



FINAL REPORT

Project Title: Therapeutically targeting aberrant chromatin remodeling in sarcomas

Project Number: SFA09-20

1. Date project was initiated: _7/2009_____
2. Period covered by this report: From _7/2009_____ To 12/2010_____
3. Publications, Abstracts, and Presentations:

- a. List all manuscripts submitted for publication during the period covered by this report resulting from this project. Include those in the categories of lay press, peer-reviewed scientific journals, invited articles, and abstracts. Each entry must include the author(s), article title, journal [book, editors(s), publisher, volume number, page number(s), and date.]

(1) Lay Press:

(2) Peer-Reviewed Scientific Journals:

Wang X, Sansam CG, Thom CS, Metzger D, Evans JA, Nguyen PTL and **Roberts CWM**. Oncogenesis caused by loss of the SNF5 tumor suppressor is dependent upon activity of BRG1, the ATPase of the SWI/SNF chromatin remodeling complex. *Cancer Research* 2009; 69: 8094-8101.

Wilson BG, Wang X, Shen X, McKenna ES, Lemieux ME, Cho YJ, Koellhoffer EC, Pomeroy SL, Orkin SH, **Roberts CWM**. Epigenetic antagonism between Polycomb and SWI/SNF complexes during oncogenic transformation. *Cancer Cell* 2010, Oct 19;18(4):316-28.

The paper below indirectly benefitted from the work funded by the SFA, although was not a direct result of the project:

Jagani Z, Mora-Blanco EL, Sansam CG, McKenna ES, Wilson B, Chen D, Klekota J, Tamayo P, Nguyen PTL, Tolstorukov M, Park PJ, Cho YJ, Hsiao K, Buonamici S, Pomeroy SL, Mesirov JP, Ruffner H, Bouwmeester T, Luchansky S, Murtie J, Kelleher J, Warmuth M, Sellers WR, **Roberts CWM***, and Dorsch M* (*Co-corresponding senior authors and contributed equally). Loss of the Tumor Suppressor Snf5 Leads to Aberrant Activation of the Hedgehog-Gli Pathway. *Nature Medicine* 2010; 16: 1374-6.

(3) Invited Articles:

Wilson BG and Roberts CWM. Nucleosome remodeling and cancer. *Nature Reviews Cancer*. This manuscript was written in response to an invitation from the editors at *Nature Reviews Cancer* and is currently under review.

(4) Abstracts:

- b. List presentations made during the last year (international, national, local societies, etc.). Use an asterisk (*) if presentation produced a manuscript.

- 2009 "The molecular basis of oncogenesis in rhabdoid tumors and implications for therapy," Children's Oncology Group/NCI Meeting for Malignant Rhabdoid Tumor and Anaplastic Wilms' Tumor, Bethesda, MD
- 2009 "Rhabdoid tumors: epigenetics, chromatin and cancer," Genetics and Biology of Childhood Cancer Symposium, Greehey Children's Cancer Research Institute, San Antonio, TX
- 2009 "Introduction to SMARCB1/INI1/SNF5 and the SWI/SNF complex" CNS Rhabdoid Tumors: Integrating Biological Insights with Clinical Success Symposium, Copley Hotel, Boston, MA
- 2009 "The molecular basis of oncogenesis in rhabdoid tumors: implications for therapy," CNS Rhabdoid Tumors: Integrating Biological Insights with Clinical Success Symposium, Copley Hotel, Boston, MA
- 2009 "SMARCB1/SNF5/INI1: epigenetics, chromatin and cancer," American Association of Cancer Research Annual Meeting, Denver, CO
- 2010 "Epigenetic antagonism between Polycomb and SWI/SNF complexes during oncogenic transformation," AACR Special Conference on Cancer Epigenetics, San Juan, Puerto Rico
- 2010 "The SWI/SNF Complex: Chromatin, Development and Cancer," Washington University Medical Scientist Training Program 40th Anniversary Celebration, St. Louis, MO
- 2010 "Aberrant Chromatin Remodeling and Epigenetic Dysregulation as Drivers of Oncogenic Transformation," University of North Carolina Lineberger Comprehensive Cancer Center's Weekly Seminar Series
- 2010 "SNF5 mutation in human cancer," FASEB Summer Research Conference on "Transcriptional Regulation During Cell Growth, Differentiation and Development", Snowmass, CO
- 2010 "The molecular basis of oncogenesis in Rhabdoid Tumors and Implications for Therapy", International Society of Pediatric Oncology Annual Meeting, Boston, MA
- 2011 "The SWI/SNF Complex: Chromatin, Epigenetics and Cancer," Institute for Cancer Genetics, Columbia University, Weekly Seminar Series

