Inference of 3D regulatory interactions from 2D genomic data

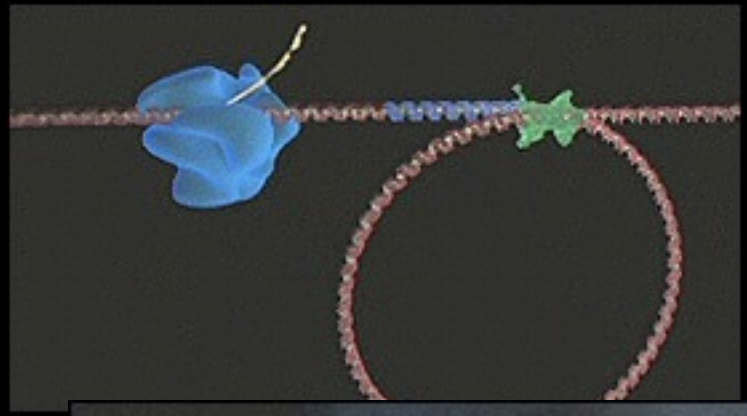
Katie Pollard

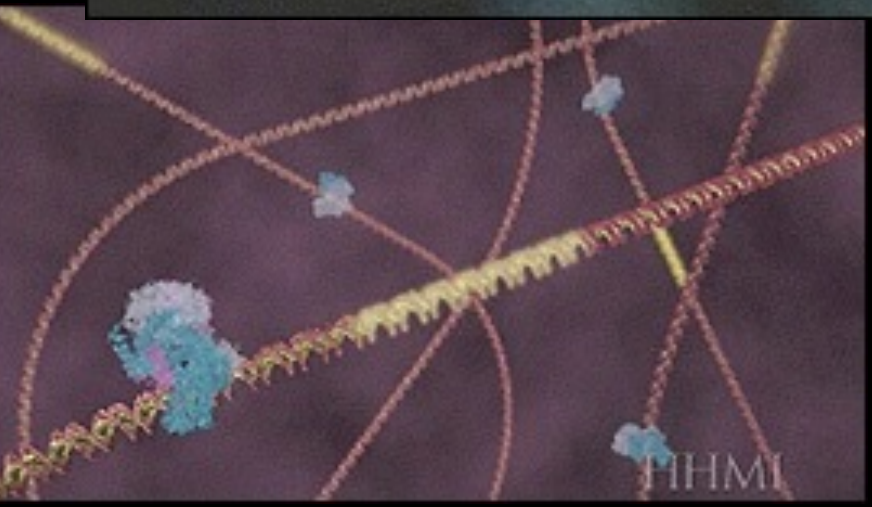
**Gladstone Institutes, Institute for Human Genetics,
Division of Biostatistics - UCSF**

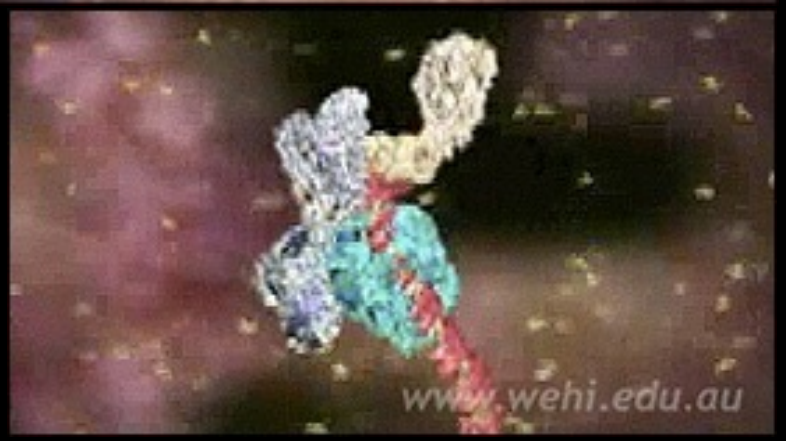
ENCODE Users Meeting

Bolger Center - June 30, 2015

Eukaryotic gene regulation is 3D and complex







Eric Keller

Drew Berry

Most phenotype associated mutations are outside coding regions

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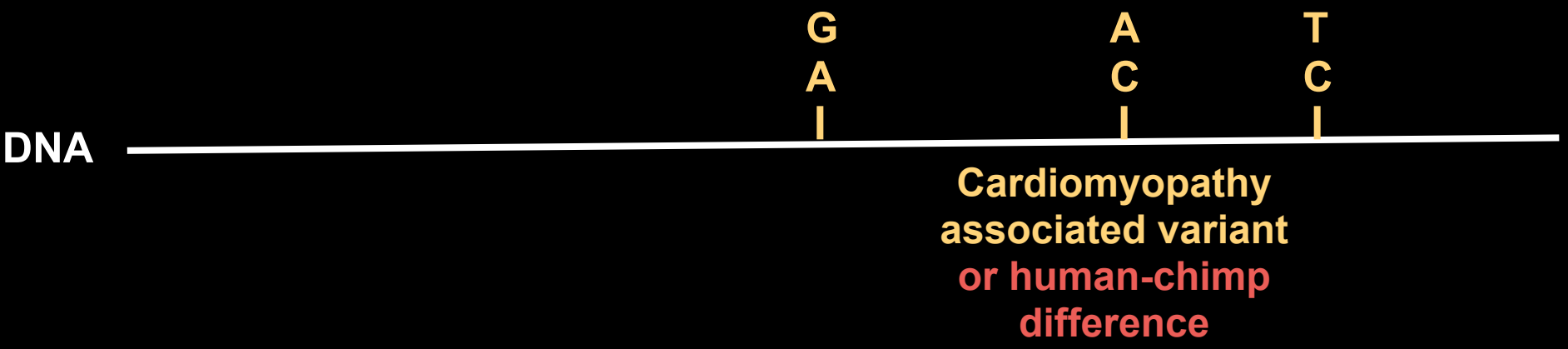

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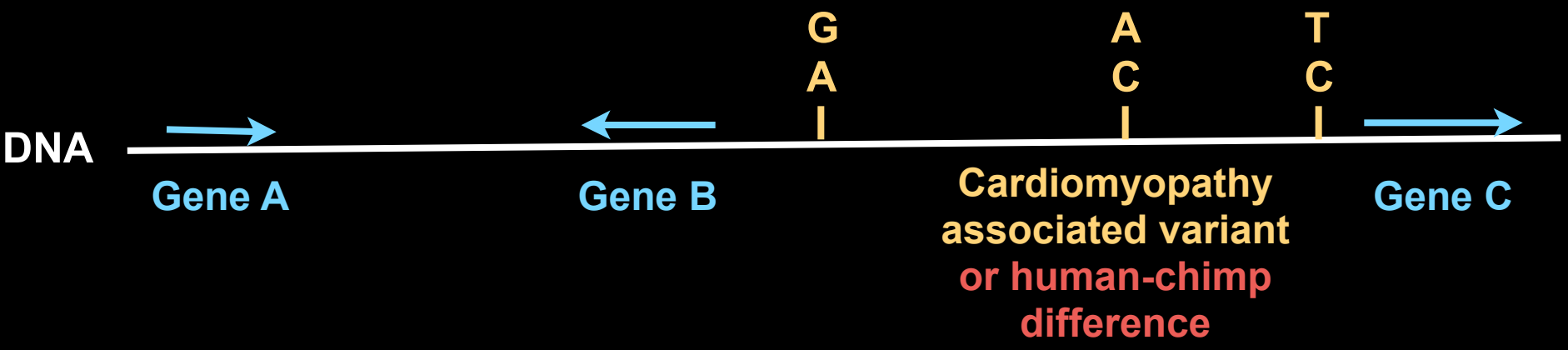

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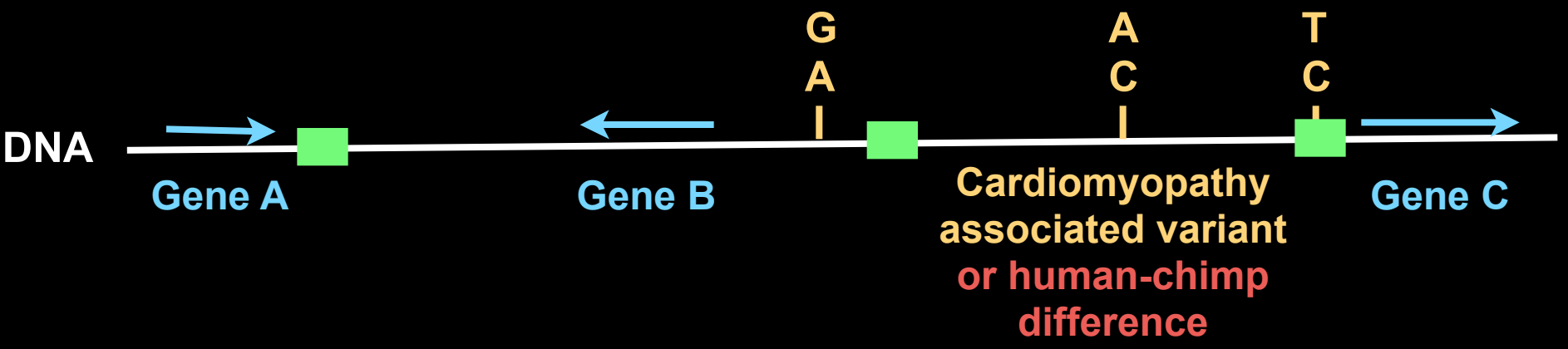

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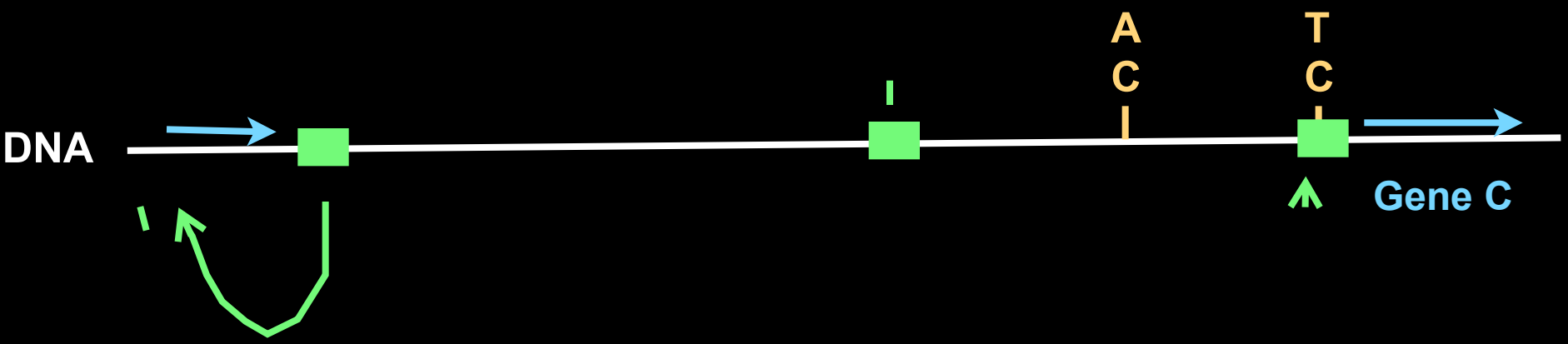

