



ANDRIJA STAMPAR
TEACHING INSTITUTE
OF PUBLIC HEALTH

Creating a Healthier Future

REVIEW 2024

SCIENTIFIC ARTICLES IN CURRENT CONTENTS
AND OTHER INDEXED JOURNALS



REVIEW 2024

Scientific articles
in Current Contents
and other indexed journals

Andrija Stampar Teaching Institute of Public Health

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FOREWORD

The *2024 Review of Scientific articles in Current Contents and other indexed journals* is an annual report of scientific articles published in *Current Contents* journals and other indexed journals by the employees of the Andrija Stampar Teaching Institute of Public Health. The first chapter brings 16 abstracts of original scientific articles, reviews and other articles published in *Current Contents* journals (there were 11 such papers in 2023 and 20 papers in 2022). In the second chapter are 14 scientific articles published in other indexed journals (13 are cited in the *Web of Science Core Collection*).

There were 54 scientists in the Institute in 2024:

- 44 PhDs and 10 with master's degrees
- 44 female scientists and 10 male scientists.

In the name of the editorial board, I congratulate the employees and their external associates for any scientific contribution.

Director of the Institute

Prof Branko Kolarić, MD, PhD

Review 2023 in digital version is available for download from

<https://stampar.hr/hr/publikacije>



1. ORIGINAL SCIENTIFIC, REVIEW AND OTHER ARTICLES IN CURRENT CONTENTS

4.1. ABBREVIATED BREAST MRI AS A SUPPLEMENT TO MAMMOGRAPHY IN FAMILY RISK HISTORY OF BREAST CANCER WITHIN THE CROATIAN NATIONAL BREAST SCREENING PROGRAM

Biomedicines. 2024;12 (10),2357. DOI:10.3390/biomedicines12102357

Impact factor: 3.9

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Abstract

Objective: To evaluate the diagnostic performance of abbreviated breast MRI compared with mammography in women with a family history of breast cancer included in the Croatian National Breast Screening Program.

Methods: 178 women with a family history of breast cancer aged 50 to 69 underwent abbreviated breast MRI and mammography. Radiological findings for each method were categorized according to the BI-RADS classification. The gold standard for assessing the diagnostic accuracy of breast MRI and mammography, in terms of suspicious BI-RADS 4 and BI-RADS 5 findings, was the histopathological diagnosis. Performance measures, including cancer detection rates, specificity, sensitivity, and positive and negative predictive values, were calculated for both imaging methods.

Results: Twelve new cases of breast cancer were detected, with seven (58.3%) identified only by abbreviated breast MRI, four (33.3%) detected by both mammography and breast MRI, and one (8.3%) diagnosed only by mammography. Diagnostic accuracy parameters for abbreviated breast MRI were 91.67% sensitivity, 94.58% specificity, 55.0% positive predictive value (PPV), and 99.37% negative predictive value (NPV), while

for mammography, the corresponding values were 41.67%, 96.39%, 45.46%, and 95.81%, respectively.

Conclusions: Abbreviated breast MRI is a useful supplement to screening mammography in women with a family history of breast cancer. Considering the results of the conducted research, it is recommended to assess whether women with a family history of breast cancer have an increased risk and subsequently provide annual abbreviated breast MRI in addition to mammography for early detection of breast cancer.

Keywords: abbreviated breast MRI, mammography, breast cancer, breast cancer screening program

4.2. ADJUSTMENTS OF THE PHYTOCHEMICAL PROFILE OF BROCCOLI TO LOW AND HIGH GROWING TEMPERATURES: IMPLICATIONS FOR THE BIOACTIVITY OF ITS EXTRACTS

International Journal of Molecular Sciences. 2024;25(7):3677. DOI:10.3390/ijms25073677

Impact factor: 4.9

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Abstract

Climate change causes shifts in temperature patterns, and plants adapt their chemical content in order to survive. We compared the effect of low (LT) and high (HT) growing temperatures on the phytochemical content of broccoli (*Brassica oleracea* L. convar. *botrytis* (L.) Alef. var. *cymosa* Duch.) microgreens and the bioactivity of their extracts. Using different spectrophotometric, LC-MS/MS, GC-MS, and statistical methods, we found that LT increased the total phenolics and tannins in broccoli. The total glucosinolates were also increased by LT; however, they were decreased by HT. Soluble sugars, known osmoprotectants, were increased by both types of stress, considerably more by HT than LT, suggesting that HT causes a more intense osmotic imbalance. Both temperatures were detrimental for chlorophyll, with HT being more impactful than LT. HT increased hormone indole-3-acetic acid, implying an important role in broccoli's defense. Ferulic and sinapic acid showed a trade-off scheme: HT increased ferulic while LT increased sinapic acid. Both stresses decreased the potential of broccoli to act against H₂O₂ damage in mouse embryonal fibroblasts (MEF), human keratinocytes, and liver cancer cells. Among the tested cell types treated by H₂O₂, the most significant reduction in ROS (36.61%) was recorded in MEF cells treated with RT extracts. The potential of broccoli extracts to inhibit α -amylase increased following both temperature stresses;

however, the inhibition of pancreatic lipase was increased by LT only. From the perspective of nutritional value, and based on the obtained results, we conclude that LT conditions result in more nutritious broccoli microgreens than HT.

Keywords: auxins, *Brassicaceae*, climate change, metabolic response, microgreens, photosynthetic pigments, polyphenolics, ROS, temperature stress, vitamin C

4.3. BLOODSTREAM INFECTIONS IN PEDIATRIC ONCOLOGY PATIENTS: BACTERIAL PATHOGEN DISTRIBUTION AND ANTIMICROBIAL SUSCEPTIBILITY AT THE UNIVERSITY HOSPITAL CENTRE ZAGREB, CROATIA — A 5-YEAR ANALYSIS

Journal of Pediatric Hematology/Oncology. 2024;46(2):e156-e163.

DOI:10.1097/MPH.0000000000002809

Impact factor: 0.9

Mareković I^{1,2}, Vujnović T³, Radanović J⁴, Herljević Z¹, Kuliš T¹, Pavlović M¹, Mucavac L¹, Dejanović Bekić S¹, Bilić E²

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Abstract

The epidemiology of bacterial pathogens causing bloodstream infections (BSIs) in pediatric hematology/oncology patients is changing and resistance to antimicrobial agents is globally spread. We retrospectively assessed demographic, clinical, and microbiologic data of BSIs during a 5-year period at a pediatric hematology/oncology unit from January 1, 2017, to December 31, 2021, at the University Hospital Centre Zagreb, Zagreb, Croatia. In 66 pediatric patients with malignancies, 93 BSI episodes were registered and 97 bacterial isolates were cultured. The Gram-positive versus Gram-negative ratio was 67 (69.1%) versus 30 (30.9%). Coagulase-negative staphylococci (48; 49.6%) were the most frequent isolates, followed by Enterobacterales (17; 17.5%) and *Staphylococcus aureus* (6; 6.2%). Multidrug resistance isolates included extended spectrum β -lactamase producers (n = 3). Resistance rates to piperacillin/tazobactam, cefepime, and meropenem in Gram-negative isolates were 15.4%, 14.3%, and 0.0%, respectively. Gram-positive bacteria are the most common cause of BSI in our patients. Resistance rates to piperacillin/tazobactam and cefepime in Gram-negative isolates make meropenem a better choice for empirical antimicrobial treatment. As national and hospital data may differ, the surveillance of pathogen distribution and antimicrobial susceptibility in pediatric hematology/oncology wards is necessary to adjust empirical treatment accordingly.

