Neurofibromatosis type 1 (NF1) is a genetic disease that affects an estimated two million people worldwide. It is caused by mutations in a gene on human chromosome 17. Patients with this genetic defect are at increased risk of developing a long list of symptoms, including skin and skeletal abnormalities; learning disabilities; and both noncancerous and cancerous tumors, many of which arise from the peripheral nervous system. The NF1 protein functions as a negative regulator of the Ras oncogene. Thus, scientists believe most NF1 symptoms are caused by excessive Ras activity. However, Ras is a central player in many molecular pathways, and the main goal of the Bernards laboratory is to identify the specific pathways responsible for different NF1 symptoms. Among our most significant recent findings is the identification of the ALK growth factor receptor as a critical component of pathways responsible for learning and other defects in a fruit fly model of NF1 and our identification of a candidate genetic modifier of tumor development in human NF1.

Neurofibromatosis-1 (NF1) affects 1 in ~3000 people worldwide. Highly variable expressivity is among the hallmarks of NF1, with common symptoms including benign and malignant tumors, abnormal skin pigmentation, skeletal defects, and learning deficiencies. NF1 is caused by loss of neurofibromin, which functions as a GTPase activating protein for Ras and R-Ras paralogs. Our NF1-related research focuses on answering three questions: 1) what are the in vivo functions of neurofibromin; 2) to what extent does defective Ras regulation explain the diverse symptoms of NF1; and 3) what is the identity of modifier genes that affect NF1 symptom development?

Our longest-running attempt to answer these questions involves a genetic study of a 60% identical Drosophila NF1 ortholog. Homozygous dNf1-null mutants are viable and normally patterned but reduced in size. Mutants also have electrophysiological, circadian rhythm and learning/memory defects. The size and cognitive deficits resemble human NF1 symptoms. Neither defect is readily modified by manipulating Ras signaling, but both are restored by increasing—or mimicked by decreasing—signaling through the cAMP/PKA pathway.

While others have reported that dNf1 has physically separable Ras and cAMP related functions, our work identified a neuronal Ras signaling defect as the proximal cause of dNf1 phenotypes. To shed further light on how excess neuronal Ras/ERK signaling causes various cAMP/PKA-sensitive phenotypes, we performed affinity capture mass spectroscopic identification of dNf1 protein complexes, determined gene expression profiles of dNf1 expressing brain and nonexpressing peripheral tissues, performed metabolomic profiling, and analyzed the spatial requirement of PKA.
expression. We also screened 570 1st and 2nd chromosome deficiencies for dominant modifiers of the cAMP/PKA-sensitive dNF1 growth defect. Responsible modifier genes, identified though the use of mutant alleles or through RNAi-mediated knockdown, can be grouped into four functional categories. Several suppressors make up the first category of genes involved in Ras/ERK signal transduction, including the neuronal dAlk receptor tyrosine kinase and its activating ligand, jellybelly. Further collaborative work implicated dAlk as a rate-limiting upstream activator of dNF1-regulated Ras/ERK signals responsible for both growth and learning defects. Importantly, NF1-regulated ALK/RAS signaling appears conserved in man, identifying ALK as an attractive therapeutic target. A second category of modifiers consists of genes involved in cAMP/PKA signaling, whereas a third includes genes involved in regulating synaptic architecture or functioning. A strong suppressor in the latter category is the CCKLR-17D1 cAMP-coupled drosulfakinin receptor, recently implicated as a regulator of neuromuscular junction growth, which is abnormal in dNF1 mutants. Finally, our screen identified several genes whose role in dNF1-mediated organismal growth control remains to be determined. Within this last group, we are particularly interested in investigating roles for the adipokinetic hormone (AKH) receptor since AKH expression is strongly suppressed in dNF1 larval brain, resulting in severely reduced trehalose and triglyceride levels.

Other projects in our lab include work that has identified a candidate tumor burden modifier in a 1.4 Mb region surrounding the human NF1 gene, a study of phosphoregulatory mechanisms impinging on two prominent human RhoGAP proteins, an ongoing genome-wide association study of NF1 patients representing the top and bottom 15% of age-adjusted tumor burden, and the complete exome-sequencing of 79 of these patients. The latter two studies involve collaborations with the lab of Dr. James Gusella at Massachusetts General Hospital and with researchers at Genentech.

Selected Publications:


