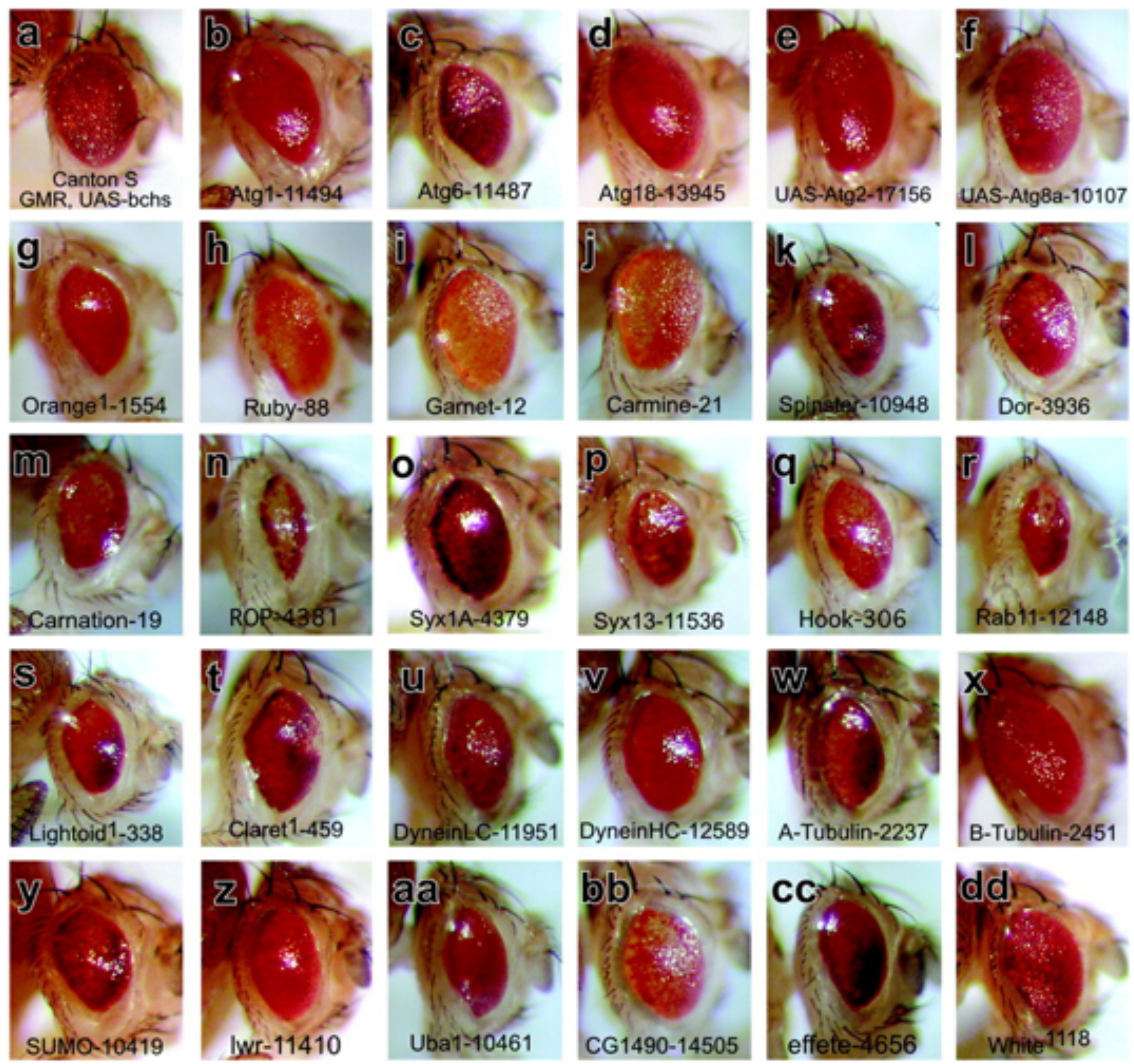
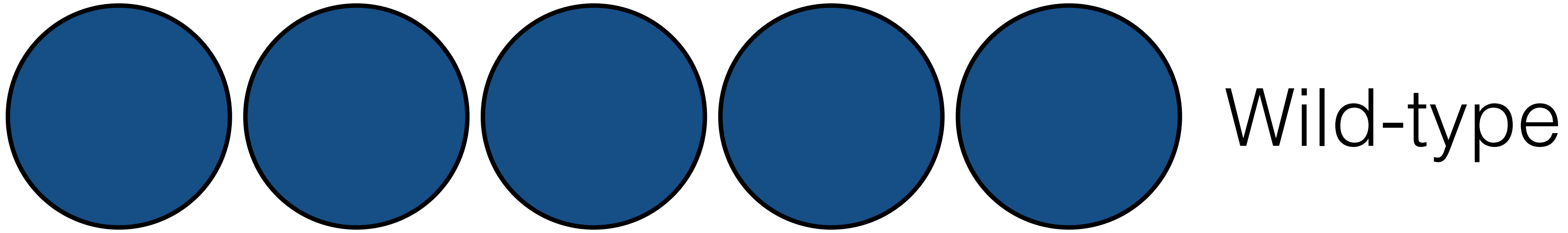
