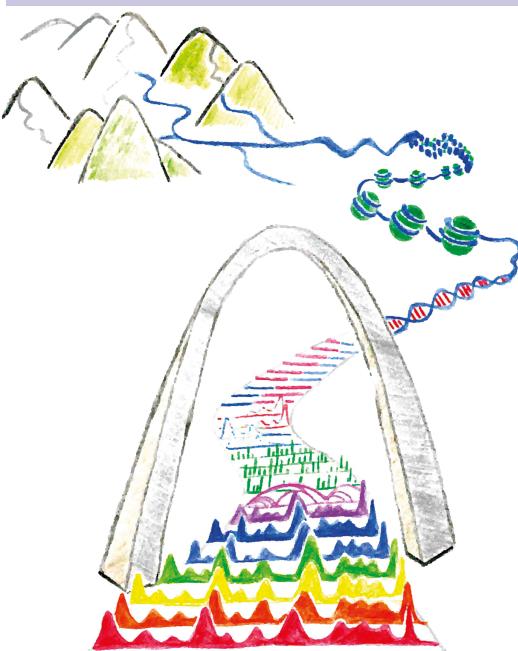


系统生物学与生物信息学
海外学者短期讲学系列课程

Current Topics in Epigenomics

表观基因组学前沿



Ting Wang
Department of Genetics
Center for Genome Sciences and Systems Biology
Washington University School of Medicine

Tsinghua University
April 15-27

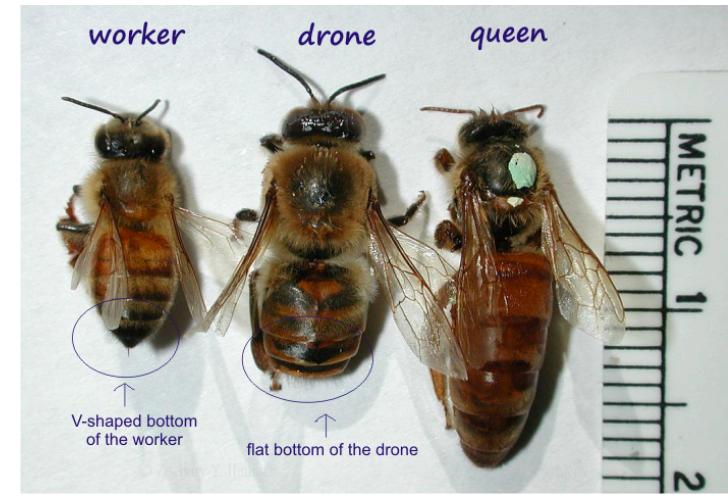
Epigenome conservation

Review of epigenome features

- What is epigenetics?
 - “Relating to, being, or involving changes in gene function that do not involve changes in DNA sequence” –Merriam-Webster
 - “The study of mechanism of temporal and spatial control of gene activity during the development of complex organisms” –Robin Holliday



Same genome but
different phenotype

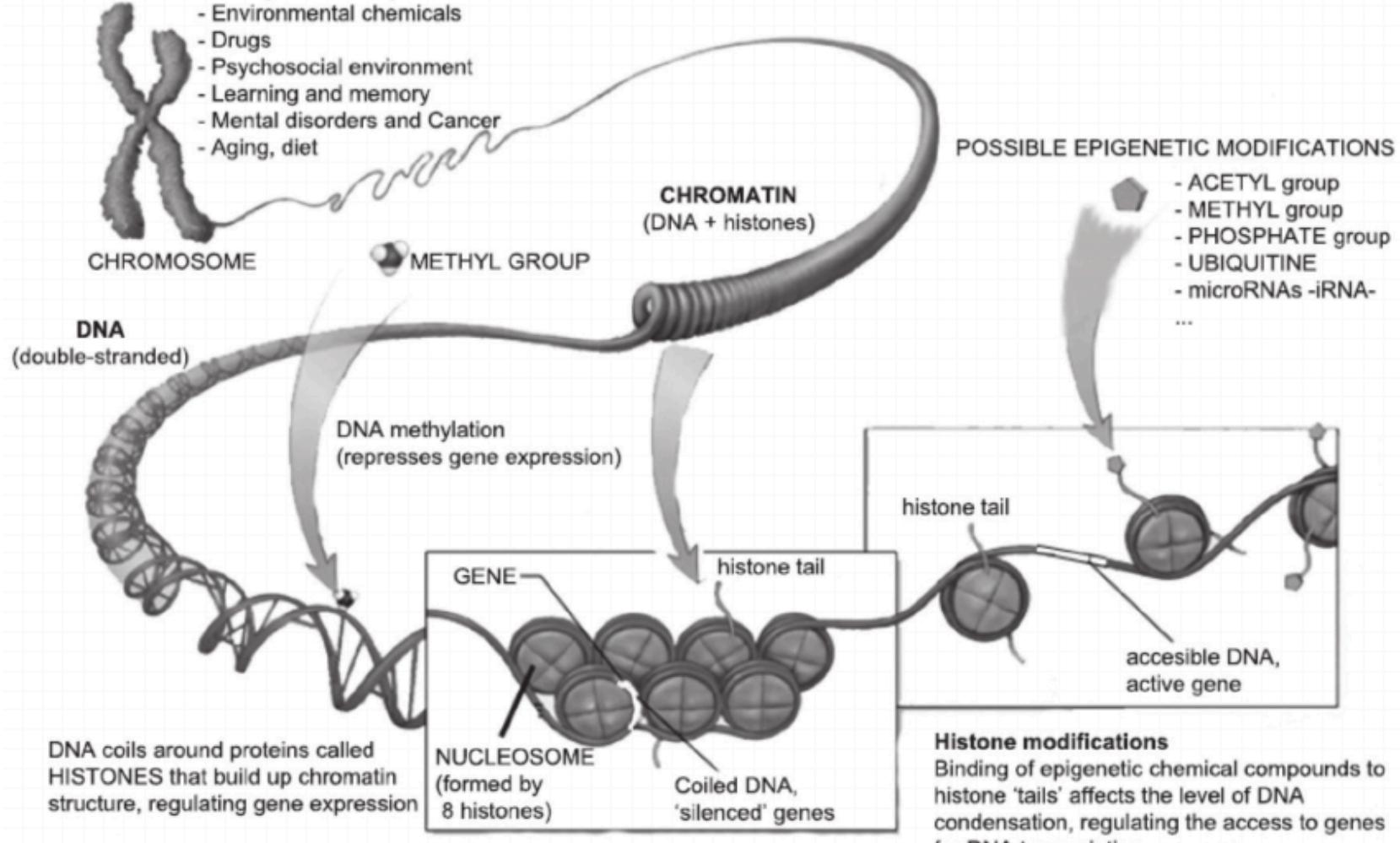


Chromatin structure

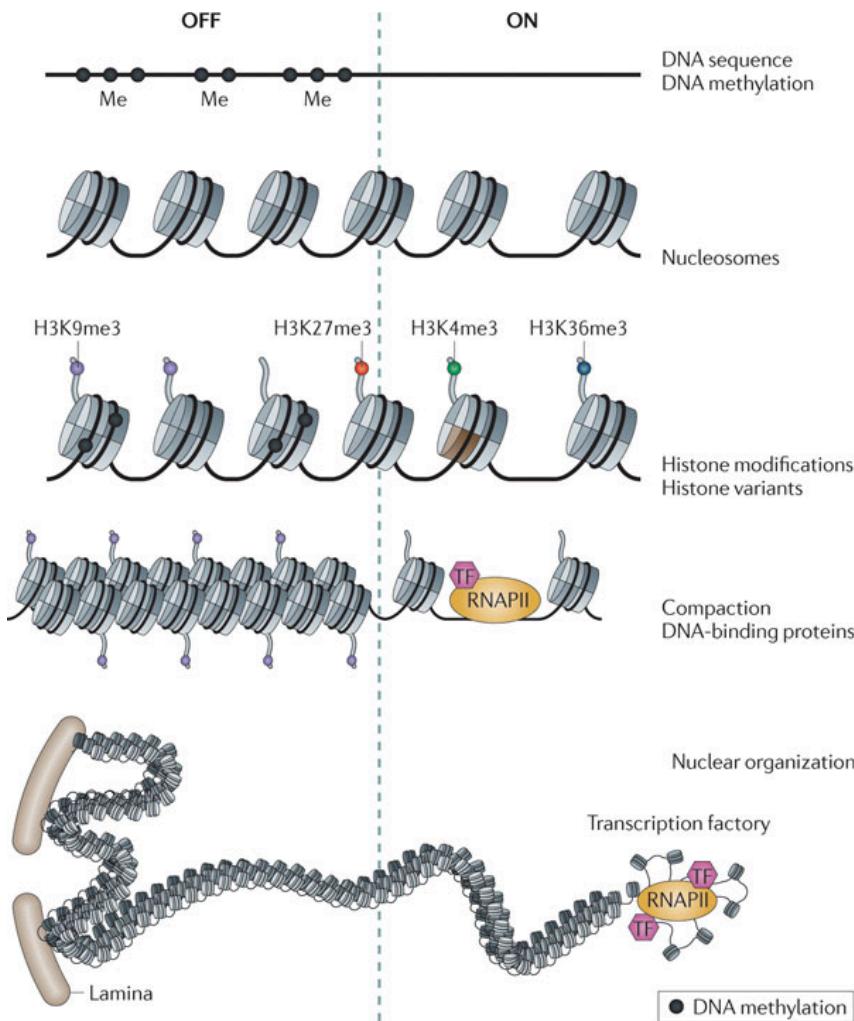
EPIGENETIC MECHANISMS

Factors and processes involved :

- Embryonic development
- Environmental chemicals
- Drugs
- Psychosocial environment
- Learning and memory
- Mental disorders and Cancer
- Aging, diet

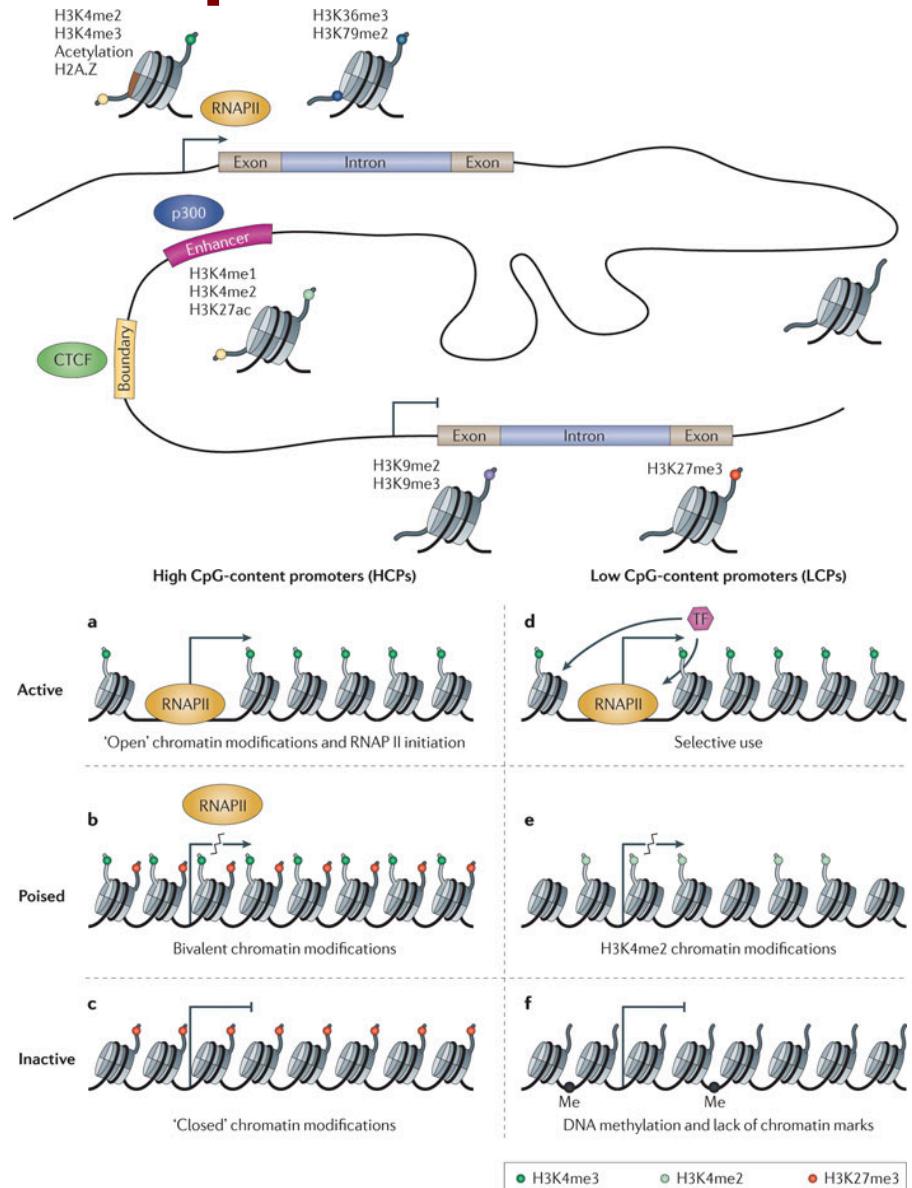


Chromatin structures' impact on function



Nature Reviews | Genetics

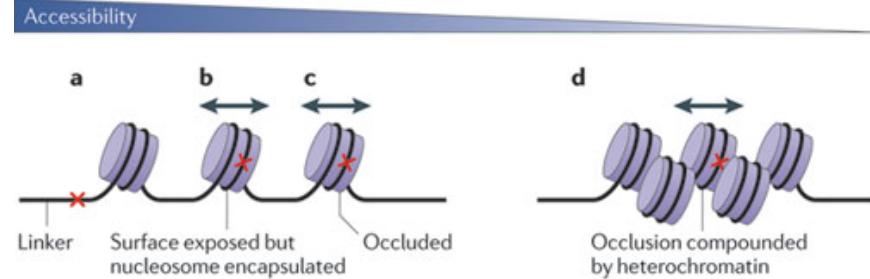
Zhou et al. 2011



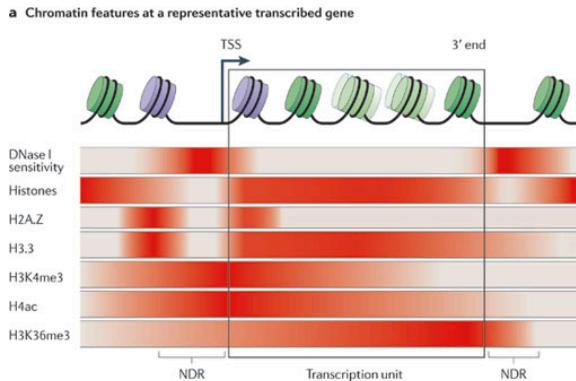
Nature Reviews | Genetics

Chromatin Organization's impact on function

Nucleosome positioning



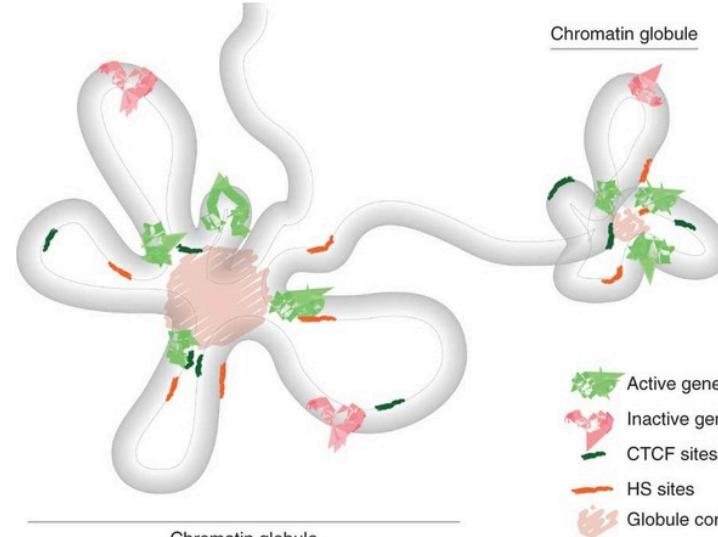
Nature Reviews | Genetics



Bell et al. 2011

Nature Reviews | Genetics

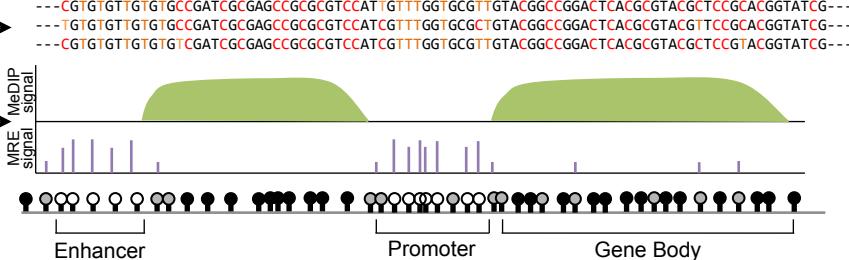
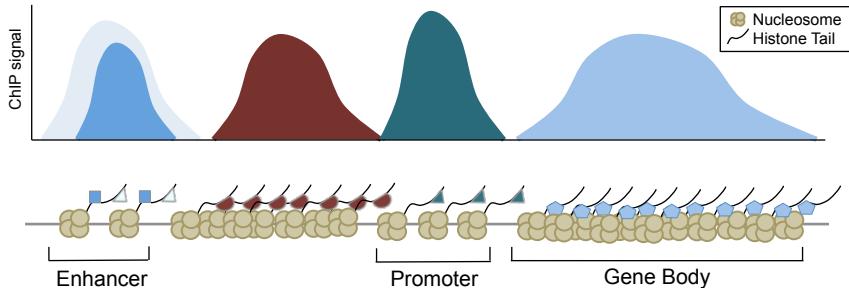
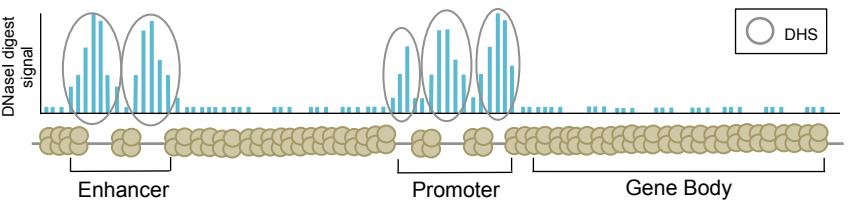
3D structure



