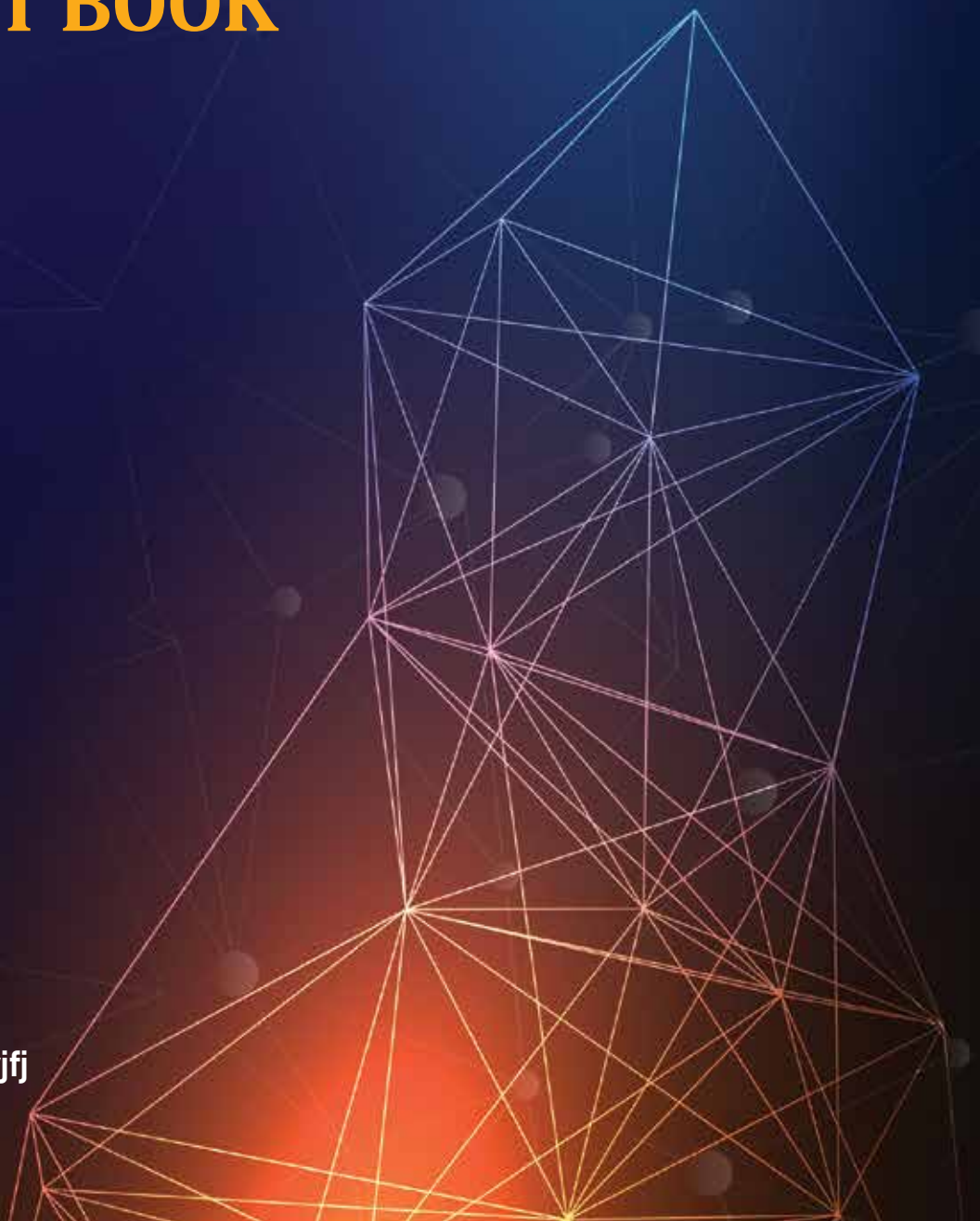
Public Health and Toxicology

16th INTERNATIONAL CONFERENCE

BIOMATERIALS AND NANOBIOMATERIALS:

Recent Advances in Safety-Toxicology and Ecology Issues

ABSTRACT BOOK



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Aims and Scope

Public Health and Toxicology is a double-blind peer-reviewed open access journal that welcomes in-vivo, in-vitro, in-silico and epidemiological research that focuses on the interaction between toxicology and health.
The journal is also interested in population-based research and the evaluation of real life scenarios and environmental risks throughout the lifespan, its impact on biological systems and the causes underlying the variability in susceptibility towards exposures.
The journal welcomes integrated epidemiological, clinical, animal and cellular biological research to provide the scientific foundation in support of hazard identification and risk assessment, resulting from exposure to chemical or biological agents (environmental toxicology) to protect public health and the environment through subsequent recommendations to abate or reduce any resulting health effects (regulatory toxicology).
This includes population-based research and the evaluation of environmental risk throughout the lifespan including pre- and postnatal development, childhood, adulthood and high-risk groups, its impact on biological systems and the causes underlying the variability in susceptibility of people to exposures.
The journal also welcomes research on public health and public health emergency preparedness related to environmental accidents, including natural and man-made disasters, that assess this exposure-outcome association from a public health and toxicology perspective.

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Editorial Board

Contents

Telomeres and aging-related diseases

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Telomere length is a well-established biomarker of biological aging. Telomeres progressively shorten with each somatic cell division, and this attrition is exacerbated by environmental stressors that induce oxidative stress and inflammation. Critically short telomeres activate the DNA damage response (DDR), leading to cellular senescence or apoptosis. Telomere characteristics -such as length and shortening rate- carry significant prognostic value for a range of age-related diseases, including cardiovascular disease, diabetes, neurodegenerative disorders, autoimmune diseases, renal conditions, solid tumors and hematological malignancies. Well-established evidence from experimental and clinical studies underscores the prognostic significance of telomere length in age-related disorders, with shorter telomeres consistently linked to higher incidence and poorer prognosis in these conditions. For instance, individuals with reduced leucocyte telomere length (LTL) demonstrate an elevated risk for metabolic disorders and cardiovascular complications, particularly in the context of type 2 diabetes. Mendelian randomization studies have highlighted the potential causal relationship between telomere length and autoimmune disease pathogenesis. In such disorders, telomere shortening correlates with autoimmune disease severity. In neurodegenerative diseases like Alzheimer's, longer telomeres may have a protective effect and could be associated with increased life expectancy. In oncology, the disruption of homeostatic mechanisms that regulate regular telomere shortening is considered one of the hallmarks of cancer. For solid tumors, short LTL has been associated with poorer outcomes in breast, prostate, colon, bladder, kidney, and skin cancer. As for hematologic malignancies, telomere length and telomerase activity have prognostic value in significant leukemia types, emphasizing the potential for telomerase-based immunotherapies. Lifeplus leverages cutting-edge telomere biology and molecular diagnostics to assess biological aging and cellular health based on individual telomere length measurements. Employing techniques such as metaphase quantitative fluorescence in situ hybridization (Q-FISH) and quantitative polymerase chain reaction (qPCR) for telomere length quantification, LifePlus provides personalized high-resolution insights into biological aging. Especially, the metaphase Q-FISH method enables chromosome-specific telomere length assessment, allowing for customized evaluation of aging rate and potential disease risk.

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Determination of macrolid antibiotics in water by fluorescence polarization method

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Introduction

The use of antibiotics in agriculture leads to their entry into food and the environment. The problem of water pollution attracts much attention, since antibiotics are not completely removed from wastewater at treatment facilities and end up in drinking water. In veterinary medicine, macrolide antibiotics are often used - tylosin (TYL) and erythromycin (ERY). Therefore, the determination of these antibiotics in the aquatic environment becomes an urgent problem. Immune methods of analysis are widely used in laboratories in various fields of medicine - enzyme immunoassay, FPIA and immunochromatographic analysis. The FPIA method allows for high specificity and sensitivity in determining low-molecular analytes in a homogeneous medium without separation, has the ability to automate the analysis system, high accuracy, label stability, as well as speed and ease of analysis.

Methods

The conjugates of TYL and ERY derivatives with BODIPY-FL-C5-OSu were obtained, purified and characterized. The FPIA methods were developed for determination of TYL and ERY. The FP signal was measured by portable fluorimeter Sentry-200.