Keywords: bloodstream infection, hematology/oncology, neutropenia, bacteria, antimicrobial susceptibility

4.4. DEVELOPMENT AND OPTIMIZATION METHOD FOR DETERMINATION OF THE STRAWBERRIES' AROMA PROFILE

Molecules. 2024;29(14):3441. DOI:10.3390/molecules29143441

Impact factor: 4.2

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Abstract

The strawberry (genus *Fragaria*) is a plant from the rose family (Rosaceae). As the fruits are likely to be picked mechanically, they are grown close to consumption centers. The aim of this work was to develop a suitable method for detecting as many molecules as possible in order to be able to distinguish between different strawberry cultivars and geographical origins in the future. Whole strawberries of the “Albion” cultivar, grown in the Jagodica Purgerica region of Zagreb, were used. Gas chromatography-mass spectrometry (GC-MS) in SCAN mode was used to analyze the aroma profile and to determine the proportion of individual components. The samples were prepared and analyzed using the solid-phase microextraction method (SPME). The impact of SPME fiber selection and GC column type was investigated, as well as sample weight, ionic strength, agitation temperature, and sampling time. A higher ionic strength was achieved by adding a 20% NaCl solution to the sample. The aroma profile of the studied strawberry cultivar consisted of furanone, esters, aldehydes, and carboxylic acids. Optimal results were achieved by adjusting the ionic strength during 15 min of extraction and incubation. The individual compounds were identified using NIST, Wiley libraries, and the “area normalization” method.

Keywords: strawberry, solid phase microextraction, gas chromatography, aroma components

4.5. DURATION OF STEAM DISTILLATION AFFECTS ESSENTIAL OIL FRACTIONS IN IMMORTELLE (*HELICHRYSUM ITALICUM*)

Horticulturae. 2024;10(2):183. DOI:10.3390/horticulturae10020183

Impact factor: 3.1

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Abstract

The composition of the essential oil depends on the duration and conditions under which the distillation of the plant material is carried out. In this study, one sample without fractionation and eight fractions (each after 15 min of steam distillation) of the essential oil of cultivated *H. italicum* were analysed by gas chromatography-mass spectrometry (GC-MS). The steam conditions for all samples were as follows: flow rate 800 L/h, temperature 104 °C, and pressure 0.4 bar. The test of the antimicrobial activity was performed with the modified Kirby–Bauer method (disc diffusion method) on non-selective nutrient media (blood agar) using the reference bacterial and fungal strains. A total of 75 different components were found in the essential oil samples obtained. A shorter distillation time makes the oil richer in monoterpenes and more suitable for the perfume and cosmetics industry. On the other hand, prolonged distillation leads to the essential oil being enriched with sesquiterpene oxides, which can have a negative effect on the fragrance of the essential oil. The essential oil of *H. italicum* showed antimicrobial activity only against *Staphylococcus aureus* ATCC 25923, and the best activity was shown by the sixth fraction.

Keywords: immortelle, essential oil, extract, fractional distillation, GC-MS, cosmetic industry

4.6. EPIDEMIOLOGY OF Q FEVER IN SOUTHEAST EUROPE FOR A 20-YEAR PERIOD (2002–2021)

Journal of Epidemiology and Global Health. 2024;14(3):1305-1318.

DOI:10.1007/s44197-024-00288-4

Impact factor: 3.8

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Abstract

This study aimed to assess epidemiological trends of Q fever in six countries of Southeast Europe by analysing surveillance data for 2002–2021 period. In this descriptive analysis, we collected and analysed data on confirmed human Q fever cases, obtained from the national Public Health Institutes of Bosnia and Herzegovina, Croatia, Greece, Montenegro, North Macedonia and Serbia. Overall, 2714 Q fever cases were registered during the 20-year period. The crude average annual notification rate was 0.82 (± 2.06) (95% CI: 0.47–1.16) per 100,000 inhabitants, ranged from 0.06 (± 0.04) (95% CI: 0.04–0.08) /100,000 in Greece to 2.78 (± 4.80) (95% CI: 0.53–5.02) /100,000 in the Republic of Srpska (entity of Bosnia and Herzegovina). Significant declining trends of Q fever age standardized rates were registered in Croatia, the Federation of Bosnia and Herzegovina, North Macedonia and Serbia, with an average annual change of -30.15%; -17.13%; -28.33% and -24.77%, respectively. An unequal spatial distribution was observed. The highest average age-specific notification rate was reported in the 20–59 age group (0.84 (± 0.40) (95% CI: 0.65–1.02) /100,000). Most cases (53.69%) were reported during the spring. Q fever remains a significant public health threat in this part of Europe. The findings of this study revealed the endemic maintenance of this disease in the including countries, with large regional and subnational disparities in notification rates. A downward trend was found in Q fever notification rates across the study countries with the average notification rate higher than in the EU/EEA, during the same period.

Keywords: *Coxiella burnetii*, epidemiology, Q fever, Southeast Europe, surveillance

4.7. EUROPEAN PAEDIATRICIANS NEED TO BE MORE AWARE OF THE RISKS FACED BY CHILDREN WHO SWALLOW BUTTON BATTERIES AND HOW TO TREAT THEM

Acta Paediatrica. 2024;113(12):2658–2663

Impact factor: 2.4

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Abstract

Aim: The aim of this study was to assess how aware European paediatricians were of the risks faced by children who swallowed button batteries and the advice for managing such cases.

Methods: An Internet-based survey was conducted among European paediatricians between 1 April 2023 and 31 May 2023. The researchers contacted members of the European Academy of Paediatrics Research in Ambulatory Setting Network and a number of European national paediatric societies.

Results: Responses were received from 605 paediatricians (66.0% females) in 41 countries and 64.5% had encountered cases of button battery ingestion. Only 31.2% had received specific training and only 35.7% were familiar with the advice issued by the European

Society for Paediatric Gastroenterology, Hepatology and Nutrition and the European Academy of Pediatrics. While 90.8% of respondents correctly recognised the need for hospitalisation of a child after button battery ingestion, only 30.1% of them identified all symptoms and 58.2% could accurately determine the appropriate diagnostic scope. One in ten underestimated the health or life risks. These deficiencies were significantly attributed to a lack of adequate training ($p < 0.01$). The overwhelming majority of the respondents expressed the need to increase awareness among physicians and childcare providers, mainly through social media campaigns and advertisements.

Conclusion: European paediatricians were insufficiently aware of button battery ingestion risks and management, with significant gaps in training and familiarity with advice. Respective education for medical practitioners and raising public awareness should be markedly strengthened.

