CHAPTER 55

ZOOLOGY

Doctoral Theses

01. AGARWAL (Surbhi)
Evaluation of Antioxidant, Anti-Inflammatory and Anti-Arthritic Properties in Ant Gut Microbes Through In-Vitro and In-Silico Analysis.
Supervisor: Prof. Vartika Mathur
Th 29290

Abstract

Rheumatoid arthritis is a chronic inflammatory disorder characterized by synovial hyperplasia, leukocyte infiltration, and elevated pro-inflammatory cytokines, necessitating the search for novel therapeutic alternatives. This study explored the anti-arthritic, anti-inflammatory and antioxidant potential of microbial cell-free extracts isolated from the gut of Camponotus compressus and Oecophylla smaragdina. In vitro assays demonstrated the significant anti-arthritic activity of eight potential microbial cell free extracts compared to diclofenac sodium and their antioxidant potential comparable to ascorbic acid. LC-Q-TOF-MS analysis revealed the presence of 85 small molecule compounds, out of which three compounds have been identified for the first time fromany microbial source. Molecular docking studies revelaed that 17 microbial metabolites exhibited strong binding affinities against COX-2, TNF-α, MMP9, IL-1β, and MCSF, surpassing the standard drug diclofenac sodium, highlighting their therapeutic potential. Given the limitations and side effects of synthetic NSAIDs and DMARDs, these findings suggest that C. compressus and O. smaragdina gut symbiont derived bioactive compounds could serve as promising natural alternative for arthritis treatment. However, further in vivo studies are required to validate their efficacy and safety in biological systems.

Contents

1. Review of Literature 2. Materials and Methodology 3. Results 4. Discussion, Supplementary Information, Summary and Conclusion, References, Annexures and List of Publications.

02. CHAUHAN (Vishesh)
Neuropeptide Y as a Regulator of Reproductive Functions in Hemidactylus Flaviviridis: Its Reproductive Phase-Dependent Expression, Role and Regulation.
Supervisors: Dr. Sunil Kumar and Prof. Umesh Rai
Th 29291

Abstract

Present Ph.D. work elucidates the first histology-based whole-brain atlas of wall lizard, Hemidactylus flaviviridis. The lizard brain showed conserved vertebrate brain plan comprising forebrain (telencephalon and diencephalon), midbrain, and hindbrain. The telencephalon included pallium and subpallium, while diencephalon comprised epithalamus, thalamus, and hypothalamus. Tectum and tegmentum were

present in midbrain and hindbrain consisted of cerebellum and medulla oblongata. Further, microscopic analysis revealed presence of four ventricles and several conserved nuclei in lizard brain. In addition, current work investigated reproductive phase–dependent expression, role, and regulation of neuropeptide Y (NPY) and its receptor in diencephalon region and gonads of male and female H. flaviviridis. The highest expression of npy/npyr in late recrudescent diencephalon and early as well as late recrudescent ovary indicated involvement of NPY in the onset of female reproductive functions. This was corroborated by in vitro experiments wherein NPY treatment upregulated diencephalonic gnrh/gnrhr along with ovarian markers of cell proliferation, differentiation, survival (bcl-2, scf, c-kit, pcna, gdf-9, bmp-15, fshr, er-α, er-β, pr), and steroidogenesis (star, cyp19). Further, differential regulation of steroidogenesis was evidenced by increased progesterone but reduced estradiol production in NPY-treated ovaries. Further, kisspeptin and leptin upregulated while substance P and FSH downregulated diencephalonic and ovarian npy/npyr. Nesfatin-1, DHT, and E2 showed tissues-specific effect, inhibiting diencephalonic but stimulating ovarian npy/npyr. In males, both diencephalonic and testicular npy/npyr peaked during recrudescence. Further, NPY treatment enhanced gnrh/gnrhr expression in diencephalon and stimulated testicular expression of bcl-2, scf, c-kit, pcna, gdf-9, bmp-15, fshr, er-α, er-β, ar. NPY also stimulated star expression and upregulated testosterone production. Testicular npy/npyr was stimulated by kisspeptin, DHT, E2, leptin, and nesfatin-1 but inhibited by substance P and FSH. Interestingly, diencephalonic npy/npyr was suppressed by all except leptin. Thus, present work provides first comprehensive evidence of involvement of NPY in regulation of reproductive functions in wall lizard.