Results

In this work, fluorescently labeled tracers were obtained for determining tylosin and erythromycin with the classic fluorescein label and with the new BODIPY label. Pairs of immunoreagents (tracers and antibodies) for determining these antibiotics were selected and characterized. The conditions for conducting FPIA were optimized, calibration curves were obtained and the analytical characteristics of the method were determined: detection limit, range of detectable concentrations, sensitivity and cross-reactivity. Using the developed FPIA methods for determining antibiotics most commonly used in veterinary medicine, water samples taken from reservoirs in Moscow and the Moscow region were tested.

Conclusion

Thus, in this work, methods for determining tylosin and erythromycin by the FPIA method with a detection limit of 5 and 1.6 ng/ml, respectively, were developed, the advantages of using the new fluorescent label BODIPY were demonstrated, the accuracy of the method was verified by the added-found test, and real water samples were tested.

Conflicts of interest

The authors declare that they have no conflict of interest in the publication of this article. The authors have no conflicts of interest to report in this work.

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Amiodarone, antiarrhythmic drug delivery via albumin nanoparticles

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Introduction

Heart rhythm disorders are major health issues that significantly increase the risk of cardiovascular complications. Atrial fibrillation plays a leading role, with a prevalence of 2–4%, and is expected to rise in the near future due to longer life expectancy and improved diagnostic methods. Amiodarone is a widely used antiarrhythmic drug. However, its application is limited by poor water solubility and severe side effects caused by its accumulation and off-target effects. Albumin-based nanoparticles are particularly attractive as

macromolecular carriers because they are biodegradable, well tolerated by patients, and possess a high drug-loading capacity and strong binding affinity for many drugs. The aim of this study was to obtain albumin nanoparticles loaded with amiodarone and to investigate their physicochemical characteristics and release profile in phosphate buffer.

Methods

Drug loading into albumin-based nanoparticles was performed via simple mixing in the presence of EtOH. Particles were purified by ultrafiltration and characterized. In vitro drug release studies were performed by dialysis at 37 °C in a phosphate buffer pH 7.4. The amiodarone content was determined spectrophotometrically at a wavelength of 260 nm (Spectramax and Nanodrop). The hydrodynamic size of the obtained particles was determined by Dynamic Light Scattering (DLS) and Nanoparticle Tracking Analysis (NTA) methods.

Results

In the present study, loaded and unloaded bovine serum albumin particles were obtained by the desolvatation method using aqueous solutions of amiodarone and albumin by adding 96% EtOH. The resulting complexes were then cross-linked with 3,3' dithiobis (sulfosuccinimidyl) propionate. The hydrodynamic diameter of unloaded albumin particles was approximately 212 ± 5 nm (PDI 0.140) before the use of the cross-linking agent and 249 ± 17 nm (PDI 0.244) after cross-linking. The size of the amiodarone-loaded particles was 219 ± 1 nm (PDI 0.240) and 238 ± 6 nm (PDI 0.249) before and after cross-linking, respectively. The encapsulation efficiency, defined as the ratio of loaded amiodarone to the initial amount, was 36.8%. The loading capacity, calculated as the ratio of the loaded drug amount to the residual protein amount, was 30.6%. The in vitro release profile indicated that 55.8% of the drug was released within 48 hours, establishing developed albumin nanoparticles as the promising extended release platform.

Conclusions

Thus, amiodarone-containing protein complexes based on albumin were obtained and characterized. The results demonstrate that these complexes can prolong the release of amiodarone, making them a promising platform for antiarrhythmic drug delivery.

Conflicts of interest

The authors declare no conflicts of interest in the publication of this article.

Funding

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Public Health Toxicol 2026;6(Supplement 1):A3

Functionalization of magnetic nanoparticles with low-molecular-weight ligands and study of their interaction with eukaryotic cell lines in the presence of a low-frequency magnetic field

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Introduction

For many years, one of the central challenges in nanomedicine has been the design of delivery systems capable of selectively releasing therapeutic agents into target cells. Recently, increasing attention has been directed toward nanostructures that can also function as efficient diagnostic platforms. Magnetite-based magnetic nanoparticles (MNPs) are of particular interest due to their high biocompatibility, surface modification versatility, and controlled navigation under external magnetic fields, all making them promising candidates for the development of theranostic agents. In contrast to conventional biopolymeric and inorganic matrix-based delivery systems, MNPs are able to exhibit magnetomechanical effects when exposed to alternating magnetic fields, which enables their consideration not only as drug carriers but also as autonomous therapeutic agents. The aim of this study was to investigate the effects of magnetic nanoparticles coated with hydrophilic low-molecular-weight ligands on eukaryotic cell lines in the presence of a low-frequency magnetic field.

Methods

In this work, reproducible synthesis of MNPs was achieved by thermal decomposition of iron pentacarbonyl. TEM analysis revealed an average particle diameter, while XRD confirmed the magnetite structure. Magnetic properties of synthesized MNPs were investigated by VSM. Hydrophobic nanoparticles were further functionalized with hydrophilic ligands such as dopamine (DOPA), folic acid (B9) and 3,4-dihydroxyphenyl acetic acid (DOPAC). Size of nanoparticles transferred into aqueous medium were characterized by DLS. The presence of ligands on the nanoparticle surface was confirmed by IR spectroscopy. Cytotoxicity and the intracellular iron content in cell lysates were also evaluated both with and without exposure to a low-frequency alternating magnetic field.

Results

Spherical magnetite MNP with an average diameter of 8.2 ± 1.6 nm were synthesized. The crystal lattice parameters (8.373 ± 0.002 Å) and the diffraction pattern obtained by XRD corresponded to a partially oxidized magnetite structure. The CSR calculated from the diffraction pattern was 9.2 ± 0.1 nm, which coincides with the TEM results. Analysis of magnetostatic properties showed the production of superparamagnetic MNP with partial preservation of interactions between crystallites. Hydrophilized MNP with DOPA, DOPAC and B9 ligands were obtained. The average hydrodynamic size of the MNP-ligand determined by the DLS method was about 100 to 400 nm depending on ligands. A series of experiments were carried out on eukaryotic cell lines to study the behavior of MNPs in the presence/absence of low-frequency magnetic field (LFMF). In the absence of LFMF, MNP-DOPA-B9 demonstrated lower cytotoxicity towards the MCF-7 cell line compared to that towards the HEK293 (up to 50%) line as was determined spectrophotometrically (Spectramax). After incubation of MNP-ligand with MCF-7 in low-frequency PVMP, a temporary increase in viability was observed with MNP-DOPAC and MNP-DOPA-B9, exhibiting distinctive biological response to magnetic field effect.

Conclusions

Thus, hydrophilized MNPs were obtained and characterized. It was demonstrated that magnetic nanoparticles are capable of enhancing cell viability following exposure to a low-frequency magnetic field. The obtained result highlights the potential of MNPs for applications in regenerative medicine.

Conflicts of interest

The authors declare no conflicts of interest in the publication of this article.

Funding

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Production of new carbon-magnetic particles and their immunoanalytical application

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Introduction

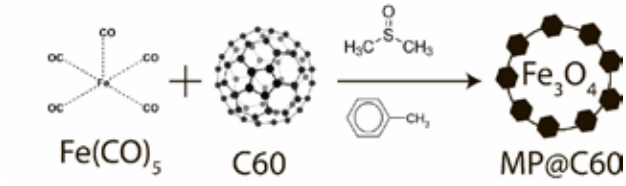
Conjugates of magnetic particles (MPs) with antibodies are effective components of analytical systems, providing selective binding of controlled compounds from large sample volumes, simple and rapid concentration upon application of an external magnetic field. The use of native metal MPs for conjugation is complicated by their tendency to aggregation and relatively low optical density. In this study, we studied the possibilities of modifying MPs with fullerene C60, which isolates the MPs surface from oxidation and provides an intense black color of the product.

Methods

The preparation of unmodified MPs was obtained by thermal decomposition of a 2% solution of iron pentacarbonyl in dimethyl sulfoxide (DMSO). The reaction was implemented at 130 °C with the formation of iron oxide and carbon dioxide. Fullerene-coated MPs were obtained by boiling together at 130 °C a 2% solution of iron pentacarbonyl and a 10% solution of C60 in a mixture (50/50 v/v) of DMSO and toluene. The solution was colored first yellow, and then black-violet. The MPs coated with C60 were precipitated using a magnet and washed first with DMSO, then with water. Based on the obtained MPs and MP@C60, conjugates of the particles with anti-species antibodies were synthesized by adsorption for testing in the format of immunochromatographic analysis.