Keywords: alkaline batteries, emergency, gastroscopy, oesophageal injury, paediatric training

4.8. EVALUATING ACADEMIC DETAILING AS AN ANTIBIOTIC STEWARDSHIP INTERVENTION IN PRIMARY HEALTHCARE SETTINGS IN CROATIA

BMC Primary Care. 2024;25(1):426. DOI:10.1186/s12875-024-02679-9

Impact factor: 2

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Abstract

Background. Acute respiratory tract infections are common in primary healthcare care settings and frequently result in antibiotic prescriptions, despite being primarily viral. There is scarcity of research examining impact of academic detailing (AD) intervention on prescribing practices for these infections in resource-constrained healthcare settings

like southeastern Europe. Therefore, aim of this study was to evaluate impact of AD intervention as an antimicrobial stewardship measure on antibiotic prescribing for acute respiratory tract infections in primary setting in Croatia which is located in southeastern Europe. Secondary goal included examining incidence of *Clostridioides difficile* infections (CDI) which are often associated with antibiotic consumption.

Methods. AD intervention was implemented from 1st to 30th April 2020 and led by hospital healthcare professionals (infectious disease physician, clinical microbiology physician and clinical pharmacist). They focused on enhancing prescribing behaviors of primary care physicians (PCPs) by presenting local data, supplemented by examples from everyday practice, research and guidelines highlighting negative consequences of imprudent antibiotic use. This feasibility quasi-experimental study had two control groups in two counties. Impact of AD intervention was assessed by analyzing antibiotic prescription patterns using log-linear model, adjusting for seasonality. Study focused on prescribed daily defined doses (DDD) per day among PCPs pre-intervention (from 1st January 2018 to 31st March 2020) and post-intervention (from 1st May 2020 to 31st December 2022).

Results. Data was collected from sixteen out of fifty-seven eligible PCPs with mean 29 years (SD 11.38) in practice. Statistically significant difference results ($p < 0.05$) favored AD intervention, leading to 30% decline in antibiotic prescribing in adjusted DDD per day for acute pharyngitis (21.14 post-intervention/30.27 pre-intervention), 33% decline for acute tonsillitis (24.91/37.38), 23% decline for acute upper respiratory infection (21.26/27.62) and 36% decline for acute bronchitis (8.13/12.77). Although there was 14% decline for acute sinusitis post-intervention, it did not reach statistical significance (30.96/35.93) ($p = 0.617$). Incidence of CDI cases decreased in investigated county while in control county stayed the same. Inter-county difference in these changes was not statistically significant (ratio=0.749, 95% CI, 0.460–1.220; $p = 0.246$).

Conclusions. This feasibility study showed reductions in antibiotic prescribing for acute respiratory tract infections, emphasizing the efficacy of targeted, educator-led programs. Tailored healthcare strategies are vital, especially in Croatia and southeastern Europe, for promoting sustainable practices and addressing antimicrobial resistance challenges.

Keywords: antimicrobial resistance, antibiotic prescribing, primary care, acute respiratory tract infections, antimicrobial stewardship, academic detailing, feasibility study

4.9. HEAVY METALS AND PESTICIDE RESIDUES IN SMALL FARM CHEESE PRODUCTION IN CROATIA — CHALLENGE BETWEEN QUALITY AND QUANTITY

Sustainability. 2024;16(4):1356. DOI:10.3390/su16041356

Impact factor: 3.3

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Abstract

The beneficial health effects of cheese as a source of bioactive compounds with antioxidant, antimicrobial, anti-inflammatory, immunomodulatory, and analgesic effects are well known. The aim of this study is to determine the presence of pesticide residues and heavy metals in 79 cheese samples from small farms in Croatia. The samples were analyzed over a period of three years for the quality parameters of fat, protein, dry matter, salt, and pH to determine whether metrological conditions affect the quality of cheese and to test the correlation between the variables in different types of cheese. A total of 509 pesticide residues were analyzed using liquid and gas chromatography with tandem mass spectrometry. Piperonyl butoxide was found in two samples. Inductively coupled plasma with mass spectrometry was used for a metal content analysis, and Cd, Cr, Mn, Ni, and Pb were found in the range of <0.005–0.012 mg kg⁻¹, <0.02–0.84 mg kg⁻¹, 0.031–1.128 mg kg⁻¹, <0.03–0.67 mg kg⁻¹, and <0.01–0.12 mg kg⁻¹, respectively. Cd was detected in just three samples. Mn was found in all analyzed samples. All tested samples complied with EU regulations and directives, and at the point of analysis, none posed a direct health risk for consumers. Sustainability on small farms could be ensured with the responsible use of pesticides and through a consistent and reliable supply of fresh, high-quality milk.

Keywords: cheese production, Croatia, health, heavy metals, pesticides, sustainability

4.10. INVASIVE PNEUMOCOCCAL DISEASE IN ADULTS AFTER THE INTRODUCTION OF PNEUMOCOCCAL VACCINATION: A RETROSPECTIVE STUDY IN THE METROPOLITAN AREA OF ZAGREB, CROATIA (2010–2022)

Frontiers in Public Health. 2024;12:1480348. DOI:10.3389/fpubh.2024.1480348

Impact factor: 3

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Abstract

Introduction: Invasive pneumococcal disease (IPD) is a severe form of illness caused by *Streptococcus pneumoniae* with high morbidity and mortality rate in the general population, particularly in children <5 years of age, adults ≥65 years of age and the immunocompromised. As known, pneumococcal vaccination lowers the risk of IPD so the aim of this study was to investigate whether the introduction of pneumococcal vaccination has influenced the incidence and mortality of IPD in adults in Croatia.

Materials and methods: A retrospective study was conducted among adult patients (aged ≥ 18 years) hospitalized due to IPD in the metropolitan area of Zagreb from 1st January 2010 to 31st December 2022. Number of vaccine doses distributed were obtained from the healthcare system.

Results: During the study period, 389 patients were hospitalized, of whom 214 (55.5%) were male. The annual incidence of IPD ranged from 0.6 to 4.1/100,000 population. A total of 185 (47.6%) patients were ≥ 65 years of age and 309 (79.4%) were ≥ 50 years of age. In 331 (85.1%) of the patients, at least one risk factor was identified, with age ≥ 65 years being the most common. Bacteremic pneumonia was the most frequent clinical presentation of IPD (66.3%). Indication for vaccination had 249 patients (64%) but only 11 patients (4.4%) were vaccinated. Also, 64 patients (16.5%) died. Serotype was determined in 233 (59.9%) of the isolates, with serotype 3 being the most frequent (49, 21%), followed by serotype 14 (38, 16.3%) and 19A (15, 6.4%). A total of 180 isolates (77.3%) were included in the 13-valent conjugate vaccine, 208 (89.3%) in the 20-valent conjugate vaccine and 212 (91%) in the 23-valent pneumococcal polysaccharide vaccine.

Discussion: The introduction of pneumococcal vaccination has led to a significant decrease in the incidence and mortality of IPD in adults. To further reduce morbidity and mortality from IPD, it is necessary to increase vaccine coverage in adults, particularly in individuals with risk factors. It may be beneficial to lower the recommended vaccination age from ≥ 65 to ≥ 50 years as the substantial difference in the incidence rates of IPD between these age groups was noticed.

