Inaugural Annual Molecular and Translational Medicine Symposium

1. **A Novel Robust Screening Strategy for NMD-Modulatory Molecules**

Sika Zheng, Ph.D.

School of Medicine, Division of Biomedical Sciences, University of California, Riverside

In the era of precision medicine, small molecules affecting cellular efficiency of nonsense-mediated mRNA decay (NMD) could potentially treat millions of individuals who are afflicted with genetic diseases caused by nonsense mutations. Previous therapeutic development for nonsense mutations has focused on nonsense suppression strategy, or read-through of premature stop codons (PTC), which is designed to generate a full-length protein by promoting incorporation of a near-cognate amino acid at the PTC. Nonsense suppression therapy shows promise but has not yet become an effective clinical treatment, probably because nonsense mutations cause two major problems for an affected protein: quality defect and insufficient quantity. NMD is a cellular surveillance mechanism that selectively targets nonsense transcripts for degradation. Therefore, NMD inhibition as a pharmacological approach to increase the level of truncated proteins merits consideration. We developed a novel and sensitive method, the first of its kind to monitor cellular NMD activity. Our new method was genetically validated for distinguishing NMD regulation from transcriptional control and alternative splicing regulation. We are now using this robust method to screen small chemicals with the goal of identifying NMD-modulatory molecules.

2. Novel strategy of developing therapeutics for anti-influenza virus and drug resistant strains by targeting host factors

Jiayu Liao, Ph.D.

Department of Bioengineering, Bourns College of Engineering, University of California, Riverside

Our long-term goal is to develop novel bioengineering technologies for basic and translational research and development. In last few years, we developed a novel quantitative Förster resonance energy transfer (FRET) technology platform for both basic kinetics parameter determinations and high-through screening(HTS) assays targeting protein-protein interactions and applied these assays for SUMOylation cascade. The novel theoretical and experimental procedures for protein interactions affinity(Kd) and protease kinetics, Kcat/KM determinations in the SUMOylation cascade in a systems biology manner. As proof-of-concept, new classes of host-targeting and broad-spectrum antiviral therapeutics that target essential protein-protein interactions (PPI) and post translational modifications (PTM) of host-virus interactions with high barriers to resistance. We successfully discovered selective and potent SUMOylation inhibitor through a novel FRET-based high throughput screening(HTS) assay and determined its specificities and cellular activities in both SUMOylation and anti-influenza virus, including drug resistant strains. Successful accomplishment of our aims will yield an exciting novel class of broad-spectrum anti-influenza and other antivirals. We are also in the process of testing the hypothesis of synthetic lethality using SUMOylation inhibitor for cancers with “un-druggable” gene mutations, such as c-Myc and KRas mutations. In addition, we are also developing big data based disease predicting model and intervention method using combinatorial herb medicine for diabetes prevention.

3. A Monoclonal Antibody Treatment for Neuropathic Pain

Xin Ge, Ph.D.

Chemical and Environmental Engineering, Bourns College of Engineering, University of California, Riverside

Chronic pain affects millions of Americans but there continues to be an unmet need for safe and effective pain medications. Opioids and nonsteroidal anti-inflammatory drugs dominate the clinical landscape despite limited effectiveness and considerable side-effect profiles. The discovery of matrix metalloproteinases (MMPs) as important mediators of pain initiation and maintenance represents a novel therapy mode of mechanism- rather than symptom-based treatments. However, the catalytic domains of MMPs share a high degree of fold and sequence homology, therefore achieving specific inhibition using small compound inhibitors is extremely difficult. With inherent high specificity, monoclonal antibody (mAb) based MMP inhibitors therefore hold great promises to deliver the needed selectivity. Inspirited by the convex-shaped paratopes of camelid antibodies, Ge Lab at UCR advanced mAb technology to facilitate the discovery of protease inhibitory antibodies [PNAS 2016]. Using this patent pending technology [WO2018067198A1; US16/223,943], a panel of mAb inhibitors of MMP-9 was isolated through our unique approach of functional selection [UC Disclosure 2019-118]. These mAbs exhibited nanomolar potency, exclusive selectivity and proteolytic stability. More significantly, UCR’s mAb L13 inhibited MMP-9 but not MMP-2/-12/-14 and significantly relieved neuropathic pain development in paclitaxel (PTX)-induced pain in mice.

4. Building a Drug Discovery and Development Pipeline at UCLA

Robert Damoiseaux, Ph.D.

University of California, Los Angeles

Academic Drug Discovery has become a major source of innovation for the next generation of drugs for all disease indications. The University of California is especially well situated with an enviable faculty body that has a high appetite for finding cures for diseases and translating their finding into the clinic. This is enabled by many resources – the Molecular Screening Shared Resource being a prime example. We offer a comprehensive set of services covering assay development, high throughput screening, data analysis and follow up work such as potency and toxicity determination as well as selectivity testing. The MSSR offers access to 200,000 compounds and 280,000 functional genomics probes. Projects are supported by three expert MSSR scientists on four fully automated, robotic screening systems with a per diem capacity exceeding 100k samples in 384 well plate format. All plate reader based assay readouts and plate formats are addressable, as are advanced assay readouts such phenotypic screening by true confocal spinning disk microscopy. In this talk we will provide an overview of our capabilities, sample projects we have worked on and present the science behind the first drug based on a molecule discovered at the MSSR that is entering clinical trials for myelodysplastic syndromes and acute myeloid leukemia.

5. Identification of Small Molecule Inhibitors of Zika Virus Infection

Stephanie Thurmond[1], Boxiao Wang[2], Jikui Song[2*], **Rong Hai[1*]**

1 Department of Microbiology and Plant Pathology, University of California, Riverside
2 Department of Biochemistry, University of California, Riverside

Zika virus (ZIKV) belongs to the single-stranded RNA-containing flavivirus family. Its recent outbreak and implication in human diseases (e.g. neurological disorders) have raised a global health alarm, and urgency to develop a therapeutic strategy against ZIKV infection. However, there are no currently approved antivirals against ZIKV available yet. Here, we tested an antiviral strategy against the non-structural protein 5 (NS5) of ZIKV, which is an RNA dependent polymerase (RdRP), responsible for virus-specific genomic replication. Since humans do not encode the RdRP, NS5 is the ideal drug target for flaviviruses. To provide an antiviral for ZIKV in timely manner, we proposed to re-purpose the currently identified flavivirus NS5 inhibitors for potential ZIKV inhibition. As a proof of principle, we tested whether ZIKV infection could be inhibited by the thiophenyl propargyl alcohol (TPA) compounds, which have been identified as inhibitors for Dengue virus (DENV) NS5, in vitro and ex vivo. Our results indicated that this compound suppressed ZIKV NS5-mediated de novo RNA synthesis. Furthermore, we obtained the structure of ZIKV NS5 in the presence of the TPA compound, which provided a basis for structure-based drug optimization for ZIKV NS5. In summary, our results supply key mechanistic insights into the NS5-mediated genome replication and establish a foundation for development of effective inhibitors against ZIKV.

6. Targeting Neuroblastoma and Age-Related Macular Degeneration Through Structure-Based Drug Design Efforts

Jeff Perry, Ph.D.

Department of Biochemistry, College of Natural and Agricultural Sciences, University of California, Riverside

Neuroblastoma is the most common cancer in infants and the third most common cancer in children. We are targeting a unique cancer-associated isoform of PCNA protein that has essential functions in neuroblastoma cells, in collaboration with Linda Malkas and colleagues at the City of Hope Medical Center, to further develop a selective lead compound. We are utilizing macromolecular crystallography, in solution small angle x-ray scattering, and fragment-based drug discovery (FBDD) and coupling these to in vitro biochemical analyses, to aid in our rational-based drug discovery approaches. We are also focusing age-related macular degeneration (AMD), the leading cause of blindness in the developed world. HtrA1 protein is central to wet-AMD disease progression, which is the more rare form of the disease, but contributes to 80% of the blindness cases. Through collaborative studies, we have been applying artificial intelligence (AI) docking approaches in our drug discovery efforts, and we have now identified several 'hit' compounds that inhibit HtrA1 at low micro-Molar concentrations in our in vitro activity assays. We are now focusing on hits-to-leads studies, leveraging our HtrA1 crystals, activity assays and selectivity screens, AI-based approaches and collaborative cell-based assays, to develop a lead compound suitable for testing in models of AMD.

7. Genomics in Drug Design

Adam Godzik, Ph.D.

School of Medicine, Division of Biomedical Sciences, University of California, Riverside

Cancer genomics exploded in last several years, providing us with detailed knowledge of genetic alterations in many cancer types and giving us new insights into molecular aspects of cancer. To extract more information from genomic data, we developed the Cancer3D database at http://www.cancer3d.org and series of associated algorithms as an open and user-friendly way to analyze cancer missense mutations in the context of structures of proteins coded by the mutated genes. Analyzed in relation to patients' clinical data information from cancer3D allowed us to find novel candidate driver genes for specific cancer sub- groups or markers of drug responses and immunity status of tumors. In particular, we found that many cancer mutations affect protein-protein and protein-DNA interfaces, disrupting integrity of specific cellular pathways. By combination of genetic and protein structure analyses and modeling we identified several possible drug targets, where therapeutically intervention my affect disease outcomes, sensitize tumors to already developed drugs or modify host immune response. This approach can be generalized to other diseases beside cancer and with the rapidly increasing amount of genetic information could enhance drug discovery and improve therapeutic targeting across many fields of medicine.

8. The MRB1 Incubator Space and Other Initiatives Supporting UCR Spin Off Companies in Translational Medicine

Mark Leibowitz, Ph.D. - Interim Director EPIC

Eric Gosink, Ph.D. - Entrepreneur in Residence

Office of Technology Partnerships, University of California, Riverside

The Office of Technology Partnerships at UCR is focused on accelerating the translation of UCR discoveries for the benefit of society. Since inception, MOLMED and OTP have collaborated in delivering resources to support translation and partnership. With support from the State of California AB2664, OTP has funded over $300,000 in proof of concept studies to assist with the generation of data relevant to secure additional funds for translation. In this presentation, we will highlight progress to date of such funded projects and will present more information on our newest addition: the life science incubator. A collaborative 3000 ft2 space for UCR and community startups to conduct foundational studies to validate their discoveries for the marketplace.

9. The UC Drug Discovery Consortium

Joseph Wagner, Ph.D.

University of California, Davis

The University of California Drug Discovery Consortium (UC DDC) is a multi-campus initiative created by the University of California Biomedical Research Acceleration, Integration & Development (UC BRAID) Drugs, Devices, Diagnostics Development (D4) group. Building on the strengths of the drug discovery community within the University of California, the synergy of the UC DDC will create teams of industry partners and investigators from multiple UC campuses to facilitate research.

Our goals are to:

- Support the development of therapeutics with mentoring & pilot funding
- Strengthen drug discovery education & training
- Expedite access to drug discovery-related core facilities across the UC system
- Build partnerships with industry to increase support for projects and develop sustainability
- Provide seed funding that will enable faculty members with promising ideas to advance drug discovery or drug development projects

The development of the UC DDC is supported by a 3-year Multi-Campus Research Proposal Initiative grant (MRPI) from the UC Office of the President.

The UC DDC currently involves representatives from UC San Francisco, UC Los Angeles, UC Irvine, UC Davis and UC San Diego. In the future, we hope to include all UC Campuses.

10. **The UC San Diego Center for Drug Discovery Innovation (CDDI)**

Michael Gilson, Ph.D.

University of California, San Diego

The UC San Diego CDDI was established in late 2013 to foster the creation of new medicines by helping faculty members advance successful drug discovery projects and developing new training and educational programs in the field. I will discuss CDDI's initiatives and progress toward these goals, as well as approaches to building sustainability of this program.

11. EPDS and MarketPlace by Covance

Vinita Roy, MBA

Covance

Vinita Roy will describe the Covance MarketPlace initiative that is a tool that could help advance UCR translational research efforts. Marketplace is a web-portal and a value-added solution that can assist UCR investigators showcase their molecules and find a licensing partner. This model brings together pharmaceutical companies and research enterprises sharing the common thread of being Covance current or past customers, through a secure IT platform that is simple to use and provides easy access to Covance's strategic pharma/biotech partners. The net result is that molecules presented in the MarketPlace repository are seen as highly prized assets. While Covance would hope to be eventually a part of the eventual product development activities, there is no contractual or formal obligation by the University or the licensing company to use Covance services to develop their molecules.

12. Targeting tumor metastases via the EphA2 receptor: changing the face of chemotherapy

Maurizio Pellecchia, Ph.D.