Results

The synthesis of modified MPs was carried out according to the following scheme: The hydrodynamic diameters of MPs and MP@C60 were 190±52 nm and 733±190 nm, respectively. The efficiency of magnetic separation of the synthesis products from their colloidal solutions was confirmed. The obtained preparations of conjugates of MPs and MP@C60 with anti-species antibodies were characterized in the immunochromatographic system. It was shown that the intensity of staining of the binding zone when using MP@C60 is 5 times higher compared to using MP, which confirms the advantages of MP@C60 as analytical markers.



Conclusions

An effective method for obtaining contrast-colored MP@C60, which combines magnetic and colorimetric properties of the original components, is proposed. These preparations are promising components of immunoanalytical systems.

Conflicts of interest

The authors declare no conflicts of interest.

Funding

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Ensuring safe irrigation in Crete: Monitoring water quality and addressing risks of wastewater reuse

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Crete faces increasing water scarcity driven by climate change, tourism, and intensive agriculture, leading to a growing reliance on alternative water sources - including treated wastewater - for irrigation. While this approach supports water conservation, it raises critical toxicological and environmental concerns. Our laboratory plays a key role in the routine monitoring and quality control of both conventional and reclaimed water used for agricultural irrigation across the island. This study presents findings from multi-year surveillance programs evaluating the chemical, microbiological, and emerging contaminant profiles of irrigation water sources in Crete. Particular attention is given to the detection of nutrients, heavy metals, and microbial indicators in treated effluents considered for reuse. Data trends reveal seasonal and regional variability, and in some cases, exceedances of regulatory thresholds, underscoring the need for stricter controls and the application of advanced treatment processes. We highlight the importance of robust, ISO-accredited laboratory practices in supporting safe wastewater reuse and preventing long-term soil contamination, crop uptake of toxicants, and potential health risks. Our work also emphasizes the need for greater integration of private laboratories in regional water management strategies, farmer education, and real-time risk assessment.

Conflicts of interest

The authors declare no conflicts of interest.

Funding

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Determination of dibutyl phthalate in natural waters and plastic extracts by fluorescence polarization method

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Introduction

Dibutyl phthalate (DBP) is widely used as a plasticizer in the production of polymeric materials to impart flexibility, strength, and extensibility. Due to its negative impact on human health, DBP content should be monitored in food and environmental objects. The aim of the work was to develop a method for polarization fluorescence immunoassay (FPIA) and to test the method using the example of analyzing natural waters of Lake Onega and its tributaries, as well as extracts from plastic.

Methods

New immunoreagents for FPIA were synthesized - a conjugate of a succinic derivative of DBP with a fluorescent label 5-aminomethylfluorescein (AMF) and dichlorotriazinylaminofluorescein (DTAF). The DBP-AMP and

DBP-DTAF was purified by thin-layer chromatography using methanol: chloroform in a ratio of 1:4 (v/v) as an eluent. The FPIA methods were developed for determination of DBP. The FP signal was measured by portable fluorimeter Sentry-200.

Results

We have developed an FPIA method that allows rapid and specific quantitative analysis of DBP in water. In this work, we used immunoreagents to determine DBP: monoclonal antibodies against DBP and a conjugate of the succinic derivative of DBP with AMF and DTAF. A calibration dependence of DBP detection in water was obtained. The volume of water sample in the analysis was 0.1 ml, while the sample dilution factor was 10 times, which allows reducing sample preparation by only filtering water from macroparticles. The detection limit of this method was 20.0 ng / ml, which is significantly lower than the maximum permissible concentration of DBP in water established in Russia. The obtained calibration dependence does not require repetition for each sample analysis, but can be used for a selected pair of immunoreagents.

Conclusion

The advantages of this homogeneous FPIA method are the almost complete absence of sample preparation, simple analysis, which comes down to mixing reagents and measuring the signal, and a low detection limit of 20 ng/ml. The FPIA method can be used as a cheaper and more express alternative to instrumental methods for analyzing natural waters.

Conflicts of interest

The authors declare that they have no conflicts of interest in the publication of this article. The authors have no conflicts of interest to report in this work. The abstract was not submitted elsewhere and published here firstly.

Funding

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Soil biodegradation of poly(3-hydroxybutyrate) films under different climatic conditions

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Introduction

The annual increase in polymer production leads to the accumulation of waste from synthetic polymers, especially microplastics, which are hazardous to the environment. The transition to fully degradable polymer materials, including polyhydroxyalkanoates (PHA), will help solve this problem. Poly(3-hydroxybutyrate) is a polymer from the PHA group, which is included in the global cycle and is transformed by microbial enzymes into carbon dioxide and water. In the environment, the degradation of PHA products is highly variable and depends on climate, hydrothermal conditions, and microbial activity, which poses the challenge of studying the rates of PHA biodegradation in different geographic regions.

Methods

Thin films of the biopolymer poly(3-hydroxybutyrate) were placed in two experimental sites with significantly different soil and climatic conditions: 1) India (Kottayam, 9°39′ N, 76°32′ E), tropical climate, red ferallite soils; 2) Siberia (Krasnoyarsk, 56°08′ N, 92°52′ E), sharply continental climate, chernozem soils. After exposure, the mass loss of the films and the physicochemical properties of the polymer samples were studied. Strains of P(3HB) degrading microorganisms were isolated and identified.

Results

During the biodegradation process, a decrease in molecular weight and an increase in the degree of crystallinity of P(3HB) samples were detected in the soils of both geographic regions. In the tropical soil, the mass loss of films occurred 2 times slower than in chernozem, which is due to a decrease in the humidity and microbial activity of red soil. The community of microorganisms – primary degraders of P(3HB), isolated from red soil, differed from the set of degrader species isolated from chernozem.

Conclusion

The metabolic potential of microorganisms involved in the biodegradation of fully degradable polymers such as PHA may be limited by climatic parameters, requiring the creation of favorable conditions for the disposal of biopolymer waste.

Conflicts of interest

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Application of the method of fluorescence polarization analysis for lysozyme assay in tablet dosage form

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Introduction

Lysozyme, a natural enzyme with muramidase and chitinase activity, is used medicinally for its antimicrobial properties. While pharmacopoeial methods rely on turbidimetric Micrococcus lysodeikticus assays—which are labor-intensive, time-consuming, and require unstable biological reagents—this study developed a fluorescence polarization assay (FPA) based on lysozyme's interaction with a fluorescently-labeled peptidoglycan trisaccharide. The aim was to validate this FPA for rapid, reliable quantification of lysozyme in tablets.

Methods

Egg white lysozyme (≥15,000 U/mg; Sisco Research Laboratories) and a fluorescently-labeled chitin oligosaccharide tracer (Mukhametova et al.) were used. Analyses employed a Sentry 200 fluorescence polarization analyzer (Ellie LLC). Commercially available tablets (20 mg lysozyme + 10 mg pyridoxine; six manufacturers) were analyzed. Validation assessed analytical range, specificity, accuracy, and precision.