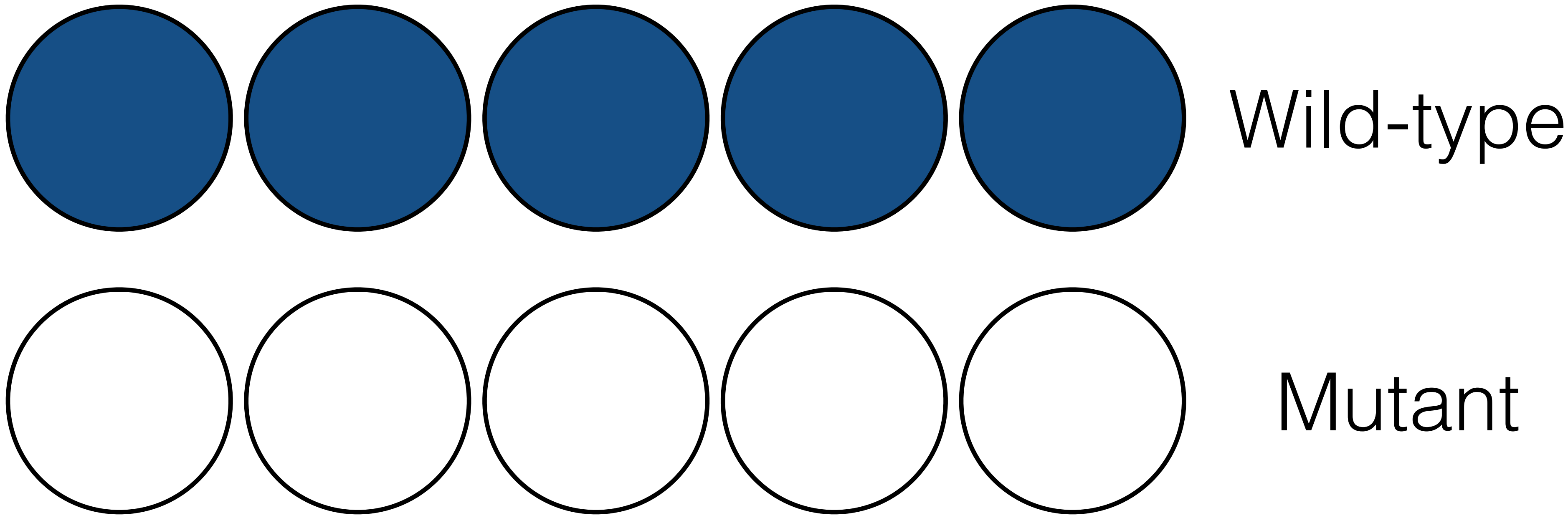
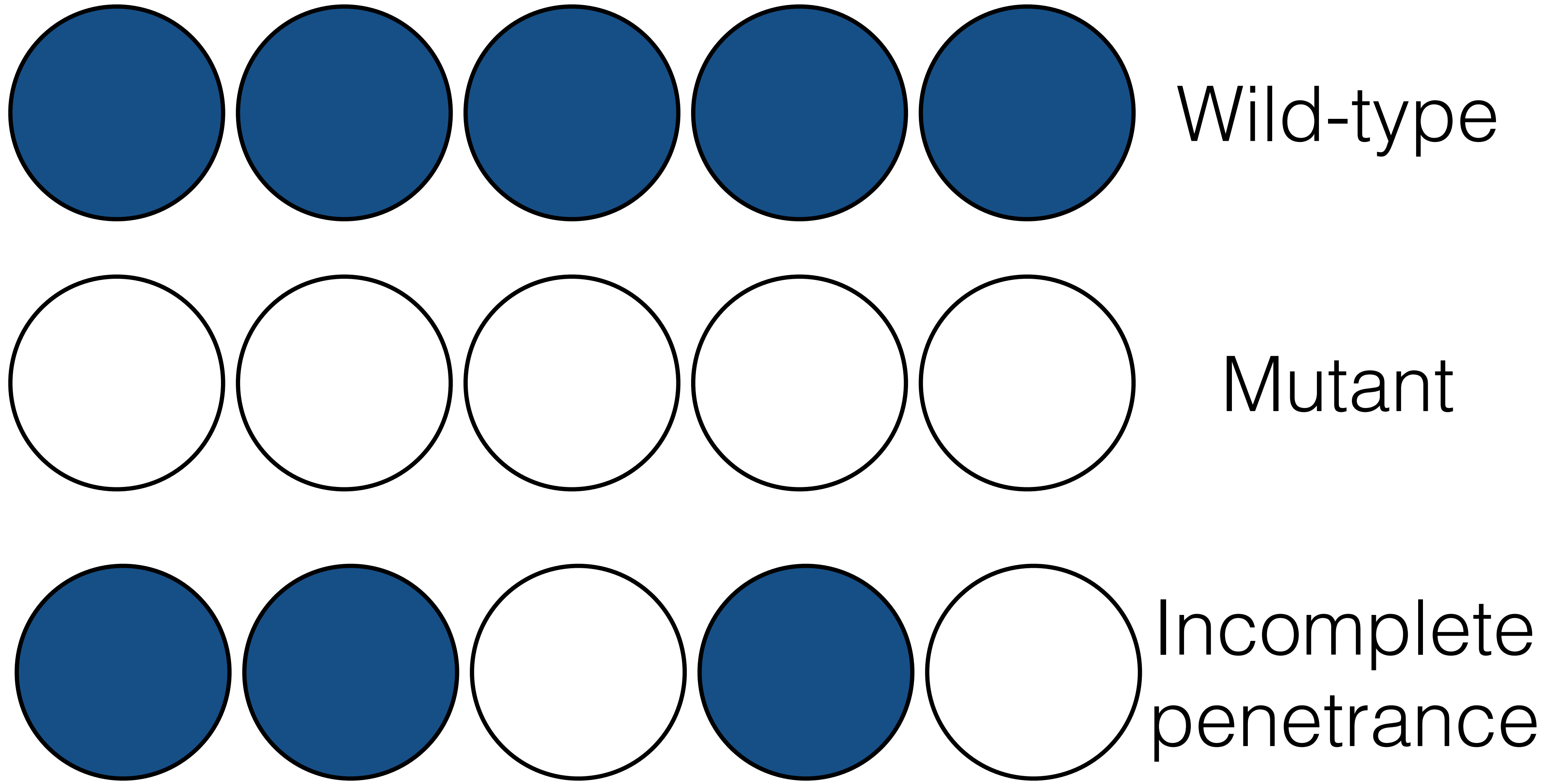
Bio393: Genetic Analysis