School of Medicine, Division of Biomedical Sciences, University of California, Riverside

Eph receptors are a family of tyrosine kinase receptors involved in neuronal connectivity, blood vessel development, and cell-cell interactions. The receptor sub-type EphA2 was identified in cancer cells where it is often highly expressed, mediating communication not only between individual cancer cells, but also between cancer cells and surrounding stromal or vascular cells. Despite EphA2 overexpression, expression of ephrinA1, its ligand, often remains normal even in a cancerous state. This can lead to the accumulation of un-activated EphA2 and subsequent oncogenic activity. In metastatic and drug resistant pancreatic, breast, and prostate cancers, EphA2 expression is dramatically inversely correlated with survival. We have recently developed potent and effective EphA2 agents that we intend to further develop in combination with chemotherapy to target breast, prostate and pancreatic cancer metastases. The agents cause receptor activation and internalization, hence can also we used to selectively deliver chemotherapy to cancer cells as we recently demonstrated in mice models of breast cancer. We feel we have gathered sufficient data to start moving the project to next stages of IND enabling studies and phase I/II trials.

13. Anticancer Proteasome Inhibitors

Michael C. Pirrung, Ph.D.

Department of Chemistry, College of Natural and Agricultural Sciences, University of California, Riverside

This study describes the synthesis, computational affinity assessment, and preclinical evaluation of a syrbactin structural analog TIR-199, a natural product-derived proteasome inhibitor. Molecular modeling and simulation suggested that TIR-199 covalently binds each of the three catalytic subunits of the proteasome and revealed key interaction sites. In vitro and cell culture-based proteasome activity measurements confirmed that TIR-199 inhibits the proteasome in a dose-dependent manner and induces tumor cell death in multiple myeloma and neuroblastoma cells as well as other cancer types in the NCI-60 cell panel. It is particularly effective against kidney tumor cell lines. Adverse drug reaction screens in a kidney panel revealed no off-targets of concern. In vivo studies in mice revealed a 25 mg/kg maximum tolerated dose of TIR-199. The anti-tumor activity of TIR-199 against colon cancer cell lines was confirmed in hollow fiber assays in mice. This is the first study to examine the anti-tumor efficacy of a syrbactin in an animal. Taken together, the results suggest that TIR-199 is a potent new proteasome inhibitor with promise for further development into a clinical drug for the treatment of multiple myeloma and other forms of cancer.

14. Structural Biology, Bioinformatics and Drug Discovery for Cancer and Tuberculosis: Fighting the Emergence of Resistance

Tom L. Blundell, F.R.S., F.Med.Sci.

Department of Biochemistry, Cambridge University, UK

Knowledge derived from genome sequences of humans and pathogens has the potential to accelerate diagnosis, prognosis and cure of disease. We are moving quickly into an era of precision medicine, not only in familial diseases where a mutation in a human gene is important, but also for understanding somatic mutations in cancer. Equally important, the genome sequences of pathogens, for example in tuberculosis or leprosy, can give clues about the choice of existing drugs, repurposing of others, and the design of new ones to combat the increasing occurrence of drug resistance. High-throughput X-ray crystallography using synchrotron sources plays a major role in assessing the druggability of candidate targets identified from the genome sequences.

One approach is to exploit state-of-the-art methods to bring new drugs for different targets to the market, but this will be difficult to finance if patient populations are small. Structure-guided fragment-based screening techniques have proved effective in lead discovery not only for classical enzyme targets but also for less “druggable” targets such as protein-protein interfaces. Initial screening involves small fragments with very low, often millimolar affinities, and biophysical methods including X-ray crystallography are used to explore chemical space of potential ligands. The approach involves a fast initial screening of a library of around 1000 compounds, followed by a validation step involving more rigorous use of related methods to define three-dimensional structure, kinetics and thermodynamics of fragment binding. The use of high-throughput approaches, with X-ray synchrotron sources playing a major role, does not end there, as it becomes a rapid technique to guide the elaboration of the fragments into larger molecular weight lead compounds. I will discuss progress in using these approaches for targets in cancer and in mycobacteria tuberculosis, abscessus and leprae infections.

I will also review our computational approaches using both statistical potentials (SDM) and machine learning methods (mCSM) for understanding mechanisms of drug resistance. These are dependent X-ray crystallographic and comparative modeling to define structures. We have demonstrated that resistance does not only arise from direct interference of the resistance mutation to drug binding but can also result allosteric mechanisms, often modifying target interactions with other proteins. This has led to new ideas about repurposing and redesigning drugs.