Results
Lysozyme quantification in tablets employed parallel analysis of 200.0, 400.0, and 600.0 mg samples with mean calculation, essential for enzymatic activity-based methods. This approach evaluated matrix effects, verified dilution-independent activity consistency, and optimized working dilution to minimize interferences. Parallel testing demonstrated convergent results across all dilutions, enabling reliable analysis across the entire validated mass range. The validated method demonstrated analytical range 150-650 µg/mL and specificity with no significant cross-reactivity to excipients. The accuracy of the method was confirmed via a standard addition approach by spiking 10 mg and 20 mg of lysozyme reference standard into homogenized tablet powder, demonstrating satisfactory recovery rates of 100.2–106.0%. Precision, assessed via six replicates, demonstrated an RSD ≤ 13.7%.
Conclusion
A validated method for lysozyme quantification in tablets was developed for quality control, offering rapid analysis, reduced labor, and elimination of live microbiological reagents.
Conflicts of interest
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New tracers for fluorescence polarization immunoassay of herbicide 4-chlorophenoxyacetic acid

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Introduction
Chlorophenoxy herbicides are a widely used class of agricultural compounds employed for weed control and as plant growth regulators. Among these, 4-chlorophenoxyacetic acid (4-D) is commonly utilized as both a herbicide and a growth regulator. However, herbicides are often applied at higher concentrations than necessary, increasing the risk of contamination in food products, which may pose hazards to human health and the environment. Therefore, developing a method to detect residual traces of 4-D is highly relevant. Fluorescence polarization immunoassay (FPIA) presents a promising solution for this purpose. The aim of this study was to synthesize tracers for 4-D detection and optimize FPIA conditions.
Methods
Two tracers of 4-chlorophenoxyacetic acid were synthesized using the carbodiimide method, conjugating the compound with fluorescein derivatives: ethylene-diamine-fluorescein thiocarbamate (EDF) and 5-aminohexyl-amino-carbonyl-fluorescein (5AHCF). The tracers' structures were confirmed by mass spectrometry. Monoclonal antibodies (M1 and M2) against 4-D were sourced from China.
Results
Both tracers yielded comparable results. However, further work proceeded with the 4D-EDF tracer and M1 monoclonal antibody. The FPIA for 4-D was optimized by determining the ideal concentrations of the tracer and M1 antibody, achieving a bound fraction of 0.5. The optimal concentrations were 2.5 nM for the tracer and 4 nM for the antibody. The assay demonstrated a detection limit of 1.2 ng/mL in aqueous solutions, with an analysis time of 15 minutes. Accuracy was confirmed via a spike-and-recovery test, yielding recovery rates between 95% and 115%. Parallel ELISA validation showed a relative standard deviation of 3%, confirming the reliability of 4-D detection in real water samples.
Conclusion
The 4D-EDF tracer is suitable for detecting 4-D in water. FPIA offers several advantages, including rapid analysis, high sensitivity, and minimal sample preparation, making it a practical tool for environmental monitoring.
Conflicts of interest
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Film packaging from degradable bioplastic synthesized on waste fish oil (wfo)

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Introduction
The global production of containers and packaging is the largest area of processing synthetic plastics (about 40%), the production volumes of which exceeded 400 million tons per year. The service life of many types of packaging is calculated on several days. Used packaging and its waste replenish the content of landfills, pollute natural ecosystems and the waters of the world ocean. The accumulation of plastic waste, primarily due to giant waste of plastic packaging, has become a global environmental problem. A relevant and highly sought-after direction is the transition to degradable packaging and the search for affordable polymeric materials for this. The aim of this study was to obtain and characterize degradable film packaging for food products.
Methods
Single-layer films were obtained by flat-slot extrusion of polymer melt using a Brabender laboratory extruder. Comprehensive studies of surface properties (SEM, AFM, measurement of contact angles), mechanical strength (Young's modulus, elongation at break), sanitary and chemical studies, resistance to model liquids and typical microflora of meat and dairy products were carried out taking into account current international standards.
Results
The technology of extrusion production of smooth films from poly-3-hydroxybutyrate melts synthesized using WFO as a carbon substrate has been developed and implemented. The physical and mechanical properties of the polymer during melting and extrusion, the structure and characteristics of the film surface, moisture and vapor permeability, resistance to the effects of model liquid media simulating food products, and antibacterial activity against the test microflora of meat and dairy products (Bacillus subtilis, Micrococcus luteus, Pseudomonas fluorescens) in forced contamination tests have been studied.

Conclusion

Comprehensive studies have shown that the obtained samples of degradable film products meet the indicators of food packaging, according to the requirements of current international ISO standards.

Conflicts of interest

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Legionella surveillance and risk assessment in high-risk environments: A laboratory-based preventive approach

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Legionella spp. are opportunistic waterborne pathogens commonly found in artificial water systems, posing a significant risk of Legionnaires' disease, particularly in healthcare facilities, hotels, and other public infrastructures. High-risk environments with complex plumbing systems, such as hospitals and resorts, require proactive strategies to monitor and control Legionella proliferation. This study presents the integrated approach used by our laboratory to assess and manage the risk of Legionella contamination in various environments, with a focus on minimizing public health hazards. Environmental sampling was conducted across multiple high-risk sites including hospitals, hotels, and wellness centers. Water samples were analyzed using accredited culture-based techniques (ISO 11731:2017) for Legionella spp. detection and quantification. Risk assessments were conducted following national and international guidelines (e.g., NPHO, WHO, ECDC), incorporating site-specific factors such as water temperature, system design, and historical contamination data. Customized mitigation strategies were developed, including thermal disinfection, biocide treatment, and infrastructure improvements. Our surveillance identified Legionella presence in over 30% of sampled sites, with higher prevalence in older and poorly maintained systems. The implementation of targeted risk management plans led to a significant reduction in Legionella counts and an improvement in water system safety profiles over a 6–12 month follow-up period. A laboratory-driven, risk-based assessment model is essential for effective Legionella control in high-risk environments. Regular monitoring, continuous training of involved personnel, data-driven interventions, and close collaboration with facility management are key components in reducing infection risk and ensuring compliance with public health standards.

Conflicts of interest

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Development of immunoassay for linezolid monitoring in critical patients

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Linezolid (LNZ) is a vital oxazolidinone antibiotic widely used for treating severe Gram-positive infections, including methicillin-resistant Staphylococcus aureus (MRSA) and vancomycin-resistant Enterococcus (VRE), as well as multidrug-resistant tuberculosis. Despite its clinical importance, the therapeutic efficacy of LNZ is challenged by its narrow therapeutic window and significant interpatient pharmacokinetic variability, especially in critically ill patients, making therapeutic drug monitoring (TDM) essential to optimize dosing and minimize toxicity. Currently, available analytical methods are primarily instrumental, such as LC-MS/MS, which, while accurate, are costly and require specialized infrastructure for routine clinical use. In response to this clinical need, an immunoassay designed to specifically and cost-effectively monitor serum LNZ concentrations was developed[1]. This work represents the first report on the design and synthesis of hapten conjugates that serve as immunogens capable of inducing high-affinity specific antibodies to LNZ. To realize this objective, a series of conjugated antigens based on four novel LNZ derivatives (H1–H4) were synthesized which provided aliphatic and heterocyclic spacer arms of different lengths (0–6.2–9.4–11.8 Å) between carrier proteins and LNZ pharmacophore. Tetanus toxoid and bovine serum albumin conjugated to H1-H3 were immunogens for rabbits and simultaneously served as heterologous H1/H4-coating antigens in direct competitive ELISAs. Three different types of antibodies to haptens H1–H3 were generated, and the corresponding heterologous ELISAs demonstrated equally high affinity and specificity to LNZ regardless of the hapten spacer length. The established sensitivities of the ELISAs (IC_{50} range: 11.5–28.3 ng/mL), low detection limits (0.8–1.6 ng/mL) and wide dynamic ranges (1.4–260 ng/mL) allowed quantitative determination of therapeutic levels of LNZ in serum and plasma. The detailed investigation into sample pretreatment methods showed that deproteinization with trichloroacetic acid was highly effective, providing recovery rates from 82.1% to 99.6%, ensuring measurement accuracy in complex biological matrices. The real serum samples (n = 36) from surgical patients with associated MRSA/VRE infection treated with LNZ were tested in parallel using ELISAs based on different reagents (antibodies and antigens). A high degree of concordance between the obtained results confirmed the reliability of the measurements and the developed assay systems suitable for TDM purposes. Thus, the study addresses the real clinical need for a reliable, sufficiently rapid (1.5 h) and sensitive tool for linezolid TDM, especially in critically ill patients with variable pharmacokinetics, which could help improve dosing accuracy and efficacy of antibiotic therapy.

