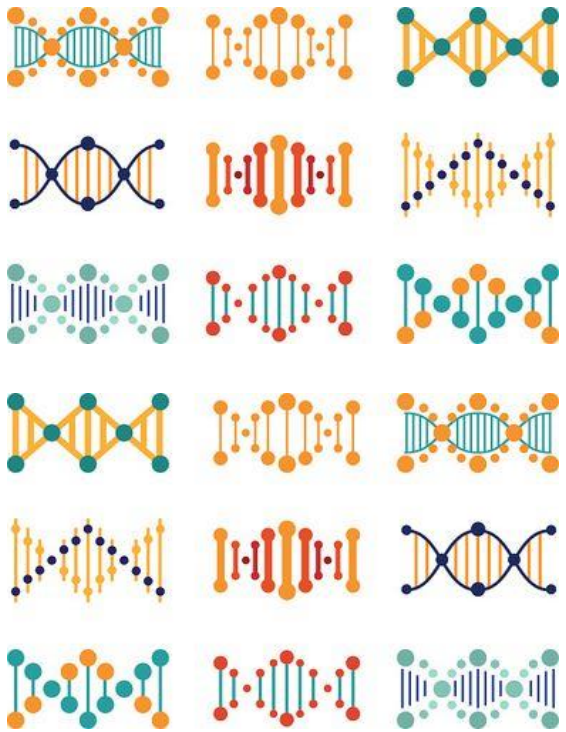

Exploring Distal Enhancers Through Histone SNPs

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