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CONFERENCE ABSTRACTS

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New generation and pharmaceutical scientists

Pharmacist-led and audit & feedback intervention to reduce antibiotic prescribing in primary care (AFA study)

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Background: The World Health Organization (WHO) has recognized antibiotic resistance as one of the top 10 global public health threats facing humanity. It remains a major challenge, primarily driven by inappropriate and excessive prescribing. Primary care plays a crucial role, accounting for 90% of total antibiotic prescriptions. In Spain, antibiotic consumption continues to rise, showing significant regional variability and remaining far from the European goal of a 20% reduction. Addressing this issue requires effective and efficient implementation strategies to promote sustainable changes in prescribing behaviours.

Objectives: to evaluate the efficacy of a face-to-face pharmacist-led intervention and an Audit and Feedback (A&F) strategy in general practitioners (GPs) to reduce antibiotic prescriptions (total number of prescriptions per 100 GP visits) in the primary care setting. Additionally, as secondary objectives, this study aims to assess the reduction in the percentage of patients initiating antibiotic treatment, as well as changes in total antibiotic prescriptions categorized by therapeutic group and prescribing indication. It will also examine prescribing patterns based on the sex of both GPs and patients. Furthermore, the study will evaluate the reach and engagement of the intervention, its adoption, implementation fidelity, maintenance, feasibility, and perceived acceptability.

Methods: this study is a randomized controlled trial conducted in primary care settings across three Spanish regions (Balearic Islands, Catalonia, Valencia, and the Basque Country). GPs will be randomized and allocated into control or intervention group (1:1). As inclusion criteria, all participating GPs will have an assigned patient list from the past three months. GPs in the intervention group will receive individualized graphical feedback and tailored messages regarding their antibiotic prescribing patterns, including information on dose, duration, prescribing indications, and point-of-care test usage (e.g., streptococcal rapid tests and C-reactive protein tests). Additionally, they will have access to an online training course. GPs in the highest prescribing quartile (≥ P75) and those who request further support will receive a tailored face-to-face intervention by the primary care pharmacist. This session will include goal-setting, action planning, and case discussions. GPs will also have the option to receive lists of their patients with recent antibiotic prescriptions. The intervention will align with the prescribing criteria established by the Plan Nacional Resistencia Antibióticos (PRAN). The control group will receive an active A&F intervention aimed at reducing prescriptions of benzodiazepines, proton pump inhibitors, or antipsychotics.

Ethics and Dissemination: The study was approved by the Balearic Islands Ethics Committee. Findings will be disseminated through research conferences and peer-reviewed journals. The AFA (PI22/01742) Study received funding from the Carlos III Institute of Health (ISCIII), the Ministry of Science and Innovation (Spain), and European Union ERDF funds (European Regional Development Fund). It

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A new participation model – early career pharmacists

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Introduction: The Council of Young Pharmacists (CYP) of the Portuguese Pharmaceutical Society (PPS) developed an initiative based on the Citizens' Assembly in collaboration with MOAI-Consulting to actively engage early-career pharmacists in shaping the future of the profession. The initiative aimed to encourage discussions and develop evidence-based recommendations addressing critical challenges in pharmaceutical practice, particularly in healthcare system integration, digital transformation, and professional development.

Method: The project was inspired by the Citizens' Assembly and Economic Co-operation and Development (OECD) Deliberative Toolbox, and conducted in three phases: pre-assembly planning, assembly discussions, and post-assembly initiatives. An initial survey was disseminated to 1,500 contacts with 160 respondents (10.6%). To maximise the response rate, PPS contacted the mailing list via phone calls, emails, and SMS. For the selection of participants the CYP followed a random selection process based on two main criteria: professional area and responses to the main questionnaire. For the assembly, respondents of the questionnaire were also randomly selected to have representativity of the different areas, including community pharmacy, clinical, hospital pharmacy, industry, and regulatory affairs, ensuring the participation of 23 pharmacists under 35 years old from diverse professional backgrounds. Participants collaborated in thematic groups to develop recommendations, which were later voted on using a consensus-based methodology. To facilitate discussions and guide participants within the groups, the CYP and MOAI invited a group of experts with relevant expertise on the discussed topics to collaborate as mentors. The final recommendations were ranked according to feasibility, impact, and urgency for implementation.

Results: The assembly discussions generated 22 key recommendations, focusing primarily on enhancing pharmaceutical integration into Local Health Units (ULS), expanding the clinical pharmacy role, improving access and interoperability of health data, and implementing artificial intelligence (AI) in pharmacy practice. High-priority recommendations included the establishment of standardised protocols for pharmaceutical services within ULS, the structured integration of clinical pharmacists into healthcare networks, and the reinforcement of continuous professional development models, integrating mandatory requirements for pharmacists adapted to their level of specialization, competences, and roles. Digital transformation emerged also as a key topic, with recommendations advocating for bidirectional communication between pharmacists and other healthcare professionals, standardisation of health data access models, and the development of digital tools for pharmaceutical interventions. Additionally, proposals underscored the need for AI-focused education and ethical guidelines to ensure responsible AI implementation in pharmaceutical practice. The final prioritisation combined consensus scores with impact assessments, resulting in a strategic hierarchy of recommendations to guide future implementation.

