
Nicolae Testemitanu State University of Medicine and Pharmacy,
Chisinau, Republic of Moldova

Joint programming (JP) has been a significant tool for planning and implementing research and development actions in Europe in this millennium. The European Union, along with its Member States and partners from associated and third countries, has endeavored to adopt a more coordinated approach to financing, planning, and implementing research and innovation activities through joint actions.

The Joint Program Initiative on Antimicrobial Resistance, JPIAMR, is an international collaborative platform involving 29 countries and the European Commission in the fight against antimicrobial resistance (AMR). JPIAMR coordinates national research funding and supports collaborative efforts to address AMR knowledge gaps from the One Health perspective. The JPIAMR program identifies key areas that require attention and provides guidance to countries, aiming to harmonize national and international AMR research programs to combat antimicrobial resistance globally. Aligned with the six strategic directions of the general research and innovation agenda of JPIAMR, the partnership engages in a wide range of activities with a One Health approach, seeking and supporting solutions to reduce the transmission of antimicrobial-resistant bacteria.

The Moldovan team from the Nicolae Testemitanu State University of Medicine and Pharmacy, Republic of Moldova, is a partner in an international consortium implementing the project Phage treatment and wetland technology as an intervention strategy to prevent the dissemination of antibiotic resistance in surface waters (PhageLand). This initiative is part of the Joint Transnational Call for Proposals 2021, titled "One Health interventions to prevent or reduce the development and transmission of antimicrobial resistance" (JPIAMR HARISSA).

PhageLand aims to develop a novel intervention strategy that combines the cost-effective and envi-

ronmentally friendly treatment capabilities of constructed wetlands with specific phage-based treatments. The goal is to prevent the transfer of antibiotic resistance from wastewater to surface water. The PhageLand project, a multi-stakeholder study, utilizes public health methods to identify multidrug-resistant bacterial pathogens in the Republic of Moldova. Phage-based treatments have been adapted to eliminate these multidrug-resistant pathogens from wastewater. Simultaneously, PhageLand assesses, in Spain and Moldova, the capacity of real-scale wetlands to self-purify from antibiotic residues, antibiotic-resistant bacteria, and antibiotic resistance genes. The project also explores the potential risks associated with the dissemination of the latter in native bacterial communities and among wetland animals.

The project serves as a commendable model for the involvement of researchers from the Republic of Moldova in multilateral projects addressing significant global and national issues. It facilitates the development of capacities among Moldovan researchers, fosters the establishment of networks with colleagues from various countries, and enhances the visibility of Moldovan researchers on the international stage.

Proiectul multilateral transnațional „Phage treatment and wetland technology as intervention strategy to prevent dissemination of antibiotic resistance in surface waters" (*PhageLand*)

Sveatoslav POSTORONCĂ,
Direcția cooperare internațională și dezvoltare durabilă,
Agenția Națională pentru Cercetare și Dezvoltare
Membru în Consiliul de administrare al I.C.P. RAM
Chișinău, Republica Moldova

Stimate cititor,

În condițiile actuale, caracterizate de profunde crize sistemice globale, în contextul cărora ne confruntăm cu serioase provocări și amenințări pentru cele mai importante ramuri vitale ale societății, așa ca: sănătate, climă, securitate alimentară, cibernetică, militară și energetică, exercițiul de alocare a finanțărilor din fondurile naționale sau internaționale, dedicate activităților științifice necesită implicare profesionistă. Pentru a crește gradul de reziliență, performanță și competitivitate economică a unei societăți, subiectele de utilizare cât mai eficientă a resurselor alocate, au fost și rămân „cap-de-listă" pe agendele managementului sistemului domeniilor de cercetare și inovare. La fel de important este și rolul prioritizării tematicilor procesate de comunitatea științifică. În urma analizelor statisticilor ce cuantifică prejudiciile aduse la nivel mondial atât omului, cât și mediu ambiant în care el vețuiește, ca urmare a utilizării insuficient de raționale a preparatelor antibiotice, domeniul Rezistenței la Antimicrobiene (RAM) se plasează pe un loc distinct. Savanții au descifrat deja foarte bine circuitul preparatelor folosite de acest gen în natură, ajungând la concluzia că răspândirea efectelor RAM nu se supun limitărilor condiționate de hotare dintre țări sau părți ale lumii, tipuri de organizare a orânduirilor sociale, nivelul de dezvoltare economică a țărilor etc. Anume din aceste considerente și sunt constituite instituții la nivel internațional, cu mandat dedicat de gestionare a arealului de probleme de acest gen. Important este exemplul implicării Organizației Mondiale a Sănătății, Organizației Națiunilor Unite prin intermediul ODD-urilor și a altor asociații profesionale de anvergură. Același format participativ persistă și la partea de activități de cercetare, urmărind consolidarea efortului oamenilor de știință într-o manieră sistematizată, focusată pe sănătatea oamenilor, a animalelor și pe starea mediului, viziu-

ne care a formulat conceptul „O Singură Sănătate" („One Health").

Programul-cadru pentru cercetare și inovare Orizont Europa al UE este în derulare, al nouălea la număr, de-a lungul anilor. Calea parcursă în organizarea și desfășurarea activităților de cercetare și inovare pe continentul European, totodată stabilind legături de colaborare pe întregul mapamond a permis crearea unor mecanisme și instrumente capabile de a lansa acțiuni de finanțare a proiectelor realizate în comun de consorțiile transnaționale, practică preluată de la programele anterioare. În lista acestora se regăsesc cele zece Inițiative Comune de Program, din care face parte Rezistența la Antimicrobiene (Joint Programme Initiatives on Antimicrobian Resistance - JPIAMR). I.C.P. RAM este o platformă internațională colaborativă, ce întrunește Comisia Europeană cu 29 de state ale lumii în dezideratul de a contracara fenomenul RAM, unde, din afara continentului European fac parte Canada, Argentina, Chile, India, Japonia, Coreea de Sud, Africa de Sud și Egipt. Anual, în cadrul inițiativei RAM sunt lansate concursuri multilaterale pentru propuneri de proiecte prin intermediul acțiunilor de co-finanțare din bugetele naționale ale țărilor participante, cu contribuția CE. Foarte important este de a menționa faptul implicării tot atât de colaborative a tuturor reprezentanților țărilor semnatare în procesul de elaborare nu doar a documentelor pentru lansarea concursurilor, proces în care sunt agreate toate condițiile expuse de parteneri pentru a se conforma la prevederile cadrului legal național din domeniu, dar și a documentelor strategice și operaționale de politici, ce reglementează dezvoltarea științifică a domeniului. Respectiv, în asemenea exercițiu sunt implicați alături de reprezentanții managementului în cercetare și inovare, experți din rândurile savanților cu recunoaștere internațională.

Republica Moldova, prin adresarea scrisorii de intenție din partea Agenției Naționale pentru Cercetare și Dezvoltare (ANCD) către organele de guvernanță ale I.C.P. RAM, în 2019 a fost acceptată în calitate de membru cu drepturi depline, astfel, în perioada anului 2020, participând în procesul de prefigurare a concursului pentru anul 2021. Este un rezultat reprezentativ al colaborării ANCD cu reprezentanții organizațiilor naționale din domeniile cercetării și inovării, precum Universitatea de Stat de Medicină și Farmacie „Nicolae Testemițanu", Agenția Națională pentru Sănătate Publică și a altor instituții din sistem, exemplu ce merită a fi propagat și replicat în rândurile comunității științifice din țară. La momentul de debut al țării noastre în concursul I.C.P RAM, lansat în 2021 cu genericul „Intervenția abordării O Singură Sănătate pentru a preveni sau a reduce evoluția și transmisia Rezistenței la Antimicrobiene" din lista celor depuse, câștigătoare a fost

propunerea proiectului „Phage treatment and wetland technology as intervention strategy to prevent dissemination of antibiotic resistance in surface waters" cu acronimul „PhageLand", având USMF „Nicolae Testemițanu" ca partener cu instituțiile omoloage din Spania, Polonia, Belgia, Principatele Unite și Țările de Jos. Un alt exemplu de colaborare a cercetătorilor moldoveni este proiectul „Bench, Bedside, Business, and Beyond: innovative solutions for AMR diagnostics" cu acronimul „B2B2B AMRDx", unde sunt implicați reprezentanții Institutului de Ftiziopneumologie „Chiril Draganiuc". Participarea ANCD la concursurile lansate de I.C.P. RAM continuă. Anul curent 2024 suntem iarăși parte a noului apel „Intervenții care avansează pentru a promova acțiuni pentru a contracara apariția și răspândirea rezistenței bacteriene și fungice și pentru a îmbunătăți tratamentele". Nutrim speranțe în depunerea noilor propuneri reușite, cu accesarea granturilor de finanțare pentru implementarea proiectelor.

Revenind la cele menționate la începutul acestui mesaj, este cazul de a reitera despre faptul conștientizării autorităților asupra necesității finanțării tematicilor menționate în context, ca fiind din aria celor de importanță majoră pentru sănătatea populației țării, integrată în sănătatea la nivel global. Despre aceasta mărturisește și direcția „Supravegherea și controlul rezistenței la preparatele antimicrobiene" din cadrul priorității strategice dedicată sănătății, regăsindu-se în documentul recent aprobat la nivel de țară „Programul național în domeniile cercetării și inovării pentru anii 2024-2027". De asemenea, la partea de cooperare internațională, ANCD intenționează participarea în noi forme de parteneriate profesionale dedicate combaterii RAM.

Mult succes cercetătorilor și inovatorilor, celora ce aspiră către explorarea platformelor unor asemenea activități în cooperare, apreciată implicare cu dăruire, ce ne asigură o societate sănătoasă și prosperă.

TERAPIA CU BACTERIOFAGI: METODĂ DE COMBATERE A REZISTENȚEI LA ANTIMICROBIENE *(ANALIZĂ NARATIVĂ)*

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Keywords: *bacteriophages, phage therapy, antimicrobial resistance, human health.*

BACTERIOPHAGE THERAPY: A METHOD TO COMBAT ANTIMICROBIAL RESISTANCE (NARRATIVE ANALYSIS)

Introduction*. Antimicrobial resistance is a 50-year pandemic requiring interventions that are harmless to human health. In this context, bacteriophage therapy has been rediscovered, which, according to the literature, is considered the optimal solution to mitigate this phenomenon.*

The aim *of this literature review is to construct a clear understanding of the possibilities, challenges, and needs that this innovation presents in terms of reducing antibiotic resistance.*

Material and methods. *For this study, a search of the SCOPUS and PubMed databases was conducted using the keywords and BOOLEAN operators "AND," "OR," and "NOT." The search yielded 911 scientific papers. After applying the inclusion/exclusion criteria, 40 papers were ultimately included in the final stage of the study.*

Results. *Based on the literature reviewed, a SWOT analysis of the potential of bacteriophage therapy as a method to combat antimicrobial resistance was conducted. The research identified several advantages in favor of bacteriophages, such as high specificity and low toxicity, among others. Additionally, the study revealed several opportunities that modern medicine can exploit, including personalized therapy assisted by genetic engineering and the use of phages as adjuvants in antimicrobial and antifungal therapy.*

Conclusions. *Antimicrobial resistance poses a significant global public health challenge, demanding prompt action. Bacteriophage therapy emerges as an alternative with multiple advantages and a promising long-term perspective.*

Cuvinte-cheie: *bacteriofagi, terapia cu fagi, rezistența antimicrobiană, sănătatea umană.*

Introduction*. Rezistența antimicrobiană este o pandemie care durează 50 de ani și necesită intervenții inofensive pentru sănătatea umană. În acest context,a fost redescoperită terapia cu bacteriofagi care, conform datelor din literatură-, este soluția optimală pentru a reduce acest fenomen.*

Scopul *acestui studiu de sinteză a literaturii este de a construi o imagine clară cu privire la posibilitățile și, pe care le aduce această inovație în materie de reducere a antibiorezistenței.*

Material și metode. *În cadrul acestui studiu s-a efectuat o căutare în bazele de date SCOPUS și PubMed utilizând cuvintele-cheie și operatorii BOOLEAN „AND", „OR" și „NOT". Căutarea a generat 911 lucrări științifice. După aplicarea criteriilor de includere/excludere, în etapa finală a studiului au fost incluse 40 de lucrări.*

Rezultate. *Pe baza literaturii cercetate a fost realizată analiza SWOT a potențialului terapiei cu bacteriofagi ca metodă de combatere a rezistenței la antimicrobiene. Cercetarea a determinat mai multe avantaje în favoarea utilizării bacteriofagilor – specificitate înaltă, toxicitate scăzută etc., și cîteva oportunități pe care medicina modernă le poate valorifica: terapia personalizată asistată de inginerie genetică, utilizarea fagilor ca adjuvanți în terapia antimicrobiană și antifungică.*

Concluzii. *Rezistența antimicrobiană este o problemă de sănătate publică globală care necesită o intervenție promptă. Terapia cu bacteriofagi este o alternativă cu multiple avantaje și o perspectivă favorabilă.*

Abrevieri: *ADN – acid dezoxiribonucleic; IDSA – Societatea de Boli Infecţioase a Americii (eng. Infectious Disease Society of America); EMA – Agenţia Europeană a Medicamentului; FDA – Administraţia Alimentelor şi a Medicamentelor (eng. Food and Drug Administration); GMO (eng.) – organism modificat genetic; MDR (eng.) – bacterii multi-drog rezistente; OMS – Organizaţia Mondială a Sănătăţi; PDR (eng.) – bacterii panrezistente; PRISMA – elemente de raportare preferate pentru revizuiri sistematice şi meta-analize (eng. Preferred Reporting Items for Systematic Reviews and Meta-Analyses); RAM – rezistenţa la antimicrobiene; SWOT – puncte tari, puncte slabe, oportunitati, amenintari (eng. Strengths, Weaknesses, Opportunities, Threats); UE – Uniunea Europeană.*

INTRODUCERE

Descoperirea penicilinei acum 95 de ani a revoluţionat medicina la nivel global, mai ales după ce Howard Florey şi Ernst Chain au reuşit, un deceniu mai târziu, să producă penicilina industrial (1). Astfel, începând cu anii ’40, descoperirea şi producerea pe scară largă a antimicrobienelor a permis eradicarea sau, cel puţin, controlul bolilor infecţioase. Până în anii 1960 au fost descoperite în jur de 20 de clase de antibiotice. Pe parcursul anilor, ritmul descoperirii şi aprobării produselor farmacologice cu efect antimicrobian a scăzut de aproximativ opt ori (1, 2, 3).

Utilizarea excesivă şi necorespunzătoare a antibioticelor a indus dezvoltarea de către agenţii patogeni a rezistenţei la antibiotice (RAM). Acum 15 ani, Societatea de Boli Infecţioase a Americii (IDSA) a catalogat RAM drept o criză globală, iar OMS, la rândul său, a clasificat rezistenţa la antimicrobiene drept o problemă majoră de sănătate publică din cauza căreia sunt înregistrate aproape cinci milioane de decese şi pierderi economice de aproape 20 de miliarde de dolari (4–7). Evoluţia agenţilor patogeni, combinată cu consumul crescut de antibiotice (cu sau fără indicaţie terapeutică) a favorizat, începînd cu anul 1942 când a fost depistată prima tulpină de *S.aureus* rezistentă la penicilină, apariţia tulpinilor bacteriene rezistente, multirezistente (MDR) şi chiar panre-zistente (PDR). Aceste tulpini reprezintă o problemă a sistemului sanitar la nivel mondial (3, 4, 5). Printre toţi agenţii patogeni care pre-zintă rezistenţă, OMS a identificat 12 bacterii, grupate în trei clase de prioritate care urgent necesită dezvoltarea de noi antibiotice sau de metode alternative pentru a fi contracarate. Primele cinci poziţii sunt ocupate de patogenii din grupul ESKAPE care înglobează *Acineto-bacter spp., Pseudomonas aeruginosa* şi Enterobacteriaceae (*Klebsiella pneumoniae* şi *Escherichia coli)* din grupul cu prioritate critică şi *Enterococcus faecium* şi *Staphylococcus aureus* din clasa cu prioritate înaltă (8, 9).

Necesitatea intervenţiilor rapide, în contextul în care RAM devine şi o problemă de mediu, a determinat cercetătorii să reexploreze capacită-ţile bacteriofagilor de a elimina bacteriile, mai ales cele rezistente (10–14). Bacteriofagii sunt virusuri capabile să distrugă bacteriile prin liza acestora după integrarea materialului genetic în genomul lor (15, 16). Fagii sunt omniprezenţi în natură şi în interacţiune permanentă cu bacteriile şi cu populaţia umană (15, 17). Istoria bacteriofagilor este mai veche decât a antibioti-celor, prima expunere cu privire la existenţa acestora anticipând descoperirea lui *Alexander Fleming* cu peste 30 de ani (5). De la prima lor utilizare de către d’Herelle pentru tratarea unui copil de 12 ani la Spitalul de Copii din Paris şi până în prezent, bacteriofagii şi-au demonstrat eficacitatea în tratamentul diverselor maladii în diverse ţări: în Franţa contra infecţiilor provo-cate de *S.aureus, P.aeruginosa* şi *E.coli*; în SUA, Polonia şi URSS pentru tratarea rănilor solda-ţilor şi bolilor de campanie (dizenteria şi alte boli diareice) (5, 16, 18). După 50 de ani, terapia cu bacteriofagi revine în atenţia cercetătorilor şi a clinicienilor în două forme distincte posibile – preparate gata pentru utilizare contra agenţilor infecţioşi identificaţi în mediu sau pentru tratamentul uman în cazul maladiilor comune (răceală, laringită, boli diareice), sau cac preparate personalizate pentru tratamentul afecţiunilor pacientului infectat cu o tulpină specifică (prioritar, MDR sau PDR) (19).

Scopul acestei lucrări este de a realiza analiza narativă a oportunităţilor şi a provocărilor pe care le aduce terapia cu bacteriofagi pentru medicina modernă prin realizarea analizei SWOT după cercetarea literaturii de specialitate.

MATERIAL ŞI METODE

Pentru realizarea studiului de sinteză a literaturii, a fost realizată o căutare avansată în bibliotecile electronice PubMed şi SCOPUS utilizând cuvintele-cheie: „bacteriofagi", „rezistenţa antimicrobiană", „terapia cu fagi" şi „sănătate umană" şi

operatorii BOOLEAN „AND", „OR" și „NOT". Căutarea a generat 911 publicații – 644 în baza de date PubMed și 267 în baza de date SCOPUS. Dinte acestea, 28,21% (n=257) au fost dublicate. Articolele rămase au fost evaluate conform ghidurilor PRISMA (20). La prima etapa de cercetare a publicațiilor au fost verificate 654, din acestea, 74,9% (n=490) nu au corespuns scopului studiului. Testului de eligibilitate au fost supuse 164 de lucrări științifice, conform criteriilor de includere după cum urmează:

a) Articole publicate în perioada 2000-2023.
b) Limbile de publicare – engleza și/sau româna.
c) Publicații în acces deschis.
d) Articole de sinteză a literaturii, articole de cercetare, metaanaliză.

Nu au fost incluse în cercetare articolele găsite publicate în limbile rusă, franceză, poloneză și chineză; articolele publicate între 1980 și 1999; scrisorile editoriale și corecțiile articolelor, precum și publicațiile cu acces restricționat (fig. 1). Pentru studiul final au fost selectate 40 de publicații.

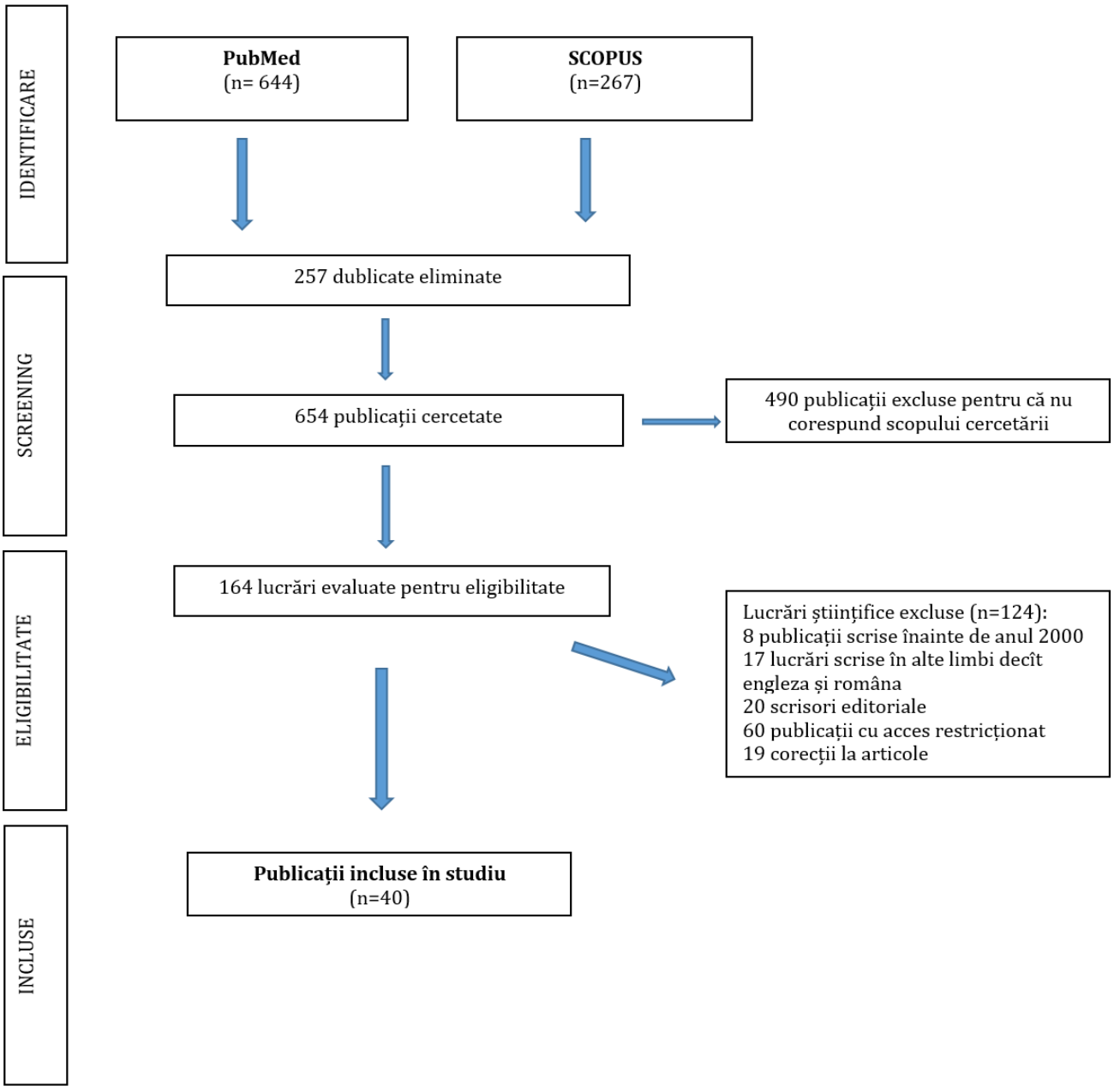

Figura 1. Criteriile de selecție a publicațiilor confom Ghidului PRISMA 2020.

REZULTATE

Analiza SWOT a terapiei cu bacteriofagi ca metodă de combatere a rezistenței la antimicrobiene

A. Puncte forte

În cadrul cercetării publicațiilor selectate au fost identificate 13 puncte forte. Toate studiile au enunțat drept atu al terapiei cu bacteriofagi specificitatea înaltă a fagilor, care determină și alte puncte forte. Qadri H. et al. susțin că activitatea țintită asupra bacteriilor rezistente ar fi principalul argument în favoarea utilizării lor pentru că ar avea efect bactericid doar asupra agentului cauzal., fără a induce modificări ale microbiotei normale. De aceeași părere a fost și Mousavi, accentuând că anume această proprietate îi face siguri în utilizare și pentru pacienții cu malabsorbție, malnutriție și alte maladii ale tractului gastrointestinal (4, 9, 19, 21-28). Al doilea cel mai frecvent punct forte identificat a fost toxicitatea scăzută. În susținerea acesui punct de vedere, autorii articolelor au citat concluzia raportului elaborat în 2020 de Grupul de Inițiativă pentru Rezistența la Antimicrobiene al Institutului Național pentru Alergii și Boli Infecțioase din SUA. De altfel, unii autori au considerat că toxicitatea minimă a fagilor se datorează compoziției acestora – materie organică care este ușor descompusă și asimilată, și nu are efecte negative asupra organelor cu funcție de detoxifiere (4, 11, 19, 23, 29).

Un alt punct forte important este capacitatea lor de a penetra și distruge biofilmele bacteriene. Formarea biofilmului este una din formele prin care agenții patogeni obțin rezistență față de factorii de mediu și agenții antimicrobieni care nu penetrează eficient clusterul bacterian. Schooley et al., Aranaga și coautorii au argumentat ideea de eficiență a fagilor contra biofilmelor prin capacitatea virusurilor de a produce enzime hidrolitice de tipul hidrolizinelor și depolimerazelor care pot distruge matricea biofilmului (11, 19, 23, 26, 28). Câteva din punctele forte se determină reciproc: bacteriofagii sunt microorganisme ubicuitare prezente permanent în orice mediu și rezistente la condițiile de mediu, de asemenea, sunt ușor de găsit și de izolat din probele biologice și de mediu (apă, sol, aer). Aceste proprietăți determină costeficiența înaltă a terapiei cu fagi (4, 9, 10, 17, 21, 22, 29).

O altă pereche de puncte forte identificate care se autodetermină au fost: capacitatea de autoreproducere și necesitatea unei singure doze pentru a asigura eficiența tratamentului. În 12,5% din studii, capacitatea de autodozare a bacteriofagilor a fost evidențiată ca punct forte în susținerea terapiei cu bacteriofagi contra RAM. Acest punct forte a fost argumentat din două poziții: (a) multiplicarea bacteriofagilor în organismul recipient reduce necesitatea unei doze repetate aproape la zero pentru că pacientul răspunde rapid la tratament (conform observațiilor lui d'Herelle de la începutul sec. XX și altor studii clinice din perioada postbelică și (b) o singură administare reduce efectul toxic generalizat asupra organismului și riscul dezvoltării rezistenței (5, 19, 22, 23, 29, 30). Alte două puncte forte identificate sunt virulența minimă a bacteriofagilor și lipsa efectelor adverse. Studiul lui Qadri et al. a considerat virulența minimă un punct forte în legătură cu incapacitatea acestor virusuri să provoace o maladie de etiologie infecțioasă sau alte efecte adverse la oameni, animale sau alte organisme vii pe care le infectează (cu excepția bacteriilor-țintă). În baza studiilor efectuate, autorii au concluzionat că terapia cu fagi nu provoacă efecte adverse asupra macroorganismului, deci este sigură pentru utilizare la scară largă (11, 22, 26).

B. Puncte slabe

În urma analizei literaturii incluse în studiu, au fost determinate opt puncte slabe ale terapiei cu fagi. Cinci dintre acestea au fost dependente de proprietățile și comportamentul fagilor *in vivo*, două au fost determinate de politicile în sănătate, iar unul – de condițiile necesare pentru producerea fagilor pentru terapie. Printre cel mai des identificate puncte slabe ale terapiei cu bacteriofagi (în 17,5% din publicații) a fost imunogenitatea acestor virusuri. Șase din cele șapte studii au determinat că răspunsul imun, provocat la pătrunderea bacteriofagilor în organism, reduce doza infectantă și scade efectul lor bactericid până a ajunge la locul infecției. Brüssow H. a avut un alt argument, bazat pe memoria imunologică a celulelor care poate împiedica administrarea repetată a aceluiași bacteriofag la un pacient cu infecție reemergentă (9, 19, 21-23, 30, 31). Mutațiile spontane au fost considerate un punct slab din considerentul că bacteriofagul litic ar putea deveni temperat, astfel răspândind RAM în loc să o

combată. Deşi rata de apariţie a mutaţiilor spontane în genomul bacteriofagului este de 1000 de ori mai scăzută decât în genomul bacterian, 10% din articolele analizate consideră mutaţiile spontane un punct slab (10, 24, 25, 32).

Autorii au identificat şi alte proprietăţi ale bacteriofagilor care pot fi catalogate drept puncte slabe: sensibilitatea bacteriofagilor faţă de razele UV, de temperaturile înalte şi de aciditatea sucului gastric au fost considerate de Adhikari et al şi Cesta et al principalele neajunsuri care scad eficienţa bacteriofagilor, deoarece scad concentraţia lor din doza administrată (2, 33). Hibstu şi Furfaro au considerat necesitatea unui contact direct cu bacteria-ţintă un punct slab pentru că, în opinia lor, alte căi de administrare decât topic sau prin acces direct ar limita eficacitatea terapiei din cauza micşorării concentraţiei de fagi şi extinderea timpului necesar ca bacteriofagii să ajungă la locul infecţiei (21, 34). Rahbarnia et al şi Hatfull et al au considerat drept punct slab incapacitatea fagilor de a elimina agenţii patogeni bacterieni strict intracelulari, reieşind din proprietatea fagilor de a nu afecta celulele şi ţesuturile umane (23, 35).

Politicile şi regulamentele restrictive din domeniul sănătăţii reprezintă un alt punct slab atribuit terapiei cu bacteriofagi. Conform autorilor, există mai multe obstacole în aprobarea de către organismele internaţionale din domeniu precum Food and Drug Administration (FDA, SUA) sau Agenţia Europeană a Medicamentului (EMA, UE) a terapiei cu bacteriofagi pentru combaterea bacteriilor rezistente la antimicrobiene. Printre acestea pot fi enumerate: (a) bacteriofagii sunt organisme vii, cu o structură chimică şi biologică instabilă, deci trebuie tratate ca GMO, produs biologic pentru utilizare medicală sau produs pentru terapie medicală avansată; (b) lipsesc date concludente, obţinute din studiile clinice controlate, asupra siguranţei şi eficacităţii lor la utilizarea în tratamentul pacienţilor umani; (c) imposibilitatea de înregistrare a dreptului de proprietate intelectuală asupra bacteriofagilor care sunt ubicuitari şi apar în mod natural în mediu (chiar şi mutanţi) (7, 10, 16, 21, 30, 34, 36, 37). Lipsa unui cadru legal standardizat, cel puţin la nivel regional, determină secundar accesibilitatea scăzută a pacienţilor la terapia cu bacteriofagi. Chiar dacă terapia are o istorie veche şi este bine reglementată în diverse ţări precum Polonia (începînd cu 1996), Georgia, Federaţia Rusă şi China (începînd cu 1958), pacienţii din alte ţări (Belgia, Canada, Japonia etc.) pot doar recurge la turism medical sau tratament experimental cu bacteriofagi pentru a se vindeca (36, 37, 38).

C. Oportunităţi

În cadrul acestui studiu am determinat nouă oprtunităţi de utilizare a terapiei cu bacteriofagi în combaterea RAM. Şase studii (15%) au expus argumente în favoarea posibilităţii de utilizare a lizogenităţii bacteriofagilor ca mecanism şi instrument pentru restabilirea sensibilităţii bacteriilor rezistente la antimicrobiene prin incorporarea genelor care să modifice structura capsulei bacteriene sau funcţia pompelor de eflux (10, 19, 21, 28, 39, 40). Studiile lui Schooley RT şi Suh GA. et al menţionează că proprietatea bacteriofagilor de a penetra biofilmele bacteriene poate fi utilizată şi în scop profilactic dacă dispozitivele implantabile (catetere, implanturi dentare, şuruburi şi tije) ar fi prelucrate cu o soluţie cu concentraţie înaltă de bacteriofagi (11, 28). Posibilităţile medicinei moderne precum ingineria genetică, secvenţierea întregului genom ar putea permite utilizarea produselor biologice ale bacteriofagilor (enzime, proteine) şi a bacteriofagilor ca agenţi antimicrobieni contra bacteriilor MDR şi PDR, contra unui spectru mai larg de agenţi patogeni şi ca vehicul sau matrice în producerea vaccinurilor (5, 10, 18, 21, 33).

Tendinţele medicinei moderne implică orientarea spre medicina personalizată şi integrarea intervenţiilor din sănătatea umană în conceptul „O singură sănătate". Publicaţiile cercetate au arătat că terapia cu bacteriofagi poate fi utilizată în aceste direcţii prin intermediul ingineriei genetice atât în medicina umană, cât şi veterinară. Şase studii (15%) au susţinut utilizarea bacteriofagilor ca metodă de combatere a tulpinilor bacteriene MDR şi PDR izolate de la pacienţi. Bioingineria şi secvenţierea genomică ar fi, după părerea lor, instrumente de primă importanţă şi necesitate în tratamentul individualizat al pacientului purtător de bacterie rezistentă cu ADN unic, dar este necesară o bibliotecă genomică extinsă (7, 19, 41-44). Sulakvelidze A. a susţinut în lucrarea sa că fagii pot fi folosiţi ca metodă de prevenţie sau de tratament al afecţiunilor tractului gastrointestinal apărute ca rezultat al antibioterapiei (16). Autorii a 12,5% din publicaţiile incluse în studiu (n=5) s-au pronunţat în favoarea utilizării terapiei cu bacteriofagi pentru profilaxia bolilor în domeniile zooteh

nie, acvacultură cu înlocuirea parţială sau integrală a antibioticelor, susţinând că bacteriofagii au sau ar avea în perspectivă capacitatea să reducă rata infecţiilor de origine zoonotică la oameni (5, 32, 42, 43, 44). Pe lângă aceste oportunităţi pe care le prezintă terapia cu bacteriofagi pentru sănătatea umană în vederea combaterii RAM, autorul unui articol a considerat drept oportunitate şi potenţialul terapiei cu bacteriofagi de a revitala industria farmaceutică, sectorul de bioinginerie şi cel de afaceri. Poziţia a fost argumentată prin cost-eficienţa izolării şi dezvoltării de noi fagi pentru terapie şi rambursarea rapidă a investiţiilor în caz de comercializare a produselor finite (38).

D. Ameninţări

Au fost identificate şapte ameninţări care ar putea diminua eficienţa terapiei cu bacteriofagi. Transducţia genică mediată de bacteriofagi a fost menţionată drept cel mai mare obstacol (12,5% din studii) în utilizarea terapiei cu bacteriofagi. Această proprietate a virusurilor de a media transferul genelor ce codifică rezistenţa la antimicrobiene între două bacterii este motivul principal al întârzierii aprobării de către organizaţiile internaţionale de profil a terapiei cu fagi pentru combaterea RAM (11, 21, 24, 25, 45). Pe locul secund s-a clasat posibilitatea bacteriilor să dezvolte rezistenţă la fagi datorită adaptabilităţii rapide a acestora la condiţiile nefavorabile de mediu. Această ameninţare a fost determinată în 10% din studii (n=4) (11, 19, 26, 32).

Al treilea obstacol identificat a fost lizogenia, proprietatea bacteriofagilor de a-şi integra materialul genetic în genomul bacterian fără a induce liza bacteriei. Autorii celor trei studii care au înaintat acest obstacol drept o provocare la adresa siguranţei terapiei cu fagi pentru pacienţi l-au argumentat prin faptul că materialul genetic viral poate altera ADN-ul bacteriei şi induce hiperexprimarea factorilor de virulenţă, care, combinat cu RAM, devine o dublă ameninţare pentru viaţa pacientului (21, 25, 45). La celălalt capăt, bacteriofagii litici cu eficacitate înaltă au fost consideraţi la fel de periculoşi din cauza dezvoltării şocului septic la pacienţi când detritul, produsele de metabolism şi toxinele bacteriene ajung în circuitul sangvin în cantităţi ce depăşesc norma admisă de protocoalele de tratament în vigoare (21, 34).

Alte două ameninţări identificate au fost dilemele bioetice legate de tratamentul pacienţilor cu organisme vii, posibil modificate genetic şi înalt mutagene, având în vedere că tratamentul administrat, chiar şi experimental (în cadrul studiilor clinice) nu trebuie să dăuneze, iar pacientul are dreptul la autodeterminare dacă terapia este una experimentală (7) şi perspectiva unui cost ridicat pentru tratamentul cu bacteriofagi dacă în costul pentru terapia personalizată vor fi incluse cheltuielile pentru producerea fagilor individualizaţi, e pentru spitalizare, pentru transportul soluţiei de fagi la patul pacientului (11) (fig. 2).

DISCUŢII

Analiza SWOT realizată pentru evaluarea terapiei cu bacteriofagi ca metodă de combatere a RAM a dat rezultate promiţătoare. Punctele forte prevalează comparativ cu punctele slabe, cu un raport de 13:8, şi există de 1,28 ori mai multe oportunităţi decât ameninţări în perspectiva implementării terapiei cu bacteriofagi ca metodă de tratament. Unele puncte slabe sau obstacole listate de unii autori, sunt considerate oportunităţi în lumina inovaţiilor în medicina moleculară şi bioinginerie sau avantaje importante comparativ cu antibioterapia.

Cu precădere, capacitatea bacteriofagilor de a muta spontan şi de a atenua sau de a pierde proprietăţile litice au fost considerate de patru grupuri de autori drept un punct slab (10, 24, 25, 32), lizogenia şi probabilitatea transducţiei genice – ameninţări în cinci publicaţii (11, 21, 24, 25, 45), de cealaltă parte, 22,5% din studii consideră aceste proprietăţi ale fagilor drept oportunităţi în contextul evoluţiei fulminante a RAM (5, 10, 18, 19, 21, 28, 33, 39, 40). Saha D. şi coautorii consideră mutagenitatea fagilor un atu important în depăşirea rezistenţei bacteriei-ţintă faţă de bacteriofagul specific, ceea ce minimizează pericolul pe care îl prezintă apariţia rezistenţei faţă de bacteriofagi în timpul tratamentului (29).

Lipsa cadrului legal standardizat la nivel regional, mai ales în spaţiul european, aduce unele dificultăţi în privinţa popularizării şi implementării terapiei cu bacteriofagi în practica medicală. Este necesară realizarea unui studiu extins, multicentric şi multidisciplinar care să cuprindă analiza comportamentului fagilor *in vitro*, în mediu, *in vivo* (pe modele animale) şi în practica clinică. Astfel, ar fi depăşit blocajul aprobării şi adoptării terapiei cu fagi ca metodă de tratament şi de combatere a fenomenului RAM. Studiile clinice con-

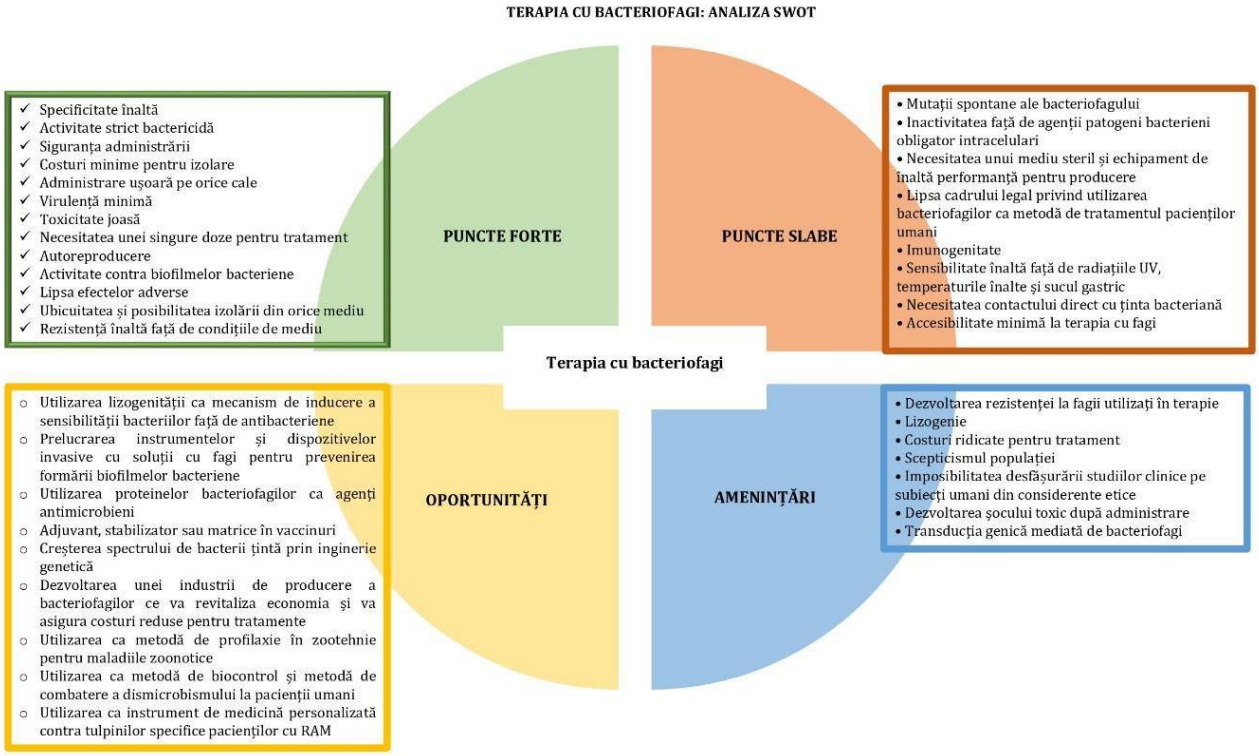

Figura 2. Terapia cu bacteriofagi prin prisma metodei de analiza SWOT.

trolate, considerate de EMA standardul de aur și unicul argument acceptabil la înregistrarea unei noi metode de tratament sau a unui nou preparat, ar fi una din metodele de cercetare ale aspectelor enumerate.

Caracteristicile fagilor precum ubicuitatea, toxicitatea redusă, specificitatea înaltă, neafectarea altor celule decât bacteriile-țintă, capacitatea de dezintegrarea a biofilmelor bacteriene sunt argumente plauzibile în favoarea implementării terapiei cu bacteriofagi pe scară largă. Dilemele bioetice privind siguranța acestor virusuri pentru sănătatea umană se reduc considerabil dacă tratăm terapia cu bacteriofagi drept analog al administrării unui vaccin viu atenuat cum ar fi vaccinul BCG contra tuberculozei, vaccinul ROR sau Polio care sunt considerate sigure pentru utilizare. Acest punct de vedere a fost susținut și de Anomaly J. în publicația sa (46).

Așadar, terapia cu bacteriofagi este o alternativă plauzibilă pentru a suplini sau a înlocui terapia cu antimicrobiene în tratarea pacienților cu infecții cauzate de agenți patogeni MDR sau PDR (9, 23, 30, 33).

CONCLUZII

1. Rezistența la antimicrobiene este o problemă de sănătate publică care necesită intervenții rapide și eficiente. Terapia cu bacteriofagi este o alternativă plauzibilă care și-a demonstrat eficacitatea în cei peste 100 de ani de evoluție.
2. Punctele forte și oportunitățile care caracterizează terapia cu bacteriofagi depășesc riscurile utilizării acestei metode ca instrument de combatere a fenomenului RAM, cu un raport puncte forte/puncte slabe de 13:8 și oportunități/amenințări de 9:7.
3. Inovațiile în medicina moleculară și în ingineria genică transformă dezavantajele bacteriofagilor precum mutagenitatea înaltă, lizogenia și capacitatea de transducție genică în oportunități pentru dezvoltarea terapiei personalizate pentru combaterea RAM.
4. Autoritățile din domeniul sănătății publice ar trebui să promoveze și să întreprindă activități de cercetare în direcția implementării terapiei cu bacteriofagi atât în sănătatea umană, cât și în contextul abordării „O singură sănătate".

CONFLICT DE INTERESE

Autorii declară lipsa conflictelor de interese în ceea ce priveşte studiul şi acest articol.