Contents

1. Preparation of histology-based whole-brain atlas of wall lizard, Hemidactylus flaviviridis 2. Exploring NPY as a modulator of reproductive functions in female wall lizard 3. Unveiling NPY-mediated regulation of male reproductive functions in Hemidactylus flaviviridis. Future Plan, Synopsis, Appendices, References, Publications and Poster/Oral Presentations.

03. NAGAR (Garima)
Structure-Based Virtual Screening and In-Vitro Evaluation of Novel Acvr1 Inhibitors.
Supervisor: Prof. Indrakant Kumar Singh
Th 29294

Abstract

The serine–threonine kinase ACVR1/ALK2 is a Type I BMP receptor, and its normal function is disrupted by disease-related genetic changes, leading mainly to a rare genetic skeletal disorder, FOP. The research investigates novel ACVR1 inhibitors, combining computational structure-based analysis with experimental validation. The ACVR1 gene pathogenic mutations were first identified through multiple in silico pathogenicity assessment tools, which predicted their existence. We employed structural modelling and comparative analysis to investigate how these mutations affect protein structure and their consequences for protein stability and ligand-binding properties. Following this step, we also conducted virtual screening through molecular docking to identify small molecules that could block the ACVR1 kinase domain. Molecular dynamics simulation studies confirmed the binding properties, protein-ligand interaction patterns, and stability of specific protein-ligand complexes, revealing their dynamic behaviour and ability to inhibit targets. Further, we used an in vitro functional system based on BMP-2-induced C2C12 osteogenic differentiation to confirm their computational results experimentally. The virtual screening process

identified multiple lead compounds for testing their potential to inhibit BMP-2-triggered osteogenic cell differentiation. Functional assays, including Alkaline phosphatase and Alizarin Red S assays, and subsequent RT-PCR analysis, were employed to confirm the osteogenic marker expression and matrix mineralisation during the evaluation of these compounds. A novel ACVR1 inhibitor was identified through an integrated approach combining pathogenic mutation analysis, structure-based virtual screening, molecular dynamics simulations, and in vitro functional assays. These findings establish a comprehensive framework for elucidating mutation-induced structural alterations in the ACVR1 gene and highlight the therapeutic potential of two newly identified compounds for the treatment of ACVR1-associated disorders, thereby providing a foundation for future therapeutic development.

Contents

1. Introduction 2. All Materials and Methods 3. To identify pathogenic non-synonymous ACVR1 variants and assess their predicted effects on protein stability, structural integrity, and conformational dynamics using integrated computational tools 4. To perform structure-based virtual screening, molecular docking, and molecular dynamics simulations to identify selective ACVR1 inhibitors with stable and energetically favourable binding within the active site 5. To establish a BMP-2–induced C2C12 osteogenic differentiation model and evaluate shortlisted ACVR1 inhibitors using cytotoxicity, biochemical, mineralisation, and transcriptional assays. Future Plan, Synopsis, References, List of Publications and List of Abstracts (Poster/Oral) in Conferences.

04. PINKI
Albumin Associated Peptides and their Role in Cancer Pathophysiology.
Supervisor: Prof. Rita Singh
Th 29381

Abstract

Cancer progression is frequently accompanied by proteomic alterations generating low-molecular-weight peptides with diagnostic and functional relevance. BRIP1 (BRCA1-interacting protein 1), a DNA helicase involved in damage repair, is implicated in cancer susceptibility; however, the biological significance of its peptide derivatives remains poorly understood. This study investigates how the cancer-associated proteolytic processing of BRIP1 generates circulating peptide fragments that associate with serum albumin and actively contribute to tumor progression. In vitro studies using A549 lung adenocarcinoma cells demonstrated rapid, energy-dependent internalization of the BRIP1 peptide, indicating an active cellular uptake mechanism. Functional assays revealed that this peptide treatment significantly enhanced cell migration and invasion, supporting its role in promoting oncogenic phenotypes. Furthermore, immunoblotting indicated alterations in the expression of cancer-associated proteins following exposure. To evaluate physiological relevance, in vivo studies were conducted in syngeneic BALB/c mice. Systemic administration of the BRIP1 peptide resulted in notable histological alterations, including epithelial hyperplasia in lung and breast tissues, along with augmented terminal end bud (TEB) morphogenesis, confirming an active role in driving abnormal proliferative responses. Additionally, in silico analyses unveiled crucial insights into BRIP1`s structural characteristics, highlighting significant intrinsically disordered regions (IDRs). These IDRs suggest dynamic roles in cellular signaling vital for cancer development. Polyclonal antibodies were also generated against a selected 23-amino-acid BRIP1-derived peptide to enable precise detection and functional characterization. Collectively, these findings provide evidence that a BRIP1-derived