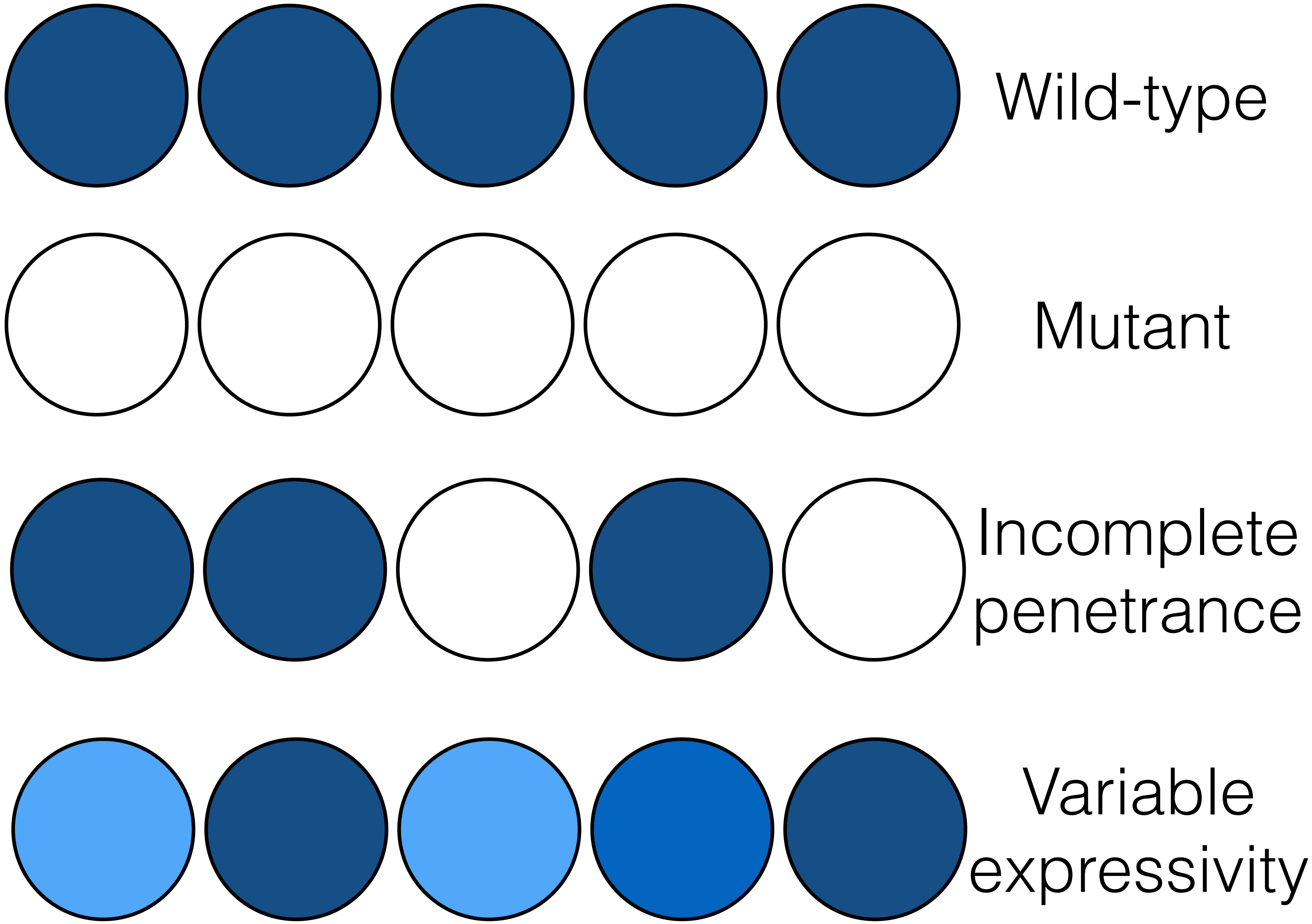
Genetic interactions: suppression and enhancement