Conclusion: This initiative demonstrated the effectiveness of a participatory model in addressing pressing challenges within the pharmaceutical profession through the opinion and prioritisation of early career pharmacists. By fostering discussions and evidence-based decision-making, the project provided a framework for policy development and professional advocacy. The integration of young pharmacists into strategic decision-making processes ensures that emerging professionals contribute to shaping a dynamic, responsive, and future-ready pharmaceutical sector. The implementation of these recommendations requires continuous stakeholder engagement, structured action plans, and ongoing evaluation to ensure meaningful advancements in pharmaceutical practice.

Formulation and characterisation of chitosan-loaded Guiera Senegalensis nanoparticles for the treatment of Malaria

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Background: Malaria remains a critical public health issue in Africa, necessitating innovative treatment strategies. In Africa, the integration of natural products with nanomedicine is pioneering new frontiers in malaria treatment. This study focuses on the formulation and characterization of chitosan-loaded Guiera senegalensis nanoparticles, combining the therapeutic potential of natural products with advancements in nanomedicine. Guiera senegalensis, a plant native to Africa, is renowned for its medicinal properties, including antimalarial activity. By integrating this natural product with chitosan, a biocompatible polymer, and utilizing nanotechnology, we aim to enhance the efficacy and delivery of antimalarial drugs.

Methodology: The dried leaves of the plant sample were pulverised and extracted using a maceration method with ethanol. A bottom-up, approach (green synthesis) was used to synthesise the nanoparticles. The formation of the nanoparticles was confirmed visually when the resultant solution changed its color. The pH of the resultant was taken, while the mixture was centrifuged at 3000rpm for 10 minutes. The nanoparticles were then subjected to comprehensive physicochemical characterization, including dynamic light scattering (DLS) for size distribution, Differential scanning calorimeter (DSC) for heat flow, and Fourier-transform infrared spectroscopy (FTIR) for chemical structure confirmation. The in vitro antimalarial efficacy of the nanoparticles was evaluated.

Results/Findings: The synthesised nanoparticles met the standards and exhibited a uniform size distribution with an average diameter of 150-200 nm. SEM analysis confirmed the spherical morphology of the nanoparticles, while FTIR spectra validated the successful encapsulation of Guiera senegalensis within the chitosan matrix. In vitro studies demonstrated that the chitosan-loaded nanoparticles provided sustained drug release over 72 hours and significantly enhanced antimalarial activity.

Conclusion/Recommendation: This research highlights the potential of combining traditional medicinal knowledge with advanced nanotechnology to develop novel antimalarial therapies. The chitosan-loaded Guiera senegalensis nanoparticles demonstrated promising results in terms of sustained drug release and enhanced antimalarial efficacy. Future work should focus on both in vitro, in vivo studies and by extension clinical trials to further validate the safety and effectiveness of this innovative approach.

A supervised machine learning model for clinical decision support in asthma diagnosis

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Background: Asthma, a chronic respiratory condition characterised by airway inflammation and narrowing, is a major global health issue, affecting millions worldwide. This disease is particularly prevalent in low- and middle-income countries (LMICs), where it contributes significantly to morbidity and mortality due to under-diagnosis and inadequate treatment. In Zimbabwe, a typical LMIC, asthma management is hindered by limited healthcare infrastructure, a shortage of trained medical personnel, and inadequate access to diagnostic tools. Addressing these challenges is critical, as early diagnosis and appropriate management are essential to improving health outcomes. This study explores the potential of machine learning (ML) models to enhance asthma diagnosis in Zimbabwe, thereby offering a scalable solution to bridge diagnostic gaps in resource-constrained settings.

Method: The research employed the Cross-Industry Standard Process for Data Mining (CRISP-DM) methodology, a structured and iterative approach comprising six phases: business understanding, data understanding, data preparation, modelling, evaluation, and deployment. The dataset utilised in this study included health-related information from 2,392 patients, focusing on demographic, environmental, and clinical features. To address the significant class imbalance between asthmatic and non-asthmatic cases, the Synthetic Minority Over-sampling Technique (SMOTE) was applied. Three machine learning models were developed and evaluated: a Random Forest Classifier, a Support Vector Classifier (SVC), and a Decision Tree Classifier. The models were assessed using metrics such as accuracy, precision, recall, F1 score, and area under the Receiver Operating Characteristic (ROC) curve.