peptide, stabilized through albumin association, is not merely a tumorigenesis by-product but actively modulates cancer-associated cellular behaviors. This highlights the potential utility of albumin-associated BRIP1 peptides as circulating biomarkers of lung cancer progression and offers mechanistic insight into their contribution to tumor pathophysiology, warranting further diagnostic and therapeutic investigation.

Contents

1. Review of literature 2. Materials and methods 3. To investigate the functional role of brip1 peptides in lung cancer progression 4. To evaluate the in vivo effect of brip1 peptide and elucidate its role in cancer pathophysiology using syngeneic mice model 5. To characterize the physicochemical properties of brip1 protein to understand its degradation. Summary and conclusions, Future perspectives, Synopsis, supplementary data, References and publications.

05. SIWACH (Sneha)
Occurrence of Microplastics in the River Yamuna, their Bioaccumulation and Organ Specific Translocation in Freshwater Fish Species.
Supervisor: Prof. Ram Kishan Negi
Th 29382

Abstract

Despite the widespread occurrence and environmental impact of microplastics (MPs) as a contaminant, a complete assessment including distribution patterns, biological uptake, tracking studies, and toxicology is still largely lacking, especially with regard to the Indian ecosystem. The present thesis contributes a holistic evaluation study on the fate of microplastics in the Yamuna River basin, including environmental occurrence, bioaccumulation studies, advanced imaging, and review synthesis. An extensive survey at 9 different sites across Uttarakhand, Haryana, and Delhi yielded 2,423 microplastics in surface and subsurface waters (36–686 MPs/L), with fibre-shaped MPs (57%) being most common and polyethylene (PE; 64%) the predominant polymer, as confirmed via FTIR-ATR analysis. Urban-industrial stretches affected by barrages were identified as significant pollution hotspots with Potential Ecological Risk Indices (PERIs) of 21.87, primarily owing to high polymer hazard due to PVC. Biological surveys conducted on 220 fish specimens representing 12 ecologically diverse species found a total of 1,678 microplastics mainly localized in the gastrointestinal tract, gills, and muscle tissue, with a Polymer Hazard Index of 4,780 (Category IV). High inter-organ correlations and marked downstream urban pollution gradient indicated that trophic group, feeding ecology, and pollution intensity determine microplastic bioaccumulation and organ translocation processes, which have clear consequences for food security and human exposure to MPs. Limitations of existing destructive techniques were addressed by developing a novel non-destructive imaging strategy incorporating micro-computed tomography (μ-CT) and fluorescent microscopy in Labeo rohita, allowing for the first time a full three-dimensional reconstruction of microplastic localization in entire organisms. In addition to route-dependent accumulation patterns, water exposure was associated with a higher multi-organ burden than diet exposure. Cross-modality correlations ($\rho = 0.71$) provided strong validation for this new method. A literature review on microplastics as carriers of persistent organic pollutants, their potential toxicity via oxidative stress, immune dysregulation, and homeostasis disturbance, and even a new syndrome, such as `plasticosis`, was synthesized. Overall, the present work provides insights into both the environmental occurrence and biological impact of microplastics in aquatic environments, while developing an innovative imaging strategy for ecological toxicology studies.

Contents

1. Review of literature 2. Material and methods 3. Results and discussion 4. Results and discussion 5. Results and discussion. Summary and conclusion, Future plan, Synopsis, References, Publication and conferences.