Wild-type


Wild-type
Mutant


Wild-type
Mutant
Incomplete penetrance


Wild-type
Mutant
Incomplete penetrance
Variable expressivity

Wild-type worms have one vulva

Multivulva mutant worms have multiple vulvae

Incomplete penetrance is when not every mutant animal has the mutant phenotype

Wild-type worms have one vulva

Multivulva mutant worms have multiple vulvae

Incomplete penetrance is when not every mutant animal has the mutant phenotype

117/129 animals are multivulva
91% penetrant

Wild-type worms have one vulva

Multivulva mutant worms have multiple vulvae

Variable expressivity is when each mutant animal is not completely mutant

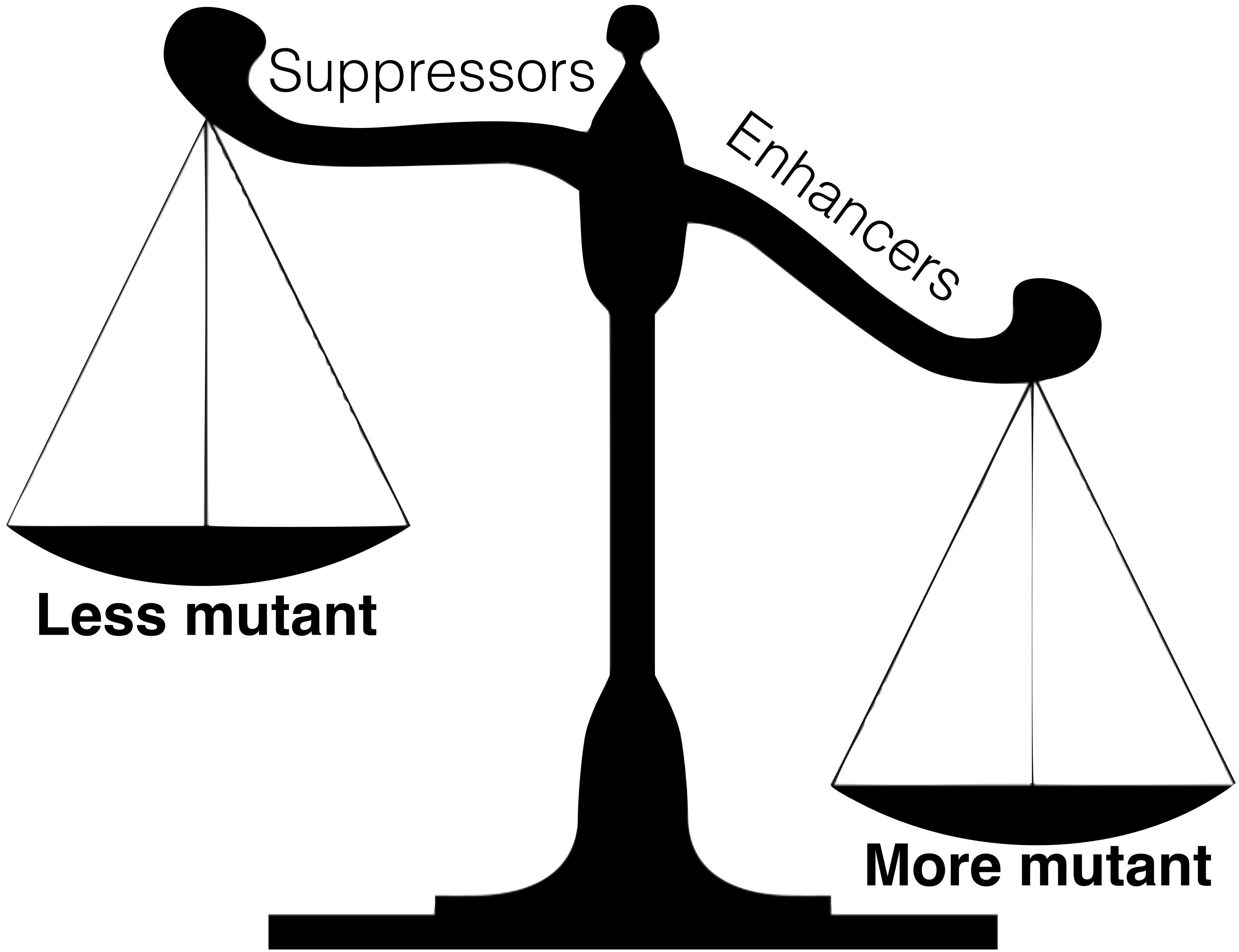
Wild-type worms have one vulva

Multivulva mutant worms have multiple vulvae

Variable expressivity is when each mutant animal is not completely mutant

An animal only has two extra vulvae instead of three.

Identification of mutants that modify an existing mutant phenotype



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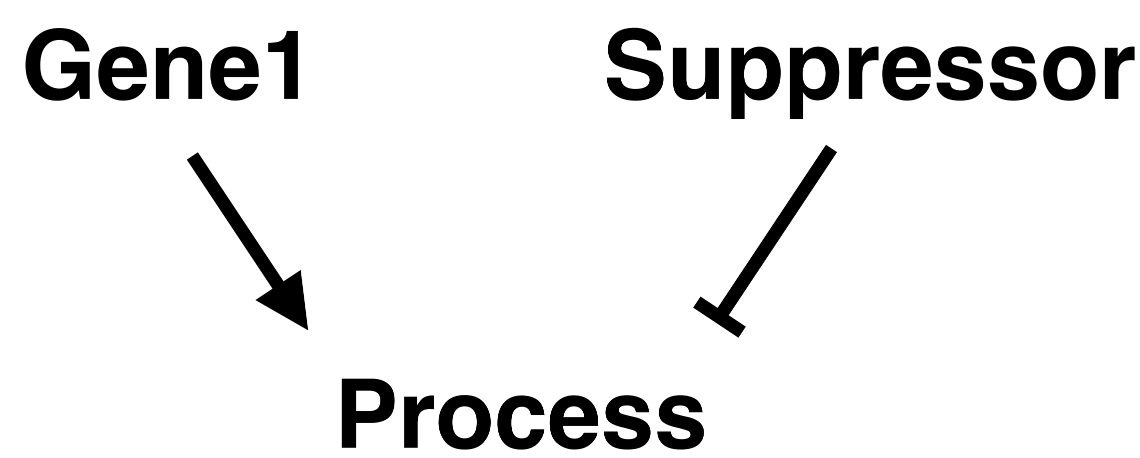

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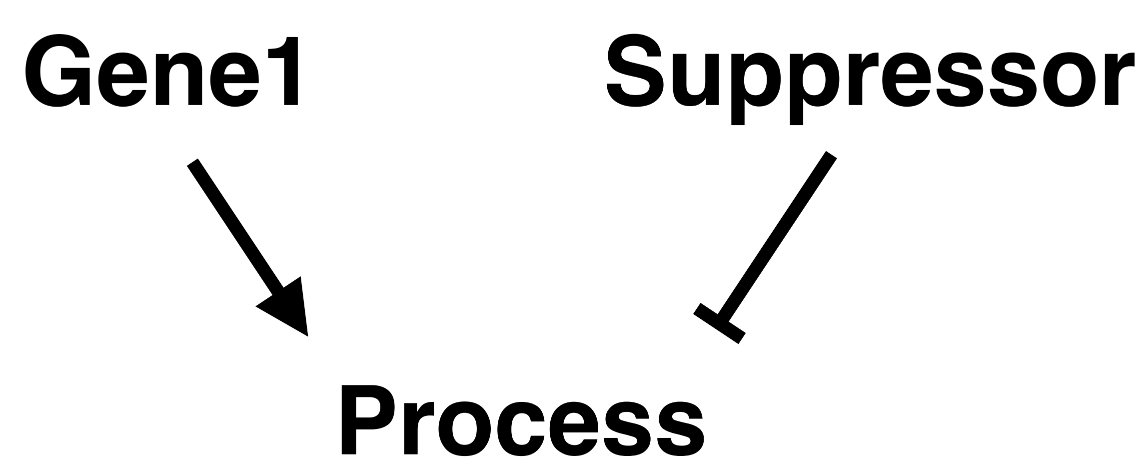
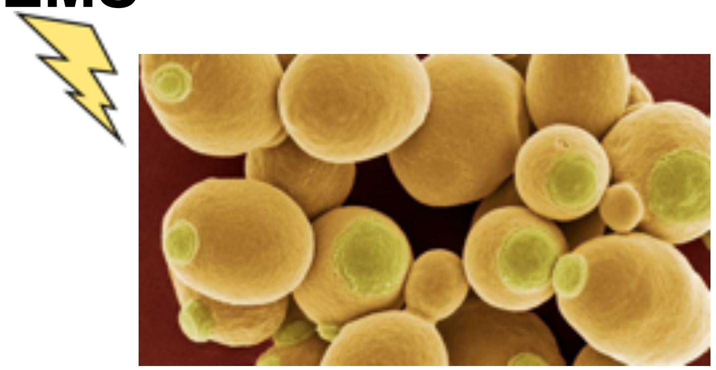