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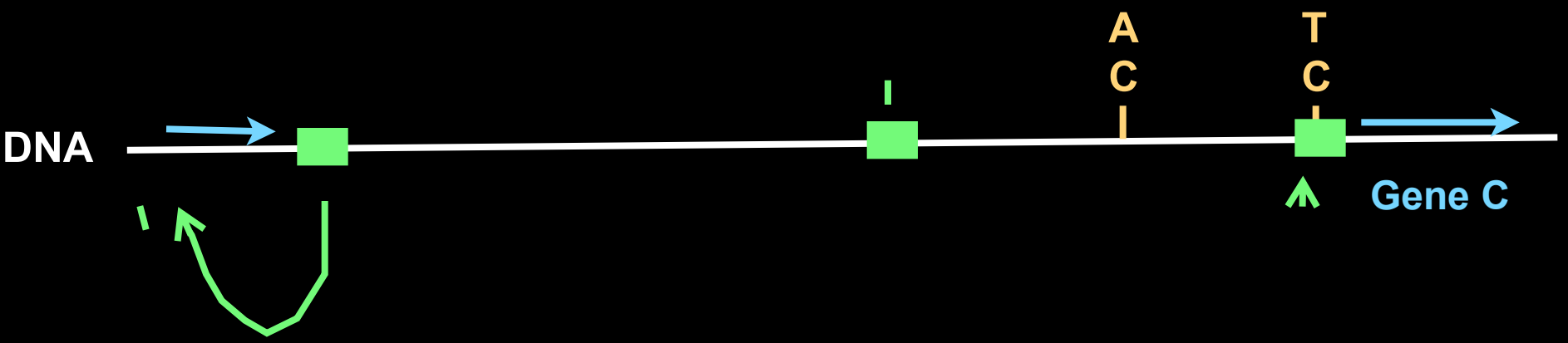
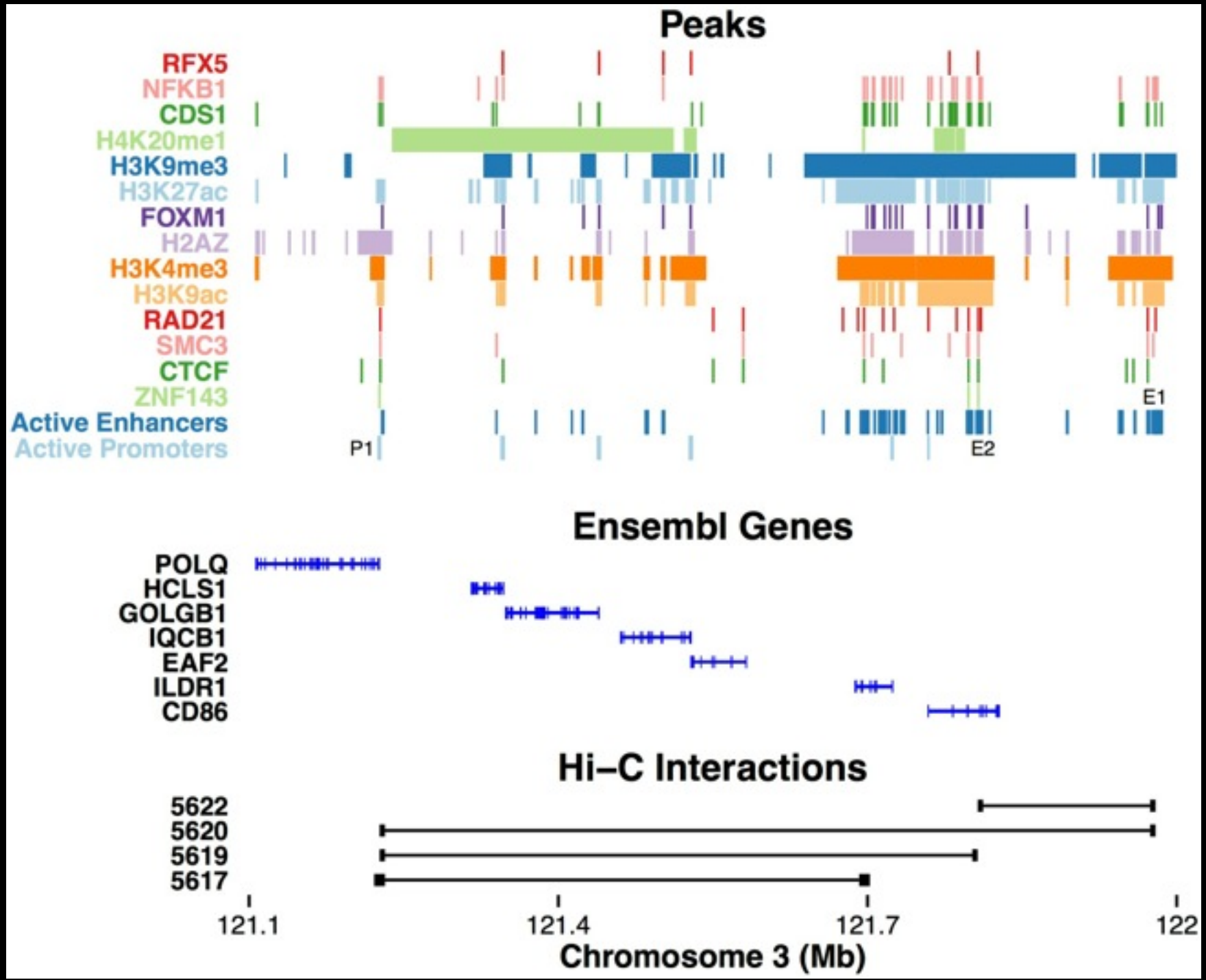


- **Enhancers:** individual ChIP-seq data sets identify <50% of known enhancers, plus many false positives
- **Gene targets:** closest gene is right ~10% of time

Can we reconstruct 3D interactions between enhancers and promoters from 2D genomic data?


Peaks
RFX5
NFKB1
CDS1
H4K20me1
H3K9me3
H3K27ac
FOXM1
H2AZ
H3K4me3
H3K9ac
RAD21
SMC3
CTCF
ZNF143
Active Enhancers
Active Promoters
P1
E1
E2
Ensembl Genes
POLQ
HCLS1
GOLGB1
IQCB1
EAF2
ILDR1
CD86
Hi–C Interactions
5622
5620
5619
5617
121.1
121.4
121.7
122
Chromosome 3 (Mb)

TargetFinder: Training

Teach a machine learning algorithm to discriminate true versus false enhancer-promoter interactions based on their features.

Training Data

Active enhancer expressed gene

Positives = Hi-C +

Negatives = Hi-C -

Rao et al 2014, 1-Kb resolution

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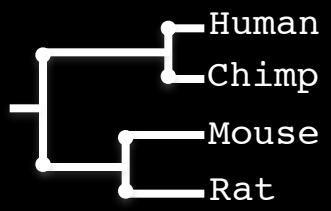
Evolutionary Conservation



Conserved synteny of enhancer and promoter

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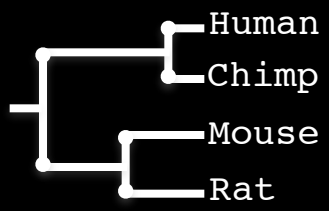

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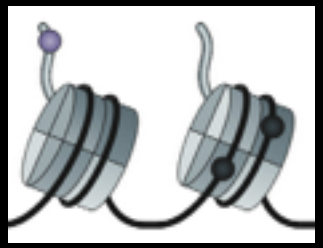
Functional Genomics



ChIA-PET

RNA-seq

ChIP-seq (TFs, histones)

enhancer/promoter/window

ChromHMM & Segway

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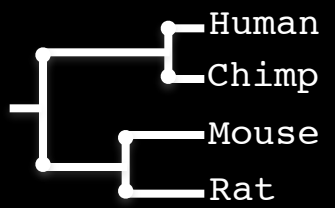

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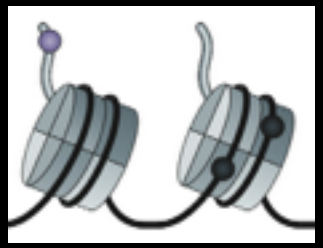

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K-mer correlation
Annotated functions and pathways of gene and enhancer bound TFs

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Computational Algorithm

Decision trees: good for interacting features

Ensemble learning: build many imperfect classifiers and combine them to improve prediction accuracy

Features

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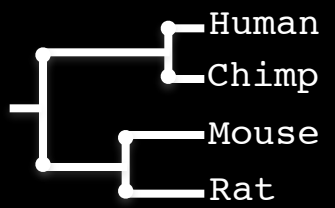

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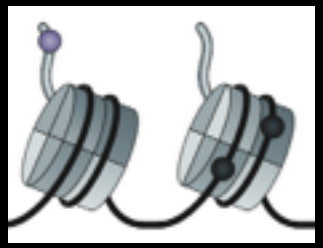