Conflicts of interest

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Choosing parameters of nanolabels and their conjugates with antibodies for efficient detection of antibiotics in food products

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Conjugates of nanoparticles and antibodies are efficient bioanalytical reagents that facilitate the formation and separation of analyte-containing complexes, as well as the generation of detectable signals. In this regard, it is important that the conjugates maintain the functionality of their components and enable the detection of analytes at low concentrations. This report focuses on immunochromatographic test systems (ICTSs), which are widely used for rapid on-site testing. The study considers the parameters of nanolabel-antibody conjugates and the conditions of assays implementation providing the desired detection limits and working ranges of ICTSs in accordance with regulatory requirements for controlled analytes. The target antigens in the ICTSs investigation are veterinary drugs of different classes. Different approaches for varying ICTSs sensitivity are explored, including using new labels, label complexes, and additional chemical reactions, are characterized. Features of spherical and branched gold nanoparticles, magnetite particles, and semiconductor quantum dots as carriers for antibody immobilization are considered. The specific requirements for nanocarriers/nanolabels in ICTSs are outlined. The sensitivity changes of competitive immunoassays with varying sizes of nanoparticles or degrees of their coating by antibodies are discussed. Additionally, alternative ways to modulate the detection limit, such as pre-incubation of reagents, transferring heterogeneous reactions into moving liquids, displacing binding zones, adjusting flow velocity and geometry, and altering the sequence of reagents introduction to reaction mixtures and test strips. Finally, the developed ICTSs for detecting antibiotics of different classes, based on the use of the selected nanoparticles-antibodies conjugates, are presented. Their efficiency for control of raw and processed meat in combination with rapid samples preparation was demonstrated.

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Study on the toxicity of hemostatic materials based on sodium alginate

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Introduction

One of the most urgent issues in disaster medicine is providing first aid to stop bleeding. Finding new materials for hemostatic agents should focus not only on their effectiveness in stopping bleeding, but also on their toxicological safety. This paper presents a study on the toxicity for new hemostatic materials: sodium alginate, with and without aminocaproic acid added.

Methods

The study of hemostatic materials involved the stages of sanitary and chemical testing, toxicological testing, and determination of hemostatic activity. Sanitary and chemical tests include the determination of pH changes, UV absorption, metal content, formaldehyde and volatile compounds in aqueous extracts.

Results

All samples comply with the specified standards: there are no toxic metals (<0.001 mg/dm^3), formaldehyde and residual organic solvents. The samples do not exhibit cytotoxicity (<10% cell death), sensitization, or irritant effects, and are apyrogenic. Hemostatic efficacy was tested, with the sample containing aminocaproic acid showing better results (70-78%) compared to pure alginate (6-30%).

Conclusion

New hemostatic materials based on sodium alginate are safe and meet the requirements for biological and toxicological safety. The combination of alginate with aminocaproic acid significantly enhances the effectiveness of hemostasis. Further research into the use of these combined hemostatic agents in disaster medicine and surgery holds promise. Keywords. hemostatic materials, toxicity study of medical devices, sodium alginate, aminocaproic acid, assessment of hemostatic activity.

Conflicts of interest

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Development of a fluorescence polarization immunoassay for quantitative analysis of acetochlor In corn silk (zea mays)

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Introduction

Acetochlor is a commonly employed herbicide that can accumulate in agricultural products, including corn (Zea mays), as well as in medicinal plant materials such as corn silk, posing a potential health risk. The objective of the research was to develop a method for the quantitative determination of acetochlor in medicinal plant raw materials (corn silk) using the fluorescence polarization immunoassay (FPIA) method.

Methods

Polyclonal rabbit antibodies to acetochlor and a tracer labeled with fluorescein acetochlor AMPA-EDF were used to develop the method. The analysis was performed using the fluorescence polarization method on a Sentry-200 portable fluorimeter (Ellie LLC), the data were processed using the Sigma Plot 11 software package (Systat Software Inc., USA). The analysis was conducted using medicinal raw materials of

corn silk, purchased at a local pharmacy. Method validation was performed for the following parameters: analytical range, specificity, accuracy, and precision.

Results

A method for quantitative determination of the pesticide acetochlor in corn silk (Zea mays) raw materials using FPIA was developed, optimized, and validated. Acetochlor extraction from plant material was performed using water followed by liquid-liquid extraction into hexane. The method demonstrated a detection range of 2.4×10^{-3} to 0.072 mg/kg, corresponding to 8-240% of the maximum permissible level (0.03 mg/kg) established by regulatory standards. No cross-reactivity with butachlor or metolachlor was observed at concentrations corresponding to IC_{20} (0.04 μg/mL). The method was proven to be specific, precise, and accurate. Analysis of corn silk samples revealed acetochlor concentrations ranging from 0.3 to 1.3 μg/kg.

Conclusion

A method for rapid quantification of the pesticide acetochlor in corn silk (Zea mays) medicinal raw materials was developed using fluorescence polarization immunoassay (FPIA).

Conflicts of interest

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Establishment of dynamic data of trace elements homeostasis based on speciation analysis as the function of preventive health

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Introduction

Speciation analysis — the determination of chemical forms of trace elements alongside total content — is a powerful tool in bioanalytical chemistry and clinical medicine. Research by A.V. Skalny et al.[1] using ICP-HPLC in studies of Parkinson's disease (PD) and animal models[2,3] demonstrates that alterations in metal distribution between high-molecular-mass (protein) and low-molecular-mass (amino acid) complexes constitute critical pathological biomarkers. In contrast, total elemental levels frequently remain within reference ranges despite underlying disease.

Results

In healthy subjects, copper is primarily bound to ceruloplasmin (94.8%), with a smaller fraction associated with albumin (4.3%) and negligible amounts present as low-molecular-mass (LMM) species. Manganese is principally bound to transferrin (49.7%), with minor proportions complexed with albumin (2.7%) or present as hydroxyacid complexes (26%). In PD patients, ceruloplasmin-bound copper is significantly reduced, and LMM copper species are detectable. Additionally, manganese–albumin complexes exhibit a more than fourfold increase. These results redefine physiological normality not as a static concentration range, but as a dynamic equilibrium among specific chemical species, dependent on tissue type, biological system, and physiological status. This paradigm shift enables the derivation of novel diagnostic indices for clinical assessment such as set of minimum necessary and sufficient similar coefficients (markers) can be included in the calculation and outpatient clinical control: Labile Copper Index (ILM): (Cu-LMM / Cu-Ceruloplasmin). The norm is <1.5%, the excess is >2.5% is a marker of the risk of neurodegeneration and oxidative stress, as well as a pronounced risk of rheumatoid arthritis or the Coefficient of manganese redistribution (KPM): (Mn-Albumin / Mn-Transferrin). The ratio 0.05–0.25 is higher than the norm — the risk of toxic accumulation of Mn in the central nervous system and the Zinc-albumin ratio (CAC): (Zn-Albumin / Zn-LMM) - decrease in this ratio reflects impaired albumin transport function and inflammatory status, associated with metabolic syndrome.

Conclusion

This approach can be extended to other essential trace elements. For selenium, for instance, the ratio of selenoprotein P (SELENOP) to non-specific selenium compounds serves as a functional indicator; a shift toward non-specific forms denotes deficiency. Disruption of these speciated patterns—termed "speciation imbalance" — represents an early and often reversible manifestation of physiological dysfunction, preceding detectable changes in total element content, which typically reflect prolonged pathology. The application of speciation analysis holds significant promise for broader applications beyond PD, including Alzheimer's disease (via metal-amyloid interactions), Wilson's disease (copper metabolism), cardiovascular diseases (oxidative stress), and oncology (altered metalloprotein metabolism in tumors).

Conflicts of interest

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Toxicological assessment of herbicides from the class of sulfonylurea exposure in long-term administratione

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Introduction

Over the last thirty years, new, fourth-generation herbicides from the class of sulfonylurea derivatives have been widely used. They are used in small doses (5–50 g/ha), providing

high efficiency. Considering the possibility of contact with pesticides by the general population and their entry into the body with food, water and air, it is important to assess the degree of toxicity and danger of pesticides in different ways of drug administration.

Methods

To establish the degree of damaging effect on the body with long-term administration, a study of the chronic action of a drug based on chlorsulfuron was conducted. The experiments were performed on outbred sexually mature white male rats weighing 220–240 g, 20 animals per group. The statistical group included 6 animals. Chlorsulfuron was administered with a metal probe in a 30% solution into the stomach. The doses tested were 0.5; 5.0; 25 and 50 mg/kg b.w. The toxic effect was judged by changes in integral, hematological, biochemical and physiological indices. A morphofunctional assessment of the long-term effect of chlorsulfuron on the body of warm-blooded animals was carried out.