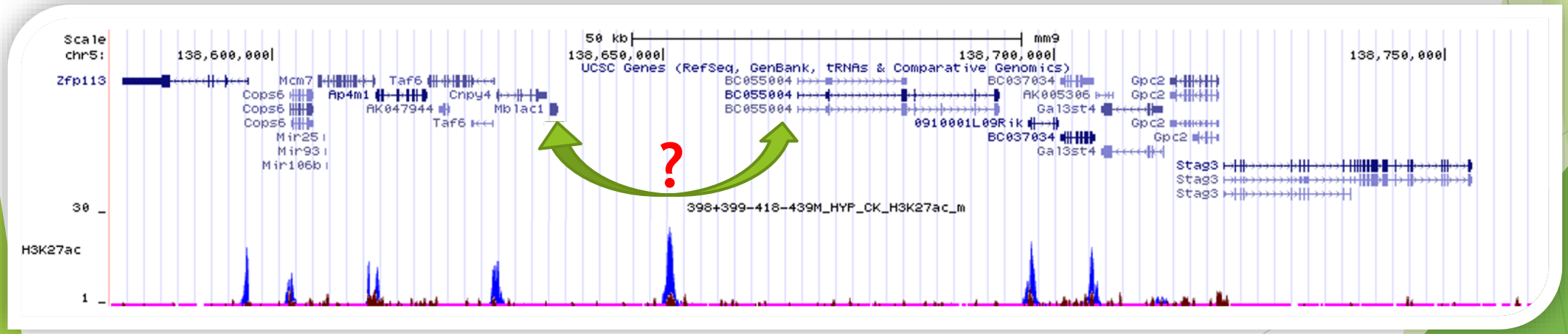
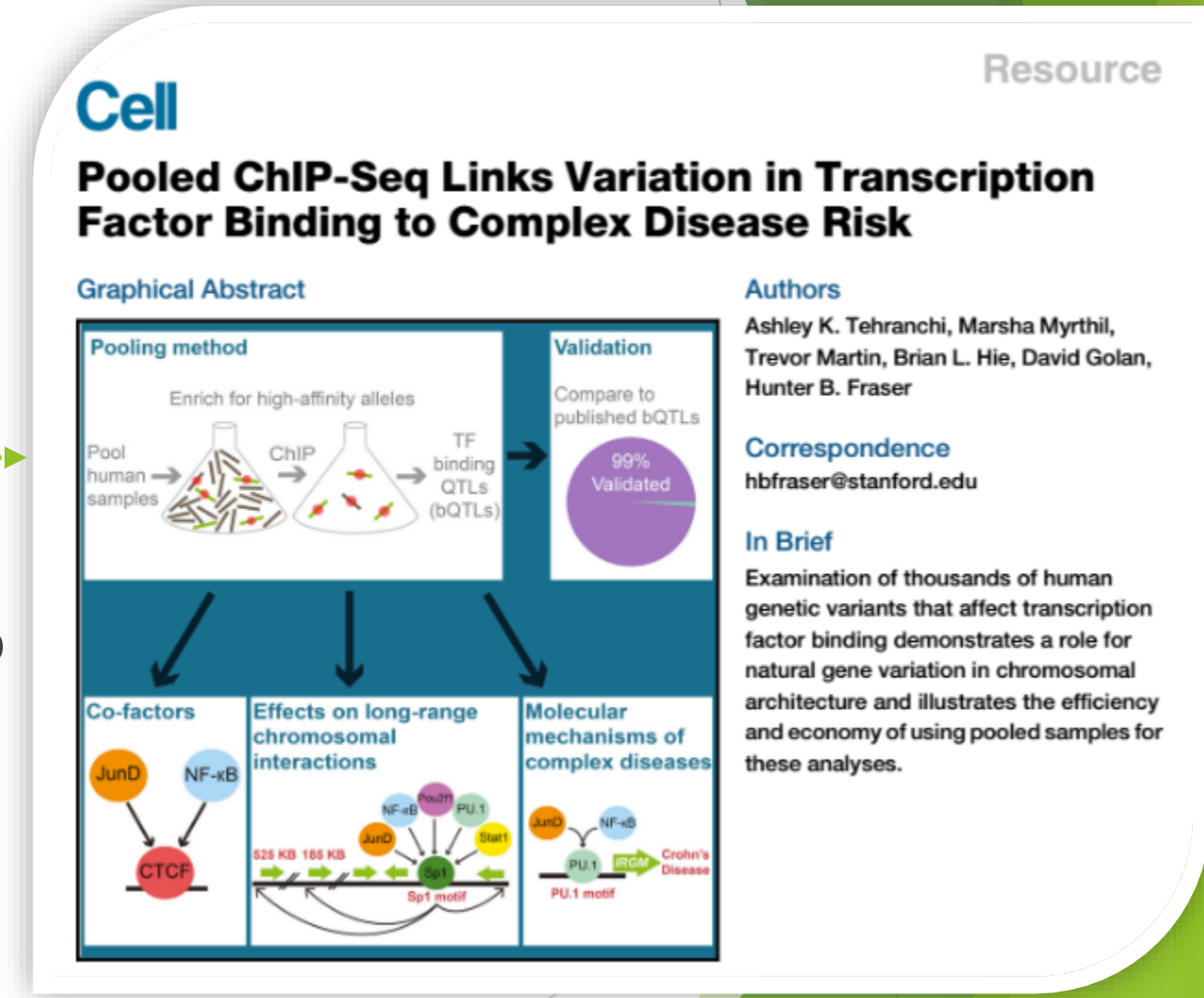
Molecular Roots of the Social Brain

