136. A SCREEN FOR GENES SYNTHETICALLY LETHAL WITH *lin-35* Rb

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The class B synMuv gene *lin-35* encodes a C. elegans protein similar to the product of the Retinoblastoma gene (Rb), a tumor suppressor. Many human solid tumors contain mutations in Rb or in genes encoding proteins that regulate Rb. *lin-35* mutant animals provide an in vivo model for mammalian cells harboring mutant Rb genes. Since *lin-35* mutations are not lethal, we are screening for genes with functions required for viability in *lin-35* mutants but not in wild-type animals to identify potential targets for cancer therapy. Such targets, if inactivated pharmacologically, could cause the specific death of Rb-deficient cells.

We are using the chromosome I RNAi library described by Fraser et al. (2000)[1] to screen for genes that are essential specifically in *lin-35(n745)* animals. *lin-35(n745)* contains an early stop codon and is considered a null allele[2]. We are comparing the phenotypes following RNAi of *lin-35(n745)* animals to the published results for N2[1] . At this point, we have screened 50% of chromosome I (1309 genes). We have seen severe phenotypes (embryonic lethality, sterility, larval arrest, larval lethality or severe growth delay) for 244 (18.6%) genes. Of those, 162 (12.4%) appeared to have the same or very similar phenotypes for *lin-35* and N2 animals. 82 (6.3%) of those tested apparently had severe phenotypes in *lin-35* but not in N2 animals, while 23 (1.8%) apparently had severe phenotypes in N2 but not in *lin-35* animals. Some of the RNAis of genes that caused severe phenotypes in *lin-35* animals but not N2 animals have been retested to confirm both the *lin-35* and N2 phenotypes. Of the 22 retested, 17 continued to display more severe phenotypes for *lin-35(n745)* animals. These genes do not fit into obvious classes based on homology. Extrapolating from the current data set, we expect to find 75-150 genes on chromosome I the RNAi of which are synthetically lethal with the *lin-35(n745)*) mutation.

Because *lin-35(n745)* animals are less healthy than N2 animals (decreased brood size and rare sterile animals[3,4]), it is possible that some of the synthetic phenotypes seen in *lin-35* mutants but not in N2 animals are caused by the non-specific additive effects of two harmful mutations. Similarly, RNAi of some genes may affect one cell type and the *lin-35* mutation another so that together these two distinct defects result in severely affected animals. To identify genes the RNAi of which are cell autonomously synthetically lethal with *lin-35*, I am developing an assay to assess the effect of inactivation of *lin-35* in combination with inactivation of any of the genes that are identified in the primary screen within a single tissue.

1Fraser, A. G., Kamath, R. S., Zipperlen, P., Martinez-Campos, M., Sohrmann, M., and J. Ahringer. (2000). Nature 408: 325. 2Lu, X., and H. R. Horvitz. (1998). Cell 95: 981. 3Fay, D. S., Keenan, S., and M. Han. (2002). Genes and Development 16: 503. 4M. Hurwitz and H. R. Horvitz, unpublished results.