Epigenome Editing

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Recent publications

- Targeted DNA demethylation and activation of endogenous genes using programmable TALE-TET1 fusion proteins http://www.nature.com/nbt/journal/v31/n12/full/nbt.2726.html
- Locus-specific editing of histone modifications at endogenous enhancers http://www.nature.com/nbt/journal/v31/n12/full/nbt.2701.html
- Optical control of mammalian endogenous transcription and epigenetic states http://www.nature.com/nature/journal/v500/n7463/abs/nature1246

TALE

- All three groups achieve this long-awaited goal using customizable Transcription Activator Like Effector (TALE) proteins - previously applied to editing the genome - to recruit chromatin-modifying enzymes to specific loci
- In plants: they recognize plant DNA sequences through a central repeat domain consisting of a variable number of ~34 amino acid repeats. There appears to be a one-to-one correspondence between the identity of two critical amino acids in each repeat and each DNA base in the target sequence
- Data on wikipedia outdated

Target sequence of interest

- Genome editing technology has relied on a handful of DNA-binding proteins, primarily TALEs and zinc finger proteins, that can be engineered to target sequences of interest
- Expanding the scope of site-specific recombinases for genetic and metabolic engineering http://onlinelibrary.wiley.com/doi/10.1002/bit.25096/full

Targeting

- Fusing TALEs to chromatin modifiers, the new studies (on previous slides) are the first to use TALEs for locus-specific editing of the epigenome rather than the genome
- Recently, an even more tractable system has been developed based on a bacterial Cas9 nuclease in complex with a guide RNA that recruits the nuclease to complementary DNA sequences http://www.nature.com/nmeth/journal/v10/n8/abs/nmeth.

Applications

- Potential enhancers locations
- Enhancers are marked by histone H3 lysine 4 mono- and dimethylation (H3K4me1 and H3K4me2) and H3K27 acetylation (H3K27ac).
- Influence of epigenome modifications on transcription