- Project to identify **gene networks** that respond to **social stimuli** in mice, honeybees, and stickleback fish.
- Utilized **RNASeq** and **ChIP-seq** after social stimulus to identify differentially expressed genes and differentially accessible regulatory elements
- In Honeybee, studies revealed stronger differences in histone modifications **between colonies**, than due to social stimuli
- Additionally, it is **always** difficult to assign distal enhancers to the genes they regulate



Differential Histone SNPs

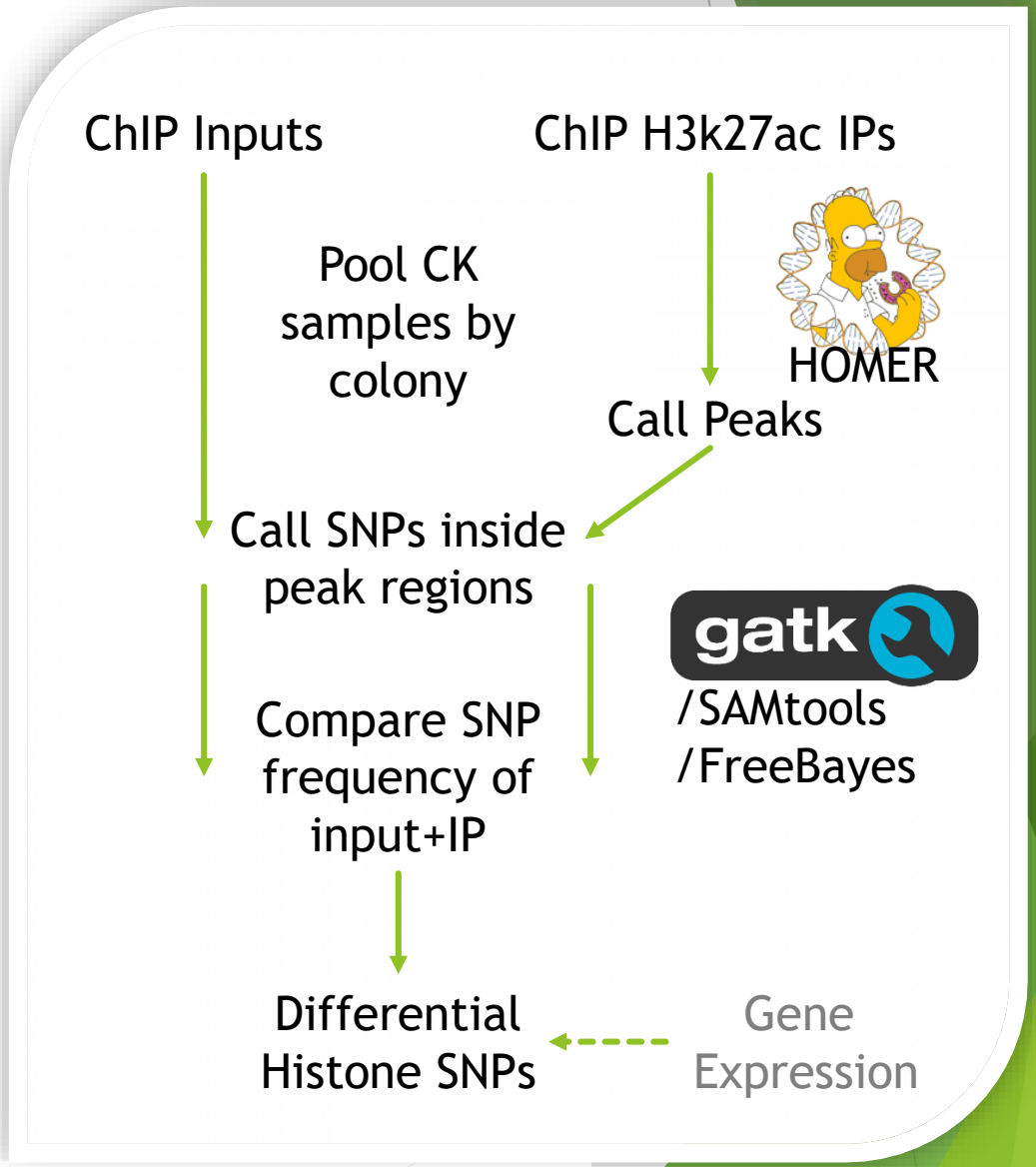
- Can SNPs explain differences we see between colonies in histone peaks?
- Recent study in humans examined **SNP preference** in pooled histone and TF ChIP peaks
- Can we apply a similar approach to data we have **already collected** from honey bee and mouse?
- Can we also use this data to help **link distal peaks to genes**?



Differentially Enriched Histone SNPs Pipeline

- When we do ChIP we collect input DNA, basically genomic DNA
- Pooling of all input DNA gives ~30x coverage ➔ call SNPs in population

- Also pooled control H3k27ac peaks from each colony ➔ call peaks
- Then checked SNP frequency in gDNA vs. inside Histone peaks

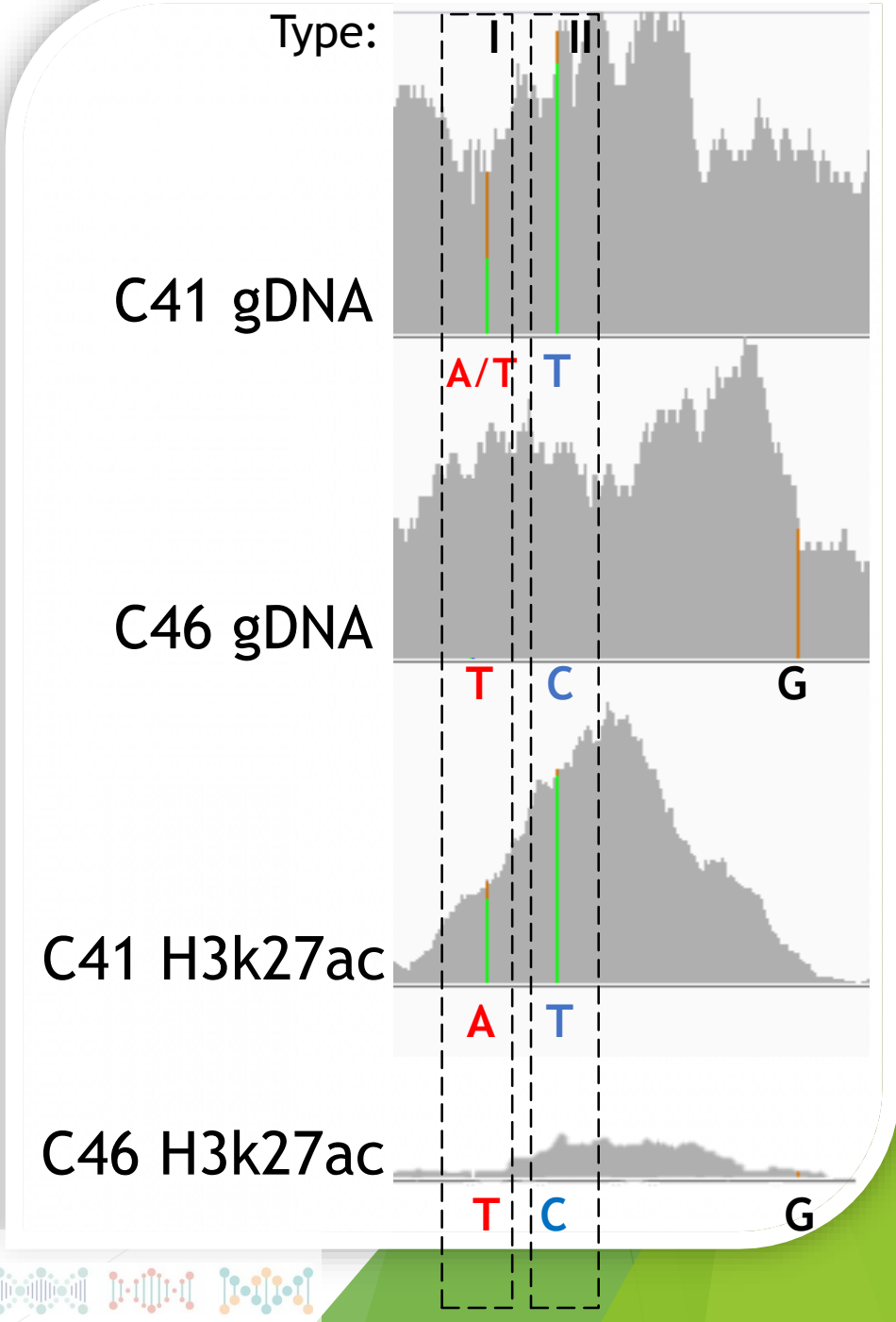


Two Possible Histone SNP Effects

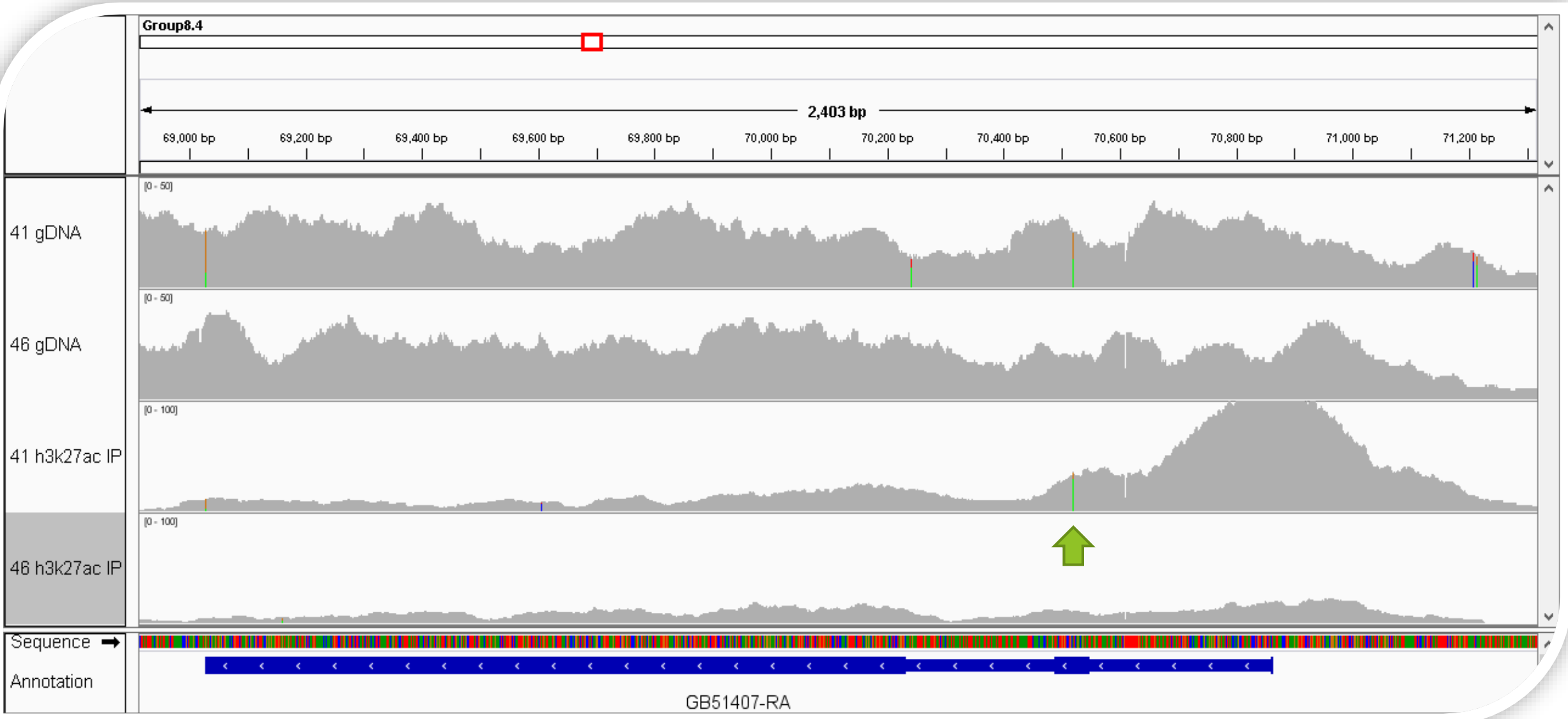
I. Genomic DNA has a particular SNP frequency in the pool, eg 50%, but Histone peak has a significantly different SNP frequency, eg 90%

II. One of the two colonies has a SNP, but the other colony does not. Colony with the SNP has a peak, but the other colony does not.