Keywords: invasive pneumococcal disease, pneumonia, meningitis, primary bacteremia, incidence, mortality, adults, vaccination

4.11. OPTIMIZATION OF SAMPLE PREPARATION PROCEDURE FOR DETERMINATION OF FAT-SOLUBLE VITAMINS IN MILK AND INFANT FOOD BY HPLC TECHNIQUE

Processes. 2024;12(7):1530. DOI:10.3390/pr12071530

Impact factor: 2.8

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Abstract

Background: The analysis of vitamins in baby food is a challenging task given the complexity of the food matrix, vitamin stability, and strict regulations of the European Union regarding permissible deviations from declared values. Vitamins in food exist in different concentrations and forms and have different stabilities; thus, the preparation of samples for a reliable analysis using the same procedure is not straightforward. Therefore, significant attention has been devoted to optimizing sample preparation in the analysis of vitamins. **Methods:** This study aims to determine which of the sample preparation and extraction methods is the most efficient for the simultaneous determination of vitamins A, D, E, and K in milk and baby food using high-performance liquid chromatography (HPLC). Different samples of baby food were prepared in seven different ways based on four methods (saponification, enzymatic hydrolysis, solvent extraction, and solid-phase extraction). **Results and Conclusions:** According to the validation parameters, the optimal preparation method proved to be solid-phase extraction with a C18 stationary phase, with recoveries of 97.4%, 96.1%, 98.3%, and 96.2% for vitamins A, D, E, and K, respectively, and HPLC with a UV–Vis detector was identified as a sufficiently sensitive technique for the identification and quantification of fat-soluble vitamins in milk and baby food.

Keywords: vitaminized baby food, vitamin determination, solid-phase extraction, fat-soluble vitamins, reversed-phase HPLC

4.12. PACKAGING MATTERS: PRESERVATION OF ANTIOXIDANT COMPOUNDS OF FRESH STINGING NETTLE LEAVES (*URTICA DIOICA* L.)

Applied Sciences. 2024;14(15):6563. DOI:10.3390/app14156563

Impact factor: 2.5

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Abstract

Green leafy vegetables are very challenging in terms of storage and preservation, while packaging in controlled conditions with the selection of appropriate polymer material is crucial for maintaining their nutritional value and quality. Various packaging materials have different gas and water vapor permeability as well as physicochemical properties that can create a specific environment inside the package, therefore affecting the chemical composition, sensory characteristics, and overall quality of packed leafy vegetables. Stinging nettle is an edible plant with a high antioxidant content, making it a valuable leafy vegetable. Therefore, this study aimed to evaluate the influence of four packaging materials (biaxially oriented polypropylene (BOPP), low-density polyethylene (LDPE), polyamide/polyethylene (PA/PE), and polylactic acid (PLA)) on the antioxidant content of packed fresh nettle leaves during 14-day storage. Ascorbic acid content was the highest after 6 days of storage, equally well preserved in all tested films, with an average of 86.74 mg/100 g fm (fresh mass). After 14 days of storage, the total phenolic content was best preserved when packed in LDPE. The content of caffeoylmalic and chlorogenic acids was the highest in LDPE after 6 days. In addition, leaves packed in LDPE after 6 days of storage had the highest content of all photosynthetic pigments. According to FRAP analysis, the antioxidant capacity was best maintained in LDPE (at the 14th day, the measured capacity was 43.61 $\mu\text{mol TE/g}$). This study shows that the type of packaging material (BOPP, LDPE, PA/PE, and PLA) and storage duration (6 and 14 days) have a great impact on the level of antioxidant compounds in the nettle leaves, where LDPE and BOPP can be highlighted as the most favorable for the preservation of total and individual phenolic compounds, photosynthetic pigments, and antioxidant capacity.

Keywords: storage, packaging, polymer packaging materials, BOPP, LDPE, PA/PE, PLA, polyphenolic compounds, ascorbic acid, antioxidant capacity

4.13. PHYSICAL AND CHEMICAL PROPERTIES, HYGIENIC QUALITY AND FATTY ACID PROFILE IN MILK OF LACTATING LACAUNE DAIRY SHEEP

Archives Animal Breeding. 2024;67(1):37-49. DOI:10.5194/aab-67-37-2024

Impact factor: 1.6

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Abstract

In recent years, there has been globally increasing interest in dairy sheep breeding, including Lacaune sheep, which is supported by a high demand for sheep's milk on the market. This paper elaborates on the influence of a sheep's lactation stage on the physical and chemical properties, hygienic quality and content of fatty acids in milk produced by Lacaune sheep kept in intensive breeding. The research was conducted on 30 Lacaune sheep, which were tested in the early (60th day), middle (120th day) and late (180th day) stages of lactation. Density, freezing point and titration acidity were determined by applying the infrared spectrometry method, and indicators of the hygienic quality of milk, such as somatic cell count (SCC), were determined by the fluoro-opto-electronic method, and the total count of aerobic mesophilic bacteria (CFU) was determined by the flow cytometry method. The fatty acid profile of feed and milk was obtained by gas–liquid chromatography. Depending on the stage of lactation, results referring to the chemical composition of Lacaune sheep's milk showed a significant increase in the content of fat, protein, total dry matter and casein together with a significant decrease in the content of lactose in the late stage of lactation. There was also a significant increase confirmed for the concentration of urea and the freezing point in milk along with the lactation progress. Depending on the stage of lactation, milk yield in Lacaune sheep significantly decreased as lactation progressed. Analysis of the fatty acid composition in milk of Lacaune sheep proved a significant decrease in the

concentrations of C4:0, C6:0, C11:0, C12:0, C13:0, C15:0, C17:1, C18:2n6 and C18:3n6 as well as the n6 concentrations and the n6 / n3 ratio. The opposite trend was observed for concentrations of C10:0, C14:1, C16:0, C16:1, C18:0, C20:2, C18:3n3, C20:3n6, C20:5n3 and C22:6n3 as well as for the n3 concentrations. When compared to the early lactation stage, the C18:3n6 and n6 concentrations were significantly lower in the late lactation stage, while the C20:2 and C20:5n3 concentrations were significantly lower in the middle lactation stage when compared to the late lactation stage. There were many significant positive and negative correlations determined between the researched properties of milk. The research results obtained with Lacaune sheep's milk can be compared to the results of other studies, except for the lower content of milk fat. This confirms the good adaptability of Lacaune sheep to different breeding conditions and the necessity to provide sheep with quality pastures for grazing.

Keywords: breeding, dairy, fatty acids, milk, sheep

4.14. PREVALENCE OF CHRONIC HCV INFECTION IN EU/EEA COUNTRIES IN 2019 USING MULTIPARAMETER EVIDENCE SYNTHESIS

The Lancet Regional Health – Europe. 2024;36:100792. DOI:10.1016/j.lanepe.2023.100792

Impact factor: 13.6

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Abstract

Background. Epidemiological data are crucial to monitoring progress towards the 2030 Hepatitis C Virus (HCV) elimination targets. Our aim was to estimate the prevalence of chronic HCV infection (cHCV) in the European Union (EU)/European Economic Area (EEA) countries in 2019.

Methods. Multi-parameter evidence synthesis (MPES) was used to produce national estimates of cHCV defined as: $\pi = \pi_{\text{rec}}\rho_{\text{rec}} + \pi_{\text{ex}}\rho_{\text{ex}} + \pi_{\text{non}}\rho_{\text{non}}$; π_{rec} , π_{ex} , and π_{non} represent cHCV prevalence among recent people who inject drugs (PWID), ex-PWID, and non-PWID, respectively, while ρ_{rec} , ρ_{ex} , and ρ_{non} represent the proportions of these groups in the population. Information sources included the European Centre for Disease Prevention and Control (ECDC) national operational contact points (NCPs) and prevalence database, the European Monitoring Centre for Drugs and Drug Addiction databases, and the published literature.