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CUNOȘTINȚE, ATITUDINI ȘI PRACTICI PRIVIND REZISTENȚA LA ANTIMICROBIENE ÎN ȚĂRILE CU VENIT MIC ȘI MEDIU: SINTEZĂ NARATIVĂ

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Keywords: *knowledge, attitudes, practices; antibiotic resistance; low- and middle-income countries.*

KNOWLEDGE, ATTITUDES AND PRACTICES REGARDING ANTIMICROBIAL RESISTANCE IN LOW- AND MIDDLE-INCOME COUNTRIES - NARRATIVE SYNTHESIS

Introduction*. Antimicrobial resistance (AMR) is an acute medico-social problem, and in order to minimize its effects, a comprehensive approach is needed at the individual level and in the formulation of public health policies.*

Material and methods. *A narrative synthesis of KAP studies results on AMR in LMICs was conducted. The search for sources was performed in public databases. After identifying articles and removing duplicates, 23 relevant ones were selected, of which 14 were fully analyzed.*

Results. *Studies have indicated that knowledge is generally deficient in LMICs. Negative or neutral attitudes and practices concerning antibiotic prescribing and administration prevail, with widespread self-medication. Determining factors are diverse, and addressing the AMR phenomenon needs a comprehensive approach, including policy-level initiatives aimed ad raising awareness and educating the population.*

Conclusions. *The determinants of population KAP in LMICs are influenced by the economic situation and the health system. Clear objectives, correct sampling techniques, and standardization of research instruments are necessary for KAP studies to ensure the representativeness of the data.*

Cuvinte-cheie: *cunoștințe, atitudini, practici; rezistența la antibiotice; țări cu venit mic și mediu.*

Introducere*. Rezistența la antimicrobiene (RAM) este o problemă medico-socială stringentă, iar pentru minimizarea efectelor acesteia este necesară o abordare comprehensivă, la nivel individual, și formarea politicilor de sănătate publică.*

Material și metode. *A fost realizată o sinteză narativă a rezultatelor studiilor CAP (Cunoștințe, Atitudini și Practici) privind RAM în țările cu venit mic și mediu. Sursele au fost căutate în bazele de date publice. După identificarea articolelor și dedublare, au fost selectate 23 de surse relevante, dintre care 14 au fost analizate integral.*

Rezultate. *Studiile au arătat că în țările cu venit mic și mediu, cunoștințele cu privire la prescrierea și administrarea antibioticelor sunt, în general, slabe, atitudinile negative sau neutre, iar practicile necorespunzătoare, fiind pe larg răspândită automedicația. Creșterea nivelului de conștientizare și educarea populației cu privire la fenomenul RAM necesită o abordare complexă, inclusiv la nivel de politici.*

Concluzii. *Determinantele CAP ale populației în țările cu venit mic și mediu sunt influențate de situația economică și de sistemul de sănătate. Pentru studiul CAP, care asigură reprezentativitatea datelor, sunt necesare obiective clare, tehnici de eșantionare corecte și standardizarea instrumentelor de cercetare.*

INTRODUCERE

Rezistența la antimicrobiene (RAM) este o amenințare globală pentru sănătate, cu impact semnificativ asupra sistemului de sănătate și a economiei (1). Utilizarea abuzivă și excesivă a antimicrobienelor rămâne principala cauză a apariției agenților patogeni rezistenți la acțiunea acestora. Identificarea lacunelor în cunoștințe și planificarea intervențiilor care ar putea duce la utilizarea rațională a antimicrobienelor sunt de o importanță decisivă pentru a reduce RAM (2). Minimizarea efectului rezistenței la antimicrobiene necesită un răspuns politic adecvat, care se bazează pe o bună guvernare și coordonare (3). Factorii sociali și comportamentali ai utilizării inadecvate a antibioticelor au fost identificați ca unul dintre factorii-cheie care contribuie la apariția RAM. Sondajele privind cunoștințele, atitudinile și practicile populației (CAP) fac parte din cadrul de monitorizare și de evaluare propus de Planul global de acțiune al OMS privind rezistența la antimicrobiene (4).

Scopul lucrării a fost identificarea caracteristicilor studiilor despre cunoștințele, atitudinile și practicile RAM, determinantele CAP ale populației privind fenomenul RAM în țările cu venit mic și mediu (LMIC).

MATERIAL ȘI METODE

A fost realizată o sinteză narativă care prezintă rezultatele a 14 studii CAP ale populației privind rezistența la antimicrobiene din cadrul țărilor cu venit mic și mediu (LMIC). Căutarea surselor de informații a fost realizată în bazele de date *PubMed*, *Google Scholar* și *Hinari* cu ajutorul următoarelor cuvinte-cheie: „antimicrobial resistance AND kap", „antimicrobial resistance NEAR knowledge AND attitude AND practice", „antimicrobial resistance NEAR knowledge attitude practice".

Criteriile de includere a publicațiilor în studiu au fost: articol publicat în perioada ianuarie 2015 – ianuarie 2023; tipul de studiu – reviuri narative, studii descriptive și observaționale despre CAP al populației privind rezistența la antimicrobiene, metodologie clară și explicită; articole scrise în limba engleză.

Criteriile de excludere a publicațiilor din studiu au fost: studii descriptive privind CAP ale studenților facultăților de medicină, lucrătorilor din domeniul medical sau veterinar, articolele care nu au metodologie sau aceasta este prezentată neexplicit.

Textul integral al studiilor a fost evaluat în baza variabilelor: scopul, metodologia, numărul cumulat de participanți/eșantionul și rezultatele.

Din informația colectată, au fost selectate evidențele privind cunoștințele, atitudinile și practicile populației în legătură cu rezistența la antimicrobiene care au fost interpretate și prezentate luând în considerare numărul, nivelul de evidență și calitatea studiilor. În *Google Academics* au fost identificate în total 1 760 de citări, în baza de date *PubMed* – 511, în *HINARI* – 523. După eliminarea înregistrărilor duplicate și screeningul abstractelor, au fost selectate 23 de articole, dintre acestea au fost excluse nouă care nu se refereau la țările cu venit mic și mediu, fiind revizuite integral 14 articole.

Studiul este realizat în cadrul Proiectului de cercetare „Phage treatment and wetland technology as intervention strategy to prevent dissemination of antibiotic resistance in surface waters (PhageLand)" cifrul 22.80013.8007.1M și a fost aprobat la Comitetul de Etică a Cercetării al Universității de Stat de Medicină și Farmacie „Nicolae Testemițanu", proces-verbal nr. 7 din 9.01.2022 (5).

REZULTATE

După screeningul textului integral, au fost incluse în reviul integrativ 14 articole enumerate în tabelul 1, cu un număr cumulat de participanți la studiu de 9380 de persoane din populația generală.

Toate cele 14 studii incluse în analiză au avut obiective clare privind evaluarea nivelului de cunoștințe, atitudinii și practicii populației în legătură cu rezistența la antimicrobiene și consumul lor. Au fost folosite metodologii adecvate pentru studii transversale bazate pe populație în raport cu întrebările cercetate.

Studiile au demonstrat că în majoritatea țărilor LMIC populația are cunoștințe slabe privind utilizarea antibioticelor (6, 7, 8, 12, 13, 14, 16, 18, 19). În unele țări totuși există o tendință de îmbunătățire a cunoștințelor în acest domeniu datorită diferitor practici de creștere a gradului de conștientizare și de educare a populației (9, 11, 15, 17), îmbunătățirea factorilor socio-culturali (10).

Tabelul 1. Studii Cunoștințe, Atitudini și Practici ale populației cu privire la fenomenul de rezistență la antimicrobiene în țările cu venit mic și mediu.

Autorul	Țara	Anul	Scopul	Tipul studiulu	Criteriile de eligibilitate	Eșantion	Sursa
Sa'ed H. Zyoud	Palestina	2012	Analiza cunoştinţelor, atitudinilor şi practicilor privind utilizarea antibioticelor în managementul infecţiilor tractului urinar la copii pe un eşantion mare de părinţi	Studiu transversal	Studiu CAP organizat pe părinţii copiilor cu infecţii ale tractului respirator superior cu vârsta cuprinsă între 18 şi 50 de ani.	385 părinţi	(6)
Grace-Ange Elong Ekambi et. al.	Cameroon	2015	Acest studiu a fost conceput pentru a determina cunoştinţele; atitudinile şi practicile faţă de utilizarea antibioticelor într-o comunitate urbană şi evaluează factorii asociaţi cu utilizarea antibioticelor	Studiu transversal şi prospectiv	Studiu CAP organizat pentru comunitatea urbană, clienţii farmaciilor, vârsta respondenţilor >15 ani	1192 respondenţi	(7)
Inocêncio Mate et. al.	Maputo, Mozambic	2016	Obţinerea datelor despre cunoştinţele, atitudinile şi practicile privind prescrierea şi utilizarea antibioticelor în Africa sub-sahariană	Studiu transversal	Sudiu CAP pe populaţie adultă >18 ani, chestionar semistructurat	1019 participanţi	(8)
Komal Raj Rijal	Nepal	2017	Explorarea cunoştinţelor, atitudinea şi practicile de utilizare a antibioticelor în rândul pacienţilor, lucrătorilor din domeniul sănătăţii, laboratoarelor, vânzătorilor de medicamente şi fermierilor din Nepal	Studiu transversal	Sondaj faţă în faţă privind CAP, inclusiv pe pacienţi cu vârsta preponderent (50%) între 26–50 de ani	516 participanţi, dintre care 324 pacienţi	(9)
Moyukh Chowdhury et. al.	Matlab, Banglad esh	2017 - 2019	Explorarea factorilor şi a practicilor privind accesul şi utilizarea antibioticelor, înţelegerea rezistenţei antimicrobiene în comunităţile rurale din Bangladesh din perspectivă socio-culturală	Studiu calitativ, faza a doua a unui studiu multi-statal ABACUS	Studiu CAP organizat pe locuitorii din diferite categorii de vârstă din zone rurale, vârsta 16-60 de ani	59 informatori	(10)
Jane Mingjie Lim et. al.	Cambod gia	2018	Evaluarea cunoştinţelor, atitudinilor şi practicilor publice cu privire la antibiotice şi rezistenţa la antibiotice în Cambodgia, oferind informaţii de bază în funcţie de care va fi monitorizat progresul intervenţiilor viitoare	Studiu descriptiv	Sondaj CAP legat de utilizarea antibioticelor de către populaţia urbană şi rurală din trei provincii cambodgiene: Phnom Penh, Siem Reap şi Prey Veng. Vârsta > 18 ani	2005 participanţi	(11)
Hassan Waseem et. al.	Sialkot, Pakistan	2018 - 2019	Evaluarea cunoştinţelor despre rezistenţa antimicrobiană şi a atitudinii faţă de acest fenomen în rândul membrilor comunităţii, farmaciştilor/proprietari de farmacii şi medicilor (Sialkot, Pakistan)	Studiu transversal	Studiu CAP bazat pe trei chestionare autoadministrate, destinate populaţiei gene rale cu vârsta cuprinsă între 18 şi 45 de ani, farmaciştilor şi medicilor	473 participanţi din po pulaţia generală	(12)

Amit Khelgi et. al.	Mangaluru, India	2019	Evaluarea cunoştinţelor privind utilizarea antibioticelor şi rezistenţei antimicrobiene în rândul populaţiei rurale din Mangaluru. Determinarea atitudinilor oamenilor din mediul rural privind utilizarea antibioticelor. Analiza practicilor acestora în utilizarea antibioticelor	Studiu transversal	Studiu CAP bazat pe chestionar, destinat populaţiei generale din mediul rural, vârsta 18-60 de ani	130 participanţi	(13)
Emelda E. Chukwu	Nigeria	2020	Evaluarea nivelului actual de conştientizare şi de cunoaştere a RAM de către publicul nigerian (Nord Central, Nord-Est, Nord-Vest, Sud-Est, Sud-Sud şi Sud-Vest)	Studiu transversal	Studiu CAP, Chestionarul pretestat şi validat folosit pentru a obţine informaţii de la publicul larg în şase zone geopolitice, vârsta 18–≥75 de ani	482 participanţi	(14)
Calvin Sindato et. al.	Tanzania	2020	Determinarea cunoştinţelor, atitudinilor şi practicilor (CAP) cu privire la utilizarea antimicrobienelor şi RAM	Studiu transversal	Studiu CAP, Chestionar semistructurat (Ilala, Kilosa şi Kibaha, Tanzania), vârsta 29-54 de ani	828 participanţi	(15)
Wudneh Simegn, Getachew Moges	Dessie, Etiopia de Nord	2021	Evaluarea nivelului de conştientizare şi de cunoaştere a rezistenţei antimicrobiene şi a factorilor asociaţi în rândul adulţilor din Dessie, Etiopia	Studiu transversal	Studiu CAP bazat pe comunitate cu implicarea persoanelor adulte	407 adulţi participanţi	(16)
Matru-jyoti Pattnaik, Ashish Kumar Nayak, et. al.	Odisha, India	2021	Determinarea cunoştinţelor, atitudinilor şi practicilor (CAP) cu privire la utilizarea de antimicrobiene şi AMR în rândul comunităţilor rurale din Tigiria (Odisha), India	Studiu transversal	Studiu CAP bazat pe chestionar standardizat, folosind un dispozitiv electronic cu Open Data Kit., vârsta participanţilor 18 - > 60 de ani	1003 participanţi	(17)
Haileyesus Dejene	Gondar, Etiopia de Nord	2021	Evaluarea cunoştinţelor, atitudinilor şi practicilor locuitorilor oraşului Gondar cu privire la utilizarea şi rezistenţa la antimicrobiene	Studiu transversal	Studiu CAP folosind un chestionar semistructurat pretestat, (ora;ul Gondar)	400 participanţi	(18)
Zahraa N. Fakhreldain, Hayder CH. Assad	Iraq	2021 - 2022	Colectarea datelor despre nivelul cunoştinţelor atitudinea şi utilizarea antibioticelor, în comunităţi irakiene şi explorarea factorilor predictivi ai pacientului asociaţi cu acestea	Studiu observaţional transversal	Studiu CAP bazat pe chestionar autoadministrat validat, pretestat. (oraşul Al-ALSadar)	475 pacienţi	(19)

În ţările LMIC, pe lângă cunoştinţele reduse ale populaţiei cu privire la administrarea corectă a antibioticelor, persistă o atitudine negativă faţă de acest fenomen (6, 12, 13, 14, 16, 18) sau neutră (19). În alte ţări, pe lângă nivelul mai înalt de cunoştinţe, se observă şi o atitudine conştientă, astfel populaţia cunoaşte unii termeni referitori la antimicrobiene şi rezistenţă la antibiotice. Majoritatea participanţilor au recunoscut că rezistenţa la antibiotice este o problemă (9, 11, 15, 17). Practicile populaţiei în legătură cu rezistenţa la antibiotice sunt inadecvate (6, 7, 8, 12, 13, 14, 16) acolo unde este foarte răspândită automedicaţia şi prescrierea inadecvată a antimicrobienelor (7, 8, 14, 16), unde lipseşte accesul la facilităţi de asistenţă medicală (10). În general, practicile privind administrarea antibioticelor sunt corelate cu nivelul de cunoştinţe al populaţiei. Astfel, în unele cazuri s-au identificat practici corespunzătoare la populaţia anchetată, dar care totuşi necesită îmbunătăţire (9, 11, 15, 17, 19). Sunt binevenite unele campanii de conştientizare pentru a atenua utilizarea inadecvată a antibioticelor (9), precum şi politicile referitoare la utilizarea corespunzătoare a antibioticelor, interzicerea comercializării acestora fără prescripţie şi abordarea *One Health* (9, 10, 16).

DISCUŢII

În urma analizei publicaţiilor ştiinţifice au fost identificaţi determinanţii principalelor compartimente. Nivelul redus de cunoaştere de către populaţie a fenomenului RAM în LMIC este influenţat, în primul rând, de nivelul scăzut de educaţie al populaţiei (8) şi de accesul limitat la informaţii medicale (9), inclusiv cu privire la antibiotice şi la administrarea corectă a acestora. Atitudinile negative ale populaţiei privind fenomenul RAM sunt, în mare parte, agravate de nivelul redus de trai (13) şi de atitudinea indiferentă faţă de problemele stringente din sănătatea publică (15, 18), lipsa de încredere în sistemul de sănătate (10), comunicare defectuoasă între medici şi pacienţi. Practicile slabe ale populaţiei privind fenomenul RAM sunt determinate, în mare parte, de sărăcie şi de situaţia economică precară, de reutilizarea antibioticelor rămase (6, 14) şi de automedicaţie (7, 9, 13, 17). Principalele motive invocate pentru practicarea automedicaţiei în ţările în curs de dezvoltare sunt lipsa accesului la asistenţă medicală aproape de populaţie, lipsa surselor financiare, ignoranţa (7) şi, cel mai important, lipsa sau controlul redus din partea autorităţilor cu privire la consumul de antibiotice, ceea ce duce inevitabil la agravarea situaţiei privind RAM (9, 10, 12).

Studiile CAP pot ajuta factorii de decizii să planifice acţiuni şi strategii de combatere a RAM şi de atenuare a efectelor acestui fenomen (20). Pentru a maximiza efectele utilizării studiilor CAP este necesar suportul programelor de sănătate publică pentru a face progrese în rezolvarea problemelor RAM (21). Creşterea nivelului de conştientizare şi educarea populaţiei cu privire la rezistenţa la antibiotice are un rol important în prevenirea răspândirii acestui fenomen (22).

CONCLUZII

1. Această sinteză a identificat câteva caracteristici ale studiilor privind cunoştinţele, atitudinile şi practicile privind rezistenţa la antimicrobiene şi consumul acestora.
2. Sunt necesare obiective foarte clare ale sondajului/studiului.
3. Pentru asigurarea reprezentativităţii datelor obţinute sunt necesare tehnici de eşantionare corespunzătoare şi corecte din punct de vedere ştiinţific.
4. Pentru a obţine date credibile şi comparabile sunt necesare teste de probă ale sondajelor şi metode de implementare a chestionarelor, precum şi standardizarea instrumentelor de cercetare.
5. În timpul elaborării chestionarului, trebuie luate în considerare caracteristicile sistemelor de sănătate, accesul la serviciile de sănătate.
6. Studiile CAP sunt un instrument de valoare mai ales dacă este asigurat suportul programelor de sănătate publică în scopul elaborării măsurilor de reducere a fenomenului de RAM.

CONFLICT DE INTERESE

Autorii declară lipsa conflictului de interese.

MULŢUMIRI ŞI FINANŢARE

Studiul este realizat în cadrul Proiectului de cercetare „Phage treatment and wetland technology as intervention strategy to prevent dissemination

of antibiotic resistance in surface waters (*PhageLand*)" cifrul 22.80013.8007.1M.

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MAPPING THE ANTIBIOTIC CONSUMPTION IN THE REPUBLIC OF MOLDOVA

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Keywords: *mapping, antibiotic consumption, DDD, WHO, hospitals.*

Introduction*. Antibiotic consumption represents a crucial aspect of modern healthcare. Abuse and inappropriate use of antibacterial agents contribute to the phenomenon of bacterial resistance. The aim of the study was to determine antibiotic consumption in hospital conditions and map the results in order to highlight regions in the country with the highest levels of antibiotic consumption and the classes of antibiotics used.*

Material and methods. *Antibiotic consumption in hospitals (64 facilities) over a 4-year period (2018-2021) was determined using the defined daily dose (DDD) as the unit of measurement. The quantity of antibiotics consumed was expressed in DDD/1000 patient-days (hospitalization days), followed by mapping the results.*

Results. *A "very high" level of antibiotic consumption, expressed in DDD/1000 patient-days, was observed in the Falesti district, while a "medium" level of antibacterial agent consumption was observed in the Taraclia, Comrat, Ialoveni, Singerei, Glodeni, and Dondu seni districts. The total consumption per antibiotic class revealed an increased use of Cephalosporins, Imidazole derivatives, Macrolides, Tetracyclines, Fluoroquinolones, and Aminoglycosides.*

Conclusions. *Mapping antibiotic consumption is an efficient tool in highlighting administrative-territorial regions with high antibiotic consumption and identifying the most frequently used antibiotic classes. Providing this information is useful for developing policies aimed at optimizing antibiotic use and minimizing resistance.*

Cuvinte-cheie: *cartografiere, consum de antibiotice, DDD, OMS, spitale.*

CARTOGRAFIEREA CONSUMULUI DE ANTIBIOTICE ÎN REPUBLICA MOLDOVA

Introducere. *Consumul de antibiotice reprezintă un aspect crucial al asistenței medicale moderne. Abuzul și utilizarea necorespunzătoare a preparatelor antibacteriene favorizează fenomenul de rezistență bacteriană. Scopul studiului a fost determinarea consumului de antibiotice în condiții de staționar în Republica Moldova și cartografierea rezultatelor obținute.*

Material și metode. *În acest studiu a fost determinat consumul de antibiotice în condiții de staționar (64 de spitale), pe o perioadă de patru ani (2018-2021), folosind ca unitate de măsură doza zilnică definită (DDD). Cantitatea de antibiotice consumată a fost exprimată în DDD/1000 zile-pacient (zile de spitalizare), ulterior fiind efectuată cartografierea rezultatelor.*

Rezultate. *Rezultatele înregistrate atestă un nivel „foarte mare” de consum de antibiotice exprimat în DDD/1000 zile de spitalizare în raionul Fălești și un nivel „mediu” de consum de preparate antibacteriene în raioanele Taraclia, Comrat, Ialoveni, Sîngerei, Glodeni, Donduşeni. Consumul total per clase de antibiotice a scos în evidență un consum sporit de cefalosporine, de derivați de imidazol, de macrolide, de tetracicline, de fluorochinolone și de aminoglicozide.*

Concluzii. *Cartografierea consumului de antibiotice reprezintă un instrument eficient de evidențiere a regiunilor administrativ-teritoriale în care consumul de antibiotice este ridicat, și a claselor de antibiotice cel mai frecvent utilizate. Aceste informații sunt utile pentru elaborarea de politici menite să optimizeze utilizarea antibioticelor și să minimizeze rezistența acestora.*

INTRODUCTION

Antibiotics are crucial for treating infectious diseases, but bacterial resistance poses a rapidly increasing global threat. A key driver of resistance is antibiotic use (1). Monitoring antibiotic consumption patterns over time and across countries could inform policies aimed at optimizing antibiotic prescribing and minimizing antibiotic resistance. This may involve setting and enforcing per capita consumption targets or supporting investments in alternatives to antibiotics (2).

According to the European Centre for Disease Prevention and Control, hospital consumption represents only 9% of the overall European Union/European Economic Area (EU/EEA) total consumption of antibacterials for systemic use. The continued decline in consumption between 2020 and 2021 is not immediately apparent when hospital and community sectors are combined. The overall EU/EEA population-weighted mean consumption in the hospital sector decreased linearly by 0.04 DDD per 1,000 inhabitants per day between 2016 and 2019. The largest annual decrease occurred in 2020 (0.11 DDD per 1,000 inhabitants per day), followed by a reduction of 0.05 DDD per 1,000 inhabitants per day in 2021. At the country-level, eight countries experienced increases in hospital consumption between 2019 and 2020, and 11 had increases between 2020 and 2021. Five countries (Bulgaria, Croatia, Greece, Portugal, and Slovakia) had higher hospital consumption in 2021 than in 2019. The overall EU/EEA population-weighted proportion of Antimicrobial Medicines Consumption (AMC) from broad spectrum sub-groups also showed consecutive annual increases during the period 2019–2021. The largest annual increase in the past 10 years occurred between 2020 and 2021 (3).

In the Republic of Moldova, antibiotics constitute approximately 10% of the total number of medicines, with a prevalence of antibiotic consumption at 42.7%. National results, based on data regarding the antimicrobial resistance profile of microorganisms involved in systemic infections, indicate a high level of resistance. Isolates of K. pneumoniae show increased resistance to cephalosporins (>96%), fluoroquinolones (>50%), and aminoglycosides (>90%). For *A. baumannii*, the resistance profile to fluoroquinolones is 100%, carbapenems >90%, and aminoglycosides >95%. *E. coli* strains exhibit resistance rates of >80% to penicillins, >65% to cephalosporins, and >55% to fluoroquinolones.

Currently, in the Republic of Moldova, 505 drugs are registered and classified according to the international code - ATC (Anatomical Therapeutic Chemical Classification) as systemic antibacterial agents. According to the Medicines and Medical Devices Agency (AMDM), there has been a constant increase in the number of antibiotics used in the last three years. In 2021, over 70 million packages were imported, representing a 10% increase compared to previous years, or over 20 boxes per capita within a year, which translates to almost two boxes per person per month (4, 5).

Therefore, there is a need to enhance the national surveillance system for antibacterial resistance and monitoring of antibiotic consumption both in hospital and outpatient settings. Studies on antibiotic consumption in hospital conditions in the Republic of Moldova are limited, which is why determining and mapping antibiotic consumption are crucial steps in preventing antibacterial resistance and promoting the rational use of antibiotics.

The aim of the study was to determine the consumption of antibiotics in hospitals in the Republic of Moldova and to map the results in order to highlight which regions of the country exhibit the highest levels of antibiotic consumption and the classes of antibiotics used.

MATERIAL AND METHODS

A comprehensive descriptive study was conducted to determine the consumption of antibacterial agents based on the geographical location of Public Health Institutions, Hospitals and the type of antibiotic delivered to IMSP.

The research utilized reports on the execution of public procurement contracts from the years 2018-2021, along with the number of hospitalization days of the healthcare units included in the study during the analyzed period.

The determination of antibacterial agent consumption was carried out in 64 IMSPs in the Republic of Moldova (the study did not include the Transnistrian region), using Defined Daily Dose (DDD) as the measurement unit recommended by the World Health Organization (WHO) for drug utilization studies (6).

The quantity of antibiotics consumed was expressed in DDD/1000 hospitalization days, with the DDD values from the 2022 version of the WHO's Anatomical Therapeutic Chemical Classification/Defined Daily Dose (ATC/DDD) system used for each antibiotic.

The practical calculation method of DDD/1000 patient days (hospitalization days) followed this algorithm:

Stage I: Examination of the antibacterial delivery report for each IMSP individually.
Stage II: Conversion of all concentrations into grams or million units (MU).
Stage III: Tabular insertion of columns: WHO DDD, and respective WHO DDD Unit.
Stage IV: Calculation of DDD.
Stage V: DDD calculation per 1000 hospitalization days.
Stage VI: Arrangement of antibacterial agents by classes.
Stage VII: The results obtained were entered into electronic databases and statistically processed using Microsoft Office Excel and IBM SPSS Statistics programs.

Based on the geographical location of IMSPs, the quantity of consumed antibiotics was centralized and distributed per district/municipality, followed by mapping the results.

Districts/municipalities were classified into 5 levels of antibiotic consumption based on the values obtained from the determination of DDD/1000 hospitalization days over the years 2018-2021.

Thus, the following levels were outlined: “very high” – 25.4-30 DDD/1000 hospitalization days, “high” – 20.7-25.4 DDD/1000 hospitalization days, “medium” – 16-20.7 DDD/1000 hospitalization days, “low” – 11.4-16 DDD/1000 hospitalization days, “very low” – 5-11.4 DDD/1000 hospitalization days.

RESULTS

After analyzing the distribution of districts/municipalities based on the level of antibiotic consumption, the research results revealed a “very low” level of antibacterial agent usage from 2018 to 2021 in the following districts of the Republic of Moldova (fig. 1): Cahul, Cimislia, Hincesti, Nisporeni, Straseni, Calarasi, Orhei, Telenesti, Rezina, Soldanesti, and in the Municipality of Balti.

The total antibiotic consumption in these districts during the years 2018-2021 ranged between 5-11.4 DDD/1000 hospitalization days.

A "low" level of antibacterial agent consumption was recorded in the following districts: Cantemir, Leova, Basarabeasca, Stefan Voda, Causeni, Anenii Noi, Criuleni, Ungheni, Floresti, Soroca, Drochia, Edinet, Briceni, and in the Municipality of Chisinau.

The total antibiotic consumption in these districts during the years 2018-2021 ranged between 11.4-16 DDD/1000 hospitalization days.

A "medium" level of antibacterial agent consumption was recorded in the following districts: Taraclia, Comrat, Ialoveni, Singerei, Glodeni, and Donduseni.

The total antibiotic consumption in these districts during the years 2018-2021 ranged between 16-20.7 DDD/1000 hospitalization days.

Districts with a "high" level of antibiotic consumption, ranging between 20.7-25.4, were not identified.

A "very high" level of antibacterial agent consumption was recorded in Falesti district, where the total antibiotic consumption during the years 2018-2021 ranged between 25.4-30 DDD/1000 hospitalization days.

The trend of consumption for antibacterial preparations for the years 2018-2021 within the Public Health Institutions (IMSP) of the Republic of Moldova, as shown in Figure 2, is as follows:

a) In 2018, the average total antibiotic consumption was 12.3 DDD/1000 hospitalization days.
b) In 2019, the average total antibiotic consumption was 11 DDD/1000 hospitalization days.
c) In 2020, the average total antibiotic consumption was 15.6 DDD/1000 hospitalization days.
d) In 2021, the average total antibiotic consumption was 12.01 DDD/1000 hospitalization days.

Thus, there is an observed trend of increased antibiotic consumption in hospitals in the Republic of Moldova in 2020, a period associated with the COVID-19 pandemic. Additionally, in 2021, the trend of antibiotic preparations consumption is decreasing in the hospital sector, constituting 12.01 DDD/1000 hospitalization days.

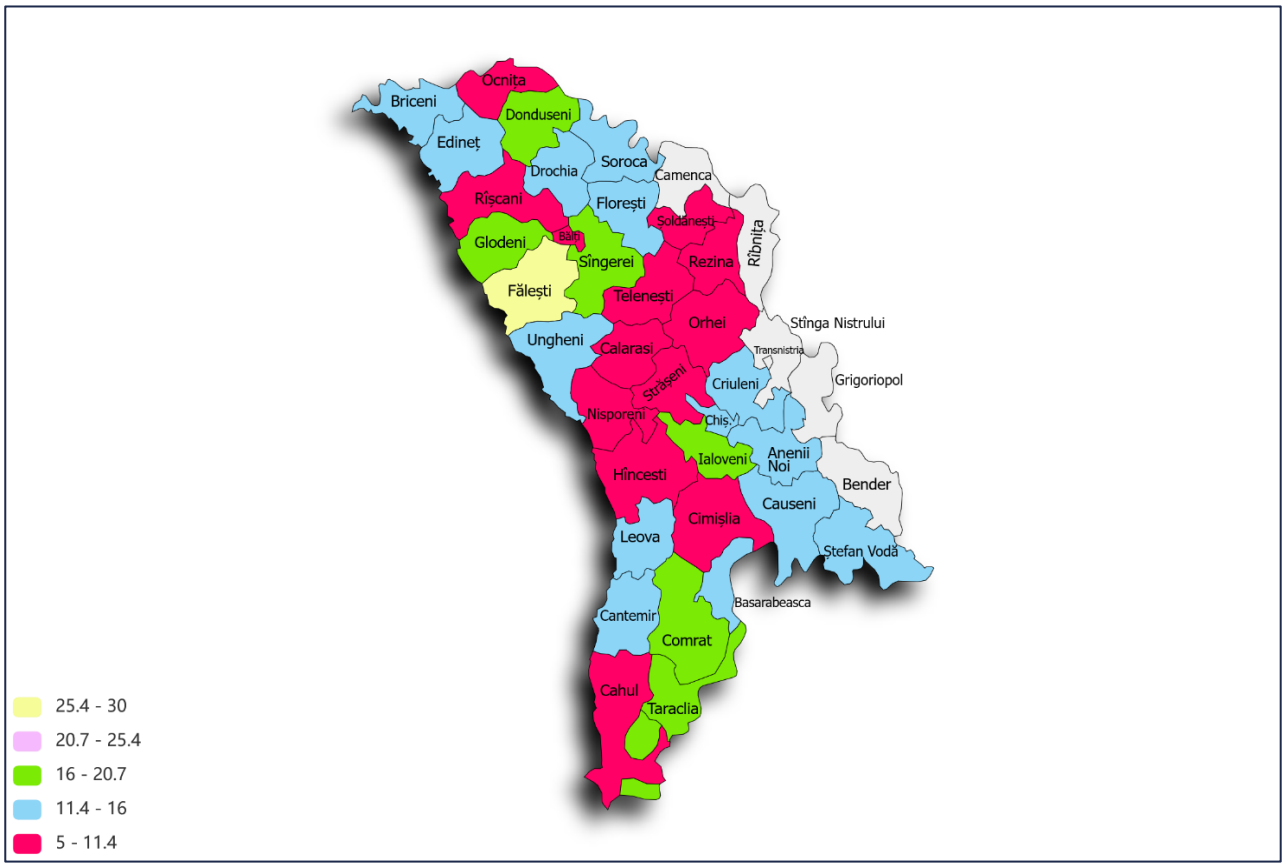

Figure 1. Mapping the antibiotic consumption in the Republic of Moldova based on administrative-territorial zone for the years 2018-2021, Expressed in DDD/1000 hospitalization days.

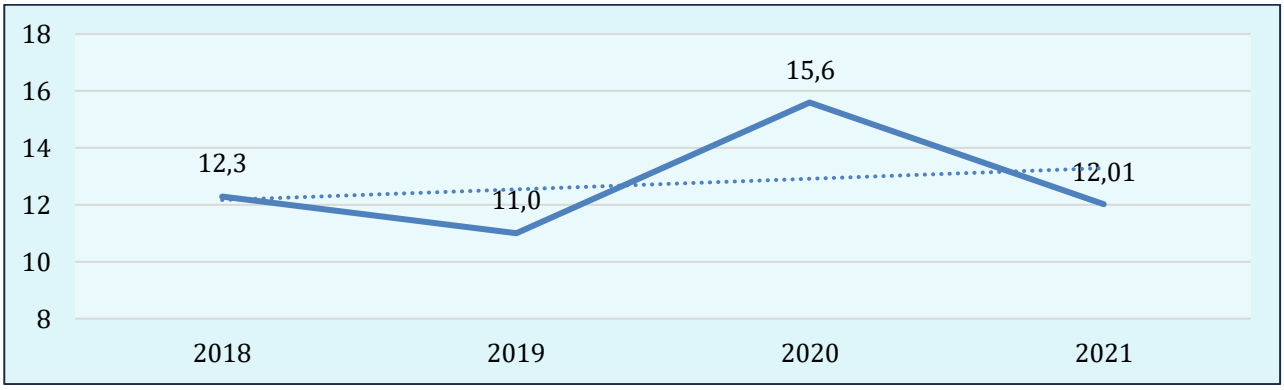

Figure 2. The trend of antibiotic consumption for the years 2018-2021, expressed in DDD/1000 hospitalization days.

Thus, there is an observed trend of increased antibiotic consumption in hospitals in the Republic of Moldova in 2020, a period associated with the COVID-19 pandemic. Additionally, in 2021, the trend of antibiotic preparations consumption is decreasing in the hospital sector, constituting 12.01 DDD/1000 hospitalization days.

Regarding the classes of consumed antibacterial preparations, as shown in Figure 3, it is presented as follows:

a) In 2018, there was an increased consumption of antibiotics from the following classes: Cephalosporins (21.2 DDD/1000 hospitalization days), Imidazole derivatives (16.1 DDD/1000 hospitalization days), Quinolones (13.6 DDD/1000 hospitalization days), and Tetracyclines (14.4 DDD/1000 hospitalization days).
b) In 2019, there was an increased consumption of antibiotics from the following classes: Cephalosporins (18.4 DDD/1000 hospitalization days),

Tetracyclines (15.5 DDD/1000 hospitalization days), Imidazole derivatives (14.4 DDD/1000 hospitalization days), and Macrolides (10.4 DDD/1000 hospitalization days).
c) In 2020, there was an increased consumption of antibiotics from the following classes: Cephalosporins (25.6 DDD/1000 hospitalization days), Macrolides (21.7 DDD/1000 hospitalization days), Tetracyclines (19.7 DDD/1000 hospitalization days), and Imidazole derivatives (15 DDD/1000 hospitalization days).
d) In 2021, there was an increased consumption of antibiotics from the following classes: Cephalosporins (17.4 DDD/1000 hospitalization days), Fluoroquinolones (15.4 DDD/1000 hospitalization days), Macrolides (12.9 DDD/1000 hospitalization days), Nitrofuran derivatives (9.9 DDD/1000 hospitalization days), and Aminoglycosides (9.5 DDD/1000 hospitalization days).

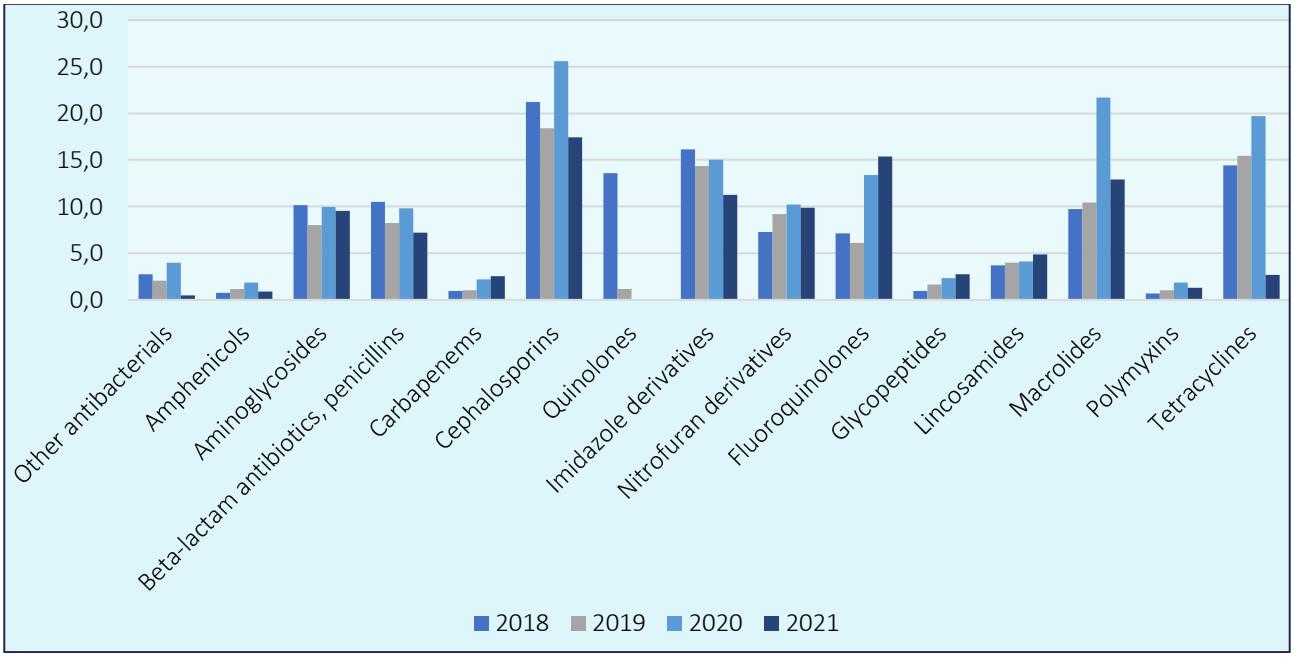

Figure 3. The consumption per class of antibiotics for the years 2018-2021, expressed in DDD/1000 hospitalization days.

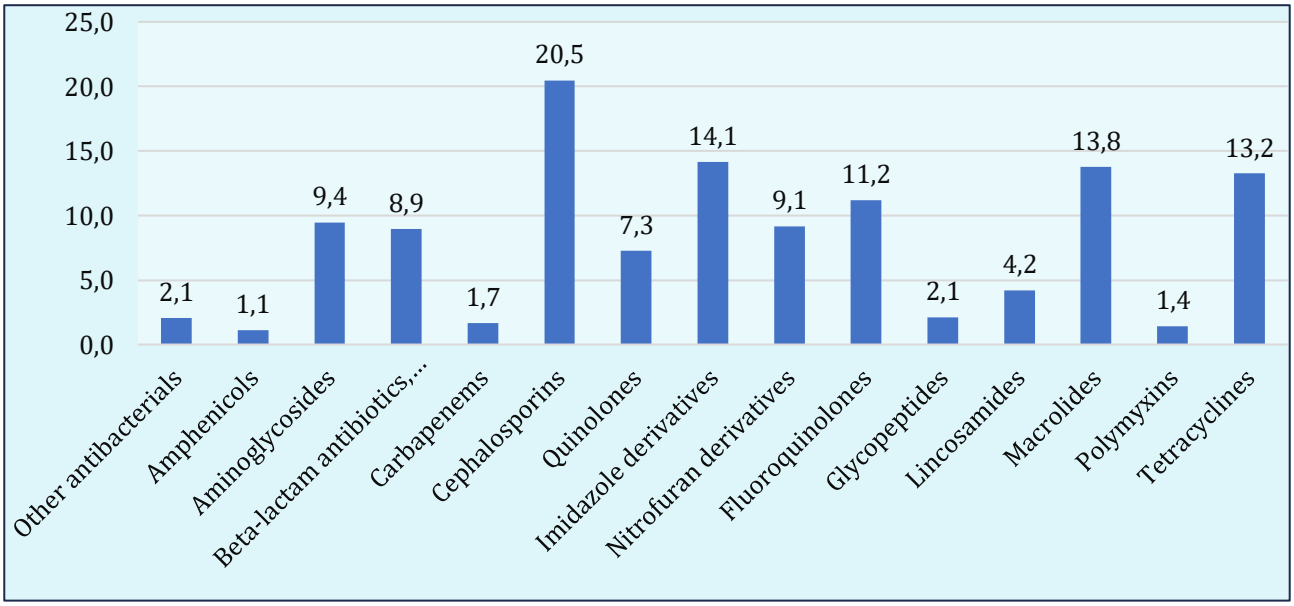

Figure 4. The total consumption per class of antibiotics for the years 2018-2021, expressed in DDD/1000 hospitalization days.

Following the centralization of data for the years 2018-2021, the total consumption per class of antibiotics highlighted an increased consumption for the following antibiotic groups (fig. 4): Cephalosporins (20.5 DDD/1000 hospitalization days), Imidazole derivatives (14.1 DDD/1000 hospitalization days), Macrolides (13.8 DDD/1000 hospitalization days), Tetracyclines (13.2 DDD/1000 hospitalization days), Fluoroquinolones (11.2 DDD/1000 hospitalization days), and Aminoglycosides (9.4 DDD/1000 hospitalization days).

DISCUSSIONS

Providing reliable national data on the consumption of antibacterial agents is mandatory to understand the epidemiology of antibiotic resistance, as antibiotic misuse is a key factor promoting bacterial resistance (5). While the majority of antibiotics are used in outpatient settings, the administration of antibiotics in hospitals is a major driver for the spread of multidrug-resistant bacteria responsible for nosocomial infections.

Thus, the conducted study serves as an efficient tool in providing valuable information for the development of new policies to prevent and combat nosocomial infections. The determined consumption levels reflect the situation only in hospital conditions; for a more comprehensive overview, it is necessary to assess antibiotic consumption in outpatient settings as well.

Regarding the antibiotic classes used in hospitals during the years 2018-2021, there is a predominant high consumption for antibiotics in the following classes: Cephalosporins, Imidazole derivatives, Macrolides, Tetracyclines, and Fluoroquinolones, and lower consumption for antibiotics in the classes: Amphenicols, Polymyxins, Carbapenems, Glycopeptides, and Lincosamides. The trend in antibiotic consumption is decreasing for the year 2021 by 23% compared to 2020.

Studies on antibiotic consumption are limited, as a major challenge in conducting these studies is the lack of data on antibiotic consumption in low- and middle-income countries (7). Strengthening national networks in the field of antimicrobial resistance and expanding subsequent participation in global and regional networks will enable the Republic of Moldova to adjust quality standards and operational procedures to international rigor and obtain comparable, standardized, and accurate data for evidence-based decision-making (4). Currently, monitoring of antimicrobial consumption relies on import data, and there is a clear need for the use of additional data sources, such as retail trade, including disaggregation by community and hospital sectors.

CONCLUSIONS

1. Mapping antibiotic consumption represents an efficient tool for highlighting the administrative-territorial regions with increased antibiotic use, contributing to the improvement of the national system for monitoring antimicrobial resistance and antibiotic consumption based on contemporary approaches. The study reveals a "very high" level of antibiotic consumption expressed in DDD/1000 hospitalization days in Falesti district and a "medium" level of antibacterial preparations consumption in the districts of Taraclia, Comrat, Ialoveni, Singerei, Glodeni, and Donduseni. Continuous monitoring of antibiotic consumption trends provides important information to competent authorities for implementing measures to reduce irrational antibiotic use, thereby decreasing the risk of resistant bacterial strains.
2. Determining the consumption of antibacterial preparations used in a hospital setting is an important step in estimating the costs associated with acquiring these preparations. It can signal the onset of bacterial resistance to certain classes of antibiotics and serve as an efficient tool to limit the misuse of drugs from a specific antibiotic class.