Results

The toxicological parameters of chlorsulfuron were determined during long-term oral exposure. It was found that the studied substance has a polytropic effect during chronic oral administration to warm-blooded animals at doses of 25 and 50 mg/kg b.w. The experimental animals showed changes in a number of hematological and biochemical parameters, as well as changes in the activity of antioxidant defense enzymes. At a dose of 50 mg/kg b.w., after 3 months from the start of the challenge, a reliable dose-dependent increase in SOD activity was observed. With an increase in the exposure time (after 12 months), a reliable dose-dependent decrease in SOD activity was recorded in the rats' bodies. The structural and functional parameters of the studied organs revealed their varying degrees of sensitivity to the effects of different doses of chlorsulfuron. At the maximum effect of chlorsulfuron at a dose of 50 mg/kg (Group IV), among the studied structural and functional parameters, no statistical differences from the control values were found in the following organs: heart, testicle, thymus, spleen, large intestine, pancreas and thyroid gland, which allows us to assume that this drug does not affect these organs. The absence of pathological changes in the internal organs of the experimental animals and morphofunctional changes and activity of antioxidant defense enzymes when exposed to chlorsulfuron at a dose of 0.5 and 5.0 mg/kg b.w. allowed to establish a no-effect (NOEL) dose of 5 mg/kg.

Conclusions

Based on the results of the studies, the no-effect level (NOEL) of chlorsulfuron was set at 5 mg/kg b.w., and the effective level was 25 mg/kg b.w.

Conflicts of interest

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Study of elemental homeostasis of essential elements Fe and Cu in children and adolescents in remission after treatment of malignant neoplasm

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Introduction

The study of metabolism in children and adolescents with various malignant neoplasms (MN) is due to their high social significance. The reason for the imbalance of chemical elements in patients with MN is the etiopathogenetic mechanisms of carcinogenesis, as well as complications of antitumor therapy.

Methods

Our research was performed at the Russian Field Medical and Rehabilitation Research Center. All patients aged 12 to 18 years and 18+ were in remission after treatment of MN. Inductively coupled plasma mass spectrometry (ICP-MS) was used to comprehensively assess the elemental status of patients.

Results

The most informative results of changes in the homeostasis of the essential elements Fe and Cu were found in patients in the ICD-10 C 40.0 group (Malignant neoplasm of the scapula and long bones of the upper limb). There is a change in the content of the studied bioelements in girls (F) and boys (M). There is a tendency to increase the level of both iron and copper in the body of children and adolescents, especially in saliva. The greatest average deviation of excess iron b is observed in boys 18+. A significant accumulation of iron in saliva is typical for boys aged 12-18 and older. For copper, there is no tendency to accumulate in the hair. The average deviation of excess copper in the blood serum is relatively evenly observed in all age and gender groups, most clearly in the group of boys 18+. Significant accumulation of copper in saliva is typical for girls aged 12-18 years.

Conclusions

The data obtained clearly demonstrate that such measures as timely correction of deficiencies and excesses of individual bioelements at the level of detoxification of the body, nutrition correction and pharmacocorrection should become an important link in full-fledged remission in children and adolescents after MN treatment.

Conflicts of interest

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Homogeneous assay of aflatoxin b1 based on modulated fluorescence of fluorescein labeled derivative of aflatoxin b1 upon immune complexes formation

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Introduction

Immunochemical assays are widely used to detect various compounds in complex media. Their conduction under homogeneous conditions provide simple, rapid and accurate testing. However, realization of immunoassays in a solution

without separation of reactants needs a parameter changing after immune complexes formation. It is known that binding of antibodies to a fluorescently labeled hapten often leads to modulation (quenching or enhancement). of registered fluorescence. By this way, direct measurement of fluorescence can provide information on the formation of immune complexes in solution. In the presented work this approach was used to detect aflatoxin B1 (AFB1), an important toxic contaminant of food stuffs, in grain samples.

Methods

AFB1 was conjugated with a fluorescein derivative (FAM5) using 3-(O-carboxymethyl)oxime as a bridge molecule. The conjugate was purified by thin-layer chromatography. Its ability to interact with antibodies was confirmed by fluorescence polarization immunoassay (FPIA). For immunoassay with modulated fluorescence of the label, competition between the conjugate and AFB1 in the sample for binding to antibodies was used. AFB1 was extracted from ground wheat grain with acetonitrile, filtered, and the filtrate was evaporated at room temperature by a factor of 6. Then, the filtrate was diluted to the original volume with phosphate-buffered saline, pH 7.4, containing monoclonal antibodies to AFB1 and AFB1-FAM5 conjugate, and fluorescence of FAM5 was recorded (ex - 495 nm, em - 520 nm).

Results

Mixing the AFB1-FAM5 conjugate (5 nM) with antibodies to AFB1 (3 nM) resulted in an approximately 2.3-fold increase in fluorescence. Thus, the bound form of AFB1-FAM5 fluoresces significantly more strongly than the free form. Addition of free AFB1 to the mixture resulted in restoration of the fluorescence level up to the initial one. This effect was used to determine the AFB1 content in grain extracts. For contaminated grain samples, the possibility of determining AFB1 at concentrations up to 0.25 ng/mL was demonstrated, which is significantly lower than the established MRL value of 2 ppb. The degree of AFB1 revealing varies from 70 to 97%.

Conclusions

The proposed method is characterized by methodical simplicity and high sensitivity. Conducting a homogeneous analysis allows reducing the time of results obtaining and excluding the stages of separation of bound and free reagents, typical for heterogeneous immunoassays, such as ELISA. Measuring fluorescence requires simpler and cheaper equipment than fluorescence polarization in the case of FPIA.

Conflicts of interest

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The level of trace elements and cytochrome cyp2d6 polymorphism in survivors of hemoblastosis complicated by the development of metabolic syndrome

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Introduction

More than 30% of patients treated for childhood malignancies develop various metabolic disorders (dyslipidemia, insulin resistance, and obesity) during rehabilitation. The formation of insulin resistance and obesity may be associated with impaired metabolism of essential elements, as well as exposure to toxic metals. The levels of trace elements and minerals in the blood serum can lead to metabolic changes in obese individuals[1]. Hereditary predisposition, including polymorphisms in cytochrome P450 genes involved in chemotherapy drug metabolism, is also significant, as these variations are associated with clinical outcomes and toxicity[2]. This study aimed to analyze the association between metabolic disorders, levels of essential and toxic trace elements, and the frequency of CYP450 allelic variants in survivors.

Methods

The study included 214 children, 95 girls and 119 boys with hemoblastosis, who got rehabilitation course at the Russian Field Medical Center. The average age was 10.2 years (from 4.9 to 17.6 years), the duration of remission was 4.6 years (from 0.8 to 12.5 years). Body mass index (BMI), fasting glucose, NOME-IR index, and cholesterol levels were determined. The NGS method was used to study the coding regions of genes involved in the metabolism of antitumor drugs. The levels of toxic and essential trace elements were determined by the ICP-MS method. The search for correlations was carried out using Spearman's criterion. Various machine learning models were used and the permutation method was applied.

Results

43% of patients were overweight, including 16% with obesity (+2.0 SDS BMI). Insulin resistance (HOMA-IR >3.2) was found in 23% of children, and a combination of at least two metabolic disorders (insulin resistance and overweight) occurred in 14% of patients. Strong positive correlation was observed in the patients' hair between the levels of toxic elements, primarily lead and cadmium, as well as arsenic, manganese and iron, and their negative correlation with zinc levels. The insulin resistance index and body mass index were positively correlated with the level of zinc and negatively with the level of toxic metals in the hair, which makes it possible to use the value of trace elements as prognostic risk factors for metabolic syndrome.

Conclusion

Using machine learning methods, the importance of the genetic polymorphism of the CYP2D6 gene associated with the individual body's ability to detoxify has been revealed for the development of insulin resistance and an increase in BMI.