Results: Among the models tested, the Random Forest Classifier achieved the highest performance, particularly in distinguishing between asthmatic and non-asthmatic patients, as evidenced by the highest area under the curve (AUC) in the ROC analysis. Key predictors of asthma diagnosis included chest tightness, hay fever, age, body mass index, and pollen exposure. Despite expectations, there was little correlation between certain variables like Forced Expiratory Volume (FEV1) and Forced Vital Capacity (FVC), often used together as a ratio in clinical assessments. This lack of correlation necessitated separate inclusion of these variables

in the model, impacting the approach to feature engineering. Nevertheless, the model's robustness and accuracy in prediction highlight its potential utility in clinical settings.

Conclusion: The study demonstrates that machine learning models, particularly the Random Forest Classifier, can substantially improve the accuracy and timeliness of asthma diagnosis in LMICs like Zimbabwe. By identifying significant predictors and effectively handling complex, non-linear relationships among variables, these models offer a promising tool for enhancing clinical decision-making. The findings suggest that integrating ML-based diagnostic tools into the Zimbabwean healthcare system could mitigate some of the current challenges in asthma management, such as the reliance on symptom-based diagnosis though limited access to spirometry still presents challenges. Future research should focus on expanding the dataset to include more diverse and larger samples, which would improve model training and validation. Additionally, further efforts are needed to explore the real-world deployment of these models, including considerations for integration into existing healthcare systems and ongoing model updates to maintain accuracy. By adopting such innovative approaches, Zimbabwe could improve asthma outcomes and reduce the public health burden of this chronic disease.

Evaluation of copper-dependent enzymes activity in a murine model of Menkes disease.

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Background: Menkes disease, a genetic disorder of the copper transport protein ATP7A, causes systemic copper deficiency. In patients, the activity of copper-dependent enzymes, including cytochrome *c* oxidase (C*c*O), the last enzyme in the electron transport chain, and lysyl oxidase (LOX), an essential enzyme in connective tissue development, is reduced. Currently, patients are treated with copper-histidine, but copper transport to the brain is difficult after blood-brain barrier (BBB) maturation. Additionally, this approach cannot deliver copper to the Golgi apparatus, resulting in low LOX activity regardless of the timing of administration. Consequently, many patients die in early childhood. Given these challenges, more effective novel therapeutic agent is required.

Aims: The present study aimed to establish an evaluation system for the C*c*O and LOX activity in macular mice, a murine model of Menkes disease and to assess the efficacy of a novel candidate therapeutic agent, copper complex A which has a dithiazine framework.

Methods: Either copper complex A or the vehicle was administered intraperitoneally to male macular mice (12-15 weeks old), while male C3H/HeNCrl mice (control mice, 12-15 weeks old) received only the vehicle. The administration was three times daily for four days. On the fifth day, one hour after the final administration, the mice were euthanized, and the cerebral cortex and liver were collected.
1) C*c*O activity: C*c*O exhibits enzymatic activity as a holoenzyme upon copper incorporation into cytochrome c oxidase subunit 1 (COX1). A correlation between C*c*O activity and COX1 level has been previously reported. To evaluate C*c*O activity in the brain, COX1 level in homogenates was quantified by Western blot analysis using ATP5A as an internal standard.
2) LOX activity: To evaluate LOX activity in liver homogenates, the fluorescence intensity produced by the redox reaction between LOX in the sample and the substrate supplied with the LOX Activity Assay Kit (Abcam) was measured. The obtained fluorescence intensity was then divided by protein concentration and reaction time to calculate LOX activity.

Results: 1) C*c*O activity: The average band intensity ratio of COX1 to ATP5A in the vehicle-treated macular mice was significantly reduced to less than one-tenth of that in the control mice, indicating a reduced COX1 level in macular mice for the first time. Furthermore, the average band intensity ratio was increased by more than fivefold in macular mice treated with copper complex A compared with vehicle-treated macular mice, suggesting that the administration of copper complex A improves C*c*O activity. 2) LOX activity: The average LOX activity was reduced by 30% in vehicle-treated macular mice compared with control mice, but no statistical differences were observed among the 3 groups due to large inter-individual variability.

Conclusion: Copper complex A may effectively cross the BBB and enhance C*c*O activity in the brain. On the other hand, copper complex A did not enhance LOX activity probably because C*c*O and LOX acquire activity through different pathways. These findings are expected to provide insights into development of a more effective therapeutic agent for Menkes disease based on the improvement of the activity of copper-dependent enzymes.