Variation could be from heterozygous DNA OR from multiple individuals in pool



Example: Unnamed Zinc Finger

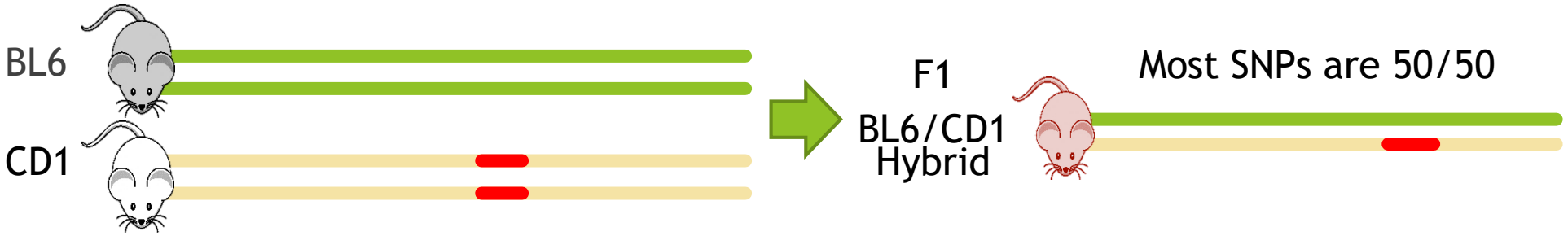


Histone peak is only present in colony that has SNPs

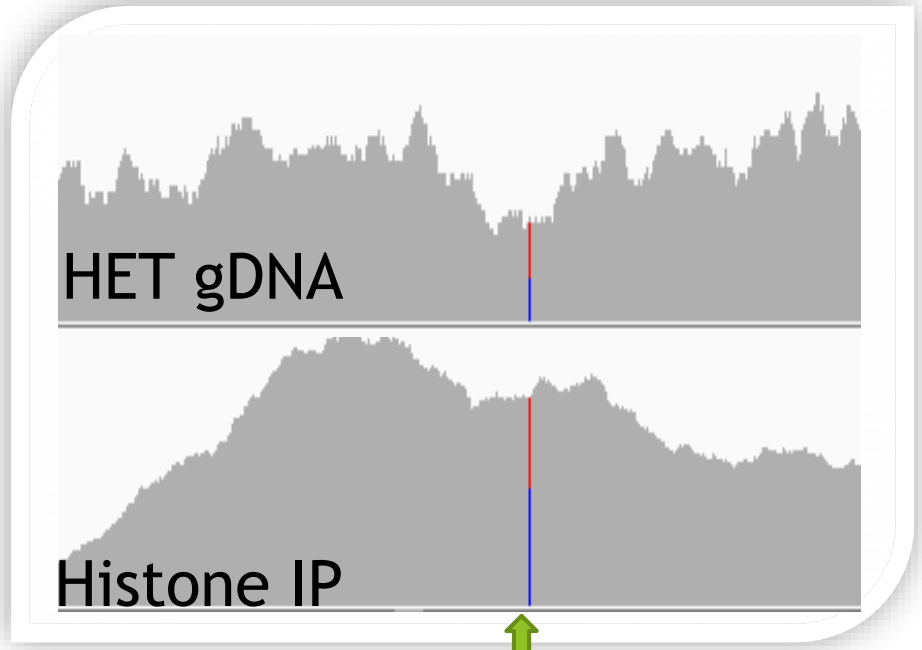
One SNP allele is preferentially found in 83% of histone reads vs. 48% of genomic reads

