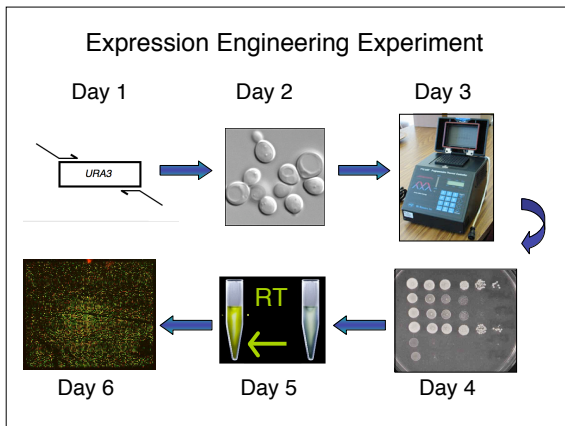


20.109: Expression Engineering

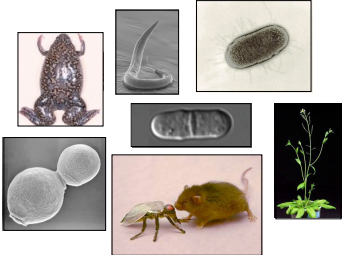
Spring 2007
Lecture 2
April 5th, 2007



Expression Engineering Experiment	
<p style="text-align: center;">Lecture 1</p> <ul style="list-style-type: none"> eukaryotic gene expression <ul style="list-style-type: none"> ✓chromatin parts ✓euk exp'n + obstacles 	<p style="text-align: center;">Lecture 2</p> <ul style="list-style-type: none"> intro to yeast genetics yeast SAGA complex
<p style="text-align: center;">Lecture 3</p> <ul style="list-style-type: none"> IMRD (Susan Ruff) 	<p style="text-align: center;">Lecture 4</p> <ul style="list-style-type: none"> yeast genetic analysis
<p style="text-align: center;">Lecture 5</p> <ul style="list-style-type: none"> measuring gene expression 	<p style="text-align: center;">Lecture 6</p> <ul style="list-style-type: none"> microarray analysis (Rebecca Fry)

Model genetic organisms: modern

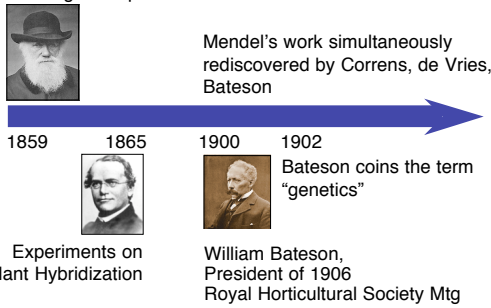
- S. cerevisiae
- S. pombe
- E. coli
- C. elegans
- D. melanogaster
- A. thaliana
- M. musculus
- X. laevis



For how many of these organisms do we have full sequence information?

Model genetic organisms: historical

On Origin of Species



Mendel's work simultaneously rediscovered by Correns, de Vries, Bateson

1859 1865 1900 1902

Experiments on Plant Hybridization

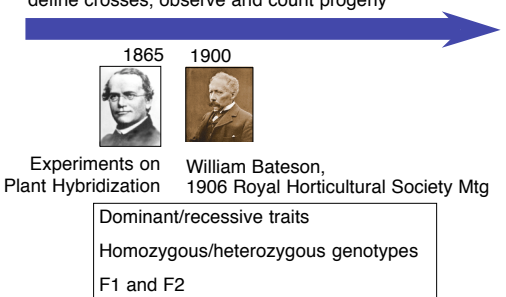
William Bateson, President of 1906 Royal Horticultural Society Mtg

Bateson coins the term "genetics"