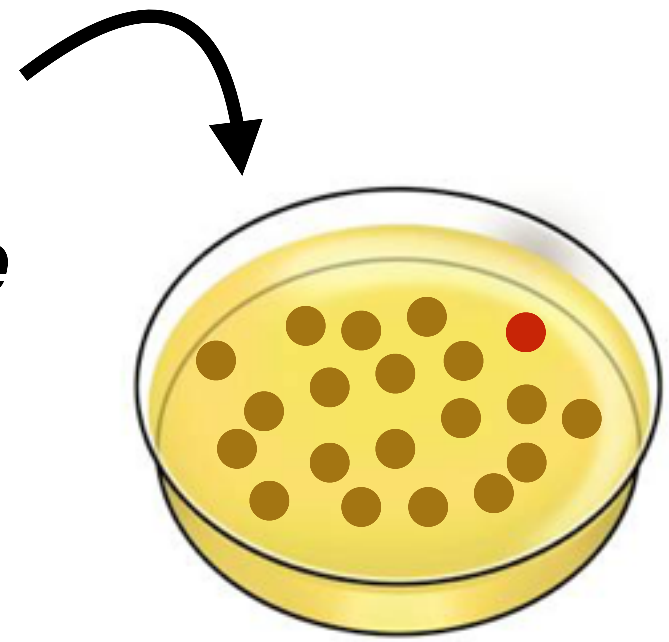
Why do we care?

We want to identify suppressors of the *ade2* mutant phenotype

EMS



S. cerevisiae



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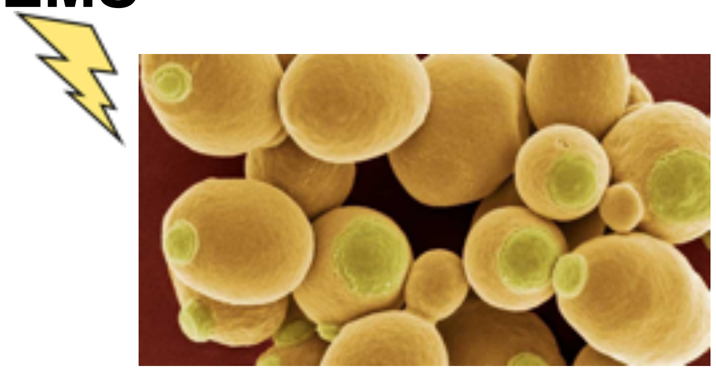

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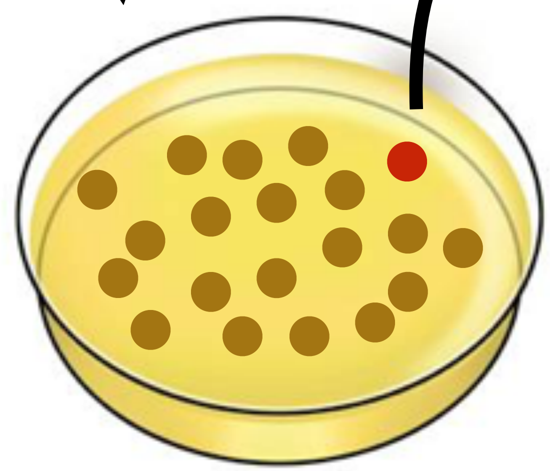