Findings. The cHCV prevalence in 29 of 30 EU/EEA countries in 2019 was 0.50% [95% Credible Interval (CrI): 0.46%, 0.55%]. The highest cHCV prevalence was observed in the

eastern EU/EEA (0.88%; 95% CrI: 0.81%, 0.94%). At least 35.76% (95% CrI: 33.07%, 38.60%) of the overall cHCV prevalence in EU/EEA countries was associated with injecting drugs.

Interpretation. Using MPES and collaborating with ECDC NCPs, we estimated the prevalence of cHCV in the EU/EEA to be low. Some areas experience higher cHCV prevalence while a third of prevalent cHCV infections was attributed to PWID. Further efforts are needed to scale up prevention measures and the diagnosis and treatment of infected individuals, especially in the east of the EU/EEA and among PWID.

Keywords: HCV, Hepatitis C, chronic hepatitis, prevalence, elimination, Europe

4.15. TEMPORAL ACTIVITY AND DISTRIBUTION OF THE INVASIVE MOSQUITOES *Aedes albopictus* AND *Aedes japonicus* IN THE ZAGREB AREA, CROATIA

Tropical Medicine and Infectious Disease. 2024;9(11):263. DOI:10.3390/tropicalmed9110263

Impact factor: 2.8

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Abstract

Aedes albopictus and *Aedes japonicus* are invasive mosquito species that are causing great public concern. *Aedes albopictus* have successfully spread in Croatia, established in both the coastal and continental parts of the country, while *Aedes japonicus* is invading temperate climate areas. The invasive *Aedes* species are particularly attracted to the black plastic water containers and flower vases in cemeteries where they oviposit their eggs. Therefore, monitoring of this species was carried out in 12 cemeteries in Zagreb, using ovitraps with masonite strips as a substrate for oviposition. The monitoring was carried out from 2017 to 2020. The traps were inspected and the substrate was replaced every two weeks. This study showed that these two invasive species were present and very abundant in the cemeteries. In the case of *Ae. albopictus*, a higher population density and a greater number of occupied cemeteries were detected. This species was identified in all 12 cemeteries. *Aedes albopictus* was identified as the dominant species at all study sites. The spread of *Ae. japonicus* increased during 2018 in comparison to the previous year. Although this species was present in approximately 9% of the ovitraps, the observed population remained consistent throughout the course of the study. The findings indicate that cemeteries can be considered as significant public health hotspots, as the invasive *Aedes* mosquitoes tend to develop and reproduce in such environments. Consequently, the mosquito population of these two invasive species may only be reduced by applying integrated mosquito management measures, focused on the education of citizens.

Keywords: invasive species, *Aedes albopictus*, *Aedes japonicus*, mosquitoes in Croatia, vectors

4.16. UNDECLARED PHOSPHODIESTERASE TYPE 5 INHIBITORS (PDE5Is) IN FOOD SUPPLEMENTS ON THE CROATIAN MARKET ANALYZED BY LIQUID CHROMATOGRAPHY TIME-OF-FLIGHT MASS SPECTROMETRY (LC-QTOF-MS)

Microchemical Journal. 2024;203:110917. DOI:10.1016/j.microc.2024.110917

Impact factor: 4.9

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Abstract

This paper presents the development and validation of an analytical method using liquid chromatography-time of flight mass spectrometry (LC-QTOF-MS) suitable for screening and identifying suspect PDE5I analogs and quantifying four target PDE5Is in various food supplements. The method validation comprised of selectivity, linearity, accuracy, the limit of detection (LOD) and quantification (LOQ), and evaluation of matrix effects. The method showed good linearity with coefficients of determination higher than 0.992 for all analytes. LODs and LOQs ranged from 0.2 to 2.2 ng/mL and 2 to 20 ng/mL, respectively. The accuracy and precision of the method were deemed satisfactory, with values ranging from 88.5 to 115.9% for intra- and 86.9 to 111.2% for inter-day experiments, and RSDs ranged from 0.4 to 4.5% and 0.9 to 5.2%, respectively. Out of 49 analyzed food supplements collected on the Croatian market, 65% were not in compliance with EU regulations and pose possible risks to human health.

Keywords: food supplements, adulteration, PDE5 inhibitors, analogs, LC-QTOF-MS

2. ORIGINAL SCIENTIFIC AND REVIEW ARTICLES IN OTHER INDEXED JOURNALS

5.1. A DNA BARCODE REFERENCE LIBRARY OF CROATIAN MOSQUITOES (DIPTERA: CULICIDAE): IMPLICATIONS FOR IDENTIFICATION AND DELIMITATION OF SPECIES, WITH NOTES ON THE DISTRIBUTION OF POTENTIAL VECTOR SPECIES

Parasites & Vectors. 2024;17:216. DOI:10.1186/s13071-024-06291-9

Impact factor: 3

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Abstract

Background: Mosquitoes pose a risk to human health worldwide, and correct species identification and detection of cryptic species are the most important keys for surveillance and control of mosquito vectors. In addition to traditional identification based on morphology, DNA barcoding has recently been widely used as a complementary tool for reliable identification of mosquito species. The main objective of this study was to create a reference DNA barcode library for the Croatian mosquito fauna, which should contribute to more accurate and faster identification of species, including cryptic species, and recognition of relevant vector species.

Methods: Sampling was carried out in three biogeographical regions of Croatia over six years (2017–2022). The mosquitoes were morphologically identified; molecular identification was based on the standard barcoding region of the mitochondrial COI gene and the nuclear ITS2 region, the latter to identify species within the *Anopheles*

maculipennis complex. The BIN-RESL algorithm assigned the COI sequences to the corresponding BINs (Barcode Index Number clusters) in BOLD, i.e. to putative MOTUs (Molecular Operational Taxonomic Units). The bPTP and ASAP species delimitation methods were applied to the genus datasets in order to verify/confirm the assignment of specimens to specific MOTUs.

Results: A total of 405 mosquito specimens belonging to six genera and 30 morphospecies were collected and processed. Species delimitation methods assigned the samples to 31 (BIN-RESL), 30 (bPTP) and 28 (ASAP) MOTUs, with most delimited MOTUs matching the morphological identification. Some species of the genera *Culex*, *Aedes* and *Anopheles* were assigned to the same MOTUs, especially species that are difficult to distinguish morphologically and/or represent species complexes. In total, COI barcode sequences for 34 mosquito species and ITS2 sequences for three species of the genus *Anopheles* were added to the mosquito sequence database for Croatia, including one individual from the Intrudens Group, which represents a new record for the Croatian mosquito fauna.

Conclusion: We present the results of the first comprehensive study combining morphological and molecular identification of most mosquito species present in Croatia, including several invasive and vector species. With the exception of some closely related species, this study confirmed that DNA barcoding based on COI provides a reliable basis for the identification of mosquito species in Croatia.