CONFLICT OF INTEREST

Authors have no conflict of interest to declare.

ETHICAL APPROVAL

Favorable opinion from the Research Ethics Committee No. 5 dated 12.07.2022, ref. no. 42 dated 26.05.2022, for the scientific research project titled: "Evaluation of antibiotic consumption and rational use of antibacterial preparations in hospitals."

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RATIONAL USE OF ANTIBACTERIALS IN PLEURAL EMPYEMA

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Keywords: *pleural empyema, antibacterial drugs, pathogenic flora, rational use.*

Introduction. *Pleural empyema remains a significant cause of morbidity and mortality globally. The rising incidence of this complex pathology necessitates an evaluation of diagnostic methods, surgical treatment, and antibiotic therapy strategies.*

Material and methods. *We selected and analyzed 50 observation records of patients with pleural empyema admitted to the thoracic surgical ward at Timofei Mosneaga Republican Clinical Hospital. A literature review was conducted to elucidate the rationale behind the administration of antibacterial treatment.*

Results. *Bacteriological examination of the pleural fluid revealed the presence of pathogenic flora, predominantly gram-negative (15 cases) - P. aeruginosa, Acinetobacter, K. pneumoniae, E. aerogenes, S. marcescens, Corrynebacterium, P. mirabilis, and in 5 cases, a polymicrobial etiology was observed. Evaluation of microbial susceptibility allowed for a rational choice of antibacterial treatment. Beta-lactam antibiotics were most frequently administered, either in monotherapy or in combination. Other groups of antibacterials administered included fluoroquinolones, aminoglycosides, macrolides, polymyxins, nitroimidazole derivatives, and glycopeptides.*

Conclusions. *Effective and harmless antibacterial treatment can only be achieved by identifying the causative pathogens and their antimicrobial susceptibility, ensuring adequate concentrations in the pleural space, determining the routes of administration, the duration of therapy, and the rational combination of antimicrobials.*

Cuvinte-cheie: *empiem pleural, antibacteriene, floră patogenă, utilizare rațională.*

UTILIZAREA RAȚIONALĂ A ANTIBACTERIENELOR ÎN EMPIEMUL PLEURAL

Introducere. *Empiemul pleural continuă să fie o cauză importantă de morbiditate și de mortalitate în lume, iar creșterea incidenței acestei patologii complexe necesită o evaluare a metodelor de diagnostic, de tratament chirurgical și a strategiilor de antibioterapie.*

Material și metode. *Au fost selectate și analizate 50 de fișe de observație ale pacienților cu empiem pleural internați în Secția chirurgie toracică a IMSP SCR „Timofei Mosneaga". Pentru elucidarea raționalității administrării tratamentului antibacterian a fost efectuat review-ul literaturii.*

Rezultate. *Examenul bacteriologic al lichidului pleural a permis depistarea florei patogene, preponderent floră gramnegativă (15 cazuri) - P. aeruginosa, Acinetobacter spp., K. pneumoniae, E. aerogenes-, S. marcescens, Corrynebacterium spp., P. mirabilis, iar în cinci cazuri – de etiologie polimicrobiană. Evaluarea sensibilității microorganismelor la antimicrobiene a permis alegerea rațională a tratamentului antibacterian. Cel mai frecvent, în monoterapie sau în combinație, au fost administrate antibioticele beta-lactamice. De asemenea au fost administrate antibacteriene din următoarele grupe: fluorochinolone, aminoglicozide, macrolide, polimixine, derivați de nitroimidazol, glicopeptide.*

Concluzii. *Tratamentul antibacterian eficient și inofensiv în emfizemul pleural poate fi realizat doar prin stabilirea agenților patogeni etiologici și a sensibilității lor la preparatele antimicrobiene, asigurarea unor concentrații adecvate în spațiul pleural, determinarea căilor de administrare, duratei curei de tratament, asocierea rațională de antimicrobiene.*

INTRODUCTION

Pleural infections present a significant challenge for medical practice and public health, with a mortality rate of 10-20%, and even 35% in elderly and immunocompromised patients. Recent epidemiological data has indicated a global increase in the incidence of pleural infections, attributed to factors such as the growing elderly population with multiple chronic comorbidities, frequent use of immunosuppressive medications, evolution of pathogenic microbial flora and bacterial resistance, and improved accessibility of diagnostic methods, including in outpatient settings. The treatment of pleural infections is considered one of the most expensive among all lung infections. Various causes of pleural infections have been identified, including community-acquired and hospital-acquired pneumonia, lung abscess, chest injuries and trauma, bronchopleural fistula, esophageal perforation, post-surgical complications, bronchogenic cancer, immunocompromised conditions, post-operative infections, patients on hemodialysis, and those undergoing antitumor therapy. The subacute onset of the pathology leads to delayed admission of patients to specialized medical services and thus the therapeutic management of pleural empyema is associated with long duration of hospitalization, significant use of healthcare resources, long-term antibiotic therapy, chest tube drainage, and/or surgery. While mortality from pleural infections has decreased considerably in the antibiotic era, the increased use of antimicrobial preparations, including irrational use, has led to a rise in bacterial resistance, as evidenced by the epidemics and pandemics of the early 21st century (SARS CoV, MRES, Ebola, SARS CoV-2, etc.). In this context, it is imperative to adjust the antibacterial therapy of pleural infections by addressing several key questions, such as the selection of antibacterial preparations for empirical therapy, improvement of methods for detecting pathogens and their susceptibility to antimicrobial preparations, determination of optimal routes of antibiotic administration and duration of antibacterial treatment, rationality of stepwise antibacterial therapy, pharmacokinetic studies on drug penetration into the site of infection, ensuring sufficient inhibitory concentrations to combat pathogens, and advocating for combinations of antimicrobial preparations (1 - 4).

The aim of the study was to analyze the spectrum of pathogens causing pleural empyema and their susceptibility to antimicrobial preparations, to characterize the groups of antibacterial preparations used, the duration of antibacterial treatment, and to justify the prescription of antimicrobial preparations based on pharmacokinetic data.

MATERIAL AND METHODS

The study was retrospective, involving the selection and analysis of 50 medical records of patients with pleural empyema as the primary (46 cases) or secondary (4 cases) clinical diagnosis, with or without fistulas, who were admitted to the thoracic surgery ward at the *Timofei Mosneaga* Republican Clinical Hospital. Bacteriological results from the medical records were analyzed to identify pathogens and their sensitivity to antibacterial preparations, and the prescription records were examined to analyze the antibiotics and synthetic chemotherapeutic agents used in the treatment. Additionally, relevant literature on antibacterial treatment for pleural infections was selected and analyzed in the PubMed database using keywords such as pleural infection, pleural empyema, pathogens, antibiotics, and bacterial resistance.

RESULTS

Based on the analysis of the medical records, it was found that the average age of the patients was 58 years (ranging from 31 to 77 years), with a predominance of male patients (41 males and 9 females). The duration of hospitalization ranged from 4 to 35 days, with an average of 12 days, and 8 patients required transfer to other wards, including intensive care. The causes of pleural empyema included pneumonia (29 cases), chest trauma (7 cases), pulmonary gangrene (4 cases), post-surgical infection (4 cases), lung abscess (2 cases), healthcare-associated empyema (2 cases), mesothelioma (1 case), and sepsis (1 case). The study was retrospective, and as a result, the data from observation records were not sufficient to differentiate between hospital-acquired and community-acquired etiologies.

The bacteriological examination of pleural fluid was conducted in all patients included in the study to identify the pathogen and antibacterial susceptibility. Our results indicate that bacterial growth was present in the pleural fluid in only

28 cases (56%). Among these, a single pathogen was identified in 23 cases (82.2%), while the etiology was polymicrobial in 5 cases (17.8%). Gram-positive flora (including *Staphylococcus aureus-5, Staphylococcus hominis-1, Staphylococcus epidermidis-1, Actinomyces odontolyticus-1*) was identified in 8 patients, and gram-negative flora (including *Pseudomonas aeruginosa-8, Acinetobacter-1, Klebsiella pneumoniae-2, Enterobacter-1, Serratia marcescens-1, Corrynebacterium-1, Proteus mirabilis-1*) was found in 15 patients. The predominance of gram-negative agents and Staphylococcus aureus suggests that the pathogenic flora of the pleural infection was most likely acquired in a hospital setting.

The analysis of bacterial susceptibility results to antibacterial preparations revealed a high resistance of the identified agents. Specifically, *Staphylococcus aureus* MRSA was found to be pandrug-resistant, *Pseudomonas aeruginosa* exhibited polyresistance, with sensitivity only to colistin or amikacin, *Enterococcus faecium* showed sensitivity to vancomycin, and *Corrynebacterium* was sensitive to linezolid.

Antibacterial treatment was administered to 45 patients, with 17 receiving a single antimicrobial preparation and 28 receiving a combination of two or more antibacterials. Among these patients, bacterial growth was not observed in the pleural fluid of 3 individuals, and following surgery, their clinical condition improved, leading to a recommendation for continued antibacterial treatment at home. In 2 patients with detected pathogenic flora, no antibacterials were prescribed as the condition improved after pleural cavity drainage. The results of the bacteriological examination of pleural fluid prompted a modification of the drug regimen. The absence of bacterial growth in the pleural fluid may be attributed to the administered antibacterial treatment or to the presence of anaerobic agents, the identification of which is challenging using standard tests. Considering the advantageous nature of anaerobic infection growth in pleural fluid, the use of an antibacterial drug with a spectrum of action against anaerobic agents is essential. The study identified the use of metronidazole, meropenem, cefoperazone, ceftazidime, and piperacillin as monotherapy or combination therapy, providing coverage against anaerobic agents in 29 cases.

Based on the analysis of the prescription records, the following medications were prescribed to patients with pleural empyema: beta-lactams (49); fluoroquinolones (9); aminoglycosides (11); polymyxins (4); macrolides (2); glycopeptides (1); and nitroimidazole derivatives (12). Among the beta-lactams, penicillins with beta-lactamase inhibitors were used in 16 cases (amoxicillin+clavulanic acid-9, piperacillin+tazobactam-7), second-generation cephalosporins (cefuroxime-4), and third-generation cephalosporins alone (ceftriaxone-4, cefotaxime-5) or in combination with beta-lactamase inhibitors (cefoperazone+sulbactam-14, ceftazidime+avibactam-5), as well as carbapenems (meropenem-3). Fluoroquinolones such as ciprofloxacin (7), levofloxacin (1), and moxifloxacin (1) were also used. Additionally, aminoglycosides like gentamicin (4) and amikacin (7), polymyxins specifically colistin (4), macrolides such as azithromycin (2), glycopeptides like vancomycin (1), and nitroimidazole derivatives like metronidazole (12) were prescribed.

In our study, the third generation of cephalosporins was the most frequently used beta-lactams (56% of cases). Their broad spectrum of action, which covers aerobic gram-positive, gram-negative (including anti-pseudomonas preparations such as cefoperazone and ceftazidime), and anaerobic bacteria, allows for effective coverage of the pathogenic flora commonly associated with pleural infections. Penicillins with beta-lactamase inhibitors were used in 15 cases either as monotherapy or in combination with other antibacterials. Amoxicillin+clavulanic acid, with its broad spectrum of action against predominantly aerobic bacteria, was frequently administered concomitantly with metronidazole to provide anti-anaerobic action. Patients showed negative bacteriological results, and the average duration of intravenous administration was 7 days. Piperacillin+tazobactam was used in 7 patients, either alone or in combination, for its anti-pseudomonas action as well as its anaerobic spectrum. An important issue is the increased resistance of Pseudomonas aeruginosa, Enterobacteriaceae, Acinetobacter spp, and Klebsiella spp, as detected in the bacteriological examination of pleural fluid in patients treated with beta-lactam antibiotics. Combining them with beta-lactamase inhibitors extends their spectrum and increases efficacy. Meropenem, with its ultra broad spectrum of action against both gram-positive and gram-negati-

ve aerobic and anaerobic bacteria, can therefore be used in polymicrobial pleural infections.

According to the results of the bacteriological examination of pleural fluid, aminoglycoside-sensitive pathogens were identified, including Staphylococcus hominis, Staphylococcus epidermidis, Pseudomonas aeruginosa, and Klebsiella pneumoniae, leading to the intravenous use of gentamicin or amikacin in 13 patients diagnosed with pyothorax. Gentamicin and particularly amikacin demonstrate activity against gram-negative aerobic flora and less activity against gram-positive flora, although this may be enhanced when combined with beta-lactams. Amikacin is considered to be an aminoglycoside with higher efficacy against Pseudomonas aeruginosa, attributed to its resistance to enzymes produced by microbes.

Vancomycin was administered in combination with amoxicillin/clavulanic acid to a patient whose pleural fluid showed growth of both *Enterococcus faecalis* (susceptible to amoxicillin/clavulanic acid, ampicillin, nitrofurantoin, ofloxacin, piperacillin/tazobactam, and vancomycin) and *Enterococcus faecium* (susceptible to linezolid, nitrofurantoin, and vancomycin). Creatinine values were monitored during treatment and showed insignificant variations. The patient was discharged with clinical improvement and recommended internal administration of ampicillin 500 mg twice daily (sources recommend administration every 6 hours-4 times daily).

Colistin was administered to 2 patients with polymicrobial pyothorax (*P. aeruginosa* with *Acinetobacter* and *P. aeruginosa, K. pneumoniae, P. mirabilis, S. saprophyticus*) and to 2 patients with P. aeruginosa in pleural fluid, susceptible to colistin. The disease progression in these patients was severe, requiring an average hospitalization period of 30 days, repeated surgical interventions for diagnostic and therapeutic purposes, and stays in 2 or 3 hospital wards during their hospitalization. Combined antibacterial therapies were ineffective, and colistin administration was the final treatment option. One patient received both inhaled and intravenous administration. The optimal dosing regimen is the administration of 9 million units per day, considering the severity of the pleural pathology.

Due to the diversity of causative pathogens and the presence of mixed flora (aerobic, anaerobic, fungal) in the treatment of pyothorax, it is recommended to administer antibiotics with a spectrum of action against anaerobic agents, such as metronidazole. Metronidazole-resistant anaerobic bacteria (Actinomyces odontolyticus) and facultatively anaerobic bacteria (Proteus mirabilis, Serratia marcescens), which are not covered by the spectrum of action of metronidazole, were identified in the study. The dosing regimen, to ensure efficacy, involves administration of 500 mg internally or intravenously every 8 hours. In the study, metronidazole was administered intravenously in 10 cases, with a 100 ml of 5% solution twice daily, and internally in 2 cases, with 500 mg administered twice daily at 12-hour intervals.

DISCUSSIONS

The agents of community-acquired pleural infection included *Streptococcus viridans, Streptococcus pneumoniae*, methicillin-sensitive *Staphylococcus aureus* (MSSA), *Enterobacteriaceae, Klebsiella*, and *Pseudomonas*. Hospital-acquired infections involved methicillin-resistant *Staphylococcus aureus* (MRSA), *Enterobacteriaceae, Enterococcus, Streptococcus viridans, Pseudomonas*, and *Klebsiella*. Additionally, atypical flora (*Mycoplasma* spp., *Legionella* spp.) and fungal flora (*Candida, Aspergillus*) were rarely found, especially in immunocompromised patients. Anaerobic infections, which are difficult to detect, were thought to accompany pleural infection in about 25% of cases (1-5).

Pleural infection caused by resistant pathogens was found in 37% of isolates from community-acquired infections and 77% of isolates from hospital-acquired infections, which were resistant to at least one of the prescribed antibiotics for respiratory infections. The high rate of resistant bacteria limits therapeutic options, and in *vitro* susceptibility of pathogens does not always correlate with the therapeutic efficacy of antibacterials, given the importance of pharmacokinetic features (1, 2).

Prompt initiation of antimicrobial therapy is crucial for the treatment of pleural infection. The initial selection of antibacterials is empirical, based on the etiology of the pathology (community-acquired, hospital-acquired), the patient's condition, and clinical manifestations (2, 6).

For the selection of antibacterial therapy in a patient with pleural empyema, it is necessary to evaluate patient -dependent factors, pathology-

dependent factors, and the characteristics of the drug, which may influence the disease's progression and response to treatment. Among patient-dependent factors, it is essential to consider age, comorbidities, previous antibacterial treatment, and so on. Among pleural pathology-dependent factors, the stage of empyema, pleural effusion size, pleural thickening, degree of pleural inflammation, and duration of symptom onset to hospital admission are important. Significant characteristics of the antimicrobial preparation include its spectrum and mechanism of action, dose- and time-dependent antibacterial effect, duration of action and dosing regimen, pharmacokinetic properties, ability to penetrate pleural fluid, availability of parenteral and enteral forms, and adverse reactions (4).

In pleural infections, pleural effusions can rapidly progress from uncomplicated parapneumonic effusions to empyema. During the exudative stage of empyema, pleural fluid accumulates due to increased permeability of the visceral pleura. As the infection progresses, fibrin accumulation on the pleural membranes leads to pleural thickening and septum formation. When pyothorax develops, the pleural fluid becomes more acidic and purulent due to inflammation, and there is an increased flow of protein into the pleural space. Consequently, the penetration of antibiotics may be difficult due to the thickened pleura, with pleural fluid characteristics varying depending on the stage of pleural empyema. Mesothelial cells, which line the pleural cavity, play a crucial role in the filtration of pleural fluid, as it results from differences in hydrostatic and colloid-osmotic pressure between the pleural fluid and capillary blood. Furthermore, it has been suggested that mesothelial cells have the ability to reabsorb antibiotics (6, 7).

Cephalosporins are commonly prescribed and administered as first-line therapy for respiratory, urinary, and CNS infections, spanning from mild to severe, due to their broad spectrum of action, bactericidal effect, and low risk of adverse reactions. However, widespread use has led to the development of resistance to these drugs. The microorganisms mainly involved in conferring resistance to this antibiotic can be identified by the acronym *ESCKAPE*, which stands for *Enterococcus faecium, Staphylococcus aureus, Clostridium difficile, Klebsiella pneumoniae, Acinetobacter baumanii, Pseudomonas aeruginosa*, and *Enterobacteriaceae*. The rapid emergence of this resistance poses a serious threat to the continuous relevance of the antibiotic (8).

First-generation fluoroquinolones (such as ciprofloxacin) exhibit greater activity against gram-negative flora and moderate activity against atypical agents and gram-positive cocci. They also possess advantageous pharmacokinetic properties, allowing for effective penetration into body fluids and tissues, and maintenance of bactericidal concentrations for 12-24 hours. Moxifloxacin, on the other hand, demonstrates activity against both Gram-positive and Gram-negative bacilli, including anaerobes. In both animal and human studies, moxifloxacin has shown favorable pleural penetration and could be considered as a de-escalation option, particularly for internal administration (3, 9).

Polymyxins demonstrate significant activity against aerobic Gram-negative bacteria, including most pathogens of the *Enterobacteriaceae* family, such as *E. coli, Enterobacter, Klebsiella, Citrobacter, Salmonella*, and *Shigella*. They are also effective against common Gram-negative non-fermentative pathogens, including *Pseudomonas aeruginosa* and *Acinetobacter baumannii*. Gram-negative bacteria that are naturally resistant to polymyxins include *Pseudomonas mallei, Morganella morganii, Vibrio cholerae, Serratia marcescens, Proteus spp., Providencia spp., Burkholderia cepacia, Chromobacterium spp., Edwardsiella spp., Legionella, Brucella*, and *Campylobacter*. Polymyxins are not active against gram-negative cocci (*Neisseria spp*.), Gram-positive and anaerobic bacteria, parasites, or fungi. Orally administered polymyxins are used only for disinfection of the digestive tract due to poor absorption when taken internally. Additionally, polymyxins do not efficiently diffuse into tissues or penetrate the cerebrospinal fluid, pleural and peritoneal cavities. However, they are used systemically by intravenous administration for serious infections caused by pathogens resistant to other therapies. Carbapenem-resistant Enterobacteriaceae, *Pseudomonas aeruginosa*, and multidrug-resistant or pan-drug-resistant *Acinetobacter baumannii* pose major problems for antimicrobial therapy due to extremely limited treatment options. Polymyxins, along with fosfomycin, ceftazidime/avibactam, and the recently approved meropenem-vaborbactam, are among the last-resort antibiotics that are still effective against such pathogens (10, 11).

Along with aminoglycosides, polymyxins can be administered by inhalation. In ICU patients, pneumonia caused by carbapenem-resistant gram-negative bacteria is a frequent and serious complication. When treating lower respiratory tract infections, the pulmonary penetration of antimicrobials is considered an important factor that can affect their efficacy. For patients with pneumonia caused by multidrug-resistant bacteria, the intravenous administration of colistin combined with nebulized inhalation is more effective than intravenous infusion alone. However, the utility of polymyxins is currently facing increasing resistance worldwide, mainly due to the plasmid-encoded colistin resistance gene present in pathogens such as *Escherichia coli* and *Klebsiella pneumoniae* (10, 11).

Macrolides, including azithromycin, may be recommended for coverage of atypical microorganisms. They have a minor role in the etiology of pleural empyema, which is why they are rarely recommended as empirical therapy (4).

In community-acquired pleural infection, antibiotic regimens typically include either parenteral second or third-generation cephalosporins (e.g., ceftriaxone) combined with metronidazole for anaerobic coverage, or a β-lactam/β-lactamase inhibitor combination (e.g., amoxicillin-clavulanic acid) in monotherapy. For patients with penicillin allergy, moxifloxacin used alone or a combination of levofloxacin and metronidazole are optimal therapeutic alternatives. In hospital-acquired pleural infection, antipseudomonal and anti-anaerobic antibiotics are required. Appropriate combinations include cefepime-metronidazole, piperacillin-tazobactam, or carbapenem. Additionally, coverage of S. aureus with vancomycin or linezolid should be considered (12, 13).

The therapeutic efficacy of antibacterials administered to patients with pleural empyema is determined by their ability to penetrate the pleural space and provide a concentration that reaches or exceeds the minimum inhibitory concentration (MIC) at the site of infection. Penetration of antibiotics into the lung follows various mechanisms, such as passive diffusion (beta-lactams), permeation (macrolides), and active transport (quinolones, clindamycin). The differences in these mechanisms lead to variations in antimicrobial concentration between pulmonary sites and serum. For example, aminoglycosides are hydrophilic, so they can enter cells very slowly via endocytosis and accumulate almost entirely in lysosomes. Macrolides enter the lungs through membrane penetration, and the rate is limited by the degree of lipid solubility. Quinolones and clindamycin penetrate cell membranes by active, energy-dependent transport and therefore can be saturated, leading to differences between serum and tissue levels. Another important pharmacokinetic parameter is the ability of antibiotics to penetrate pleural fluid. Studies recommend the use of beta-lactams in the treatment of pleural empyema, demonstrating efficacy due to high penetration into pleural fluid (1, 3, 4, 14, 15).

It has been suggested that antibiotic levels in pleural fluid are lower than those in serum in patients with empyema, due to the lower permeability of the thickened pleura and the more acidic local environment. However, during acute infection characterized by inflammation, vasodilation, edema, and increased membrane permeability, the penetration of antimicrobial agents may be enhanced. In this context, for the assessment of antibiotic penetration into the pleural space, special and careful consideration should be given to the underlying pathophysiology and mechanisms of fluid formation. In empyema, during the exudative stage, pleural fluid accumulates in the pleural space due to inflammation and increased permeability of the visceral pleura. Progression of infection, bacterial invasion of the pleural space, fibrin deposition on pleural membranes, and the formation of septations cause thickening of the pleura and loss of elasticity. Regarding malignant effusions, an exudate predominantly forms due to the deregulation of pleural space drainage caused by the obstruction of blood and lymphatic vessels in the lungs and pleura. These features would determine the penetration of antimicrobial agents into the pleural fluid depending on the etiology and pathophysiological mechanisms. Based on the reported results, it was concluded that there is very little difference between chemically different antimicrobial agents in their degree of pleural penetration (16).

Most antibiotics exhibit good lung penetration. Blood concentrations of ampicillin and penicillin G were higher than those in lung sites. The pulmonary and blood concentration ratios of orally and intravenously administered amoxicillin and cefotaxime were greater than 1 at 5-6 hours after the last dose. Oral administration or intravenous in

fusion of linezolid and levofloxacin resulted in higher concentrations in pleural fluid than in serum. The higher concentration of the drug at the sites of infection compared to that in the blood may be associated with changes arising from the inflammatory process. Inflammatory conditions cause vasodilation of capillaries and loss of selectivity of penetration of large molecules through membrane cells. Increased membrane permeability facilitates the ability of antibiotics to penetrate the lung. The degree of expression of inflammation and the stage of empyema may influence lung penetration capacity, as demonstrated by the use of β-lactams, clindamycin, oleandomycin, and erythromycin. Lung penetration decreases with a reduction in the expression of the inflammatory process (14).

In pharmacokinetic studies, the penetration of antibiotics into the pleura is assessed by calculating the ratio of the total area under the curve (AUC) for the concentration of a given antibiotic in the pleural fluid to the serum concentration. In an experimental pleural empyema model in animals, penicillin exhibited the highest pleural fluid to serum AUC ratio (AUCLP/S), followed by metronidazole, ceftriaxone, and clindamycin. Most studies have indicated that aminoglycosides have poor pleural penetration and are inactivated by the acidic environment of the infected pleural space. Gentamicin had the lowest ratio in this study. Therefore, due to the low pleural penetration and the tendency of aminoglycosides to be inactivated in the acidic environment of the infected pleural space, this group is not recommended for the management of pleural infections. In a study of humans with parapneumonic effusions, ceftriaxone concentration remained above the minimum inhibitory concentration level for most susceptible organisms for 53 hours after a single parenteral dose. In patients with *Staphylococcus aureus* MRSA mediastinitis given linezolid, the drug had an AUCLP/S of 1.64, suggesting that it is a rational therapeutic option in resistant pleural infections (2, 4, 5, 13).

Fluoroquinolones, antimicrobial preparations with a large volume of distribution, exhibit extensive tissue penetration. The respiratory fluoroquinolones, moxifloxacin or levofloxacin, are recommended as initial empirical antibiotic therapy for patients with respiratory infections, including pleural effusion, due to their high activity against gram-positive and gram-negative bacteria, including anaerobes. Several studies have investigated ciprofloxacin penetration into pleural fluid. Analysis of the structure of moxifloxacin, specifically the absence of the piperazinyl ring, revealed that the antimicrobial activity is not affected by acidic conditions. Thus, novel fluoroquinolones, including moxifloxacin, may be an attractive treatment option for pleural infections. The study determined inter-individual variability in moxifloxacin pharmacokinetics due to patient condition, concomitant drug administration, and competition for the same metabolic pathways. Examination of similar pharmacokinetic parameters (Cmax, AUC) confirmed moxifloxacin's ability to penetrate tissue compartments independent of the degree of inflammation or pH reduction. Therefore, the empirical use of moxifloxacin in the treatment of parapneumonic effusion and empyema is supported by its sufficient penetration into the pleural space and a favorable pharmacokinetic profile, regardless of the origin of pleural fluid (15).

There are few reports describing vancomycin penetration into the pleural cavity, and the available data are inconclusive. In pleural empyema, the parietal pleura, which consists of the mesothelial cells lining the pleural cavity, may already be thickened, and the minimum inhibitory concentration (MIC) of vancomycin may not be sufficient for a bactericidal effect. However, other studies report the efficacy of vancomycin administration in patients with pleural empyema caused by sensitive gram-positive bacteria. In some countries, increasing cases of resistance have been observed, especially in enterococci, including multiresistant strains of *Enterococcus faecium* (7).

An important clinical issue is the duration of antibacterial treatment. The duration of antimicrobial therapy in pleural infection is an area lacking in evidence and is largely based on expert opinion and extrapolation from recommendations for the treatment of lung abscess. The recommended duration ranges from 2 to 6 weeks, and the authors suggest a minimum of 4 weeks (including both intravenous and oral treatment). Discontinuation of treatment (less than 2 weeks) increases the risk of recurrence. However, the duration should be guided by the response to therapy, improvement in clinical symptoms, positive dynamics and/or radiographic resolution, and improvement in laboratory markers of infection (2-4, 6, 13).

Another issue that needs to be assessed is the optimal time to switch from intravenous to oral antibiotics or to perform de-escalation therapy (stepwise). This could be done in the context of analyzing the clinical response and biochemical parameters and could coincide with the time of chest tube removal. Current guidelines recommend switching from intravenous to oral antibiotics when there has been clinical improvement with normal body temperature, resolution of inflammation, and radiological improvement. A retrospective study showed that 3 weeks of antimicrobial therapy was usually adequate to prevent treatment failure. Treatment options with intrapleural enzyme therapies and surgery could serve as additional arguments for reducing the duration of antibiotic regimens (5, 13).

CONCLUSIONS

1. In patients with pleural empyema, gram-negative pathogens, commonly associated with hospital-acquired infections, were detected. Most of the bacterial isolates exhibited a high level of resistance to commonly prescribed empirical therapies.
2. Antibacterial preparations (beta-lactams, fluoroquinolones, polymyxins, glycopeptides, nitroimidazole derivatives) were carefully selected, taking into account their beneficial pharmacokinetic properties for penetration into pleural fluid and their activity against anaerobic flora.
3. The dosing regimen needs to be reviewed based on pharmacokinetic properties and the ability to penetrate the pleural space for patients with pleural infection.

CONFLICT OF INTEREST

Authors have no conflict of interest to declare.

ETHICAL APPROVAL

Since the article does not involve ethical risks, no research approval was required from the Ethics Committee.

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There is no information.

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THE CONCEPT OF THE BIOBANK INFORMATION SYSTEM AT *NICOLAE TESTEMITANU* STATE UNIVERSITY OF MEDICINE AND PHARMACY, REPUBLIC OF MOLDOVA

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Keywords: *Biospecimens, Biobank, research, database, donors.*

Introduction*. During the implementation of the Informational Record of Biospecimens within the Biobank project, the Register of Informational Record of Biospecimens within the Biobank software was developed and implemented.* ***Material and methods.*** *A narrative synthesis on the informational aspect of bio-specimens within the Biobank at the international level was conducted. Sources were searched in public databases, and after identification and deduplication, 54 relevant sources were selected, out of which 15 were comprehensively analyzed. The Register of Informational Record of Biospecimens within the Biobank software fully captures the flow of information within the Biobank at the Nicolae Testemitanu State University of Medicine and Pharmacy and serves as a tool for managing the biospecimens stored in the Biobank. It offers various capabilities to ensure diverse and representative data regarding potential donors of biospecimens.* ***Results.*** *The software enables rapid processing of biospecimens and their associated data, ensuring traceability with extensive management capabilities for diverse types of biospecimens at all stages throughout the Biobank circuit. The relevance of this data facilitates the improvement and acceleration of research and decision-making in the field of health, to the benefit of the donors. The software allows for the rapid registration and processing of biospecimens from donors in various research studies, adhering to the principles of research ethics and ensuring the anonymity of donors' personal data, as well as the biospecimen data and associated information.* ***Conclusions.*** *The Register of Informational Record of Biospecimens within the Biobank software is relevant for initiating scientific research. Potential beneficiaries, including scientific researchers, collaborators in scientific laboratories, and the academic staff at Nicolae Testemitanu State University of Medicine and Pharmacy, will benefit from comprehensive informational support for biospecimen records within the Biobank and have opportunities to streamline research activities.*

Cuvinte-cheie: *biospecimene, biobancă, cercerare, bază de date, donatori.*

CONCEPTUL SISTEMULUI INFORMAȚIONAL AL BIOBĂNCII UNIVERSITĂȚII DE STAT DE MEDICINĂ ŞI FARMACIE „NICOLAE TESTEMIȚANU" DIN REPUBLICA MOLDOVA

Introducere. *În perioada realizării proiectului „Evidența informațională a biospecimenelor din cadrul Biobăncii" s-a elaborat și implementat softul „Registrul de evidență informațională a bio-specimenelor din cadrul Biobăncii".* ***Material și metode.*** *A fost realizat o sinteză narativă asupra aspectului informațional al bio-specimenelo în cadrul Biobancii la nivel internațional. Sursele au fost căutate în baze de date publice/deschuse, iar după identificare și deduplicare, au fost selectate 54 de surse relevante dintre care 15 au fost analizate cuprinzător. Softul „Registrul de evidență informațională a biospecimenelor din cadrul Biobăncii" reflectă pe deplin mișcarea fluxului informațional din cadrul Biobăncii USMF „Nicolae Testemițanu" și servește ca instrument de gestionare a biospecimenelor stocate în Biobancă și are o serie de posibilități pentru a asigura date diversificate și reprezentative privind potențialii donatori de biospecimene.* ***Rezultate.*** *Softul permite prelucrarea rapidă a biospecimenelor și a datelor asociate acestora, și asigură trasabilitatea cu posibilități vaste de gestionare a biospecimenelor de natură diferită la toate etapele circuitului în Biobancă. Relevanța acestor date facilitează îmbunătățirea și accelerarea cercetării și luării de decizii în domeniul sănătății în beneficiul donatorilor. Softul permite înregistrarea și prelucrarea rapidă a biospecimenelor de la donatori în diverse studii de cercetare, respectând principiile eticii cercetării, și asigură anonimatul datelor personale ale donatorilor, precum și a datelor biospecimenelor și a informațiilor adiacente acestora.* ***Concluzii.*** *Software-ul „Registrul informațional al biospecimenelor din cadrul Biobăncii" este relevant în inițierea cercetării științifice. Beneficiari potențiali, cercetători științifici, colaboratori ai laboratoarelor științifice și personalul didactico-științific al USMF „Nicolae Testemitsanu", vor beneficia de un suport informațional cuprinzător pentru evidența biospecimenelor din cadrul Biobăncii și vor avea posibilități de eficientizare a activității de cercetare.*

INTRODUCTION

In the Republic of Moldova, the ongoing development of activities in the field of the human genome, technological innovations in cell processing, genetic research to study and describe the connections between genes, the physical and social environment, and people's lifestyles has led to the establishment of the national Biobank within the *Nicolae Testemitanu* State University of Medicine and Pharmacy (SMUPh), Republic of Moldova. Given the various types of Biobanks, it is essential to define the objectives of the Biobank at the outset of its formation.

The establishment of the Biobank requires accreditation and strict compliance with current quality standards. The Biobank at *Nicolae Testemitanu* SUMPh contains organized collections, storing human biological samples and associated data that are of great importance for research and personalized medicine. In other words, the Biobank serves as a repository of biological materials (biospecimens).

The purpose of the Biobank is to conduct the collection, reception, encoding/decoding, processing, storage, preservation, freezing, thawing, distribution, and/or destruction of biospecimens collected from patients involved in various research studies, along with their associated data, in order to support scientific investigations.

The individuals participating in the studies are assured of anonymity regarding their personal data after signing the consent form. Subsequently, samples are taken for the necessary biomaterial. The collected data are used exclusively for scientific and research purposes. During the biomaterial sampling and processing, personal data anonymization methods are employed, adhering to the limits established by the Personal Data Security Policy and the legal norms outlined in Law no. 133 of July 8, 2011, concerning the protection of personal data, as well as Law no. 71 of March 22, 2007, regarding registers. Ethical concerns surrounding biospecimens and the involvement of a number of people (operators) require special attention in information processing, with the incorporation of technological means. These factors constitute the critical components of the Biobank, with efforts made to ensure efficiency and safety in this aspect.

MATERIAL AND METHODS

A narrative synthesis was conducted on the informational subject of biospecimens within the Biobank at the international level. Simultaneously, data were systematized to fully reflect the movement of the information flow within the *Nicolae Testemitanu* SUMPh Biobank. Information sources were searched in PubMed, Google Scholar, and Hinari databases. The search strategies involved using keywords such as "biospecimens," "biobank," "database," "donors," and "data." The inclusion criteria for publications were articles published between January 2015 and January 2024. These publications were identified in databases such as Google Scholar, PubMed, HINARI, etc. After removing duplicate records and screening abstracts, 54 scientific sources were selected, of which 15 articles were fully reviewed.

The Biobank at *Nicolae Testemitanu* SUMPh features a specialized laboratory (scientific center) that enables the research, collection, processing, and storage of biospecimens obtained as a result of research projects. These biospecimens can be utilized in clinical trials or in prospective or retrospective studies (1). The increasing demand for tissues for translational studies is leading to a rise in the number of Biobanks (2).

RESULTS

The purpose of biobanks. To standardize the activities of the Biobank, it is necessary to implement standard operating procedures in all aspects, from acquisition, shipment, storage, and destruction to the safety of biospecimens (3). However, the development of these Biobank practices involves specialists to address potential ethical, legal, and social issues (4). The expanding field of biotechnology, which requires the establishment of an information system to monitor the collected biospecimens, highlights the increasing importance of financial support for Biobanks (5).

Biobanks serve as collections of biospecimens, which can later be used to elucidate medical diagnoses and personalized treatments (6). With this in mind, aliquoting will allow the use of biospecimens for both clinical and research purposes (7). To achieve this objective, the Biobank at *Nicolae Testemitanu* SUMPh collects, stores, and processes human biological materials, ensuring security in the collection of biospecimens intended for research or their use to guarantee a high level of

protection and promotion of human health. Various standard operating procedures and best practice protocols for biobanks are available, and the determination of which to use often depends on the specific needs of the clinic or study being conducted. The Best Practices for Operating Procedures, published by the Biospecimen Research Branch (NCI BBRB), can be accessed online (8).

Within the Biobank at *Nicolae Testemitanu* SUMPh, internationally standardized procedures, adjusted to the current legislation, have been implemented to assist researchers in making clinical decisions.

Screening the donor population can elucidate the specific alleles underlying a unique disease phenotype. By conducting such studies, it becomes possible to evaluate the environmental effects alongside the genetic ones, studying both monozygotic and dizygotic twins. This prospective approach can be employed in preventive medical programs and epidemiological studies (9).

The efficient management of biospecimens in the Biobank requires prompt and accurate collection, proper storage, and the recording of related information in an electronic system.

The information system allows for the accumulation and analysis of information related to biospecimens, including their storage in the Biobank and the monitoring of the flow of biospecimens from donors in various research studies. It ensures traceability with vast possibilities for managing biospecimens of different natures at all stages in the Biobank circuit.

The software contributes to the effectiveness and efficiency of personalized medicine processes, ensuring the qualitative management of information about research subjects. It serves as a strategic tool for the competitive clinical management of health services.

The information received from the medical institutions participating in the research projects is stored in the biospecimen record system using a standardized recording form. The data transmission procedure is outlined in a standardized instruction. The form encompasses primary patient data from various research studies, including passport information, epidemiological data, clinical details, paraclinical data, and more. These details are then presented to the operator responsible for records within the Biobank.

The Biobank operator accumulates and stores data in the *Register of Informational Record of Biospecimens within the Biobank* software, developed as part of the *Informational Record of Biospecimens within the Biobank* project, number 21.70086.37ŞD.

Collecting pertinent data on researched patients enables the assessment of management potential, efficiency options, and effectiveness in enhancing patients' quality of life while minimizing the impact on the health system.

Following the processing of all the data obtained at the Biobank level, information is obtained about the patients included in the research, with the elaboration of reports on the state of health regarding various medical conditions as well as their genealogy.

Donors' personal data, including biomaterial coding, are accessible to a limited number of people. Confidentiality regarding their identity and background is maintained throughout and after the completion of their studies.

Software requirements. Accreditation of a Biobank ensures that the laboratory (scientific center) controls and optimizes the use of biospecimens in accordance with good professional practice, as defined by internationally established standards. Accreditation requirements include the implementation of an operational quality management system and continuous control of the methods used for diagnostic purposes (10). Consequently, the Biobank is recognized for maintaining high-quality processes that produce reliable results. The accreditation process provides the Biobank with acknowledgment by ensuring consistency and standardization of practices.

Clinical research involving the Biobank requires approval from the Medical Ethics Committee (11). The cornerstone of bioethics is informed consent, which mandates providing patients and/or subjects with an overview of the study, a discussion of the protocol specifics, and disclosure of potential benefits and risks.

Equally important to a Biobank's role in acquiring and processing biospecimens is the management of clinical and research data relevant to each of those biospecimens. The *Nicolae Testemitanu* SUMPh Biobank's information record software has important capabilities that are recommended

in the practice guidelines and provides a secure environment that protects the data, includes a unique and unchangeable identifier for all biological samples, and the ability to continuously monitor the location and status of the evidence. The software system, with its implemented encryption component, operates 24/7, has a resilient infrastructure, and can effectively handle disasters and downtime. Moreover, it has the capability to recover stored data from regular backups using hard drives, external hard drives, USB sticks, and other portable memories, all properly encrypted to meet the latest information technology standards.

The biospecimen information record software within the Biobank satisfies the complete structure of the medical information of donors' samples. This adherence is in accordance with the sample collection form developed by the work team and provided to the Biobank at *Nicolae Testemitanu* SUMPh. It fully meets the needs for storage, processing, and validation of the obtained data. The software encompasses instruments enabling the accumulation and storage of information about the collected samples in the Biobank. It includes processing elements for primary donor data, diagnostic data, bio-specimen type, and more. It allows access to information about biospecimen donors from medical institutions, both separately by institution and in full, including donor data. The software facilitates the storage of sample information about donors through the software component created for the *Nicolae Testemitanu* SUMPh Biobank, which owns the database. Moreover, it grants access to collaborators at *Nicolae Testemitanu* SUMPh who are authorized to work with the IT system in question. The software enables the processing and analysis of results, such as the selection of samples based on criteria like nosology and those related to a specific period, on an institutional level. Additionally, it offers the capability to create reports through graphical representation and the use of statistical formulas. It is equipped with functionalities to analyze correlations between the data stored in the software from all medical institutions as a whole.

The software is equipped with interactive subsystems tailored to facilitate the tasks of operators. One group of operators focuses on collecting information from various medical institutions, while another operator, situated within the Biobank at *Nicolae Testemitanu* SUMPh, processes the gathered data. Hence, the software, referred to as the *Register of Informational Record of Biospecimens within the Biobank*, is organized into modules, each comprising two primary components. The first component is designed to input information about donors and the details acquired during the collection of their samples from medical institutions. The second component is integrated within the Biobank at *Nicolae Testemitanu* SUMPh, and its role is to manage the collection and preservation of human biological material for scientific purposes.

Each system component encompasses functions such as input-output, data validation and control, and performing relevant calculations to ensure the provision of information flows based on various parameters (grouping criteria). This shared functionality across different system services enhances the efficiency and quality of information related to the collection of donor sample data.

The computer system can manage information collected at the time of receiving biospecimens, as well as details about previously stored samples (both current and stored over time). The software is equipped with a service and maintenance tool for the database. The system includes essential components for reporting statistical data (a set of standardized reports) about donor samples stored in the database. Diagnoses are recorded according to the international codes of ICD revision X.

The computer system, named *Register of Informational Record of Biospecimens within the Biobank*, implemented within the Biobank at *Nicolae Testemitanu* SUMPh, can group, adjust, and process information within an individual medical institution, as well as across all medical institutions whose donor samples are included in the register. It also has the capability to process data from any period within the stored records.

Taking into account the potential dangers that may arise when collecting donors' personal data as well as the real risks when storing and processing them, the software provides the possibility of anonymizing and encrypting personal data.

Within the computerized system of informational record of biospecimens within the Biobank, technical and organizational measures will be imple-

mented to prevent the destruction or unauthorized modification, processing, and access to the data in the system. The primary and traditional measure for protecting personal data is the obligation of the staff engaged in using the IT system to maintain the confidentiality of patients' medical data. Given the various potential biosecurity hazards involved in working with bio-specimens, it is crucial for all staff members to adopt appropriate security measures. The relevant Biobank staff will regularly review biosafety information documents for all materials used. These documents include Material Safety Data Sheets, providing information on health hazards associated with chemicals and infectious agents, as well as guidelines on how to use biosafety items (12).