Non-animal approach in polyphenols research

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Introduction: Animal studies are considered the gold standard for preclinical drug validation in pharmaceutical development. However, the accuracy and reproducibility of test results obtained from animal studies are compromised in humans due to species differences between animal and human systems. European regulatory bodies support the replacement of animal testing by promoting the 3Rs (Replacement, Refinement and Reduction) Polyphenols, a diverse group of naturally occurring compounds found in plants are known for their potent antioxidant, anti-inflammatory, antiviral, anticarcinogenic, and neuroprotective properties. The primary objective of this research was to explore the integration of in vitro models with polyphenols for enhanced therapeutic applications while improving the understanding of polyphenol characteristics.

Methods: A literature review is conducted to ascertain what is known from the existing literature about in vitro studies on polyphenol compounds. To achieve the objective, literature data from relevant scientific articles were used. The medical database (Medline) and internet search engines (Google Scholar, PubMed) were used for the search. The following keywords were used in the search: in vitro, preclinical trials, polyphenols, curcumin, thymoquinone, resveratrol, cell culture, 3D cell culture, organ-on-a-chip.

Results: Testing active substances and drugs using Non-Animal Approach (NAA) enables precise assesment of the biological activity and toxicity of drugs under controlled conditions, which is crucial for the development of therapy. The broad-spectrum biological effects of polyphenols have drawn a lot of attention. Numerous studies have been conducted so far, allowing significant advancement in understanding polyphenols` mechanisms of pharmacological action. These studies allow researchers to assess drug/active substance efficacy, interactions with target molecules, as well as potential side effects, before tested in vivo. These technologies are widely considered by academia, pharmaceutical industry and regulatory bodies as key developments in biomedicine. These models enable researchers to investigate how polyphenols modulate cytokine levels, oxidative stress responses, and inflammatory pathways in a controlled microenvironment, making them valuable tools for screening therapeutics based on polyphenolic compounds. There are studies that use organ-on-a-chip platforms to investigate the anti-inflammatory and other effects of polyphenols in in vitro models. The results demonstrate polyphenols' potential to modulate cytokine levels, highlighting the importance of such systems for studying inflammation and assessing therapeutic compounds. Polyphenols are characterized as compounds with variable stability and metabolism, which can significantly affect their bioactivity. This can be considered a significant challenge on polyphenol research. Despite their potential, current in vitro models, including organ-on-a-chip platforms, still have limitations in replicating the full complexity of human physiology.

Conclusion: Monitoring of polyphenol concentration and degradation within NAA models is crucial for accurately assessing their biological effects. Organ-on-a-chip technology and other Non-Animal Approach models, combined with polyphenol research, holds great potential. It provides an advanced platform for evaluating bioactive compounds with higher accuracy and translational relevance. In further research, it is necessary to pay more attention to the analysis of the stability of polyphenolic substances during in vitro studies in order to avoid misinterpretation of the results.

Injecting oncolytic virotherapy for high-grade gliomas

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Background information: Oncolytic virotherapy represents a novel frontier in the treatment of high-grade gliomas, leveraging the ability of engineered viruses to selectively target and destroy malignant cells while sparing normal tissue. Why is this study critically important? Amidst the growing urgency to improve therapeutic outcomes for glioblastoma, an aggressive and treatment-resistant brain tumor, this approach has gained increasing attention.

Purpose: The injected viruses are typically modified to enhance their ability to target cancer cells, replicate within them, and trigger an immune response that further attacks the tumor. Additionally, these viruses can be engineered to carry therapeutic genes that boost the body's ability to fight cancer.

Method/Study design: In a clinical investigation assessing the efficacy and safety of DNX-2401, patients received intratumoral viral injections designed to initiate direct oncolysis and stimulate immune-mediated tumor clearance. A structured dose-escalation framework guided administration, with treatment groups experiencing distinct intervention sequences. Tumor response was evaluated through a combination of radiographic imaging, histological analysis, and immune profiling, revealing key insights into viral replication, immune infiltration, and tumor regression patterns.

Dose Administration

•Group A: Single DNX-2401 dose ranging from 1×10^7 to 3×10^{10} vp in 1 mL.
•Group B: Initial dose via catheter ($1 \times 10^7 – 3 \times 10^8$ vp), followed by surgical resection and second DNX-2401 injection.
•Dose escalation followed a 3 + 3 design in Group A, beginning at 1×10^7 viral particles (vp), with Group B enrolling only after the safety of the dose level was confirmed. Study Evaluations and management
•Medical history, MRI, and blood tests were documented at baseline and follow-ups.
•Adverse events (AEs) were graded
•Dose-limiting toxicity was defined as any DNX-2401–related, nonhematologic AE ≥ grade 3.
•Tumor response was evaluated using the Macdonald criteria Response classifications:
•Complete response (CR): ≥ 95% tumor reduction
•Partial response: > 50% tumor reduction
•Progressive disease: > 25% tumor increase

Results: The presence of inclusion bodies further confirmed active adenovirus replication providing evidence of glioma cells lyses and successful replication in human tumors. Immune Effects
Preclinical studies suggest DNX-2401 induces a TH1-skewed CD8+ cytotoxic T-cell response. Consistent with this, MRI scans of complete responders initially showed increased contrast enhancement, followed by tumor regression.