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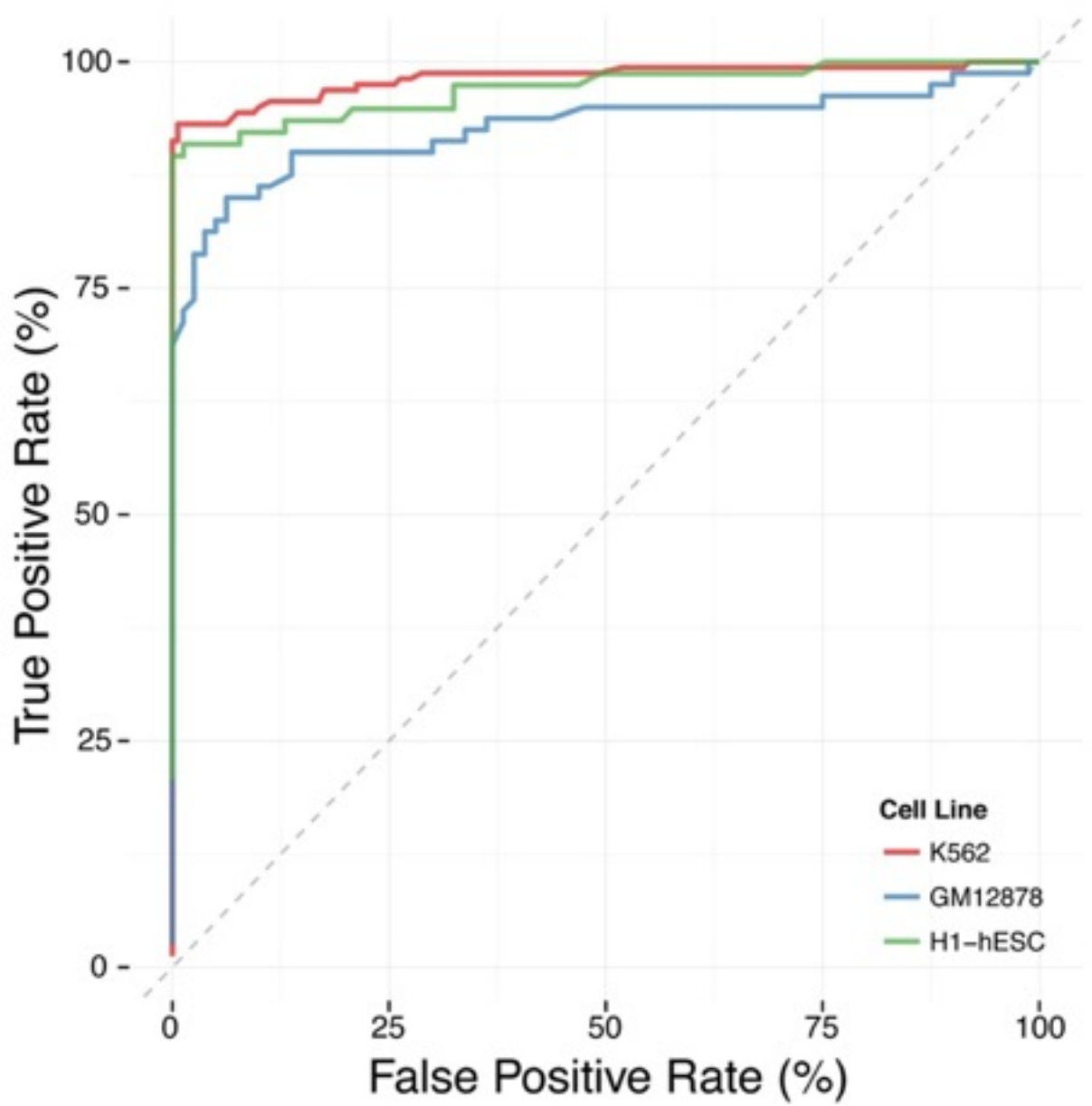


AUC=0.94-0.96

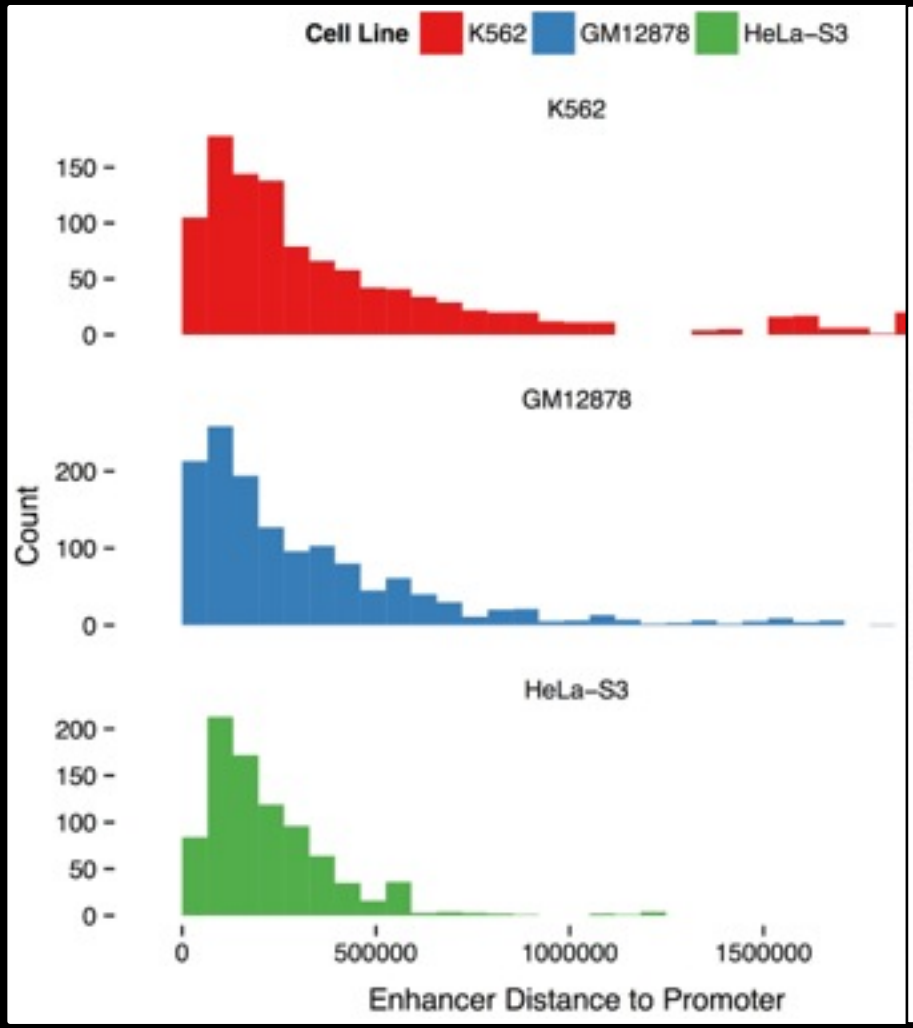
Precision =90-95%

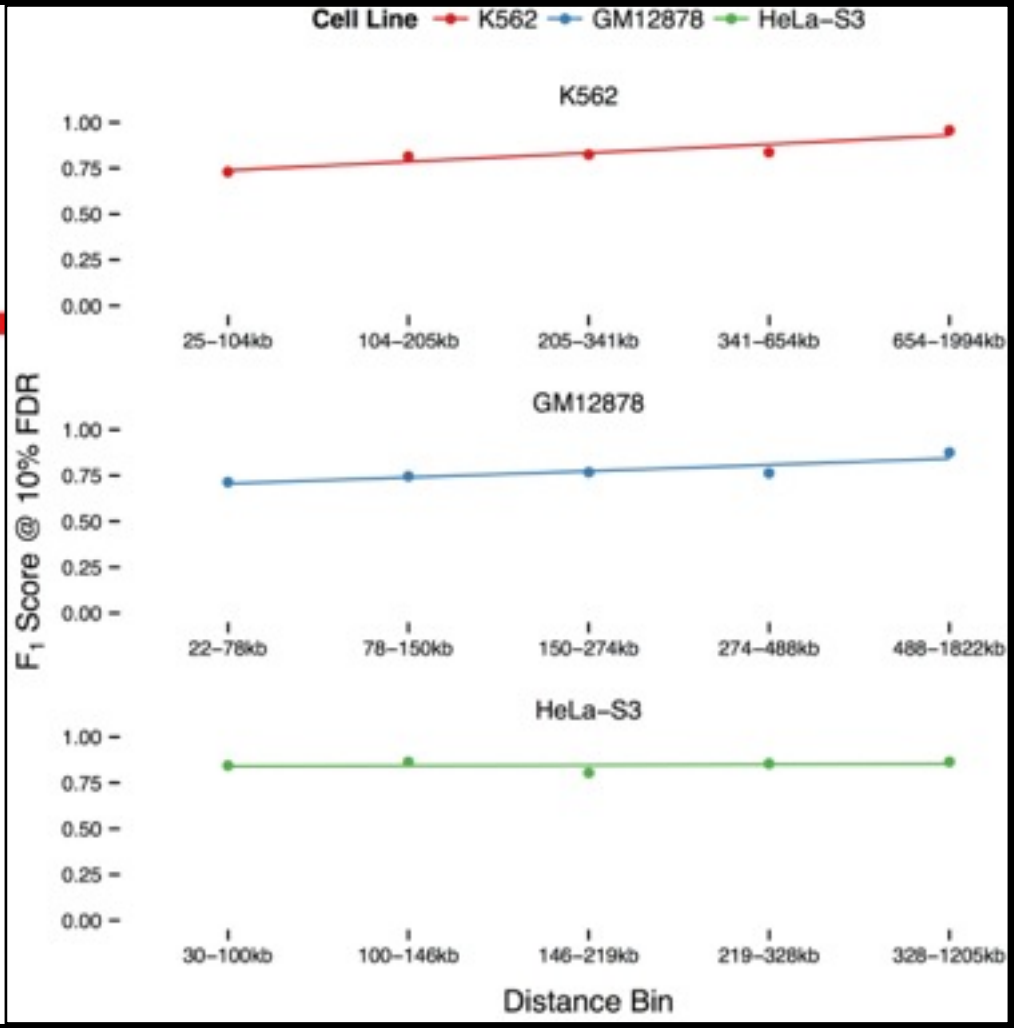
Recall=76-83%

Power=85-89% at 10% FPR

Significantly better than random and logistic regression

TargetFinder performs well at very long distances





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Most predictive features mark the *window between* the enhancer and the promoter

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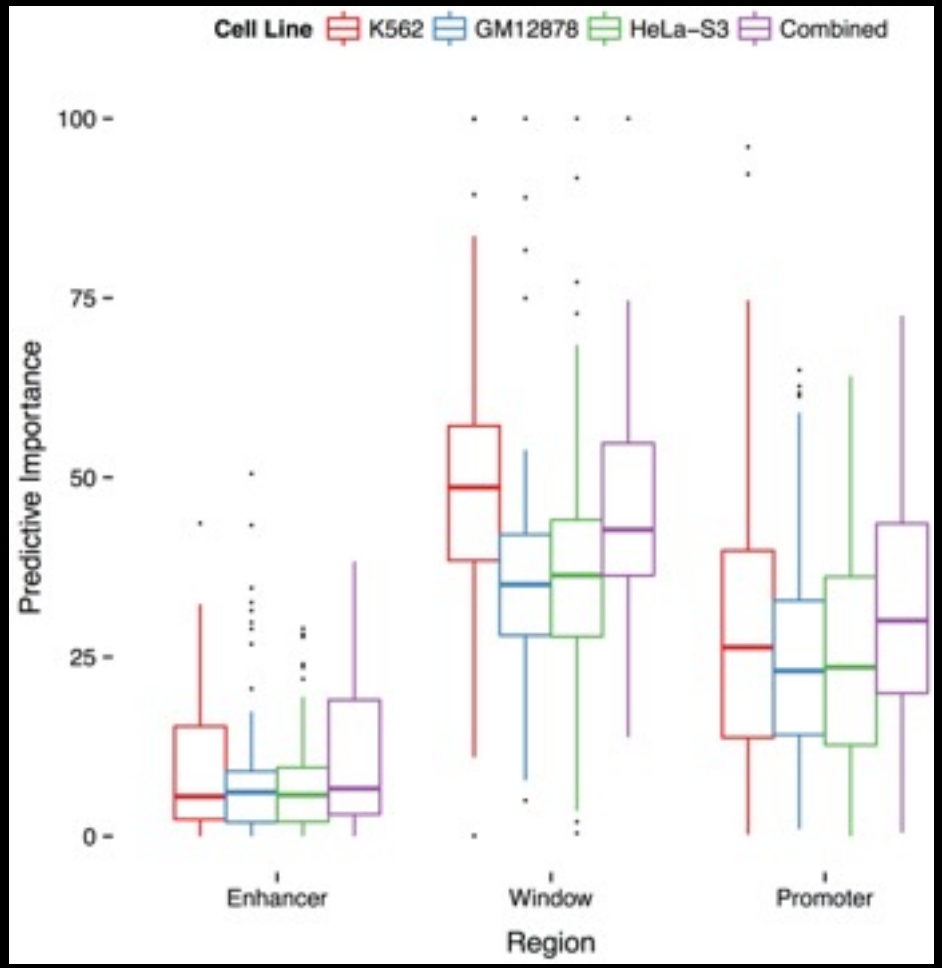