To ensure the confidentiality of donor sample data and medical institutions, the software should restrict access to unauthorized individuals. Depending on the size, various types of staff may be required for proper Biobank operation (13, 14, 15). Therefore, when hiring staff for a Biobank, factors such as professional training, work ethic, promptness, intellectual capacity, teamwork, and communication skills must be taken into consideration. Additionally, employees should be enrolled in educational programs to obtain the required certifications. Biobank staff may have a range of responsibilities such as purchasing, processing, storing, and maintaining the database.

DISCUSSIONS

The system's integrity is assured by the proper functioning of the developed tools. Data processing for scientific medical research and statistical purposes must adhere to suitable safe guards. The computer system will provide regular backups of data. The Biobank oversees the management and storage of data and generates electronic registers based on information authenticated and validated according to pre-established criteria.

The software developed for the Biobank provides data that will describe the biological material, its origin, and outcome. It facilitates information exchange with medical institutions through network traffic established between the Biobank and these institutions. Electronic records are created online, and the system encompasses various functionalities:

- Sample Collection Register - activates a screen with a table containing all the recorded samples;
- Project Log - opens a screen with a table containing all existing projects in the platform and related information;
- Donor register - opens a screen with a table containing the data of all the donors registered in the Biobank platform who have developed analyses, according to their agreement, for different scientific researches and projects;
- Results register - opens a screen with a table containing all the results of samples registered in the platform;
- Archived samples register - opens a screen with a table containing all the samples that have been destroyed, removed from research or have completed their research cycle.

The software implemented in the Biobank of the *Nicolae Testemitanu* SUMPh has an archiving system for sample records.

CONCLUSIONS

1. The development of the Biobank at *Nicolae Testemitanu* SUMPh adheres to the same requirements as other Biobanks for research. Emphasizing ethical considerations and respecting personal data through encryption are primary elements of the software development. This Biobank has the potential to offer unique research-based services to the scientific and medical community. Biospecimens are essential resources for research focused on human diseases. It is crucial for funding agencies, governments, and private entities to continue recognizing the importance of Biobanks in the ongoing development of human genome activities and genetic research.
2. Scientific researchers, collaborators of scientific laboratories, teaching and scientific staff at *Nicolae Testemitanu* SUMPh benefit from comprehensive informational support for recording biospecimens within the Biobank at *Nicolae Testemitanu* SUMPh, and they have the opportunity to enhance the efficiency of their research activities.

CONFIRMATION

This work was developed at *Nicolae Testemitanu* State University of Medicine and Pharmacy as part of the research project *Informational Evidence of Biospecimens within the Biobank* during the years 2021-2023, under the strategic direction of *Information Technology and Digital Development*. The project is registered in the State Register of projects in the field of science and innovation under the number 21.70105.37ŞD.

CONFLICT OF INTEREST

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

ETHICAL APPROVAL

The study was conducted and approved by the Ethics Committee no. 35/03.05.2022 of *Nicolae Testemitanu* State University of Medicine and Pharmacy of the Republic of Moldova.

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A NEW PROCEDURE FOR THE RAPID DETERMINATION OF *PSEUDOMONAS AERUGINOSA* BACTERIA IN MEDICATIONS

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Keywords: *procedure, rapid determination, Pseudomonas aeruginosa, culture medium, medication.*

Introduction. *The microbiological quality of non-sterile products covers two relevant aspects: firstly, the assessment of microbial load, and secondly, the absence of pathogenic microorganisms. The absence of certain pathogenic microorganisms in a preparation depends on their route of administration. Oral pharmaceutical products require the absence of microorganisms such as Salmonella and coliform bacteria, such as Escherichia coli. On the other hand, topical products require the absence of species like Staphylococcus aureus and Pseudomonas aeruginosa. The methods commonly used for identifying microorganisms in medications are laborious and time-consuming. Considering these factors, the development of rapid methods for determining unacceptable microorganisms in medications is an urgent necessity.*

Material and methods. *This is an experimental study in which reference bacterial strains, medicinal preparations, components, ingredients, selectivity factors, and inhibition factors were utilized. For the preparation of the culture medium, high-purity reagents registered in the Republic of Moldova were used.*

Results. *Following the conducted studies, a rapid procedure has been developed for determining P. aeruginosa bacteria in medications. Detection of individual P. aeruginosa cells is possible after 9-12 hours of incubation, while results for concentrations of 104-105 CFU are obtained within 5-6 hours. The medium stands out for its high selectivity and specificity, predominantly for these bacteria, and is economical and straightforward to apply. The shelf life of the medium is two years (observation period).*

Conclusions. *The developed procedure is valuable for the rapid determination of P. aeruginosa bacteria, and the accuracy of the results will enable the timely implementation of microbiological monitoring strategies for large batches of medications.*

Cuvinte-cheie: *procedeu, determi-nare rapidă, Pseudomonas aeruginosa, mediu de cultură, medicament.*

UN NOU PROCEDEU DE DETERMINARE RAPIDĂ A BACTERIILOR *PSEUDOMONAS AERUGINOSA* ÎN MEDICAMENTE

Introducere. *Calitatea microbiologică a produselor nesterile acoperă două aspecte relevante: evaluarea încărcăturii microbiene și absența microorganismelor patogene. Absența unor microorganisme patogene într-un preparat este în funcție de calea lor de administrare. Astfel, în produsele farmaceutice de uz oral nu trebuie să existe microorganisme din genul Salmonella și bacterii coliforme, cum ar fi Escherichia coli, iar în produsele topice sunt inadmisibile speciile Staphylococcus aureus și Pseudomonas aeruginosa. Întrucât metodele actuale de identificare a microorganismelor în medicamente sunt laborioase și necesită mult timp, dezvoltarea de metode rapide este o necesitate stringentă.*

Material și metode. *În prezentul studiu experimental au fost utilizate tulpini bacteriene de referință, preparate medicamentoase, componente, ingrediente, factori de selectivitae și de inhibiție. Pentru prepararea mediului de cultură au fost utilizate reactive de puritate înaltă înregistrate în Republica Moldova.*

Rezultate. *În urma studiilor efectuate s-a elaborat un procedeu rapid pentru determinarea bacteriilor P. aeruginosa în medicamente. Indicarea celulelor unice de P. aeruginosa este posibilă peste 9-12 ore de incubare, iar la concentrații de 10^4-10^5 UFC rezultatele se obțin timp de 5-6 ore. Mediul de cultură se distinge prin selectivitate și specificitate înaltă preponderent pentru această specie bacteriană, este econom și simplu în aplicare și are un termen de păstrare de doi ani (termen de observare).*

Concluzii. *Procedeul elaborat este util pentru determinarea rapidă a bacteriilor P. aeruginosa în medicamente, iar veridicitatea rezultatelor permite aplicarea în timp util a strategiilor de monitorizare microbiologică a loturilor mari de medicamente.*

INTRODUCTION

The microbiological control of medication is one of the essential missions in ensuring the quality surveillance of human pharmaceuticals, in accordance with the provisions of the current analytical and normative documentation. Currently, medical practice and observations in the pharmaceutical field demonstrate that numerous pharmaceutical forms, often overlooked, may contain bacteria and/or fungi in significantly varied proportions and diverse species, mostly saprophytic. Their contamination with pathogenic, conditionally pathogenic, or even saprophytic microorganisms increasingly raises public, collective, or individual health concerns. These medications pose a significant risk, especially in the treatment of burns, wounds, and severe ulcerations on large denuded epidermal surfaces. Additionally, indigenous intravenous medications prepared in hospitals, which carry a high risk of microbial contamination, have a short shelf life, leading to incidents that have resulted in fatalities (1, 2).

The increasing number of pharmaceutical recalls in recent years has once again made us aware of how crucial the microbiological quality of medications is. The presence of microorganisms in medications can become a source of infectious disease spread through them, especially when contaminated products are processed industrially in large batches (3).

The stability of a medication, alongside efficacy, purity, and safety, is a crucial factor in ensuring its quality. A medication considered stable maintains the quality characteristics imparted during preparation within the limits specified by official standards for a specified period. The medication should exhibit physical, chemical, and microbiological stability. Any change in the medication, whether apparent or not, as long as it falls outside the specifications provided by the manufacturer, can compromise its efficacy and safety. Medication incompatibilities may arise, and the presence of unacceptable microorganisms can introduce sources of contamination (1, 4).

In turn, the metabolic activity of microorganisms can induce physico-chemical instabilities, leading to the production of pyrogenic metabolites, a decrease in pharmacokinetic properties, and the nullification of therapeutic effects. This can instill doubt in the patient regarding the benefits offered by the medication.

In the treatment of various diseases, various pharmaceutical forms are used based on their content, mode of administration, and therapeutic action. These forms may be exposed to the unfavorable factors mentioned above. These preparations fulfil the indispensable conditions for the development of microorganisms; hence, a primary concern is to avoid microbial pollution from the technological preparation phase to the administration phase. The increasing number of pharmaceutical product recalls in recent years has once again made us aware of how crucial the microbiological quality of medications is (2, 4).

The *aim* of the study is to develop a procedure for the rapid detection and identification of unacceptable microorganisms in medications focusing on *P. aeruginosa*, which involves the culture medium for rapid multiplication and detection, as well as a minimal set of tests for swift identification.

MATERIAL AND METHODS

Bacterial strains

In this study, seven bacterial strains from the American Type Culture Collection (ATCC) and the National Collection of Strains from the United Kingdom were utilized, belonging to four species: *Pseudomonas aeruginosa* ATCC 27853, *Pseudomonas aeruginosa* ATCC 15442, *Pseudomonas aeruginosa* ATCC 49189, *Pseudomonas aeruginosa* NCTC 6750, *Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 25923, and *Staphylococcus epidermidis* ATCC 149990. Prior to the study, the strains were precultivated in Tryptic Soy Broth. All microbial strains were cultured on specific media for each species. The utilized media included Tryptic Soy Agar, Mannitol Salt Agar, MacConkey Agar, Blood Agar, Peptone Agar (BioMérieux, France), and MCS-Ps medium. The study also involved the use of medications labelled as "sterile," oral and topical preparations, components of formulations for micro-pellicle media, reagents, and chemical compounds for achieving selectivity, factors promoting multiplication, and inhibition factors obtained from official distributors.

A microbiological method was employed in the study, comparing known methods for the detection and identification of *Pseudomonas aeruginosa* with the proposed rapid determination proce-

dure, the investigation of the selectivity of the concentrated micro-pellicle medium, the examination of the specificity of the rapid procedure, a comparative study of established identification means with the developed one, and the microbiological control of the medication regulated by the European Pharmacopoeia compared with the control of the medication using the proposed procedure and algorithm (5).

Development of the culture medium

The proposed procedure consists of the micro-pellicle culture medium, which includes in its composition casein hydrolysate, gelatin, sodium malonate, cetyltrimethylammonium bromide, sodium chloride, water-soluble bromothymol blue, monopotassium phosphate, and disodium phosphate. The research was conducted using materials and high-purity reagents from "Sigma-Aldrich".

For the preparation of the medium, stock solutions of phosphates are prepared in advance to obtain the phosphate buffer solution at pH 6.9-7.0, a 10% gelatin solution, and a 0.5% bromothymol blue solution.

The preparation of phosphate stock solutions takes place using the following method: monopotassium phosphate (KH2PO4) – 9.078 g is dissolved in 1000 ml of distilled water, and disodium phosphate (Na2HPO4) – 11.876 g in 1000 mL of distilled water until complete dissolution.

The 10% gelatin solution is prepared in a sterile tube by introducing 1.0 g of crystallized gelatin and 9.0 mL of distilled water. It is left for 45-50 minutes to swell, then heated in a water bath at a temperature of 85-90°C, stirring with a glass rod until completely dissolved.

For the preparation of the 0.5% bromothymol blue solution, in a sterile tube, 10.0 mL of sterile distilled water is poured, and 0.05 g of water-soluble bromothymol blue is added. The tube is then heated for 45 minutes in a water bath at a temperature of 85-90°C.

The preparation of the culture medium involves introducing casein hydrolysate, sodium malonate, cetyltrimethylammonium bromide, sodium chloride, sterile distilled water, disodium phosphate, and monopotassium phosphate into a clean and sterile chemical retort. The mixture is stirred, and gelatin solution and bromothymol blue solution are added. The content is mixed to obtain a liquid culture medium, which is then dispensed in 0.2 ml portions into sterile bottles using a measured pipette. The bottles are dried at a temperature of +37°C for 24-48 hours and sterilized under the action of ultraviolet rays for 90-120 minutes. The tubes are sealed with sterile rubber stoppers and then with metal caps. Thus, the developed medium presents a sterile micro-pellicle fixed at the bottom of a 10.0 mL bottle.

RESULTS

For the determination of *P. aeruginosa* bacteria in the bottle with the medium, 2.0 ml of sterile distilled water and the strain of *P. aeruginosa* in a concentration of 10^1 .... 10^9 are applied. The medium dissolves within 2-3 minutes, and then it is incubated in a thermostat at a temperature of 37°C for up to 9 hours.

In the presence of *P. aeruginosa* bacteria in the examined material, the color of the mixture in the bottle changes from green to blue. Meanwhile, in the control bottle, the color remains unchanged (remains green).

The time for the detection of *P. aeruginosa* bacteria depends on the initial concentration of the germs in one milliliter of the examined material. The detection of individual P. aeruginosa cells is possible after 9-24 hours of incubation, while the detection of concentrations of 10^4-10^5 CFU/mL takes 5-6 hours of incubation at a temperature of 37°C (tab. 1).

We determined the sensitivity and selectivity of the proposed medium. To assess the selectivity of the medium, we conducted a series of experiments with four batches of microorganisms in association, in 32 repetitions. When forming the associations, we selected the microorganisms deemed unacceptable in the majority of medications.

I. *P. aeruginosa* ATCC 27853 (10^2) + *S. aureus* ATCC 25923 (10^6); *P. aeruginosa* ATCC 27853 (10^2) + *E. coli* ATCC 25922 (10^6); *P. aeruginosa* ATCC 27853 (10^2) + *S. epidermidis* ATCC 149990 (10^6).

II. *P. aeruginosa* ATCC 27853 (10^3) + *S. aureus* ATCC 25923 (10^6); *P. aeruginosa* ATCC 27853 (10^3) + *E. coli* ATCC 25922 (10^6); *P. aeruginosa* ATCC 27853 (10^3) + *S. epidermidis* ATCC 149990 (10^6).

III. *P. aeruginosa* ATCC 27853 (10^4) + *S. aureus* ATCC 25923 (10^6); *P. aeruginosa* ATCC 27853 (10^4) + *E. coli* ATCC 25922 (10^6); *P. aeruginosa* ATCC 27853 (10^4) + *S. epidermidis* ATCC 149990 (10^6).

IV. *P. aeruginosa* ATCC 27853 (10^5) + *S. aureus* ATCC 25923 (10^6); *P. aeruginosa* ATCC 27853 (10^5) + *E. coli* ATCC 25922 (10^6); *P. aeruginosa* ATCC 27853 (10^5) + *S. epidermidis* ATCC 149990 (10^6) (tab. 2).

Table 1. The time of detection of *P. aeruginosa* based on their initial concentration (CFU/mL).

Microorganism concentration (CFU/mL)	**Detection time, hours**									
	1	2	3	4	5	6	7	8	9	24
10^1	-	-	-	-	-	-	-	+	++	++
10^2	-	-	-	-	-	-	-	+	++	++
10^3	-	-	-	-	-	-	-	++	++	++
10^4	-	-	-	-	-	+	++	++	++	++
10^5	-	-	-	-	+	++	++	++	++	++
10^6	-	-	+	++	++	++	++	++	++	++
10^7	-	+	+	++	++	++	++	++	++	++
10^8	-	-	+	++	++	++	++	++	++	++
10^9	-	+	++	++	++	++	++	++	++	++
>2 mld.	++	++	++	++	++	++	++	++	++	++

Note: „++" – pronounced positive reaction; „+" – positive reaction; „-" – negative reaction.

Table 2. The selectivity of the culture medium.

Microbial species	**Exp. No.**	**Detection, hours**			**p**	
		6	9	24	6.9	9.24
***P. aeruginosa* (10^2) *S. aureus* (10^6)**	32	0	90.6±1.9	96.9±2.1	-	<0.05
***P. aeruginosa* (10^2) *E. coli* (10^6)**	32	0	93.8±2.0	100±0.0	-	<0.05
***P. aeruginosa* (10^2) *S. epidermidis* (10^6)**	32	0	87.5±1.7	96.9±2.1	-	<0.05
***P. aeruginosa* (10^3) *S. aureus* (10^6)**	32	0	93.8±2.0	100±0.0	-	<0.05
***P. aeruginosa* (10^3) *E. coli* (10^6)**	32	0	96.9±2.1	96.9±2.1	-	<0.05
***P. aeruginosa* (10^3) *S. epidermidis* (10^6)**	32	0	100±0.0	100±0.0	-	-
***P. aeruginosa* (10^4) *S. aureus* (10^6)**	32	9.4±0.6	100±0.0	100±0.0	<0.001	-
***P. aeruginosa* (10^4) *E. coli* (10^6)**	32	6.2±0.4	96,9±2.1	96.9±2.1	<0.001	<0.05
***P. aeruginosa* (10^4) *S. epidermidiss* (10^6)**	32	9.4±0.6	96.9±2.1	100±0.0	<0.001	<0.05
***P. aeruginosa* (10^5) *S. aureus* (10^6)**	32	28.1±1.1	96.9±2.1	100±0.0	<0.001	<0.05
***P. aeruginosa* (10^5) *E. coli* (10^6)**	32	31.3±1.2	100±0.0	100±0.0	<0.001	-
***P. aeruginosa* (10^5) *S. epidermidis* (10^6)**	32	28.1±1.1	96.9±2.1	100±0.0	<0.001	<0.05

Based on the obtained results, we have determined that the medium exhibits selectivity towards *P. aeruginosa* bacteria, depending on the initial concentration of pseudomonads and the microorganisms in association.

To determine the sensitivity of the medium, we conducted a series of experiments with four reference strains of *P. aeruginosa* at two concentrations, 10^4 and 10^5 CFU/mL, in 14 repetitions. In parallel, we carried out the cultivation of bacteria on blood agar and peptone agar media (tab. 3).

Experimentally, we determined that the proposed medium is much more sensitive compared to blood agar and peptone agar media, allowing for the detection of *P. aeruginosa* bacteria within 6 hours.

The composition and optimal ratio of ingredients create a medium in the form of a micro-pellicle fixed at the bottom of a bottle, which serves as both a container for preserving the medium and, at the same time, for conducting the analysis. The storage term of the medium is 2 years (observation period).

Table 3. The sensitivity of the detection of *P. aeruginosa* bacteria.

Microbial species	Exp. No.	The concentration of microorganisms CFU/ml and their detection after 6 hours of incubation at 37ºC					
		The micro-pellicle medium		Blood agar		Peptone agar	
		10^4	10^5	10^4	10^5	10^4	10^5
***P. aeruginosa* ATCC 27853**	14	71.4±1.3	100±0.0	0	0	0	0
***P. aeruginosa* ATCC 15442**	14	83.3±1.4	100±0.0	0	0	0	0
***P. aeruginosa* ATCC 49189**	14	70.0±1.4	100±0.0	0	0	0	0
***P. aeruginosa* NCTC 6750**	14	80.0±1.6	100±0.0	0	0	0	0

DISCUSSIONS

The control of "microbiological purity" carried out in support of pharmaceutical and biopharmaceutical production falls into three main categories: indication (qualitative), enumeration (quantitative), and characterization/identification. Traditional microbiological methods are listed in compendia and discussed using conventional techniques based on growth and multiplication, which are bulky and time-consuming. Generally, such tests require several days of incubation to determine microbial contamination, and therefore, managing them can lead to proactive corrective measures. Additionally, microbial growth is limited by the culture medium used and incubation conditions, influencing the sensitivity, specificity, and reproducibility of testing (6).

The most discussed topics revolve around the development of various technological platforms for rapid microbiological methods, and many have been readily adopted by microbiology laboratories and the pharmaceutical industry. Their use could enable drug companies to adapt to deadlines for manufacturing processes and product release. Some rapid methods also provide the possibility of real-time microbiological control, allowing management to respond to microbial contamination events over a more extended period and potentially offering cost savings and increased efficiency in quality control testing laboratories. Despite the numerous proven advantages in quality management and the initiatives of international associations to promote the use of analytical process technology, including rapid microbiological methods, the pharmaceutical and biopharmaceutical industry has been somewhat slow to embrace alternative microbiological control methodologies due to reported divergent results (6, 7).

The use of rapid methods is a dynamic field in applied microbiology that has garnered increased attention both nationally and internationally over time (8, 9, 10). This topic has been widely addressed in conferences and published documents worldwide. More recently, the use of alternative methods for microbiological quality control of

pharmaceutical products and materials used in pharmaceutical production has been extensively discussed in various guidelines and compendia in an attempt to facilitate the implementation of these technologies by pharmaceutical companies (6, 11).

Based on the above, the issue of developing procedures and means for the rapid detection and identification of bacteria from the *Pseudomonas spp., Staphylococcus, Candida*, and *Enterobacteriaceae* groups, as microorganisms unacceptable in medications, is current and aligns with the purpose of the proposed study.

The procedure pertains to culture media for the detection of *Pseudomonas aeruginosa* bacteria in various samples, environmental objects, including human pharmaceuticals. In technical essence, closer is the reagent for the detection of *P. aeruginosa* bacteria, which contains all optimal ingredients, with reference to the specific substrate of sodium citrate, used for rapid detection within 9 hours of incubation at a temperature of 37°C. The disadvantage of the known reagent is its lower specificity, as it allows the detection of other citrate-positive microbial species.

The problem addressed by the proposed procedure lies in the development of a culture medium that allows a significant improvement in the specificity of detecting *Pseudomonas aeruginosa* bacteria.

A new culture medium is proposed that includes all optimal ingredients for the detection of *P. aeruginosa* bacteria, with sodium malonate as a specific substrate, water-soluble bromothymol blue as an indicator, sodium hydrogen phosphate, and potassium dihydrogen phosphate. The results of the conducted studies have shown an increase in the specificity of the medium by including sodium malonate as a specific substrate, which *P. aeruginosa* uses as the sole carbon source for multiplication. The other ingredients are included to promote and facilitate the multiplication of P. aeruginosa. Detection occurs under the pH conditions formed by sodium hydrogen phosphate and potassium dihydrogen phosphate, and the cleavage products of sodium malonate are indicated using bromothymolblue.

CONCLUSIONS

1. The use of the developed procedure allows for the rapid and specific isolation of P. aeruginosa strains and facilitates timely monitoring of pharmaceutical preparations and resources to limit contamination throughout the pharmaceutical manufacturing process.
2. The conducted studies indicate a clear increase in the sensitivity and speed of detection of P. aeruginosa bacteria in pure culture, mixed culture, or other research materials.
3. The described procedure is simple to implement, cost-effective, and can be used in microbiological laboratories of various levels.

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BACTERIAL NEUROLOGICAL DISEASES INFLUENCED BY GLOBAL WARMING

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Keywords: *central nervous system, bacterial diseases, neuroinfections.*

Introduction. *In recent years, the incidence of infectious diseases affecting the central nervous system has increased. Among the reasons for the expansion of these diseases and the emergence of new neuropathogens are globalization, global warming, increased proximity between humans and wild animals due to human activities such as deforestation, the rising cases of antimicrobial resistance, etc.*

Material and methods. *A narrative synthesis of specialized bibliographic references was conducted, selecting sources from Google Scholar and ResearchGate using the keywords "bacterial diseases of the nervous system," "neuroinfections," and "global warming" in English. A total of 39 bibliographic sources were identified, excluding 3 abstracts and 4 duplicated articles. Additionally, 19 irrelevant articles were excluded, and 4 articles highlighting the topicality of the subject were added.*

Results. *Bacterial diseases affecting the nervous system tend to become increasingly severe due to the critical functions performed by the brain, spinal cord, peripheral nerves, and cranial nerves. Infections can occur in nervous tissue or in the meninges (covering membranes). Specialized literature mentions that there is no direct connection between neuroinfections and global warming. The development of neuroinfections is linked to exposure to bacterial agents in the environment and is associated with human behaviors. Although global warming can influence the distribution of disease vectors, there is a significant distance between this phenomenon and the direct transmission of neuroinfections. Global warming can impact human health in various ways, including the expansion of the range of vector-borne diseases or changes in the distribution of pathogens. However, these connections are complex and depend on numerous factors.*

Conclusions. *The effects of global warming can enhance the adverse impact of microorganisms on the nervous system.*

Cuvinte-cheie: *sistem nervos central, maladii bacteriene, neuroinfecții.*

MALADII BACTERIENE NEUROLOGICE INFLUENȚATE DE ÎNCĂLZIREA GLOBALĂ

Introducere. *În ultimii ani a crescut incidența bolilor infecțioase care afectează sistemul nervos central. Printre motivele expansiunii acestor boli și apariției de noi neuropatogeni se numără globalizarea, încălzirea globală, apropierea crescută dintre oameni și animalele sălbatice din cauza activităților umane precum defrișările, creșterea cazurilor de rezistență antimicrobiană etc.*

Material și metode. *A fost realizată o sinteză narativă a referințelor bibliografice de specialitate. Sursele au fost selectate în Google Scholar și ResearchGate folosind cuvintele-cheie „maladii bacteriene ale sistemului nervos", „neuroinfecții" și „încălzire globală" în limba engleză. Astfel au fost identificate 39 de surse bibliografice, dintre care au fost excluse trei abstracte, patru articole dublate, 19 articole netematice și au fost adăugate patru articole ce reflectă actualitatea temei.*

Rezultate. *Bolile bacteriene, care afectează sistemul nervos, au tendință de a deveni tot mai grave din cauza funcțiilor critice pe care le îndeplinesc creierul, măduva spinării, nervii periferici și cranieni. Infecțiile pot apărea în țesutul nervos sau în meninge (membranele de acoperire ale creierului). În literatura de specialitate se menționează că nu există o conexiune directă între neuroinfecții și încălzirea globală. Dezvoltarea neuroinfecțiilor are legătură cu expunerea la agenții bacterieni din mediu și cu comportamentele umane. Deși încălzirea globală poate influența distribuția unor vectori ai bolilor, există o distanță semnificativă între acest fenomen și transmiterea directă a neuroinfecțiilor. Încălzirea globală poate afecta sănătatea umană într-o varietate de moduri, inclusiv prin extinderea ariei de răspândire a unor boli transmise de vectori sau prin schimbări în distribuția unor agenți patogeni, dar aceste legături sunt complexe și depind de numeroși factori.*

Concluzii. *Efectele încălzirii globale potențează acțiunea nefavorabilă a microorganismelor asupra sistemului nervos.*

INTRODUCTION

The progress in human health and diseases has been substantial in today's world, with a growing emphasis on exploring the causal relationships between various illnesses and diverse causative factors, both internal and external. There is limited conclusive data or few pieces of evidence to suggest that climate change/global warming is responsible for altering disease patterns. However, extreme temperature fluctuations, such as heatwaves and cold spells, coupled with changes in precipitation levels leading to famine and floods, and the presence of airborne allergens with increased air pollution, impact human health at an individual level. Vector-borne diseases are directly affected by changes in temperature and precipitation levels. Water-related diseases arising from poor water quality, its quantity, and insufficient personal hygiene have complex associations. The loss of human lives, damage to health infrastructure, and public properties due to recent disasters like storms, cyclones, and hurricanes serve as a warning for the future world to address this situation now or never. On the other hand, the vulnerable population in low- or middle-income countries bears the brunt of the health burden related to global warming (1). Together, these factors will have adverse consequences on the health status of the population, including the health of the human brain (2).

Neurotropism, which affects the normal function of the brain, is characteristic of certain microorganisms such as prions, viruses, bacteria, fungi, and parasites (3). Bacterial infections of the central nervous system are significant causes of morbidity and mortality. Neuroinfections caused by these agents activate the immune response, inducing neuroinflammation, excitotoxicity, and neurodegeneration. Purinergic signaling is an evolutionarily conserved signaling pathway associated with these neuropathologies (4).

Global warming is a phenomenon of great concern, given the increasing temperature and frequency of extreme weather conditions directly impacting all life on Earth. The planet's temperature is rising at an unprecedented rate, leading to the loss of biodiversity. Biodiversity is a term that encompasses both macro and microenvironments. Altered biodiversity, the duration of pollination seasons and geography, coupled with the production of harmful gases, released toxic substances, and chemicals, underscore the rise in disturbances related to the epithelial barrier (5, 6).

The incidence of infectious diseases affecting the central nervous system (CNS) has increased in recent years. Among the reasons for the expansion of these diseases and the emergence of new neuropathogens are globalization, global warming, and increased proximity between humans and wild animals due to human activities such as deforestation (4).

The anatomy of the brain and meninges determines the unique nature of infections of the central nervous system, often referred to as neuroinfections.

Many inflammatory conditions have been influenced by a disrupted epithelial barrier, best explained by the hypothesis of the epithelial barrier (6).

The increasingly frequent extreme weather events can synergistically damage the integrity of the epithelial barrier. A compromised epithelial barrier induces pro-inflammatory activation of epithelial cells. An "open" epithelial barrier contributes to the entry of the external exposome into and beneath the epithelium, triggering an expulsion response led by local inflammatory cells and chronic inflammation. These changes are associated with microbial dysbiosis with opportunistic pathogenic colonizers and decreased commensal presence. These cellular and molecular events are key mechanisms in the pathogenesis of numerous chronic inflammatory disorders (5, 7).

Thus, mitigating the impact on health should be tailored to regional health threats. Effective strategies need to be devised at all levels of the healthcare delivery system (1, 2).

The *aim* of the research was to analyze the issues related to bacterial diseases of the CNS in association with global warming.

MATERIAL AND METHODS

The research represents a narrative synthesis of specialized bibliographic references from both domestic and international sources. The synthesis relied on the theoretical evaluation of scholars' experiences regarding the addressed phenomenon. Sources were selected through the *Google Scholar* search engine and the *ResearchGate* social

network. The keywords "bacterial diseases of the nervous system," "neuroinfections," and "global warming" were used in English. The primary criterion for selecting bibliographic sources was the availability of full, free access to the articles.

The first stage of the work included selecting articles that contain two components of interest in the research: bacterial diseases of the nervous system and global warming. At this stage, 39 bibliographic sources were identified on *Google Scholar* and *ResearchGate*. In the *second stage*, 3 abstracts and 4 duplicated articles were excluded. After a detailed analysis of titles and abstracts (*third stage*), articles containing relevant information for the research topic with both requested components were selected. Thus, the final number for analysis consisted of 12 scientific articles, supplemented with 4 articles used in the introduction = 16 scientific articles (fig. 1).

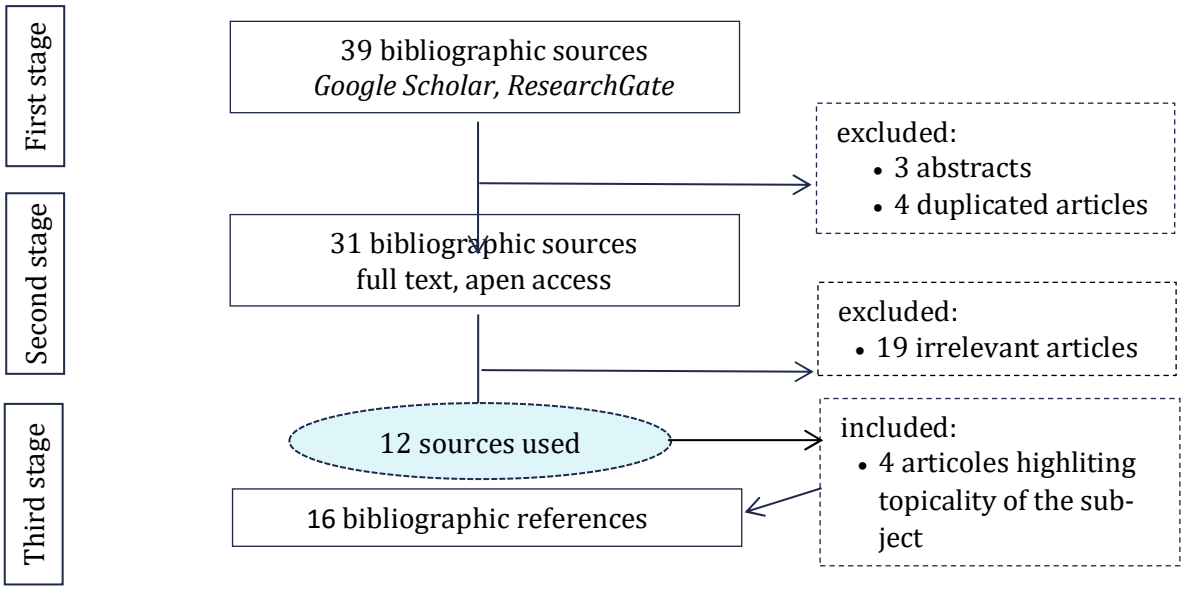

Figure 1. Article selection algorithm.

RESULTS

Bacterial diseases of the nervous system are very rare but can be life-threatening.

Microbiology of the nervous system

Bacterial diseases of the CNS continue to increase in incidence due to conditions contributing to the emergence of pathogens, such as global travel, climate changes, and human encroachment into animal territories. The severity and complexity of these diseases are influenced by the diversity of etiological agents and their routes of neuroinvasion (8).

Among the bacterial diseases affecting the nervous system are: meningococcal meningitis, pneumococcal meningitis, listeriosis, Lyme disease, tuberculoid leprosy, lepromatous leprosy, tetanus, botulism, etc. (4, 9). Epidural abscesses remain localized, while subdural abscesses spread over one of the cerebral hemispheres. Infections in the subarachnoid space tend to spread over the brain and spinal cord (8, 10).

The major bacterial causes of central nervous system infections are *Haemophilus influenzae, Streptococcus pneumoniae*, and *Neisseria meningitidis* (9). Most of these agents invade through the bloodstream. In these infections, the number of bacteria rapidly increases in the cerebrospinal fluid. Brain abscesses often present a mixed flora of aerobic and anaerobic bacteria (4).

The bacterial world and global warming

Microorganisms provide long- and short-term feedback reactions to global warming and climate changes, which can be both positive and negative. Since they can recycle and modify fundamental elements such as carbon and nitrogen that make up cells, they play a significant role as producers or consumers of these gases in the environment. In unpopulated areas that were previously too cold for bacteria to survive, they have been able to spread and can cause fatal infections (11).

New infections could arise as the gap between ambient temperatures and human body temperature narrows. Floods and other natural disasters are more likely, driven by climate changes, leading to an increased risk of bacterial growth in people's homes. In this context, certain bacteria can lead to severe contamination of the lungs and brain (11).

The capacity of the medical system to treat bacterial diseases is compromised by antimicrobial resistance (a threat in which microorganisms, especially bacteria, become resistant to antibiotics and disinfectants, thereby complicating the ability to treat and prevent infections). Resistance is often developed through sub-inhibitory exposures to drugs and/or disinfectants but can also be caused by (often negative) environmental changes as a response mechanism to environmental conditions such as air temperature, water salinity, metals (potential toxic elements), and organic pollutants. Possible mechanisms through which climate change could affect the transfer of antimicrobial resistance genes between bacteria include:

- direct action, through changes in air temperature and/or precipitation,
- indirect impact, such as changes in human populations, disease vectors, agricultural processes, water availability, glacial and hydrological processes (1).

The impact of global warming on neurological diseases

Exposure to heat can lead to hyperthermia and heatstroke in extreme cases, which can be fatal. As global temperatures continue to rise, the incidence of hyperthermia and heatstroke is also inevitably expected to increase. Hyperthermia, along with atmospheric changes associated with global warming, is linked to an increased prevalence of migraines, seizures, stroke, and certain forms of dementia, including Alzheimer's disease. Prolonged exposure to heat can elevate levels of proinflammatory cytokines, such as tumor necrosis factor-alpha (TNF-α) and interleukin-1 (IL-1), and may induce gliosis and a decrease in synaptic density in the brain (2).

With global warming and the extremely hot summers witnessed worldwide in recent years, there is an increase in the incidence of fatal encephalitis caused by the so-called "brain-eating amoebas." (12).

The major cause of global warming is greenhouse gases that trap the thermal energy reflected by the Earth's surface. The main greenhouse gases include carbon dioxide, methane, nitrous oxide, and ozone. A new bacterium called *Methylokorus infernorum* has been discovered, which could help address the greenhouse gas issue. Found in geothermal areas with acidic and hot conditions, this bacterium utilizes methane gas. These bacteria can consume vast amounts of methane, approximately 11 kg/year, and could be useful in reducing ethane emissions from methane-producing facilities and landfills (13).

Bacteria can be involved in stopping global warming

Global warming has direct and indirect opposing effects on the structure and function of the microbial community in permafrost lake sediments.

Northern lakes disproportionately influence the global carbon cycle and may do so even more in the future, depending on how their microbial communities respond to global warming. Microbial communities can change due to the direct effects of global warming on their metabolism and the indirect effects of global warming on the connectivity of groundwater from permafrost thaw in the surrounding area (2).

Bacteria play a crucial role in the global carbon, nitrogen, and methane cycles. They capture carbon from non-living sources, making it available for living organisms through the process of carbon fixation. Bacteria can cycle carbon molecules under anaerobic conditions to produce energy through the fermentation process. Certain bacteria, such as *Thiobacillus ferrooxidans*, sulfur bacteria, and *Clostridium butyricum*, may participate in the carbon cycle (which describes the continuous movement of carbon atoms from the atmosphere to Earth and back to the atmosphere) (11, 14).

The main component of air, gaseous nitrogen (N_2), enters the biosphere through biological fixation. Bacteria exhibit specificity to certain plants. As a result of attaching to one of the plant's root hairs, the bacterium creates a hollow tube that leads into the root. Through this tube, bacteria develop and eventually form a nodule on the root. For bacteria, the plant provides food and energy, while nitrogen from the air is supplied in a form usable by the plant through fixation by the bacteria. *Rhizobium trifolium* contains the nitrogenase

enzyme, which fixes atmospheric nitrogen into a form of ammonium ions that is chemically advantageous for higher species. The plant transforms the fixed ammonium ion into nitrogen oxides and amino acids to create proteins and other compounds like alkaloids, as part of their symbiotic association. Bacteria contribute to the collection of energy or the accumulation of nitrogen in a form necessary for their growth and development. Bacteria that consume methane (CH_4), a greenhouse gas, are imperative for maintaining Earth's climate stability. Methane serves as an energy source for bacteria metabolism (11).

DISCUSSIONS

Neuroinfections begin nonspecifically, without signs of involvement of the nervous system.

Bacterial diseases affecting the nervous system tend to become more severe due to the critical functions performed by the brain, spinal cord, peripheral nerves, and cranial nerves. Infections can occur in nervous tissue or in the meninges (covering membranes).

Climate change is a global existential challenge that has introduced unprecedented pressures on the planet, already translating into significant consequences for human health.

The study conducted in Canada, sampling 19 lakes along a 1600 km latitudinal transect covering all major permafrost regions, assessed the potential impacts of global warming through spatial substitution over time. The direct effects of regional warming, such as temperatures and permafrost coverage extent, were compared with its indirect impacts on local groundwater connectivity and lake water chemistry. It was found that methanogens and metabolically diverse Euryarchaeota genes involved in all major pathways of methanogenesis responded particularly positively to the direct effects of regional global warming (15).

Some authors mention that increases in sequences representing methanogens and methanogenesis could be partially compensated by greater groundwater connectivity associated with warmer temperatures, as the abundance of these sequences has been negatively correlated with groundwater connectivity (16).

The opposing views among scientists on different aspects of global warming help explain why considerable variation remains unexplained in methane emission predictions by Earth system models, even when microbial dynamics are considered. The potential methane production can be further enhanced in warmer locations due to the inadequate increase in its oxidation.

The specialized literature mentions that there is no direct connection between neuroinfections and global warming. The development of neuroinfections is linked to exposure to bacterial agents in the environment and is associated with human behavior. Although global warming can influence the distribution of disease vectors, there is a significant distance between this phenomenon and the direct transmission of neuroinfections. Global warming can impact human health in various ways, including expanding the range of vector-borne diseases or altering the distribution of pathogens, but these connections are complex and depend on numerous factors (6, 17).

Nevertheless, it is important to understand and address the consequences of global warming on human health, including potential public health risks and possible impacts on the distribution of infectious diseases. Studies in this field are ongoing to investigate these connections and to develop effective adaptation and mitigation strategies.

The increase in methanogenesis is supported by concurrent measurements of methane fluxes at the study sites. Together, the results provide a new understanding of why methane emissions are highly sensitive to rising temperatures in the study region. Future predictions of methane emissions could now be improved by considering how microbial dynamics vary with the direct and indirect effects of global warming associated with temperature and hydrology (14).
Environmental changes can create additional stress on microorganisms and the distribution of pathogens. Further research is needed to understand the microbiome, resistance, and environmental stress factors (such as temperature) that modify the impact, wastewater treatment, pathogens discharged into the environment, and the evolution and dissemination after exposure to ecological stress (1).

Diagnostic tests for neurological disorders often involve examining cerebrospinal fluid, and antibiotic therapy must use medications that can cross the blood-brain barrier.

CONCLUSIONS

1. Climate change represents a shift in balance, and the outcomes of the interaction between environmental factors and bacteria, the connections between these components, are complex and challenging to predict.
2. The effects of global warming enhance the unfavorable action of microorganisms on the nervous system.

CONFLICT OF INTEREST

Authors have no conflict of interest to declare.

ETHICAL APPROVAL

As the article does not involve ethical risks, approval from the Ethics Committee was not deemed necessary for the research.