Keywords: Culicidae, cytochrome c oxidase I (COI), internal transcribed spacer 2 (ITS2), species identification, species delimitation, cryptic species, species complex, invasive species

5.2. ANTIBIOTICS PRESCRIBING PATTERN AND QUALITY OF PRESCRIBING IN CROATIAN DENTAL PRACTICES — 5-YEAR NATIONAL STUDY

Antibiotics. 2024;13:345. DOI:10.3390/antibiotics13040345

Impact Factor: 4.3

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Abstract

Purpose: Antibiotic resistance is one of the biggest threats to global health today. The aim of this study was to analyze antibiotic prescribing patterns and quality of prescribing in Croatian dental practices over a 5-year period.

Methods: This is a retrospective observational study based on the analysis of the electronic prescriptions (medicines in ATC groups J01 and P01) from dental practices in Croatia prescribed from 1 January 2015 to 31 December 2019. Prescriptions were retrieved from the Croatian Health Insurance Fund (HZZO). The analyses included the number of prescriptions, type and quantity of prescribed drugs, indication, and the patient's and prescriber's characteristics.

Results: The consumption increased from 1.98 DID in 2015, to 2.10 DID in 2019. The most prescribed antibiotic was Amoxicillin with clavulanic acid followed by Amoxicillin, Clindamycin, Metronidazole and Cefalexin. The analyses showed that 29.79% of antibiotics were not prescribed in accordance with the contemporary guidelines for the proper use of antibiotics. Additionally, 22% of antibiotics were prescribed in inconclusive indications.

Conclusion: The research showed an increase in antibiotic consumption over five years along with unnecessary prescribing of antibiotics in cases with no indications for its use. The development of national guidelines for antibiotic use is necessary.

Keywords: prescribing pattern, antibiotics, quality of prescribing, dentists, electronic prescription

5.3. ASSOCIATION BETWEEN MEDITERRANEAN DIET AND ADVANCED GLYCATION END PRODUCTS IN UNIVERSITY STUDENTS: A CROSS-SECTIONAL STUDY

Nutrients. 2024;16(15):2483. DOI:10.3390/nu16152483

Impact factor: 4.8

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Abstract

The aim of this study was to evaluate the association between the Mediterranean diet (MD) and the accumulation of advanced glycation end products (AGEs) measured by skin autofluorescence. This cross-sectional study included 1016 healthy students from the University of Split, Croatia. Participants completed a self-administered questionnaire. Adherence to the MD was assessed using the Mediterranean Diet Serving Score (MDSS), and tissue AGEs accumulation was measured using the AGE Reader mu (DiagnOptics). Multivariate linear regression was used in the analysis. Students' age and female gender were associated with higher levels of AGEs, which was likewise found for greater coffee intake, adequate olive oil consumption, smoking, and lower levels of physical activity.

Higher consumption of vegetables and eating breakfast regularly were associated with lower AGEs levels. The overall MD adherence was not associated with AGEs, possibly due to very low overall compliance to the MD principles among students (8.3% in women and 3.8% in men). Health perception was positively associated with the MD and nonsmoking and negatively with the perceived stress level, while AGEs did not show significant association with self-rated students' health. These results indicate that various lifestyle habits are associated with AGEs accumulation even in young and generally healthy people. Hence, health promotion and preventive measures are necessary from an early age.

Keywords: Mediterranean diet, advanced glycation end products, coffee, physical activity, smoking, health, university students

5.4. BAT BITES AND RABIES PEP IN THE CROATIAN REFERENCE CENTRE FOR RABIES 1995–2020

Viruses. 2024;16:876. DOI:10.3390/v16060876

Impact Factor: 3.8

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Abstract

Seroprevalence of lyssaviruses in certain bat species has been proven in the Republic of Croatia, but there have been no confirmed positive bat brain isolates or human fatalities associated with bat injuries/bites. The study included a retrospective analysis of bat injuries/bites, post-exposure prophylaxis (PEP) and geographic distribution of bat injuries in persons examined at the Zagreb Antirabies Clinic, the Croatian Reference Centre for Rabies. In the period 1995–2020, we examined a total of 21,910 patients due to animal injuries, of which 71 cases were bat-related (0.32%). Of the above number of patients, 4574 received rabies PEP (20.87%). However, for bat injuries, the proportion of patients receiving PEP was significantly higher: 66 out of 71 patients (92.95%). Of these, 33 received only the rabies vaccine, while the other 33 patients received the vaccine with human rabies immunoglobulin (HRIG). In five cases, PEP was not administered, as there was no indication for treatment. Thirty-five of the injured patients were biologists or biology students (49.29%). The bat species was confirmed in only one of the exposure cases. This was a serotine bat (*Eptesicus serotinus*), a known carrier of *Lyssavirus hamburg*. The results showed that the bat bites were rather sporadic compared to other human injuries caused by animal bites. All bat injuries should be treated as if they were caused by a rabid animal, and according to WHO recommendations. People who come into contact with bats should be strongly advised to be vaccinated against rabies. Entering bat habitats should be done with caution and in accordance with current recommendations, and nationwide surveillance should be carried out by competent institutions and in close collaboration between bat experts, epidemiologists and rabies experts.

Keywords: bat bite, rabies virus, Zagreb, bat lyssaviruses, post-exposure prophylaxis (PEP), pre-exposure prophylaxis (PrEP), bat monitoring

5.5. COMPARISON OF CARBAPENEMASES AND EXTENDED-SPECTRUM β -LACTAMASES AND RESISTANCE PHENOTYPES IN HOSPITAL- AND COMMUNITY-ACQUIRED ISOLATES OF *KLEBSIELLA PNEUMONIAE* FROM CROATIA

Microorganisms. 2024;12(11):2224. DOI:10.3390/microorganisms12112224

Impact factor: 4.1

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Abstract

K. pneumoniae harbors various antibiotic resistance determinants like extended-spectrum and plasmid-mediated AmpC β -lactamases and carbapenemases. In the last three years, in the period of intense population aging, migrations and climate changes in Europe and Croatia as well, we observed changes in antibiotic resistance patterns of carbapenem-resistant *K. pneumoniae* (CRKP) isolates obtained routinely in community and inpatient setting. The aim was to compare and subsequently analyze CRKP hospital and community isolates resistance mechanisms, traits and molecular epidemiology, in order to analyze the dynamic of resistance trends, carbapenemase types and plasmid epidemiology. Disk diffusion and broth dilution method were the methods of choice to determine antibiotic susceptibility. β -lactamases were screened by phenotypic methods and confirmed with PCR. In total 113 isolates were analysed. Resistance to amoxicillin-clavulanate and ertapenem was confirmed in all strains. High resistance rates (over 90%) were observed for extended-spectrum cephalosporins, and ciprofloxacin. OXNV (OXA-

48, KPC, NDM, VIM) testing and PCR detected OXA-48 in 106, NDM in 7 and KPC in only one isolate. ESBLs accompanied carbapenemases in 103 isolates. IncL, associated with OXA-48, was the dominant plasmid type. No significant differences in the resistance profile and resistance determinants were found between hospital and community isolates plasmid type. The predominance of OXA-48 carbapenemase is in line with the reports from the neighbouring countries.