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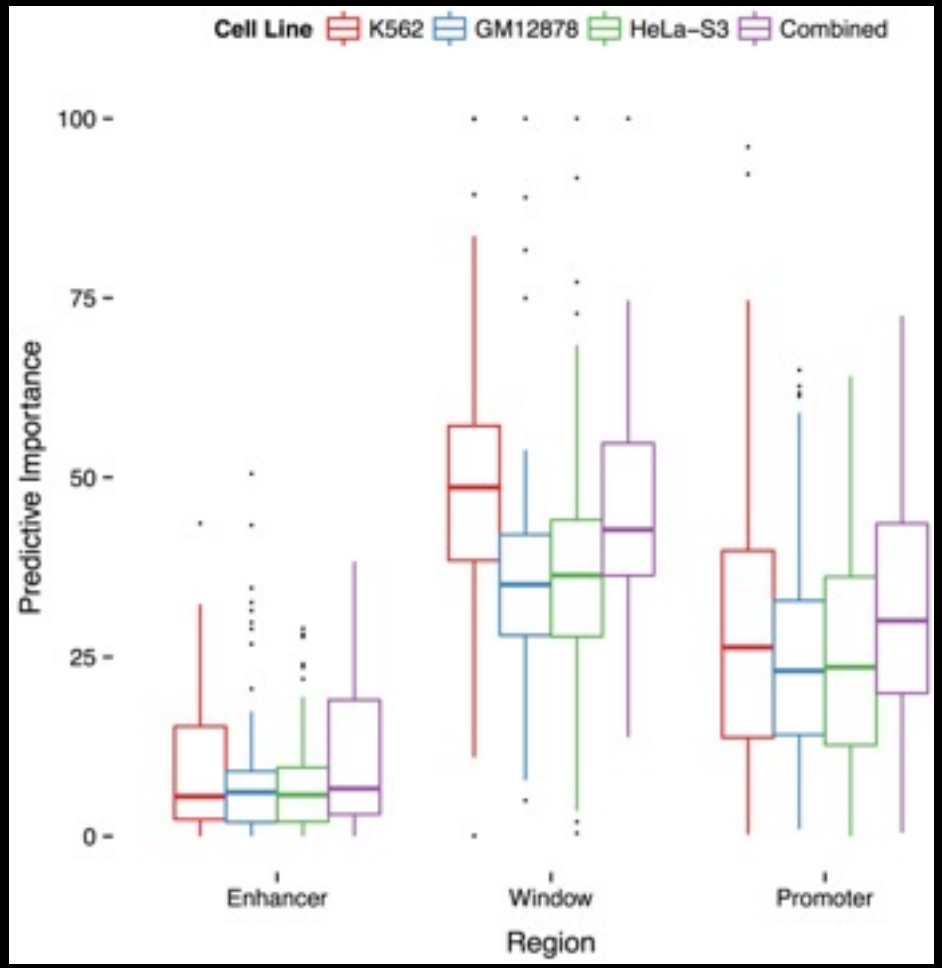

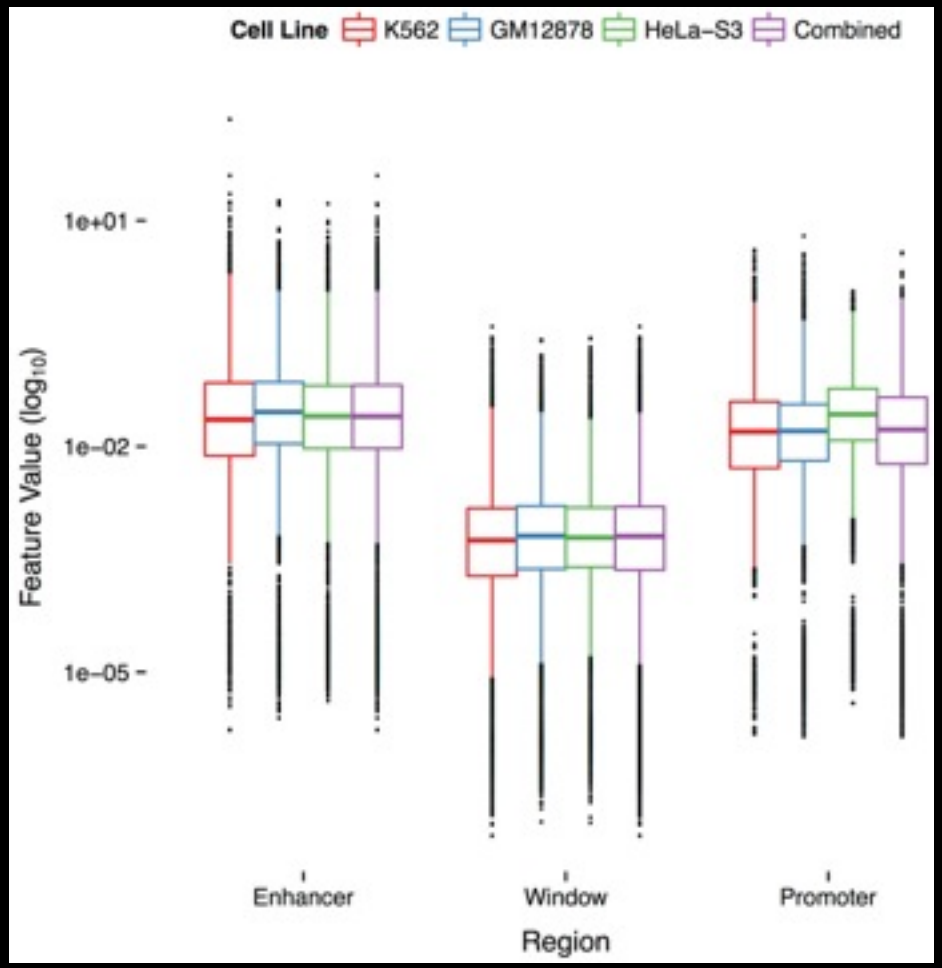

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- **True interactions**
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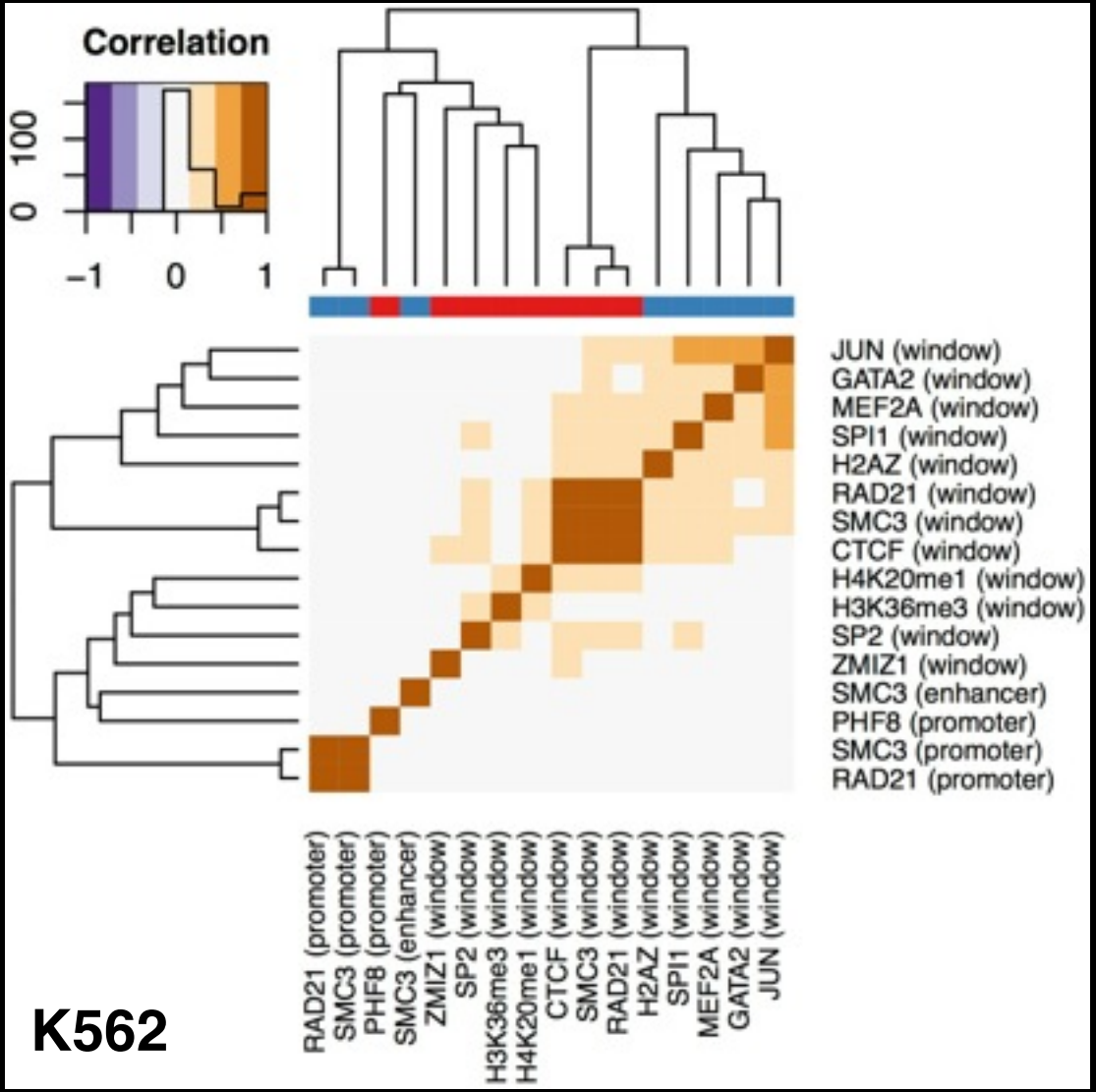
Many “window” features have a different meaning when marking promoters and enhancers (e.g., cohesin)

Predictive features colocate and form complexes



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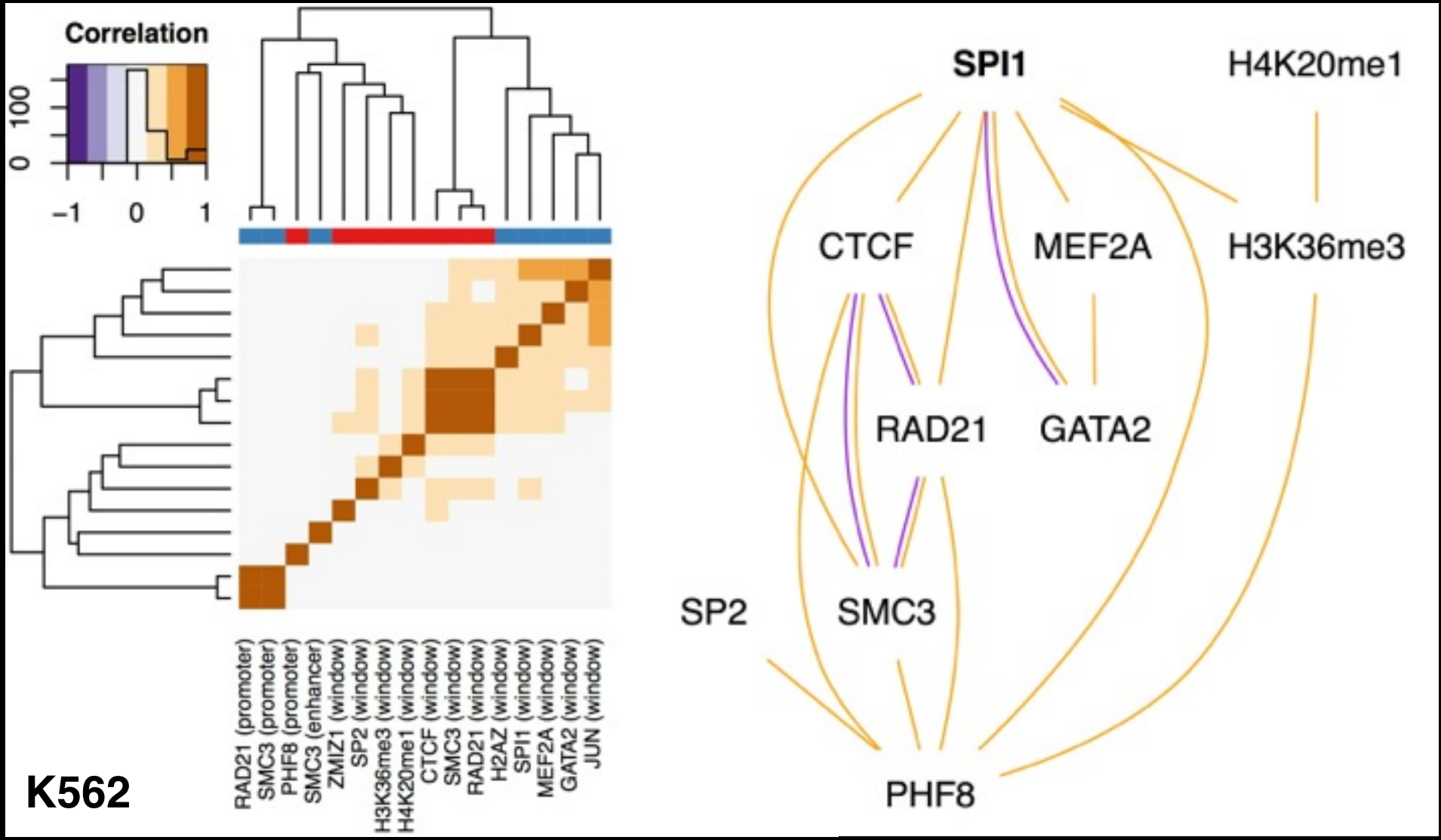

Can TargetFinder work outside ENCODE cell lines?

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What is a minimal set of experiments for accurate prediction?

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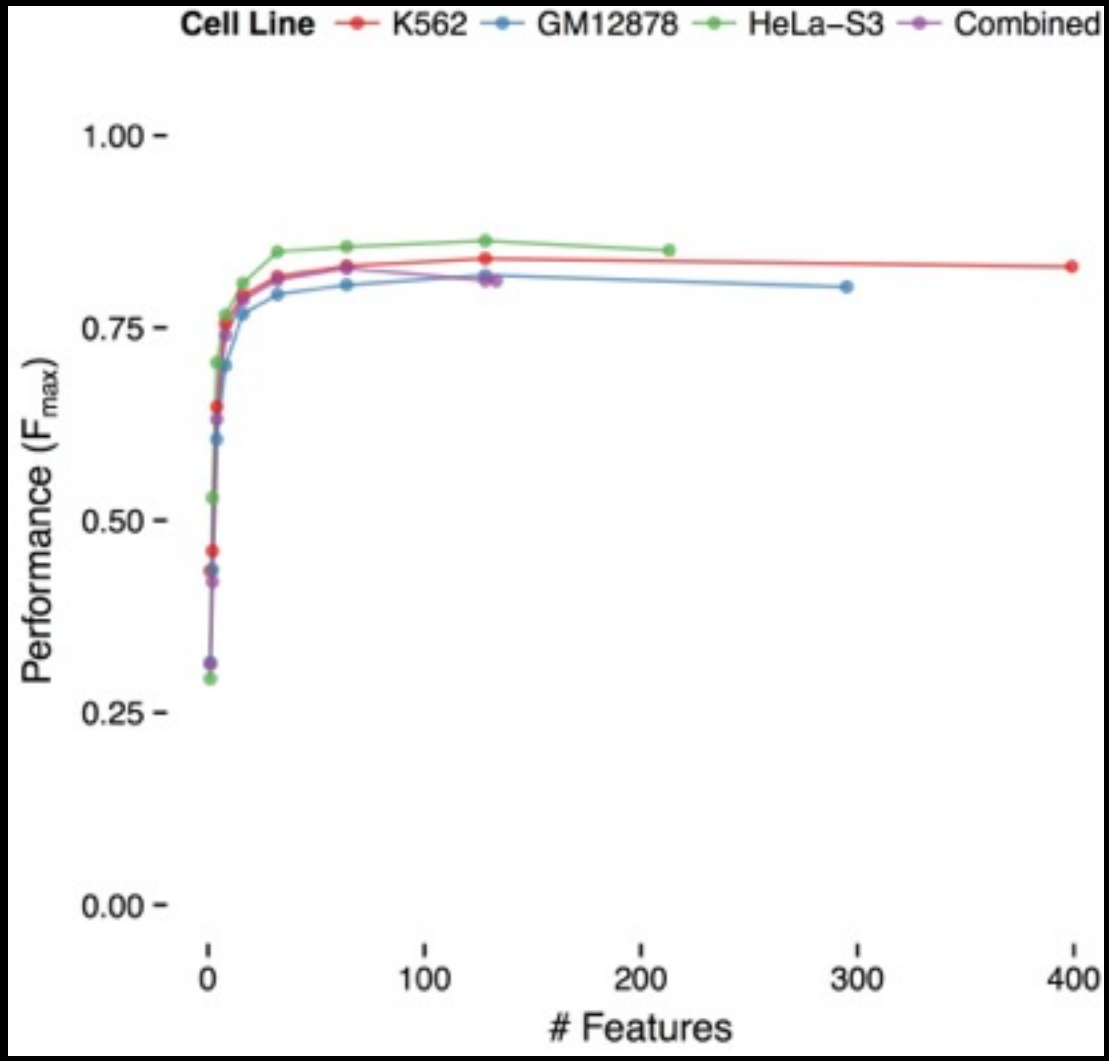
What is a minimal set of experiments for accurate prediction?

optimal: 16+

minimal: 8



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Test if models generalize across cell types

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EVALUATE

TRAIN Fmax values	GM12878	K562	HeLa-S3	HUVEC
GM12878	0.83	0.40	0.43	0.39
K562	0.46	0.85	0.45	0.44
HeLa-S3	0.43	0.38	0.88	0.41
HUVEC	0.39	0.40	0.38	-

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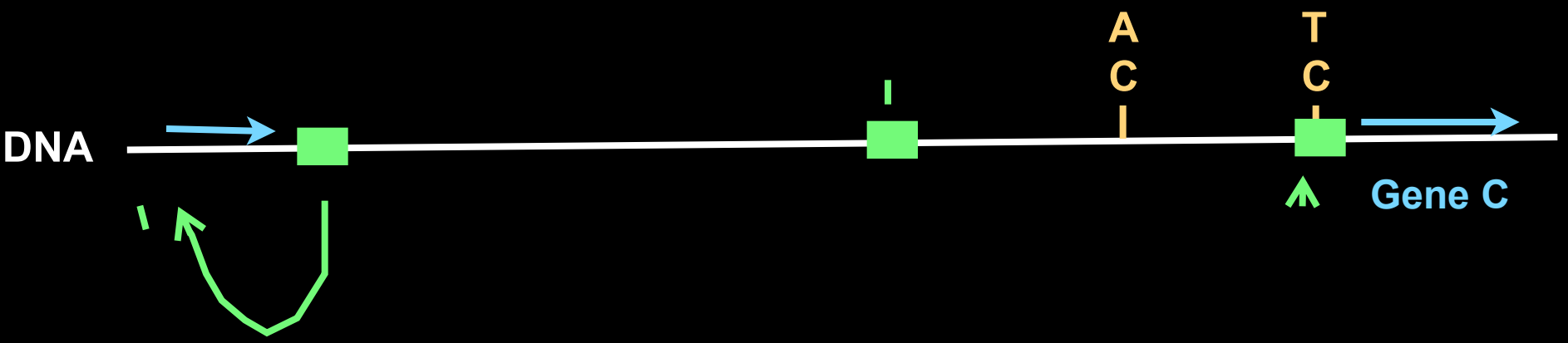
Expect ~35% precision and 55% recall on a new cell type with ~10 ChIP-seq datasets

TargetFinder accurately annotates enhancer-promoter pairs



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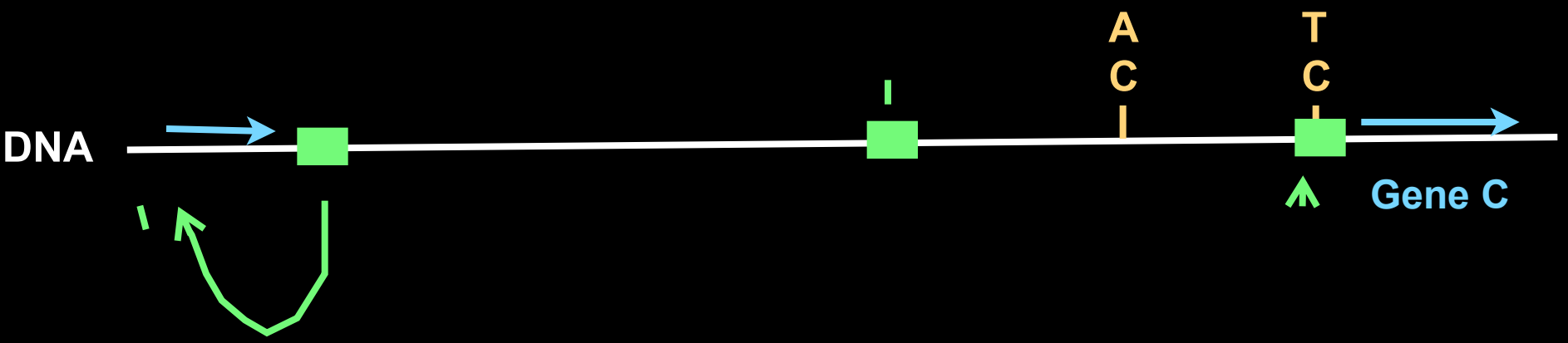

Massive data integration improves prediction

- **Closest gene**
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  - Many false positives

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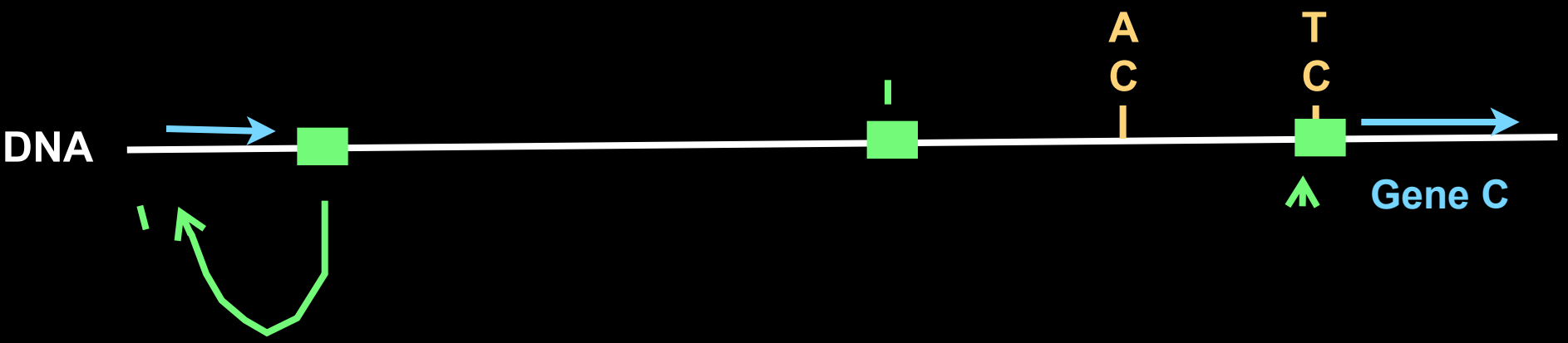

Massive data integration improves prediction

- **Closest gene**
  - Usually fails to identify the right promoter
  - Many false positives
- **TargetFinder**
  - Identifies 95-90% of known pairs (55% with less data)
  - Few false positives

Which human genome sequences function as long-range enhancers?

EnhancerFinder: Training

Teach a machine learning algorithm to identify developmental enhancers active in different tissues based on their features.

Training Data

VISTA Browser


+


–

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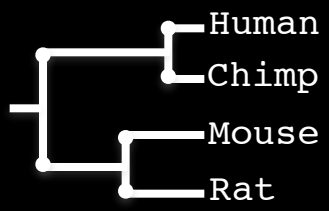

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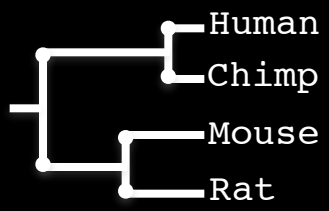

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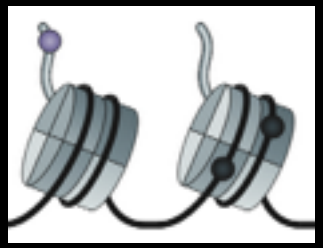

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DNase Hypersensitivity
ENCODE
Epigenomics Roadmap
Bench-to-Bassinet

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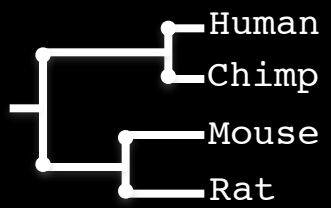

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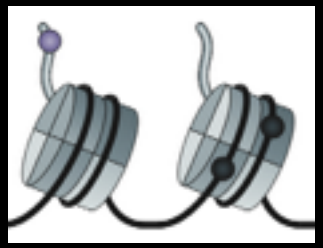

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short k-mers
known TF motifs

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Computational Algorithm

Support vector machine: separates 2 groups

Multi-kernel: good for combining heterogeneous data types with different weights

$$f(x) = \sum_{i=1}^{N} \alpha_i \sum_{j=1}^{M} \beta_j k_j(x, x_i) + b$$

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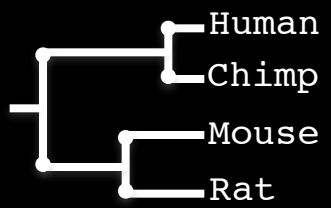

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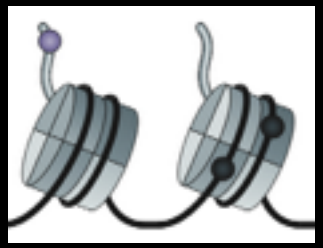

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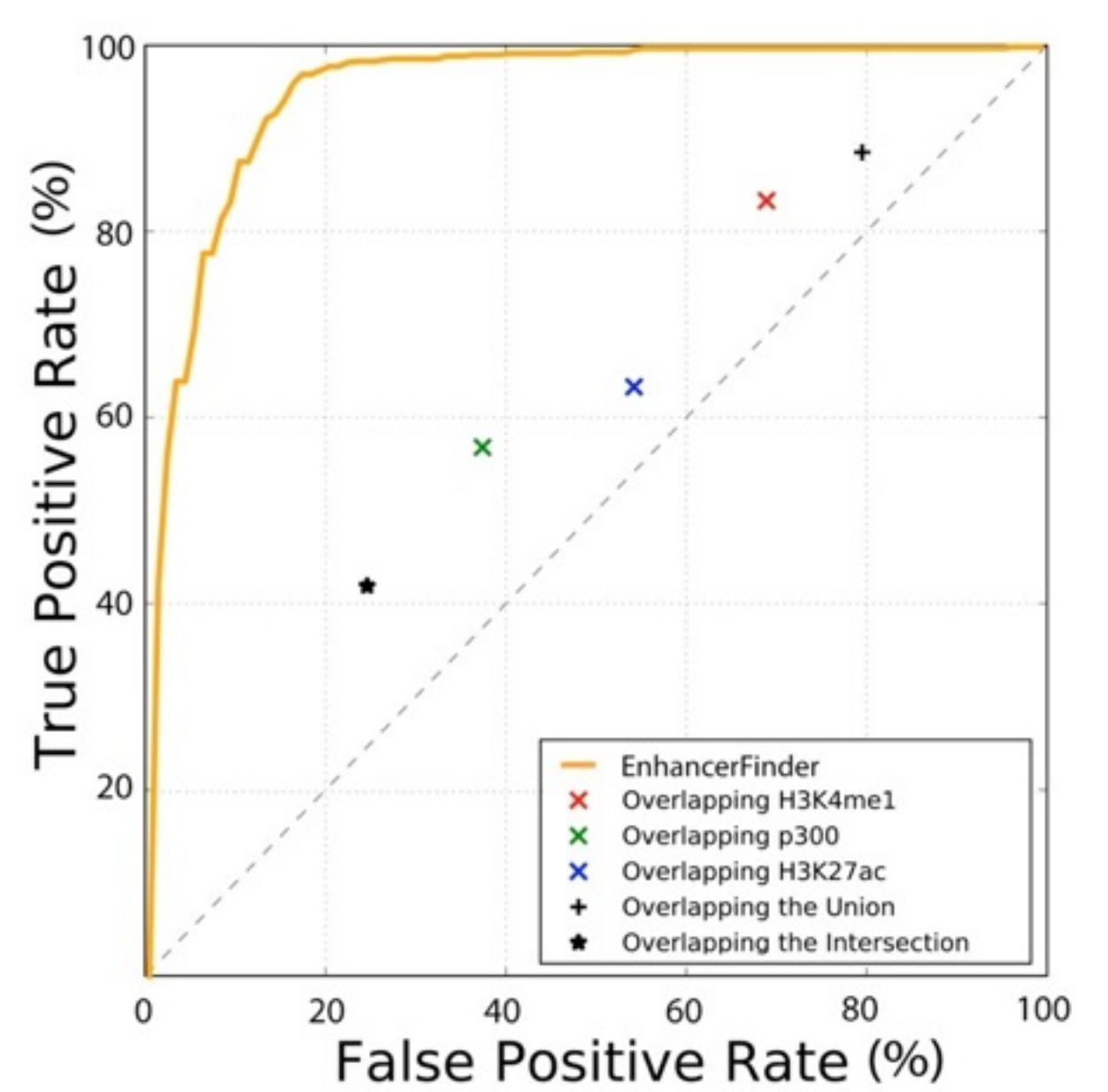

AUC=0.96
Power=85%
at 10% FPR,
Recall=85% at
93% Precision

FDR ~10-50%
Significantly
better than
other methods

>80% in vivo
validation rate

Erwin et al. (2014)
PLoS Comp Bio

EnhancerFinder: Predictions

Erwin et al. (2014) PLoS Comp Bio, Capra et al. (2014) PTRSB

EnhancerFinder: Predictions

- 84,301 developmental enhancer predictions
  - Cover 2% of the human genome
  - Nearby genes have high expression and annotated functions in the relevant fetal tissue
  - Significant overlap with disease mutations
  - Cluster around developmental transcription factors and signaling genes

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- 239 predictions overlap a Human Accelerated Region (33% of HARs), 25/30 validated *in vivo*