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UNILATERAL DYSKINETIC CEREBRAL PALSY: TIME FOR A CHANGE IN CEREBRAL PALSY CLASSIFICATION *(CASE PRESENTATION)*

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Keywords: *dyskinesia, unilateral, case study, rehabilitation.*

Introduction. *Unilateral dyskinesia is a rare neurological disorder characterized by involuntary, abnormal movements on one side of the body. In children, understanding its clinical presentation and age-appropriate rehabilitation strategies is essential for the best outcome.*

Material and methods. *In this case-presentation, we conducted an in-depth examination of a 10-year-old boy with unilateral dyskinesia, previously diagnosed with bilateral spastic cerebral palsy. Extensive pediatric neurological assessments, including neuroimaging and child-friendly clinical evaluations, were performed to comprehensively document the patient's condition.*

Results. *Birth data: 39 weeks gestation, singleton, weight – 3950 g, length – 56 cm, head circumference – 36 cm, Apgar score at 5 minutes – 7, without birth-related complications. Paraclinical investigations revealed MRI-identified basal ganglia lesions in the putamen. Functional classifications indicated GMFCS II, MACS II, epilepsy, normal vision, normal hearing, EDACS level I, Viking speech scale III, and moderate intellectual disability. The pediatric patient exhibited pronounced dyskinesia limited to the right side of the body, which has progressively worsened over the past six years.*

Conclusions. *The results are in line with specific variables of dyskinetic CP. This study emphasizes the challenges in diagnosing pediatric patients with unilateral dyskinesia during the early stages. Clinical features are evident; therefore, rehabilitation strategies should be selected accordingly.*

Cuvinte-cheie: *dischinezie, unila-teral, studiu de caz, reabilitare.*

PARALIZIA CEREBRALĂ DISCHINETICĂ UNILATERALĂ IMPUNE O SCHIMBARE A CLASIFICĂRII PC *(STUDIU DE CAZ)*

Introducere. *Dischinezia unilaterală este o afecțiune neurologică rară, caracterizată prin mișcări involuntare, anormale pe o parte a corpului. La copii, înțelegerea prezentării sale clinice și a strategiilor de reabilitare adecvate vârstei este esențială pentru obținerea rezultatului scontat.*

Material și metode. *În prezentarea de caz, a fost efectuată o examinare aprofundată a unui băiat de 10 ani cu dischinezie unilaterală, diagnosticat anterior cu paralizie cerebrală spastică bilaterală. Pentru a documenta în mod cuprinzător starea pacientului, au fost efectuate evaluări neurologice, pediatrice extensive, inclusiv neuroimagistice, și evaluări clinice prietenoase pentru copii.*

Rezultate. *Date la naștere: 39 de săptămâni de gestație, greutate – 3950 g, talie – 56 cm, perimetrul cranian – 36 cm, scor Apgar la 5 min. – 7, fără complicații legate de naștere. Cu ajutorul RMN s-au evidențiat leziuni ale ganglionilor bazali în putamen. Clasificările funcționale au indicat GMFCS II, MACS II, epilepsie, vedere normală, auz normal, EDACS I, scara de vorbire Viking III și dizabilitate intelectuală moderată. Pacientul pediatric a prezentat dischinezie pronunțată, limitată la partea dreaptă a corpului care s-a agravat progresiv în ultimii șase ani.*

Concluzii. *Rezultatele sunt în concordanță cu specificul PC dischinetice. Acest studiu subliniază provocările în diagnosticarea pacienților pediatrici cu dischinezie unilaterală în stadiile incipiente. Caracteristicile clinice fiind evidente, strategiile de recuperare ar trebui selectate conform acestora.*

INTRODUCTION

Dyskinetic cerebral palsy (CP), a subtype of cerebral palsy marked by uncontrolled, irregular movements, arises from damage to the extrapyramidal regions of the brain responsible for movement coordination. This particular form of CP is relatively uncommon, constituting between 6% and 17% of all cerebral palsy cases (1, 2). Unilateral dyskinesia, an unusual manifestation of this disorder, is characterized by involuntary movements on one side of the body and presents unique management challenges. Children with dyskinetic CP generally experience more severe motor impairments and are more likely to have associated disabilities compared to those with spastic CP (3, 4). Recognized risk factors for dyskinetic CP include sentinel events during childbirth, and characteristic lesions in the basal ganglia and thalamus are typically observed in neuroimaging (4, 5). Despite a general decline in the overall birth prevalence of CP in high-income countries (6) over the last two decades, data indicates an increase in the prevalence of dyskinetic CP in Europe from the 1970s to the 1990s (7 – 12). It's uncertain whether the observed decrease in dyskinetic CP prevalence from 2005 to 2010 represents a sustained trend or a temporary dip. The rise in dyskinetic CP's birth prevalence towards the late 20th century (7), concurrent with the stable prevalence of spastic quadriplegic CP despite significant advances in obstetric and perinatal care, suggests that these CP subtypes predominantly originate antenatally and are rarely preventable. This perspective was further supported in 1998 when an international consensus statement identified dyskinetic CP or spastic quadriplegic CP as key criteria for defining an acute intrapartum event (13). Understanding and preventing dyskinetic CP remains complex and challenging. Given its severity and resistance to treatment, a comprehensive, multi-disciplinary approach is essential for the effective management and support of individuals with this condition.

CASE PRESENTATION

In this study, we conducted an in-depth examination of a 10-year-old boy with unilateral dyskinesia, previously diagnosed with bilateral spastic CP. Extensive pediatric neurological assessments, including neuroimaging and child-friendly clinical evaluations, were performed to comprehensively document the patient's condition.

1. Birth data

Born in 2013 at a level II hospital, the pregnancy was the first, and it resulted in a single birth. The gestational age was 39 weeks, with a weight of 3980 g, length of 56 cm, head circumference of 36 cm, chest circumference of 34 cm, and abdomen circumference of 33 cm. The APGAR score was 4/7. The birth was through caesarean section due to hypoxia caused by a circular umbilical cord around the neck. The newborn was transferred to a tertiary level hospital on the same day and was hospitalized for 14 days in intensive care. The diagnosis at birth was moderate asphyxia and cerebral ischemia grade I-II, along with a convulsive syndrome. The child underwent repeated rehabilitation treatments and on 15.05.15, Botulinum Toxin Type A (Dysport) 200 IU was administered.

2. Clinical and paraclinical evaluations:

1. Cerebral Computed Tomography 2015 - cystiform dilatation of the cistern magna is determined. 2. Electroencephalography (EEG). During artifact-free periods, disorganized brain bioelectric activity is noted, without pathological interhemispheric asymmetry and epiphenomena. Functional probe reactivity attenuated. 3. M-Echo, 12.2023 structural averages are not deviated LS=LD=61mm Ventricle III=3 mm 4. Nuclear Magnetic Resonance (NMR), 2015 - Lesions of the basal ganglia in the putamen 5. Ophthalmologist consultation-07.2015 F.O. Moderate dilated veins, 03.2019 F.O. No changes.

3. Current functional assessments

3.1. Neurological status:

During the first year of life with a delay in motor development: the child was not sitting at 6-8 months, at 1.5 years he was not walking. At the age of 1 year, the diagnosis of Cerebral Palsy in the form of Spasticity was established. Since birth, he has had seizures, treated with Clonazepam. Last seizure 2 years ago. Last treatment in 12.2023. Hypertonic muscle tone, emphasis on the left. Bilateral muscle strength in upper limbs slightly diminished D=S=4p. In the bilateral lower limbs proximally 3 points; distal flexion 4 points; extension 2 points. ROT live superior D=S, inferior: patellar -exaggerated; Achilles-D=S; Babinski positive bilaterally, more pronounced on the left side. Unstable Romberg pose, the coordination tests performed with more pronounced inaccuracy on the right (when performing the index-

nose test, a tremor appears in the right hand).

3.2. Speech therapist evaluation
Dysarthria, global language disorders grade II. Viking speech scale III. EDACS (Eating and Drinking Ability Classification) level I.

3.3. Psycho-pedagogue evaluation
Purpose and objectives - training and development of attention. Conclusion - the cognitive level does not correspond to the chronological age, IQ=62.

3.4. Gross motor function evaluation
The patient is diagnosed with Cerebral Palsy, bilateral spastic form. After a more detailed analysis of the gross motor function and the biped movement pattern, the patient presents a mixed form - spatial form on the left side and dyskinetic - on the right. The patient experiences difficulties while moving, finding it hard to maintain balance with a swaying, inclined movement on the left side. While walking, the child stabilizes the right leg with the left upper limb. During movement with the legs, they go into more pronounced hyperextension on the right, and the patient has an asymmetrical posture with an inclination on the left side. Movement control is better in the left hemisphere. Gross Motor Function Level II (GMFCS II).

3.5. Assessment of fine motor function
While performing simple abduction and adduction movements, the patient demonstrates more concrete, coordinated, and precise movement with the left hand than with the right. The left side, being spastic, is better controlled by the patient than the right side, which is dyskinetic. When reaching for a fitball, the right hand is placed ahead of the left. When holding the ball, the left hand is placed below, the right above, and the head looks to the right. When holding an object with the left hand, the right hand goes into abduction, supination, and dorsiflexion, which demonstrates that the extrapyramidal system is damaged, and the brain eliminates the presence of the right hand. However, if asked to hold his hand close to his body, he struggles, and movements with his left hand become clumsier. With his right hand, he can only grasp and hold objects the size of a ping-pong ball and then throw it. The interdigital grip and the terminal opposition grip cannot be performed with the right hand. With the left hand, he performs prehension through terminal opposition only with the ring finger and the little finger, and with the index and the medius only by sub-terminal opposition. When building a wall with blocks, he holds the pieces very tightly with his right hand due to the lack of co-contraction, while with his left hand he makes an effort to join the pieces. Fine movements of the hand and fingers are performed with difficulty and may be accompanied by intentional tremors. Manual Ability Classification (MACS) level II.

Lower limbs
Standing and walking are achieved with a large base of support. During the orthostatic position, support is performed only on the left leg, with the right leg initially lifted and then the heel lowered. The patient cannot maintain a standing position without movement for more than 3 seconds. Due to the lack of coordination of the activity of agonists, antagonists, and synergists, movements become uncontrolled and are carried out with high amplitude. This results in a “mowing gait” characterized by an abduction pattern along with dorsiflexion and eversion of the leg, and as compensation, hyperextension of the hip and knee. In activities of daily living (ADL), all actions are performed with the left hand, with the right hand not involved in the action. During dressing and undressing, the left hand engages and dresses the right hand, while the right hand is put aside, with the feet fixed on the floor. When putting on and taking off pants, socks, and shoes, coordinating the right leg is difficult. He manages, with difficulty, to fasten and unfasten buttons, and can easily unfasten and fasten zippers.

DISSCUSIONS

When conducting the rehabilitation process for dyskinetic cerebral palsy, whether it is unilateral or bilateral, it must be customized to fit each person's individual needs, abilities, and limitations. However, there are some general differences in the rehabilitation approach between the two types:

Unilateral Dyskinetic Cerebral Palsy: 1. Specific Focus: Rehabilitation will often be more focused on the part the affected body, which allows an intensive concentration on improving the function and coordination of that part. 2. Physical and Occupational Therapy: It will be aimed at improving strength, flexibility, and coordination of the affected body part. Exercises and activities are often designed to stimulate the use of the affected limb

and to encourage the development of fine and gross motor skills. 3. Use of Assistive Equipment: May be required on the affected side, such as orthotics or other support devices, to improve alignment and operation. 4. Compensatory Strategies: Patients can learn to use more effectively the unaffected side of the body to compensate and perform daily activities.

Bilateral Dyskinetic Cerebral Palsy: 1. Broad Approach: Rehabilitation requires a more global approach because both parts of the body are affected. This involves more coordination complex and careful planning to address the needs on both sides of the body.2. Physical and Occupational Therapy: This will aim to improve motor control, strength, and coordination on both sides of the body. Therapists may use a variety of techniques, including whole-body exercises, activities involving both hands and feet, and relaxation techniques to control involuntary movements. 3. Extensive Adaptations in Daily Life: Due to bilateral involvement, a wider range of assistive equipment and modifications of the home or school environment may be necessary to facilitate independence and safety. 4. Management of Involuntary Movements: Complex approaches may be required for managing uncontrolled movements, including medication, therapy with controlled movement, and in some cases, surgery.

Common Considerations: 1. Speech and Language Therapy: Both types can benefit from speech and language therapy, especially if facial muscle control is affected and coordination is needed for speech and eating. 2. Psychosocial Support: Emotional and psychological support is crucial in both cases, as cerebral palsy can have a significant impact on self-esteem, independence, and quality of life. 3. Multidisciplinary Approach: A multidisciplinary team involving doctors, therapists, educators, and other specialist services is essential to provide comprehensive care.

CONCLUSIONS

1. The results are in line with specific variables of dyskinetic CP. This study emphasizes the challenges in diagnosing and rehabilitating pediatric patients with unilateral dyskinesia during the early stages. In conclusion, although there are common basic principles in CP rehabilitation, the specific approaches and intensity of interventions may vary considerably between unilateral and bilateral cases.
2. An individualized assessment is essential for creating a personalized treatment plan to effectively address each person's needs and goals.

CONFLICT OF INTEREST

Authors have no conflict of interest to declare.

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We have obtained permission from the parents to present this information.

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CHANGES IN YOUNG PEOPLE'S ATTITUDES TO THE PROBLEM OF COVID-19, FLU, AND OTHER UPPER RESPIRATORY INFECTIONS DURING THE WAR IN UKRAINE

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Keywords:
COVID-19, pandemic, war, antiepidemic measures, Ukraine.

Introduction. *The population of all countries of the world was affected by the COVID-19 pandemic, but the most severe consequences were for low- and middle-income countries (LMICs). Following Russia's invasion of Ukraine, there have changes in people's attitudes to the challenges posed by the COVID-19 pandemic. The aim is to study the attitude of young people to the problem of COVID-19, influenza, and other upper respiratory infections (URI) during the war in Ukraine.* ***Material and methods****. The materials consist of the results of an online survey of young people in Chernivtsi and neighboring regions of the western part of Ukraine in 2023/2024 (N=269). The methods of sociological analysis, descriptive statistics, pair correlation, and logistic regression using PIVOT and SPSS were applied.* ***Results.*** *According to the results, 65.1% of respondents suffered from COVID-19, with 70.3% of them being vaccinated. During the pandemic, only 4.2% of respondents admitted that they did nothing to prevent infection. Since the beginning of the war, this percentage increased to 43.2%, with an additional 8.2% citing a lack of opportunity for prevention. At the end of the second year of the war, only 25.7% of respondents expressed concern about COVID-19, influenza, and other URIs. A direct correlation was found between this concern and the problem of restricting men's travel abroad during the pandemic (r=0.239), psychological issues (r=0.198), and financial problems (r=0.224) among females. Female anxiety was also influenced by job loss (r=0.152) and engagement in activities such as volunteering in aid delivery to the front (r=0.288) and cooking food for soldiers (r=0.152). Significance was established at the 95% CI, p<0.05.* ***Conclusions.*** *At the end of the second year of the war, the situation with COVID-19, influenza and other URIs was a concern of only one in four respondents (25.7%), which was also associated with vaccination (70%) and a transmitted disease (65%).*

Cuvinte-cheie:
COVID-19, pandemie, război, măsuri antiepidemice, Ucraina.

SCHIMBĂRI ÎN ATITUDINEA TINERILOR FAȚĂ DE COVID-19, GRIPĂ ȘI ALTE INFECȚII ALE CĂILOR RESPIRATORII SUPERIOARE ÎN TIMPUL RĂZBOIULUI DIN UCRAINA

Introducere. *Populația tuturor țărilor lumii a fost afectată de pandemia COVID-19, îndeosebi cea din țările cu venituri mici și mijlocii. Invazia Rusiei în Ucraina a indus schimbări în atitudinea oamenilor față de provocările pandemiei COVID-19. Scopul prezentului studiu constă în elucidarea atitudinii tinerilor față de COVID-19, gripă și alte infecții ale căilor respiratorii superioare în timpul războiului din Ucraina.* ***Material și metode.*** *Rezultatele unui sondaj online al tinerilor din Cernăuți și din regiunile învecinate din vestul Ucrainei din perioada 2023/2024 (N269) au fost analizate aplicând metode de sociologie, statistici descriptive, corelație de perechi, regresie logistică folosind PIVOT, SPSS.* ***Rezultate.*** *Sondajul a arătat că 65,1% dintre respondenți au suferit de COVID-19, dintre care 70,3% au fost vaccinați. În timpul pandemiei, doar 4,2% dintre respondenți au recunoscut că nu au făcut nimic pentru a nu se infecta, iar de la începutul războiului – 43,2%, 8,2% din respondenți motivând inactivitatea de prevenție prin faptul că „nu există posibilitate". La sfârșitul celui de-al doilea an de război, situația privind COVID-19, gripa și alte URI-uri a provocat îngrijorare doar pentru 25,7% dintre respondenți. În cazul bărbaților, o corelație directă a fost stabilită între această preocupare și restricționarea călătoriilor bărbaților în străinătate în timpul pandemiei (r=0,239). Pentru femei mai acute sunt problemele psihologice (r=0,198) și financiare (r=0,224). Anxietatea femeilor a fost determinată și de pierderea locului de muncă (r=0,152), de livrarea de ajutoare pe front (r=0,288) și gătitul mâncării pentru soldați (r=0,152). Semnificația a fost aleasă la nivelul de 95% IÎ, p<0,05.* ***Concluzii.*** *La sfârșitul celui de-al doilea an de război, situația privind COVID-19, gripa și alte infecții ale căilor respiratorii superioare era o preocupare doar pentru unul din patru respondenți (25.7%), fapt determinat de vaccinare (70%) și de caracterul transmisibil al acestor afecțiuni (65%).*

INTRODUCTION

Previously, it was believed that low- and middle-income countries (LMICs) faced a triple problem in healthcare, namely, major non-communicable diseases and road traffic accidents were added to the traditional infectious disease burden (1). However, the COVID-19 pandemic, against the background of the socio-economic crisis it caused, has made its own adjustments. It has deepened existing disparities between countries, posed challenges for management at various levels, and exacerbated the adverse effects of both communicable and non-communicable diseases (2, 3).

The WHO has emphasized that, although the COVID-19 pandemic has affected the populations of all countries around the world, the most severe consequences have been felt by those who were already in a vulnerable state, such as those with poorer health and less access to necessary quality healthcare. This group has suffered the most due to anti-epidemic restrictions, particularly in LMICs (4). Despite the fact that the COVID-19 pandemic is no longer considered a public health emergency of international concern, the disease continues to pose a serious threat to human health globally. With the pandemic now spanning four years, new strains of the virus are emerging, and WHO experts predict that it will persist for a significant period of time (5). The latest Human Development Report 2021/22 has highlighted the "stunning reality" of a two-year decline in the global Human Development Index (HDI) due to the COVID-19 pandemic, which has led to an unprecedented synchronized multidimensional crisis (6). Ukraine, despite belonging to the LMICs in terms of per capita income, has demonstrated an almost steady increase in the HDI since 1996. However, in 2020, progress gave way to regression, with the main factor being a reduction in life expectancy, primarily as a result of the pandemic (7).

The situation in the country worsened significantly when, amidst the COVID-19 pandemic, the Russian armed forces launched a large-scale invasion of Ukraine on February 24, 2022. Civilian casualties increased, and large areas came under occupation. More than 15 million civilians were displaced within Ukraine or abroad (8). Military shelling of civilian objects continues in Ukraine. The humanitarian and psychological crises in areas near and far from the war zone worsened during the winter of 2023 due to repeated bombardments of civilian infrastructure (9).

Against the backdrop of military operations, the public's attitude toward disease prevention, compliance with anti-epidemic measures, and their implementation face significant challenges. *The purpose* of our study was to examine the attitudes of young people toward COVID-19, influenza, and other upper respiratory infections (URIs) during the period of martial law in Ukraine.

MATERIAL AND METHODS

The materials were based on the results of an online survey of young people in Chernivtsi and nearby regions of the western part of Ukraine. The participants included students (3-6 years), graduates of Bukovinian State Medical University (BSMU), and individuals from other educational institutions. The survey, titled "Let's summarize," was conducted through a Google form from December 20, 2023, to January 03, 2024, and involved 269 respondents, comprising 68 males and 201 females. For comparative analysis, the results of an online survey conducted by the Ptukha Institute of Demography and Social Studies of the National Academy of Sciences of Ukraine in May 2022 were utilized. This survey encompassed 523 respondents (130 males and 393 females) from 16 educational institutions across various regions of Ukraine, aiming to investigate the impact of the COVID-19 pandemic and the Russo-Ukrainian war on the lives and health of students. The specifically designed "Let's summarize" questionnaire included questions about the most significant challenges young people encountered during the COVID-19 pandemic (2020/21), the full-scale invasion (from 02/24/2022), and at the end of the second year of the war (2023/24). A separate section addressed whether the current situation with COVID-19, influenza, and other upper respiratory infections as of early 2024 is a cause for concern. Respondents could choose multiple answers, and open-ended questions were also included. The research employed sociological methods, descriptive statistics using PIVOT, as well as the method of pairwise correlation, logistic regression, with the determination of the probability of an event (P) using the SPSS statistical program. Reliability was set at the 95% confidence interval with $p<0.05$. To compare the frequency of individual responses among males and females, the *Student's t*-test was used for relative values.

RESULTS

The respondents were requested to outline their individual challenges over the past 4 years in light of the COVID-19 pandemic and Russia's full-scale invasion with the outbreak of hostilities in Ukraine. The socio-demographic characteristics of the respondents and significant events that occurred in their lives during the war are detailed in Table 1. A majority of the respondents, constituting 92.2%, were young. Changes in the educational process (start/completion) were reported by 28.3% of respondents, while changes in employment were noted by 42.5%. Among those who found employment, 18.8% either lost their jobs or changed their field of activity. Additionally, one in ten respondents initiated a family, and 7.5% of female respondents gave birth to a child. According to the survey, a significant proportion of young people, up to 65.1% (64.7% males, 65.2% females) had contracted COVID-19, as confirmed by testing and clinical examination. For comparison purposes, in the 2022 survey, 36.3% (37.7% of males, 35.9% of females) reported having had the disease, confirmed by testing. At the time of the survey, 70.3% of respondents were vaccinated against COVID-19.

In addition to health problems, the pandemic has also caused financial difficulties, as indicated by more than a quarter of respondents in both the 2023/24 survey, with 25.3% (20.6% males; 26.9% females), and in the 2022 survey with 25.8% (23.1% males, 26.7% females). Financial problems associated with the spread of coronavirus disease were found to be less significant than the same problems caused by the war, as reported by 49.5% and 35.7% of respondents, respectively. A correlation was found between financial problems during the pandemic (2020-2021) and feelings of anxiety about the situation with COVID-19, influenza, and other URIs in early 2024 among females ($r=0.224$, $p<0.01$).

A correlation was observed between the presence of psychological health complaints, encompassing mood swings, stress, and depression, at the time of the survey and the financial problems that existed at the beginning of the war among males ($r=0.286$, $p<0.05$) and females ($r=0.174$, $p<0.05$). Financial problems during the 2020-2021 pandemic were found to be important for females' psychological health ($r=0.289$, $p<0.01$).

According to the results of the 2023/24 survey, 82.5% of respondents reported significant problems during the COVID-19 pandemic (2020-2021). The structure of these problems was predominantly associated with the restriction of usual communications, movement within the country, and foreign travel, accounting for 47.7% in total. A slightly smaller share (40.8%) was attributed to psychological problems, including fear of death, safety, and the health of relatives (fig. 1).

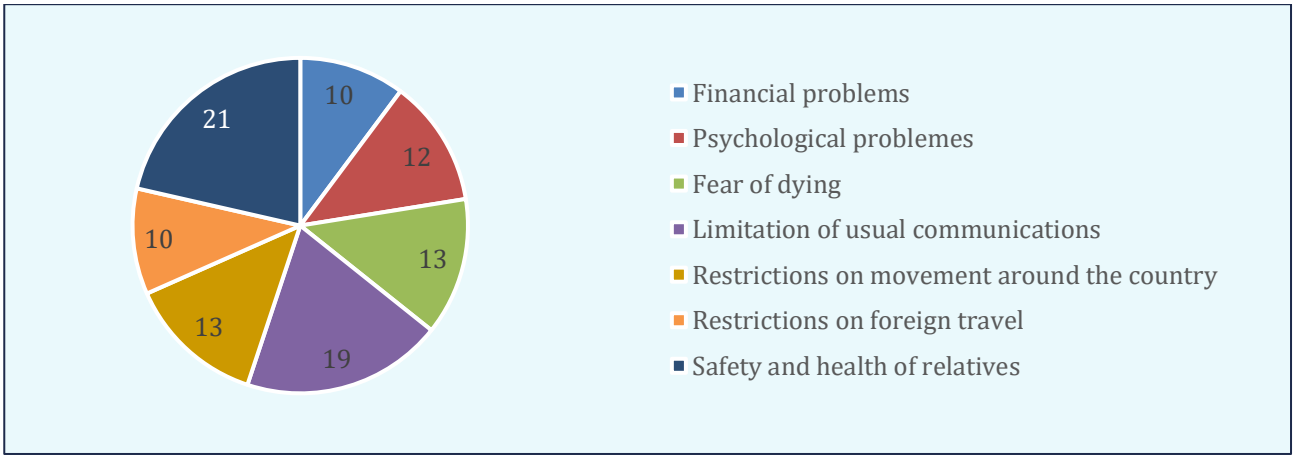

Figure 1. The structure of problems that occurred during the COVID-19 pandemic (2020-2021) according to respondents (survey 2023/24).

Psychological problems during the war were more relevant than during the pandemic and accounted for 58.5% of the structure. At the same time, the share of problems related to the restriction of usual communications and movement decreased from the beginning of the pandemic to 29.2% during the war (fig. 2).

Table 1. Socio-demographic characteristics of respondents.

Determinants		Quantity	Percentage (%)
Total		**269**	**100**
Males		68	25.3
Females		201	74.7
Age	18-24	209	77.7
	25-44	39	14.5
	45 and older	21	7.8
Education	Higher	85	31.6
	Incomplete higher	126	46.8
	Secondary specialized	32	11.9
	Secondary	26	9.67
Studying	***Total***	***211***	***78.4***
	Males	52	76.5
	Females	159	79.1
Working	***Total***	***133***	***49.4***
	Males	40	58.8
	Females	93	46.3
Working with those who are studying	***Total***	***88***	***41.7***
	Males	25	48.1
	Females	63	39.6
During the period of martial law	***Total***	***46***	***17.1***
	Males	6	8.8
	Females	40	19.9
Completed studying	***Total***	***30***	***11.2***
	Males	4	5.9
	Females	26	12.9
Started a family	***Total***	***28***	***10.4***
	Males	5	7.4
	Females	23	11.4
Found employment	***Total***	***69***	***25.7***
	Males	16	23.5
	Females	53	26.4
Lost a job	***Total***	***19***	***7.1***
	Males	6	8.8
	Females	13	6.5
Changed their field of activity	***Total***	***26***	***9.7***
	Males	10	14.7
	Females	16	8.0

Financial problems had almost the same share in the structure of complaints – 11.5% during the pandemic and 12.3% at the beginning of the war, with no statistically significant difference between males and females.

Among all COVID problems (593), each respondent named an average of two (2.2), while since the beginning of the war, 780 have been named, with an average of three (2.9).

The frequency of psychological problems differed significantly. During the war, they were reported more often than during the 2020-21 pandemic, with more than twice as many females (77.1% vs. 34.8%) and three times as many males (52.9% vs. 17.6%). The concern for the safety and mental health of relatives during the war was also twice as significant to respondents (51.7% vs. 26.4%) compared to during the pandemic. During the pandemic, psychological problems in various manifestations, including the fear of death, safety, and mental health of relatives, affected the vast

majority of respondents (59.5%, comprising 42.6% males and 65.2% females), but the impact of the war surpassed this, affecting 84.8% of respondents (66.2% males and 91.0% females).

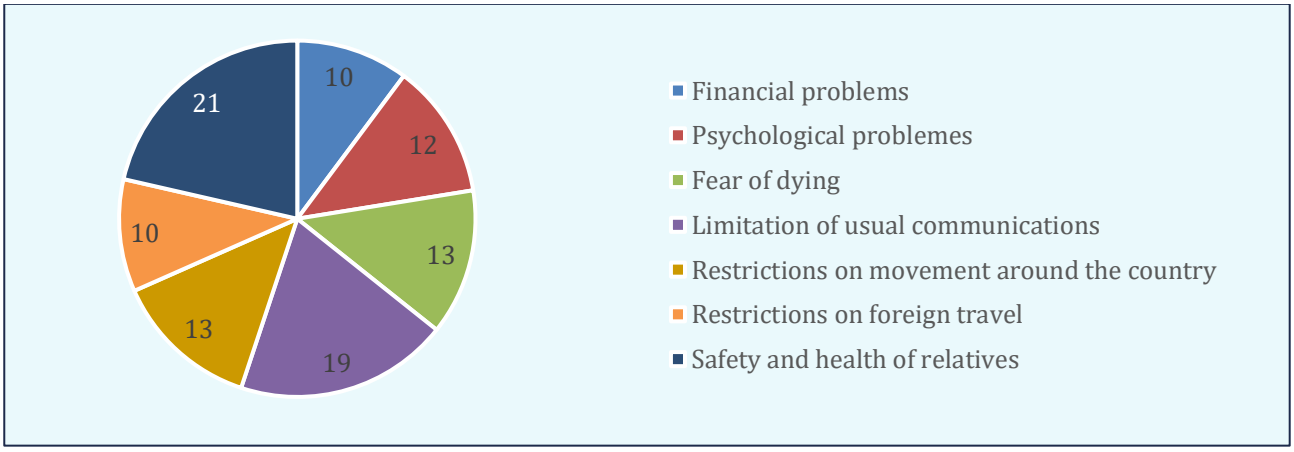

Figure 2. The structure of problems that occurred during the full-scale Russian invasion (from 02/24/2022) according to respondents (survey 2023/24).

According to a survey of students in higher education institutions in Ukraine at the beginning of the war in 2022, the vaccination rate was 69.6% (with 70% among males and 69.5% among females). Trust in vaccines and vaccination was observed in 51.6% of respondents, with an additional 18.0% being immunized on demand. Prior to the war, students were meticulous in following anti-epidemic measures against COVID-19: 89.1% (87.7% males, 89.6% females) used masks in public places, 73.4% (68.5% males, 75.1% females) frequently and thoroughly washed their hands or regularly disinfected them, 36.5% (35.4% males, 36.9% females) limited their movements and avoided unnecessary outings. Students often ventilated the room they were in for an extended period (44.7%, with 36.9% males and 47.3% females), maintained social distance (49.3%, with 44.6% males and 50.9% females), avoided crowds and public events (55.3%, with 55.4% males and 55.2% females), and did not use public transport, preferring to walk when possible (40.0%, with 40.0% males and 39.9% females). Only 4.2% of respondents (4.6% males, 4.1% females) admitted to doing nothing to prevent infection.

With the outbreak of war, the attitude towards the implementation of anti-epidemic measures has changed: 43.2% (44.6% males, 42.7% females) answered "no action." Another 8.2% (10.8% males, 7.4% females) added "no possibility." Partial measures could be taken by 35.0% of respondents, with fewer males (22.3%) than females (39.2%) ($t=3.84$, $p<0.001$). However, 13.2% of respondents did not change their infection prevention actions since the beginning of the war, with more males (22.3%) than females (10.2%) ($t=3.06$, $p<0.01$).

As a result, at the end of the second year of the war (1 year and 10 months), the situation with COVID-19, influenza, and other URIs was a concern for only one in four respondents (25.7%), with females (27.9%) expressing this concern more often than males (19.1%) ($t=2.13$, $p<0.05$). A direct correlation was found between male anxiety about the situation with COVID-19, influenza, and URIs and the problem of restricting travel abroad during the 2020/21 pandemic ($r=0.239$, $p<0.05$) and an inverse correlation with continuing education during this period ($r=-0.259$, $p<0.05$). Male anxiety was also influenced by the reassessment of the importance of preserving their own health and that of their loved ones during the war ($r=0.238$), with an error probability of 5.1%.

There was a correlation between female anxiety about the situation with COVID-19, influenza, and other URI and psychological problems that arose during the 2020/21 pandemic ($r=0.198$, $p<0.01$), except for financial problems, which have already been discussed. Female anxiety was also influenced by job loss ($r=0.152$, $p<0.05$), volunteering in the form of participation in the delivery of aid to the front ($r=0.288$, $p<0.01$), and cooking ($r=0.152$, $p<0.01$). A correlation was found bet-

ween the presence of psychological complaints in females at the time of the survey and anxiety about COVID-19, influenza, and other upper respiratory infections ($r=0.163$, $p<0.05$). When applying logistic regression, a statistically significant inverse characteristic was found to be mastery of first aid skills. Thus, with a high probability (P=0.99%), the concerns of female respondents about the current situation can be described as follows: $Z = -8.220 + 1.21a - 0.728b + 2.030c$, *where a - financial problems during the 2020/21 pandemic; b – mastering first aid; c - participation in the delivery of aid to the front line.*

DISCUSSIONS

The online survey of young people in Chernivtsi and nearby regions of the western part of Ukraine successfully complements the previous 2022 study of students studying at higher education institutions in various regions of Ukraine. The aim is to explore the most significant challenges faced by young people during the COVID-19 pandemic (2020/21) and Russia's full-scale invasion of Ukraine. This study aligns with a survey conducted by the independent think tank Cedos in Ukraine among the civilian population (10, 11, 12).

The research results revealed that in Ukraine (across most of its regions), unlike other LMICs, challenges arising from the war distort commonly accepted strategies for combating infectious diseases. The population, particularly young people, struggle to adhere fully to anti-epidemic measures across all territories. During the war, psychological concerns displaced fears of the pandemic. Psychological issues during the pandemic may lose their relevance in retrospect, potentially impacting the survey results in late 2023 and early 2024. Challenges related to communication and transportation decreased to 29.2% due to adaptation to the current situation. The smaller proportion of financial issues in the survey conducted among students and graduates of BSMU was attributed to the survey being conducted in the relatively less war-affected territory of Bukovyna.

At the time of the survey, 70.3% of respondents were vaccinated against COVID-19, which could have reduced the fear of getting sick, but was not statistically significant. This may be due to the fact that a significant number of students had already had these infections at the time of the survey. For example, the inverse correlation showed that male students were less likely to be concerned about the situation with COVID-19, influenza, and other URI. Young people seemed to have moved on from COVID-19 as an experience. In the comments to the questionnaire, we read: "Why ask about COVID-19? Are you looking to resume it?" Female anxiety regarding the situation with COVID-19, influenza, and Upper Respiratory Infections (URI) was influenced by job loss (effectively a financial problem) and volunteering, manifested through participating in delivering aid to the front lines and cooking (effectively demonstrating concern for the health and safety of soldiers). Studies among Czech university students [12] associated the female gender with higher levels of anxiety and depressive symptoms. Logistic regression identified an additional stress reduction factor: mastering first aid skills. Among males, this supportive component could be characterized as a reassessment during the war of the importance of preserving their own health and that of their loved ones. Consequently, more males (22.3%) than females (10.2%) did not alter their actions to prevent COVID-19 since the beginning of the war ($p<0.01$).

CONCLUSIONS

1. At the end of the second year of the war, the situation with COVID-19, influenza, and other URIs concerned only one in four respondents (25.7%), and no statistically significant relationship was found between experiences and vaccination (70%) or past illnesses (65%).
2. The psychological challenges faced by young people in Ukraine during the war (84.8%) overshadowed those caused by the pandemic (59.5%), leading to adverse changes in compliance with COVID-19 anti-epidemic measures in the initial months of the war. Notably, 43.2% of respondents reported engaging in no preventive activities.
3. A weak direct correlation was observed between the concerns of males about the situation with COVID-19, influenza, and URIs during the war (2023/24 survey) and the problem of restricting travel abroad during the 2020/21 pandemic ($r=0.239$, $p<0.05$). Additionally, there was an inverse

correlation with the pursuit of continuing education during the survey period.

4. There is a weak direct correlation between females' concerns about the situation with COVID-19, influenza, and other URIs and various factors, which include psychological issues (r=0.198, p<0.01), financial problems (r=0.224, p<0.01) arising during the 2020/21 pandemic, job loss (r=0.152, p<0.05), volunteering in the form of participation in the delivery of aid to the front (r=0.288, p<0.01), cooking (r=0.152, p<0.01), and the presence of psychological complaints at the time of the survey (r=0.163, p<0.05).

CONFLICT OF INTEREST

The authors declare no conflict of interest regarding the study and this paper.

ETHICAL APPROVAL

The study was approved by the Ethics Committee of Bukovinian State Medical University (approval number 4 from 21.12.2023).

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SYNERGISTIC ACTION OF CHEMICAL COMPOUNDS AND SPIRULINA EXTRACTS

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Keywords: *chemical compounds, biological compounds, synergistic action, antimicrobial action.*

Introduction. *Antimicrobial resistance is a critical global issue, and the demand for new drugs is urgent. The current model of antimicrobial discovery is not providing sufficient new agents to address current levels of antimicrobial resistance. In this study, we aim to assess the antimicrobial activity of recently synthesized chemical compounds and spirulina extracts, along with their associative effects.*

Material and methods. *It is an experimental study, which includes seven newly synthesized chemical compounds and three extracts from Spirulina platensis.*

Results. *The in vitro results of the present study determined the MIC values of chemical compounds and spirulina extracts against gram-negative, gram-positive microorganisms, and fungi of the genus Candida. The optimal combination of two compounds can enhance benefits over time and minimize side effects. The synergistic or partially synergistic effects of biological compounds in conjunction with chemical compounds strongly corroborated this assertion. Time-kill curves and FICI scores confirmed the ability of the compounds to synergistically reduce the microbial count below the lowest detectable limit within 24 hours.*

Conclusions. *Our study offers a potential therapeutic option for antibiotic-resistant microbial agents by combining natural extracts with a variety of chemical compounds from different classes. The results of the present study are promising, and this knowledge holds potential utility for the development of future therapeutic strategies.*

Keywords: *chemical compounds, biological compounds, synergistic action, antimicrobial action.*

Cuvinte-cheie: *compuși chimici, compuși biologici, acțiune sinergică, acțiune antimicrobiană.*

ACȚIUNEA SINERGICĂ A COMPUȘILOR CHIMICI ȘI A EXTRACTELOR DIN SPIRULINĂ

Introducere. *Rezistența la antimicrobiene reprezintă o problemă globală gravă, de aceea obținerea unor noi remedii cu acțiune antimicrobiană este o urgență. Modelul actual de descoperire a antimicrobienelor nu furnizează agenți noi, suficienți pentru combaterea nivelurilor actualre de rezistență la antimicrobiene. În acest studiu ne-am propunem să evaluăm activitatea antimicrobiană a unor compuși chimici noi sintetizați și a extractelor din spirulină, precum și efectele asociative ale acestora.*

Material și metode. *În prezentul studiu experimental au fost testați șapte compuși chimici noi sintetizați și trei extracte din Spirulina platensis.*

Rezultate. *In vitro au fost determinate valorile CMI ale celor șapte compuși chimici noi sintetizați și a trei extracte din spirulină împotriva microorganismelor gramnegative, grampozitive și a micetelor din genul Candida. Combinația adecvată a doi compuși poate mări beneficiile în timp și minimaliza reacțiile adverse. Efectele sinergice sau parțial sinergice ale compușilor biologici în combinație cu compușii chimici au susținut acest efect. Curbele de omorâre în timp și scorurile FICI au confirmat capacitatea compușilor testași de a reduce sinergic numărul de microorganisme sub cea mai mică limită detectabilă în 24 de ore.*

Concluzii. *Studiul oferă o opțiune terapeutică potențială pentru agenții microbieni rezistenți la antibiotice prin combinarea extractelor naturale și a compușilor chimici din diferite clase. Rezultatele obținute sunt promițătoare și pot fi utile pentru dezvoltarea viitoarelor strategii terapeutice.*

INTRODUCTION

Bacterial resistance to conventional antimicrobials has become a significant and life-threatening issue worldwide, imposing a substantial economic burden on healthcare systems.

Over the recent decades, the number of antimicrobial-resistant bacteria has increased dramatically due to the widespread use of antibiotics (1, 2). Despite the emergence of new antibiotics with different mechanisms of action, the process of drug discovery and development typically spans between 10 and 17 years, with a success rate below 10% (3). Furthermore, while new single-target antimicrobials are widely used, new antibiotic-resistant strains will inevitably arise.

This ever-growing challenge makes the discovery of new antibiotics inevitable, as well as the development of new alternative approaches. Increased attention among these alternative approaches has been given to antimicrobials combined with plant extracts. The latter approach, namely, the combination therapy or synergistic therapy used in combating resistant microorganisms can lead to new ways of treating infectious diseases, which is likely to outline a potential area for future investigations. Combination therapy is also efficient in patients with severe infections caused by antibiotic-resistant pathogens. Synergism consists of intensifying the pharmacodynamic effects of two or more associated compounds (4).

Combination therapy is the most common empirical treatment recommended in cases of bacterial infections within intensive care units, where monotherapy may not be effective against all the potential disease-causing pathogens. Moreover, combination therapy may reduce the drug dosage and thus minimize the side effects, which might result in overcoming the toxicity problem and the spread of resistant microbial strains (5).

MATERIAL AND METHODS

Chemical compounds

The study included the following chemical compounds, which were synthesized at the Department of Inorganic Chemistry, Department of Chemistry, at State University of Moldova: C1 ($C_{14}H_{19}CuN_7O_4S$), C2 ($C_{13}H_{16}Br_2CuN_4S$), C3 ($C_{10}H_{14}CuN_4O_5S_2$), C4 ($C_{13}H_{17}ClCuN_4S$), C5 ($C_{18}H_{20}CuN_4O_2S$), C6 ($C_{14}H_{20}N_4S$) and C7 ($C_{14}H_{19}ClCuN_4S$).

ES1, ES2 and ES3 Spirulina extracts

The biologically active complexes *ES1, ES2* and *ES3* extracts were obtained through a biotechnological process from the cyanobacterial strain of *Spirulina platensis* CNMN CB-02 (Spirulina), stored within the National Collection of Non-Pathogenic Microorganisms, at the Institute of Microbiology and Biotechnology.

Spirulina biomass was obtained from a cyanobacterial growth culture and through controlled synthesis of its biologically active compounds. Biologically active complexes, encompassing free amino acids, oligopeptides, proteins, sulphated polysaccharides, and phospholipids, were extracted from the Spirulina biomass. These extracts were successively fractionated and purified by using benign solvents and techniques. Relevant formulas were developed and standardized for complex compositions of extracts, based on biologically active complexes derived from Spirulina biomass. All extracts were natural and devoid of herbicides, toxins, or preservatives.

ES1 spirulina extract is an amino acid/oligopeptide complex, which contains non-essential (glycine, alanine, serine, cysteine, tyrosine, aspartic acid, glutamic acid, and proline) and essential amino acids (arginine, phenylalanine, histidine, isoleucine, leucine, lysine, threonine, tryptophan, and valine), being in their free state or combined in oligopeptides (up to 10kDa), as well as biologically functionalized macro- and microelements. *In vitro* tests were used for *ES1 form*, which is an alcoholic solution, having 10mg/ml of extract concentration and 50% of alcohol concentration. *ES2 spirulina extract* is a synergistic combination of amino acid/oligopeptide complex, sulfated polysaccharides, proteins, and biologically functionalized macro- and microelements derived from spirulina. For the *ES2 form*, *in vitro* tests were conducted using an alcoholic solution with a concentration of 20 mg/ml of extract and 45% alcohol concentration.

ES3 spirulina extract is a glycosidic carotenoid. *In vitro* tests were conducted using an 80% aqueous-ethanol solution.

Microbial strains

Microbial strains of *Staphylococcus aureus* ATCC 25923, *Bacillus subtilis* ATCC 6633, *Escherichia coli ATCC 25922, Pseudomonas aeruginosa ATCC*

27853, Acinetobacter baumannii ATCC BAA-747, and Candida *albicans ATCC 10231* were used in the study. The microbial strains were cultured on appropriate nutrient media at their optimum growth temperature. Overnight cultures of the bacterial strains were used for further investigations.

Antimicrobial activity

Serial culture dilutions were used to determine the antibacterial activity of the plant, allowing for the assessment of the minimum inhibitory concentration (MIC) and the minimum bactericidal/fungicidal concentration (MBC/MFC). MIC and MBC/MFC were determined using a discontinuous gradient of extract concentrations tested in Muller-Hinton broth, followed by the addition of 100µL of bacterial suspension according to the 0.5 McFarland turbidity standard. The tubes were then incubated at 35-37°C for 18-24 hours, and the MIC value was determined by macroscopic analysis of the tubes, based on the presence or absence of bacterial growth. MBC/MFC was determined by replicates of Muller-Hinton agar dilution. The MBC/MFC value represented the lowest concentration of extract that reduced the number of microbial colonies by up to 99.9% (6, 7).

The negative control sample consisted of Muller-Hinton broth containing the studied extracts, while the positive control sample involved Muller-Hinton broth inoculated with the studied microorganisms. All experiments were conducted in triplicate.

Methods of synergy testing

The combined antibacterial effects of two preparations were assessed using the checkerboard method, followed by the determination of the FIC index (Fractional Inhibitory Concentration). Stock solutions and double dilutions of each compound were prepared in accordance with EUCAST recommendations immediately before testing (6). The assays were conducted in multiwell plates, with 50µL of Mueller-Hinton broth added per well. The first combined compound was serially diluted along the y-axis (y), while the second compound was diluted along the abscissa axis (x). A suspension corresponding to the McFarland 0.5 turbidity standard was prepared from the tested strain, and subsequently, 100 µL of microbial suspension ($5x10^5$CFU / ml) was added to each well. The plates were incubated at 35°C for 24 hours aerobically.

Fractional inhibitory concentration was calculated using the MIC of the combined compounds, as well as the MIC of each compound obtained through parallel testing, according to the following formula:

where,

$$\Sigma FIC = FIC\ (A) + FIC\ (B)$$

$$FIC\ (A) = \frac{MIC\ (A)\ combination}{MIC\ (A)\ alone}$$

and

$$FIC\ (B) = \frac{MIC\ (B)\ combination}{MIC\ (B)\ alone}$$

The ΣFICI values were interpreted as follows: ∑FIC ≤ 0,5 = Synergistic; 0,5 < ∑FICI ≤ 1 = Additive; 1 < ∑FICI ≤ 4 = Indifferent; ∑FIC > 4,0 = Antagonistic.

