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.