Sloan / DOE Computational Molecular Biology Postdoctoral Fellowship

EVOLUTIONARY DYNAMICS OF CANCER: FINAL REPORT

CARLO C. MALEY

May 1, 2003

1. AIMS

We hypothesized that a subset of the mutations observed in the progression to cancer confer beneficial selective effects on the cell. Our aim was to identify these selective mutations and to infer the interactions between the mutant clones in Barrett's esophagus (BE) that eventually lead to the development of esophageal adenocarcinoma. The results were to be a set of predictions about the roles of specific mutations in the progression to cancer.

2. ACTIVITIES

My Sloan/DOE computational molecular biology postdoctoral fellowship began September 2000. This allowed me to make the transition from the department of Computer Science at the University of New Mexico, to a postdoctoral position at the Fred Hutchinson Cancer Research Center, working with Brian Reid and co-sponsored by Leonid Kruglyak. In April 2001, I received a K01 award from the NCI. With the approval of the DOE and the Sloan Foundation, we used the remaining funds from the first year of the grant to support Patricia Galipeau, the person in our lab most directly responsible for producing the data that I analyze. The second year's funding from the Sloan/DOE fellowship was relinquished with our gratitude for your generous support. Thus, this report covers our activities of that first year of funding.

2.1. **Data Generation.** During the three months spanning June through August 2001, one of Ms. Galipeau's responsibilities was to supervise the analysis of laboratory data from loss of heterozygosity (LOH) assays in Barrett's biopsies. Using a custom analysis program, over 35,000 genotypes were analyzed from 206 patients for whom the laboratory had previously assessed baseline LOH status. This represented the first attempts at evaluating LOH over time and has provided preliminary data about LOH incidence in early stage Barrett's esophagus. This dataset has been used to study clonal competition, LOH dynamics (interstitial loss vs terminal losses), and cell behavior before and after important genetic transitions during neoplastic progression in BE. I worked closely with Ms. Galipeau to

develop rules for an LOH analysis algorithm that would incorporate all of this data and summarize it for subsequent quality control.

2.2. **Selective Effects of Mutations.** Perl scripts were developed to analyze the frequency of loss of heterozygosity (LOH) lesions in each marker, in each patient of our 325 patient database. A large expanse of a lesion suggests a selective sweep of a clone. However, lesions in some loci may have expanded across the tumor simply because they co-occurred with an advantageous mutation in the same cell. These are called "hitchhikers." We developed scripts to separate the hitchhikers from selectively advantageous mutations. The results of this suggest that lesions in p16 (LOH, mutation or methylation) are selective both in the hemizygote and the null cells. Since the period of the grant we have been mainly focused on cleaning up the data so that it will be publishable. The paper is in preparation.

2.3. **Interactions.** During the period of the grant we also examined the data for signs of interactions between p16 and p53 lesions. It seems that p53 lesions do not expand in BE if they occur in p16 wildtype cells. Counterintuitively, there does not seem to be a strong selective effect of p53, even after the loss of p16. These initial results guided our work in the second year and we now have a paper in preparation on this interaction.

2.4. **Phylogenies.** In the first year, we developed algorithms to reconstruct the phylogenies of the clones for each patient in our database. Almost all other cancers allow only cross-sectional data to be collected. While phylogenetic reconstruction is designed for cross-sectional data, we realized that with longitudinal data, we could validate the inferences in the technique. This project was then put on hold until a full second time point of data could be generated and verified.

3. SUMMARY

The Sloan/DOE computational molecular biology postdoctoral fellowship funded the initial stages of my collaboration with the Reid lab. In doing so, it has launched my career and provided the foundation of a productive interaction between my quantitative approaches and the Reid lab's wonderful experimental system for understanding cancer progression. Our work has resulted in a set of predictions about the effects of mutations on the evolutionary dynamics of BE. We currently have 4 manuscripts in preparation and we anticipate that our basic work on the evolutionary dynamics of cancer in BE will have a significant impact on how the community approaches cancer biology and treatment over time.

I am deeply grateful for the support of the DOE and the Sloan Foundation.