National Cancer Institute

Mayo Clinic Arizona/Rochester and U. Minnesota

Overcoming Drug Resistance in Multiple Myeloma

- **Project 1:** Develop high throughput drug screening platform for myeloma cell lines and >500 primary patient tumor samples. Determine frequency of specific mutations or cellular subsets resistant to active drugs or combinations. (Dr. A. Keith Stewart - Mayo Clinic Arizona)

- **Project 2:** Examine mechanisms of IMiD resistance in patients and humanized mice with MM. Explore links between IMiD resistance and activity of Ikaros and modulation by BET and CBP/p300 inhibitors. (Dr. P. Leif Bergsagel - Mayo Clinic Arizona)

- **Project 3:** Model proteasome inhibitor response in MM lines and patient samples with single cell analysis to model gene signatures of response. Perform functional validation of gene signatures with CRISPR-Cas gene engineering. (Dr. Brian Van Ness - University of Minnesota and Dr. Shaji Kumar - Mayo Clinic Rochester)

National Cancer Institute

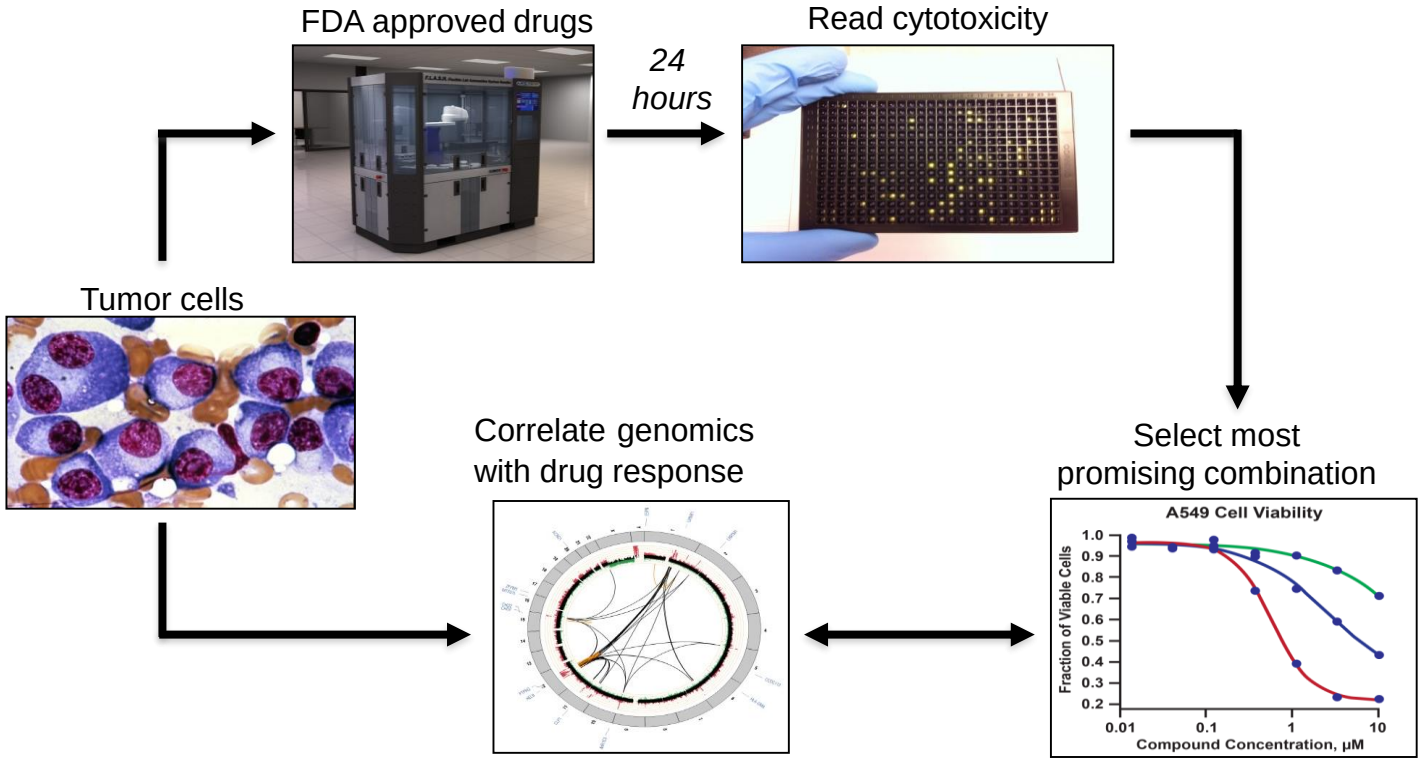
Project 1: Tailoring Therapeutics

Specific Aims

1. We will analyze *in vitro* responsiveness of 500 primary myeloma patient samples against 79 clinically available compounds.