Time-Kill Assay

Time-Kill Assay enables the measurement of changes in the population of aerobic microorganisms over a defined period in the presence of antimicrobial agents. The assay was conducted on *S.aureus, E.coli* and *C.albicans* strains, based on the aforementioned method with minor modifications (8). A saline suspension was prepared from an 18-24 hour microbial culture, by finally obtaining an inoculum of 1 x 10^6 CFU/mL. This suspension was evenly distributed into four tubes: I tube (culture control sample) - Mueller Hinton broth; II tube - Mueller Hinton broth + 0.5×CMI extract; III tube - Mueller Hinton broth + 1xCMI extract; IV tube - Mueller Hinton broth + 2×CMI extract. The tubes were incubated at 35°C for 24 hours. 100µL from each tube was replicated on the plate medium at specific intervals. Afterwards, the plates were incubated at 37°C for 24 hours and the CFU/plate was measured, followed by CFU/mL (the mean number of colonies multiplied by dilution). The tests were performed in triplicate (three independent experiments).

Time-Kill curves were graphically represented by $\log_{10}$CFU mL^{-1} versus the 24-hour time period. Bactericidal activity (99.9% of killing) was defined as a ≥3-$\log_{10}$CFU mL^{-1} reduction in the number of colonies from the initial inoculum.

Statistical analysis

Data are expressed as mean standard deviation. Statistical analysis involved a one-way analysis of variance (ANOVA). P<0.001 was considered to indicate a statistically significant difference.

RESULTS

The antimicrobial effect of chemical compounds and biologically active complex extracts derived from the cyanobacterium *Spirulina platensis*, namely *ES1, ES2,* and *ES3,* were observed during the initial stage of the present research.

Based on this, we conducted in vitro assays to assess the bactericidal and antifungal effects of seven new chemical compounds on gram-positive bacteria, gram-negative bacteria, and fungi of the genus Candida. The tests demonstrated that all chemical compounds exhibited superior bactericidal and bacteriostatic actions against gram-positive bacteria. In comparison to control drug preparations (Furacillin and Miconazole), most compounds were effective at lower doses (tab. 1).

Table 1. The antibacterial activity of chemical compounds against some microbial strains (µg/mL).

Chemical compounds		*S. aureus*	*B. subtilis*	*A. baumannii*	*E. coli*	*P. aeruginosa*	*C. albicans*
C1	**MIC**	0.976	0.976	0.976	7.812	250	15.62
	MBC	1.953	0.976	1.953	15.62	500	31.25
C2	**MIC**	0.976	0.976	15.62	15.62	125	3.906
	MBC	1.953	1.953	31.25	31.25	250	7.812
C3	**MIC**	0.122	0.122	1.953	3.906	15.62	31.25
	MBC	0.244	0.244	3.906	7.812	31.25	62.5
C4	**MIC**	0.488	0.122	7.812	15.62	>500	1.953
	MBC	1.953	0.122	15.62	31.25	>500	31.25
C5	**MIC**	0.488	0.244	3.906	7.81	125	7.81
	MBC	0.976	0.488	7.812	15.62	250	15.62
C6	**MIC**	0.061	1.953	0.25	>500	>500	0.976
	MBC	0.244	3.906	0.50	>500	>500	1.953
C7	**MIC**	0.976	0.488	62.50	31.25	>500	7.812
	MBC	1.953	0.976	125	62.50	>500	15.62
Furacillinum	**CMI**	4.67	4.67	4.67	4.67	4.67	-
	MBC	4.67	4.67	9.35	4.67	9.35	-
Miconazole	**MIC**	-	-	-	-	-	16.0
	MFC	-	-	-	-	-	32.0

Note: *S. aureus* (*Staphylococcus aureus* ATCC 25923); *B. subtilis* (*Bacillus subtilis* ATCC 6633); *A. baumannii* (*Acinetobacter baumannii* BAA-747); *E. coli* (*Escherichia coli* ATCC 25922); *P. aeruginosa* (*Pseudomonas aeruginosa* ATCC 27853); *C. albicans* (*Candida albicans* ATCC 10231). MIC – minimum inhibitory concentration; MBC – minimum bactericidal concentration; MFC – minimum fungicidal concentration.

C6 (*S. aureus* MIC 0.061 µg/mL), C4 and C5 (*S. aureus* MIC 0.488 µg/mL) were identified as the most active compounds against gram-positive strains. Compounds C6 (*A. baumannii* MIC 0.25 µg/mL) and C1 (*A. baumannii* MIC 0.976) exhibited a higher activity on gram-negative strains. Only four compounds were active against *P. aeruginosa* strains, the most active being C3 (MIC 15.62).

C. albicans strain demonstrated sensitivity to all tested compounds, whereas C6 (MIC 0.976) and C4 (MIC 1.953) compounds showed a higher low-dose activity.

The experimental testing of the biological compounds showed that all extracts of *S. platensis* exhibited promising antimicrobial activity against both gram-positive and gram-negative bacterial strains used in the present research (tab. 2).

The *ES3* extract exhibited a high antibacterial and antifungal activity compared to *ES1* and *ES2* extracts. The highest activity of *ES3* extract was recorded against *B. subtilis* strains (MIC 0.004 mg/mL) and *C. albicans* strains (0.004 mg/mL), as well as against gram-negative bacillus strains, being active at MIC 0.009 mg/mL. The *ES2* extract displayed bacteriostatic activity against all tested species at higher concentrations, compared to *ES1* and *ES3* extracts.

Table 2. The action of biologically active complex extracts of *Spirulina platensis* against some bacterial strains.

Species	*ES1*		*ES2*		*ES3*		Ampicillin
	MIC mg/mL	MBC mg/mL	MIC mg/mL	MBC mg/mL	MIC mg/mL	MBC mg/mL	MIC µg/mL
Gram-positive bacteria							
***S. aureus* ATCC 25923**	0.625	1.25	2.5	5.0	0.018	0.037	0.08
***B. subtilis* ATCC 6633**	0.156	0.156	2.5	5.0	0.004	0.004	0.078
Gram-negative bacteria							
P. aeruginosa* ATCC *27853	0.625	1.25	1.25	2.5	0.009	0.018	0.012
***E. coli* ATCC 25922**	0.625	1.25	2.5	5.0	0.009	0.018	0.025
***A. baumanni* BAA-747**	0,625	1.25	1.25	2.5	0.009	0.018	0.25
Yeast	MIC mg/mL	MFC mg/mL	MIC mg/mL	MFC mg/mL	MIC mg/mL	MFC mg/mL	Miconasole MIC µg/ml
***C. albicans* ATCC 10231**	0.625	1.25	1.25	2.5	0.004	0.009	16.0

Subsequently, the synergistic action of the chemical compounds combined with the biological compounds was assayed on six reference strains. The determination of the Fractional Inhibitory Concentration Index revealed synergistic actions in 87.2% of cases, additive actions in 6.8%, and indifferent actions in 6.0%. The tested compounds did not show any antagonistic relationship (FICI> 4) (ta. 3).

The testing of the chemical compound C1 combined with three biological compounds revealed the synergism phenomenon in most cases, except for *P. aeruginosa* bacteria, where the additive and indifferent phenomenon were registered in combinations with *ES1* and *ES2*, respectively.

The C2 compound showed synergism against the tested bacteria in all combinations, with the exception of its combination with the ES2 compound. In this particular case, an indifferent action (FICI-1.56) was observed against the *P. aeruginosa* strain.

The C3 compound, in combination with biological compounds, exhibited synergistic action in 72.2% of cases, additive action in 22.2% of cases, and indifferent action in 5.6% of cases against the tested strains, whereas the C4 compound showed predominantly synergistic action (94.4%) and additive action in only 5.6% of cases.

The C5 compound, when combined with *ES2*, showed indifferent action against *E. coli* and *P. aeruginosa* strains. Additionally, when combined with *ES1*, it showed indifference against *P. aeruginosa* strains only.

C6 exhibited indifferent action against *A. baumannii* strain when combined with *ES2* and showed additive action in combination with *ES1*. On the other hand, the C7 compound, when combined with *ES2*, revealed an additive effect against *E. coli* strains.

The chemical compounds combined with the biological compound *ES3* showed a synergistic action against all tested bacterial species, except for C3 that showed additive effects (FICI 0.56) against *P. aeruginosa* strains.

The minimum inhibition concentrations for the combined chemical and biological compounds resulted in a 4 to 32-fold decrease in the MIC of the parent compound (tab. 4). The additive and indifferent actions were particularly recorded against gram-negative bacilli and fungi of the *Candida* genus.

To determine the synergistic activity of the chemical compounds in combination with the biological compounds, along with the treatment duration and efficiency on the viability of microbial cells, the time-kill behavior was assessed.

Table 3. Interaction between chemical compounds and biological compounds expressed by FICI.

Tested compounds		*S. aureus* ATCC 25923	*B. subtilis* ATCC 6633	*A. baumannii* BAA-747	*E. coli* ATCC 25922	*P. aeruginosa* ATCC27853	*C. albicans* ATCC 10231
C1	***ES1***	0.35 (S)	0.12 (S)	0.5 (S)	0.32 (S)	0.61 (Ad)	0.32 (S)
	ES2	0.32 (S)	0.35 (S)	0.5 (S)	0.37 (S)	1.37 (I)	0.35 (S)
	ES3	0.35 (S)	0.12 (S)	0.40 (S)	0.32 (S)	0.5 (S)	0.32 (S)
C2	***ES1***	0.32 (S)	0.35 (S)	0.30 (S)	0.35 (S)	0.5 (S)	0.44 (S)
	ES2	0.35 (S)	0.35 (S)	0.30 (S)	0.35 (S)	1.56 (I)	0.5 (S)
	ES3	0.12 (S)	0.12 (S)	0.32 (S)	0.35 (S)	0.44 (S)	0.37 (S)
C3	***ES1***	0.37 (S)	0.35 (S)	0.32 (S)	0.32 (S)	0.75 (Ad)	0.5 (S)
	ES2	0.44 (S)	0.32 (S)	0.44 (S)	0.56 (Ad)	1.23 (I)	0.61 (Ad)
	ES3	0.32 (S)	0.12 (S)	0.25 (S)	0.32 (S)	0.56 (Ad)	0.44 (S)
C4	***ES1***	0.32 (S)	0.35 (S)	0.5 (S)	0.44 (S)	0.5 (S)	0.37 (S)
	ES2	0.37 (S)	0.32 (S)	0.5 (S)	0.5 (S)	0.84 (Ad)	0.44 (S)
	ES3	0.32 (S)	0.32 (S)	0.44 (S)	0.44 (S)	0.44 (S)	0.32 (S)
C5	***ES1***	0.32 (S)	0.25 (S)	0.32 (S)	0.5 (S)	1.23 (I)	0.44 (S)
	ES2	0.32 (S)	0.32 (S)	0.37 (S)	1.24 (I)	1.42 (I)	0.5 (S)
	ES3	0.30 (S)	0.35 (S)	0.25 (S)	0.44 (S)	0.5 (S)	0.37 (S)
C6	***ES1***	0.35 (S)	0.37 (S)	0.56 (Ad)	NT	NT	0.32 (S)
	ES2	0.32 (S)	0.44 (S)	1.07 (I)	NT	NT	0.37 (S)
	ES3	0.12 (S)	0.32 (S)	0.5 (S)	NT	NT	0.35 (S)
C7	***ES1***	0.35 (S)	0.25 (S)	0.44 (S)	0.5 (S)	NT	0.39 (S)
	ES2	0.32 (S)	0.32 (S)	0.5 (S)	0.84 (Ad)	NT	0.44 (S)
	ES3	0.32 (S)	0.25 (S)	0.37 (S)	0.44 (S)	NT	0.35 (S)

Note: Synergistic (S) actions- FICI≤0.5; Additive (Ad) actions - 0.5<FICI≤1; Indifferent actions (I) - 1<FICI≤4; Antagonistic actions (An) FICI>4; NT – not tested.

Distinct time-kill profiles were recorded for each bacterial strain within 24 hours after inoculation. The results indicated no increase in the microbial strains tested in the first 30 minutes after inoculation. However, variations in antimicrobial activity among the tested compounds or bacterial strains were observed over the subsequent 90 minutes of incubation. No decrease in the number of CFUs was noted in the control tubes, and the use of chemical or biological compounds alone at a concentration of 0.5MIC did not induce bacterial death unless observed over 24 hours.

The combined chemical and biological compounds showed a significant reduction in the number of microorganisms. The combination of chemical compounds with *ES3* exhibited the best results on the tested species, as the microorganisms were killed over 8, 12, 16 and 20 hours. The combinations of 0.25MIC C1 + 0.25×MIC *ES3* and 0.25×MIC C2 + 0.25×MIC *ES3* completely inhibited the growth of *S. aureus* strains over 8 hours.

The combinations of chemical compounds with the *ES2* biological compound showed bactericidal action for 16 and 20 hours. Notably, only the combination of *ES3* with C2 displayed a bactericidal action against *S. aureus* strains over 8 hours.

DISCUSSIONS

Arthrospira platensis (also called *Spirulina platensis*), one of the most well-known cyanobacteria produced on an industrial scale, has attracted the attention of researchers as a natural compound with therapeutic properties and potential antimicrobial effect (antibacterial, antifungal, and antiviral). During the investigation of *Arthrospira platensis* as a source of proteins, vitamins (such as vitamin B12 and provitamin A), and essential fatty acids such as γ-linolenic acid, biologically active compounds with antimicrobial activity against some species of microorganisms were obtained. Therefore, commercial production of spirulina has gained importance worldwide due to

its multiple benefits. Spirulina can suppress the growth of several microorganisms due to its rich content of bioactive ingredients with antimicrobial activity. The effectiveness of spirulina extracts on microorganisms with multiple resistance to antimicrobials was experimentally demonstrated. Their antimicrobial activity is attributed to the presence of carbohydrates, phenolic compounds, flavonoids, and tannins in their composition. One objective of this study was to investigate the antimicrobial activity of spirulina, and the results indicated that spirulina extracts have a greater potential to inhibit the growth of gram-positive bacteria compared to gram-negative bacteria. This effect can be attributed to the complicated structure of the cell wall (outer membrane) of gram-negative bacteria (9, 10, 11).

Table 4. The synergistic antimicrobial effects of chemical and biological compounds against the reference strains.

Tested compounds		*S. aureus* ATCC 25923	*B. subtilis* ATCC 6633	*A. baumannii* BAA-747	*E. coli* ATCC 25922	*P. aeruginosa* ATCC27853	*C. albicans* ATCC 10231
C1	*ES1*	$1/32_C + 1/16_B$	$1/32_C + 1/32_B$	$1/4_C + 1/4_B$	$1/16_C + 1/8_B$	-	$1/16_C + 1/8_B$
	ES2	$1/16_C + 1/8_B$	$1/32_C + 1/16_B$	$1/4_C + 1/4_B$	$1/8_C + 1/8_B$	-	$1/16_C + 1/32_B$
	ES3	$1/32_C + 1/16_B$	$1/32_C + /32_B$	$1/16_C + 1/4_B$	$1/8_C + 1/16_B$	$1/4_C + 1/4_B$	$1/16_C + 1/8_B$
C2	*ES1*	$1/8_C + 1/16_B$	$1/32_C + 1/16_B$	$1/8_C + 1/32_B$	$1/32_C + 1/16_B$	$1/4_C + 1/4_B$	$1/8_C + 1/4_B$
	ES2	$1/16_C + 1/32_B$	$1/32_C + 1/16_B$	$1/8_C + 1/32_B$	$1/16_C + 1/32_B$	-	$1/4_C + 1/8_B$
	ES3	$1/32_C + 1/32_B$	$1/32_C + 1/32_B$	$1/16_C + 1/8_B$	$1/32_C + 1/16_B$	$1/4_C + 1/4_B$	$1/8_C + 1/8_B$
C3	*ES1*	$1/8_C + 1/8_B$	$1/32_C + 1/16_B$	$1/16_C + 1/8_B$	$1/16_C + 1/8_B$	-	$1/4_C + 1/4_B$
	ES2	$1/8_C + 1/4_B$	$1/16_C + 1/8_B$	$1/4_C + 1/8_B$	-	-	-
	ES3	$1/16_C + 1/8_B$	$1/32_C + 1/32_B$	$1/16_C + 1/16_B$	$1/16_C + 1/8_B$	-	$1/4_C + 1/8_B$
C4	*ES1*	$1/16_C + 1/8_B$	$1/32_C + 1/16_B$	$1/4_C + 1/4_B$	$1/4_C + 1/8_B$	$1/4_C + 1/4_B$	$1/8_C + 1/8_B$
	ES2	$1/8_C + 1/8_B$	$1/16_C + 1/8_B$	$1/4_C + 1/4_B$	$1/4_C + 1/4_B$	-	$1/8_C + 1/4_B$
	ES3	$1/8_C + 1/16_B$	$1/8_C + 1/16_B$	$1/8_C + 1/4_B$	$1/8_C + 1/4_B$	$1/8_C + 1/4_B$	$1/16_C + 1/8_B$
C5	*ES1*	$1/16_C + 1/8_B$	$1/16_C + 1/16_B$	$1/16_C + 1/8_B$	$1/4_C + 1/4_B$	-	$1/8_C + 1/4_B$
	ES2	$1/8_C + 1/16_B$	$1/8_C + 1/16_B$	$1/8_C + 1/8_B$	-	-	$1/4_C + 1/4_B$
	ES3	$1/32_C + 1/8_B$	$1/32_C + 1/16_B$	$1/16_C + 1/8_B$	$1/8_C + 1/4_B$	$1/4_C + 1/4_B$	$1/8_C + 1/8_B$
C6	*ES1*	$1/32_C + 1/16_B$	$1/8_C + 1/8_B$	-	NT	NT	$1/16_C + 1/8_B$
	ES2	$1/16_C + 1/8_B$	$1/8_C + 1/4_B$	-	NT	NT	$1/8_C + 1/8_B$
	ES3	$1/32_C + 1/32_B$	$1/16_C + 1/8_B$	$1/4_C + 1/4_B$	NT	NT	$1/32_C + 1/16_B$
C7	*ES1*	$1/32_C + 1/16_B$	$1/16_C + 1/16_B$	$1/8_C + 1/4_B$	$1/4_C + 1/4_B$	NT	$1/4_C + 1/16_B$
	ES2	$1/8_C + 1/16_B$	$1/8_C + 1/16_B$	$1/4_C + 1/4_B$	-	NT	$1/4_C + 1/8_B$
	ES3	$1/8_C + 1/16_B$	$1/16_C + 1/16_B$	$1/8_C + 1/8_B$	$1/8_C + 1/4_B$	NT	$1/16_C + 1/32_B$

In recent years, the number of works dedicated to the research of coordination complexes has increased significantly, which indicates the increased interest of researchers in these chemical compounds. The main advantages of these compounds are their homogeneity, stability, the possibility of exact dosing, the ease of assessing metabolic processes, as well as the strict observance of technological operations. Most of the studies emphasize the potential for controlling the properties and biological effects of these compounds (12, 13).

Coordination chemistry remains one of the most important and current areas of inorganic chemistry. Of particular interest is the synthesis and study of the physico-chemical properties of coordination complexes formed by transition metals with organic ligands. Ligands with a high coordination tendency form compounds with diverse compositions, structures, and properties. Due to their valuable biological activity, these compounds are used in medical practice. Among them are heterocyclic thiosemicarbazones with antibacterial, antifungal, antimalarial, and antiviral properties (14, 15).

In recent decades, the development of new antimicrobial drugs has been based on combining traditional antimicrobials with various coordinating

compounds. The remarkable results of these studies have been confirmed by the intensification of activity and broadening of the spectrum of action when using compounds with both synergistic and cumulative effects, as well as the prevention of antimicrobial resistance. This approach also allows for the reduction of treatment doses, costs, and toxic side effects (16).

The research of recent years has not only highlighted a new alternative in combating multiresistant microorganisms but has also suggested the possibility of restoring the antimicrobial effect of some antimicrobials previously categorized as ineffective. Promising results have also been observed when natural compounds with antimicrobial action were combined with synthetic antimicrobials. The combined use of natural and synthetic antimicrobial compounds in the treatment of infectious diseases potentiates the antimicrobial spectrum, reduces the toxicity of some antimicrobials, and prevents the development of antimicrobial resistance (17, 18).

Combination therapy is more effective in polymicrobial infections compared to monotherapy. Antimicrobial remedies of natural origin, when used in combination with synthetic medicines, have shown a series of effective interactions, including the synergistic amplification of antimicrobial potential and reduction of the adverse effects of synthetic medicines. These synergistic effects of combined medicines reduce therapeutic failures, increase efficacy, and shorten hospital stays (19, 20).

The synergy of natural medicines combined with antimicrobials, particularly against microorganisms prone to developing resistance, has been investigated by several researchers. Some have observed that Berberis aetnensis leaf extracts significantly reduce the minimum inhibitory concentration of ciprofloxacin, thereby restoring its efficacy in the therapy of *S. aureus, E. coli* and *P. aeruginosa* infections. In another study, a significant increase in antimicrobial activity against multiresistant *P. aeruginosa* strains was demonstrated when antibiotics were combined with clove, jambolan, pomegranate, and thyme extracts. The use of clove-ampicillin and clove-tetracycline combinations resulted in increased antimicrobial activity against *K. pneumoniae* and *Proteus* spp. strains (20).

CONCLUSIONS

1. All chemical and biological compounds included in the study have exhibited antibacterial and antifungal activity at various concentrations.
2. 87.2% of the combinations of chemical compounds with biological ones exhibited synergistic actions, while only 6.8% showed additive actions, and 6.0% were indifferent. Additive and indifferent effects were particularly observed on gram-negative bacilli and yeast-like fungi. No antagonistic actions were recorded when combining chemical compounds with biological ones. In combinations, the MIC for chemical and biological compounds decreased from four to 32 times compared to the MIC of individual compounds.
3. When using chemical and biological compounds at a concentration of 0.25 times the MIC, in most cases, there was no recorded reduction in the number of microbial cells. However, when these compounds were combined, the microorganisms were killed within 8-24 hours. The shortest time for microbial destruction (8-20 hours) was observed when combining chemical compounds with the biological compound ES3.
4. This study presents a potential therapeutic option for antibiotic-resistant microorganisms by combining natural extracts with a range of different classes of chemical compounds. The most effective approach to developing antimicrobials with minimal toxic or adverse side effects is through the use of natural products.

CONFLICT OF INTEREST

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

ETHICAL APPROVAL

The study was conducted and approved by the Ethics Committee no. 3/14.04.2023 of *Nicolae Testemitanu* State University of Medicine and Pharmacy of the Republic of Moldova.

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METHODS FOR STIMULATING COGNITIVE ABILITIES USED BY STUDENTS

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Keywords: *cognitive ability, learning activity, stimulation, students.*

Introduction. *The modern workplace requires workers to have broad cognitive and affective abilities. For continuous progress and societal improvement, students' education plays a crucial role, and for this, students need a high level of cognitive abilities to apply in their learning activities, reflecting in their academic performance. There are several methods to stimulate cognitive abilities, which can be healthy, unhealthy, or a combination of both.*
The *aim* of the research *is to identify and describe the methods used by students to stimulate cognitive abilities in their learning activities.*

Material and methods. *Google Drive questionnaire. Sample - 212 of respondents.*

Results. *It was found that students prefer physical activities to stimulate their cognitive abilities. The most common method is walking (45.7%), followed by workout (17.10%), and meditation (15.2%). They also engage in other activities such as swimming, outdoor games, practicing yoga, drawing, cooking, etc. At the same time, students have a high rate of consuming caffeinated beverages, with 30.50% drinking them systematically, 37.10% occasionally, and 19% rarely. One-third of the respondents (30.5%) occasionally consume energy drinks, and 11.4% rarely do so.*

Conclusions. *Alongside healthy methods, students use many other quick and efficient ways to stimulate their cognitive abilities. The study results raise concerns about the students' risk behavior, as some prefer unhealthy methods to stimulate their cognitive abilities in their learning activities.*

Cuvinte-cheie: *capacitate cognitivă, activitate de învă-țare, stimulare, elevi.*

METODE DE STIMULARE A ABILITĂȚILOR COGNITIVE UTILIZATE DE ELEVI

Introducere. *Pentru obținerea de performanțe academice, elevii au nevoie de un nivel ridicat de abilități cognitive pe care să le utilizeze în activitățile lor de învățare. Există mai multe metode de stimulare a abilităților cognitive ale elevilor care pot fi sănătoase sau nesănătoase.*

Scopul cercetării. Identificarea și descrierea metodelor de stimulare a abilităților cognitive utilizate de elevi în activitatea de învățare.

Material și metode. *Chestionarul Google Drive. Eșantionul - 212 respondenți.*

Rezultate. *În urma analizei răspunsurilor din chestionare s-a constatat că elevii preferă activitățile fizice pentru a-și stimula abilitățile cognitive, cele mai aplicate fiind mersul pe jos (45,7%), antrenamentul (17,10%) și meditația (15,2%). Urmează, în ordinea descreșterii preferințelor, înotul, jocurile în aer liber, practicarea yoga, desenul, gătitul etc. Printre preferințele elevilor se numără și băuturile cofeinizate, care sunt consumate sistematic (30,50%), uneori (37,10%) sau ocazional (19%). O treime dintre respondenți (30,5%) consumă băuturi energizante ocazional, iar 11,4% – rar.*

Concluzii. *Pe lângă metode sănătoase, elevii folosesc multe alte modalități mai puțin sănătoase, dar rapide și eficiente de a-și stimula abilitățile congnitive. Rezultatele studiului ridică un motiv de îngrijorare cu privire la comportamentul de risc al elevilor care preferă metode nesănătoase de stimulare a abilităților cognitive în activitatea de învățare.*

INTRODUCTION

High cognitive abilities are becoming increasingly relevant in our highly competitive world. Often referred to as "21st century skills," these abilities include being able to solve complex problems, think critically, communicate effectively across diverse cultures and using various techniques, work in collaboration with others, adapt to rapidly changing environments and conditions for performing tasks, effectively manage one's work, and autonomously acquire new skills and information (1). Both psychological and pedagogical theory and practice prove that extracting, structuring, and organizing knowledge, along with developing skills to find solutions to problems by students, are possible only as a result of their intensive cognitive activity. Common examples of cognitive activities are reasoning, thinking, perceiving, listening, observing, etc. High-level cognitive activities usually include problem-solving, decision-making, and sense-making that involve using and working with information (2). Cognitive ability is closely associated with educational attainment, occupation, and health outcomes (3).

The level of education directly influences cognitive function by enhancing brain function, and indirectly by encouraging individuals with higher education to engage in intellectually stimulating activities, thus preserving cognitive ability. Many efforts to enhance cognition are of a rather commonplace nature, with some practices dating back thousands of years. Education and training, for example, aim not only to impart specific skills or information but also to improve general mental faculties such as concentration, memory, and critical thinking. Various forms of mental training, including yoga, martial arts, meditation, and creativity courses, are also commonly employed. Caffeine is widely used to improve alertness, and herbal extracts believed to improve memory, like Ginko Biloba, enjoy significant popularity, with sales amounting to several hundred million dollars annually in the U.S.A. In an ordinary supermarket, numerous energy drinks are on display, appealing to consumers seeking to turbo-charge their brains (4).

According to some authors (Chrisantus O. Brumboiu I, Porrovecchio A, Peze T, Hurdiel R, Cazacu I, Mogosan C, Ladner J, Tavolacci P.M.), students use various methods to stimulate cognitive ability in their learning activities, such as the kinesiology method, meditation method, maintaining proper sleep, keeping a healthy and balanced diet, use of prescription drugs (beta-blockers, modafinil, and methylphenidate), use of "soft enhancers" (caffeinated products, food supplements, and energy drinks), and use of "drugs of abuse" (alcohol, cannabis, cocaine, and amphetamines) (5, 6).

As the discussion about stimulating cognitive abilities is widely covered in the literature, it has sparked empirical curiosity to know which specific methods students from our university use. Therefore, *the aim of the study* is to identify and describe the most common methods for stimulating cognitive abilities used by students in their learning activities.

MATERIAL AND METHODS

The study represents a descriptive, cross-sectional research based on the collection, interpretation, and evaluation of data. Research methods include bibliographic, statistical, and sociological approaches. A study was conducted among university students aged 18 and above from the Republic of Moldova. University students were invited by email to complete an online anonymous 20-minute Google Drive questionnaire consisting of 15 questions. Respondents' confidentiality was fully respected, as the Google Drive questionnaire did not collect emails. The criteria for respondent selection included student status, age above 18, and their agreement to respond to the questions. A total of 246 students responded to the questionnaire, but only 212 responses met the age criteria for inclusion in the research. Data about different methods for stimulating cognitive abilities (healthy or less healthy) were collected. All questions adhered to non-discriminatory and non-harm principles. The data were analyzed through an Excel database, and conclusions were formulated.

RESULTS

Students from various academic years participated in the survey, with the majority (75.5%) belonging to the second and third years of study. Out of the 212 respondents, 91.4% fell within the age range of 19 to 24, while 6.7% were older than 25, and the remaining were under 19. When queried about the time allocated to their learning activities, 46.7% of students reported spending

3-4 hours on learning, 32.40% dedicated less than 3 hours, and 21% spent more than 4 hours on learning.

Out of 212 respondents, when asked about methods to stimulate or reinforce their cognitive abilities in learning activities, 45.70% of students prefer walking, 15.2% prefer meditation, 17.1% prefer workouts, 2.90% of the students prefer swimming as well as physical exercises, 7.60% of respondents prefer outdoor games and activities, and 9.10% prefer to sleep or engage in other activities such as drawing, cooking, or playing musical instruments (fig. 1).

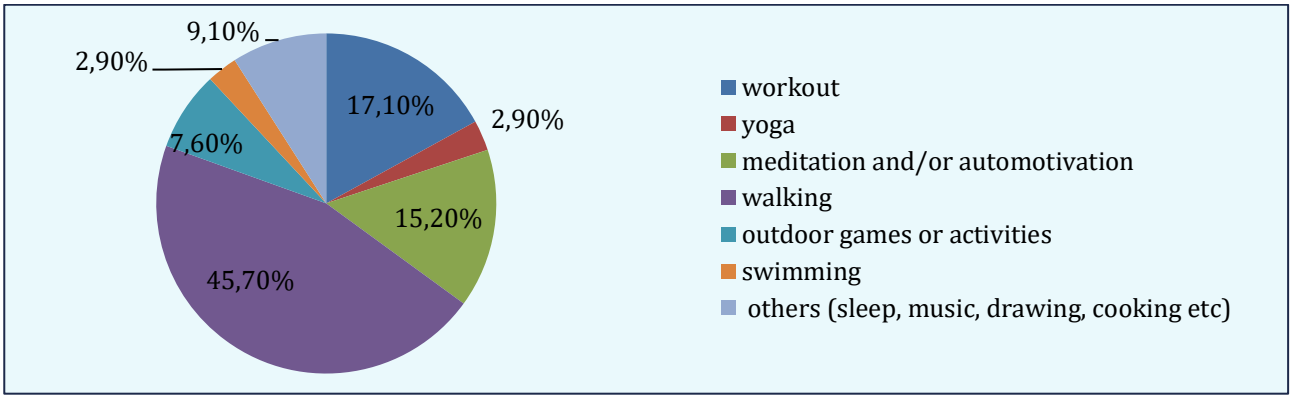

Figure 1. Students' preferred physical activities for stimulating cognitive abilities.

An important factor for stimulating cognitive abilities is sleep. Despite unanimous agreement among all respondents about the significant impact of sleep, 53.3% of students sleep less than 7 hours, 45.70% sleep for 7-9 hours, and only 1% of students reported sleeping for more than 9 hours.

When asked about their consumption of caffeinated drinks while studying, students provided varied responses. Among respondents, 37.10% and 30.50% of students drink caffeinated drinks sometimes and systematically, respectively. Additionally, 19% of students prefer only occasional consumption, and 13.30% do not consume caffeinated drinks (fig. 2).

When asked about the healthiness of their meals, more than half of the respondents (62.90%) perceive their eating habits as average in terms of healthiness. Meanwhile, 23.80% of them acknowledge following a poor healthiness of eating habit, and only 13.30% of students believe they have a good level of healthiness in their eating habits.

Among the 212 respondents, 48% of students never used energy drinks, while 38% of students drink them occasionally, and 12% use them seldom. Only 2% of the respondents use energy drinks regularly (fig. 3).

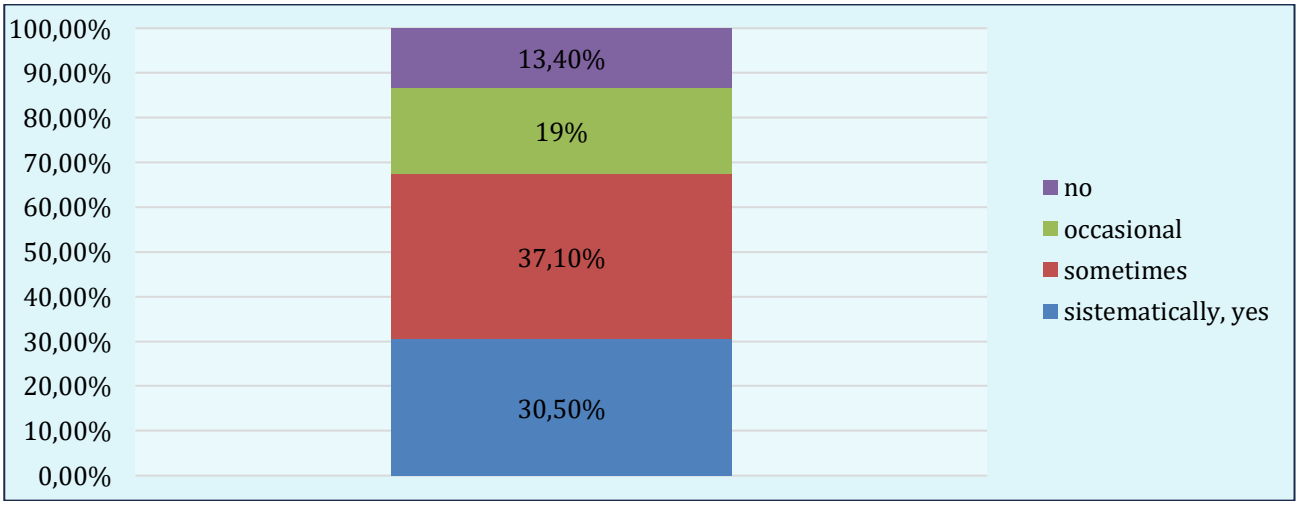

Figure 2. Consumption of caffeinated drinks while studying.

When asked about the use of drugs like Ritalin, Dexedrine, and Adderall without a doctor's prescription, all respondents (100% of the students) reported never using them. Regarding the use of psychoactive drugs such as marijuana, crack, cocaine, MDMA, codeine, morphine, dermol, and others without a doctor's prescription, the majority of the 212 respondents, 95%, reported never using them. Only a very few respondents (4%) indicated using them occasionally or trying them once or twice, and no one reported using them regularly (fig. 4).

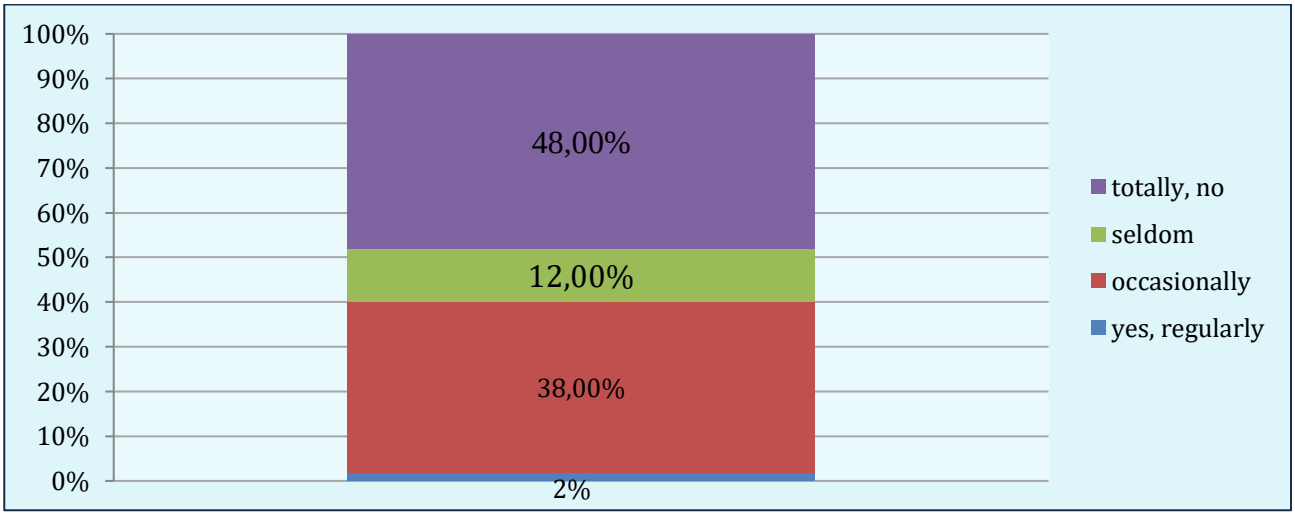

Figure 3. Energy drink consumption.

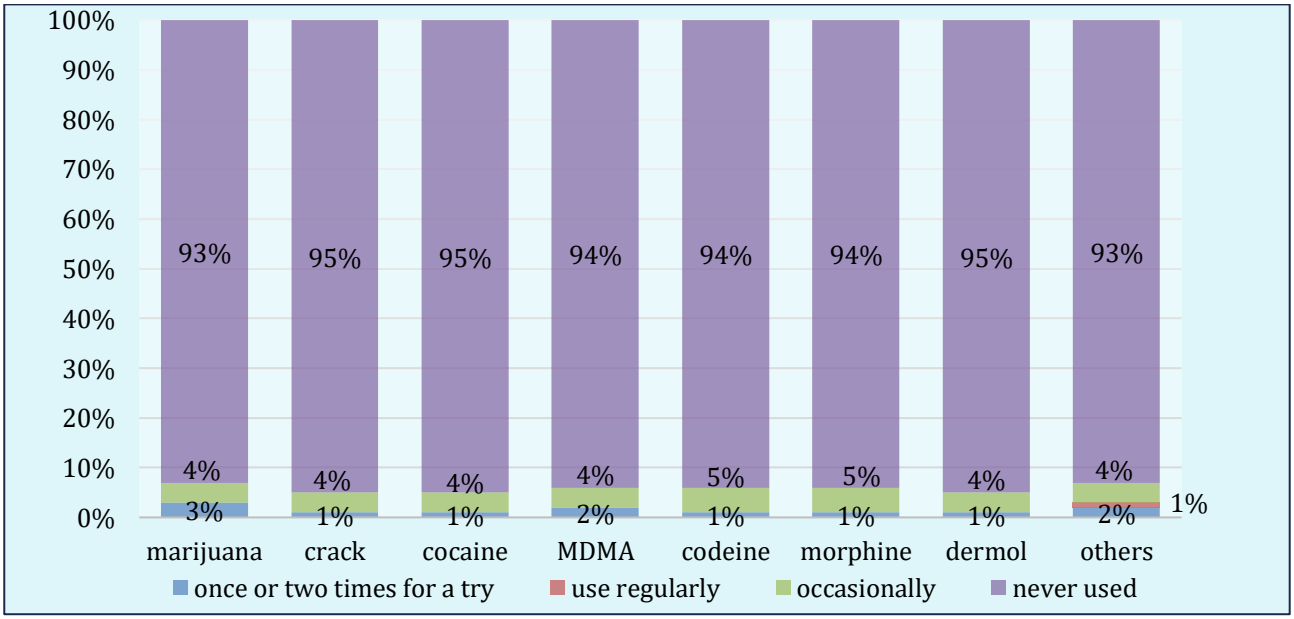

Figure 4. Use of psychoactive drugs (without doctor's prescription).

DISCUSSIONS

In this study, one of the main aspects investigated was the time spent on learning activities, and the findings reveal that a majority (46.7%) of students dedicate around 3-4 hours to studying. Additionally, 21% of students invest more than 4 hours in learning, indicating a high motivation to enhance their cognitive abilities and achieve academic success. The study also highlights that students prefer engaging in physical activities to stimulate their cognitive abilities. The most favored methods include walking (45.7%), followed by workout (17.10%), and meditation (15.2%). Furthermore, students express a preference for various other activities such as swim-

ming, playing outdoor games, practicing yoga, drawing, cooking, etc. These activities are not only essential for cognitive stimulation but also contribute to maintaining the mental and physical well-being of students.

According to the study data, students exhibit a high rate of caffeinated drink consumption, with 30.50% drinking them systematically, 37.10% sometimes, and 19% occasionally. Caffeinated beverages, such as coffee, tea, and cola, are generally socially acceptable and serve as coping strategies for students to enhance cognitive function and manage stressful academic situations. However, the misuse of caffeinated drinks, especially among systematic users, can lead to various adverse effects. Similar to caffeinated beverages, 30.5% of students use energy drinks occasionally, and 11.4% use them seldom. It is crucial to be mindful of the dosing of energy drinks, as increased consumption poses potential risks.

The study indicates that the majority of students (62.90%) maintain an average level of healthiness in their eating habits, while 23.80% follow poor dietary practices. Regarding the use of drugs like Ritalin, Dexedrine, and Adderall without a doctor's prescription, all respondents (100%) reported never using them. Additionally, the consumption of drugs of abuse for neuroenhancement, such as marijuana, cocaine, MDMA, crack, morphine, etc., is very low (4-5%), with the majority (95%) of students reporting never using them. While the data suggests a healthy and positive approach, it's important to note that in reality, many students may not disclose the use of unhealthy methods for neuroenhancement.

CONCLUSIONS

1. The information derived from specialized literature sources highlights the correlation between cognitive abilities and learning activities. The enhancement of students' education is pivotal for societal improvement, and achieving a high level of cognitive abilities is essential for students to excel in their learning activities, thereby positively influencing their academic performance.
2. Research reveals that the majority of students favor healthy methods to stimulate their cognitive abilities, such as physical exercise, walking, and meditation. These practices contribute to increased attention during learning activities. However, some students opt for a combination of both healthy and unhealthy methods. They may engage in healthy activities while also resorting to less advisable practices like smoking or using psychoactive drugs without a doctor's prescription. It appears that students utilizing systematic methods for cognitive stimulation may prioritize approaches with immediate effects. Notably, only a very small percentage of students choose to rely solely on unhealthy methods to enhance their cognitive abilities during learning activities.
3. The study results highlight a concern regarding the risk behavior of students who opt for unhealthy methods to stimulate cognitive abilities during learning activities. It emphasizes the need for educational programs and other measures aimed at increasing awareness among students about the potential consequences associated with unhealthy methods of cognitive enhancement during learning activities.

CONFLICT OF INTEREST

Authors have no conflict of interest to declare.

ETHICAL APPROVAL

Not applicable to this research.