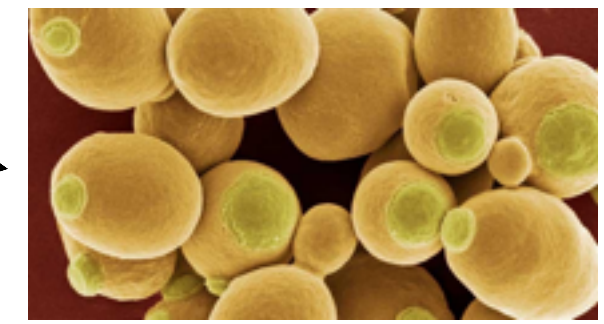

***ade2* mutants**

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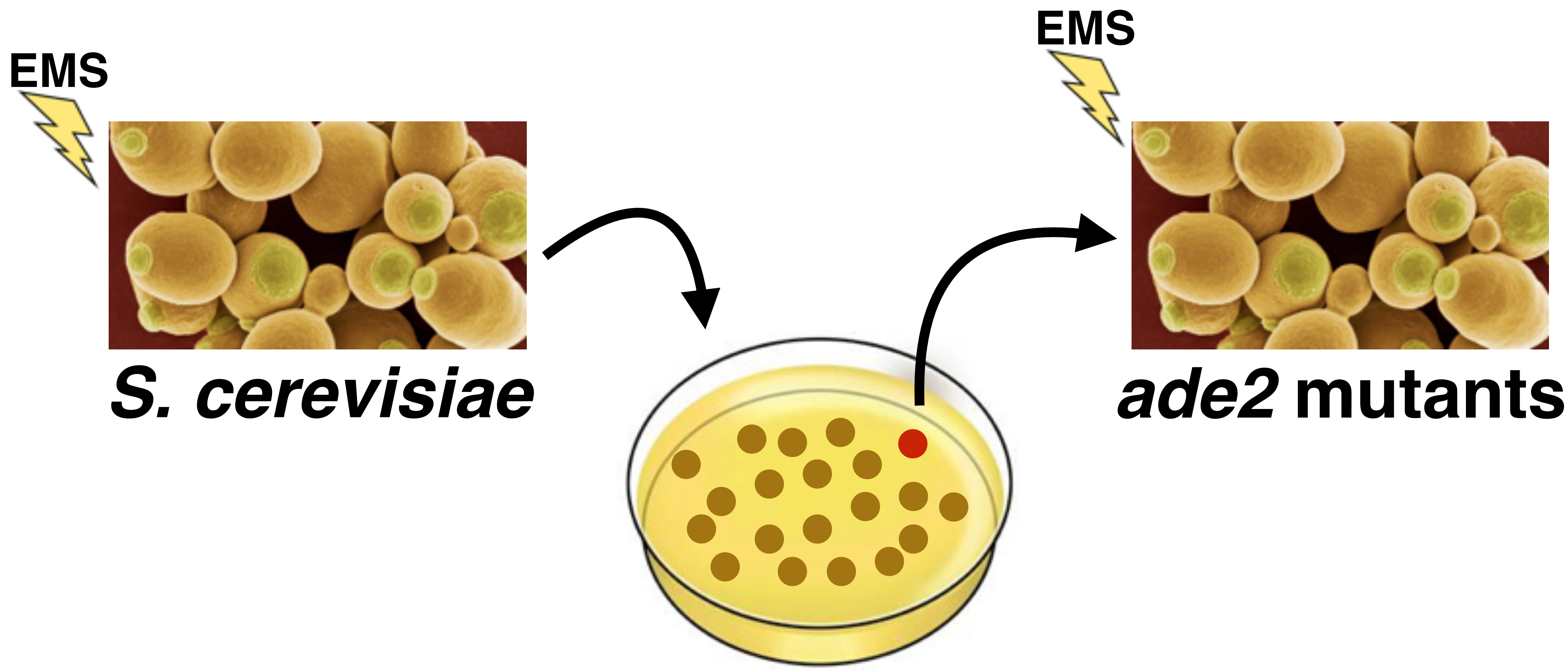

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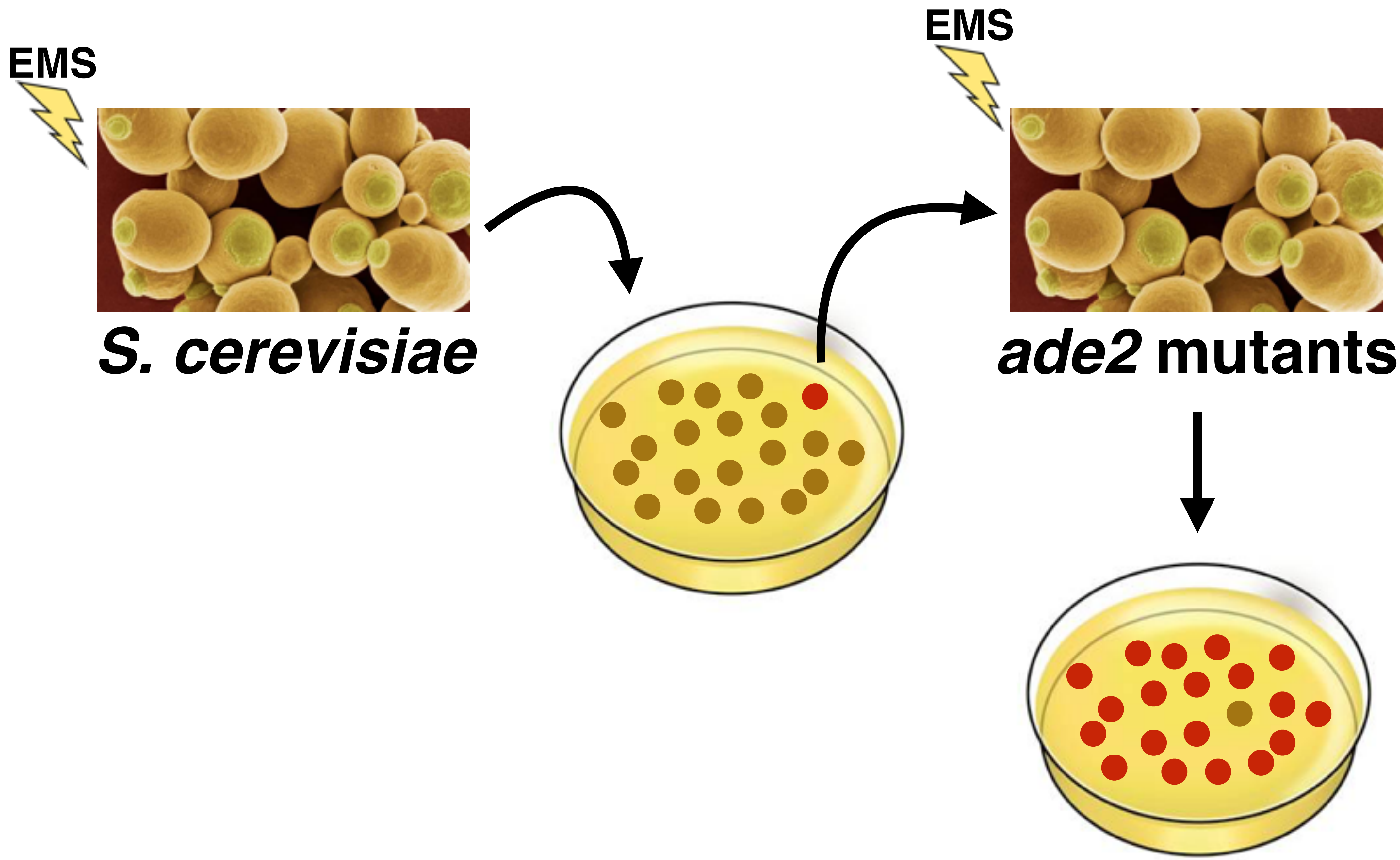

