

DNA methylation gets dynamic

Two recent papers mark a fundamental shift in our thinking about how DNA methylation affects gene expression in metazoans. In contrast to the view of an epigenetic mark that is associated with transcriptionally silent chromatin, the new studies reveal that cycles of DNA methylation are involved in transcriptional activation.

The ligand-dependent transcription factor estrogen receptor α (ER α) induces cyclical activation of its target promoters by inducing the recruitment of a series of transcription-factor complexes. Cycles are characterized by waves of transcription that are followed by repressive processes that limit the transcriptional response.

The two new studies report that DNA methylation shows a similar cyclical pattern at a set of promoters. Kangaspeska, Stride and colleagues used the methyl-binding domain



of MeCP2 — a protein that binds methylated CpG dinucleotides (the main sites of DNA methylation in mammals) — to isolate and quantify methylated DNA from the promoter of one ER α -responsive gene, *pS2*. Before release from transcriptional blockade with the DNA interchelator doxorubicin, the gene is unmethylated, but transcriptional activation following washout of doxorubicin results in cyclical methylation of the promoter. DNA methylation occurs simultaneously with the initial cycle of transcription, and again after the second cycle of productive transcription. The authors went on to show that similar cyclical DNA methylation occurs at four other genes.

In the second paper, Métivier and colleagues also show cyclical DNA methylation at several CpG sites in the *pS2* promoter, in this case using bisulphite sequencing and methylation-sensitive restriction of DNA. These experiments also revealed that some of the cyclical methylation is strand specific, and that only the transcribed strand is demethylated after the first cycle of transcription.

The same group investigated how cyclical DNA methylation is brought about. The authors showed that DNA methyltransferases (DNMTs), which catalyse the addition of methyl groups at CpGs, are present at the active promoter. Their recruitment is associated with both phases of DNA methylation and demethylation. This observation, together with the use of a DNMT inhibitor, suggested that DNMTs are involved in both the addition and removal of methyl groups — a function that was

previously unreported in mammals. The authors then provide a mechanism for this activity, showing that DNMT3a and DNMT3b deaminate methylated CpGs, thereby generating mismatches that are cleaved by a glycosylase and repaired by machineries that are involved in the base-excision pathway of DNA repair (BER).

Both groups provide evidence that the cyclical addition and removal of methyl groups from DNA is important for the normal expression of *pS2*. Métivier and colleagues further showed that this activity is essential for the sequential recruitment of chromatin remodellers and other proteins that are involved in cycles of transcriptional activation at the promoter. Meanwhile, Kangaspeska, Stride and colleagues show that stimulating cyclical DNA methylation in cells in which *pS2* expression is epigenetically silenced results in low-level expression of this gene.

The rapid cycling of DNA methylation that is reported in these studies is a previously unrecognized aspect of gene regulation by what was considered a purely epigenetic mark. Together with the surprising finding that DNMTs have roles in both the addition and removal of methyl groups, this insight promises to open up new areas of research into the role of DNA methylation in metazoans.

Louisa Flintoft

ORIGINAL RESEARCH PAPERS

Métivier, R. *et al.* Cyclical DNA methylation of a transcriptionally active promoter. *Nature* **452**, 45–50 (2008) | Kangaspeska, S. & Stride, B. *et al.* Transient cyclical methylation of promoter DNA. *Nature* **452**, 112–115 (2008)