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USE OF MICROMOLECULAR COPPER COMPLEXES OF THIOSEMICARBAZIDES AS AN ENDOGENOUS CATALASE INDUCER/ACTIVATOR

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Keywords: *copper coordinating compound, thiosemicarbazide derivatives, production or activation of CAT, multifactorial diseases.*

Introduction*. Oxidative stress is a major mechanism in the pathogenesis of many diseases, including severe multifactorial ones. Catalase (CAT), along with superoxide dismutase, is one of the first-line antioxidant defense enzymes. The development of methods/compounds that can increase/induce CAT will provide new possibilities to strengthen antioxidant protection and prevent oxidative damage to cells and tissues.*

Material and methods. *The ability of three copper coordination compounds from the class of transition metal thiosemicarbazides to enhance/induce CAT in erythrocytes of healthy white laboratory rats has been studied.*

Results. *Dichloro-(methyl-N-(prop-2n-1-yl)-2-(pyridin-2-ylmethylidene)hydrazine-carbimido-thioate)copper was found to show the highest induction and/or activation of CAT, exceeding 2.71 times the values of the control group and 1.80 times the values produced by vitamin D3. This reveals increased synthesis of CAT after exposure to this compound, a phenomenon that we have established for the first time.*

Conclusions. *The compound can be used as a therapeutic agent, which, by activating the production of CAT in the body, can prevent and/or reduce the occurrence of multifactorial diseases, due to prevention of the damage of cells associated with excessive accumulation of hydrogen peroxide. The obtained data open perspectives for the research of biologically active synthetic compounds, which will diversify the arsenal of effective tools for preventing/treating various diseases.*

Cuvinte-cheie: *compus coordinativ al cuprului, derivați de tiosemicarbazidă, producerea sau activarea CAT, boli multifactoriale.*

UTILIZAREA COMPLEXELOR MICROMOLECULARE DE CUPRU ALE TIOSEMICARBAZIDELOR CA INDUCTORI/ACTIVATORI ENDOGENI AI CATALAZEI

Introducere. *Stresul oxidativ este un mecanism major în patogeneza multor boli, inclusiv a celor multifactoriale severe. Catalaza (CAT), împreună cu superoxid dismutaza, este una dintre enzimele de apărare antioxidantă de primă linie. Dezvoltarea de metode/compuși care pot crește/induce CAT va oferi noi posibilități de a spori protecția antioxidantă și de a preveni deteriorarea oxidativă a celulelor și a țesuturilor.*

Material și metode*. A fost studiată capacitatea a trei compuși de coordonare a cuprului din clasa tiosemicarbazidelor metalelor de tranziție de a crește/induce CAT în eritrocitele șobolanilor albi de laborator sănătoși.*

Rezultate. *S-a constatat că dicloro-(metil-N-(prop-2n-1-il)-2-(piridin-2-ilmetiliden)hidrazin-carbimido-tioat)cuprul exercită cea mai potentă inducție și/sau activare a CAT, depășind de 2,71 ori valorile lotului martor și de 1,80 ori valorile produse de vitamina D3. Aceste date relevă sinteza endogenă crescută a CAT după expunerea la acest compus, fenomen care a fost stabilit pentru prima dată.*

Concluzii. *Compusul poate fi utilizat în medicină ca agent terapeutic care, prin activarea/inducerea CAT în organism, poate preveni și/sau reduce apariția bolilor multifactoriale cauzate de deteriorarea celulelor și a țesuturilor, asociate cu acumularea excesivă de peroxid de hidrogen. Datele obținute deschid noi perspective pentru cercetarea compușilor sintetici biologic activi care vor diversifica arsenalul de instrumente eficiente pentru prevenirea sau tratarea diferitor boli.*

INTRODUCTION

CAT plays a central role in the detoxification of oxygen peroxide (H_2O), which is crucial in defending cells against oxidative damage caused by H_2O_2. Hydrogen peroxide is toxic due to its ability to form other reactive oxygen species (ROS), such as the hydroxyl radical in the Fenton reaction. Additionally, H_2O_2 can act as an important signalling molecule, being involved in multiple physiological and pathological processes (1). CAT can also act as a peroxidase, thus contributing to the metabolism of micromolecular substrates such as methanol, ethanol, azide, and hydroperoxides. In the case of ethanol, the enzyme is able to oxidize it to acetaldehyde, contributing to its metabolism in the liver. Thus, CAT may have additional roles, such as detoxification or activation of toxic and antitumor compounds.

However, the molecular mechanisms regulating the expression of CAT, the oldest known antioxidant enzyme, have not yet been fully elucidated. Identifying these mechanisms will allow us to find new approaches to modulate antioxidant status in a variety of pathological conditions (2, 3).

Deficiency or impaired functioning of CAT is linked to the pathogenesis of a number of age-related diseases, such as diabetes mellitus, hypertension, anemia, vitiligo, Alzheimer's disease, Parkinson's disease, bipolar disorder, cancer, and schizophrenia (4).

Several known compounds have been identified to increase CAT activity in tissues during various pathological processes. Notable examples include the elevation of CAT activity in erythrocytes of patients with type II diabetes through metformin treatment (5). Additionally, CAT production and/or activity in the mouse macrophage-like cell line RAW264 is increased by treatment with sodium nitroprusside (SNP) and 1-hydroxy-2-oxo-3,3-bis-(2-aminoethyl)-1-triazene (NOC18). At the maximum concentration of 300 μmol/L, NOC18 statistically and conclusively increases CAT activity by up to 50%, thereby protecting macrophages from apoptosis induced by H_2O_2 (6). Furthermore, adding coffee to the feed of animals with liver cancer increases CAT activity almost to the values specific to healthy animals (7). Melatonin has been shown to increase CAT activity in peripheral blood mononuclear cells (PBMC) by 80% compared to untreated cells (8), while Fullerenol ($C60(OH)36$) increases it in red blood cells by 24% ($p<0.05$) compared to untreated ones (9). However, these methods have the disadvantage of being ineffective, as they do not provide stable, sufficiently high induction and/or activation of CAT.

The method closest in technical essence and result to increasing the production or activity of CAT in the body is based on the administration of vitamin D3 or its derivatives (10). However, this method has several disadvantages, namely that it does not provide sufficient stimulation of the production and/or activity of CAT, and the simultaneous use of vitamin D3 with its analogues increases the risk of developing hypervitaminosis D and its toxic effects.

The problem that our method solves is the generation of new copper coordination compounds from the class of thiosemicarbazides, thereby expanding the range of synthetic compounds with high CAT induction/activation potential.

MATERIAL AND METHODS

Three new local copper coordinating compounds (CC), derivatives of thiosemicarbazide, were included in the study:

1) Acetato-2-(({(metilsulfanil)((prop-2-en-1-il)amino)metiliden}hidraziniliden) metil) fenolatoaquacupru (coded (Cu(OAc)2);
2) Bromo-2-{(2-(prop-2-en-1-ylcarbamothioyl)-hydrazinylidene) methyl}phenolatocopper, coded (Cu(HL)Br);
3) Dichloro-(methyl-N-(prop-2n-1-yl)-2-(pyridin-2-ylmethylidene)hydrazine-carbimidothioate)copper, coded (Cu(L-H)Cl2).

These compounds were synthesized at Moldova State University in the “Advanced Materials in Biopharmaceutics and Technology” Laboratory, by the team of professor Aurelian Gulea, PhD, academician (11, 12, 13).

The effect of the local CC on erythrocyte CAT was evaluated in experiments on *Rattus norvegicus domestica albino*. The experiments were conducted in accordance with contemporary principles in the biological standardization of experiments and the Helsinki Declaration with subsequent amendments (Somerset West Amendment, 1996). The study was approved by the Research Ethics Board of Nicolae Testemițanu State University of Medicine and Pharmacy (approval no. 81 of 19.09.2020).

The experiments involved 50 rats weighing 180-230 g, divided into 5 groups of 10 animals each. The first group, the control, consisted of animals kept on a regular vivarium diet and injected with physiological saline 3 times a week for 30 days. CC were administered in a dose of 1.0 µM/kg, 3 times a week for 30 days in the following way: group 2 – (Cu(OAc)2), group 3 – (Cu(HL)Br), group 4 – (Cu(L-H)Cl2). The rats in group 5 (prototype) were administered vitamin D3 in a dose of 20.0 µM/kg in the same way as CC. After 24 hours from the last administration of the tested compounds, the blood was collected for the evaluation of CAT. The erythrocyte mass, obtained after decanting the blood serum, was washed 3 times with physiological saline.

CAT activity was determined according to the method described by Korolyuk M.A. et al. (1988) (14) with modifications (15). Briefly, 0.01 mL of the erythrocyte mass diluted 1000 times with distilled water was pipetted into the 96-well photometric microplates, and 0.18 ml of 0.03% H_2O_2 was added. In the control samples, instead of H_2O_2, the same amount of distilled water was added. Three parallel reference samples, which contain only H_2O_2 and distilled water, were prepared. The samples were incubated for 10 minutes at 37°C, and afterwards, 0.10 mL of 4% ammonium molybdate solution was added. The solution was shaken, and its absorbance was measured at 410 nm. The difference between the absorbance of the reference sample and the experimental sample was calculated. Enzyme activity was expressed in µmol per s per 1 g Hb (µmol/s·g Hb).

The spectrophotometric method was used to assess the levels of vitamin D in the research samples (16).

The statistical evaluation of the obtained data was conducted using StatsDirect Statistical Analysis Software (StatsDirect Ltd, UK). Mean±standard deviation (X±S) was calculated. To test the significant difference between the studied markers of the compared groups, the non-parametric "U" Mann-Whitney statistical test and the significance threshold "p" ($p<0.05$) were employed.

RESULTS

The results of the assessment of the changes in the CAT activity of erythrocytes due to the administration of certain local CC are presented in the statistical data in Table 1.

Based on the experimental data presented in Table 1, it is evident that the mentioned compounds induce a greater activation of CAT than that produced by vitamin D3, used as a prototype. The studied thiosemicarbazone derivatives increased erythrocyte CAT activity by 1.9-2.7 times compared to the control values and by 1.8 times compared to the values produced by the prototype (vit. D3). Compound Cu(L-H)Cl2 was found to exhibit the highest CAT induction/activation activity, exceeding the control values by 2.72 times and the prototype values by 1.8 times.

Table 1. The impact of local CC, derivatives of thiosemicarbazide, on erythrocyte CAT activity.

Study groups	CAT, µM/s.g Hb (X±S)
Control group	18.80±4.02 (100%)
Acetato-2-(({{(metilsulfanil)((prop-2-en-1-il)amino)metiliden}hidraziniliden) metil) fenolatoaquacupru (0,1 µM/kg) (coded (Cu(OAc)2)	35.7±5.61 *** (190%)
Bromo-2-{(2-(prop-2-en-1-ylcarbamothioyl)-hydrazinylidene)methyl}phenolato copper (0,1 µM/kg), coded (Cu(HL)Br)	50.8±5.66 *** (270%) ## (178%)
Dichloro-(methyl-N-(prop-2n-1-yl)-2-(pyridin-2-ylmethylidene)hydrazine-carbimido-thioate)copper (0,1 µM/kg), coded (Cu(L-H)Cl2)	51.2±7.98 *** (272%) ## (179%)
Vitamina D3 (prototip) (20 µM/kg)	28.6±4.26 **(152%)

Note: statistically significant difference with the control group *- $p<0.05$; ** - $p<0.01$; *** - $p<0.001$; statistically significant difference with the prototype (vitamin D3) - # $p<0.05$; ## - $p<0.01$.

DISCUSSIONS

The established properties of the studied CC are of interest for medicine in terms of expanding the range of synthetic inducers/activators of CAT. Enzyme induction is a process in which a molecule (e.g., a drug) induces an increase in the metabolic activity of an enzyme either by binding to the enzyme and activating it, or by increasing the expression of the gene encoding the enzyme (17, 18). Enzyme induction occurs when the molecule (called an inducer) facilitates gene expression. The specifics of how this occurs depend on control mechanisms as well as on differences between prokaryotic and eukaryotic cells (18).

CAT is one of the most important antioxidant enzymes, present in almost all aerobic organisms. In 1937, CAT was first crystallized from bovine liver in the laboratory of Sumner and Dounce. The gene encoding CAT in humans is located on chromosome 11 (19).

The significance and importance of the CAT induction phenomenon by CC, derivatives of thiosemicarbazide, stem from the broad range of practical applications of these inducers.

These inducers could potentially be used as agents for the treatment and prevention of renal fibrosis caused by CAT deficiency (20), or for the treatment of some forms of infertility, since CAT is found in mouse oocytes and likely plays a role in genomic protection from oxidative damage during meiotic maturation (21). It has been established that CAT expression is also altered in cancer cells, which favors cell proliferation by inducing genetic instability and activating oncogenes. Regulation of CAT expression is likely controlled mainly at transcriptional levels, although other mechanisms may be involved. In addition to transcription factors such as Sp1 and NF-Y, the transcription factors JunB and RARα are crucial regulators in breast cancer cells by recruiting proteins involved in transcriptional complexes and chromatin remodeling. Therefore, CAT may be an attractive therapeutic target in the context of cancer (22).

CAT was found to significantly decrease chromosomal aberrations and to delay or prevent spontaneous neoplastic transformation in mice fibroblasts and epidermal keratinocytes (23). Similarly, liver CAT activity decreases in the presence of a growing tumor, and after the tumor is removed, this activity returns to normal, demonstrating the importance of this antioxidant enzyme in tumorigenesis (24).

The methods for inducing CAT expression or activity are important for treating retinal oxidative stress associated with diabetic retinopathy (25).

Due to the ability of thiosemicarbazone derivatives to easily penetrate the blood-brain barrier (26) and their stability in the bloodstream, these compounds could be used to develop new effective methods for the early diagnosis of severe brain diseases, such as brain tumors and their metastases, as well as for the visualization of Aβ plaques in Alzheimer's disease.

CAT could be extremely useful for the development of effective therapies for brain and neurological dysfunctions. This is based on the fact that brain CAT activity is extremely low compared to other tissues and organs, such as the liver and kidney. The results of some studies reveal the importance of transient receptor potential (TRP) channels as a key component of the neurological Ca2+ ion entry pathway in response to the harmful action of reactive oxygen species (ROS). Exploratory data suggest that CAT may act effectively by suppressing the TRP channel activated by oxidative stress, exhibiting protective effects on neuronal mitochondrial function and neuronal survival (27). In the future, CAT could be extremely useful for the development of effective therapies for neurodegenerative diseases, such as Alzheimer's and Parkinson's disease, as well as sensory pain, because the decrease of CAT activity due to oxidative stress plays an important role in the pathogenesis of the aforementioned diseases (28).

One of the most severe multifactorial diseases is sepsis, a systemic inflammatory syndrome caused by infections that can lead to organ dysfunction with a high mortality rate (over 25%). Considering that the most susceptible to sepsis are children, people aged over 65 years, as well as patients with immunodeficiency, autoimmune diseases, tumors, kidney, and lung diseases (29), developing treatments using CAT inducers and/or activators would be a desirable and effective option. Septic shock is also one of the frequent complications of COVID-19 (30); therefore, CAT inducers could be an attractive therapeutic target

for the prevention of complications and treatment of SARS-CoV-2 infection. CAT helps regulate cytokine production, protect against oxidative damage, and suppress SARS-CoV-2 replication, as shown in human leukocytes, alveolar epithelial cells, and rhesus macaques, without noticeable toxicity (31, 32).

CONCLUSIONS

1. An effective method of inducing/activating CAT in the body has been developed using biologically active copper coordination compounds from the class of transition metal thiosemicarbazides. These compounds can prevent and/or reduce the occurrence of a variety of multifactorial diseases and the development of cell and tissue damage related to the excessive accumulation of hydrogen peroxide.
2. Therefore, the data obtained mark the beginning, opening up prospects for the development and research of new CCs, which will diversify the arsenal of means to combat various severe pathological processes. Further studies are needed to confirm the therapeutic utility of these bioactive compounds.

AUTHORS' CONTRIBUTION

The authors declare that their authorship complies with the international ICMJE criteria. All authors made a substantial contribution to the conception of the work, acquisition, analysis, interpretation of data for the work, drafting and revising the work, final approval of the version to be published, and agree to be accountable for all aspects of the work.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this paper.

ETHICAL APPROVAL

The research was approved by the Research Ethics Boar of *Nicolae Testemițanu* State University of Medicine and Pharmacy (approval no. 73 of April 26, 2017).

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UTILIZAREA POTENȚIALĂ A BACTERIOFAGILOR LA BIOAUGMENTAREA PROCESELOR BIOLOGICE DE EPURARE A APELOR UZATE

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Cuvinte-cheie: *bacteriofagi, tratare biologică, ape uzate, stație de epurare, rezistența la antimicrobiene.*

Introducere. În ultimii ani, calitatea apei a scăzut considerabil ca urmare a creșterpoii pulației și a dezvoltării industriale. Stațiile de epurare a apelor uzate primesc tone de apă uzată provenită din diverse surse, cum ar fi spitale, abatoare, ferme, unități farmaceutice și canalizare menajeră. Aceste stații adăpostesc o diversitate mare de bacterii și resturi de preparate antimicrobiene, ceea ce facilitează interacțiunea dintre diverse microorganisme și antimicrobiene, simplificând dezvoltarea mecanismelor de rezistență la antimicrobiene, precum și transmiterea genelor de rezistență de la o celulă la alta. Prin urmare, utilizarea metodelor eficiente de tratarea a apelor uzate are o importanță majoră în eliminarea microorganismelor rezistente la antimiicrobiene.

Scop. Evaluarea utilizării bacteriofagilor la bioaugmentarea proceselor de tratare biologică a apelor uzate.

Material și metode. Am realizat un studiu secundar-sinteză narativă a literaturii. Informația a fost obținută din bazele de date electronice internaționale folosind următoarele cuvinte-cheie „ape uzate”, „stația de epurare”, „rezistența la antimicrobiene”, „utilizarea bacteriofagilor”, „tratare biologică” în diferite combinații. Din numărul total de articole găsite (49), au fost selectate și analizate 25.

Rezultate. Multiple studii au raportat că în pofida utilizării diverselor procese de tratare biologică și/sau chimică, stațiile de epurare a apelor uzate nu sunt pe deplin eficiente în îndepărtarea agenților patogeni. Higgins și colab. (2018) menționează că agentul patogen oportunist *Acinetobacter baumanni* a fost depistat la toate etapele de epurare, fiind ulterior eliberat în mediu. Oliveira și colab. (2021) au constatat ineficiența proceselor convenționale de tratare a apelor uzate în eliminarea agenților patogeni rezistenți la carbapeneme. Bioreactoarele cu membrană sunt strategii avansate de tratare a apelor uzate, apreciate pentru capacitatea lor înaltă de eliminare a microorganismelor, însă formarea biofilmelor pe membrane împiedică funcționarea adecvată a acestora. O alternativă promițătoare este tratarea apelor uzate cu bacteriofagi ce oferă un remediu ecologic și rentabil în eliminarea microorganismelor din apele uzate. Bacteriofagii afectează gazda prin două moduri, fie prin modificarea procesului metabolic, adaptării și potențialului de supraviețuire al gazdelor, fie prin distrugerea directă a celulelor-gazdă. Printre proprietățile bacteriofagilor care îi fac atrăgători ca agenți terapeutici sau în calitate de biocontrol se numără liza doar a bacteriilor specifice acestora prin receptori specifici, creșterea ca număr în funcție de densitatea agenților patogeni și adaptarea ușoară la condițiile de mediu. Bacteriofagii au rol și în îndepărtarea biofilmelor bacteriene.

Concluzii. Creșterea semnificativă a rezistenței la antimicrobiene a adus în prim-plan terapia cu bacteriofagi, care are o istorie de peste 100 de ani. Aplicarea bacteriofagilor la stațiile de epurare influențează semnificativ capacitatea procesului de tratare, deci și calitatea apei uzate, precum și eliminarea agenților patogeni din biofilme și din nămol.

A REVIEW ON PREVALENCE AND ANTIMICROBIAL RESISTANCE OF SALMONELLA SPP. AND CAMPYLOBACTER SPP: ONE HEALTH PERSPECTIVE

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Keywords: *carcasses, incidence, Salmonella spp., Campylobacter spp. AMR, poultry.*

Introduction. Conducting an updated review of the antimicrobial resistance phenomenon and associated factors, as well the mechanisms and current perspectives on the "One Health" approach, including the Republic of Moldova's position in response to the World Health Organization's call, are priority directions for both human and veterinary medical services. Systematic review and gathering scientific information on the spread, incidence, resistance, and serotyping of bacteria from the genera *Salmonella spp*. and *Campylobacter spp*. remain crucial priorities worldwide.

The aim. The study focuses on analysing and assessing data regarding the incidence of infections with microorganisms from the *Campylobacter spp*. and *Salmonella spp*. genera in humans and in products of animal origins.

Material and methods. The study was conducted by analysing specialized literature from various databases, selecting relevant articles published between 2018 and 2023. The keywords used included prevalence, distribution of serotypes, antimicrobial resistance phenotypes, and genotypes of *Salmonella spp*. and *Campylobacter spp*. strains in humans and animal products. The *One Health* approach was employed. In total, 93 bibliographic sources were examined; utilizing databases such as *Embase*, *PubMed, Hinari*, *Google Scholar*, and data published by ECDC and WHO.

Results. The results regarding the proposed subjects were analysed. This was carried out within an integrative approach to the current situation in the field of human health, animal health, and the environment, as well as the position of the Republic of Moldova in combating the phenomenon of AMR. The obtained data confirmed that the highest prevalence of *Campylobacter spp*. among all types of samples investigated was found in poultry carcasses, with an average prevalence of 57% (44 out of 77 carcasses tested positive for *Campylobacter*). In comparison, based on surveillance data, on average, 38% of samples from fresh poultry meat in 22 European countries tested positive for *Campylobacter* (as of 2018).
The prevalence of pathogens in the carcasses of free-range broilers may result from the dissemination of bacteria from the genera Salmonella spp. and Campylobacter spp. in small-scale poultry farms, potentially affecting food safety.

Conclusions. The results regarding the incidence of *Salmonella spp*. and *Campylobacter spp*. argue for the importance and necessity of applying new techniques to assess the impact of zoonotic bacteria on public health. Systematic monitoring of the "environment-animal-human" components is an activity that will determine the level of quality of life. Bacteria from the genera *Salmonella spp*. and *Campylobacter spp*. are most frequently involved in foodborne infections in humans, underscoring the need for a revision of monitoring programs and mitigation strategies to reduce their impact on public health.

REZISTENŢA LA ANTIMICROBIENE ŞI FACTORII DE PATOGENITATE AI TULPINILOR CLINICE DE *STAPHYLOCOCCUS* SPP.

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Cuvinte-cheie: *Staphylococcus spp., rezistenţa la antimicrobiene, factori de patogenitate.*

Introducere. Tulpinile de *Staphylococcus* spp. izolate din secţiile de terapie intensivă şi din hemocultură sunt din ce în ce mai rezistente la un număr mare de agenţi antimicrobieni. În mod inevitabil, acest fenomen a dus la ineficienţa mai multor antimicrobiene, ceea ce pune viaţa pacienţilor în pericol. *S. aureus* este agentul patogen care trezeşte cele mai mari îngrijorări din cauza virulenţei intrinseci, a capacităţii de a provoca o gamă variată de infecţii şi de a se adapta la diferite condiţii de mediu. Mortalitatea prin bacteriemie cu *S. aureus* este în jur de 20–40%, în ciuda disponibilităţii antimicrobienelor eficiente. Persistenţa microorganismelor este determinată de multipli factori care inactivează mecanismele antimicrobiene ale sistemului imun. De aceea, este binevenită studierea factorilor de patogenitate ai microorganismelor, de a căror manifestare depinde rata de eliminare a acestora din focar și prognosticul bolii.

Scop. Evaluarea rezistenţei la antimicrobiene şi a factorilor de patogenitate ai tulpinilor clinice de *Staphylococcus* spp.

Material şi metode. Studiul a fost efectuat pe 102 tulpini de *Staphylococcus* spp. izolate din infecţii invazive în culturi pure şi ulterior identificate prin metode microbiologice clasice şi sistemul Vitek2 Compact (BioMerieux). Testarea sensibilităţii la antimicrobiene şi a capacităţii de formare a biofilmului a fost efectuată cu ajutorul metodei discdifuzimetrică Kirby-Bauer şi, respectiv, a testului cantitativ de aderenţă. Activitatea antilizozim a fost determinată după metoda descrisă de Gordina E. şi coaut, iar activitatea antcomplementară – după metoda descrisă de Bukharin O. şi coaut.

Rezultate. 74 (72,5%) de tulpini de *Staphylococcus* spp. izolate din infecţiile invazive au fost polirezistente la antibiotice, 41 (40,2%) – meticilin rezistente (MRS), 19 (15,8%) – pozitive prin testul D, iar 76 (75,5%) au produs biofilme. Tulpinile de stafilococi au prezentat sensibilitate mai înaltă la vancomicină (100%) şi la linezolid (90,6%). Rată mai mare de rezistenţă inductibilă la clindamicină a fost observată printre tulpinile MRS, în comparaţie cu cele sensibile la meticilină. Similar, rate mai mari de polirezistenţă şi rezistenţă la meticilină au fost înregistrate printre tulpinile producătoare de biofilm, în comparaţie cu tulpinile care nu produc biofilm. Activitatea antilizozim şi anticomplementară a fost pusă în evidenţă la 83,3% dintre tulpini şi la 69,6%, respectiv.

Concluzii. Pentru un management eficient al infecţiilor provocate de stafilococi este important de a include în diagnosticul de laborator de rutină testul D pentru detectarea rezistenţei inductibile la clindamicină. Vancomicina şi linezolidul pot fi indicate în tratamentul empiric al infecţiilor severe cauzate de *Staphylococcus* spp. Cunoaşterea factorilor de persistenţă a *Staphylococcus* spp va permite utilizarea lor în calitate de ţintă de atac la selectarea preparatelor antimicrobiene.

Studiul este realizat în cadrul proiectului JPIAMR „Phage treatment and wetland technology as intervention strategy to prevent dissemination of antibiotic resistance in surface waters”; (*PhageLand*), cifrul proiectului - 22.80013.8007.1M.

EVALUAREA PRACTICILOR DE UTILIZARE A ANTIBACTERIENELOR DE CĂTRE MEDICII VETERINARI DIN REPUBLICA MOLDOVA

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Cuvinte-cheie: *medic veterinar, RAM, antimicrobiene, sensibilitate, prescripție.*

Introducere. Problema utilizării excesive și incorecte a preparatelor antimicrobiene la creșterea animalelor rămâne a fi una acută. Prezentul studiu prevede analiza corectitudinii utilizării antimicrobienelor, în contextul rezistenți la antimicrobiene (RAM), în tratamentul și profilaxia bolilor bacteriene la animale. Stabilirea cauzelor RAM și reducerea impactului acesteia rămâne o prioritate în contextul abordării unei singure sănătăți.

Scop. Evaluarea practicilor de utilizare a antimicrobienelor de către medicii veterinari în Republica Moldova.

Material și metode. Studiul a fost bazat pe datele unui chestionar standardizat autoadministrat îndeplinit de către medicii/felcerii veterinari care activează în circumscripțiile teritoriale sanitar-veterinare ale Agenției Naționale pentru Siguranța Alimentelor din 35 de raioane ale țării, în perioada 20-27 septembrie 2023, pe un eșantion de 150 de respondenți.

Rezultate. La studiu au participat 123 de respondenți (82%), dintre care 32% t medici veterinari, iar restul 68%) – felceri veterinari. Vechimea medie în muncă a respondenților a fost de 27 de ani. 3% (4) dintre medicii veterinari consideră că problema RAM este importantă doar pentru sănătatea publică, 7% o consideră problemă de ordin național, iar 88% – de nivel global.. Dintre medicii veterinari chestionați, 92% prescriu antibiotice diferitor specii de animale de rentă, iar 36% folosesc antibiotice pentru a trata bolile neinfecțioase atât la animale adulte, cât și la tineret. În 80% din cazuri, prescrierea antibioticelor de către medicii veterinari are la bază examinarea clinică a animalelor de rentă bolnave. Nici unul dintre medicii veterinari chestionați nu efectuează teste de sensibilitate la antibiotice, utilizând antibiotice cu spectru larg de acțiune. Recurg la serviciile de laborator atunci când prescriu antibiotice patru (3%) medici veterinari și doar în trei (67%) cazuri motivul utilizării serviciilor de laborator a fost determinarea sensibilității la antibiotice. În 76% din cazuri, medicii veterinari nu efectuează teste de laborator înainte de a prescrie antibiotice din cauza lipsei unui laborator în apropiere, iar cinci respondenți consideră că testele de laborator nu sunt necesare.

Concluzii. (1) Chestionarea medicilor veterinari responsabili de utilizarea antimicrobienelor în creșterea animalelor reprezintă o măsură periodică obligatorie care poate contribui semnificativ la ajustarea metodelor de diminuare a fenomenului RAM. (2) Rezultatele anchetei au demonstrat principalele cauze ale neaprecierii sensibilității florei bacteriene la antimicrobiene o constituie accesul limitat la serviciile diagnosticului de laborator din cauza plasamentului la distanțe mari a laboratoarelor, obținerea tardivă a rezultatelor și costurile mari ale testelor de laborator. (3) Una din măsurile importante, care ar contribui eficient la reducerea RAM,. este utilizarea antimicrobienelor în baza prescripției veterinare care rămâne o problemă prioritară la nivel național.

SPECTRUL ŞI REZISTENŢA LA ANTIMICROBIENE ALE INFECŢIILOR TRACTULUI URINAR ASOCIATE CATETERIZĂRII

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Cuvinte-cheie: *spectrul microbian, rezistenţă la antimicrobiene, infecţii urinare asociate cateterizării.*

Introducere. Infecţiile tractului urinar reprezintă cca 40% din totalul infecţiilor asociate asistenţei medicale, majoritatea cazurilor fiind determinate de cateterizare. La pacienţii fără cateter, tractul urinar este colonizat, de obicei, de microorganisme de origine enterică. Prezenţa unui cateter creează un mediu favorabil pentru persistenţa microorganismelor şi formarea de biofilme, ceea ce creşte probabilitatea ca tractul urinar să fie colonizat de microorganisme nosocomiale nonenterice. Prin urmare, este important să cunoaştem spectrul agenţilor patogeni asociaţi cu prezenţa unui cateter urinar.

Scop. Evaluarea spectrului şi rezistenţei la antimicrobiene a tulpinilor microbiene izolate din infecţiile tractului urinar asociate cateterizării.

Material şi metode. Studiul realizat este unul secundar-sinteză narativă a literaturii. Informaţia a fost selectată din bazele de date electronice internaţionale, din 2018 până în 2023, cu privire la spectrul şi rezistenţa la antibiotice a infecţiilor urinare asociate cateterizării (ITUAC). În acest scop au fost folosite următoarele cuvinte-cheie în diferite combinaţii: cateter, infecţie urinară, bacteriurie, rezistenţă la antimicrobiene. În studiu au fost analizate cele 38 de publicaţii selectate, precum şi datele, şi recomandările Asociaţiei Europene de Urologie, Centrului de Prevenire şi Control al Bolilor (CDC), inclusiv Ghidul pentru prevenirea infecţiilor urinare asociate cateterizării.

Rezultate. Cateterismul permanent, care durează 30 de zile sau mai mult, este definit ca „pe termen lung” sau „cronic”. Durata prelungită a cateterizării instantaneu duce la formarea biofilmului pe suprafaţa acestuia. La pacienţii cateterizaţi pe termen lung, bacteriuria polimicrobiană apare în ~95% din cazuri. Cei mai frecvenţi agenţi patogeni asociaţi cu prezenţa unui cateter permanent sunt *E. coli* (21,4%) şi *Candida* spp. (21,0%), urmaţi de *Enterococcus* spp. (14,9%), *P. aeruginosa* (10,0%), *K. pneumoniae* (7,7%), *Enterobacter* spp. (4,1%) şi *A. baumannii* (1,2%). În rezumatul anual al datelor raportate de CDC, 24,8% dintre izolatele de *E. coli* şi 33,8% dintre izolatele de *P. aeruginosa* au fost rezistente la fluorochinolone. Tulpinile de *E. coli* şi de *K. pneumoniae* au prezentat rezistenţă în 5,5% din cazuri, inclusiv 21,2% la ceftriaxonă. Ratele de rezistenţă au fost relativ înalte chiar şi la carbapeneme: *E. coli* 4%, *K. pneuniae* 10%, *P. aeruginosa* 25% şi *A. baumannii* 25,6%. Rezistenţă semnificativă a fost înregistrată la vancomicină (6,1%) şi la ampicilină (3,1%) în cazul *E. faecalis*.

Concluzii. Cateterele urinare sunt recunoscute ca factor de risc major pentru dezvoltarea infecţiilor urinare asociate asistenţei medicale. Spectrul microorganismelor în ITUAC este foarte diferit, iar din cauza formării biofilmelor rezistenţa la antimicrobiene a acestor tulpini este înaltă. Agenţii patogeni din biofilme sunt bine protejaţi de antimicrobiene şi de apărarea gazdei. Deoarece terapia antimicrobală la pacienţii cateterizaţi duce la dezvoltarea rezistenţei la antimicrobiene, strategia prudentă de administrare a antibioticelor, gestionarea cateterului şi prevenirea ITUAC trebuie considerate prioritare.

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CALITATEA MICROBIOLOGICĂ A PRODUSELOR FARMACEUTICE NESTERILE

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Cuvinte-cheie: *controlul calităţii, produse farmaceutice, contaminare, microorganisme.*

Introducere. Evaluarea microbiologică a produselor farmaceutice nesterile este relevantă, deoarece contaminarea microbiană reduce sau anulează efectul lor terapeutic şi poate provoca infecţii. Microorganismele prezente în medicamente nu numai că le fac periculoase din punct de vedere infecţios, dar pot să modifice proprietăţile chimice, fizice şi organoleptice ale acestora sau conţinutul ingredientelor active. În plus, microorganismele pot produce metaboliţi cu proprietăţi toxice.

Scop. Analiza rezultatelor obţinute în urma testelor de puritate microbiologică a medicamentelor nesterile de către diferite companii farmaceutice şi prezentarea neconformităţilor înregistrate în grupele de medicamente studiate.

Material şi metode. A fost realizat un studiu secundar-sinteză narativă a literaturii. Informaţia a fost selectată din bazele de date electronice internaţionale folosind următoarele cuvinte-cheie în diferite combinaţii: produse farmaceutice, controlul calităţii, contaminare microbiologică. Prin căutare, au fost identificate 26 de publicaţii.

Rezultate. Există multe rapoarte internaţionale cu privire la neconformităţile în procesul de producere a medicamentelor care au ca rezultat acordarea autorizaţiilor de introducere pe piaţă a produselor de proastă calitate. Długaszewska şi colab. au raportat că pe o perioadă de 10 ani procentul mediu de probe neconforme a fost de 0,7%. O analiză a rezultatelor testelor de puritate microbiologică a medicamentelor combinate a arătat că până la 5,6% dintre acestea nu corespund standardelor aplicabile. Printre neconformităţile majore ies în evidenţă numărul excesiv de bacterii, de fungi şi de bacterii gramnegative. Charnock a evaluat conţinutul microbian al 77 de produse farmaceutice nesterile înregistrate ca mărci comerciale distribuite în Norvegia. Toate produsele examinate corespundeau normativelor aplicabile în ceea ce priveşte numărul şi tipurile de microorganisme izolate, ceea ce indică la eficacitatea practicilor de producere în conformitate cu standardele existente. Majoritatea bacteriilor izolate au fost bacili grampozitivi care formează endospori. Deşi bacilii gramnegativi au fost puşi în evidenţă foarte rar, unii dintre ei aparţineau unor specii indicate anterior ca agenţi patogeni oportunişti inadmisibili în produsele farmaceutice. Într-un studiu efectuat de Ratajczak şi colab., din 1285 de probe de medicamente studiate 24 de probe au prezentat neconformităţi. Cea mai frecventă neconformitate a fost depăşirea numărului maxim acceptabil de fungi. Fungii izolaţi au aparţinut genurilor: *Aspergillus, Rhizopus, Alternaria, Mucor.* Levuri nu au fost detectate. Micetele pot produce micotoxine cancerigene şi mutagene, de asemenea pot provoca intoxicaţii acute şi cronice, alergii, boli ale sistemelor respirator, digestiv şi leziuni hepatice.

Concluzii. Rezultatele obţinute pot îmbunătăţi calitatea producerii în fabricile farmaceutice, pot informa/conştientiza necesitatea controlului microbiologic al procesului de producţie al fiecărei serii de medicamente şi, prin urmare, pot îmbunătăţi siguranţa şi calitatea medicamentelor.

IMPACTUL METODELOR MICROBIOLOGICE ASUPRA DIAGNOSTICULUI INFECȚIEI CU CLOSTRIDIODES DIFFICILE

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Cuvinte-cheie: *Clostridioides difficile, metode, diagnostic, algoritm.*

Introducere. Bacteria *Clostridioides difficile* este o cauză frecventă a diareii asociate asistenței medicale. Forma clinică depinde de factorii ce țin de gazdă și de virulența tulpinii implicate în process. Acest microorganism provoacă boală care variază în severitate de la colonizare asimptomatică la diaree severă, colită pseudomembranoasă, megacolon toxic, perforație de colon și deces. Depistarea și raportarea cazurilor de ICD depinde de metodele de diagnostic utilizate. În unele țări, în special în cele cu venituri mici și medii, nu există cazuri de ICD raportate și, respectiv, nu se cunoaște amploarea poverii ICD. O altă provocare în diagnosticul de laborator al ICD o reprezintă și numeroasele metode propuse, fără a fi stabilit un algoritm de diagnostic bine determinat.

Scop. Evaluarea impactului metodelor microbiologice de diagnostic asupra diagnosticului infecției cu *C. difficile*.

Material și metode. Studiul reprezintă o sinteză narativă a literaturii de specialitate. Informația a fost selectată din bazele de date electronice internaționale folosind următoarele cuvinte-cheie în diferite combinații: *Clostridioides difficile*, metode, diagnostic, algoritm, standarde. Criteriul de bază de selectare a surselor bibliografice a fost disponibilitatea integrală și gratuită a articolelor. Pe baza acestor criterii au fost selectate 14 articole științifice.

Rezultate. Diferitele în strategiile de testare de laborator utilizate au un impact substanțial asupra ratelor de ICD raportate. Într-un studiu s-a constatat că la testarea independentă bazată pe PCR a fost obținut un număr dublu de rezultate pozitive, în comparație cu testarea independentă a toxinelor prin analiza imunoenzimatică (AIE). În mod similar, un alt studiu a demonstrat că testarea bazată pe PCR a probelor GDH-pozitive și toxine-negative aproape a dublat numărul de rezultate pozitive, în comparație doar cu testarea GDH și a toxinelor prin AIE. Testarea de laborator pentru ICD este un domeniu în schimbare permanentă și nu există un singur standard de referință acceptat sau un singur test considerat cel mai bun. Un test de diagnostic este evaluat în funcție de sensibilitate, de specificitate, de timpul de realizare, de costul și de disponibilitatea acestuia. Testul de neutralizare a citotoxicității celulare și de izolare a agentului în cultură pură sunt considerate teste de referință pentru diagnosticul de laborator al ICD. Din cauza timpului îndelungat de obținere a rezultatelor și a cerințelor tehnice, aceste teste nu sunt utilizate pe larg. De asemenea, a fost demonstrat că la utilizarea testelelor de sine stătător pentru diagnosticarea ICD nu se obțin rezultate fiabile, iar implementarea algoritmului în doi pași a redus numărul de rezultate fals pozitive și a permis divizarea pacienților în cei fără ICD, cei cu ICD și potențiali purtători de *C. difficile.*

Concluzii. Diagnosticul de laborator este foarte important pentru a diferenția ICD de alte cauze de diaree și pentru a stabili prezența infecției versus colonizare. Alegerea metodelor de diagnostic de laborator are un impact semnificativ asupra diagnosticului ICD și, prin urmare, asupra ratelor raportate de ICD.

PROBLEMA REZISTENȚEI LA ANTIBIOTICE ÎN MEDIUL OCUPAȚIONAL AL LUCRĂTORILOR MEDICALI: SINTEZĂ NARATIVĂ

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Cuvinte-cheie: *rezistența la antibiotice, factori de risc, mediul ocupațional, lucrători medicali.*

Introducere. Rezistența la antibiotice reprezintă una dintre cele mai serioase provocări la nivel global în tratarea bolilor infecțioase. Fenomenul de rezistență la antibiotice, când agenții patogeni suferă modificări care reduc sau elimină eficacitatea tratamentelor medicamentoase, devine o problemă din ce în ce mai urgentă la nivel mondial. Lucrătorii din domeniul medical se confruntă cu riscul de a contracta infecții severe și, uneori, fatale, care devin rezistente la tratamentele disponibile. Deși întreaga comunitate este expusă acestui pericol, anumite ocupații sunt mai expuse acetui pericol.

Scop. Studierea literaturii de specialitate pentru elucidarea antibiorezistenței ca factor de risc prezent la locul de muncă al angajaților din sectorul medical.

Material și metode. Studiul a fost fundamentat pe o analiză narativă a 21 de surse științifice din baze de date internaționale care abordează factorii de risc din mediul ocupațional, inclusiv rezistența la antibiotice, și care au impact asupra stării de sănătate a lucrătorilor medicali din instituțiile medicale din Republica Moldova.

Rezultate. În e spitalele și în unitățile de terapie intensivă, se conturează un mediu propice pentru proliferarea și răspândirea bacteriilor rezistente la antibiotice. Această situație este generată de utilizarea intensivă a antibioticelor în cadrul unei populații dense de pacienți, expunerea repetată a personalului medical și condițiile specifice din aceste instituții. Transmiterea bacteriilor rezistente includ poate avea loc prin contactul direct cu mâinile contaminate ale personalului, cu suprafețele infectate (mânerele ușilor și echipamentele utilizate), precum și prin contactul direct cu pacienții sau cu fluidele corporale ale acestora, care sunt o sursă primară de infecție. Numeroși agenți biologici, inclusiv bcteriile din aerul expirat sau toxinele eliberate de micete, sunt transmise prin intermediul aerului. Deficiențele în construcția clădirilor și în proiectarea sistemelor de aer condiționat nu asigură condiții optime de ventilație și de uscare a aerului

Concluzii. Starea reală din instituțiile medicale subliniază necesitatea unei abordări multidisciplinare și a unor măsuri preventive robuste pentru a gestiona rezistența la antibiotice în mediul ocupational al lucrătorilor medicali. Astfel, promovarea unor practici riguroase de igienă, gestionarea adecvată a echipamentelor și a suprafețelor, îmbunătățirea sistemelor de ventilare și a infrastructurii sunt aspecte esențiale în asigurarea unui mediu de lucru sănătos și sigur pentru personalul medical și pacienți.

RESISTANCE OF PATHOGENS CAUSING URINARY TRACT INFECTIONS IN CHILDREN

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Keywords: *urinary tract infection, children, microbial resistance.*

Introduction. Urinary tract infections are among the most common bacterial infections in children. Limited data on the pharmacokinetic and pharmacodynamic characteristics of antibacterial drugs in pediatric patients may lead to their excessive and irrational use, resulting in the development of complications. Currently, there is an alarming level of antimicrobial resistance among causative pathogens due to the inappropriate and widespread use of antibacterials. Resistant infections are associated with higher morbidity, mortality, and increased healthcare costs.

The aim of this study was to identify the causative pathogens of urinary tract infections in children and analyze their susceptibility to antibacterial preparations for optimal therapeutic management.

Material and methods. This analytical-retrospective study involved analyzing the results of bacteriological urine examinations in children admitted to the specialized wards of Valentin Ignatenco Children's Municipal Clinical Hospital in 2022, focusing on the identification of microorganisms and evaluation of susceptibility to antibacterial preparations.

Results. According to the data obtained, 39% positive urocultures were identified in children and the most common pathogen detected was *Escherichia coli* (43% cases), followed by *Staphylococcus epidermidis* (40%), *Enterococcus* (6%), *Proteus mirabilis* (5.5%), *Klebsiella pneumoniae* (4.5%), *Pseudomonas aeruginosa* (1%). *Escherichia coli* showed resistance to cephalosporins-cefaclor (47%), cefuroxime (51%), ceftazidime (40%), cefoperazone (61%), fluoroquinolones-ciprofloxacin (52%), ofloxacin (62%) and, amoxicillin/clavulanic acid in 42% cases. *Staphylococcus epidermidis* detected in 39% cases, was resistant to ceftriaxone (33%), ceftazidime (28%), cefuroxime (31%), ciprofloxacin (37%), amoxicillin/clavulanic acid in 53% cases. *Enterococcus* was resistant to ceftriaxone (32%), cefuroxime (37%), cefaclor (30%), ciprofloxacin (29%), ofloxacin (35%), amoxicillin/clavulanic acid (22%). *Klebsiella pneumoniae* was resistant to ceftazidime (35%), cefoperazone (27%), ceftriaxon (23%), cefuroxime (31%), cephalexin (27%), ciprofloxacin (38%), ofloxacin (46%), amoxicillin/clavulanic acid (27%), and *Pseudomonas aeruginosa* was resistant to ceftazidime (27%), ceftriaxon (20%), cefuroxime (40%), cephalexin (27%) ciprofloxacin (47%).