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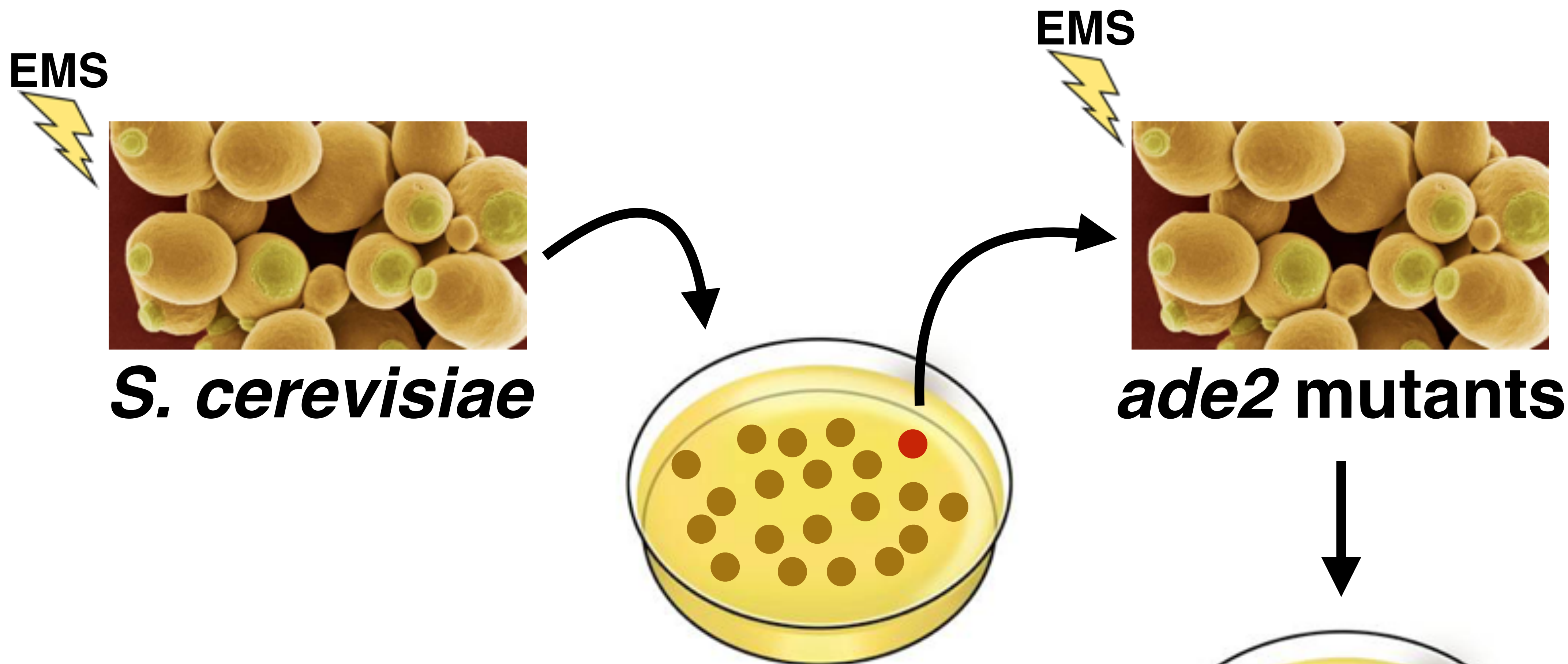

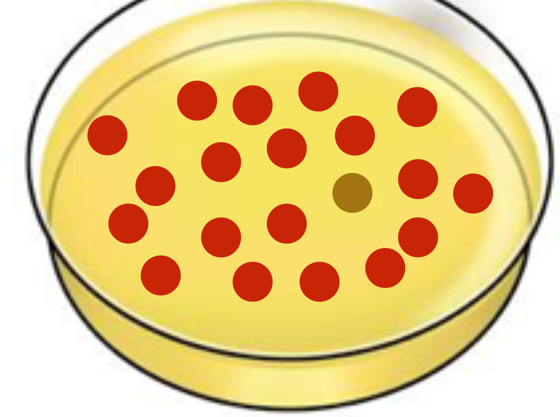

How could the mutant phenotype be suppressed?