*Reports due by 31 July 2010

4. Provide a brief list of keywords: (limit to 20 words)

Rhabdoid, Swi/Snf, sarcoma, tumor suppressor, epigenetics, chromatin, chromatin remodeling

5. Summarize the progress during the period of this report and its impact on your plans for the remainder of the project. Include a summary of the progress toward the achievement of the originally stated aims and list the significant results:

We were highly successful during the period of the funding provided by the Sarcoma Foundation of America. Specific Aim 1 was completely accomplished and the results were published in the Wang et al paper cited above. The results of this work confirmed the hypothesis we proposed for this project. Briefly, our results demonstrated that cancer formation caused by mutation of SNF5 is dependent upon the activity of the residual BRG1-containing SWI/SNF complex. These findings suggest that, much like the concept of oncogene addiction, targeted inhibition of residual SWI/SNF activity may be an effective therapeutic approach for aggressive SNF5 deficient human tumors.

With respect to Specific Aim 2, we had proposed a screen for small molecule inhibitors of BRG1, the ATPase subunit of the SWI/SNF complex. Several things have appeared in the literature that have made us want to further define the mechanism by which the residual SWI/SNF complex is driving cancer prior to initiating the screen. BRG1 itself has now been reported to be frequently mutated in lung cancer cell lines, although perhaps not as frequently in primary lung cancers. In addition, two other SWI/SNF subunits (ARID1A and PBRM) have been found to be frequently mutated in ovarian, endometrial and renal cancers. Given that BRG1 itself may have tumor suppressor activity, we are in the process of evaluating activity of the residual SWI/SNF complex in each of these cancer types. Our hypothesis now is that in each of these cases a partially functional residual SWI/SNF complex is responsible for driving cancer formation. Via our current experiments, we aim to determine whether targeted inhibition of BRG1, the closely related mutually exclusive BRM subunit (which may be substituting for BRG1 when it is mutated), or another core subunit constitutes the most promising target, both for rhabdoid tumors as well as the other cancers.

In the process of pursuing this work, we further investigated the basis of cancers driven by the aberrant chromatin remodeling caused by SNF5 loss. We found that imbalanced epigenetic antagonism between the SWI/SNF complex and the Polycomb complex PRC2 is an essential mechanism underlying the rapid onset of cancer formation following SNF5 loss. This work revealed essential roles for epigenetic modifications during tumor formation and demonstrated that inactivation of EZH2 can have therapeutic efficacy against SNF5 mutant cancers in vivo. This work was published in the Wilson et al, paper.

Collectively, we have confirmed our hypothesis and have identified potential therapeutic targets. Going forward, we are working to elucidate the activity of the residual SWI/SNF complex when the tumor suppressor subunits are mutated, and are also pursuing therapeutic targeting of EZH2 for these cancers.

6. In layperson's terms, summarize the progress during the period of this report. Explain any medical significance or implications of your results to date:

Mutations in SNF5, a core subunit of the SWI/SNF complex, are present in the large majority of malignant rhabdoid tumors (MRT), a highly lethal sarcoma that occurs in kidney, brain, and soft tissues of young children. Inactivating mutations in SNF5 have also recently been found to occur in

a variety of other sarcomas including epithelioid sarcomas, small cell hepatoblastomas, undifferentiated sarcomas, chondrosarcomas. Despite the use of intensive therapies, the outcome for patients with these SNF5-mutant tumors has been dismal, with over 80% of patients dying of their disease, mostly within one year of diagnosis.

In order to develop targeted therapies for cancers in which SNF5 is mutated, it has not been clear whether a therapeutic goal should be to replace lost function of the SWI/SNF complex or whether to inhibit aberrant function of the residual SWI/SNF complex caused by SNF5 loss. We hypothesized that SNF5 loss does not equate to inactivation of the SWI/SNF complex but rather results in abnormal residual activity of the complex that promotes cancer. The experiments we performed via support from SFA confirmed this hypothesis. Further, we demonstrated that inactivation of BRG1, a core component of the residual SWI/SNF complex, completely blocks growth of SNF5 mutant cancers. This has established BRG1, or potentially other members of the SWI/SNF complex as a potential therapeutic target. In addition, based upon aberrant function of the residual complex, we have identified another therapeutic target that is activated by SNF5 loss.

Collectively, our work has identified two new targets that, when inhibited, block the growth of these lethal cancers. Several pharmaceutical companies are in the process of developing inhibitors to one of those targets, a protein called EZH2, and we are now collaborating with them to pursue this as a new potential therapy for patients with these cancers.

Principal Investigator (signature)

Date

Department Chair (signature)

Date