Data analysis:

•Immune analyses reveal robust CD8+ T-cell infiltration, reduced TIM-3 expression, and evidence of immunogenic cell death.
•MRI showed necrosis at the injection site
•Tumor analysis: 80% necrosis, CD8+ T-cell infiltration and Macrophage activation

Conclusion: Studies confirm DNX-2401's ability to replicate in tumors, induce significant regression (≥72% response rate), and achieve durable remission.
These findings reinforce the promise of oncolytic virotherapy as a transformative strategy offering not only direct tumor cytotoxicity but also the induction of a robust and lasting immune response. Monotherapy and in combination with complementary immunotherapeutic agents such as Pembrolizumab may pave the way for enhanced patient outcomes in this intractable malignancy.

Maternal tryptophan supplementation alters offspring gut-brain axis and behaviour in a sex-specific manner

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Introduction: The bidirectional relationship between the brain and gut – known as the gut-brain axis – gained recognition for its role in regulating health. The gut microbiome, shaped by diet, plays a crucial role in the communication between the gut and the nervous system. Both neuronal and microbiota development are highly sensitive to nutritional changes during early life and can be influenced by external factors such as maternal diet. Maternal nutrition plays a crucial role in the health of fetal development and the prevention of neuropsychiatric disorders, influencing gut microbiota composition and the gut-brain axis. Poor nutrition during pregnancy has been linked to increased risks of metabolic disorders and cognitive impairments in both animal and human offspring. Disruptions of the microbiome during the perinatal period have also been associated with neurodevelopmental disorders, namely autism spectrum disorder. This supports the idea that early microbiome development influences brain function and behaviour.

Tryptophan, an essential amino acid and precursor of serotonin (a neurotransmitter vital for regulating mood and cognition), emerges as a key component in regulating both mental health and the composition of the gut microbiome. Because tryptophan is involved in the production of serotonin and of hormones such as melatonin, it is often used as a supplement for treating anxiety and sleep disturbances. Some women experience anxiety, depression, as well as sleep issues during and after pregnancy. Further, clinical studies have shown that low doses of tryptophan can be taken safely during breastfeeding, as they did not increase tryptophan levels in breast milk. However, while tryptophan is an essential nutrient in pregnancy, there is insufficient evidence to confirm the safety of high doses during pregnancy. Understanding how maternal nutrition, the gut microbiome,

and offspring brain health are interconnected is crucial for minimizing the incidence of neurodevelopmental diseases linked to maternal habits.
In this study, it was hypothesized that a tryptophan-enriched maternal diet would disrupt the gut-brain axis in healthy offspring in a sex-dependent manner.

Methods: This study aimed to investigate the effects of tryptophan supplementation in maternal nutrition and exploring its implications for the microbiome and the behaviours of offspring. For that, healthy C57BL/6J dams were feeding with a tryptophan-enriched diet from pregnancy to weaning. Subsequently, gut microbiota composition, metabolic activity, brain levels of tryptophan and serotonin, and behavioral outcomes in male and female mice offspring were analysed.

Results: The findings revealed that maternal tryptophan supplementation led to sex-specific changes. Female offspring exhibited disrupted microbiota diversity, reduced cortical tryptophan and serotonin levels, and heightened anxiety-like behaviours. On the other hand, male offspring displayed increased gut metabolic activity, elevated cortical tryptophan levels, and increased repetitive behaviours.These results suggest that maternal tryptophan supplementation influences offspring neurodevelopment and behaviour by modulating gut-brain axis components in a sex-dependent manner.

Conclusion: This work highlights the need for further research into prenatal dietary interventions. These findings contribute to a deeper understanding of how maternal diet shapes offspring neurodevelopment and may inform strategies for improving mental health outcomes.

Sustainable health education for pharmacy students: Leveraging interdisciplinary online learning and E-portfolios

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Background information: The need for sustainability education in healthcare and life sciences is evident, given the critical role these fields play in advancing sustainable practices. Although sustainability competences have been systematically integrated into the teaching at the University of Helsinki's Faculty of Pharmacy over the past decade through a curriculum reform, the need for new sustainability education content and approaches continues to grow.

This research seeks to develop an online sustainability course specifically tailored for pharmacy, health, and life science students. The course content is built on the United Nations' 17 Sustainable Development Goals, each accompanied by assignments related to human, animal, and environmental health. Primarily based on self-guided and asynchronous online learning, the course is completed by constructing an e-portfolio of sustainability challenges and solutions.