Conflicts of interest

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The elemental status of the population of the Krasnoyarsk Krai: Analysis of ionomic hair profiles and assessment of the influence of environmental factors

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Introduction
In the modern world, issues of environmental safety and the impact of the environment on human health are becoming increasingly relevant. The Krasnoyarsk Krai as the region with diverse landscape, climatic and ecological conditions, as well as a developed industry, can have a significant impact on the bioelement status of the population.
Methods
Laboratory studies to determine the concentration of chemical elements in the hair of the examined individuals were performed by mass spectrometry with induction-coupled argon plasma.
Results
In the course of the study, it was revealed that adult residents of the Krasnoyarsk Krai (especially in the southern part, including city of Krasnoyarsk) have a relatively reduced content of macro- and microelements in their hair compared to other regions of the Siberian Federal District and Moscow. In the first part of the study, the ion profiles of about 3,500 adult residents (about 75% of whom were women) and 1,000 girls and boys in the Krasnoyarsk Krai were examined. Over 20 years of observations, there has been a noticeable trend towards a decrease in the level of toxicants: the level of lead in men's and women's hair decreased by 1.7 and 1.2 times; mercury — by 1.6 and 1.4 times; cadmium — by 2.1 and 1.6 times, as well as an increase in selenium levels (1.1 and 1.3 times respectively). Children from the Krasnoyarsk Krai were found to have lower levels of many elements in their hair compared to children from the Siberian Federal District, including mercury, lead, cadmium, cobalt, vanadium, selenium, manganese and chromium, while maintaining a higher zinc content.
Conclusion
There is a positive trend in reducing the level of toxic elements and increasing the level of protective elements (for example, selenium) in the hair of both adults and children. The results of the study indicate an improvement in the ecological situation and the quality of the environment in the Krasnoyarsk Territory and may be useful for forming strategies to improve the quality of life of the population of the Krasnoyarsk Territory.
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Human hair analysis in relation to similar environmental and occupational exposure in the Krasnoyarsk Krai

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Introduction
Humans are an integral part of ecosystems. Hair is recognized as an informative and reliable biological material that reflects the environmental conditions as well as the presence of diseases and health deviations in the human body. The chemical composition of hair reflects both the internal physiological state and the impact of various exogenous factors. Due to the slow growth rate of hair, its analysis provides an average concentration of macro- and microelements over several years. Monitoring of trace element content in hair can address a wide range of scientific challenges, including the indication of environmental pollution and the assessment of harmful factors affecting human health.
Methods
10000 hair samples were collected from adults and children residing in the Krasnoyarsk Krai. Laboratory studies to determine the concentration of trace elements in the hair of the examined individuals were carried out using mass spectrometry with inductively coupled argon plasma.
Results
No significant changes in the hair elemental status were observed over the last 20 years. The concentration of chemical elements in hair reflects the ecological state of the territory, which is formed under the influence of natural and/or anthropotechnogenic factors. Database has been created that allows to obtain reference values for the content of chemical elements in hair for the studied region. For the 2015-2024 years decreasing trends were observed in hair concentrations of toxic trace elements (mercury, lead and cadmium) in presence of steady-state (calcium, magnesium, zinc) or elevated (selenium) levels of their antagonists.
Conclusions
Conducted analyses confirmed usefulness and necessity of conducting research basing on the use of sensitive biomarkers, such as hair. Hair analysis allows for an assessment of the natural environment contamination, which is particularly important for the research on populations inhabiting areas with different degree of pollution. Studies targeting subgroups of populations have shown a direct relationship between hair metal content and exposure to various environmental pollutants, especially in the northern area of the Krasnoyarsk Krai due to air, water and food pollution.
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The sensitivity of human somatic cells to the genotoxic effect of pesticides in vivo and in vitro

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The study of individual human sensitivity to adverse environmental factors is important for predicting pathology risk and developing preventive measures. Of particular concern are factors capable of disrupting genetic structures in human cells. Comparison of genotoxic effects of chemicals in the somatic cells of the same donors in vivo and in vitro makes possible to elucidate the applicability of such in vitro model for predicting individual sensitivity to genotoxicants and identifying high-risk groups. The aim of the study is a

comparative study of the sensitivity of human somatic cells to the genotoxic effects of pesticides in vivo and in vitro. The micronucleus test on buccal epithelial cells of humans contacted with pesticides in the spring-summer season due to professional exposure and the DNA comet assay on human peripheral blood lymphocytes with and without a metabolic activation were used. Before blood sampling for in vitro assessment, the study participants (48 persons) had not been exposed to pesticides at least for 3 months). A study of the ability of captan, thiram, and chlorpyrifos to induce DNA damage in vitro showed statistically significant dose-dependent genotoxic effects (based on the "% DNA in the tail comet" parameter). The levels of DNA damage in lymphocytes from different donors varied in the range of 4-20 times. A group of donors was identified whose lymphocytes were more sensitive to the DNA-damaging effects of pesticides. The micronucleus test in vivo showed statistically significant differences in the level of cytogenetic damages in the epithelial cells of different donors. Comparisson of the abnormalities observed in buccal epithelial cells in the micronucleus test and the levels of DNA damage in the lymphocytes in the DNA comet assay revealed the comparable results in the in vivo and in vitro systems. Donors whose lymphocytes showed the maximum DNA-damaging effect of pesticides also had a high risk level based on the index of accumulation of cytogenetic damage. Epithelial cells of individuals whose lymphocytes were tolerant to the action of pesticides in the model test system showed a low level of cytogenetic abnormalities and a high reparation index. The in vitro test system using human lymphocytes may be a promising model for assessing individual genetic risk.

Conflicts of interest

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Protective and anti-inflammatory effects of cardamom (elettaria cardamomum) against bee venom toxicity in an in vitro wound model

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Introduction

Elettaria cardamomum (cardamom), which contains compounds with potential anticonvulsant activity, is consumed as an aromatic spice native to Asian regions. Numerous studies have demonstrated the biological activities of E. cardamomum and its polyphenols, including antioxidant, antitumor, anti-inflammatory, and metabolic regulatory effects. Its antimicrobial and anti-inflammatory properties can reduce infection risk in the wound area and accelerate healing. Bee venom (apitoxin) is a mixture of bioactive molecules, with melittin as its major component, and is rich in proteins, lipids, and various hormones with significant pharmacological effects. Based on this knowledge, the present study aimed to investigate whether E. cardamomum could alleviate bee venom-induced inflammation in an in vitro wound.

Methods

A wound model was created on a fibroblast cell line using a 200 µL pipette tip, followed by the application of bee venom at a toxic dose (5 µg/mL) for 15 minutes. The cells were then treated with different concentrations of E. cardamomum (1, 2, and 3 µg/mL) for 48 hours. At the end of the treatment, cell viability was assessed using the MTT assay, while oxidative stress and antioxidant capacity were evaluated by TAS, TOS, LDHA, and NO analyses. Apoptotic features were examined using Annexin-V/PI staining and DAPI immunofluorescence. All data were statistically analyzed.

Results

Cell viability was reduced by 49.92% in the bee venom group ($P<0.05$). However, E. cardamomum preserved cell viability in a dose-dependent manner, with the highest effect observed at 3 µg/mL, showing approximately a 1.5-fold increase compared to the bee venom group ($P<0.01$). LDH levels in the treatment group decreased 2.3-fold compared to the bee venom control ($P<0.01$). Moreover, E. cardamomum reduced oxidative damage and lowered NO levels by approximately 2.3-fold ($P<0.01$). The vasodilatory effect of NO contributed to increased blood flow, enhanced oxygen and nutrient supply, and promoted cell proliferation. Immunofluorescence analysis revealed decreased Annexin (FITC)/PI-positive cells, indicating suppression of apoptosis.

Conclusion

These findings suggest that E. cardamomum treatment substantially mitigates bee venom-induced cytotoxicity and cellular damage in vitro.

Conflicts of interest

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Treatment of bee venom-induced toxicity with c. polygonoides: A study focusing on inflammation and apoptosis

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Introduction

Bee venom contains compounds such as adrenaline, dopamine, histamine, mellitin, phospholipases A2 (PLA2s), phospholipases B, and serotonin. Melittin is the main and most toxic compound in bee venom, comprising 50–60% of the total venom. Melittin causes allergic reactions and pain caused by bee stings. Accumulated melittin and PLA2 act synergistically with lipid membranes, leading to cell damage. Currently, various therapeutic methods are used for the treatment of bee venom-induced toxicity. C. polygonoides Linn. belongs to the Polygonaceae family and does not require any vegetation for growth. C. polygonoides (CP) flowers

contain high amounts of protein and have digestive properties. Based on this information in the literature, our study aimed to determine the effects of CP on bee venom toxicity.

