The *mir-35* Family of MicroRNAs Acts Redundantly in Embryonic Development in *C. elegans*. *Ezequiel A. Alvarez-Saavedra[1], Eric A. Miska[1,2], Allison L. Abbott[3], Nelson C. Lau[1,4], David P. Bartel[1,4], Victor Ambros[3], Bob Horvitz[1].* 1) Dept Biol, HHMI, MIT, Cambridge, MA; 2) Wellcome Trust/Cancer Research UK Gurdon Institute, Cambridge, UK; 3) Dept. Genetics, Dartmouth Medical School, Hanover, NH; 4) Whitehead Institute for Biomedical Research, MIT, Cambridge, MA.

As part of an effort to obtain deletion alleles of all microRNA genes in *C. elegans* (see abstract by Miska *et al.*), we have identified mutants with deletions in the *mir-35* family, which consists of eight microRNA genes closely related in sequence. *mir-35* through *mir-41* are located within a 1 kb genomic cluster, while *mir-42* is in a separate genomic location also on LGII.

The seven microRNAs of the *mir-35* cluster appear to be expressed only during embryogenesis as assayed by northern blots and a GFP reporter. A deletion that removes the seven microRNAs in the *mir-35* cluster results in a temperature-sensitive late embryonic or L1 lethal phenotype, while deletions that remove *mir-37* through *mir-41*, *mir-42* alone, or *mir-37* through *mir-42* do not cause lethality. Thus, expression of just *mir-35* and/or *mir-36* is sufficient for normal development, indicating either that *mir-35* and/or *mir-36* are the only microRNA(s) in this family required for development or that they function redundantly with the other members of the family. Rescue experiments show that any single member of the *mir-35* family can rescue the embryonic lethality, suggesting that all members of the family act redundantly.

Animals lacking all eight *mir-35* family microRNAs die by the two- to three-fold stage of embryogenesis with apparent defects in elongation at the two-fold stage. The embryonic lethal phenotype is fully penetrant and independent of temperature. The embryonic lethality can be rescued by maternal or zygotic contribution of the endogenous microRNAs, indicating that their function is not required until after zygotic transcription has begun.

We have attempted to identify direct targets of this family using a combination of bioinformatics and RNAi without success so far. We are currently conducting a screen for suppressors of the embryonic lethality to seek targets of this family of microRNAs.