2. We will conduct combination screens with IMiDs and proteasome inhibitors as base compounds to seek synergistic combinations of these drugs with other active MM therapeutics.

3. Utilizing our MM mutation panels, we will conduct a chemogenomic interrogation to examine specific correlates of drug sensitivity and resistance utilizing pre-treatment and surviving cells from primary patient robotic screens. These studies will determine the frequency of specific genetic mutation or cellular subsets resistant to the most active drugs or drug combinations used in MM therapy



National Cancer Institute

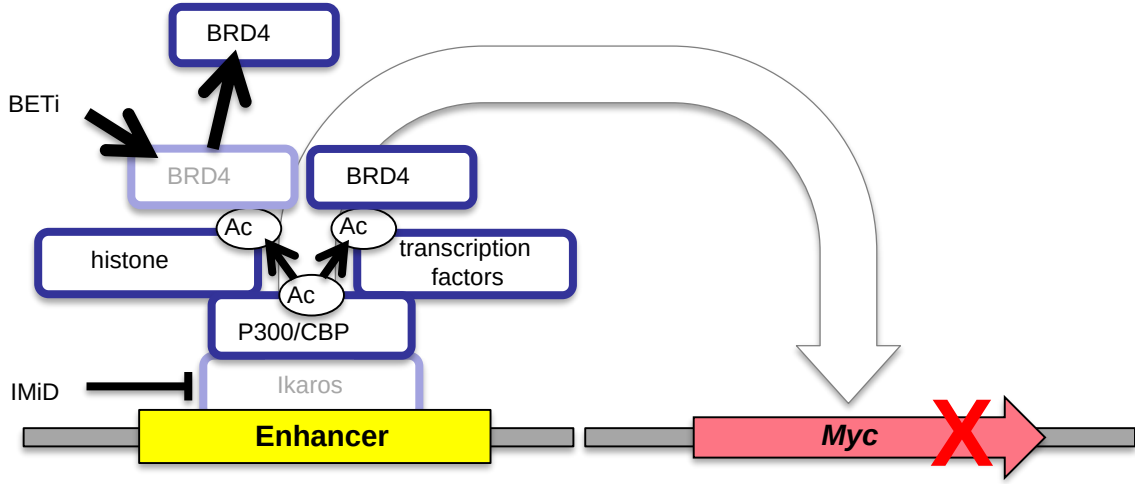
Project 2: Mechanisms of resistance to IMiDs

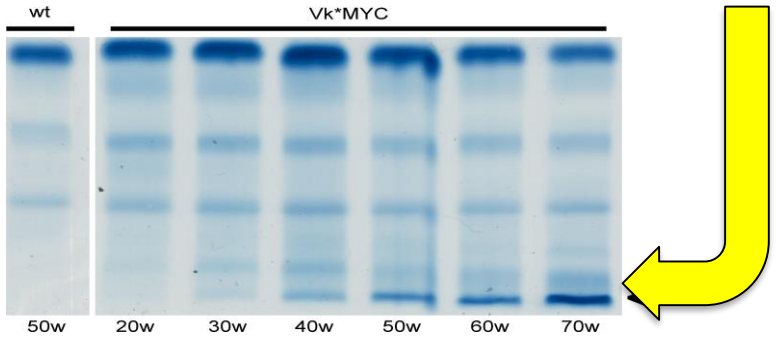
Specific Aims

1. Identify oncogene-translocated enhancers and/or gene expression signatures associated with IMiD resistance in patients. Tumors collected before and after onset of IMiD resistance will be examined for new mutations and structural variations in addition to changes in the gene expression profile to better understand acquired IMiD resistance.