Nature Reviews Genetics (2007) 8: 231

Genetics retrospective

Genetics = study of inheritance
define crosses, observe and count progeny



1865 1900


Experiments on Plant Hybridization


William Bateson, 1906 Royal Horticultural Society Mtg

Dominant/recessive traits
Homozygous/heterozygous genotypes
F1 and F2


Genetics retrospective

Genetics = study of DNA mutations





1953



1956


Identify mutants, map gene
examine linkage
epistasis
genetic interactions

“epigenetic landscape”
coined by Conrad Waddington

Genetics retrospective


Genetics = study of DNA sequences

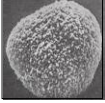
read/write DNA



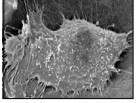
Genetics = gene use

differential expression of DNA






liver cell

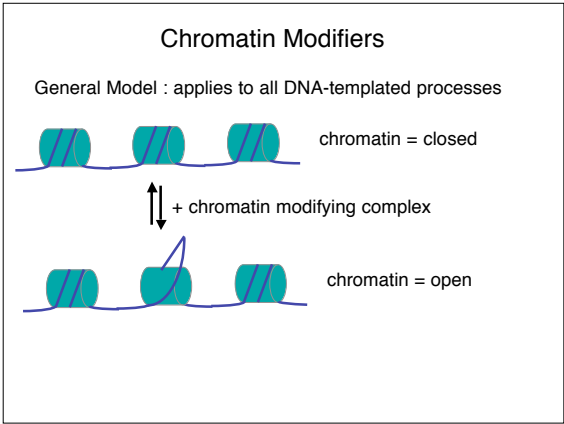


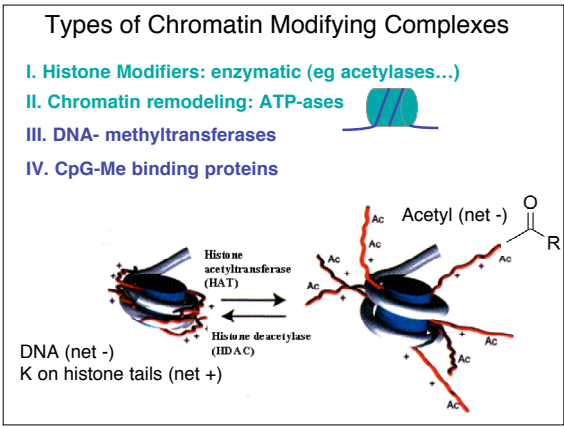
fibroblast cell

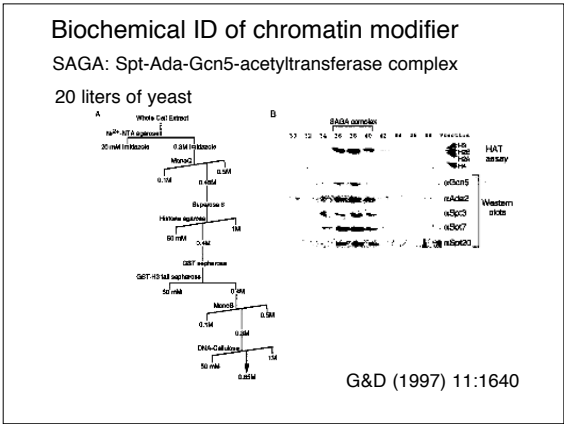
S. cerevisiae features



- Genomic stability/recombination balance
-




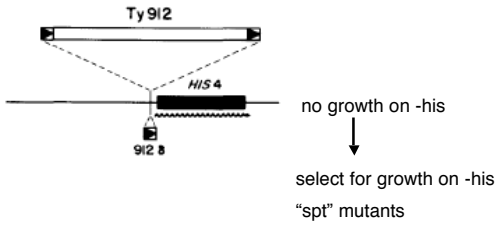




SPT: suppressor of Ty element insertions

Transposable elements = Ty

~6kb long, with 300 bp repeats called delta elements 

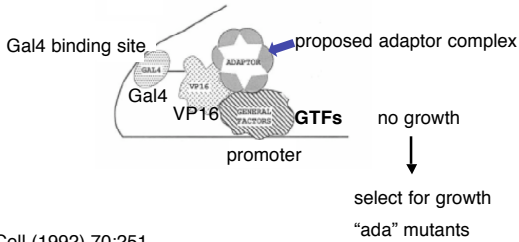


Genetics (1984) 107:179

ADA: transcriptional adaptor

GAL4-VP16 = toxic chimera

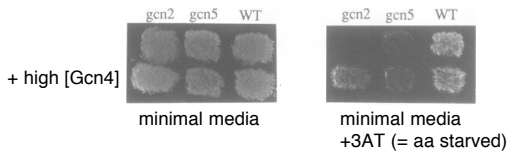
Gal4 (DNA binding domain) + VP16 (HSV activation patch)



Cell (1992) 70:251

GCN5: general control nonderepressible

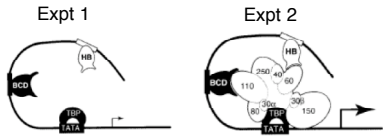
"general control" = amino acid starvation--> derepression of many biosynthetic genes by Gcn4



"Thus, GCN5 is a new member of the recently revealed general class of transcriptional regulators that collaborate with certain specific DNA binding activators to promote high levels of transcription."

EMBO J. (1992) 11: 4145

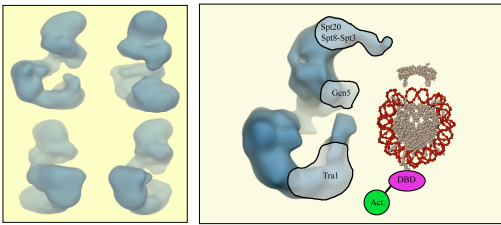
TAF: TBP associated factors



Component	Expt 1	Expt 2
TF (bicoid)	+	+
TF (hunchback)	+	+
TBP	+	+
TAFs	-----	+
<i>In vitro</i> transcript?		

TIBS (1996) 21:338

SAGA structure by cryo-EM



images from P. Schultz

Summary

