Cancer Target Discovery and Development (CTD^2)

Specific Aims

Institution: University of California, San Francisco

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The goal of this proposal is to bridge the gap between the enormous volumes of data generated by the comprehensive molecular characterization of a number of cancer types– and the ability to use these data for the development of human cancer therapeutics. Our end goal is to generate game-changing reagents, and data valuable for the development of cancer therapeutics.

Our general strategy is to take advantage of novel tools and methodologies that we have developed- EXPAND RNAi technology-based on highly complex RNAi libraries that are deconvoluted by deep sequencing. This technology greatly improves our ability to perform comprehensive quantitative RNAi screens and represents a transformative approach for systematically uncovering cancer-relevant gene interaction networks that drive tumor growth and drug resistance with the goal of aiding discovery of polytherapy approaches.

We present our collective labs current efforts which combine in-depth mining of large-scale genomic data, RNAi function-based screening, and systems biology analyses to characterize functional roles of genetic lesions, both alone and in combination, in driving tumor formation and growth. These powerful approaches and the data they yield are expected to contribute to trans-Network activities, including participation in joint pilot research projects. The specific aims of this proposal are to:

Aim 1: Develop next generation EXPAND libraries targeting cancer-specific genetic alterations. In this aim, we will build upon an existing system we have developed to create libraries that will be instrumental in our own efforts and easily disseminated among the trans-Network. Through data integration approaches we will identify candidate driver genes with recurrent mutations from forthcoming TCGA, TARGET, CGCI, ICGC and related initiatives. From these datasets we will generate libraries of full-length cDNA lentiviral constructs and validated potency-optimized shRNAs.

Aim 2: Identify recurrently mutated genes that regulate oncogenic pathways and drug responses. From genome sequencing efforts it is clear that novel recurrent mutations are often co-mutated with known oncogenes, however whether this can influence oncogenic pathways or clinical drug responses is unknown. Using novel EXPANDed libraries from Aim 1, we will identify genes whose perturbation can cooperate or interfere with the ability of known oncogenes to impart hallmark cancer phenotypes to cells in culture and to modulate therapeutic responses. The relevance of recurrent mutations in cancer phenotypes and responses to molecularly targeted inhibitors will be assessed through drug screens in highly relevant engineered primary cell lines.

Aim 3: Produce genetic interaction maps to uncover pathway relationships between candidate drivers. We have developed a novel strategy that now makes it possible to evaluate the functional impact of large numbers (100,000 per experiment) of pairwise genetic perturbations in mammalian cells. The resulting systematic genetic interaction maps provide critical insights into biological pathways and functional dependencies that can be exploited to rationally design polytherapies. We will focus on both TCGA targets and mechanisms described in aims 2 and 3 as well as explore novel targets and screening systems.

Relevance to the Cancer Target Discovery and Development (CTD^2) Network. As important as the cancer genome sequencing initiatives are, the identification and cataloguing of large numbers of variations is only the first step. An important goal is to ultimately understand how these variations contribute to cancer. Variations may produce loss- and gain-of-function and dominant negative effects, however it is impossible to distinguish these mechanisms based on sequence alone. Our program aims to fill this void and we have developed methodologies to uncover and distinguish cancer drivers and passengers, using a cutting-edge RNAi approach that allows rapid and quantitative exploration of huge numbers of perturbations individually and in combination. This provides a unique tool for the type of functional analysis that is central to the CTD^2 mission.

Value to the CTD^2 trans-Network mission. The activities described in this proposal are highly complementary to existing RNAi methods that are already being done in several labs. We are not simply using RNAi to interrogate drivers; instead, we are using an RNAi-based technology to understand how drivers interact in cancer relevant pathways. We have spent several years developing a unique set of powerful technologies that will dovetail into existing efforts. The synergy between our own expertise in mechanistic biochemistry and quantitative genetics together with the deep knowledge of cancer biology within this consortium places us in a superb position to provide high-value unique interactions to CTD^2 trans-Network investigators.