Purpose: The primary objective of this research is to study the effectiveness and impact of a healthcare and life science-oriented online course on sustainability competences. We hypothesize that student-centred learning methods will increase students' self-efficacy beliefs regarding sustainability competences.

Method: An online pre-survey assesses students' prior experiences with sustainability learning and their attitudes toward sustainability. Upon course completion, a follow-up survey measures the development of perceived sustainability competences. Statistical comparisons of pre- and post-test results will further quantify learning outcomes, while correlating background data and course activity with achievement and satisfaction will provide additional insight into how different learners respond. Semi-structured interviews with a subset of participants will offer a deeper, qualitative perspective on their experiences.

Results: Preliminary results indicate a positive shift in students' self-efficacy beliefs regarding sustainability competences. Based on pre-survey responses, most students reported limited prior exposure to sustainability education. However, initial post-course surveys indicate that students may have become more aware of the sustainability content they have encountered in their past education, suggesting improved recognition and understanding of sustainability competences. The reflective portfolios highlight that students feel more capable of contributing to sustainable practices in their professions, emphasising the course's effectiveness in promoting student agency. Flexibility and freedom to tailor portfolios to individual interests and backgrounds have received considerable praise in student feedback.

Notably, the first post-course survey respondents reported significant improvements in their perceptions of their capacity to advance sustainability and their role in promoting sustainability in their professions. Student feedback and survey responses highlight the value of an interdisciplinary approach including portfolio peer evaluation, positively impacting students' perceptions of cross-professional collaboration for sustainable development.

Conclusion: Initial findings suggest that e-portfolio-based online learning can effectively enhance sustainability competences in healthcare and life science students. This research contributes to the effort of incorporating sustainability into curricula, offering a template for similar courses across disciplines. Future work will refine the course based on feedback and expand its reach, potentially

transforming it into a MOOC. The positive feedback from the initial post-course survey underscores the course's potential in significantly enhancing students' perceptions of their ability to advance sustainability.

Exploring the potential of digital pills for improving medication adherence

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Background: Digital pills represent a novel approach to improving medication adherence in patients with chronic diseases. By providing real-time data on medication intake, they offer new opportunities for patients and healthcare providers to optimize treatment and reduce the risk of treatment failure. However, their implementation raises challenges related to technology, cost, and ethics. This research explores how real-time data from digital pills can contribute to increased adherence and examines the opportunities and challenges associated with their use.

Aim: This study investigates the potential of digital pills to enhance medication adherence and evaluates the implications for healthcare systems, focusing on the balance between benefits and implementation challenges.

Method: A systematic literature review was conducted, analyzing research articles published between 2017 and 2024. Literature searches were performed using PubMed and Oria, applying strict inclusion and exclusion criteria to ensure relevance and quality. A total of 13 studies were included, focusing on both the efficacy and challenges of digital pills.

Results: Digital pills were found to improve medication adherence by providing real-time data, enabling better monitoring and decision-making for chronic conditions such as HIV, tuberculosis, and diabetes. Patients and healthcare providers benefited from enhanced insights into treatment adherence, leading to improved health outcomes. However, challenges such as unstable data transmission, high costs, and ethical concerns related to privacy and patient autonomy were also identified.

Conclusion: Digital pills hold significant promise for addressing medication adherence challenges, particularly for chronic diseases. Successful implementation, however, requires technological advancements, patient-centered solutions, and a robust ethical framework to address concerns of privacy and autonomy.

Future research should prioritize large-scale studies, long-term effects, and solutions to technological and economic barriers. Interdisciplinary collaboration among healthcare professionals, technology providers, and policymakers will be essential for realizing the full potential of digital pills in improving medication adherence and healthcare outcomes.

Advancing drug screening for Glioblastoma: The potential of 3D bioprinting

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Background: Glioblastoma is an aggressive brain tumor with a poor prognosis, with a five-year survival rate of less than 10%. Resistance to temozolomide, the current standard treatment, and its inability to target glioblastoma stem cells contribute to frequent disease progression and recurrence. The low success rate of oncological drugs, with only 3.4% advancing from preclinical to clinical stages, highlights limitations in existing in vitro and in vivo models. Developing reliable, cost-effective in vitro models that better mimic the tumor microenvironment is crucial for improving drug screening.

Aims: This review evaluates the effectiveness of 3D bioprinted glioblastoma models and their potential to replace existing in vitro and in vivo models for drug screening. It also provides an overview of 3D in vitro systems, focusing on their advantages and limitations.

Method: A comprehensive review of recent studies on 3D bioprinted glioblastoma models was conducted. The literature search focused on advancements in bioprinting technology, biomaterials for recreating the tumor microenvironment, and the comparative effectiveness of these models versus traditional ones. Studies addressing advantages, challenges, and technical aspects of 3D bioprinting were analyzed.

Results: 3D bioprinted glioblastoma models offer significant advantages over traditional 2D cell cultures and animal models. These models better replicate the tumor microenvironment by incorporating extracellular matrix components, nutrient gradients, and cell-cell interactions, resulting in improved predictive accuracy for drug efficacy and resistance.

However, challenges persist, including the lack of standardized protocols, limited quality control measures, and the complexity of biomaterial selection and bioprinting techniques. These limitations hinder the broader adoption of 3D bioprinting in drug development pipelines.

Conclusion: 3D bioprinted glioblastoma models represent a promising advancement for glioblastoma research, offering superior tumor representation and improved drug screening potential compared to existing models. While these models enhance preclinical predictive power and may reduce clinical trial failure rates, further efforts are needed to standardize

protocols, optimize techniques, and ensure reliable quality control. Continued interdisciplinary collaboration is essential to advance these models and improve glioblastoma treatment outcomes.

Establishment of a 3D culture model using patient-derived conditionally reprogrammed breast cancer cells and evaluation of their sensitivity to anticancer drugs

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Introduction: Breast cancer is the most common malignant tumor in women, with the highest cancer-related mortality rate among women. In the era of precision medicine, accurately predicting individual sensitivity to chemotherapy drugs in vitro could enhance treatment success rates and reduce the economic burden on patients. There is a critical need for an in vitro cancer model capable of accurately predicting individual responses to anticancer drugs to improve the success rate of breast cancer treatment. Currently, cancer research heavily relies on tumor cell lines, which often fail to replicate the heterogeneity of breast cancer among patients. Applying Conditional reprogramming (CR) technology to individualized drug sensitivity testing for tumors can overcome the limitations of tumor cell lines in reproducing tumor heterogeneity among patients. Compared to traditional two-dimensional (2D) cell culture models, three-dimensional (3D) models better mimic the tumor microenvironment in vivo and provide greater physiological relevance, offering significant advantages in predicting the response of tumors to drugs. The objective of this study was to establish a long-term in vitro culture system for primary breast cancer cells derived from patients using CR technology and to construct a 3D in vitro culture model based on Alginate-Gelatin Methacryloyl (Alg-GelMA) microspheres.

Methods: CR technology was employed to culture CRBCs. Alg-GelMA microbeads were created to explore their potential use in the assessment of anti-cancer drugs. Cell proliferation was examined using the MTS assay. Live/dead staining was performed to estimate cell distribution and viability using Calcein acetoxymethyl ester/Propidium iodide double staining. Protein expression was assessed by Western blot.

Results: The Alg-GelMA microspheres prepared in this study possess a porous structure similar to the extracellular matrix of breast cells, providing an appropriate spatial scale for cell growth. Alg-GelMA microspheres exhibited good permeability, allowing for the exchange of oxygen, nutrients, and metabolic waste. It was observed that tumor cells in Alg-GelMA microspheres tended to aggregate into cell clusters, and a 3D culture model was established at a density of 10,000 cells/microsphere, maintaining good cell viability over the long term. Compared to cells cultured in 2D, the proliferation rate of cells in 3D microspheres was significantly reduced, and cell cycle analysis showed an increase in the proportion of cells in the G0 phase and a decrease in the S phase. Transwell analysis indicated enhanced migration and invasion abilities of CRBCs cultured in 3D. Furthermore, the expression levels of cancer stem cell markers (ALDH1A1, OCT-4, and LEF-1) were upregulated in CRBCs cultured in 3D microspheres. Drug sensitivity assays revealed that CRBCs in 3D microspheres exhibited greater resistance to anticancer drugs compared to those in 2D culture. Analysis of apoptosis pathway protein expression in CRBCs cultured in different dimensions after drug treatment showed that cells cultured in 3D microspheres exhibited stronger anti-apoptotic capabilities.

Conclusions: These findings suggest that 3D hydrogel cell cultures more closely mimic in vivo biological and clinical behavior which demonstrated higher innate resistance to anti-cancer drugs compared to 2D cultures, and thus provide valuable tools ready to be used in diagnosis, drug screening, and personalized medicine.

Phytochemical profile and in-vitro biological activity of novel Leonotis leonurus extracts on Monoamine oxidase B (MAO-B) – a drug target for depression

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Background: Depression is a debilitating mental disorder affecting over 120 million people worldwide. Current antidepressants are associated with many side effects and in patients with comorbidities, they present severe adverse drug-drug interactions. Medicinal plants present a possible source for new potential antidepressant drugs. They have played an important role in drug discovery, with many pharmaceutical products originating from them. Purpose: The study aimed to optimize and standardize the extraction conditions of Leonotis leonurus for optimal yield of phytochemicals with inhibitory activity of the human monoamine oxidase B (MAO-B) enzyme.

Methods: The aqueous, methanol, and hexane extracts of Leonotis leonurus were prepared using the maceration technique. The fractions of the dried leaves were obtained using an IP protected extraction technique with variations in temperature, pressure and extraction time. TLC, HPLC and FTIR were used to evaluate the chemical profile of the extracts and fractions. The qualitative phytochemical analysis, total phenolic content (TPC), total flavonoid content (TFC) and DPPH scavenging activities of the extracts and fractions were evaluated using standard methods. The in

vitro MAO-B inhibitory activity of the extracts and fractions was evaluated using standard methods.

Results: The extracts and the fractions revealed the presence of flavonoid, phenols, glycosides, tannins, saponins and terpenoids. The methanol extract had the highest TPC and TFC at 62.5ug/ml and 384.4ug/ml respectively, while the F6B2ii fraction produced the highest DPPH free radical scavenging activity of 87.46 ug/ml at 250 ug/ml concentration GAE. FTIR revealed the presence of major hydroxyl functional groups in the extracts and fractions. Fraction F3D3iii showed the highest MAO-B inhibitory activity of 64.8 % inhibition at the 100 ug/ml concentration.

Conclusion: An optimised extraction yielded a fraction of the plant with MAO-B inhibition properties higher than that of conventionally prepared extracts. The presence of prominent MAO-B inhibitory constituents in the F3D3iii fraction of Leonotis leonurus suggest its potential for eliciting pharmacological effects that might be useful in the treatment of depression.

Lactobacillus reuteri protects Parkinson's disease mice through attenuating BBB impairment mediated by VSMC apoptosis and inhibiting neuroinflammation

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Introduction: Parkinson's disease (PD) is a prevalent neurodegenerative disorder characterized by gastrointestinal (GI) and motor dysfunctions, with limited treatment options available. Emerging evidence has implicated the involvement of gut microbiota dysbiosis in PD pathogenesis. As an approach to balancing the gut microbiome, probiotics have exerted potential health benefits to PD. Based on our previous study, Lactobacillus reuteri (L. reuteri) holds promise as a therapeutic candidate for treating PD via microbiota-gut-brain axis. Here, this study aims to systematically evaluate the therapeutic potential of the probiotic L. reuteri supplementation in a rotenone-induced PD mouse model and to elucidate the underlying mechanisms.

Methods: In the current study, the chronic administration of rotenone was utilized to induce a PD mouse model. Behavioral tests and gastrointestinal function tests were performed to evaluate the protective effects of L. reuteri treatment on PD symptoms. Furthermore, several molecular experiments including metagenomic sequencing, transcriptome profiling, and metabolome analysis were conducted to investigate the underlying mechanisms.

Results: Oral administration of L. reuteri remarkably alleviated the motor dysfunctions in the rotarod test ($P < 0.001$), the pole test ($P < 0.01$), the grip strength test ($P < 0.05$), the adhesive removal test ($P < 0.05$), and the open field test (all $P < 0.001$) of rotenone-intoxicated mice. Besides, L. reuteri supplementation attenuated the GI dysfunctions of rotenone-challenged mice, with improvements in the number of pellets, water concentration, fecal output frequencies, colon length, and transit distance (all $P < 0.001$). In addition, L. reuteri treatment restored the dopaminergic neuronal loss and α-synuclein (α-syn) aggregation in the substantia nigra (SN) of PD mice (both $P < 0.01$). These data together suggested that L. reuteri administration could protect against the PD model. Mechanistic experiments revealed that L. reuteri treatment restored gut microbiota dysbiosis, reduced intestinal inflammation, and enhanced gut barrier integrity. Furthermore, transcriptomic sequencing of midbrains unveiled that L. reuteri treatment suppressed vascular smooth muscle cell (VSMC) apoptosis, thereby preserving blood-brain barrier (BBB) integrity and reducing neuroinflammation in the SN. Metabolic analysis found that serum taurodeoxycholic acid (TDCA) and taurochenodeoxycholic acid (TCDCA) levels were significantly elevated in the rotenone mice but remarkably decreased by L. reuteri treatment. Moreover, correlation analyses revealed that these two bile acids were negatively associated with the behavioral test, the GI function test, the dopaminergic neuron numbers, and the VSMC marker expression, while they were positively correlated with the α-syn expression and the colon histological score. These findings suggested that the communication between the gut and the brain might be mediated by serum TCDA and TCDCA.

Conclusion: This study demonstrates that L. reuteri supplementation protects against a PD mouse model through restoring the BBB integrity mediated by VSMC apoptosis and reducing neuroinflammation. The microbiota-gut-brain axis mediated by the microbial metabolites, TDCA and TCDCA, appears essential to these neuroprotective effects. These findings further underscore the potential of probiotics as a promising therapeutic strategy for PD.