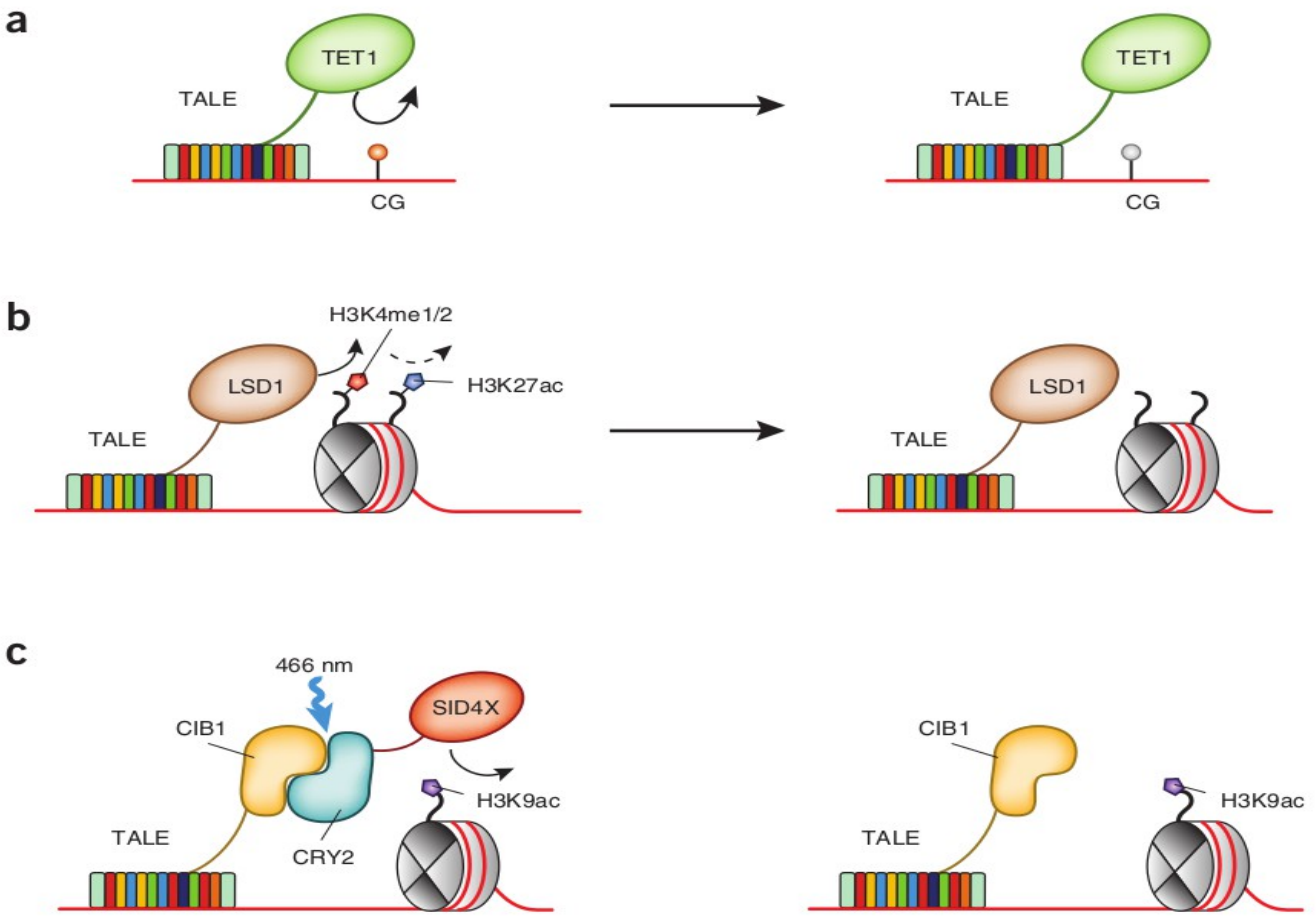

Figure 1 Site-specific recruitment of chromatin modifiers for epigenome editing. (**a**) Maeder *et al.*[1] fused TALEs to the catalytic domain of TET1 to induce demethylation of methylated cytosine (5mC) at CpG sites (shown as CG). A methylated cytosine is represented by a filled circle and an unmethylated cytosine by an open circle. This approach can be used to activate genes repressed by DNA methylation and also enables identification of functional CpG methylation events. (**b**) Mendenhall *et al.*[2] used TALE fusions to target the H3K4me1/2 demethylase LSD1 to putative enhancers. This induced demethylation (removal of red shape) as well as H3K27 deacetylation (an indirect effect, shown by dashed arrow) at target sites. For some putative active enhancers, changes in expression of neighboring genes were observed. (**c**) Konermann *et al.*[3] used TALEs to direct chromatin-modifying activities that repress gene expression, in a manner controlled by blue light (466 nm). The TALE is fused to a light-sensitive cryptochrome (CIB1), and the chromatin modifier is fused to an interaction partner of the cryptochrome (CRY2). In the example shown, a repressor domain known as SID4X directs the removal of H3K9 acetylation (H3K9ac) in the presence of blue light. In the absence of blue light (right panel), CIB1 and CRY2 do not associate.

Challenges

- Once an epigenome modifier is recruited to its proper target, depending on the design of the fusion protein, the catalytic activity of the fused chromatin modifier could be compromised
- Especially challenging in the context of multisubunit complexes such as Polycomb repressive complexes
- Access to the substrate could be occluded by other factors already bound to chromatin at the target site
- Conversely, binding sites for transcription factors or other chromatin modifiers necessary for gene activation could be blocked by the targeted TALE
- Endogenous systems that reverse the chromatin alterations could prevent the desired epigenetic changes

Questions

- Little is known about how H3K27 trimethylation and H2A monoubiquitination mediate transcriptional repression
- Can introduction of H3K4me3 at a promoter activate transcription?Will a
- modification that is introduced by an inducible system before S phase survive passage of the replication fork and be transmitted to the daughter cells even after removal of the signal that established the mark?

References

- http://scholar.google.ru/ as trigger for new articles of the particular author
- http://www.nature.com/nbt/journal/v31/n12/full/nbt.275