Second Approach: BL6/CD1 Hybrid Histone SNPs

- Generating a Hybrid Mouse line allows for testing with less coverage

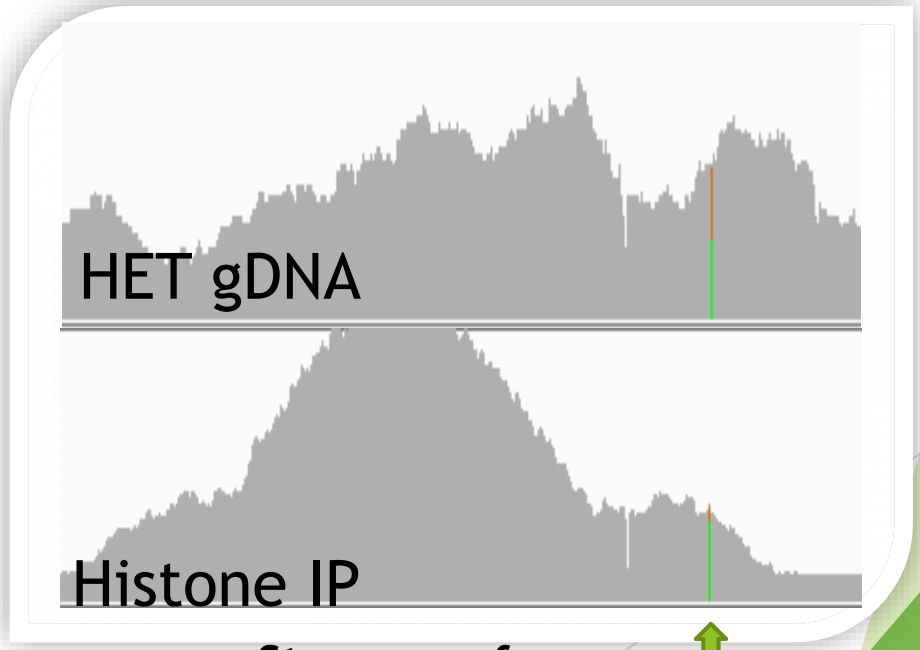


Expected 50/50 non-Influencing SNP



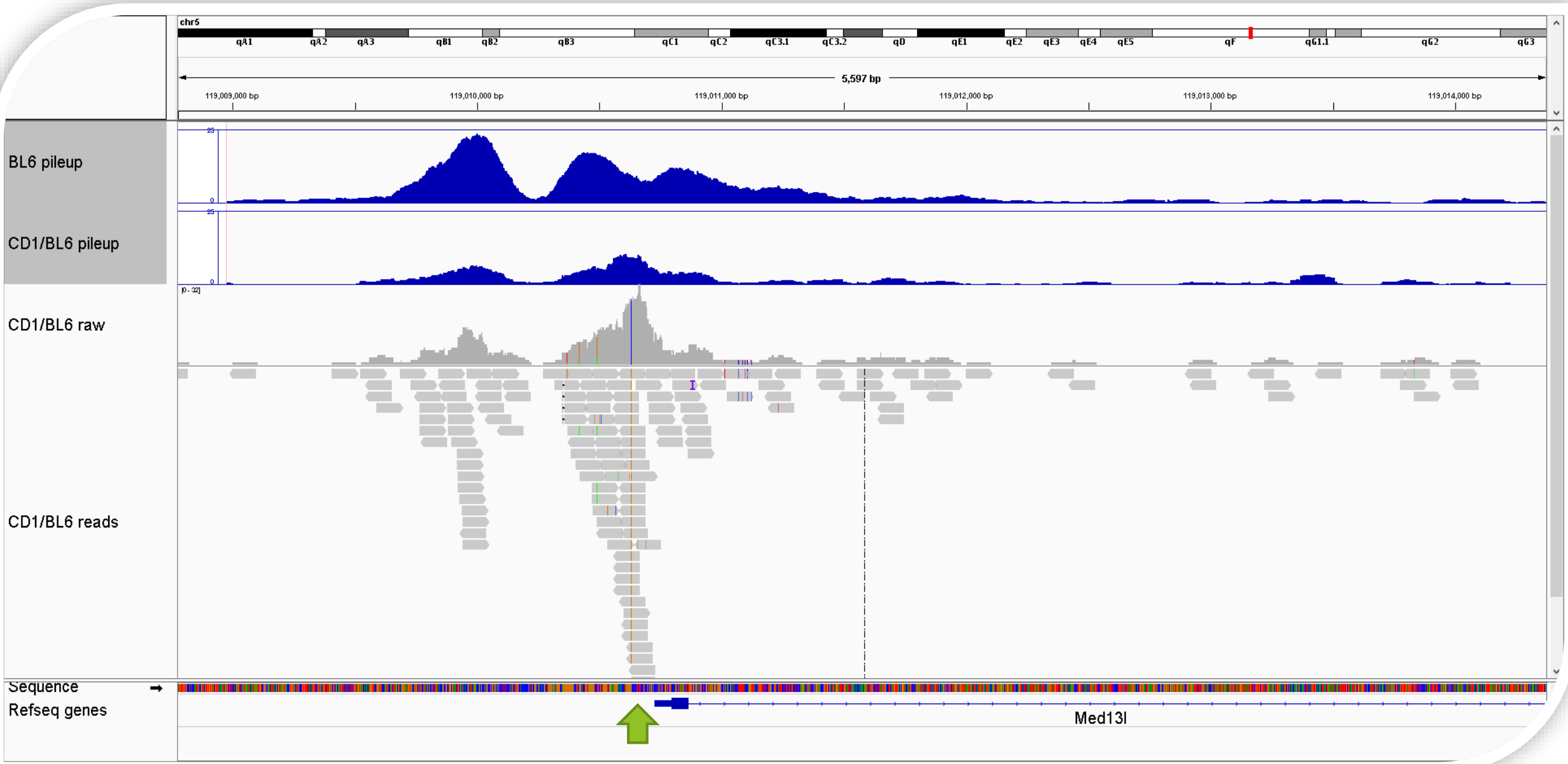
No preference

Histone Influencing SNP



Strong preference for one genotype

Example: Med13l (100/0 SNP)



Future Directions

- Re-run improved Histone SNP calling pipeline on honeybee dataset, taking **ploidy** into account
- Bring in **gene expression data** to see if Histone SNPs also change gene expression pattern of nearby genes, allowing us to link **distal peaks** to the genes they regulate
- Identify co-incidence of Histone SNPs with **regulatory motifs**
- Use **CRISPR** to test hypotheses and **validate** methods *in vivo*
  - Eg. Introduce a SNP predicted to be influential to a primary cell line and see if that alters nearby chromatin and expression

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