Keywords: *Klebsiella pneumoniae*, OXA-48, NDM, KPC, resistance

5.6. CONTINUED CIRCULATION OF MPOX: AN EPIDEMIOLOGICAL AND PHYLOGENETIC ASSESSMENT, EUROPEAN REGION, 2023 TO 2024

Eurosurveillance. 2024;29(27):2400330. DOI10.2807/1560-7917.ES.2024.29.27.2400330

Impact factor: 10

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Abstract

During the summer of 2023, the European Region experienced a limited resurgence of mpox cases following the substantial outbreak in 2022. This increase was characterised by asynchronous and bimodal increases, with countries experiencing peaks at different times. The demographic profile of cases during the resurgence was largely consistent with those reported previously. All available sequences from the European Region belonged to clade IIb. Sustained efforts are crucial to control and eventually eliminate mpox in the European Region.

Keywords: Europe, MPXV, mpox, WHO European Region, orthopoxvirus, outbreak

5.7. DETECTION OF MACROLIDE AND/OR FLUOROQUINOLONE RESISTANCE GENES IN *MYCOPLASMA GENITALIUM* STRAINS ISOLATED FROM MEN IN THE NORTHWEST REGION OF CROATIA IN 2018–2023

Genes. 2024;15(4):470. DOI:10.3390/genes15040470

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Abstract

Mycoplasma genitalium (*M. genitalium*) poses a significant public health challenge due to its association with non-gonococcal urethritis (particularly in men) and antimicrobial resistance. However, despite the prevalence of *M. genitalium* infections and the rise in resistance rates, routine testing and surveillance remain limited. This is the first study from Croatia that aimed to assess the prevalence and trends of resistance in *M. genitalium* strains isolated from male individuals by detecting macrolide and fluoroquinolone resistance genes. The study also aimed to explore the factors associated with resistance and changes in resistance patterns over time. Urine samples collected from male individuals in the Zagreb County and northwest region of Croatia between 2018 and 2023 were tested for *M. genitalium* with the use of molecular methods. Positive samples were subjected to DNA extraction and multiplex tandem polymerase chain reaction (MT-PCR) targeting genetic mutations associated with macrolide (*23S rRNA* gene) and fluoroquinolone (*parC* gene) resistance. Of the 8073 urine samples tested from 6480 male individuals (and following the exclusion of repeated specimens), we found that the prevalence of *M. genitalium* infection was 2.2%. Macrolide resistance was observed in 60.4% of strains, while fluoroquinolone resistance was found in 19.2%. Co-resistance to both antibiotics was present in 18.2% of cases. A statistically significant

increase in fluoroquinolone resistance was noted over the study period ($p = 0.010$), but this was not evident for azithromycin resistance ($p = 0.165$). There were no statistically significant differences in resistance patterns between age groups, whereas re-testing of patients revealed dynamic changes in resistance profiles over time. The high burden of macrolide resistance and increasing fluoroquinolone resistance underscore the urgent need for comprehensive resistance testing and surveillance programs. The implementation of resistance-guided treatment strategies, along with enhanced access to molecular diagnostics, is pivotal for effectively managing *M. genitalium* infections.

Keywords: *Mycoplasma genitalium*, antimicrobial resistance, molecular testing, resistance genes, macrolides, fluoroquinolones, polymerase chain reaction (PCR), men

5.8. RISING CONSUMPTION OF ANTICOAGULANTS IN CENTRAL AND EASTERN EUROPEAN COUNTRIES IN THE PERIOD 2007–2019

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Abstract

Introduction and Aim: Due to the growing incidence of thromboembolic disease and atrial fibrillation, increasing trends in anticoagulants consumption can be expected. The aim of this study was to analyse the consumption of anticoagulants in the Czech Republic, Croatia, Hungary, Romania, and Slovakia between 2007 and 2019.

Methods: A retrospective analysis of anticoagulants comprising B01AA, B01AB, B01AE, B01AF and B01AX was performed using ATC/DDD methodology. The analysis was initiated in the year before the launch of the first non-vitamin K oral anticoagulant (NOAC). The consumption of each drug was assessed based on annual data and was expressed as DDD per 1,000 population per day (DDD/TID).

Results: The overall rates of anticoagulant consumption increased in all countries. Specifically, doubled in the Czech Republic, Croatia, and Slovakia, more than tripled in Hungary and more than quadrupled in Romania. Parenteral anticoagulant consumption remained stable or decreased, while the proportion of oral anticoagulants increased

from an average of 61.41% in 2009 to 66.95% in 2019. The use of vitamin K antagonists declined, with the highest rate in the Czech Republic (11.16 DDD/TID in 2019). NOAC consumption showed substantial growth: from 0.002 to 8.33 DDD/TID in the Czech Republic, 0.001 to 6.73 in Croatia, 0.009 to 8.31 in Hungary, 0.0005 to 5.40 in Romania, and 0.03 to 10.77 in Slovakia. By 2019, rivaroxaban was the most commonly used NOAC in all countries, apart from Romania.

Conclusion: The study showed an overall increase in the anticoagulant consumption. However, specific characteristics of individual countries need to be further analysed to better understand the different factors influencing utilization patterns.

Keywords: drug utilization analysis, Europe, oral anticoagulation, parenteral anticoagulation

5.9. SCREENING FOR TORCH ANTIBODIES IN CROATIAN CHILDBEARING-AGED WOMEN, 2014–2023

Antibodies. 2024;13(2):49. DOI:10.3390/antib13020049

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Abstract

TORCH infections usually result in mild maternal morbidity, but may cause severe congenital abnormalities. Therefore, it is important to detect maternal infections, monitor the fetus after the disease has been recognized, and define the seronegative women who are at risk of primary infection during pregnancy. From 2014 to 2023, serum samples from 1032 childbearing-aged and pregnant women (16–45 years) were tested

for IgM/IgG antibodies to the most common TORCH pathogens: *Toxoplasma gondii*, rubella virus (RUBV), cytomegalovirus (CMV), and herpes simplex viruses (HSV-1 and HSV-2). The overall IgG seroprevalence rates were 20.1% for *T. gondii*, 91.3% for RUBV, 70.5% for CMV, 66.8% for HSV-1, and 3.5% for HSV-2. Only HSV-2 seroprevalence was age-related, with a significant progressive increase in seropositivity from 0% in those aged less than 26 years to 9.3% in those older than 40 years. The seroprevalence of *T. gondii* was higher in residents of suburban/rural areas than in residents of urban areas (27.4% vs. 17.1%). In addition, participants from continental regions were more often toxoplasma-seropositive than those from coastal regions (22.2% vs. 15.3%). HSV-1 seroprevalence was also higher in suburban/rural areas (71.7% vs. 64.7%). Obstetric history was not associated with TORCH seropositivity. Univariate and multivariate risk analysis showed that suburban/rural areas of residence and continental geographic regions were significant risk factors for *T. gondii* seroprevalence. Furthermore, suburban/rural area of residence was a significant risk factor for HSV-1 seroprevalence, while older age was a significant risk factor for HSV-2 seroprevalence. A declining trend in the seroprevalence of all TORCH pathogens was observed compared to previous Croatian studies (2005–2011). Similarly, the proportion of women simultaneously IgG-seropositive to two or three pathogens decreased over time. The maternal serology before pregnancy could potentially reduce the burden of congenital TORCH infections.

