

## Cloning and Characterization of Suppressors of the Synthetic Multivulva Phenotype

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The synthetic Multivulva (synMuv) genes are grouped into at least three functionally redundant classes, A, B, and C, that negatively regulate the specification of vulval cell fates. Animals mutant for one or more genes within the same class are non-Muv. Animals mutant for genes within any two classes are Muv. Some of the identified class B gene products are homologs of members of at least two transcriptional repression complexes, and some of the identified class C gene products are homologs of a putative transcriptional activation complex.

To identify loci that interact genetically with the synMuv genes, we performed two screens for synMuv suppressors. From these screens, we isolated 166 suppressors of the synMuv phenotype of *lin-15AB(n765ts)* animals and 43 suppressors of the synMuv phenotype of *lin-53(n833); lin-15A(n767)* animals. The synMuv suppressor *isw-1*, a homolog of the chromatin-remodeling ATPase ISWI, may act with the genes encoding homologs of the *Drosophila* NURF complex to antagonize the synMuv phenotype, because loss of the NURF301 and NURF38 homologs suppresses the synMuv phenotype of *lin-15AB(n765ts)* animals.

In our characterization of histone methyltransferase genes (see abstract by Andersen and Horvitz), we found that *mes-2* and *mes-4* are synMuv suppressors. Additionally, loss-of-function mutations in *mes-3* and *mes-6* but not *mes-1* suppress the synMuv phenotype. MES-2 and MES-6 are homologs of the Polycomb proteins E(z) and ESC, respectively, and are predicted to be histone methyltransferases. The Strome laboratory has shown that MES-2, MES-3, and MES-6 form a complex in embryos, methylate histone H3 lysine 27, and may play a role in the localization of MES-4. We are currently investigating how histone methylation can influence the synMuv phenotype and whether the predicted NURF complex homologs and the *mes* genes interact.

We identified *Ist-3(n2070)* as a dominant suppressor of the synMuv phenotype of *lin-15AB(n765ts)* mutants. The Greenwald laboratory has implicated *Ist-3* through bioinformatics as a putative target gene of LAG-1, a *lin-12* and *glp-1* Notch effector, and postulated that it may mediate the interplay between the RTK/Ras and Notch signal transduction pathways in vulval development. The dominant phenotype conferred by *Ist-3(n2070)* affects the putative DNA-binding domain and may increase wild-type function. We isolated several nonsense *cis*-dominant suppressor alleles and a deletion allele to study loss of *Ist-3* function. Putative null mutations of *Ist-3* confer a synMuv phenotype in combination with mutations in class A but not class B synMuv genes. These gain-of-function and loss-of-function alleles of a negative regulator of vulval development may allow for a genetic analysis of the interplay between the RTK/Ras, Notch, and synMuv pathways.

Poster

Cell-fate specification: Post-embryonic

Gene Expression

Keyword: Development: the vulva