Conclusions. Urinary tract infections in children pose a significant challenge due to the specific features of the pediatric population, including the difficulty in conducting studies, limited therapeutic options, contraindications, variability of causative pathogens, and increased resistance to antibacterial therapy. *Escherichia coli* was the most commonly detected pathogen, with an elevated rate of resistance to amoxicillin/clavulanic acid and cephalosporins, which are commonly used for the treatment of urinary tract infections in children. Rational selection of antibacterials is essential to ensure effective and safe treatment and to prevent urinary tract infections with multidrug-resistant agents.

MALADII BACTERIENE CU MANIFSTĂRI NEUROLOGICE INFLUENŢATE DE SCHIMBAREA CLIMEI

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Cuvinte-cheie: *schimbarea clomei, maladii bacteriene, maladii neurologice, sănătate mintală, neuroinfecţii.*

Introducere. Schimbările climatice reprezintă una dintre cele mai mari şi mai urgente provocări ale secolului XXI. Creşterea temperaturilor medii şi a nivelului oceanelor, modelele modificate ale precipitaţiilor şi sporirea evenimentelor meteorologice extreme afectează nu numai peisajul şi ecosistemul global, ci şi sănătatea umană. Sănătatea creierului este în pericol ca urmsaare a creşterii incidenţei infecţiilor bacteriene, influenţate de schimbările climatice.

Scop. Cercetarea realizată a avut ca scop argumentarea legăturii infecţiilor bacteriene ale creierului cu schimbarea climei.

Material şi metode. A fost realizat un studiu bibliografic, fiind căutate îmbinările de cuvinte„schimbarea climei”, „maladii bacteriene” şi „maladii neurologice” în motorul de căutare disponibil gratuit, care oferă căutare de tip text-integral de publicaţii ştiinţifice open-acces – *Google Scolar*. Ca rezultat au apărut peste 40 de surse. Din ele au fost analizate 17, care au fost relevante tematicii.

Rezultate. În ultimii câţiva ani, incidenţa afecţiunilor bacteriene, care afectează sistemul nervos central (SNC), este în creştere. În pofida faptului că joacă un rol-cheie în controlul schimbărilor climatice, bacteriile sunt rareori obiectul de studii în cercetările tematice şi nu sunt luate în considerare la dezvoltarea de noi strategii. Bacteriile reacţionează la schimbările climatice pr4in reacţii de feedback atât pe termen lung, cât şi pe termen scurt şi care pot fi atât pozitive, cât şi negative.
Neuroinfecţiile crează un micromediu inflamator care poate avea consecinţe negative asupra calităţii vieţii umane, inclusiv disfuncţii cognitive, modificări comportamentale, depresie, convulsii, şi deficienţe fizice. Simptomele neurologice în bolile infecţioase care afectează SNC sunt, de obicei, neglijate. Printre primele zece boli infecţioase care cauzează deces la nivel mondial se regăsesc encefalita şi meningita – infecţii bacteriene ale sistemului nervos. Cei mai importanţi agenţi patogeni, care provoacă aceste afecţiuni, sunt *S. pneumoniae*, *Streptococcus agalactiae*, *Neisseria meningiditis*, *Haemophilus influenzae, Listeria monocytogenes*. Sepsisul polimicrobian este de asemenea o afecţiune medicală severă care provoacă simptome neurologice şi se agravează în timpul valurilor de căldură. Infecţiile pneumococice se pot răspândi prin aerosoli şi evoluează asimptomatic în colonizarea cavităţii nazale, de unde se răspândesc în alte organe, inclusiv în creier. Ajungând în fluxul sangvin şi, traversând bariera hematoencefalică, provoacă meningită. Recent a fost stabilită o relaţie între creşterea conţinutului de mercur în aerul atmosferic şi sporirea numărului pacienţilor afectaţi de encefalită cauzată de *Naegleria fowleri* în SUA şi în sud-estul Asiei. Pacienţii afectaţi suferă de meningoencefalită, a cărei rată a mortalităţii a rămas în jur de 98%, cu mai puţin de cinci supravieţuitori care au trecut de 50 de ani. Aceşti agenţi patogeni pot provoca, şi o boală a sistemului nervos central numită encefalită granulomatoasă.

Concluzii. Schimbările climatice au impact şi asupra bacteriilor, contribuind la prelungirea ciclului lor de viaţă, la sporirea agresivităţii şi a consecinţelor pe sănătatea umană, agravând sau chiar determinând unele afecţiuni neurologice.

BENEFITS AND CHALLENGES IN USING WETLANDS FOR WASTEWATER TREATMENT

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Keywords: *wastewater, constructed wetlands, sewage, public health.*

Introduction. Being a sustainable and eco-friendly method of treating wastewater, constructed wetlands use microorganisms and natural processes to eliminate pollutants. Depending on how they are built, put into use, and maintained, they may offer a number of advantages as well as disadvantages.

The aim of the study was to improve the management of constructed wetlands by promoting their benefits and minimizing the associated risks.

Material and methods. A review of national and international scientific bibliographic sources on best practices for implementing constructed wetlands in wastewater treatment was conducted.

Results. Constructed wetlands are ecological infrastructures designed to replicate the advantages and functions of natural wetlands. These areas effectively clean and manage water, including wastewater, in a sustainable manner by harnessing the power of bacteria and natural processes. They also serve as habitats for aquatic life, including fish, plants, and microbes, which work together to filter, clean, and regenerate water. Compared to conventional wastewater treatment methods, constructed wetlands use less energy, require fewer staff for maintenance, effectively treat wastewater with dispersed pollution, help preserve biodiversity, and provide habitat for a variety of plants and animals. Additionally, they can enhance the visual appeal of metropolitan areas and offer higher resilience to climate change and extreme events. However, despite the numerous benefits of constructed wetlands, there are also challenges. These include the need for more space than traditional technologies due to the use of extensive areas of vegetation and soil, potentially longer implementation and installation times, and the potential for the development and spread of antibiotic resistance. Local conditions such as soil type, climate, and hydrological features can influence the efficiency of constructed wetlands, necessitating adjustments for each unique circumstance. These challenges can be addressed through effective design, meticulous material and plant selection, and the application of engineering techniques to enhance efficiency. Furthermore, the implementation of new biotechnologies focused on combating antibiotic-resistant pathogenic flora can help mitigate these challenges.

Conclusions. Constructed wetlands remain a valuable choice for sustainable water resource management and environmental protection, despite the challenges involved. With proper integration and management, they can serve as sustainable solutions for issues related to water quality, flood management, and biodiversity conservation.

The research was conducted as part of the JPIAMR projects: "Phage treatment and wetland technology as an intervention strategy to prevent dissemination of antibiotic resistance in surface waters" (*PhageLand*).

ASPECTE EPIDEMIOLOGICE A INFECȚIILOR DE SITUS CHIRURGICAL: ANALIZĂ NARATIVĂ

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Cuvinte-cheie: *infecţii asociate asistenţei medicale, infecţie de situs chirurgical, infecţii postoperatorii, infecţii de plagă.*

Introducere. Infecţiile la nivelul locului chirurgical sunt infecţii frecvente asociate cu îngrijirea sănătăţii, care pot provoca daune severe pacientului, inclusiv deces, şi cresc costurile de îngrijire. Rezultatele unui studiu din 2017 au arătat că ratele de readmitere asociate cu aceste infecţii variau între 1,45% şi 6,34%. Lipsa conformităţii cu ghidurile de bune practici acceptate pentru îngrijirea perioperatorie poate creşte riscul pacientului de a dezvolta o infecţie de situs chirurgical.

Scopul lucrării a fost evaluarea legităţilor de manifestare a procesului epidemic şi factorii de risc în infecţile asociate asistenţei medicale (IAAM) la pacienţii cu situs chirurgical bazându-ne pe datele de literatură.

Material şi metode. S-a realizat studiu de tip secundar, sinteză narativă, cu utilizarea publicaţilor din bazele de date PubMed, Med ONE, NLM, NCBI, raporte anuale de activitate a spitalul de nivel republican. Perioada de referinţă – 2015-2022.

Rezultate. S-a stabilit, că infecţiile de situs chirurgical sunt responsabile de 38% din decesele cauzate de complicaţiile post-operatorii, iar durata medie a spitalizării pacientului cu infecţie de situs chirurgical creşte, în dependenţă de ţară şi zonă geografică cu 5-20 de zile. Costul suplimentar al tratamentului infecţiilor de situs chirurgical se situează între 170 şi 29600 $ în ţările cu venituri mici şi medii şi pot ajunge la 35000 $ în ţările cu venituri mari. Incidenţa generală a infecţiilor de situs chirurgical la nivel global ajunge la 2,5%, cu incidenţa cea mai înaltă în Regiunea Africană (aprox. 7%). Studiile au arătat că cea mai crescută incidenţă se înregistrează în Tanzania (26%), iar cea mai mică – în China (0,2%). Cele mai frecvent izolate microorganisme din secreţiile de plagă, la nivel mondial, sunt *S.aureus*, stafilococi coagulazo-negativi, *Enterococcus sp*. şi *E.co*li. Etiologia infecţiilor postoperatorii depinde în primul rând de sediul anatomic al intervenţiei şi de tipul de plagă chirurgicală. Astfel, din infecţii ale „plăgilor curate” se izolează cel mai frecvent *S. aureus*, iar din infecţii ale „plăgilor contaminate” şi ale „plăgilor murdare” se izolează în special bacili gram-negativi: *E. coli, Proteus spp., Klebsiella spp., Enterobacter spp., Pseudomonas spp.* etc. În cazul „plăgilor contaminate”, a „plăgilor murdare” s-a constatat deseori o etiologie polimicrobiană aerobă şi anaerobă, asemănătoare cu flora endogenă a organului rezecat. Prevalenţa consumului de antimicrobiene constituie 42,7%. Fiecare al 3-a pacient spitalizat primeşte cel puţin un preparat antimicrobian. În mediu la un pacient sunt administrate 1,3 antimicrobiene. Pacienţii cu intervenţii chirurgicale invazive fac IAAM de circa 3 ori mai frecvent, prevalenţa IAAM constituind 6,9% şi de 2 ori mai frecvent consumă antimicrobiene, cu o prevalenţă de 99% de cazuri.

Concluzii. Infecţia de situs chirurgical se numără printre cele mai frecvente complicaţii postoperatorii şi este responsabilă de ratele mari de morbiditate şi mortalitate. Prevenirea acestor infecţii este o sarcină majoră şi necesită eforturi deosebite cu utilizarea complexă a mai multor măsuri, inclusiv este nevoie de mai multe cercetări. Determinarea legităţile procesului epidemic în IAAM la pacienţii internaţi în secţiile cu profil chirurgical, factorii de risc, spectrul de microorganisme izolate şi rezistenţa/sensibilitatea faţă de preparate antimicrobiene ne permite să obţinem limete de securitate postoperatorie şi argumentează necesitatea elaborării „Ghidul de prevenire şi control al IAAM”.

ANALIZA DIVERSITĂŢII GENETICE A VIRUSULUI SARS-CoV-2: REVISTA LITERATURII

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Cuvinte-cheie: *SARS-CoV-2, COVID-19, monitorizarea genetică, variante de mutaţii, secvenţierea, proteina S.*

Introducere. Pandemia COVID-19, provocată de SARS-CoV-2, un agent patogen din familia *Coronaviridae*, a evidenţiat necesitatea înţelegerii evoluţiei virusului. De la secvenţierea iniţială în ianuarie 2020, virusul a acumulat mii de mutaţii, majoritatea în proteina S şi domeniul său de legare la receptor (RBD). Aceste mutaţii influenţează afinitatea pentru receptorul celular ACE2, crescând infectivitatea şi generând noi variante. Secvenţierea genomului reprezintă una dintre metodele optime pentru monitorizarea variantelor de COVID-19, identificarea şi studierea rapidă a modificărilor în structura genomului virusului şi evoluţia acestuia.

Scop. Studiul vizează analiza variabilităţii genetice a SARS-CoV-2 pe parcursul pandemiei, cu accent pe mutaţiile semnificative şi impactul lor.

Material şi metode. Pentru realizarea studiului, a fost efectuată analiza narativă a literaturii, selectând articole relevante publicate între 2020-2023, folosind cuvintele: „genome sequencing", „SARS-CoV-2", „mutation analysis" şi „genovariants". În total, au fost examinate 48 de surse bibliografice, utilizând baze de date precum Embase, PubMed, Hinari, Google Academic şi bibliotecile naţionale.

Rezultate. Monitorizarea genetică a relevat cinci variante de îngrijorare conform OMS: Alpha (B.1.1.7), Beta (B.1.351), Gamma (P.1), Delta (B.1.617.2) şi Omicron (B.1.1.529). Aceste variante prezintă mutaţii multiple în proteina S, în special în RBD, conducând la contagiozitate crescută şi evaziunea imunitară. Alpha, detectată iniţial în Regatul Unit al Marii Britanii, are aşa mutaţii ca N501Y în RBD, care îmbunătăţesc afinitatea pentru receptorul ACE2. Beta, identificată în Africa de Sud, prezintă mutaţii precum K417N şi E484K, crescând evaziunea imună. Gamma, cu origine în Brazilia, are mutaţii similare cu Beta, influenţând atât transmisibilitatea, cât şi recunoaşterea imunitară. Delta, apărută în India, cu mutaţii ca L452R şi P681R, a generat rate mai mari de infectare şi de replicare virală. Varianta Omicron provine din linia genetică B.1.1. Genomul variantei Omicron are mai mult de 30 mutaţii în proteina S, în comparaţie cu tulpina de referinţă Wuhan-Hu-1, dintre care 15 substituţii de aminoacizi în domeniul de legare la receptor (RBD), zonele asociate cu răspunsul imun. Mutaţiile prezente în Omicron pot fi asociate cu transmisibilitate mai mare, legare alterată la receptor şi evaziune imună. Studiul subliniază importanţa proteinelor structurale în adaptarea virusului la noi gazde şi în patogeneza COVID-19, esenţială pentru dezvoltarea strategiilor de control al pandemiei.

Concluzii. Analiza datelor din literatură evidenţiază importanţa monitorizării mutaţiilor pentru înţelegerea dinamicii pandemiei şi dezvoltarea strategiilor de control, inclusiv îmbunătăţirea vaccinurilor. Proteina S, în particular, joacă un rol crucial în patogeneza infecţiei şi este un punct focal în cercetările viitoare.

MARKETING SOCIAL ÎN CAMPANIILE DE VACCINARE

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Cuvinte-cheie: *sanatatea publică, ezitare la vaccinare, marketing social „4P”, promovarea sănătăţii.*

Introducere. Vaccinurile sunt considerate una dintre cele mai importante realizări ale medicinei moderne. La nivel mondial, în fiecare an, vaccinările previn până la trei milioane de decese. Studiile au arătat că vaccinurile sunt printre cele mai sigure injecţii disponibile, reacţiile adverse fiind rare. Totuşi, în pofida creşterii disponibilităţii vaccinurilor, o bună parte a populaţiei nu se vaccinează şi nu vaccinează copiii din motive ce nu ţin de contraindicaţii medicale.

Scop. Analiza impactului marketingului social în companiile de vaccinare.

Material şi metode. Studiu secundar, sinteză narativă. Din bazele de date electronice internaţionale, inclusiv *Scopus*, *PubMed*, *Web of Science* şi G*oogle Scholar*. Au fost analizate 180 de surse, cele mai actuale informaţii, ghiduri, publicaţii recente în limbile română, rusă, engleză, privind aplicarea principiilor de marketing social în promovarea vaccinării. Perioada de referinţă au fost anii 2012-2023. Ultima căutare a literaturii a fost efectuată în decembrie 2023. Metodele de cercetare utilizate: descriptivă, analitică.

Rezultate. Imunizarea sistematică a populaţiei reduce în mod semnificativ morbiditatea, mortalitatea şi invaliditatea cauzate de bolile infecţioase care pot fi prevenite prin vaccinare, reducând astfel semnificativ daunele sociale şi economice induse de acestea. Principiile marketingului social privind promovarea campaniilor de vaccinare devine una din activităţie principale în contextul ezitării populaţiei privind vaccinarea. Sistemele de comunicare publică şi de masă, utilizate pentru a promova administrarea vaccinurilor, se încadrează în categoriile de activităţi promoţionale: publicitate, relaţii publice. În condiţiile actuale, strategiile de modificare a percepţiilor publicului cu privire la vaccinuri ar trebui să includă furnizarea de informaţii imparţiale şi cuprinzătoare, adaptate nevoilor de informare a populaţiei şi transmise prin tehnologii de comunicare multiple şi noi precum mass-media socială, intervenţiile electronice – serviciile de mesagerie text şi reţelele sociale, aspecte ce ţin de marketing.

Concluzii. În formularea strategiilor de promovare a vaccinării, în marketingul social, ca şi în cel comercial, se recurge la mixul de marketing, reprezentat de 4P: produs, preţ, plasament, promovare. Marketingul social în sănătate reprezintă un potenţial important pentru atingerea unui grup de persoane cât mai mare posibil cu costurile cele mai mici, printr-un program eficient, care să satisfacă consumatorii cât mai mult. Este necesar ca speciliştii în marketing să fie incluşi în grupurile de lucru desemnate să elaboreze planuri şi politici de promovare a sănătăţii (inclusiv campanii de imunizare) în rândul populaţiei. Aceştia, la rândul lor, vor ţine cont de elementele de fundamentare a strategiei de marketing social pentru a atinge cele mai bune rezultate în campaniile de promovare a vaccinării.

CYTOGENETIC STUDY IN MEN FROM THE POPULATION OF THE REPUBLIC OF MOLDOVA

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Keywords: *karyotype, chromosomal abnormalities, male, infertility.*

Introduction. Globally, it is estimated that 15% of couples experience infertility, with male factors contributing to half of these cases. The causes of male infertility are highly diverse, often stemming from disorders in spermatogenesis, which are clinically characterized by severe oligozoospermia and azoospermia. Genetic factors account for approximately 30% of male infertility cases associated with spermatogenesis disorders. Among the numerous genetic causes of spermatogenic insufficiency, chromosomal anomalies are particularly significant from a clinical standpoint. Approximately 15% of men with non-obstructive azoospermia and 4% of men with severe oligozoospermia have chromosomal abnormalities.

The aim of this study is to evaluate the profile of chromosomal variations in male infertility with severe oligozoospermia and azoospermia, in order to confirm the importance of exploring their cytogenetics for both diagnosis and treatment, as well as for assessment and prognosis.

Material and methods. The study involved a retrospective descriptive analysis of cytogenetic results in men with oligozoospermia and azoospermia in the Moldovan population from 2014 to 2022. The participants were individuals from infertile couples who sought assistance at the National Center for Reproductive Health and Medical Genetics. The patient group comprised 32 individuals with oligozoospermia and 156 with azoospermia. All patients underwent cytogenetic analysis using the classical G-banding technique on peripheral blood lymphocytes. Results were reported using the nomenclature outlined in the 2016 International System of Cytogenetic Nomenclature (ISCN).

Results. The average age of men was 33.8±5.3 years, (95% CI: 32.7-34.9; median 33.0) (IGR 25 – 75: 30.0-36.0). Among the 156 men with azoospermia, 101 (64.7%) had a normal karyotype of 46,XY, while variations in the number or structure of chromosomes were observed in 55 individuals (35.3%; 95% CI: 27.8-42.8). Of these, 43 patients (27.6%) exhibited variations in the X or Y sex chromosomes, and 12 patients (7.7%) showed variations in the autosomal chromosomes. Among the 43 azoospermic patients with sex chromosomal abnormalities, 32 cases involved chromosomal number abnormalities, and 11 cases involved structural variations. Among the number of chromosomal abnormalities, 29 cases were attributed to X aneuploidy (Klinefelter Syndrome), 2 cases were mosaic 45,X/46,XY, and one case presented a 46,XX karyotype in a male. The structural variations of the sex chromosomes included 9 cases of duplications of the distal arm of the Y chromosome (Yqh+) and 3 cases of deletions of the distal arm of the same chromosome (Yqh-). Among the 32 men with oligozoospermia, 28 (87.5%; 95%CI: 76.0-99.0) had a normal karyotype of 46,XY, while 4 (12.5%; 95% CI: 1.0-24.0) exhibited variations in the sex chromosomes.

Conclusions. In this study, the prevalence of chromosomal abnormalities identified in azoospermia was 35.3% and in oligozoospermia was 12.5%. Given the high frequency of chromosomal abnormalities in men with infertility, as well as the genetic risks for future generations, it is important to assess the profile of chromosomal variations before resorting to Assisted Reproduction Techniques, for diagnosis, treatment, as well as assessment and prognosis.

RISCUL VARICELOR RECIDIVANTE ŞI VARIANTELE ANATOMICE ALE VENEI SAFENE MARI

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Cuvinte-cheie:
vena safenă mare, afluenţi, vena safenă mare dublă, vene safene accesorii, joncţiunea safenofemurală.

Introducere. Conform surselor bibliografice, rata varicelor recidivante ale membrului inferior după intervenţia chirurgicală variază de la 20-40% până la 50-60% sau chiar 80% din cazuri. Cauzele dezvoltării acestora pot fi greşelile tehnice şi tactice, progresarea maladiei, neovascularizarea, precum şi prezenţa variantelor anatomice ale venei safene mari – vena safenă mare dublă şi venele safene mari accesorii.

Scopul lucrării a fost de a evalua frecvenţa variantelor anatomice ale venei safene mari la nivelul coapsei şi a afluenţilor acesteia în apropiere de joncţiunea safenofemurală.

Material şi metode. În vederea elucidării temei abordate, a fost organizat un studiu descriptiv transversal în cadrul căruia au fost disecate 22 de vene safene mari la nivelul coapsei şi afluenţii acestora. Studiul a fost efectuat pe cadavre formolizate în cadrul Catedrei de anatomie şi anatomie clinică, a Universităţii de Stat de Medicină şi Farmacie *Nicolae Testemiţanu* din Republica Moldova.

Rezultate. Segmentul femural al venei safene mari, situat pe faţa anteromedială, urca cranial pe coapsă şi se vărsa în vena femurală, formând joncţiunea safenofemurală. În nouă cazuri (40,9%), vena safenă mare era solitară la nivelul coapsei, în trei cazuri (13,6%) se dubla la nivelul tuberozităţii tibiale, iar zece cazuri (45,5%) era însoţită de vene safene accesorii (anterioară şi/sau superficială), plasate paralel cu vena safenă mare. Vena safenă mare accesorie superficială a coapsei, întâlnită în două cazuri (9,1%), era separată de vena safenă mare prin fascia safenă.

Vena safenă mare accesorie posterioară, cu un traiect ascendent pe faţa posterioară a coapsei, a fost depistată în 15 cazuri (68,2%). Ea înconjura medial coapsa, dar se vărsa în vena safenă mare evident distal de joncţiunea safenofemurală.

Segmentul terminal al venei safene mari, situat în proximitatea joncţiunii safenofemurale, a primit următorii afluenţi: vena circumflexă iliacă superficială – în 72,7 % din cazuri, vena epigastrică superficială – în 68,2%, vena pudendală externă superficială – în 68,2%, vena pudendală externă profundă – în 9,1%, vena safenă mare accesorie anterioară a coapsei în – 40,9%, vena safenă mare accesorie superficială a coapsei – în 4,55%.

Concluzii. Venele superficiale ale membrului inferior prezintă o variabilitate anatomică înaltă, încă puţin studiată. Segmentul femural al venei safene mari, în aproape jumătate din cazuri, este însoţită de vene safene accesorii (anterioară şi/sau superficială) cu un traiect paralel. Vena safenă mare accesorie posterioară a coapsei, prezentă în 68,2% din cazuri, se varsă în vena safenă mare în treimea mijlocie a coapsei.

Imposibilitatea ligaturării sau ligaturarea incompletă şi/sau greşită a tributarelor venei safene mari, prezenţa venei safene duble sau a venelor safene accesorii pot duce la apariţia recidivei varicelor membrelor inferioare.

ASSESSMENT OF THE LIFE QUALITY AFTER SURGERY FOR PERFORATED GASTRODUODENAL ULCER

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Keywords: *perforated ulcer, surgery, quality of life, GSRS, MOS SF-36.*

Introduction. Quality of life is a cornerstone in the assessment of modern medicine, surgery, and healthcare practices. This aspect is becoming increasingly important in healthcare due to the rapidly increasing power, variety, and cost of modern drugs and surgical techniques.

The aim of this study was to assess and compare the quality of postoperative recovery in patients undergoing surgery for perforated ulcer.

Material and methods. This observational, prospective study was conducted on 231 patients undergoing emergency surgery. Postoperative recovery was assessed using the GSRS and MOS SF-36. The control group for the studied parameters consisted of average data obtained from 42 healthy individuals from the general population, with similar socioeconomic status and age to the main group of the study.

Results. The research presents an analysis of 231 patients, mostly men, which is considered statistically conclusive ($p<0.0001$), with a diagnosis of perforated gastroduodenal ulcer resolved by various surgical interventions. The majority of the patients were of working age, highlighting the significant social and economic impact of these issues. The remote follow-up period extended up to 10 years (average 7 years) after the primary surgery. The correlation between the life quality index and the surgical approach applied was statistically significant ($p<0.001$). In the comparative analysis, it was noted that, among all the surgical intervention methods, the groups undergoing ulcer excision, ulcer excision with vagotomy, and gastric resection yielded the least favorable results ($p<0.001$). The data obtained confirm that the GSRS and MOS SF-36 indices of life quality in patients undergoing the laparoscopic approach are statistically significantly better and more sustainable, impacting the effectiveness of treatment resolution ($p<0.001$).

Conclusions. Life quality measures are increasingly used worldwide to assess surgical outcomes, as they focus on specific health conditions and are essential for detecting changes resulting from treatment, thereby assessing its effectiveness. The present study found that the differential selection of treatment tactics using modern minimally invasive technologies, especially laparoscopic suturing of perforated gastroduodenal ulcer, demonstrated significantly better efficiency compared to traditional, especially radical, operations.

INDICII CALITĂȚII VIEȚII LA BOLNAVII CU HEPATOPATII CRONICE DIN REPUBLICA MOLDOVA

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Cuvinte-cheie: *calitatea vieții, chestionarul SF-36, infecție cronică HBV, boala ficatului gras metabolic asociat.*

Introducere. Calitatea vieții prezintă un indicator strategic al sănătății în diferite patologii cronice, în special a celor hepatice. Conceptul general al calității vieții este utilizat pe scară largă, reflectând o corelare directă dintre calitatea vieții și bunăstarea personală.

Scop. Studierea indicilor calității vieții la bolnavii cu hepatopatii cronice în condițiile Republicii Moldova.

Material și metode. Pentru realizarea scopului au fost examinați 59 de pacienți adulți cu diferite hepatopatii cronice: 34 de pacienți cu infecție cronică cu virusul hepatic B (Lotul HBV), 25 de pacienți cu boala ficatului gras metabolic asociat (Lotul BFG) și 26 de persoane adulte aparent sănătoase (Lotul-martor). Pentru evaluarea calității vieții (QoL) a fost folosit chestionarul SF-36 cu ajutorul căruia au fost apreciate opt scoruri: PF – funcționarea fizică; RF-P – funcționarea determinată de starea fizică; BP – durerea corporală; GH – starea generală de sănătate; VT – vitalitatea; SF – funcționarea socială; RF-E – funcționarea determinată de starea emoțională; MH_2 – sănătatea mintală. Pe baza sumării indicilor acestor scoruri au fost apreciați doi parametri integrali: Indicele de sănătate fizică (PH) și Indicele de sănătate mintală (MH). Datele obținute au fost prezentate în formatul M±m și analizate utilizând programul de completare „Analiza datelor” Excel 2016. Pentru a determina nivelul de semnificație statistică (p), a fost aplicat testul Mann-Whitney neparametric. Pentru efectuarea studiului a fost obținut acordul Comitetului de Etică a Cercetării Științifice a USMF „Nicolae Testemițanu”, Chișinău, Republica Moldova, nr.10 din 28.12.2020.

Rezultate. Nivelurile cele mai scăzute ale parametrilor integrali ai QoL s-au înregistrat în lotul HBV: PH=46,69±1,22, MH=38,97±1,56. În lotul BFG s-au atestat următoarele valori ale acestor parametri: PH=47,02±1,37 și MH=45,91±1,55. Veridicitatea diferențelor între loturi pentru PH și MH a fost de 95% și de 99%, respecftiv. În lotul-martor, valorile medii pentru acești parametri au fost PH=53,01±0,91 și MH=51,51±1,50, existând diferențe veridice ale acestor parametri cu ambele loturi de studiu ($p<0,01$; $p<0,01$). Analiza rezultatelor scorurilor reflectă menținerea tendinței valorilor minimale în lotul HBV. O scădere mai accentuată se observă în scorurile RF-E al MH. Acest indicator a fost apreciat cu punctaje scăzute în loturile HBV și BFG (37,25±4,83 și 66,67±7,45) versus lotul-martor 93,59±3,71 ($p<0,01$; $p<0,01$; $p<0,01$). Scorurile PH ale QoL demonstrează o scădere mai slabă a punctajelor. Cel mai afectat scor s-a dovedit a fi RF-P: 52,94±5,76 in lotul HBV și 68,00±5,69 în lotul BFG, versus 89,42±3,45 în lot ul-martor. Scorul GH a fost apreciat cu punctaje scăzute în loturile HBV și BFG (52,32±2,84 și 53,88±3,50), versus lotul-martor: 70,54±3,05 ($p<0,01$; $p<0,01$).

Concluzii. În hepatopatiile cronice descresc toate componentele ce caracterizează calitatea vieții, cel mai mult fiind afectată componenta mintală. Scăderi mai semnificative ale calității vieții se înregistrează în lotul de pacienți cu infecție cronică cu VHB.

EFECTELE SCHIMBĂRILOR CLIMATICE ASUPRA CALITĂȚII APEI POTABILE

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Cuvinte-cheie: *schimbări climatice, apa potabilă, sănătate, maladii hidrice.*

Introducere. Schimbările climatice, una dintre cele mai presante probleme ale secolului nostru, afectează profund calitatea apei potabile. Modificările în precipitaţii, temperatură şi evenimente meteorologice extreme au un impact semnificativ asupra disponibilităţii şi purităţii apei, ceea ce constituie o provocare majoră pentru sănătatea publică şi sustenabilitate.

Scop. Studiul îşi propune să analizeze efectele schimbărilor climatice asupra calităţii apei potabile, examinând modificările în compoziţia chimică şi microbiologică, şi impactul acestora asupra sănătăţii umane.

Material şi metode. S-au folosit date din rapoarte din domeniul sănătăţii şi a mediului, studii climatologice şi analize ale calităţii apei din diverse ţări. Metodologia include analiza comparativă a datelor istorice şi actuale privind calitatea apei, cu un accent special pe zonele afectate de secetă, de inundaţii sau de alte fenomene climatice extreme.

Rezultate. Rezultatele studiului subliniază o deteriorare semnificativă a calităţii apei în diverse regiuni ale lumii. În India, în ultimii 20 de ani, nivelurile de nitraţi din apele subterane au depăşit valorile normate cu peste 50% din cauza perioadei de secetă prelungită. Mai mult de atât, rapoartele recente indică o creştere cu 35% a contaminării bacteriene în aceleaşi surse de apă. În Europa, creşterea medie a temperaturilor cu 2°C în ultimele decenii a accelerat proliferarea algelor toxice în lacuri şi în rezervoare, afectând astfel calitatea a 30% din sursele de apă potabilă. Studii efectuate în bazinul Rinului au arătat o creştere cu 40% a concentraţiilor de fitoplancton nociv. În Africa, analizele de după inundaţii au relevat că peste 60% din sursele de apă testate în zonele afectate sunt contaminate cu *E. coli* şi cu alte bacterii patogene, sporind riscul de boli diareice, care afectează anual milioane de persoane. În America de Nord, modificările modelului de precipitaţii au determinat o creştere alarmantă, cu 25, a concentraţiilor de plumb şi de alte metale grele în anumite surse de apă. Rapoarte recente din Statele Unite ale Americii indică faptul că peste 18 milioane de oameni sunt expuşi la niveluri de contaminare cu plumb care depăşesc limitele recomandate de Agenţia pentru Protecţia Mediului. Aceste tendinţe au un impact direct asupra sănătăţii publice. De exemplu, în regiunile afectate de contaminarea cu nitraţi şi cu metale grele, s-a înregistrat o creştere cu 20% a cazurilor de afecţiuni renale şi cu 15% a cazurilor de cancer asociate cu calitatea apei. Aceste cifre subliniază importanţa unei gestionări mai eficiente a resurselor de apă şi a implementării de soluţii adaptate la schimbările climatice pentru a asigura accesul la apă potabilă de înaltă calitate.

Concluzii. Schimbările climatice au un impact evident şi alarmant asupra calităţii apei potabile. De aceea, este esenţial să se intensifice eforturile de monitorizare şi de purificare a apei, precum şi să se dezvolte strategii adaptative pentru gestionarea resurselor de apă în contextul noilor provocări climatice. Protejarea calităţii apei este vitală pentru sănătatea şi securitatea populaţiilor la nivel global.

INVESTIGATION OF PANCREATIC ACTIVITY PARAMETERS IN PATIENTS DURING COVID-INFECTION IN REPUBLIC OF MOLDOVA

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Keywords: *chronic diffuse liver disease, pancreatic function, COVID-19 disease, viral infection.*

Introduction. Chronic diffuse liver diseases continue to rank high among the issues contributing to high mortality rates worldwide. The Republic of Moldova holds the leading position globally in terms of the highest age-standardized incidence rate of liver cirrhosis and related mortality cases. The 2019 coronavirus (COVID-19) pandemic has posed yet another significant challenge for healthcare systems worldwide, given its potential to affect internal organs and cellular mechanisms. The investigation of metabolic interrelations between different organs and systems of the organism, particularly those with an injured liver affected by this new virus, has garnered significant scientific interest in recent years.

The aim. The study of pancreatic activity parameters in chronic liver patients during SARS-CoV2 infection in Republic of Moldova.

Material and methods. Our study represents a retrospective analysis of consecutively hospitalized patients in the COVID-19 unit at the *Timofei Moşneaga* Republican Clinical Hospital in Chişinau, Republic of Moldova, during the period from March 2020 to February 2021. A total of 73 patients' electronic medical records were examined, comprising 40 men (54.8%) and 33 women (45.2%), with a mean age of 57.29 years. All patients were categorized into three main groups: group 1 (13) consisted of individuals with chronic hepatitis (CH) of viral etiology (HBV, HCV); group 2 (13) included patients with liver cirrhosis (LC); and Comparison group 3 (CG) comprised patients (23) without liver pathology but with some functional issues related to the pancreas. Patients with metabolically associated liver disease (2) and nonspecific reactive hepatitis (22) were not considered in this analysis. Blood serum levels of glucose, amylase, and lipase were investigated in all patients. The analysis involved the use of percentages, means, and standard error of the mean for evaluation and statistical analysis, with the significance of differences (p) assessed using the Mann-Whitney U test.

Results. Elevated blood glucose levels, indicative of disrupted carbohydrate metabolism, were observed in patients with liver cirrhosis (6.38±1.23 mmol/L) and in the Comparison group (CG) (8.11±0.79 mmol/L), exceeding the normal reference values of 4.11-5.89 mmol/L. Amylase activity was within normal limits across all patient groups. In contrast, the level of lipase, a more sensitive indicator of pancreatic function, was elevated in the CG (112.13±82.16 u/L) compared to the normal range of 13-60 u/L. These findings align with the main hypothesis and recent literature, suggesting that COVID-19 may induce pancreatic injury in patients with chronic liver disease and those with a history of pancreatic issues, highlighting the human pancreas as another potential target for the virus.

Conclusions. COVID-19 can provoke functional abnormalities in pancreatic function in patients with chronic diffuse liver disease, as well as in patients with a history of underlying pancreatic dysfunction. Further investigation of pancreatic function is needed in a larger group of patients with different liver pathologies.

DENTAL EDUCATION FOR MEDICAL STUDENTS

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Keywords: *medical students, Dental medicine discipline*

Introduction. Oral health disparities constitute a significant public health challenge, with far-reaching consequences for the overall health of the population and economic ramifications for any nation. Introducing medical students to the field of dentistry and equipping them with essential knowledge about risk factors and preventive measures for common dental conditions can foster greater responsibility within the medical community towards oral health and enhance communication among healthcare specialists.

The objective of this study was to assess the opinions of students at the Faculty of Medicine regarding the Dental Medicine discipline.

Material and methods. This study involved conducting a survey among medical students in two stages: Stage I, before the commencement of the Dental Medicine module, and Stage II, upon its completion. In Stage I, data were collected from 356 fourth-year students at the Faculty of Medicine. Among them, 77% were high school graduates, 20% graduated from medical colleges, and 3% from other educational institutions. In Stage II, 386 students who had completed the Dental Medicine module were surveyed. The questionnaire consisted of general information about the participants and inquiries about the necessity and significance of including this discipline in the curriculum at the Faculty of Medicine.

Results. According to the gathered data from Stage I, 63.6% of respondents confirmed that Dental Medicine was a new subject for them. Additionally, 60.1% expressed their willingness to study it, 67.8% believed it would be beneficial, and 85.8% recognized a close connection between oral and general health. Regarding its inclusion in the curriculum, 65% considered it should be mandatory, while 35% suggested it be optional. After completing the module, 100% of participants acknowledged a strong correlation between oral health and general health. They asserted that Dental Medicine should be a compulsory discipline in the Faculty of Medicine's curriculum and is indispensable for medical students.

Conclusions. Introducing medical professionals to the field of dentistry, coupled with comprehensive knowledge of anatomical structures, physiological functions, and pathological conditions, enhances patient trust in the medical profession and fosters better inter-professional communication. Given that many oral conditions are preventable or treatable with early intervention, training physicians and actively involving them in oral disease prevention efforts can help mitigate oral health disparities and their adverse effects on overall health. Through oral screenings, preventive measures, referrals for dental assessments, and collaborations with dentists, physicians can positively impact oral health outcomes, especially in vulnerable populations.

STRATEGII DE PROMOVARE ÎN MARKETINGUL CABINETULUI STOMATOLOGIC

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Cuvinte-cheie: *marketing social, promovare, managementul marketingului, strategie.*

Introducere. Piaţa serviciilor stomatologice este una foarte competitivă. În prezent, problema medicilor stomatologi este fluxul de pacienţi. Pentru atragerea acestora, cabinetele stomatologice se ocupă de publicitate în diverse moduri: în cabinetele mici şi mijlocii această misiune şi-o asumă proprietarul, iar în cele mari pot exista subdiviziuni specializate în publicitate care dispun de buget propriu şi organizează acţiuni de publicitate cu participarea companiilor specializate. Rolul marketingului în domeniul serviciilor de sănătate este de a contribui la furnizarea de informaţii de valoare clienţilor.

Scop. Analiza instrumentelor publicitare şi recomandarea unor tehnici moderne de promovare a cabinetului stomatologic.

Material şi metode. Populaţia studiului: pacienţii a cinci cabinete stomatologice cu următoarele specializări: terapie, ortopedie, chirurgie dentoalveolară, parodontologie. Cei 138 de pacienţi care şi-au dat acordul verbal de a participa la studiu, au completat un chestionar autoadministrat, compus din 22 de întrebări grupate în şase categorii. Perioada de colectare a datelor: decembrie 2022 – februarie 2023. În cadrul studiului a fost studiată şi documentaţia internă a cabinetelor stomatologice precum: raportul de autoevaluare semnat de conducătorul cabinetului; copia regulamentului de organizare şi de funcţionare; organigrama; schema sistemului informaţional; planul de dezvoltare strategică; planul de control a calităţii serviciilor prestate. Metode de cercetare utilizate: descriptivă, analitică, comparativă.

Rezultate. Strategiile de marketing se propun ca o modalitate sigură de a construi relaţii cu publicul cabinetului stomatologic. Pentru un stomatolog, strategia de marketing înseamnă a convinge pacienţii că se oferă o experienţă fără durere şi cu rezultate aşteptate. Conform studiului, există următoarele modalităţi de promovare: recomandarea – 31%, reţelele de socializare – 25%, google – 17%, televiziunea – 15%, plasarea – 12%. Satisfăcuţi de condiţiile prestate de cabinetele stomatologice au fost 73,18% din persoanele intervievate, 92,75% au fost mulţumite de atitudinea personalului medical, iar 56,52% au ales cabinetul stomatologic în baza reputaţiei medicilor. Aplicarea unui sistem de marketing contribuie la formarea categoriilor de pacienţi planificaţi şi pacienţi continuu. La intrebarea ce il face pe pacient să fie fidel unui anume cabinet stomatologic, 32 % au numit aplicarea unui plan de marketing, 25% – condiţii favorabile de muncă ale medicilor. S-a constatat că pacienţii doresc mai multă computerizare a serviciilor medicale.

Concluzii. Publicitatea în stomatologie oferă pacienţilor posibilitatea de a alege tratamentul, clinica, medicul. Dintre metodele şi instrumentele de promovare analizatee s-au evidenţiat recomandarea me dicului din parte pacientilor, strategiile legate de promoţii şi cele privind preţul, prezenţa saitului propriu. O strategie eficientă s-a dovedit a fi promovarea în mediul online îmbinată cu cea offline. Aplicarea unor strategii de marketing eficiente duce nu numai la creşterea satisfacţiei clienţilor, dar şi a personalului – un element component în managementul unui cabinet stomatologic.

WORK PACKAGES / PACHETE DE LUCRU

WP1

Coordonare

Partener principal: ICRA
Participare: toți partenerii

Afișați mai multe

WP2

Investigații de sănătate publică în LMIC-urile europene

Partener principal: NTSU
Participare: ICRA, UW

Afișați mai multe

WP3

Performanța zonelor umede construite în condiții de câmp

Partener principal: ICRA / NTSU
Participare: UW, QIB

Afișați mai multe

WP4

Biopurificare pe bază de fagi

Partener principal: KUL / QIB
Participare: UW, TUD

Afișați mai multe

WP5

Analiza de risc

Partener principal: SGGW / ICRA
Participare: UW, KUL, QIB, TUD

Afișați mai multe

WP6

Dovada de concept

Partener principal: TUD / UW
Participare: ICRA, QIB

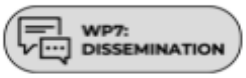
Afișați mai multe

WP7

Diseminare

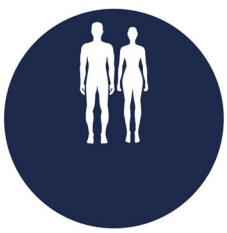
Partener principal: toți partenerii

Afișați mai multe



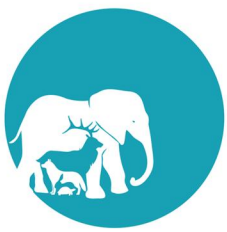
The *One Health* concept

Human health



The WHO defined health in 1946 as "a state of complete physical, mental and social well-being and not merely the absence of disease or infirmity", with the later addition of "the capacity to lead a socially and economically productive life".

Animal health



The OIE defines animal welfare in 2008: an animal is in good condition if it is healthy, enjoys comfort, is well fed, is safe, is able to display its innate (natural) behavior and does not suffer from unpleasant conditions such as pain, fear and stress.

Plant and environmental health

Environmental health refers to those aspects of human health that include the quality of life determined by physical, biological, socio-economic and psycho-social factors in the environment. The interrelationships of people with the environment concern medicine, when an ecological system is in a state of equilibrium, the health of the population prevails.

Globally, the *One Health* concept is a worldwide strategy to expand interdisciplinary collaborations and communications in all aspects related to the health care of humans, domestic animals or wildlife, which can no longer be approached separately, but only jointly.

One Health addresses not only human and animal disease concerns, but also issues related to lifestyle, diet, exercise, the impact of different types of human-animal relationships, and environmental exposures that can affect both populations. In order to achieve the expected effects, it is also necessary to educate the population to make them aware of the risk factors and benefits of prevention, as well as communication and understanding between patients and healthcare providers.