Methods

A linear wound model was studied on the fibroblast cell line (L929), and each cell group was treated with 5 µg/ml bee venom. CP doses of 1, 2.5, and 5 µg/ml were used for treatment. After treatment, cells were incubated for 48 hours, and semi-quantitative analyses of MTT, LDH, TAC, TOS, and PPAR-γ, as well as Annexin V+PI+DAPI expression, were performed using immunofluorescence staining. The findings were statistically evaluated to determine their significance.

Results

When the results were examined, bee venom (5 µg/ml) induced a significant apoptotic effect on L929 cells. Cell viability decreased by 44.58%, and the level of oxidative damage increased significantly ($P<0.01$). Following CP treatment, antioxidant and anti-apoptotic effects against bee venom occurred with increasing doses. In particular, the CP 5 µg/ml dose increased cell viability approximately 1.75-fold compared to the bee venom group ($P<0.01$), bringing it closer to the control group. Similarly, it increased PPAR-γ activation 3.2-fold, maintaining mitochondrial stability and increasing TAC levels. Contrary to PPAR-γ and TAC levels, decreased TOS and LDH levels were observed. Immunofluorescence findings indicate a decrease in Annexin (FITC)/PI antibody levels, which inhibits apoptosis.

Conclusion

This study demonstrated that C. polygonoides promotes proliferation by reducing apoptosis and inflammation in bee venom-induced toxicities. This therapeutic effect contributes to the literature for future studies.

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Therapeutic potential of ceiba speciosa in counteracting bee venom-induced cytotoxic and inflammatory responses

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Background

In wound models, bee venom (apitoxin) has both therapeutic and toxic effects through biologically active compounds such as melittin, apamin, and phospholipase A2. At high doses, the toxic effects of apitoxin become predominant. Melittin-induced membrane damage leads to fibroblast and keratinocyte death, while the excessive inflammatory response caused by phospholipase A2 results in edema, cellular damage, and necrotic areas in the wound region. C. speciosa (Flush tree), a plant rich in flavonoids (quercetin, rutin, kaempferol derivatives), phenolic acids (gallic acid, chlorogenic acid), triterpenes, and sterols, exhibit significant anti-inflammatory properties through these phytochemical constituents. This study aimed to evaluate the treatment of bee venom-induced apoptosis with C. speciosa.

Methods

An in vitro wound model was established in the L929 cell line using a 200 µL pipette tip, followed by the application of bee venom at a toxic dose (5 µg/mL) for 15 minutes. Cells were then treated with different concentrations of C. speciosa (1 µg/mL, 2 µg/mL, and 3 µg/mL) for 48 hours. At the end of 48 hours, the experiment was terminated, and cell viability was measured using the MTT assay. TAS, TOS, LDHA, and Klotho analyses were performed to determine oxidative stress and antioxidant capacity. In addition, Annexin-V-PI and DAPI immunofluorescence staining were used to detect apoptotic features. The data obtained were statistically analyzed.

Results

Examination of the data revealed that cell viability decreased by 42.08% in the bee venom group ($P<0.05$), which was correlated with an increase in LDHA levels ($P<0.05$). However, C. speciosa preserved cell viability in a dose-dependent manner, and viability increased with higher concentrations. The best result was obtained in the C. speciosa 3 µg/mL treatment group, where viability increased approximately two-fold compared with the bee venom group ($P<0.01$). At the same time, oxidative damage induced by toxicity decreased in the C. speciosa group. Klotho levels increased approximately 2.6-fold ($P<0.01$). Accordingly, antioxidant enzyme activity increased, DNA damage decreased, and apoptosis was suppressed. Immunofluorescence results showed a reduction in Annexin (FITC)/PI antibody levels, indicating the suppression of apoptosis.

Conclusion

These results demonstrate that C. speciosa has anti-apoptotic and anti-inflammatory effects against bee venom toxicity.

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Beyond IC50: Molecular determinants of dose–response curve shape for etoposide, busulfan, and melphalan in leukaemia and normal cells

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Background/Objectives

Etoposide, busulfan, and melphalan are commonly used cytotoxic agents in high-dose chemotherapy and

haematopoietic stem cell transplantation. Their effectiveness varies between malignant and normal cells due to overlapping yet distinct molecular mechanisms. This in vitro and in silico study aimed to mechanistically interpret IC50 values and dose–response curves in two leukaemia cell lines (HL-60, K-562) and normal fibroblasts (MRC-5), integrating drug–protein interactions, gene expression, and functional profiling.

Methods

IC50 values were measured in vitro using MTT assays and four-parameter logistic regression. Hill coefficients and IC80/IC20 ratios were calculated to characterise the steepness and dynamic range of cytotoxic responses. In silico analyses incorporated data from UniProt, DrugBank, and the Human Protein Atlas to identify pharmacologically relevant proteins and quantify their transcriptomic expression (nTPM).

Results

Etoposide demonstrated the highest efficacy in HL-60 cells, where elevated TOP 2 A expression and apoptotic priming outweighed efflux and repair pathways. Busulfan produced broad, flat dose–response curves across all cell lines, with limited selectivity mainly driven by glutathione-based detoxification and transporter activity. Melphalan induced steep, threshold-like responses in K-562 cells, attributable to LAT 1-mediated uptake and decreased detoxification. High Hill slopes and narrow IC_{80}/IC_{20} ratios reflected cooperative apoptosis with an abrupt transition to cell death, while shallow slopes with wide ranges suggested heterogeneous or buffered responses.

Conclusions

Parameters of dose–response curves capture biologically meaningful dynamics of cytotoxicity beyond IC50 values alone. Integration of IC50, Hill slopes, and IC80/IC20 ratios with proteomic and transcriptomic profiling identified key molecular determinants—including drug targets, transporters, detoxification enzymes, and apoptosis regulators—that shape cell-specific responses. This mechanistically informed framework enhances the predictive value of cytotoxicity assays and may support biomarker-guided drug selection and safer, personalised chemotherapy strategies.

Conflicts of interest

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From in vivo study pitfalls to in silico uncertainty: Toxicological studies of metal mixtures

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Background

Toxicological assessment of chemical mixtures is becoming more important as humans are chronically exposed to multiple agents rather than single compounds. Metal mixtures are especially concerning due to their dual role as essential trace elements and toxic pollutants, with further relevance to occupational exposures and smoking. However, many in vivo studies lack clear relevance to humans and have significant methodological flaws.

Methods

This critical review systematically examined published in vivo studies of metal mixtures, focusing on common pitfalls. Shortcomings include: (i) inadequate mixture definition without justification of human relevance; (ii) inappropriate biomarker selection, failing to capture both exposure and effects; (iii) excessive numbers of mixture components, making factorial designs unmanageable; (iv) dose selection based on uncertain reference levels and irrelevant exposure routes; (v) neglect of chemical speciation, despite organic and inorganic metal compounds exhibiting markedly different toxicokinetics and toxicodynamics. While dissociation produces the free metal ion, which binds sulfhydryl groups, disrupts metalloenzymes, redox balance, and ion exchange, the target organ determines divergent clinical manifestations (e.g., neurotoxicity vs. nephrotoxicity). Additional flaws include (vi) improper euthanasia and tissue preparation, such as the use of anaesthetics or omission of organ perfusion; (vii) neglect of species, strain, sex, and age in animal models; and (viii) flawed comparisons between studies that are not comparable.

Results

These flaws explain the variability and poor reproducibility reported in the literature. They hinder interpretation of mixture effects (synergism, additivity, antagonism), distort biomarker outcomes, and impair extrapolation to humans. Moreover, standard analytical methods such as atomic absorption spectrometry cannot differentiate whether a measured metal originates from an organic or inorganic compound, further limiting mechanistic understanding. Flawed results are often incorporated into toxicological databases, creating misleading inputs for in silico models.

Conclusion

Future studies should: (i) limit mixtures to no more than three metals relevant to humans; (ii) apply mechanistically based dose selection; (iii) use realistic exposure routes; (iv) ensure proper euthanasia and mandatory organ perfusion before tissue sampling; (v) select biomarkers that reflect both exposure and effects; and (vi) consider species, strain, sex, and age. Only with these improvements can in vivo toxicology of metal mixtures yield meaningful and translatable insights. Poorly designed studies that propagate flawed data into databases ultimately weaken the predictive power and regulatory reliability of in silico toxicology.

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Public Health and Toxicology