RNAi restores methylation

Mutation of *Arabidopsis MET1* (encoding methyltransferase) or *DDM1* (encoding ATPase chromatin remodeler) leads to >70% loss of cytosine methylation that persists in F_1 progeny despite the recessive action of *ddm1* and *met1* mutations. Teixeira *et al.* (*Science*, published online 30 January 2009; doi:10.1126/science.1165313) examined the stability of this hypomethylation in methylation-proficient plants five generations after demethylation events, using methylation-sensitive restriction enzyme digestion and quantitative PCR. Within a 500-kb heterochromatic 'knob' of chromosome 4, a consistent pattern of repeat sequences that retained hypomethylation was found interspersed with sequences that restored wild-type methylation, with little variation between lines. The authors found no parent-of-origin effect on remethylation. *ddm1rdr2* plants were then crossed to *rdr2* (lacking RNA-dependent RNA polymerase RDR2) plants to demonstrate RNAi dependence of progressive restoration of cytosine methylation. In this RNAi-compromised background, remethylation was sporadic and not progressive. Teixeira *et al.* conclude that there are three mechanisms of repeat silencing in the plant: those silenced by MTases alone that cannot subsequently become remethylated; remethylatable sequences that are targeted by both MTases and RNAi; and targets of RNAi-dependent *de novo* methylation that do not become hypomethylated as a result of the *ddm1* and *met1* mutations. *MA*

SYNGAP1 and mental retardation

Mutations at autosomal loci account for most cases of nonsyndromic mental retardation, but pinpointing of the underlying molecular lesions has been hampered by the limited availability of families suitable for linkage analysis. To circumvent this problem, Jacques Michaud and colleagues (*N. Engl. J. Med.* **360**, 599–605; 2009) used a candidate-gene resequencing strategy in 94 subjects with unexplained nonsyndromic mental retardation. Reasoning that defects in synaptic function might underlie this phenotype, the authors sequenced *SYNGAP1*, which encodes a brain-specific protein required for normal NMDA receptor signaling, and found *de novo*, heterozygous truncating mutations in three individuals. Shared phenotypes included moderate to severe mental retardation and severe language impairment. Two of the subjects also had mild epilepsy. Parallel sequencing in 142 subjects with autism-spectrum disorders, 143 subjects with schizophrenia and 190 controls revealed no similar mutations, suggesting that truncating *SYNGAP1* mutations are not commonly associated with a broader spectrum of neurological or behavioral phenotypes. Analyses in larger cohorts will be needed to establish the prevalence of *SYNGAP1* mutations among individuals with nonsyndromic mental retardation. These results should also catalyze efforts to examine other components of the NMDA-receptor complex as potential causes of unexplained mental retardation. *KV*

Footprints under water

Metagenomics studies sampling from heterogeneous environments have allowed researchers to address fundamental questions of microbial diversity and evolution but have also demonstrated the need for new methods to cope with the scale of these datasets. Mark Gerstein, Peer Bork and colleagues now report a new method to analyze metagenomics datasets and characterize how microbes adapt to different environments (*Proc. Natl. Acad. Sci. USA* **106**, 1374–1379; 2009). The method is based on canonical correlation analysis of pathways associated with combinations of defined environmental variables, termed a metabolic footprint. The authors applied this method to the Global Ocean Survey (GOS), which includes metagenomic sequences from >40 aquatic sites and quantitative environmental features. They weighted pathways, modules and operons with maximal variation to a combination of environmental variables as defined across the aquatic sites and identified metabolic footprints specific to environments or environmental factors. Comparing coastal and open sites, they found significant differences in the usage of pathways in metabolite biosynthesis, lipid transport and metabolism, amino acid metabolism and energy production. Notably, they found correlation between energy-conversion strategies and environmental variables such as temperature gradient. Such a method could be used to predict environmental features from a metabolic footprint, as well as the use of particular pathways in a given environment. *OB*

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Hirschsprung's disease modifier

Hirschsprung's disease, or aganglionic megacolon, is caused by the failure of neural crest cells to populate the hindgut during development, resulting in the absence of enteric nerves from a segment of the intestinal tract. Mutations in *RET*, encoding a key regulator of enteric nervous system development, account for a substantial fraction of familial and sporadic cases, and common variants near *RET* also act as low-penetrance susceptibility alleles. To identify other variants influencing disease risk, Paul Tam and colleagues (*Proc. Natl. Acad. Sci. USA*, published online 5 February 2009; doi:10.1073/pnas.0809630105) performed a genome-wide association study of Chinese individuals with sporadic Hirschsprung's disease and followed up the most promising candidate SNPs in a second cohort drawn from the same population. In addition to replicating known association with common variants in *RET*, they identified two variants in *NRG1* associated with disease risk at a genome-wide level of significance. Notably, one of these variants showed a statistically significant interaction with the common *RET* homozygous risk genotype, identifying this allele as a modifier of disease penetrance. The authors propose that *NRG1* variants might modify disease risk by affecting expression of the major disease gene *RET*. *KV*

Black fur coat

Natural pigment variation is controlled by the agouti–melanocortin 1 receptor (Mc1r) pathway, which modulates the amount and type of pigment produced by melanocytes. In addition, the *K* locus, which encodes a β-defensin protein that is also a component of the Mc1r pathway, has been found to promote pigment-type switching in domesticated dogs. Anderson *et al.* (*Science*, published online 5 February 2009; doi:10.1126/science.1165448) now show that a 3-bp deletion of *K* suppresses *agouti* action and triggers melanism in the gray wolf, producing a dominant black coat. Black wolves are extremely rare in the tundra, and the authors show that the frequency of the melanistic *K* allele corresponds to phenotypic frequencies of 2–33% and 33–64% for black wolves in the tundra and forests, respectively. Evaluation of the haplotype structure surrounding the *K* locus, in combination with coat color, habitat and gene frequency, revealed very low diversity for the melanistic *K* genotype, suggesting that it has been under positive selection in forest wolves. Moreover, estimated coalescent times to the most recent common ancestor indicated that the melanistic *K* allele derives from past hybridization with domestic dogs. Black coats confer a predatory advantage, and as tundra habitats decline, the frequency of the melanistic mutation may increase as a means of adaptation to a changing environment. *LK*