Bau *et al.* 2011

Dekker *et al.* 2013

Examples of Epigenetic assays

Epigenetic Mechanisms	Modification Types*	Assay Examples	Simplified Diagrams
DNA Methylation	Low Intermediate High	 <u>Chemical-based</u> Bisulfite-treatment# [99] RRBS ^[100] <u>Enrichment-based</u> MeDIP# [101] Bio-CAP# [102] <u>Enzyme digest-based</u> MRE# [101]	
Histone PTM	 H3K4me1 H3K4me2 H3K4me3 H3K9me3 H3K27ac H3K27me3 H3K36me3	<u>Enrichment-based</u> Histone-specific ChIP# [103]	
Nucleosome Occupancy	DNase I Hypersensitive sites (DHS)	<u>Enzyme digest-based</u> DNase I# [104]	

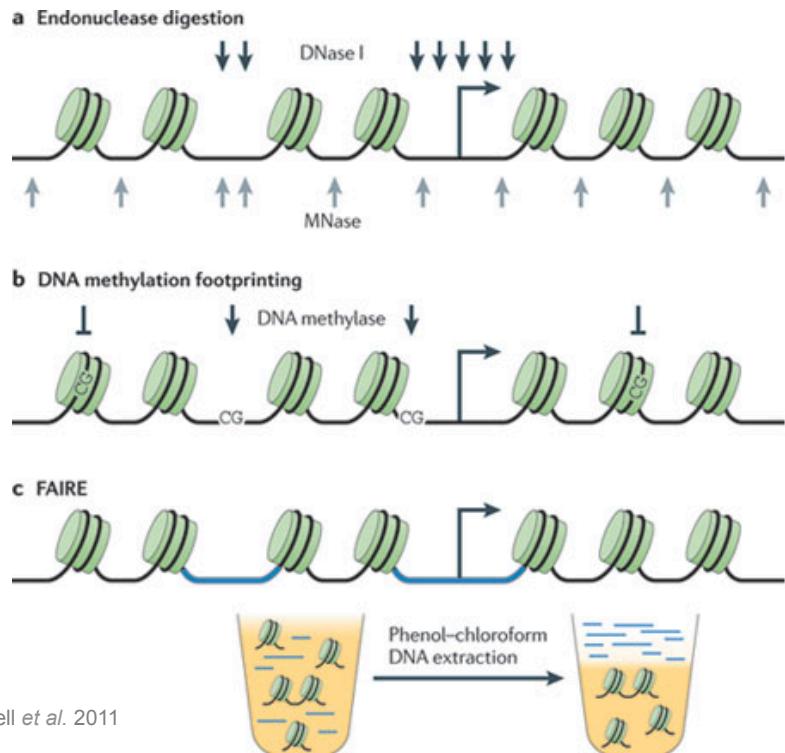
* = Red and blue terms correspond to repressive and processive state, respectively.

= These assays can be quantified by numerous techniques including, but not limited to, gel-imaging, targeted sequencing, RT-qPCR, microarrays and high-throughput sequencing.

[99] = Reference

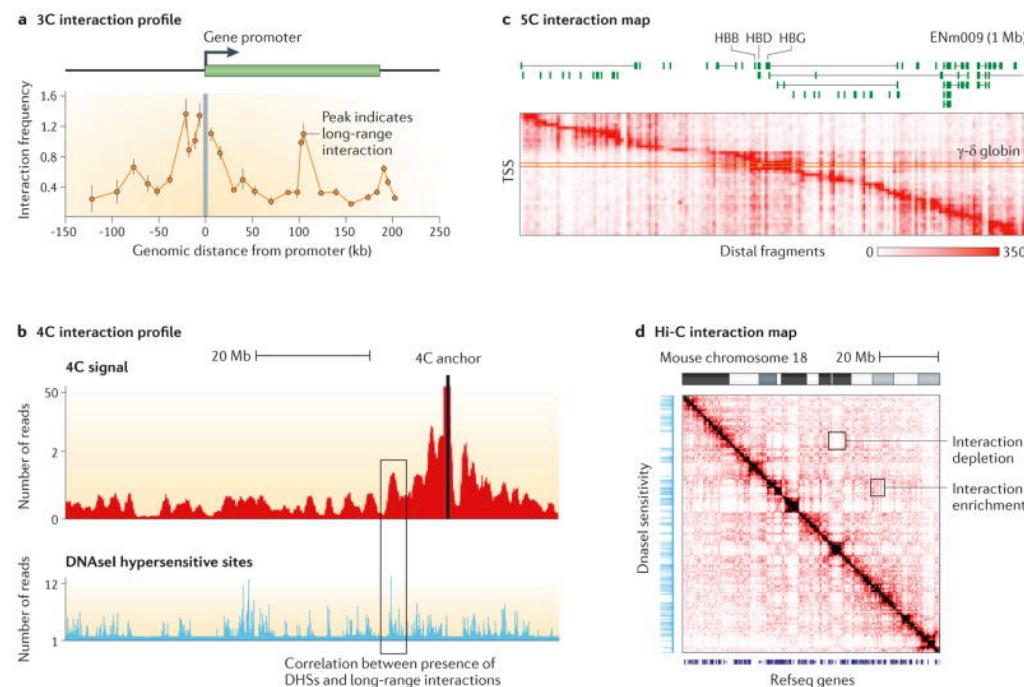
Examples of Epigenetic assays

Nucleosome positioning



Nature Reviews | Genetics

3D structure

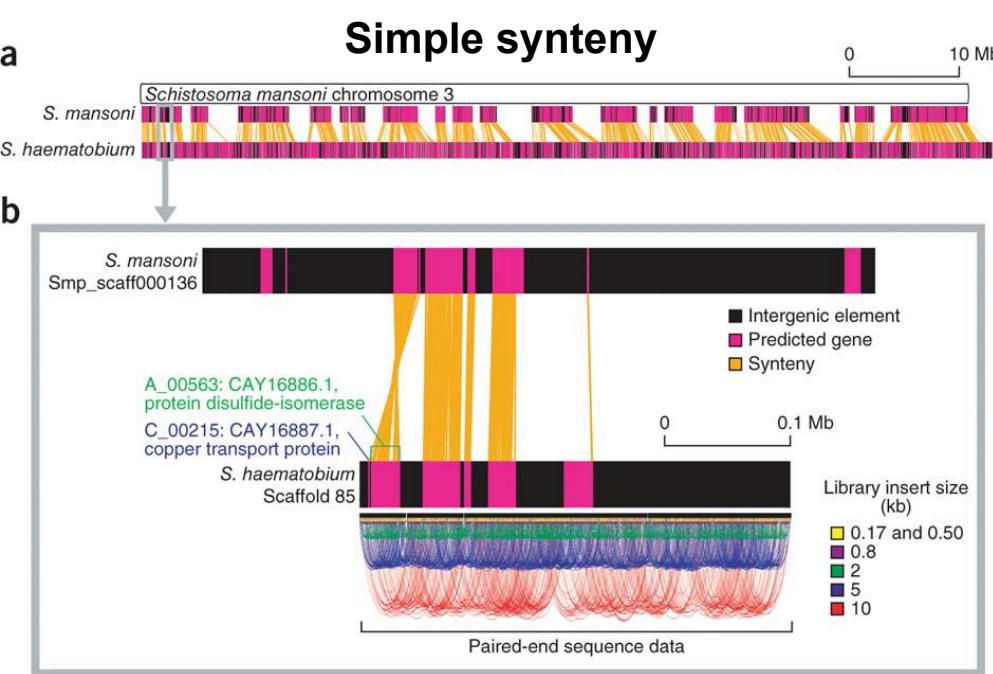


Comparative epigenomics

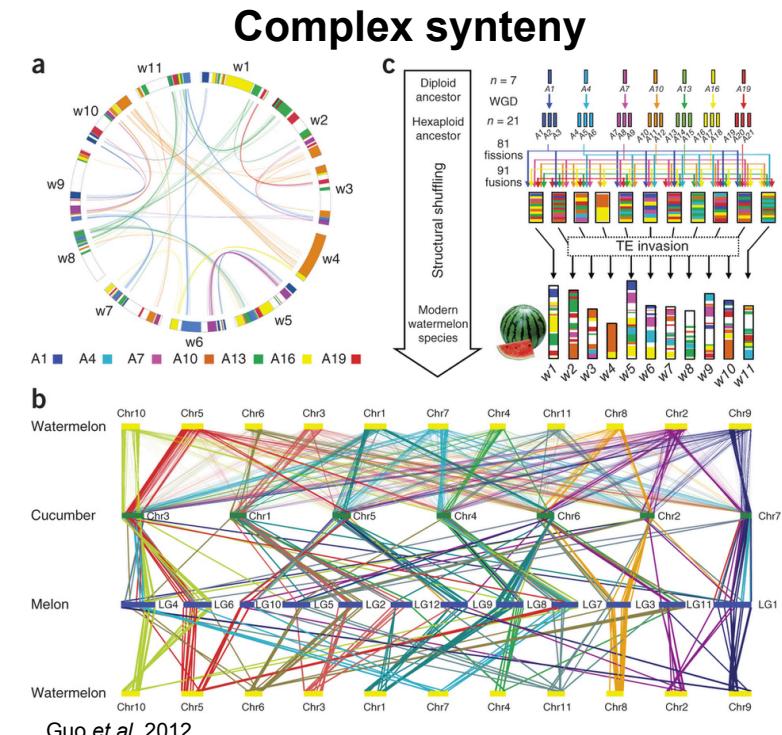
- What is it?
 - While epigenetics often refers to the study of single genes or sets of genes, epigenomics refers to more global analyses of epigenetic changes across the entire genome.
 - Comparative analysis can vary from unique cell-types to species-wide comparison of epigenomes.
- What types of epigenetic modifications are conserved or diverged over evolution?
 - For the scope of this review, we focus on how methylation and histone post-translational modifications (PTMs) evolved in syntentic regions across vertebrate organisms.

Epigenetic Evolution (EE) at syntenic sequences

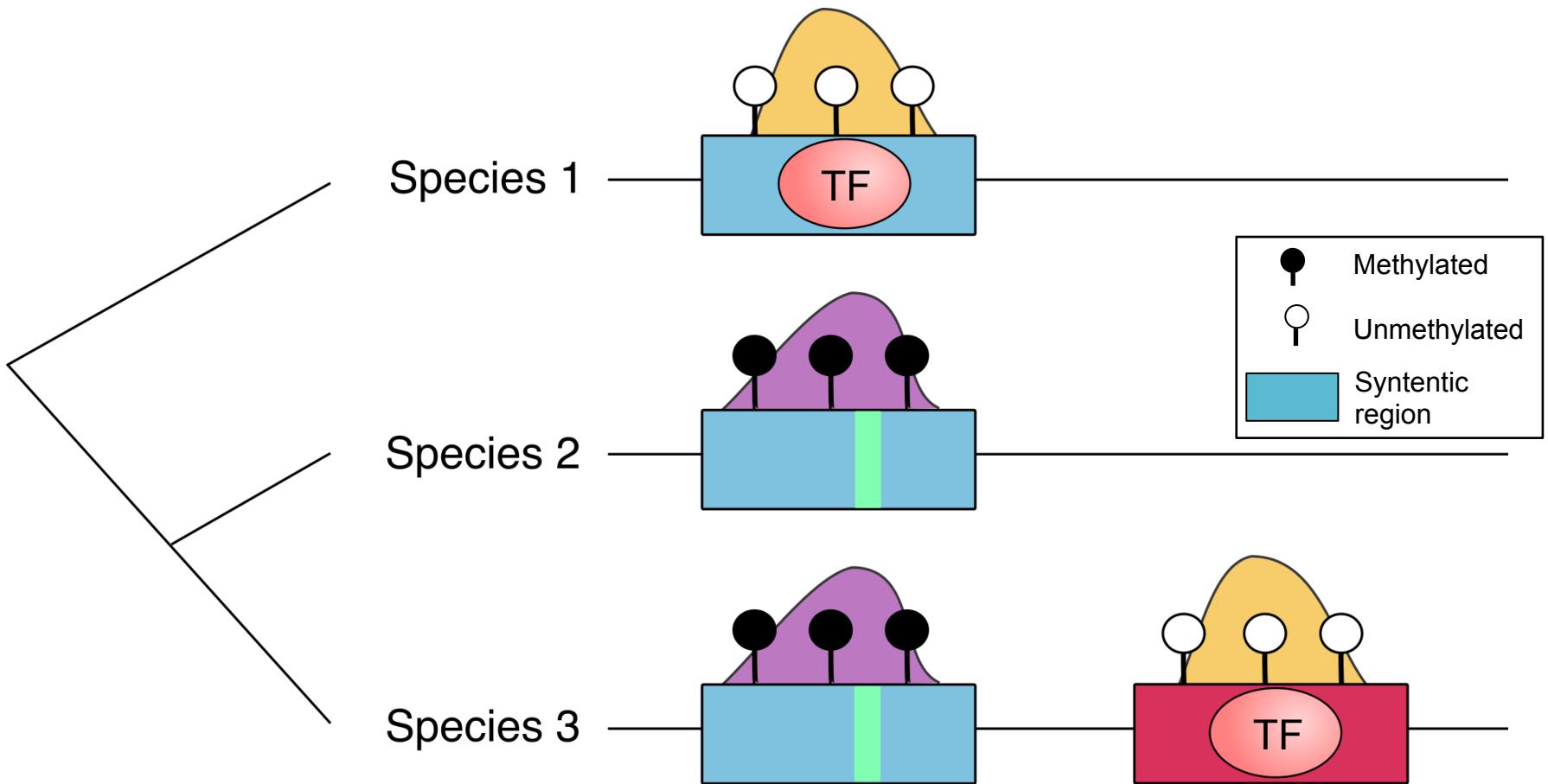
- What is synteny?
 - Conserved block of sequences or genes that are shared among related species.
 - Syntenic regions can reveal how evolution shaped the divergence or conservation of unique regulatory pathways.



Young et al. 2011



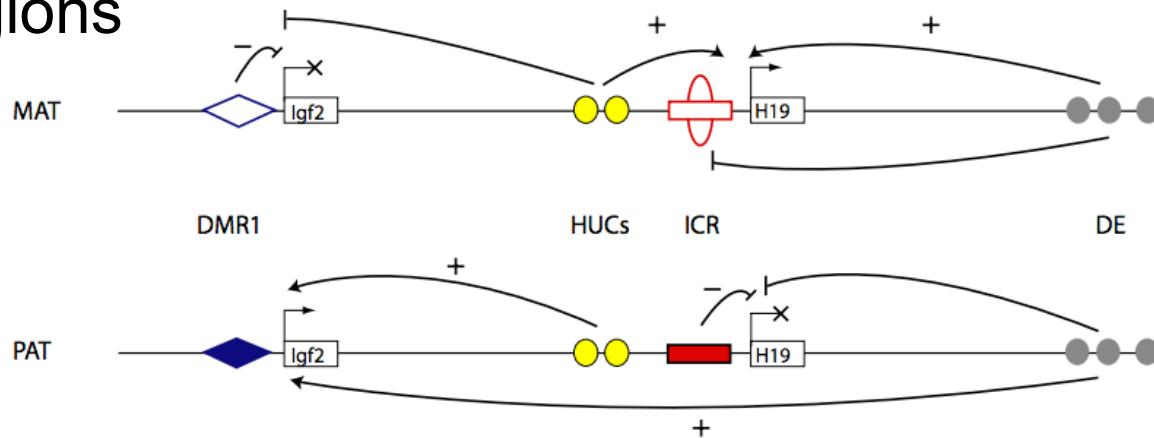
Epigenetic evolution in syntenic regions



What evidence is present that supports or refutes this model?

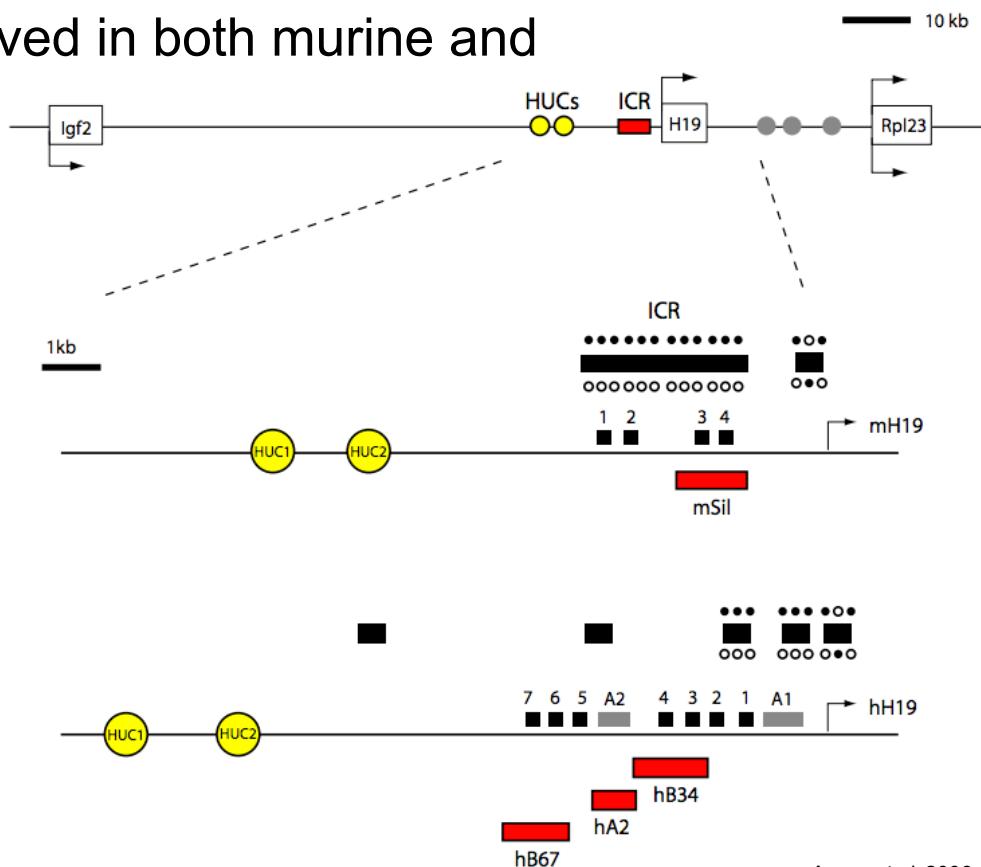
Epigenetic complexity in syntenic regions

- Example: *H19* and *Igf2* imprinted genes.
 - Syntenic regions in mouse and humans are thought to be coordinately regulated.
 - *H19* is expressed from paternal allele
 - *Igf2* is expressed from maternal allele
 - Within a single genome, there is differential expression driven by epigenetic control in syntenic regions



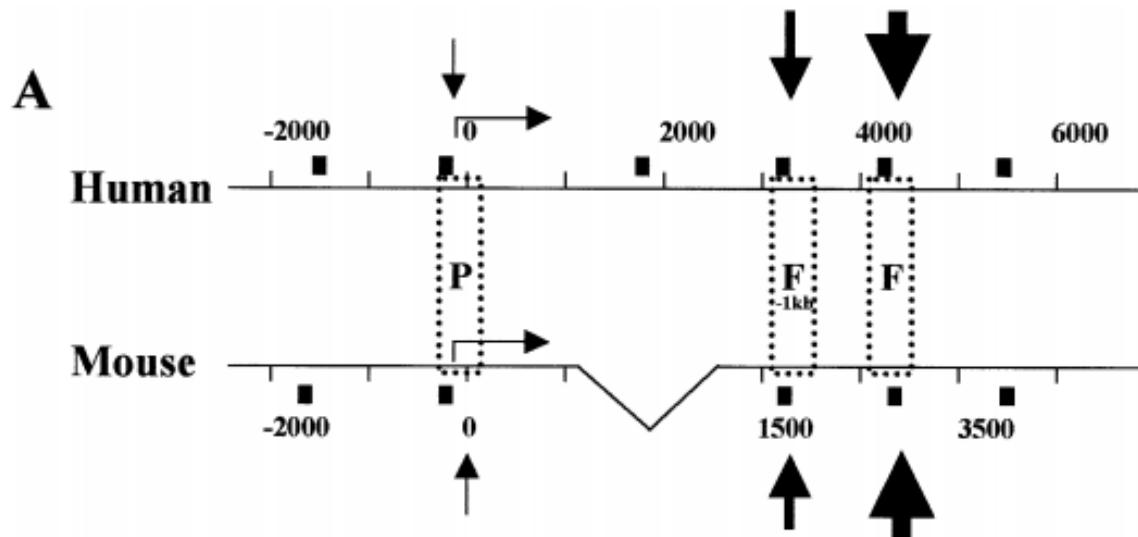
H19 and *Igf2* imprinted genes

- Same sequence yet different mono-allelic gene expression pattern. What regulates expression?
 - Imprinting control region is differentially methylated.
 - This phenomenon is conserved in both murine and human genome.
- Example of how syntetic regions across species have conserved function determined by epigenetic mechanisms.



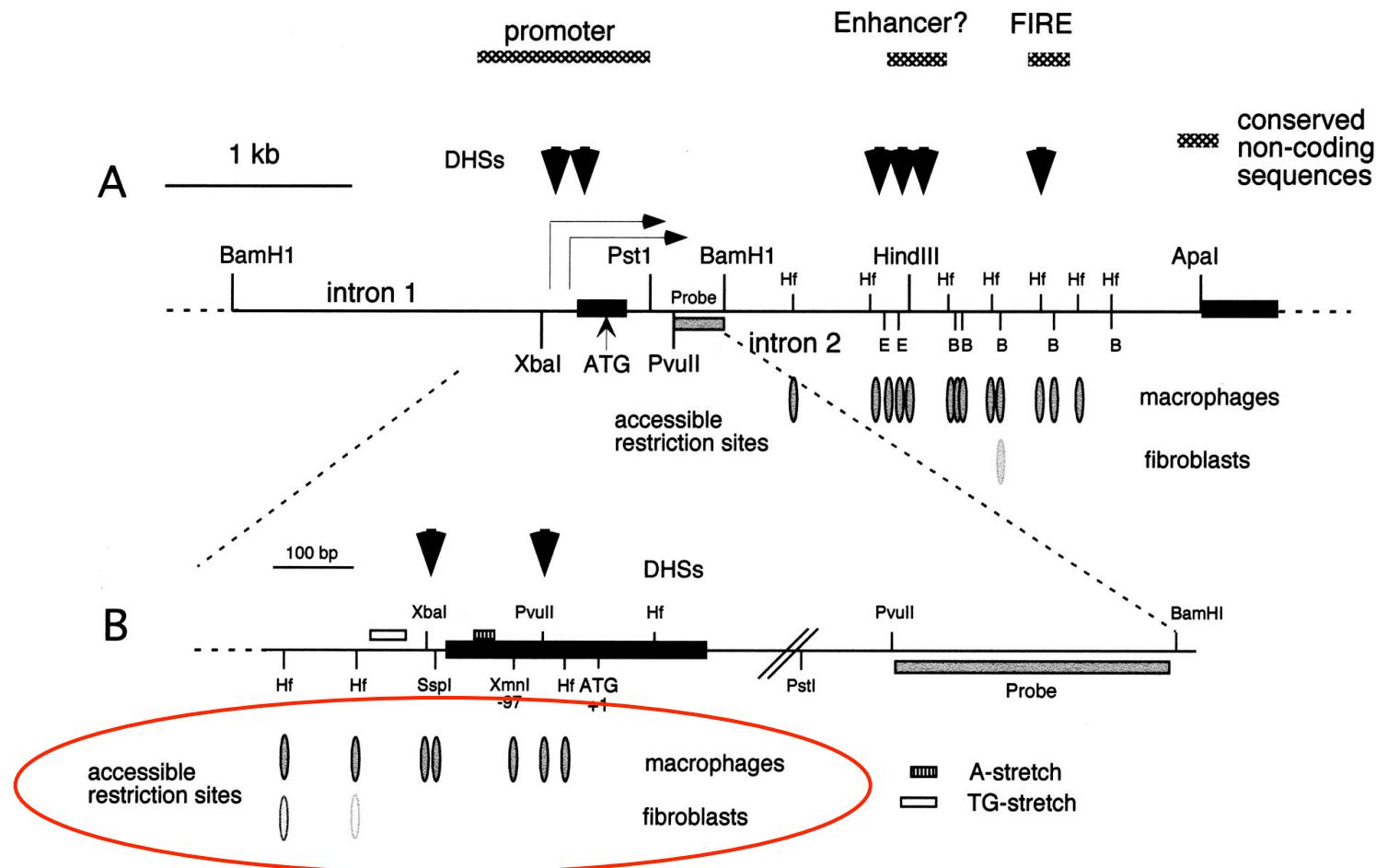
Cell-type and species-specific c-FMS regulation

- Macrophage colony stimulating factor receptor (c-FMS) gene is highly conserved across vertebrate species.
 - Highly conserved sequence yet species-specific chromatin structure has been revealed.



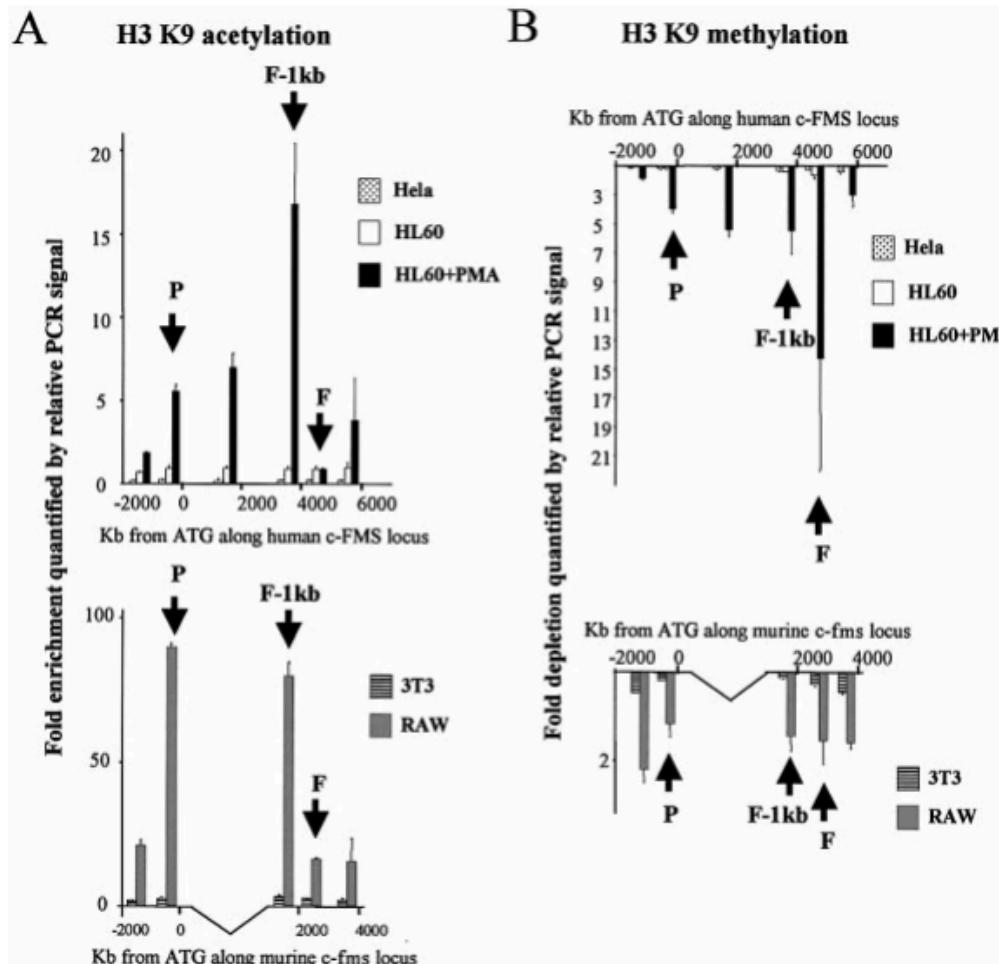
Cell-type specific c-FMS landscape

- Cell-type specific chromatin structure



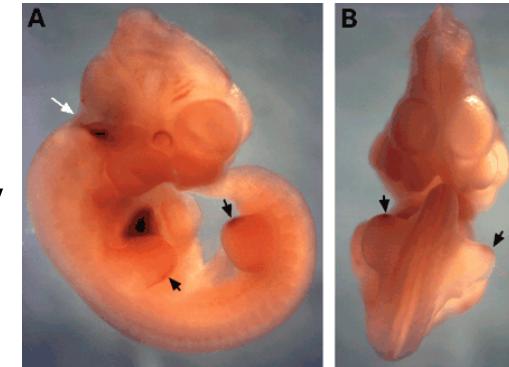
Species-specific c-FMS chromatin structure

- Accessible chromatin regions vary across species upstream of c-FMS gene



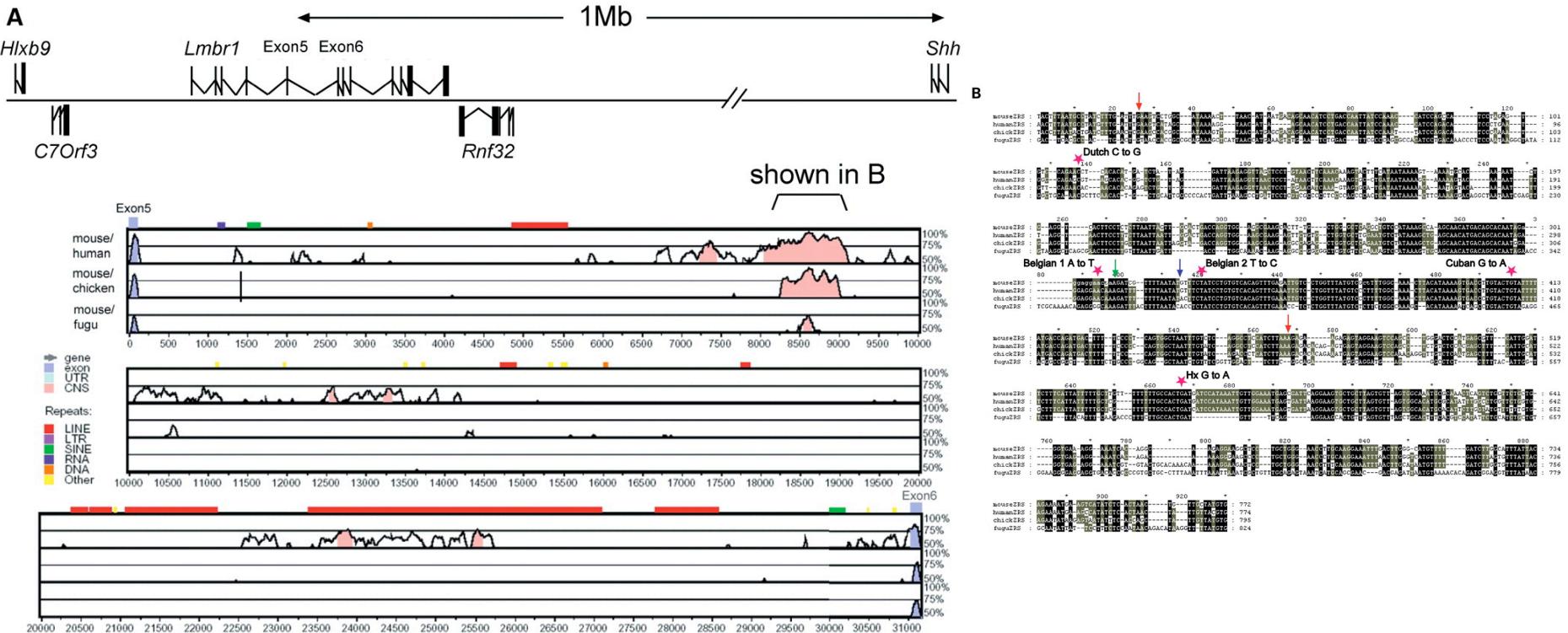
Long-range *Shh* enhancer function conservation

- Sonic hedgehog (SHH) expression in zone of polarising activity (ZPA) regulates proper limb development.
- Expression of *Shh* can be controlled by elements.
- Identification of highly conserved regions across species can reveal regulatory elements.
 - Pair-wise comparison among human, mouse, chicken and fugu identified highly conserved region in intron 5 of *Lmbr1* gene, which is believed to be regulatory.



Lettice et. al 2003

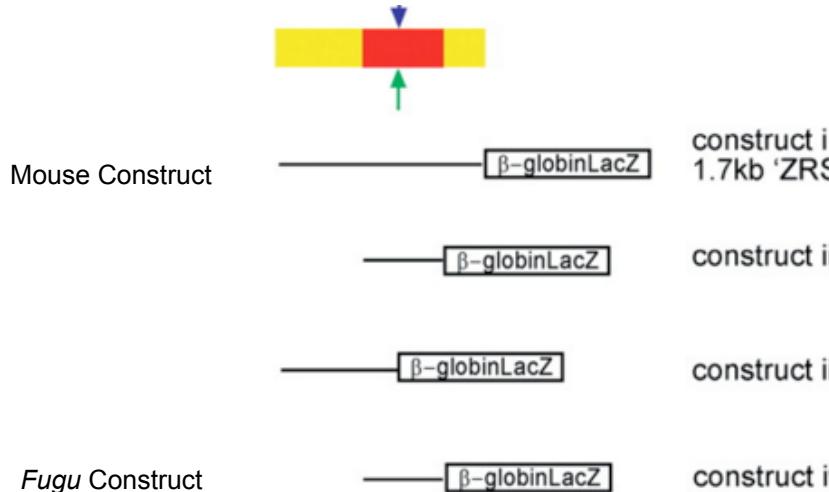
Long-range *Shh* enhancer function conservation



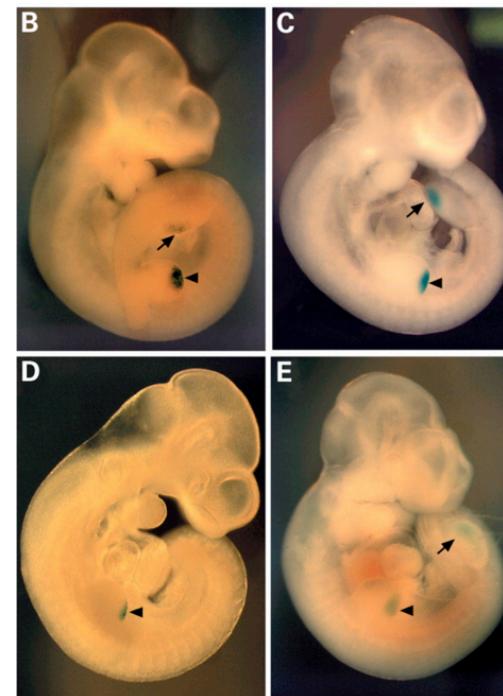
Lettice et. al 2003

Long-range *Shh* enhancer function conservation

- Mouse and *Fugu* conserved domains of *Lmbr1* intron 5 show *shh* regulatory function. This example illustrates how syntenic sequences have conserved function.

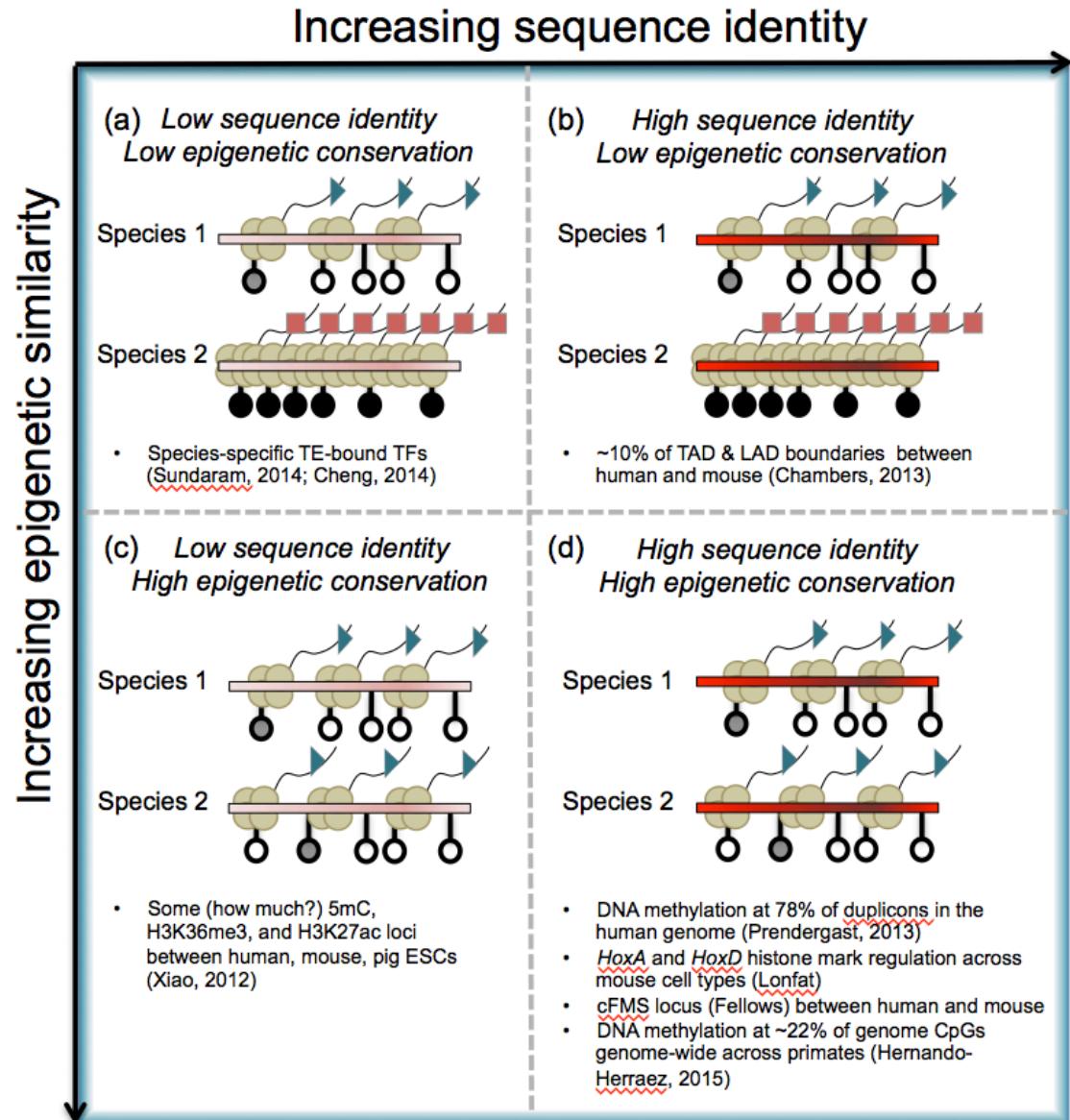


Lettice et. al 2003



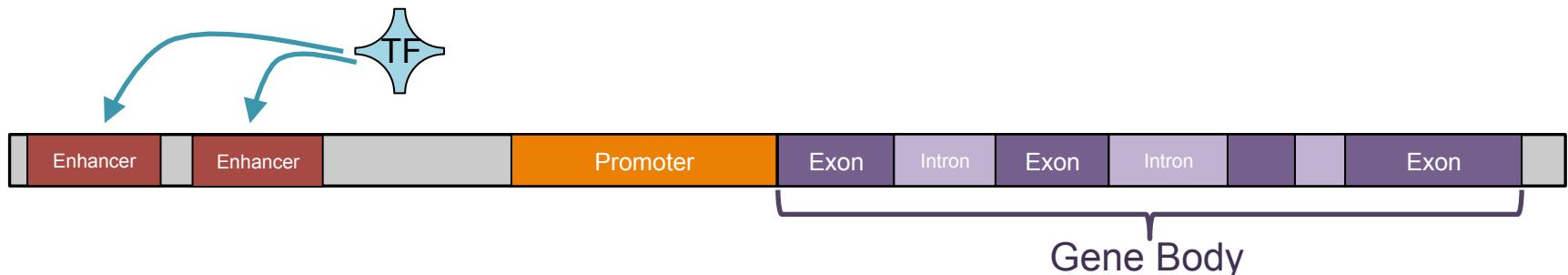
How to classify Epigenetic Evolution?

- Lot of unique examples of epigenetic conservation and divergence.
- Here, we propose 4 simplified scenarios of epigenetic evolution:



Overview of EE examples

- General DNA methylation conservation across sequence context
- General relationship between histone-PTM conservation and sequence context
- Epigenetic evolution across functional elements.
 - Promoters: Methylation and Histone PTM signatures
 - Gene body-specific epigenetic conservation
 - Evolution of epigenetic regulation at enhancers
- Transcription factor binding conservation



DNA methylation conservation

- In the human genome, 78% of paralogous CpGs had an absolute DNA methylation difference of 20% or less.
 - Duplicons tend to retain their DNA methylation signature, supporting the hypothesis that epigenetic features are correlated with underlying sequence

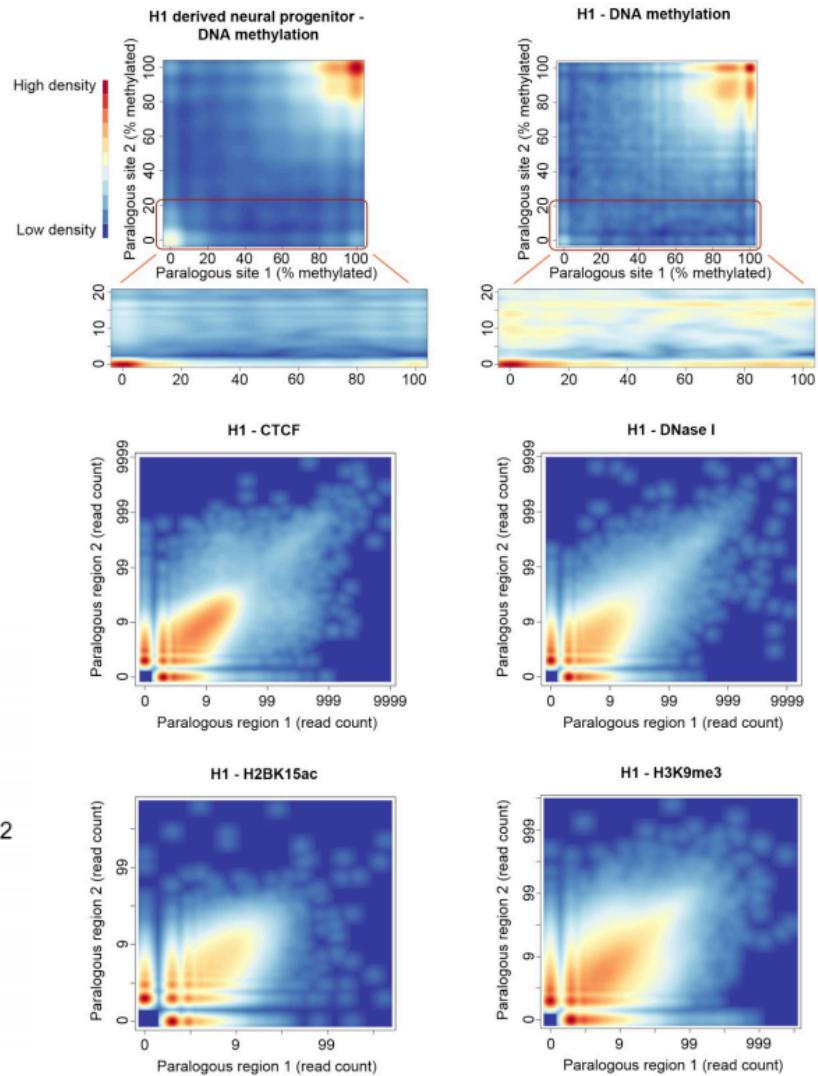
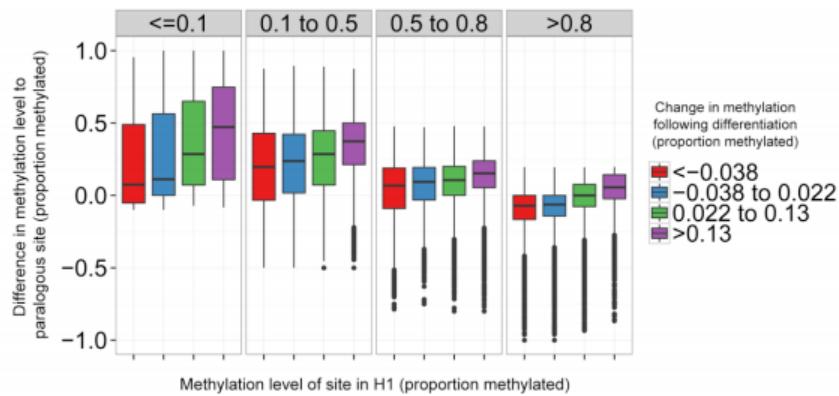
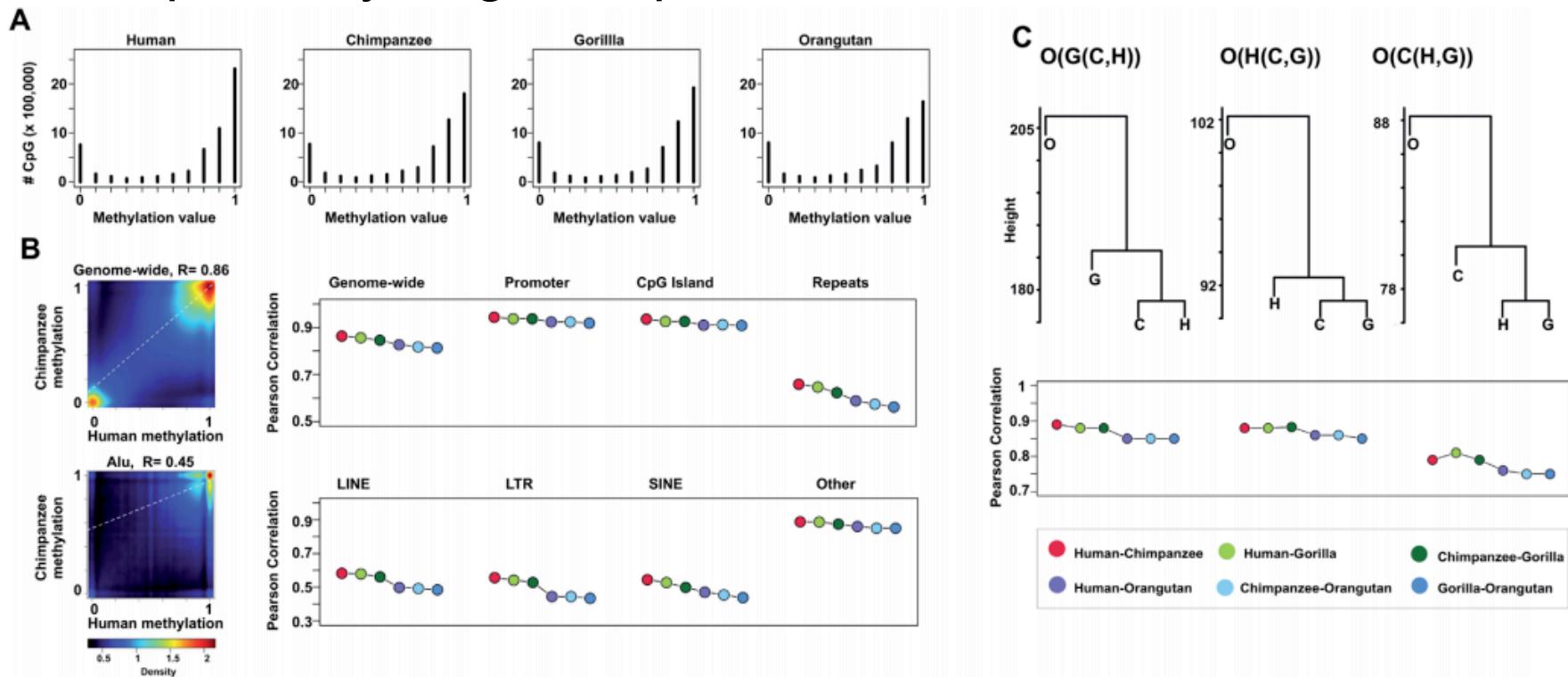


Figure 1

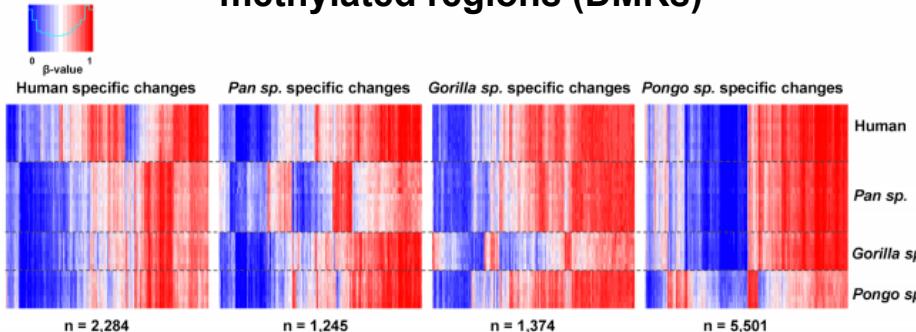
DNA methylation conservation across primates

- Comparing genome-wide DNA methylation levels between species, 70–74% and 80–82% similarities were found in peripheral blood and prefrontal cortex, respectively, in great ape somatic tissues.

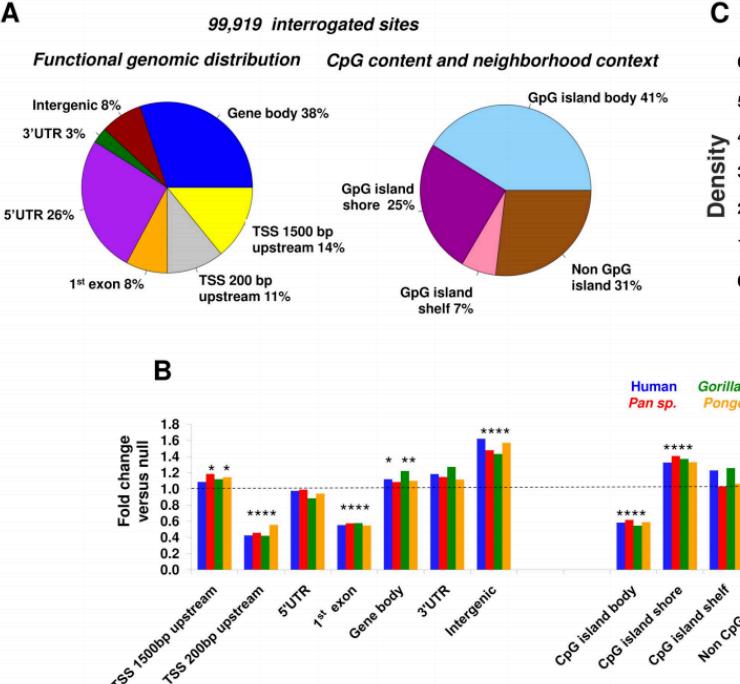


However, species-specific methylation exists

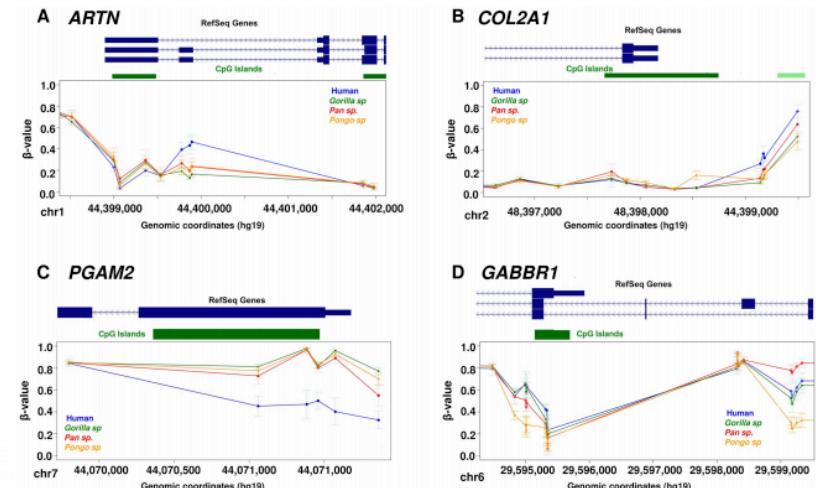
Species-specific differentially methylated regions (DMRs)



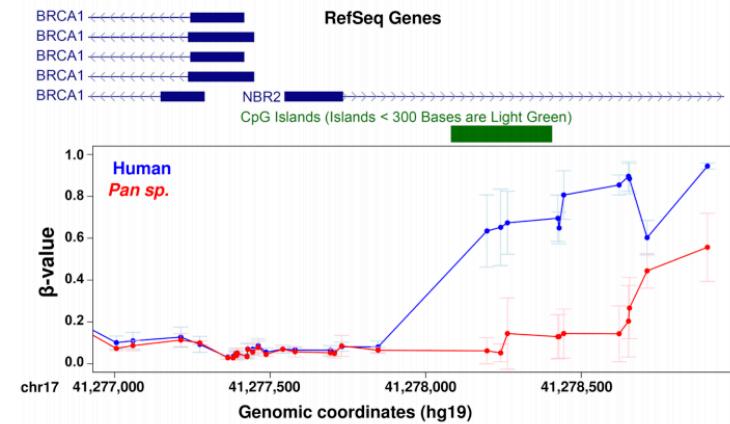
Location of DMRs



Differentially methylated genes

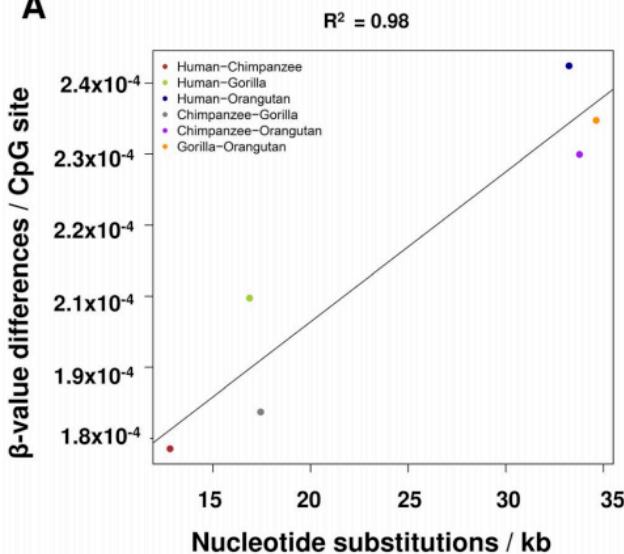


BRCA1

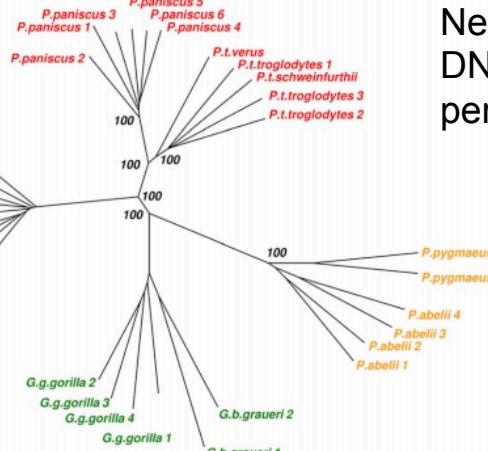


DNA methylation patterns related to sequence variation

A



B



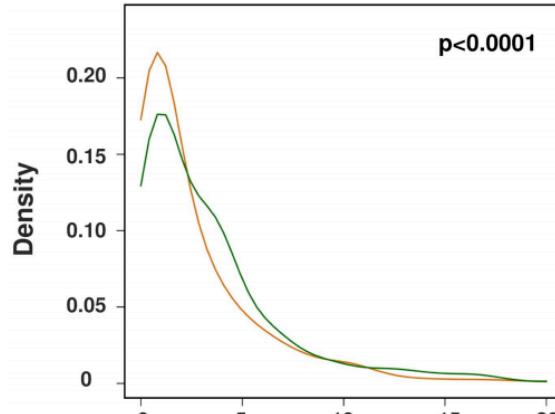
Neighbor-joining tree based on DNA methylation differences perfectly recapitulates speciation.

Hernando-Herraez et al. 2013

Significant association between rate of protein evolution and methylation differences

A

Differentially methylated genes
n=745



B

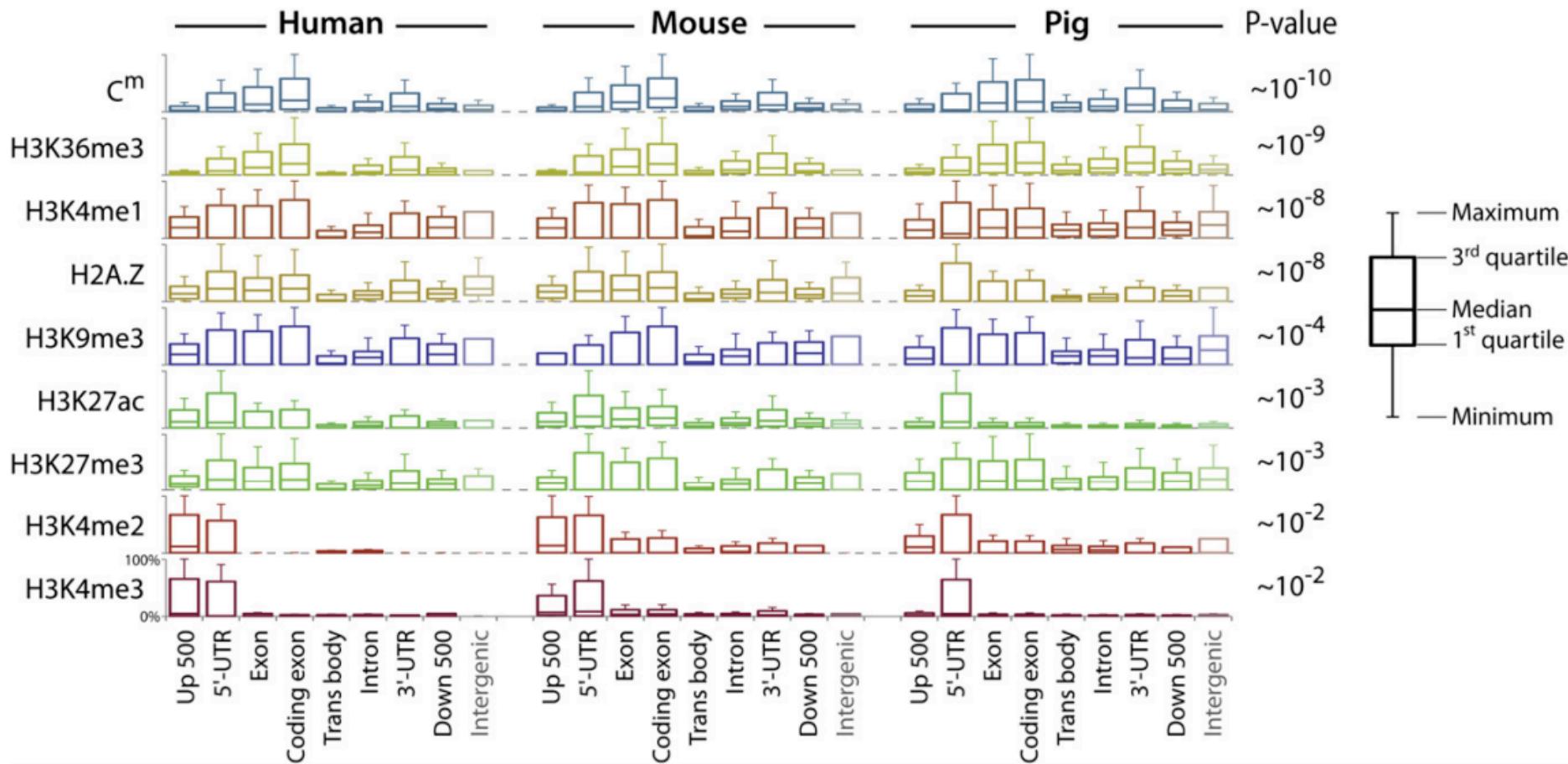
Genes without significant changes in methylation n=6,507

Number of amino acid changes per gene

K_A/K_I ratio per gene



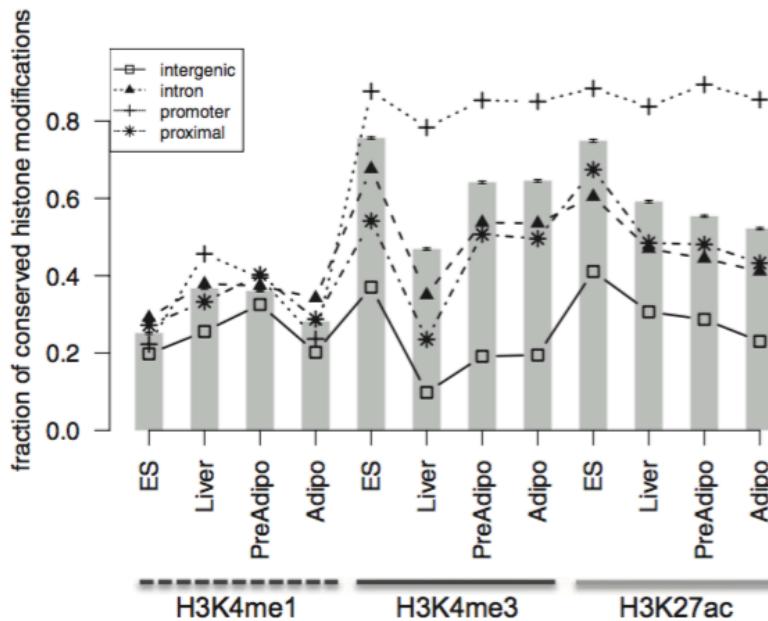
Overall Histone PTM conservation across species



-Reveals both conservation and divergence of histone PTM signatures across genomic regions and species.

Histone modification conservation over orthologous sequences

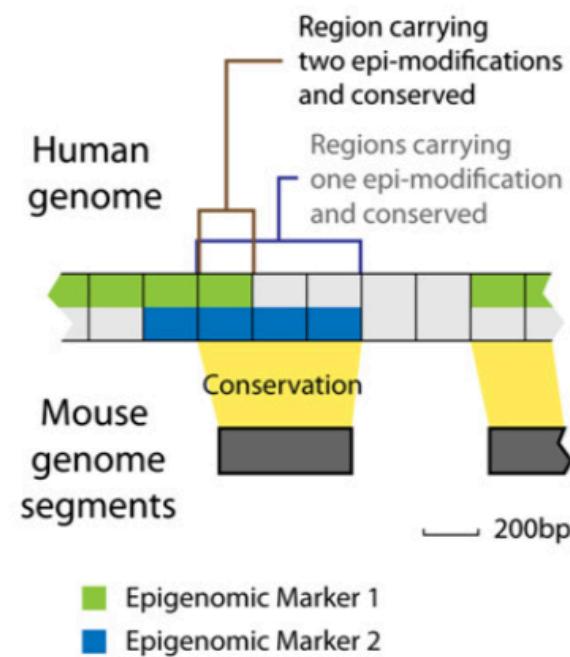
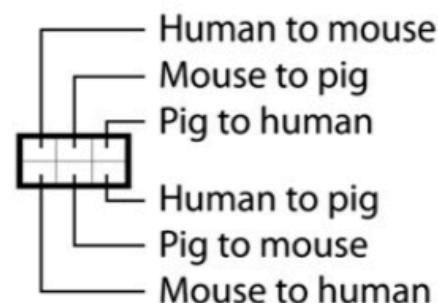
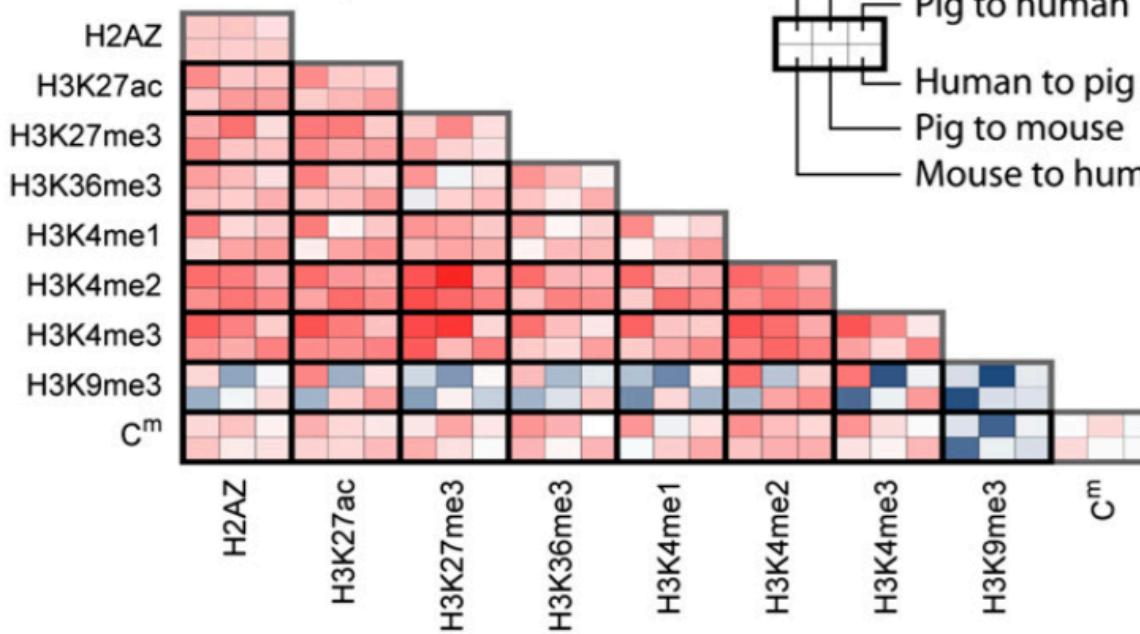
- In 100k regions in human and mouse, H3K4me3 is generally more conserved.
- Epigenetic status at promoters was generally conserved across cell types.



Histone PTM status compared to sequence conservation

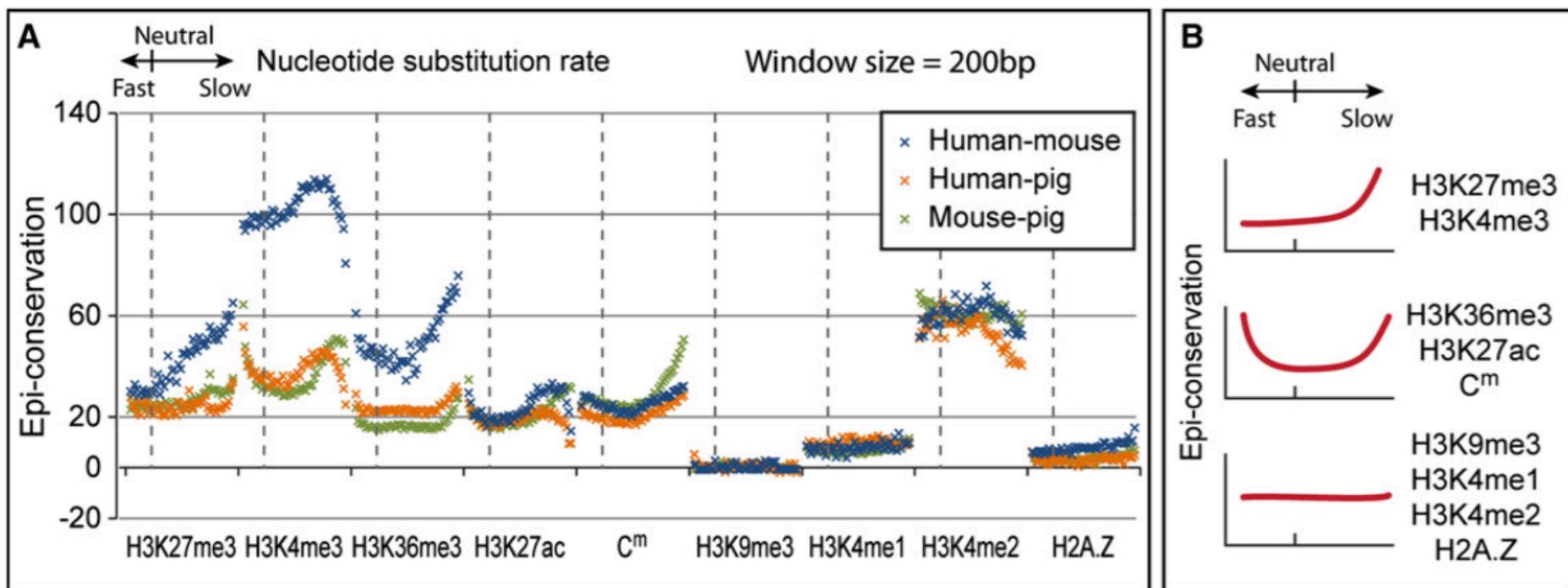
- Xiao, 2012

Overlap of epi-modified and Conserved sequences



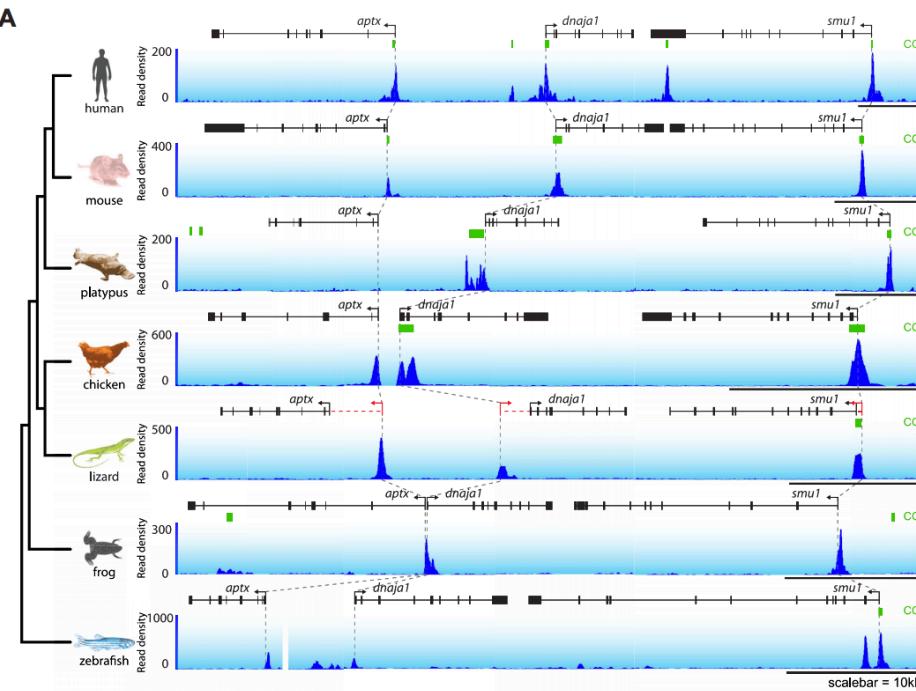
Sequence conservation is not required for Histone PTM conservation

- Xiao, 2012

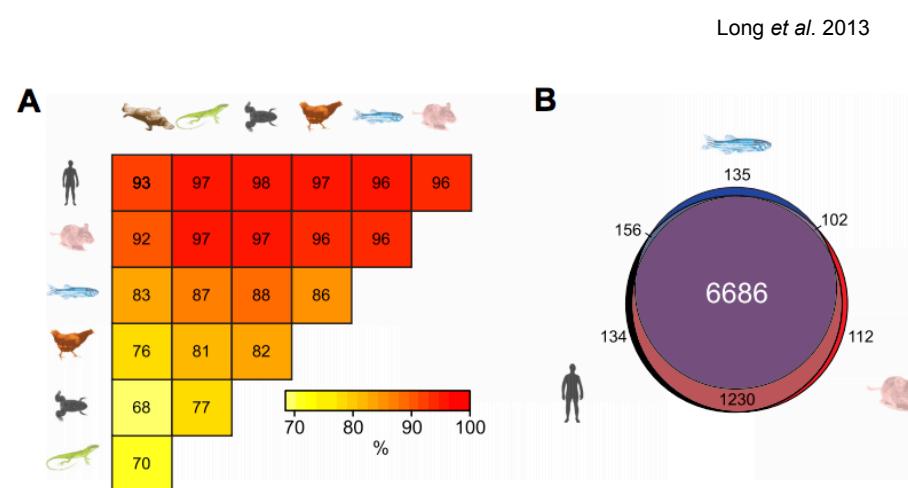


Methylation conservation in promoters

- Non-methylated islands (NMIs) show high conservation in vertebrate gene promoters.
- 2/3 of gene promoters in mammals are associated with NMIs



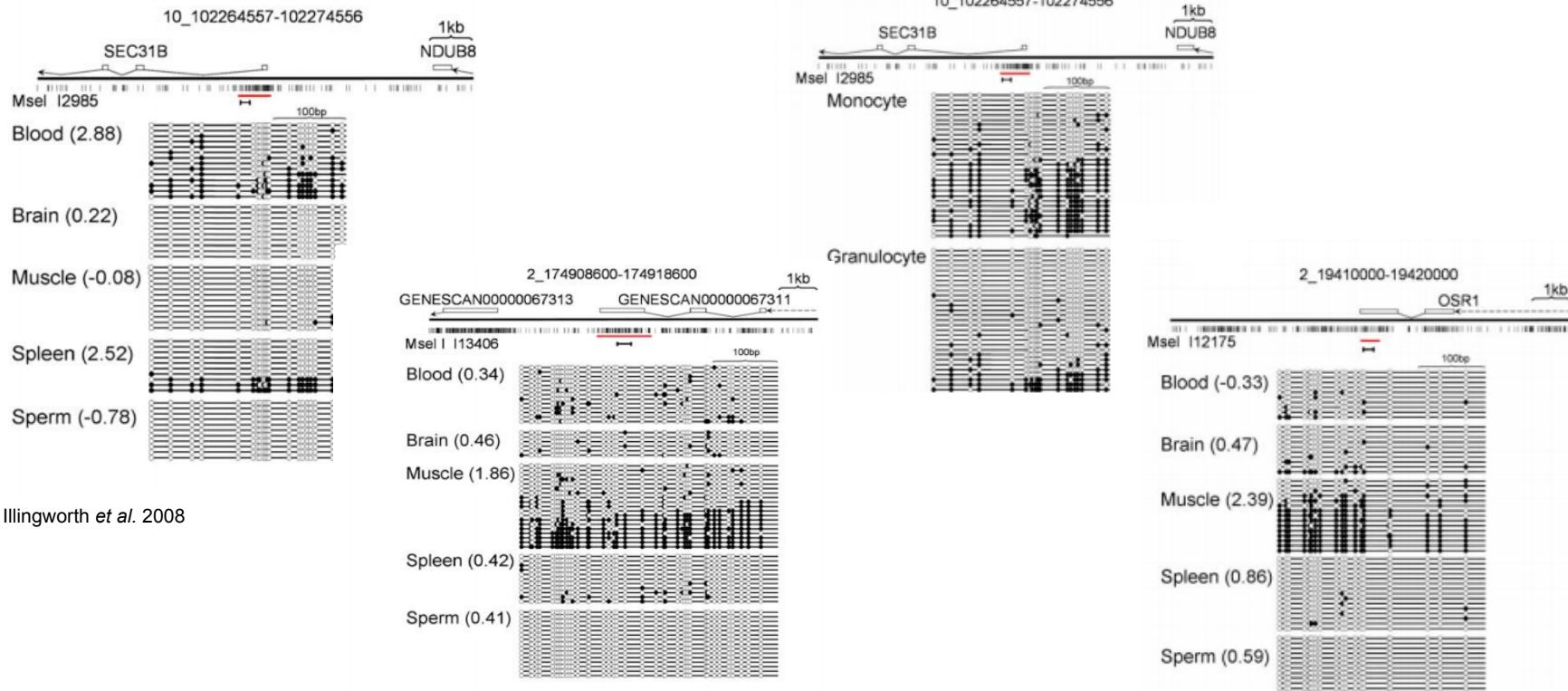
NMIs frequently overlap in syntenic promoter regions



Percentage of NMIs conserved at orthologous gene TSS across vertebrate species.

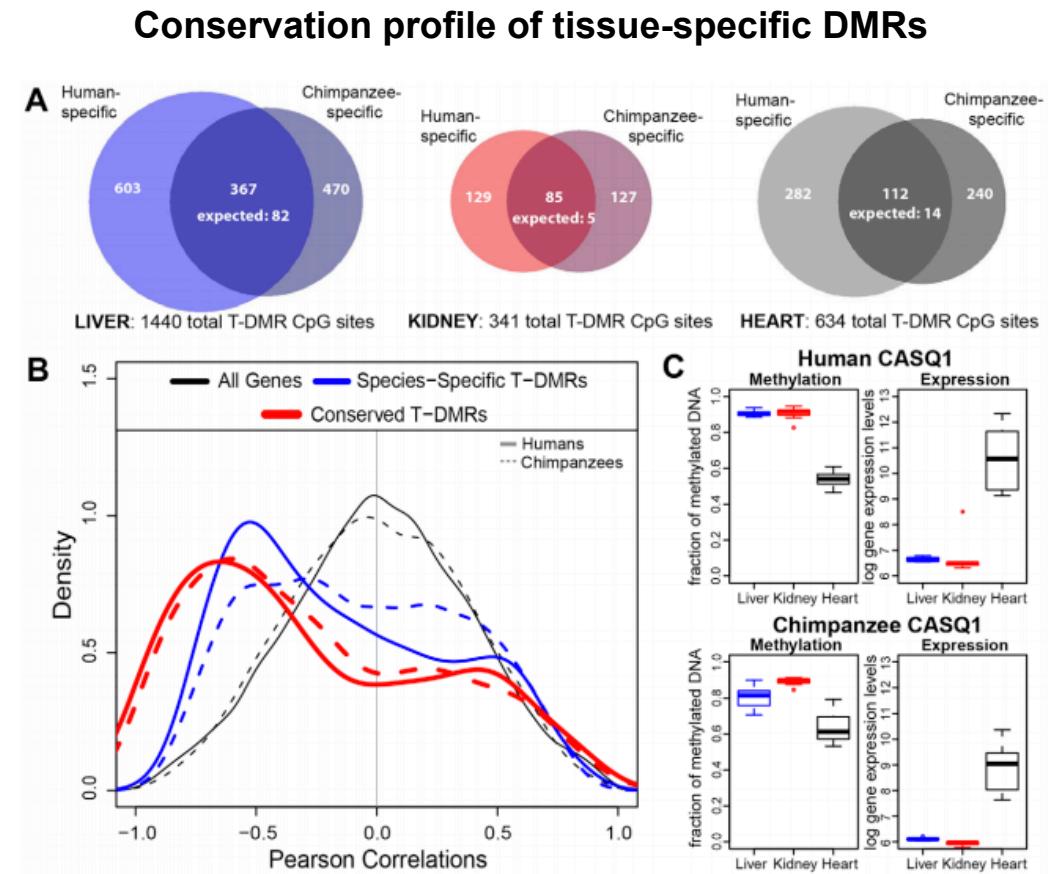
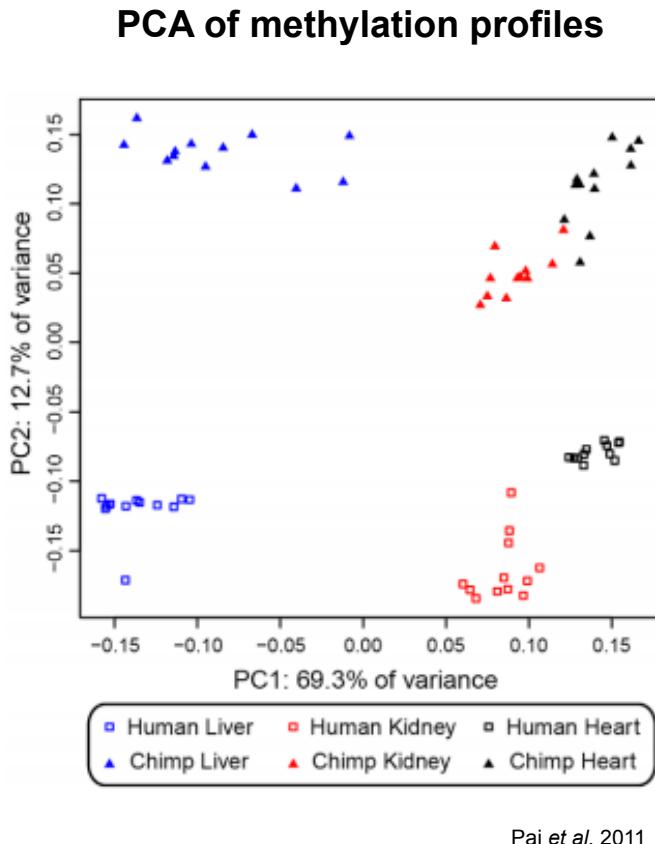
Tissue-specific DMRs in promoter regions

- Bisulfite sequencing reveal differential methylation in CpG islands in promoters across different tissue types.



Species-specific DMRs in promoter regions

- There are promoters with inter-species differential DNA methylation, and these explained 12–18% of gene expression level differences between primates.

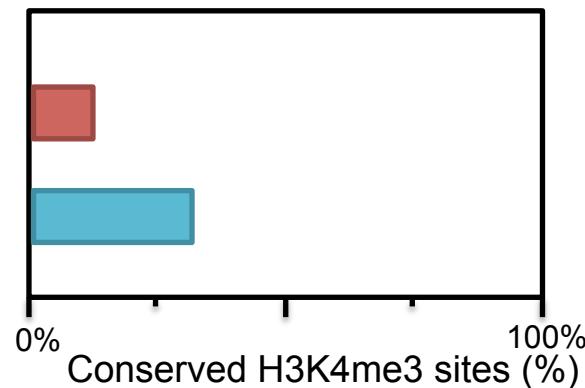


Histone modification conservation over orthologous promoters

- Epigenetic conservation of orthologous promoters decreasing as phylogenetic scope increases.

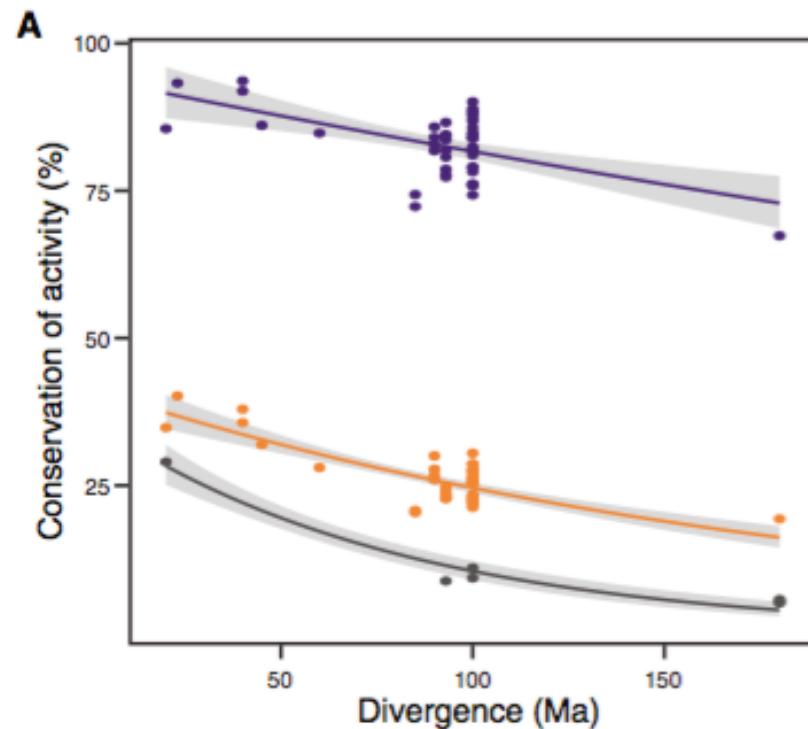
Promoters with shared H3K4me3 across 20 mammals

Promoters with shared H3K4me3 across 3 primates



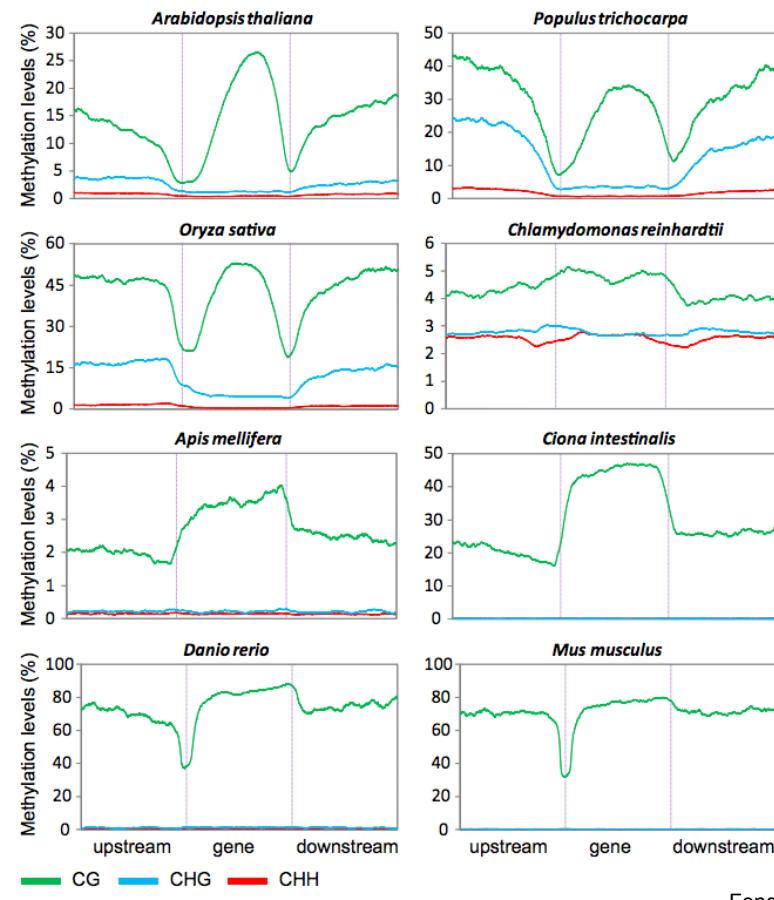
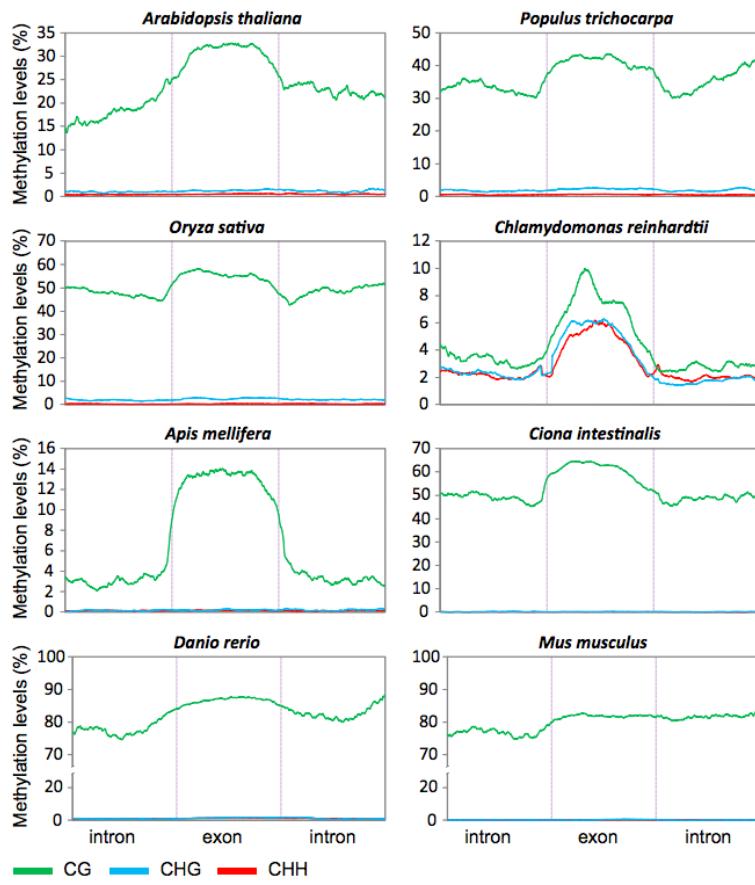
Histone modification conservation over orthologous promoters

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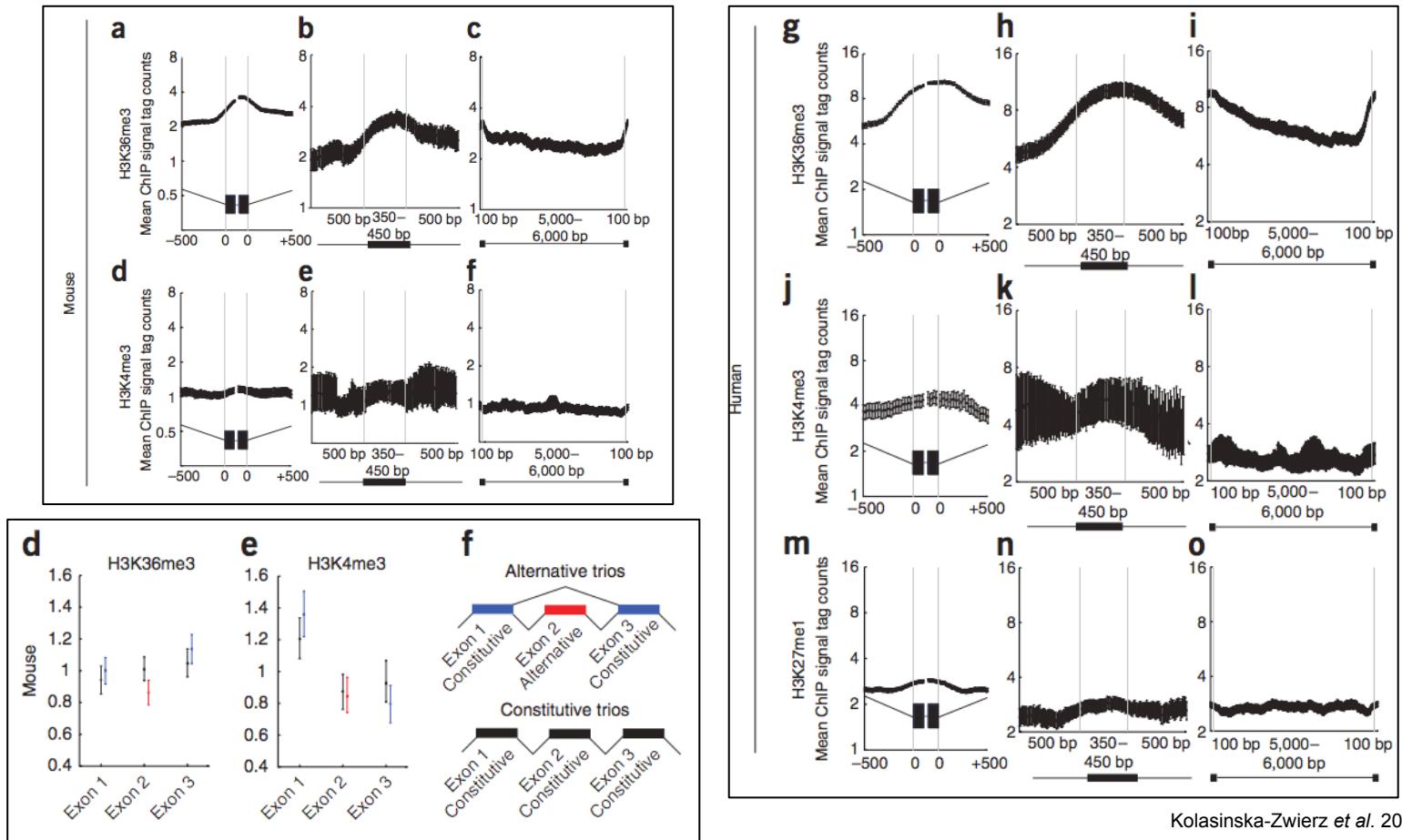
Gene body methylation conservation

- High methylation signature across gene body is conserved in multiple organisms



Gene body histone PTM

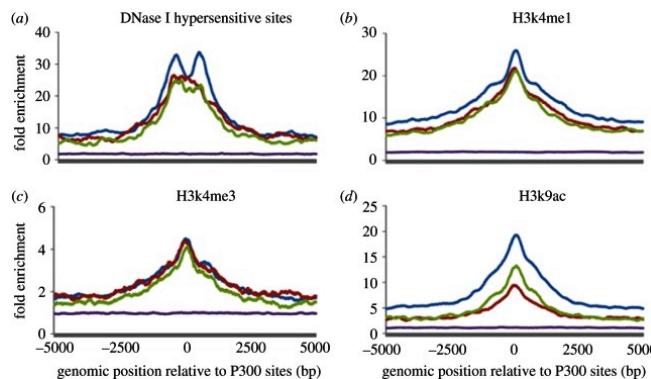
- Exon-specific H3K36me3 modifications are conserved across eukaryotes and are associated with exon inclusion.



Enhancer signature in epigenetic landscape

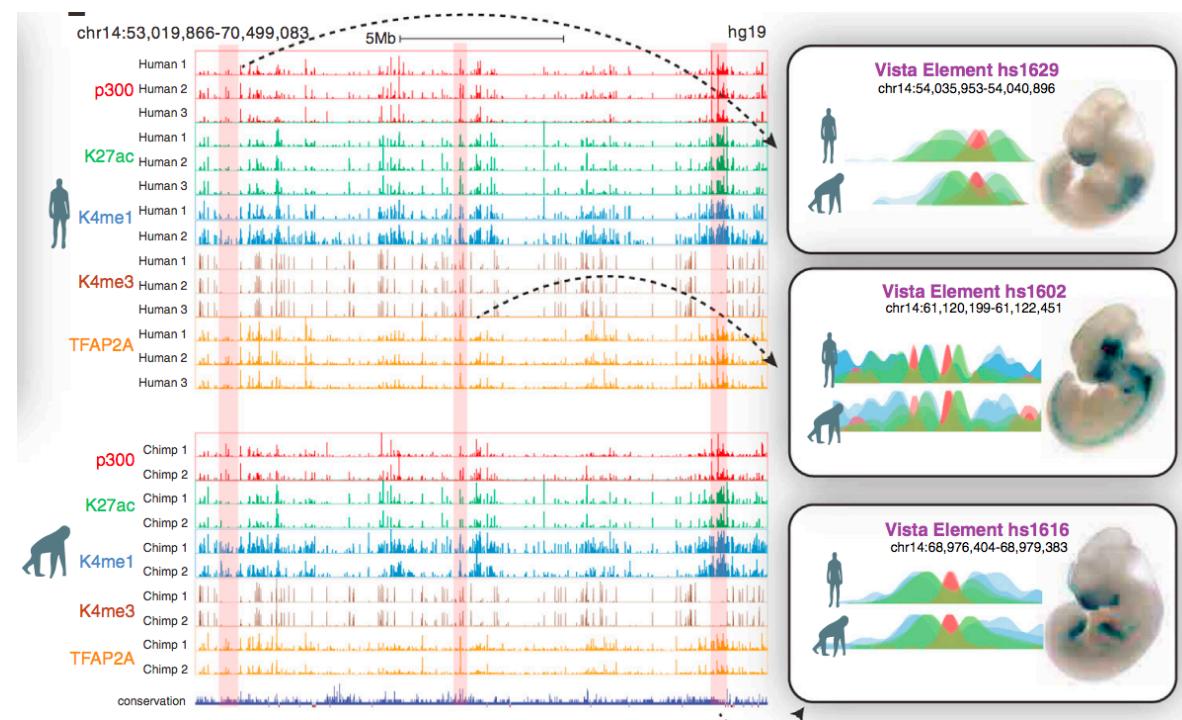
- Enhancers display a characteristic histone modification profile of H3K4me1 and H3K27ac, and are usually hypomethylated.

Distribution of histone modifications around heart enhancers



Hsu & Ovcharenko 2013

Identification of novel conserved enhancers



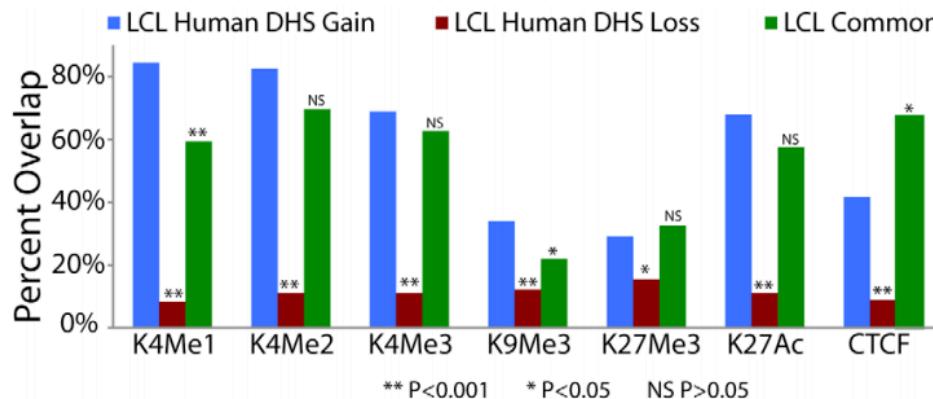
Prescott et al. 2015

Enhancer signature in Epigenetic landscape

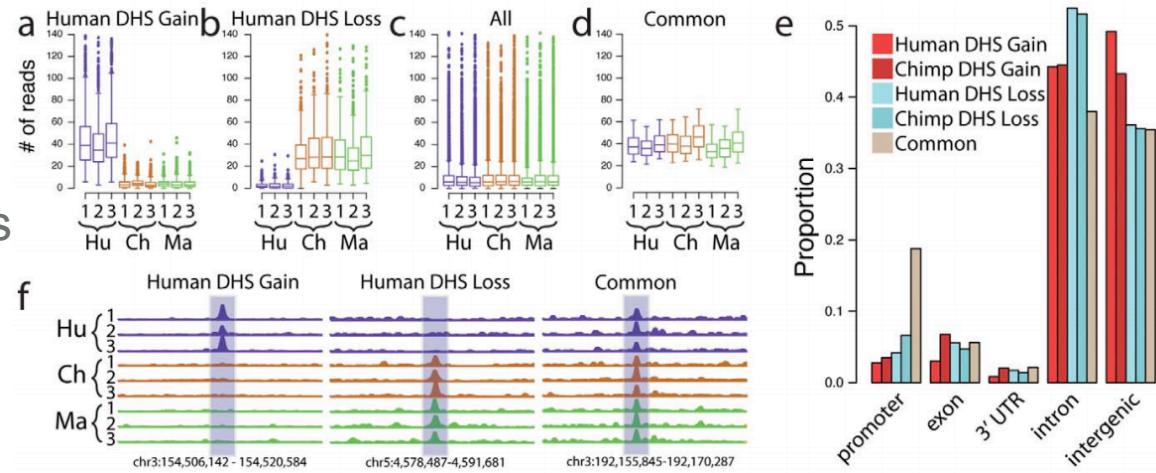
- DNase I Hypersensitive Sites (DHSs) identify region of open/accessible chromatin for transcription factor binding.
 - Often correlated with enhancer and promoter epigenetic signature.
- The field often relates transcription factor binding as enhancer activity as evidenced by increase in gene expression when binding is present.

Enhancer evolution across species

- Human DHS gains in skin and lymphoblastoid cells significantly overlapped with ChIP-seq signals for enhancer-associated chromatin marks H3K4me1 (80% overlap), H3K4me2 (80%), and H3K27ac (70%).

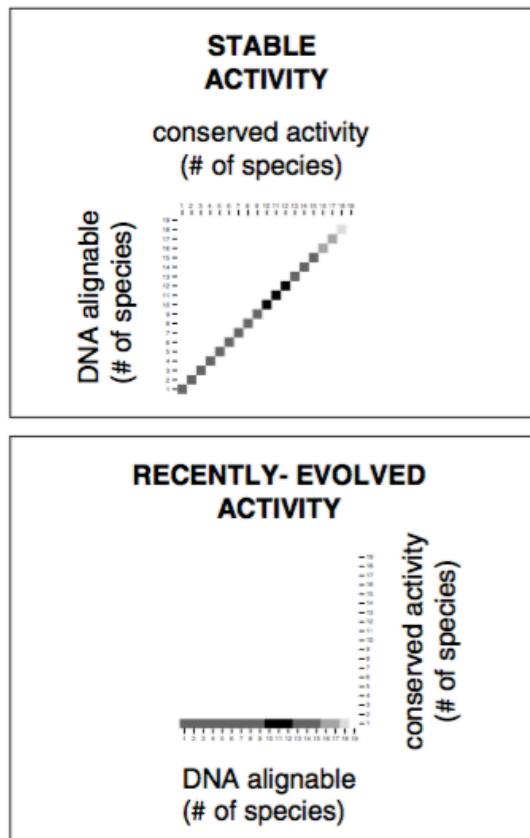


Numerous species-specific DHS gain and loss have been identified. These gain and loss could reveal insights into enhancer evolution.

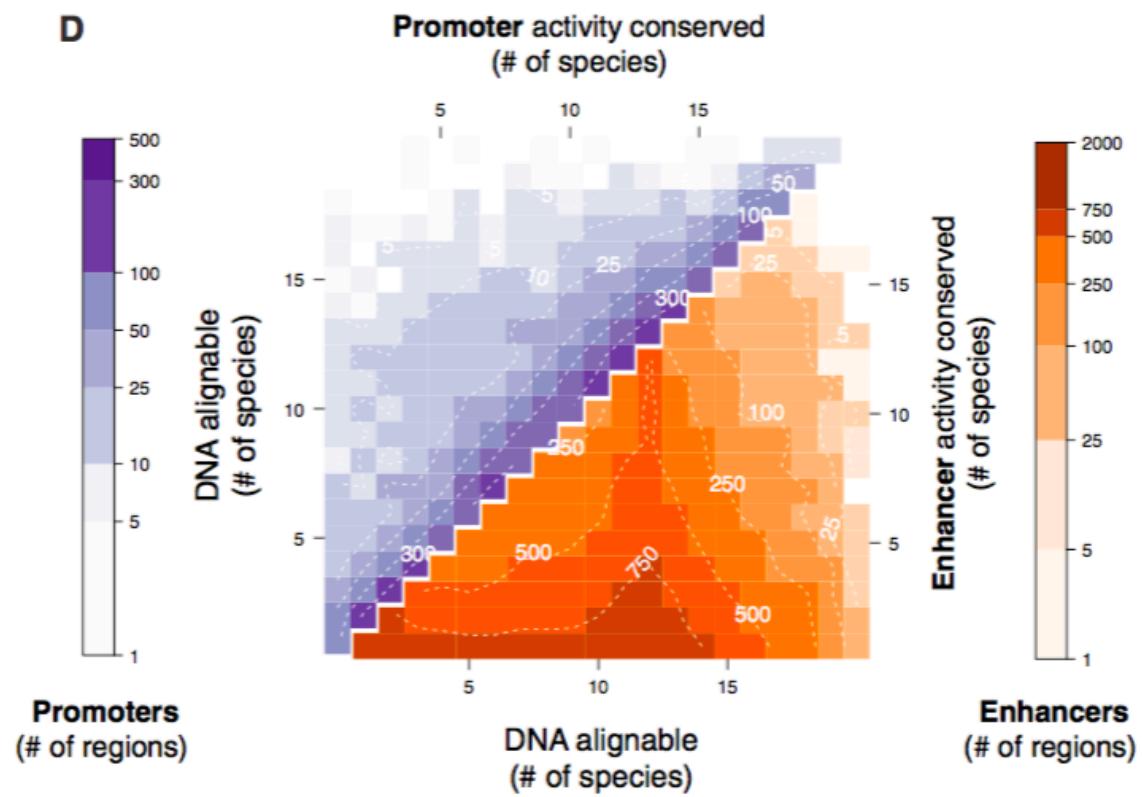


Enhancers tend to be species-specific

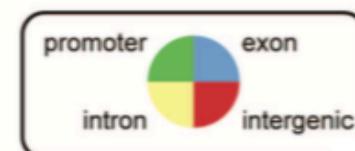
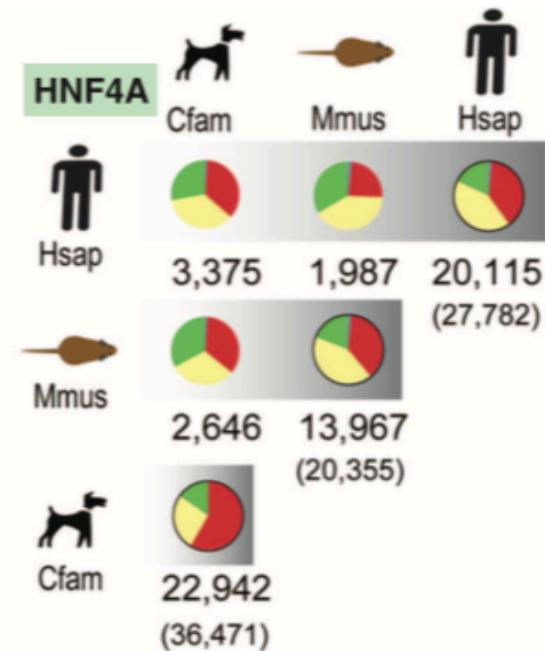
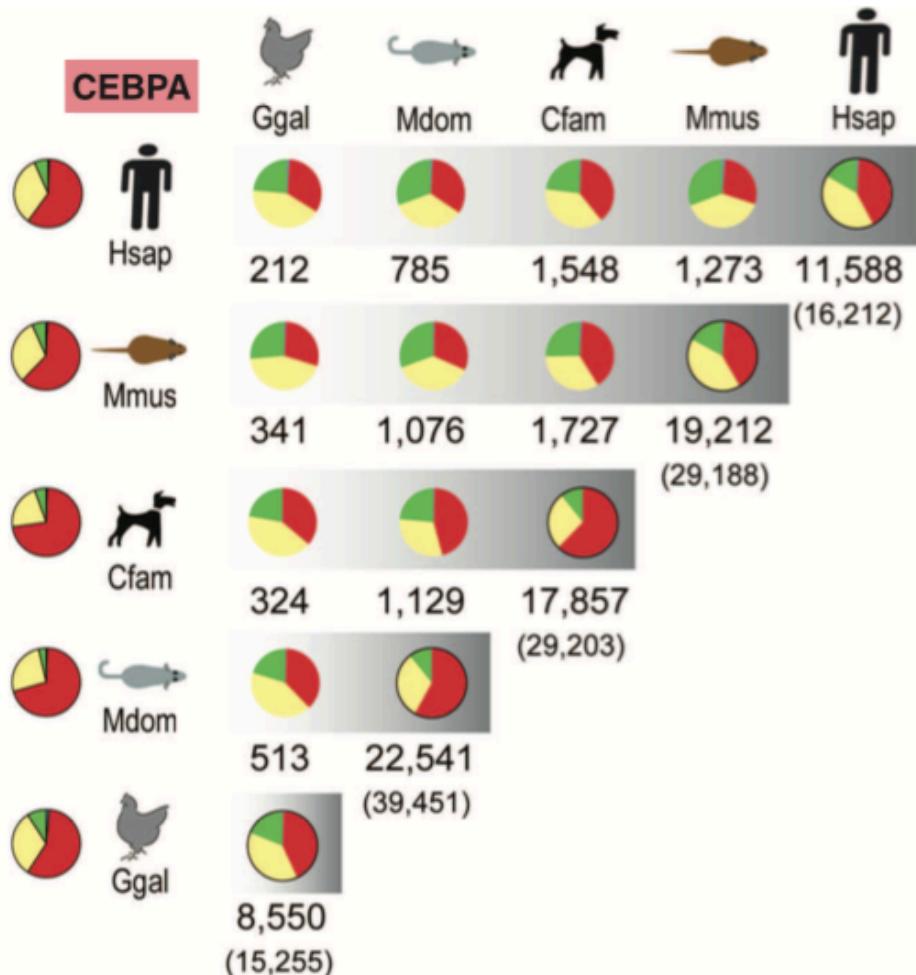
C



D

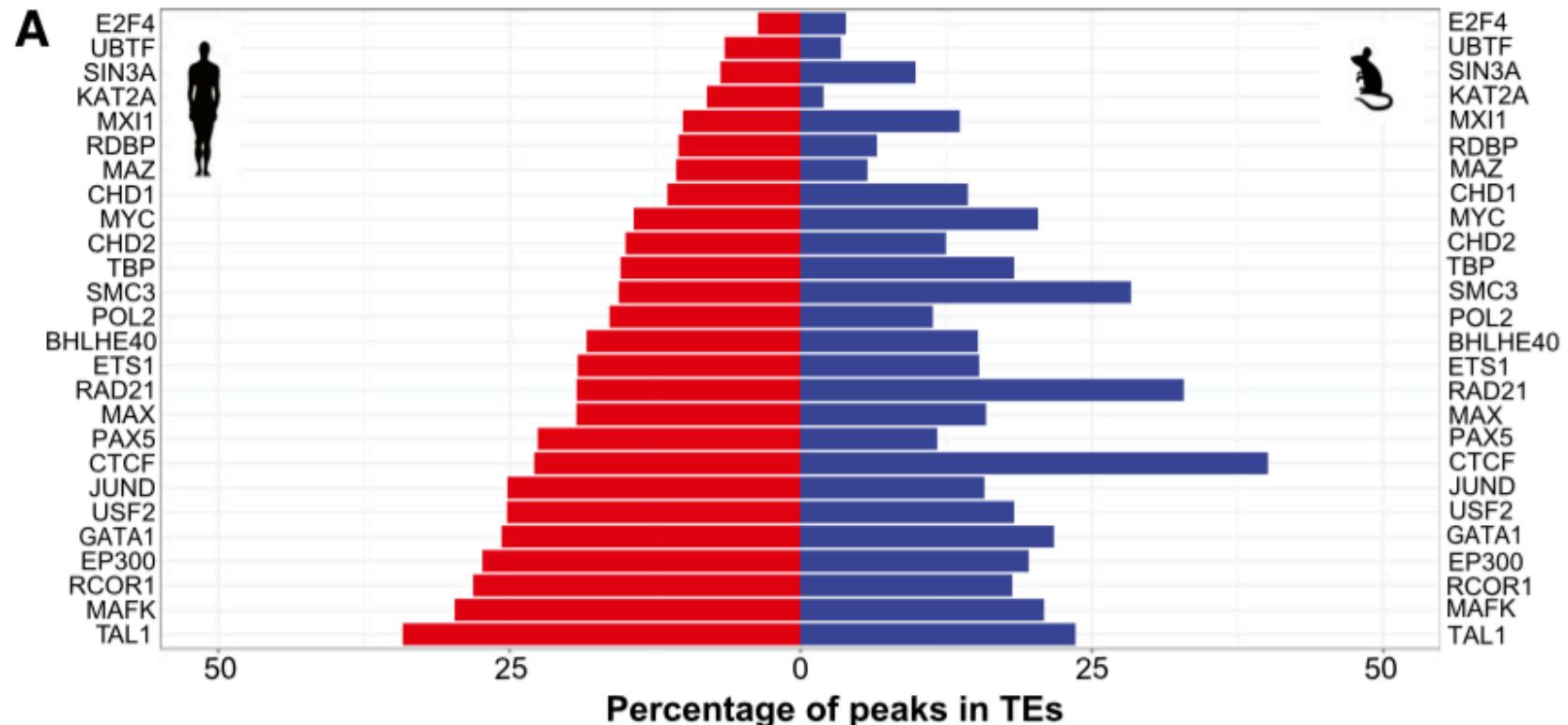


TF binding at orthologs



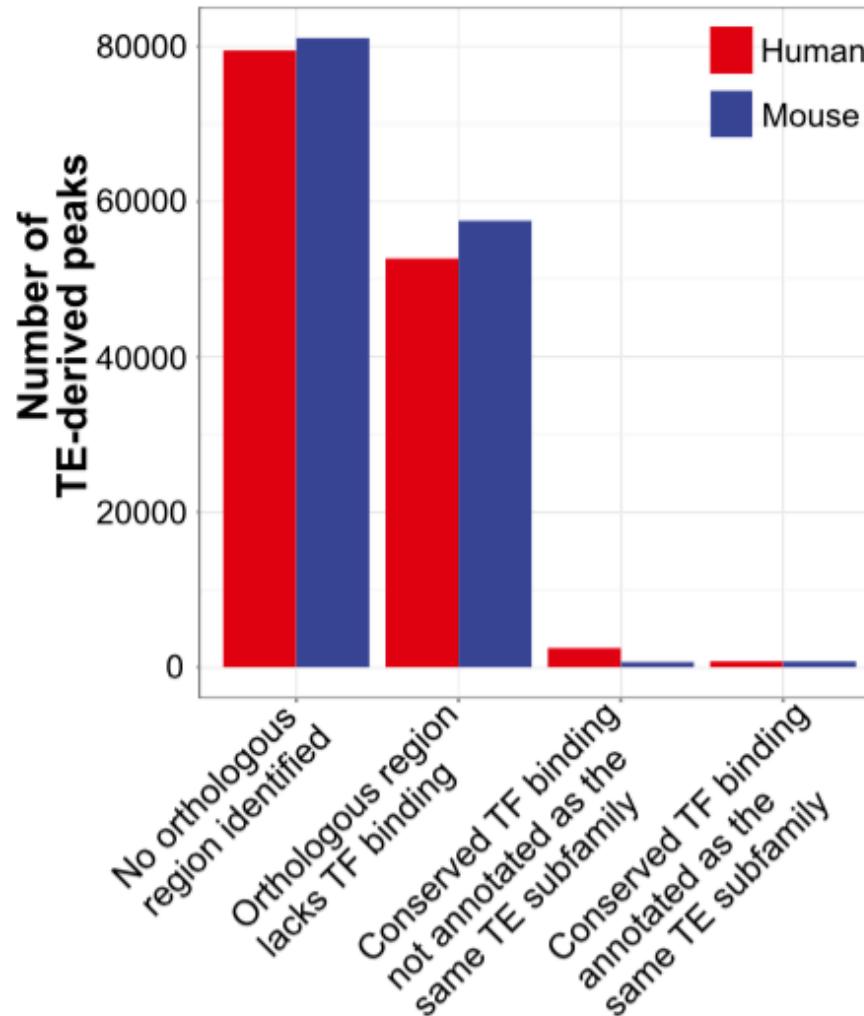
© Schmidt

2-40% of TF binding peaks occur in transposable elements

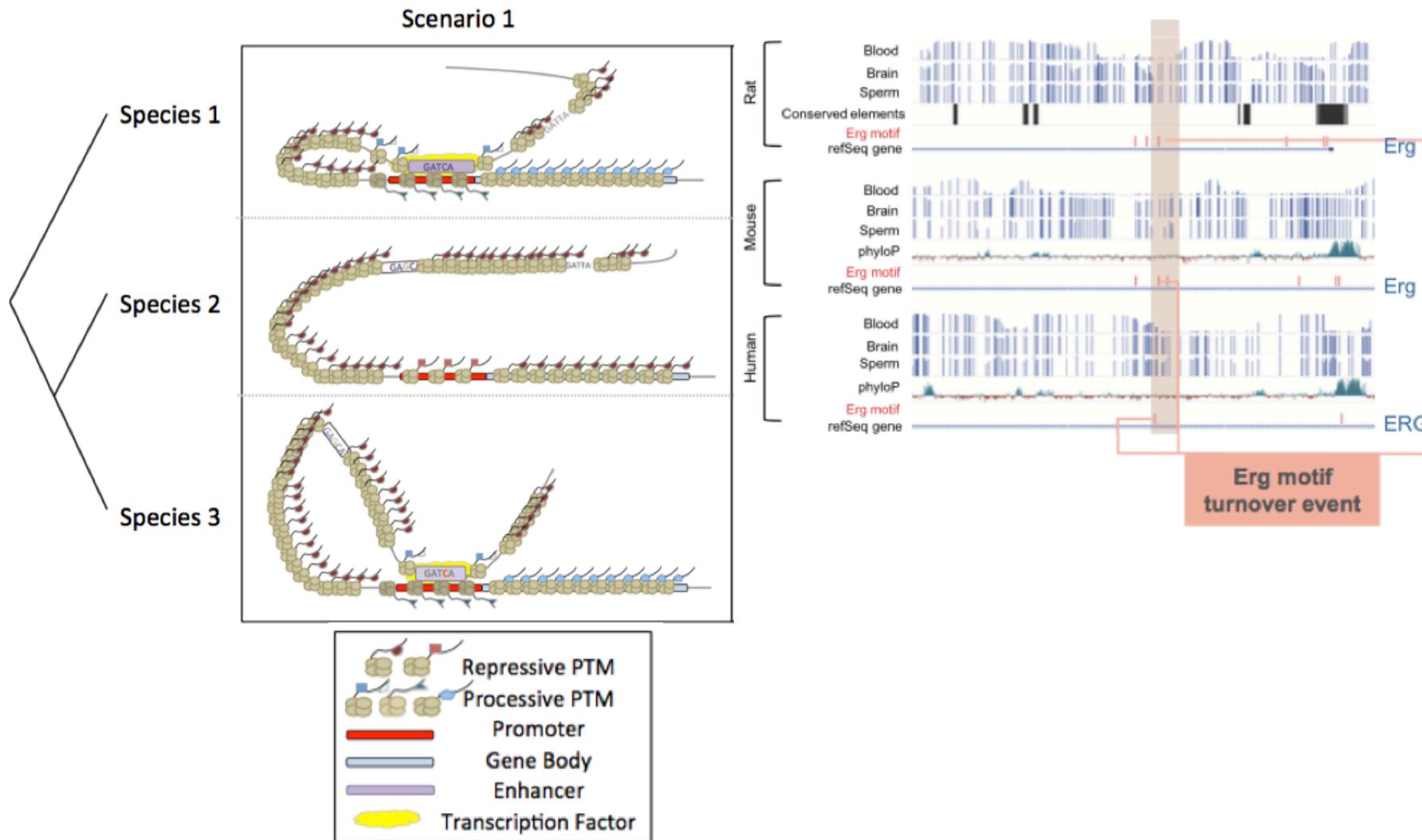


TF binding peaks derived from TEs tend to be species-specific

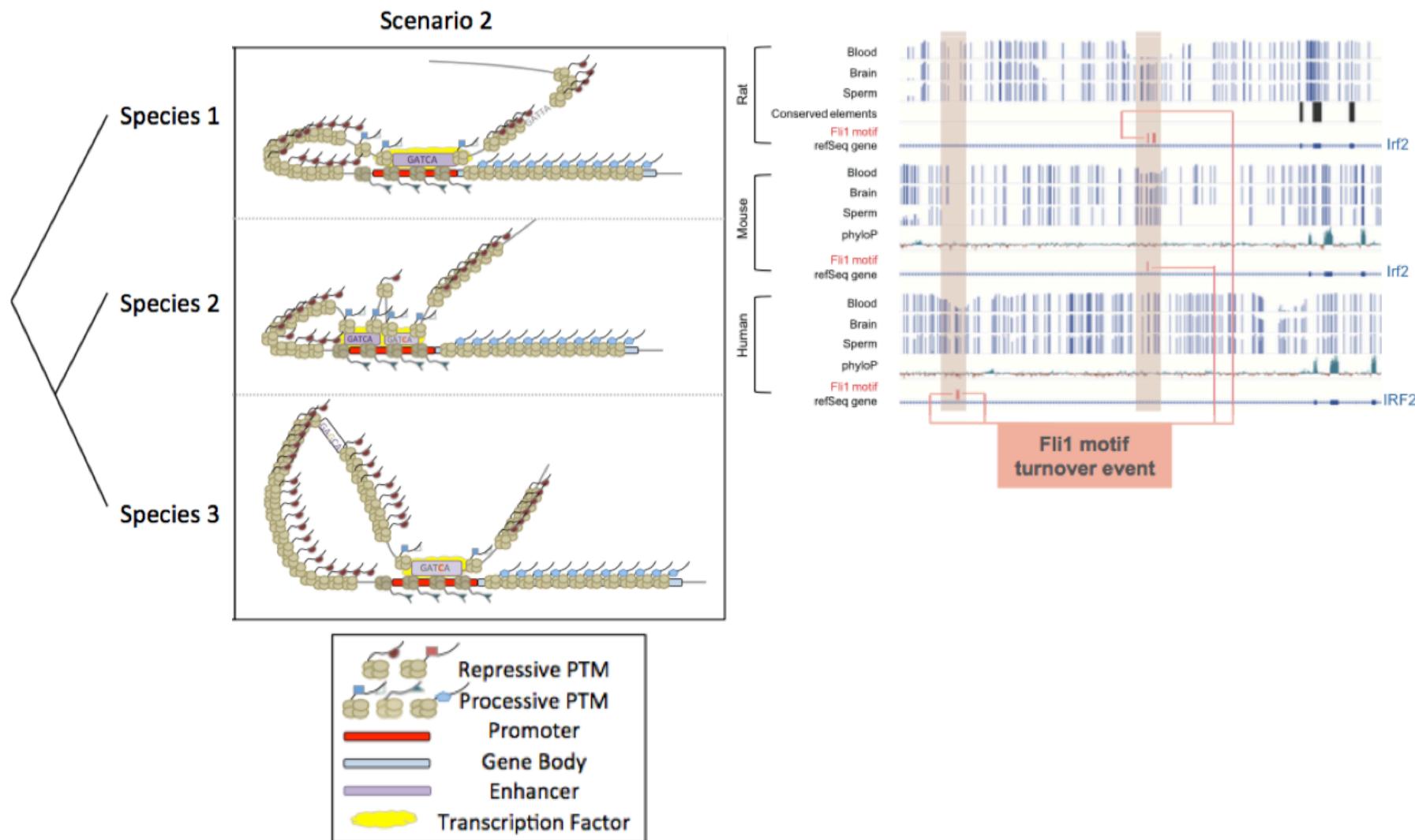
A



TFBS turnover mediates epigenome evolution at regulatory elements



TFBS turnover mediates epigenome evolution at regulatory elements



Framework for studying epigenome evolution of transcriptional regulation