Keywords: TORCH, *Toxoplasma gondii*, rubella virus, cytomegalovirus, herpes simplex viruses, childbearing-aged women, Croatia

5.10. SEROEPIDEMIOLOGY OF HERPES SIMPLEX VIRUSES TYPE 1 AND 2 IN PREGNANT WOMEN IN CROATIA

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Abstract

Background and Objectives: Herpes simplex viruses (HSV-1 and HSV-2) are one of the most widespread causes of human viral infections. In Croatia, only two published studies have analyzed the seroprevalence of HSV infections in childbearing-aged and pregnant women (2005–2010), while more recent data are lacking. This study aimed to analyze the prevalence and risk factors for HSV-1 and HSV-2 infections among pregnant women in Croatia in the period from 2011 to 2021.

Materials and Methods: This study included 667 pregnant women aged 16–45 years submitted for HSV-1 and HSV-2 serology testing. Serum samples were initially screened for HSV-1 and HSV-2 IgM and IgG antibodies using a commercial ELISA test with a confirmation of HSV-2-positive samples using an immunoblot assay.

Results: The overall IgG seroprevalence rates were 69.9% for HSV-1 and 3.8% for HSV-2. A significant gradual increase in the HSV-2 seroprevalence with age was observed from 0.5% in participants under 30 years to 8.3% in participants above 40 years. The HSV-1 seroprevalence was stable up to 40 years (70.0 and 68.3%, respectively), with an increase to 86.1%, but this difference did not reach statistical significance. Area of residence (urban or suburban/rural), geographic region (continental or coastal), and obstetric history (normal pregnancy or unfavorable obstetric history) were not associated with HSV-1 and HSV-2 seroprevalence. Older age was found to be a significant risk factor for HSV-2 seropositivity in both univariate and multivariate risk analysis.

Conclusions: HSV-1 infection is widely prevalent among pregnant women with a stable trend over time. However, a declining trend in the HSV-2 seroprevalence was observed compared to 2005–2010. Serological screening in pregnant women is important in identifying seronegative women who are susceptible to HSV infection as well as seropositive women who are at risk for genital herpes recurrence during delivery.

Keywords: herpes simplex virus, pregnant women, seroprevalence, epidemiology, Croatia

5.11. THE RE-EMERGENCE OF NEUROINVASIVE FLAVIVIRUSES IN CROATIA DURING THE 2022 TRANSMISSION SEASON

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Abstract

(Re-)emerging arboviruses, such as tick-borne encephalitis virus (TBEV), West Nile virus (WNV), and Usutu virus (USUV), are continuously increasing in incidence. We analyzed the epidemiological characteristics of flavivirus infections in humans, sentinel animals, and mosquitoes detected in the 2022 transmission season in Croatia. From April to November 2022, 110 hospitalized patients with neuroinvasive diseases (NID) were

tested for the presence of arboviruses. RT-qPCR was used to detect TBEV, WNV, and USUV RNA. An ELISA and virus neutralization tests were used for the detection of flavivirus antibodies. TBEV infection was confirmed in 22 patients with NID. WNV NID was detected in six patients. TBE showed male predominance (81.8%; male-to-female ratio of 4.5:1). All but one WNV patients were males. TBE occurred from April to August, with the majority of patients (83.3%) being detected during the May–June–July period. WNV infections were recorded in August and September. In addition to human cases, asymptomatic WNV infections (IgM positive) were reported in 10 horses. For the first time in Croatia, WNV NID was observed in one horse that presented with neurological symptoms. Furthermore, USUV was confirmed in one dead blackbird that presented with neurological symptoms. A total of 1984 mosquitoes were collected in the City of Zagreb. Two *Ae. albopictus* pools tested positive for flavivirus RNA: one collected in July (USUV) and the other collected in August (WNV). A phylogenetic analysis of detected human and avian strains confirmed WNV lineage 2 and the USUV Europe 2 lineage. The presented results confirm the endemic presence of neuroinvasive flaviviruses in continental Croatia. The continuous monitoring of virus circulation in humans, sentinel animals, and mosquitoes is needed to reduce the disease burden.

Keywords: tick-borne encephalitis virus, West Nile virus, Usutu virus, epidemiology, Croatia

5.12. THE ROLE OF QUANTITATIVE PCR IN EVALUATING THE CLINICAL SIGNIFICANCE OF HUMAN BOCAVIRUS DETECTION IN CHILDREN

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Abstract

Human bocavirus (HBoV) has emerged as a significant pathogen primarily associated with respiratory infections in children. This study aimed to evaluate the clinical relevance of HBoV infection by quantifying viral loads in nasopharyngeal swabs from hospitalized children with acute respiratory infections (ARIs) and investigating correlations with clinical outcomes. A total of 957 children were tested, with HBoV detected in 73 cases (7.6%), either as a sole infection or co-infection with other respiratory viruses. Quantitative polymerase chain reaction (qPCR) was employed to measure viral load, and a threshold of 10^4 copies/mL was used to differentiate high and low viral concentrations. Results have shown that children with lower respiratory tract infections (LRTIs) had significantly higher viral loads, most notably in cases where HBoV was the sole pathogen. Additionally, children with pre-existing health conditions were more likely to have elevated HBoV concentrations compared to those who were previously healthy.

Children with higher viral loads were more likely to require oxygen supplementation and receive empirical antibiotic therapy, indicating a more severe clinical course. This study underscores the importance of considering HBoV viral load in clinical diagnostics, as higher concentrations were associated with more severe presentations, including the need for oxygen support. Future research should focus on refining diagnostic thresholds and exploring HBoV's role in co-infections to enhance patient care strategies.

Keywords: human bocavirus, viral load, acute respiratory infection, lower respiratory tract infection, pediatric infections, polymerase chain reaction (PCR), molecular diagnostics, co-detection

5.13. TRENDS IN NEW HIV DIAGNOSES AND FACTORS CONTRIBUTING TO LATE DIAGNOSIS AMONG MIGRANT POPULATIONS IN EU/EEA COUNTRIES, 2014 TO 2023

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Abstract

We analysed trends in new HIV diagnoses and factors contributing to late diagnosis among migrants in countries in the European Union (EU)/European Economic Area (EEA) from 2014 to 2023. Of the total reported HIV diagnoses, 45.9% were in migrants, with 13.3% born in EU/EEA countries and 86.7% in non-EU/EEA countries. Late diagnosis was observed in 52.4% of migrants, particularly among non-EU/EEA migrants with heterosexual transmission, regardless of sex. Improved HIV prevention and testing strategies are essential for at-risk migrant populations.

Keywords: delayed diagnosis, HIV infections, healthcare, epidemiology, migrants, population surveillance

5.14. WORRYING INCREASE IN THE RISK OF VERTICAL TRANSMISSION OF SYPHILIS IN CROATIA, 2020 TO 2024

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Abstract

Four infants potentially exposed to syphilis infection in utero, meeting World Health Organization surveillance criteria of congenital syphilis (CS), were diagnosed in Croatia between September 2020 and January 2024. We conducted a retrospective analysis of epidemiological, clinical and laboratory data of these cases to assess compliance with surveillance case definitions. As only one confirmed CS case has been reported in Croatia in over 2 decades, these reports signal an increased risk of syphilis vertical transmission and warrant strengthening antenatal screening.

Keywords: Croatia, congenital syphilis, epidemiological surveillance, pregnancy, vertical transmission

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