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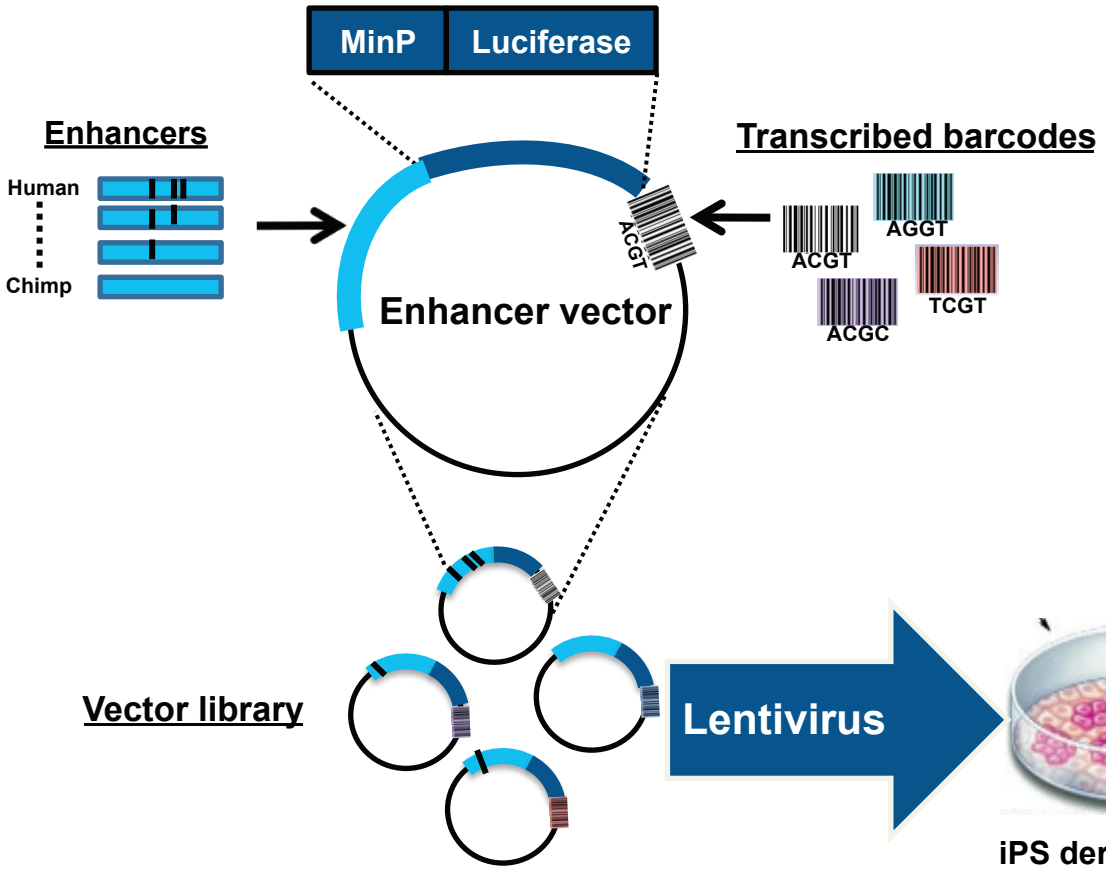
- 84,301 developmental enhancer predictions
  - Cover 2% of the human genome
  - Nearby genes have high expression and annotated functions in the relevant fetal tissue
  - Significant overlap with disease mutations
  - Cluster around developmental transcription factors and signaling genes

- 239 predictions overlap a Human Accelerated Region (33% of HARs), 25/30 validated *in vivo*

- Identify sites with fitness effects (Gulko et al 2015)

Erwin et al. (2014) PLoS Comp Bio, Capra et al. (2014) PTRSB

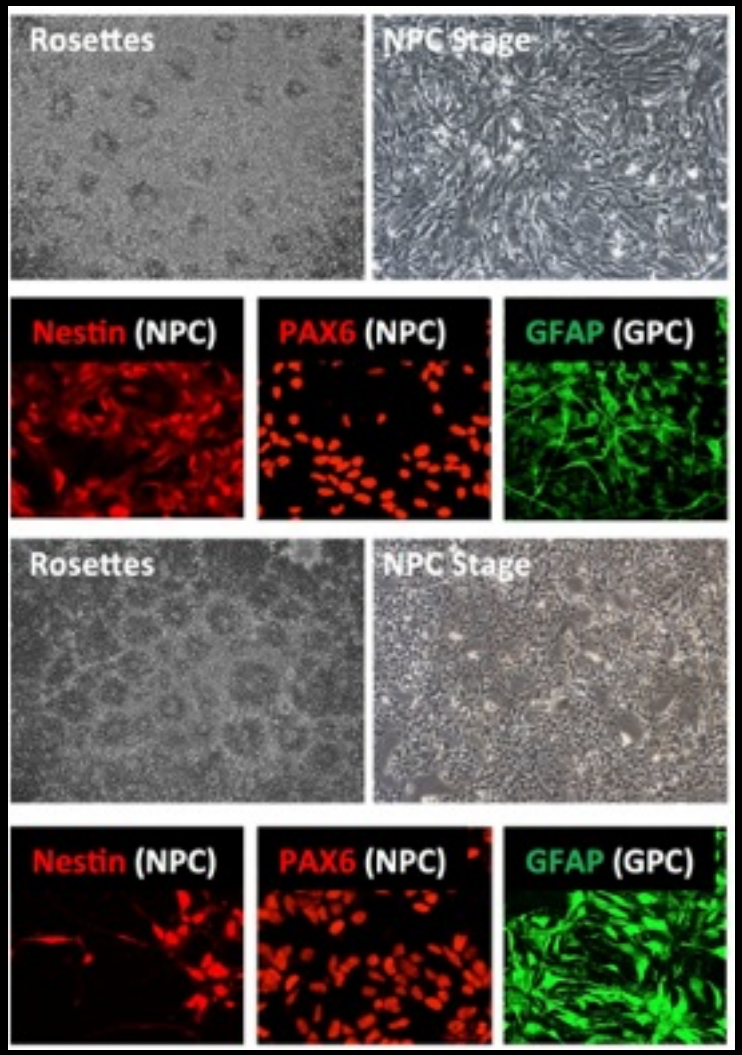
Massively Parallel Reporter Assays and capture Hi-C for validation

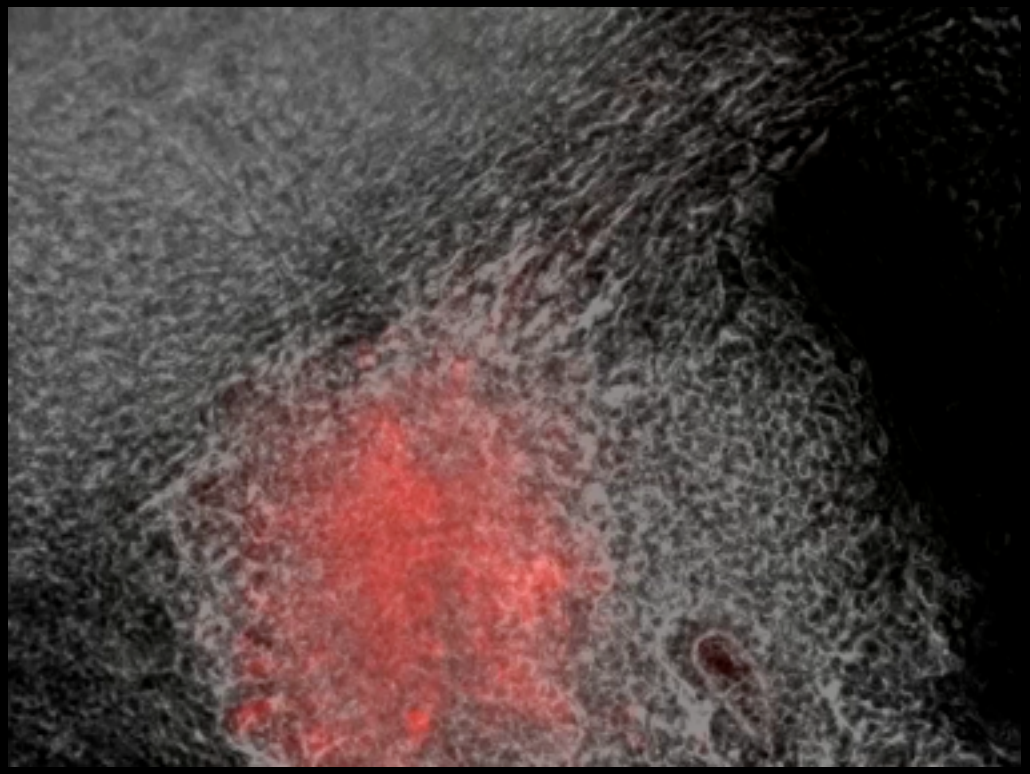


- Test >12,000 170bp enhancers in parallel
- Quantitative activity assayed via RNA-seq
- Compare genotypes
- Human vs. chimp
- Disease SNPs

Hane Ryu, Nadav Ahituv, Jay Shendure, Yin Shen

Induced pluripotent stem cell derived neuronal and cardiac lines



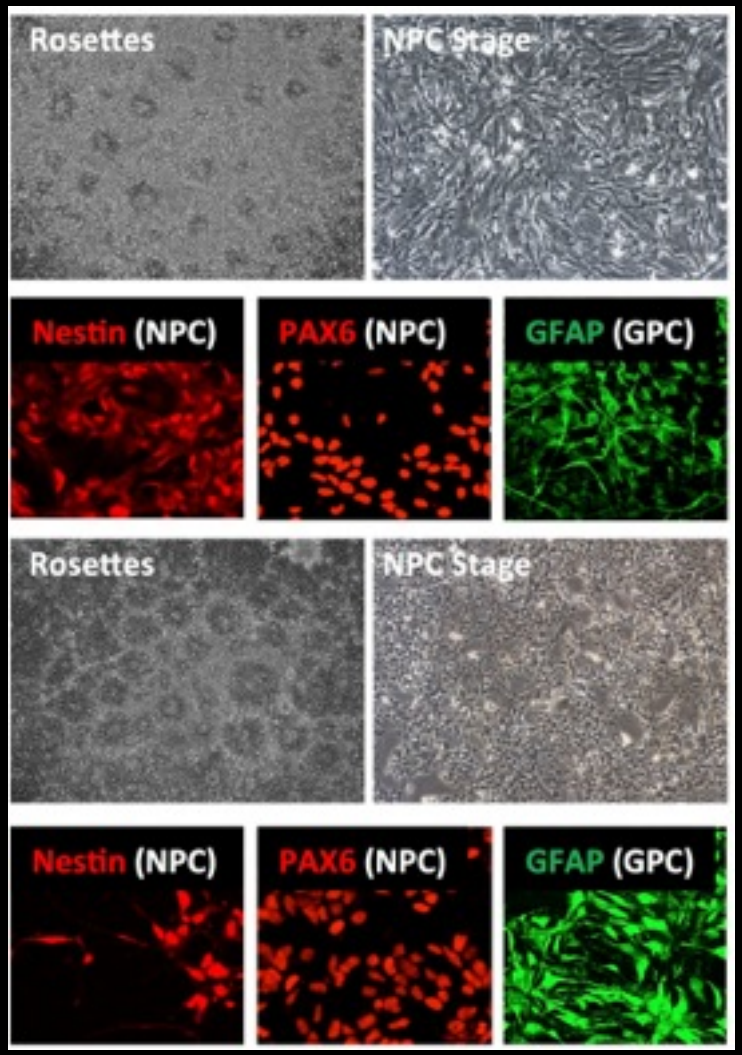


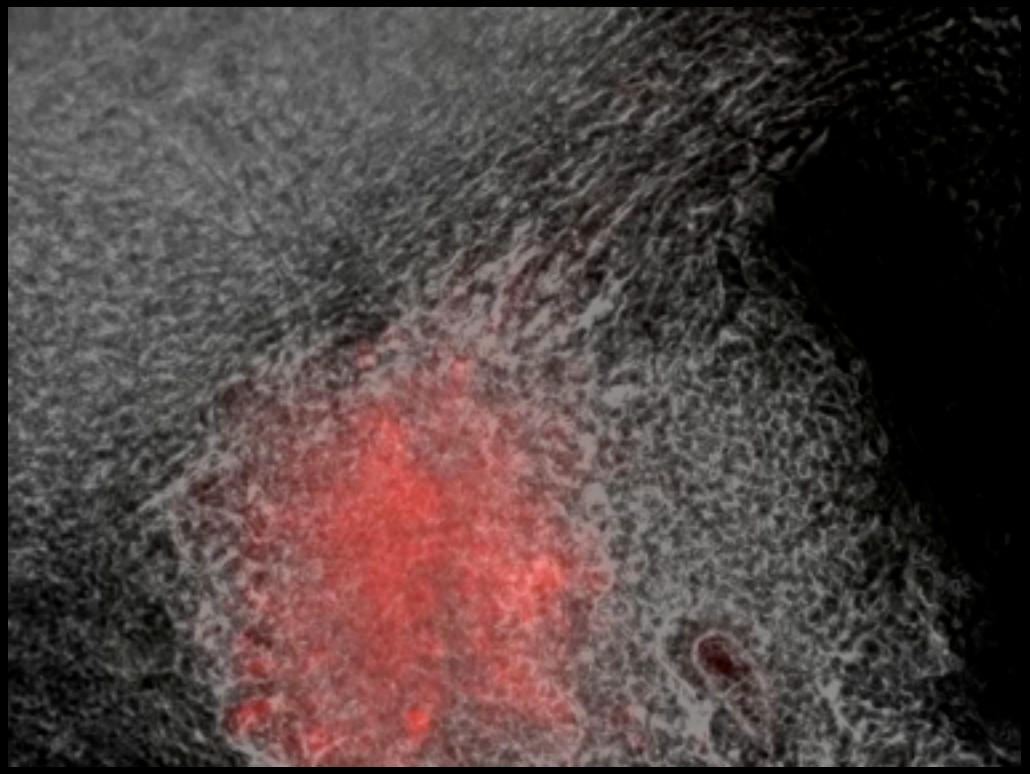
Human iPSC derived cardiomyocytes

Hane Ryu, Alex Pollen, Nadav Ahituv, Arnold Kriegstein

Bruce Conklin

Induced pluripotent stem cell derived neuronal and cardiac lines





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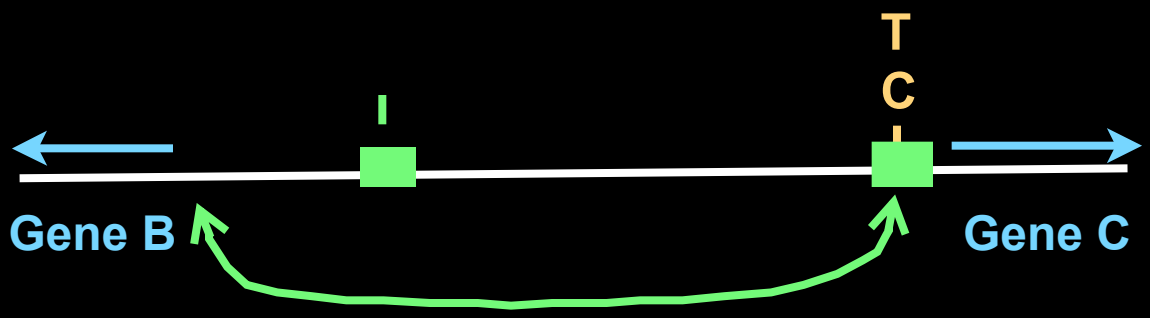
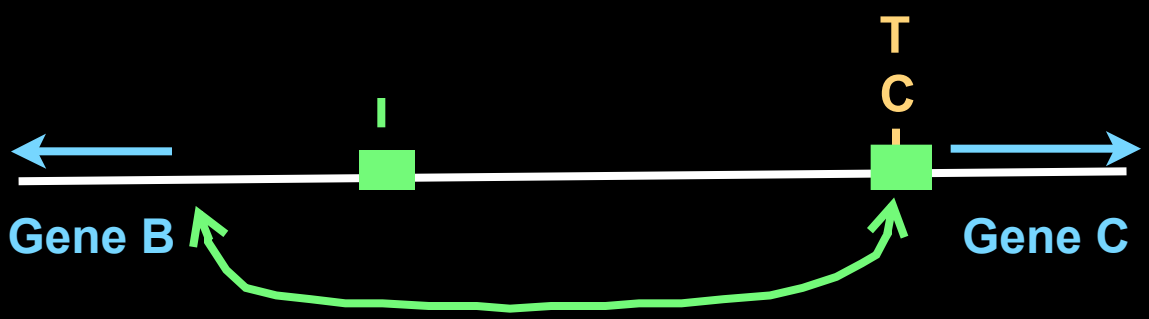
Discovering the role of enhancers in human biology and evolution

Discovering the role of enhancers in human biology and evolution

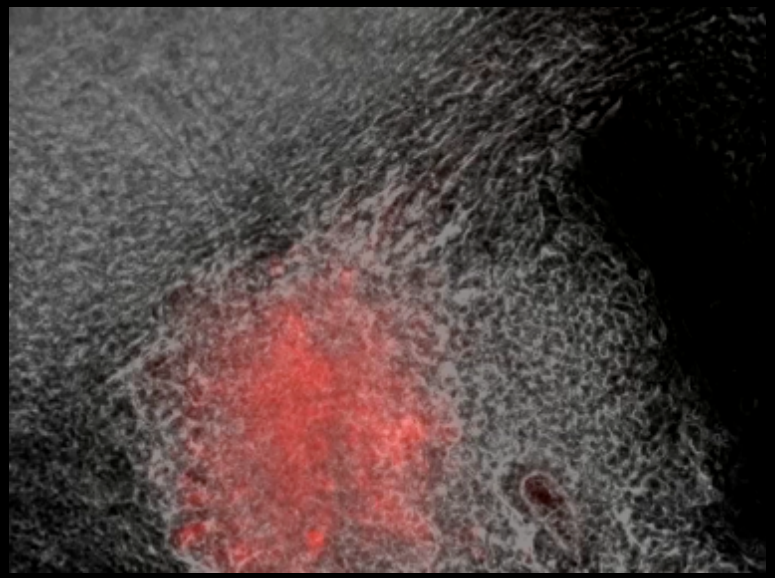


Computational Characterization

Discovering the role of enhancers in human biology and evolution

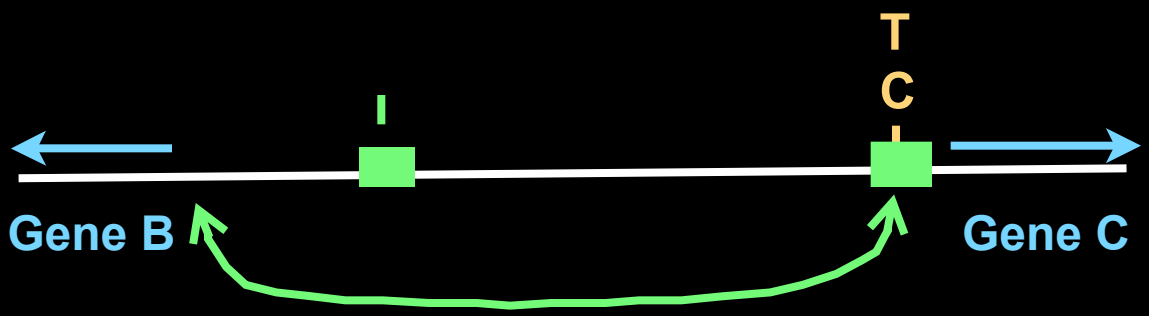


Computational Characterization

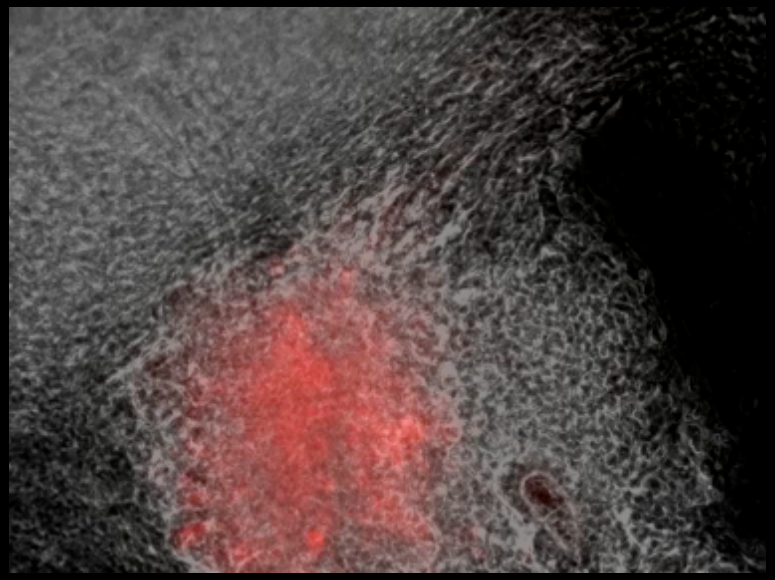


iPSC based screening

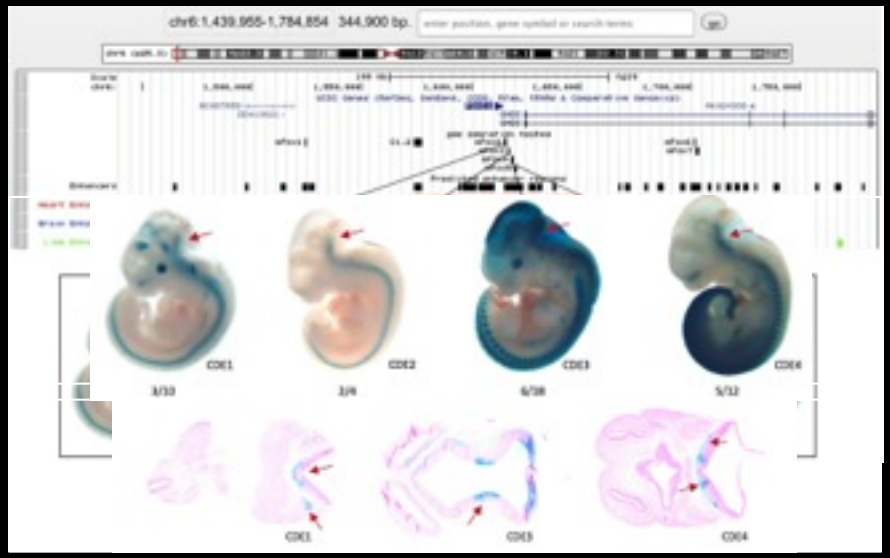
Discovering the role of enhancers in human biology and evolution



Computational Characterization



iPSC based screening



In vivo molecular studies

Collaborators

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John Rubenstein

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