Keeping the Germline Silent


A common feature of germ cell development, observed in animals as diverse as Caenorhabditis elegans and Drosophila, is widespread silencing of somatic gene transcription in germ cell precursors. Researchers have now identified a factor that seems to mediate such silencing in the ascidian Ciona intestinalis.

Maki Shirae-Kurabayashi et al. took a close look at Ci-pem-1, an RNA component of the C. intestinalis germ plasm. Germ plasm is found in the cytoplasm of embryonic germ cells in many animals, from mice to worms. For instance, in nematodes germ plasm is segregated asymmetrically during blastomere divisions into the cells that give rise to germ cells, dubbed germine blastomeres, and contains factors essential for germline development.

The researchers found that the Ci-Pem-1 protein is highly expressed in the nucleus of germline blastomeres and that it appears to silence transcription (see accompanying figure). For instance, in embryos lacking Ci-Pem-1, several zygotic RNAs that are normally expressed only in somatic cells were also expressed in germline blastomeres. Ci-Pem-1 may operate through binding of C. intestinalis homologs of Groucho, a known transcriptional repressor. Ci-Pem-1 protein contains a motif known to interact with Groucho, and it bound to two ascidian homologs in immunoprecipitation experiments.

The researchers also showed that germline blastomeres in C. intestinalis contained low levels of phosphorylated RNA polymerase II, suggestive of low levels of transcription. A question for the future is whether embryos depleted of Ci-Pem-1 have altered levels of phosphorylated RNA polymerase II in the germline blastomeres.

In other animals distinct factors keep transcription quiet in the embryonic germline. C. elegans, for instance, deploys the protein PIE-1 in germline blastomeres, and in Drosophila silencing is achieved through Pgc, a small 71 amino acid protein that directly interferes with phosphorylation of RNA polymerase II in pole cells, the germ cell progenitors. In mice, some, but not all, somatic transcription is silenced during primordial germ cell specification—this silencing is mediated by the PR domain transcriptional inhibitor Blimp1.

Female Germ Cells Guide Their Own Fate with Sex Lethal


In Drosophila and mice, determination of the sex of the germline—whether it gives rise to sperm or eggs—occurs both through cell-autonomous cues and through interactions between primordial germ cells (PGCs) and the soma. Researchers have now found that, in Drosophila, the gene Sex lethal (Sxl) acts within female PGCs to tell them that they are female.

Sxl is known to operate in the soma of XX embryos. It encodes an RNA binding protein, involved in alternative splicing and translation, and its activity results in a female-specific version of Dsx (Double sex) protein which promotes female sexual development. In males, Sxl is not turned on and, as a consequence, a male-specific form of Dsx is produced.

Kazuya Hashiyama et al. now find that Sxl operates not only in the soma, but also cell-autonomously in the germline of female fruit flies. They observed that Sxl was expressed in XX, but not XY, PGCs during their migration to the gonads. This expression was necessary for PGCs to assume a female fate. Moreover, ectopic expression of Sxl in XY PGCs prompted them to enter oogenesis and produce functional eggs when transplanted into an XX host.

Previous studies have found that the female soma also seems to provide signals to promote a female fate of PGCs. For instance, researchers have transplanted XY PGCs into the female soma and found that the PGCs exhibit a female expression profile, but they do not develop into eggs.

Sxl does not appear to operate in germline sex specification in mice, where the cell-autonomous cues involved in the process are still not clear.

Stem Cells Spurred on by MicroRNA During Sperm Production


Ci-Pem-1 protein (green) localizes to the nuclei of the germline blastomeres in the ascidian C. intestinalis. Germ plasm stains intense green. F actin in purple, DNA in blue. Photo credit: Maki Shirae-Kurabayashi.
A large-scale analysis has homed in on a microRNA that seems to keep the ‘stem’ in spermatogonial stem cells (SSCs).

Previous studies have shown that microRNAs regulate the progression of spermatogenesis. For instance, the loss of Dicer, a molecule involved in processing microRNAs, perturbs germ cell development and leads to infertility. Now, Zhiyv Niu et al. have examined microRNAs specifically in the SSC population and explored the function of an individual microRNA, MicroRNA-21 (miR-21).

The researchers compared the expression of microRNAs in the Thy1^+ testis cell population, which is highly enriched for spermatogonial stem cells, with the expression of miRNAs in the Thy1^- population, which is mostly somatic. They found several microRNAs that were preferentially expressed in Thy1^+ cells—these microRNAs were also expressed at high levels in a cell culture consisting of SSC-enriched germ cells, underscoring the value of this in vitro system.

The researchers then dissected the function of one of the highly-expressed microRNAs, miR-21. This microRNA is known to counteract apoptosis, and acts as an oncogene in some biological systems. In SSC-enriched germ cell cultures, transiently inhibiting miR-21 increased the number of apoptotic cells and decreased the number of stem cells.

The renewal and maintenance of the SSC population relies on GDNF (glial cell line-derived neurotrophic factor), that operates through the ETS transcription factor ETV5, among other molecules. ETV5, in turn, seems to regulate miR-21: ETV5 was shown to bind the miR-21 enhancer in chromatin immunoprecipitation experiments, and overexpression of ETV5 in cultured germ cells bumped up expression of miR-21. Eight of the ten most abundant miRNAs expressed in the germ cell cultures, including miR-21, have conserved regulatory binding sites for ETS transcription factors.

How miR-21 might interact with other factors, such as genes required for stem cell self-renewal or differentiation, remains an open question.

Second-Hand Smoke Smokes Sperm


Passive exposure to cigarette smoke induces mutations in sperm DNA, according to a recent study in mice. What’s more, the standard test for somatic cell mutations does not reveal this effect, highlighting the limitations of conventional safety measurements.

To assess genomic instability in response to smoke, Marchetti et al. examined the induction of mutations in expanded single tandem repeat regions (ESTR), which are especially vulnerable to damage. Previous studies have found that mutations in these repeat regions occur in response to chemical mutagens, particulate air pollution, and ionizing radiation. In mice, mainstream smoke (the smoke breathed by cigarette users) similarly induces such ESTR mutations in the germline and these mutations are transmitted to progeny. This is consistent with observations that men who smoke are at risk for abnormalities in their sperm and that their offspring are at increased risk of developing cancer.

To assess the effect of sidestream smoke (the smoke that emerges from the tip of a cigarette), the researchers exposed mice to the equivalent of 3 or 16 cigarettes per day for two weeks. The researchers examined the induction of micronuclei in the red blood cells of exposed mice, a test widely used by regulators to assess chemical safety. Whereas mainstream smoke resulted in the formation of micronuclei, sidestream smoke did not—bringing into question the value of this assay in assessing the ability of an agent to affect the germline.

More importantly, the mutation rate in response to sidestream smoke was similar to that of mainstream smoke—about 4.6 and 4 percent, respectively, in response to the 3-cigarette regimen. For comparison, the ESTR mutation frequency in control (sham-exposed) mice was about 1.5 percent. These findings raise new concerns about the impact of sidestream smoke on the genetic makeup of individuals who are regularly exposed to smokers.