2. Understand the IMiD regulation of enhancer function. A large panel of genetically annotated MM cell lines will be examined for resistance to IMiDs alone and in combination with other enhancer disrupting drugs.

3. Study IMiD response in humanized mouse models. The IMiD response of Mouse PCT cell lines expressing hCRBN will be extensively characterized both in vitro and in vivo. In parallel, the contribution of the host on the response to IMiDs will be studied in a humanized hCRBN Vk*MYC mouse model.





National Cancer Institute

Project 3: Mechanisms of resistance to proteasome inhibitors

Specific Aims

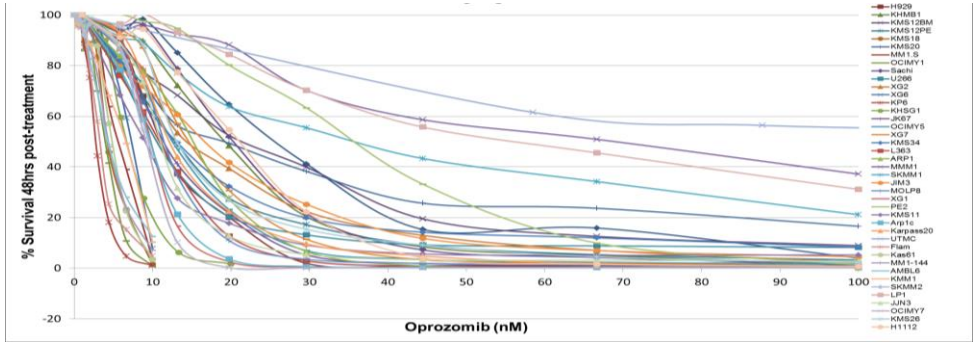
1. Expand analysis of response/resistance of drugs to 80 human myeloma cell lines
   - Develop a gene expression signature that distinguished response and resistance
   - Develop immunophenotypic signatures of subpopulations with varying responses (CyTOF)

2. Apply expression analysis to patient tumor cells
   - Correlate signatures to response, resistance, toxicities
   - Apply single cell analysis to identify subpopulations that may lead to emerging relapses
   - Apply the immunophenotypic analysis to identify response and resistant subpopulations

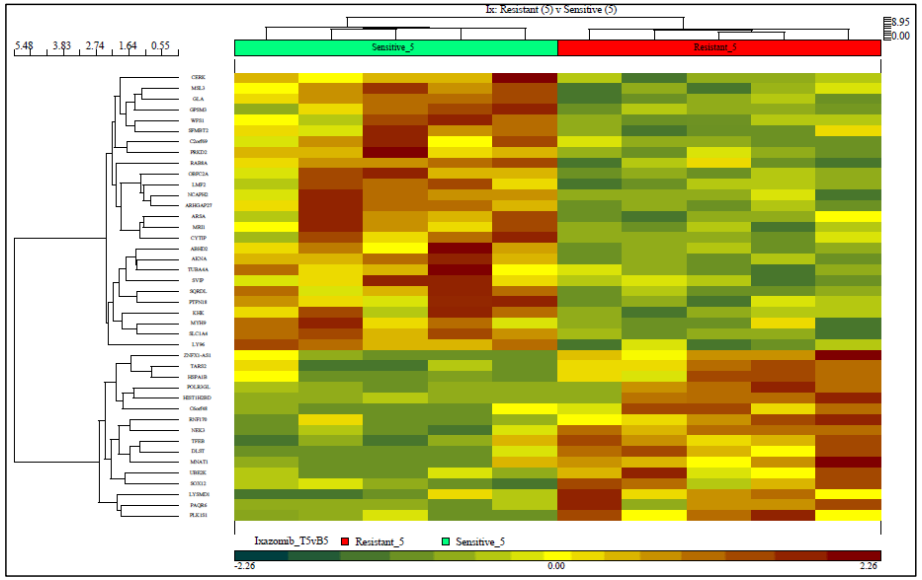
3. SNP analysis of pharmacologic targets affecting PK and PD; and correlate with related drug toxicities.

4. Functionally alter response by CRISPRi/a vectors.

Cell lines show diverse responses to proteasome inhibitors

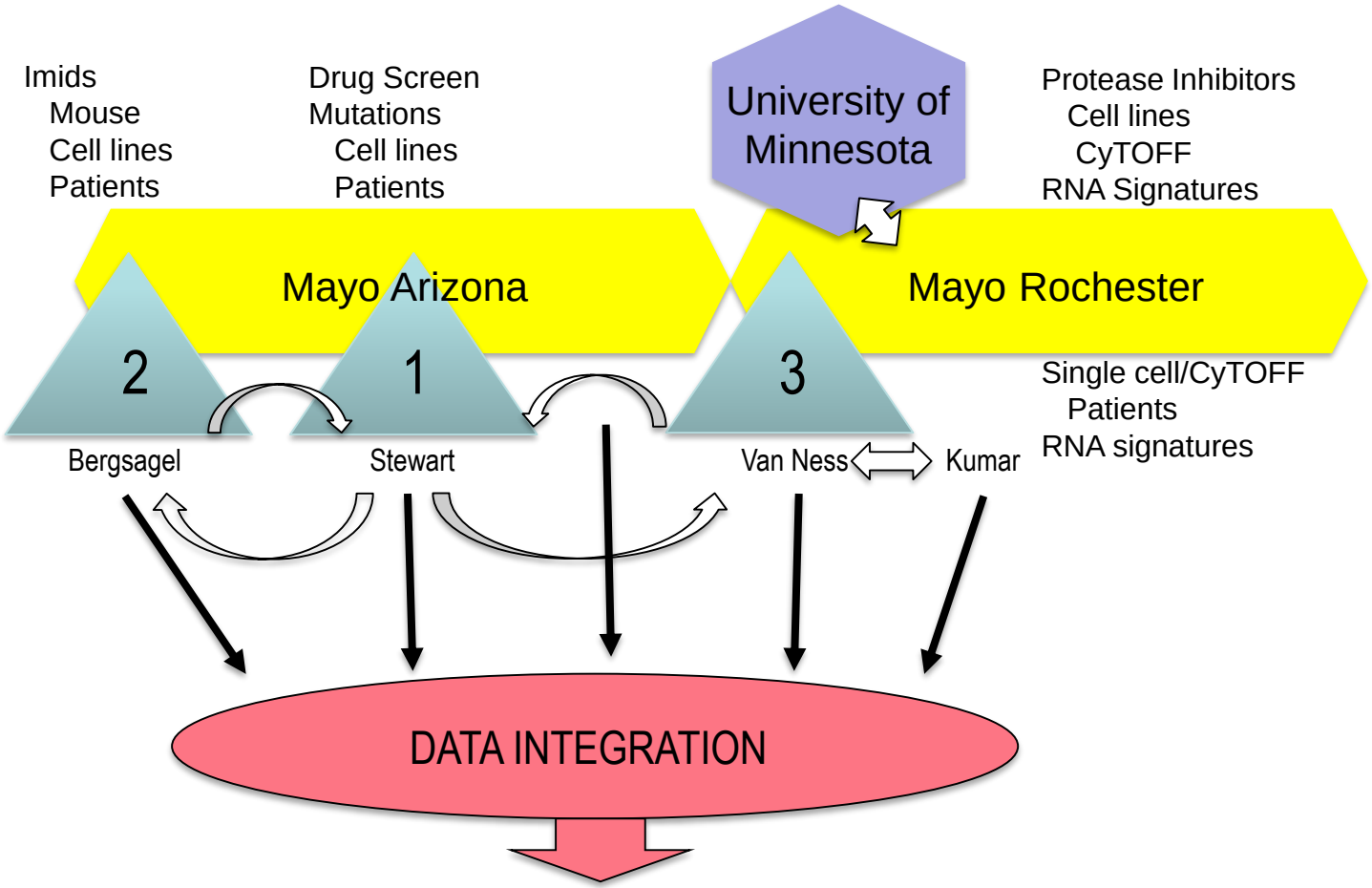


GEP profiles distinguish sensitive and resistant



National Cancer Institute

Myeloma DRSC



Identification of best drug(s) for individualized treatment
Identification of tumor inter- and intra-heterogeneity
Predicting response / resistance to IMiDs
Predicting response / resistance to proteasome inhibitors