Three general types of suppressor mutants

1. Revertant (back mutation)

Three general types of suppressor mutants

1. Revertant (back mutation)

```
>ADE2 YOR128C SGDID:S000005654
ATGGATTCTAGAACAGTTGGTATATTAGGAGGGGACAATTGGGACGTATGATTGTTGAG
GCAGCAAACAGGCTCAACATTAAGACGGTAATACTAGATGCTGAAAATTCTCCTGCCAAA
CAAATAAGCAACTCCAATGACCACGTTAATGGCTCCTTTTCCAATCCTCTTGATATCGAA
AAACTAGCTGAAAAATGTGATGTGCTAACGATTGAGATTGAGCATGTTGATGTTCCTACA
CTAAAGAATCTTCAAGTAAAACATCCCAAATTAAAAATTTACCCTTCTCCAGAAACAATC
AGATTGATACAAGACAAATATATTCAAAAAGAGCATTTAATCAAAAATGGTATAGCAGTT
ACCCAAAGTGTTCCTGTGGAACAAGCCAGTGAGACGTCCCTATTGAATGTTGGAAGAGAT
TTGGGTTTTCCATTCGTCTTGAAGTCGAGGACTTTGGCATACGATGGAAGAGGTAACTTC
GTTGTAAAGAATAAGGAAATGATTCCGGAAGCTTTGGAAGTACTGAAGGATCGTCCTTTG
TACGCCGAAAAATGGGCACCATTTACTAAAGAATTAGCAGTCATGATTGTGAGATCTGTT
AACGGTTTAGTGTTTTCTTACCCAATTGTAGAGACTATCCACAAGGACAATATTTGTGAC
TTATGTTATGCGCCTGCTAGAGTTCCGGACTCCGTTCAACTTAAGGCGAAGTTGTTGGCA
GAAAATGCAATCAAATCTTTTCCCGGTTGTGGTATATTTGGTGTGGAAATGTTCTATTTA
GAAACAGGGGAATTGCTTATTAACGAAATTGCCCCAAGGCCTCACAACTCTGGACATTAT
ACCATTGATGCTTGCGTCACTTCTCAATTTGAAGCTCATTTGAGATCAATATTGGATTTG
CCAATGCCAAAGAATTTCACATCTTTCTCCACCATTACAACGAACGCCATTATGCTAAAT
GTTCTTGGAGACAAACATACAAAAGATAAAGAGCTAGAAACTTGCGAAAGAGCATTGGCG
ACTCCAGGTTCCTCAGTGTACTTATATGGAAAAGACTCTAGACCTAACAGAAAAGTAGGT
CACATAAATATTATTGCCTCCAGTATGGCGGAATCAAACAAAGGCTGAACTACATTACA
GGTAGAACTGATATTCCAATCAAAATCTCTGTCGCTCAAAAGTTGGACTTGGAAGCAATG
GTCAAACCATTGGTTGGAATCATCATGGGATCAGACTCTGACTTGCCGGTAATGTCTGCC
GCATGTGCGGTTTTAAAAGATTTTGGCGTTCCATTTGAAGTGACAATAGTCTCTGCTCAT
AGAACTCCACATAGGATGTCAGCATATGCTATTTCCGCAAGCAAGCGTGGAATTAAAACA
ATTATCGCTGGAGCTGGTGGGGCTGCTCACTTGCCAGGTATGGTGGCTGCAATGACACCA
CTTCCTGTCATCGGTGTGCCCGTAAAAGGTTCTTGTCTAGATGGAGTAGATTCTTTACAT
TCAATTGTGCAAATGCCTAGAGGTGTTCCAGTAGCTACCGTCGCTATTAATAATAGTACG
AACGCTGCGCTGTTGGCTGTCAGACTGCTTGGCGCTTATGATTCAAGTTATACAACGAAA
ATGGAACAGTTTTTATTAAAGCAAGAAGAAGTTCTTGTCAAAGCACAAAAGTTAGAA
ACTGTCGGTTACGAAGCTTATCTAGAAAACAAGTAA
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AAACTAGCTGAAAAATGTGATGTGCTAACGATTGAGATTGAGCATGTTGATGTTCCTACA
CTAAAGAATCTTCAAGTAAAACATCCCAAATTAAAAATTTACCCTTCTCCAGAAACAATC
AGATTGATACAAGACAAATATATTCAAAAAGAGCATTTAATCAAAAATGGTATAGCAGTT
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TTGGGTTTTCCATTCGTCTTGAAGTCGAGGACTTTGGCATACGATGGAAGAGGTAACTTC
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AACGGTTTAGTGTTTTCTTACCCAATTGTAGAGACTATCCACAAGGACAATATTTGTGAC
TTATGTTATGCGCCTGCTAGAGTTCCGGACTCCGTTCAACTTAAGGCGAAGTTGTTGGCA
GAAAATGCAATCAAATCTTTTCCCGGTTGTGGTATATTTGGTGTGGAAATGTTCTATTTA
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ACCATTGATGCTTGCGTCACTTCTCAATTTGAAGCTCATTTGAGATCAATATTGGATTTG
CCAATGCCAAAGAATTTCACATCTTTCTCCACCATTACAACGAACGCCATTATGCTAAAT
GTTCTTGGAGACAAACATACAAAAGATAAAGAGCTAGAAACTTGCGAAAGAGCATTGGCG
ACTCCAGGTTCCTCAGTGTACTTATATGGAAAAGAGTCTAGACCTAACAGAAAAGTAGGT
CACATAAATATTATTGCCTCCAGTATGGCGGAATGTGAACAAAGGCTGAACTACATTACA
GGTAGAACTGATATTCCAATCAAAATCTCTGTCGCTCAAAAGTTGGACTTGGAAGCAATG
GTCAAACCATTGGTTGGAATCATCATGGGATCAGACTCTGACTTGCCGGTAATGTCTGCC
GCATGTGCGGTTTTAAAAGATTTTGGCGTTCCATTTGAAGTGACAATAGTCTCTGCTCAT
AGAACTCCACATAGGATGTCAGCATATGCTATTTCCGCAAGCAAGCGTGGAATTAAAACA
ATTATCGCTGGAGCTGGTGGGGCTGCTCACTTGCCAGGTATGGTGGCTGCAATGACACCA
CTTCCTGTCATCGGTGTGCCCGTAAAAGGTTCTTGTCTAGATGGAGTAGATTCTTTACAT
TCAATTGTGCAAATGCCTAGAGGTGTTCCAGTAGCTACCGTCGCTATTAATAATAGTACG
AACGCTGCGCTGTTGGCTGTCAGACTGCTTGGCGCTTATGATTCAAGTTATACAACGAAA
ATGGAACAGTTTTTATTAAAGCAAGAAGAAGTTCTTGTCAAAGCACAAAAGTTAGAA
ACTGTCGGTTACGAAGCTTATCTAGAAAACAAGTAA
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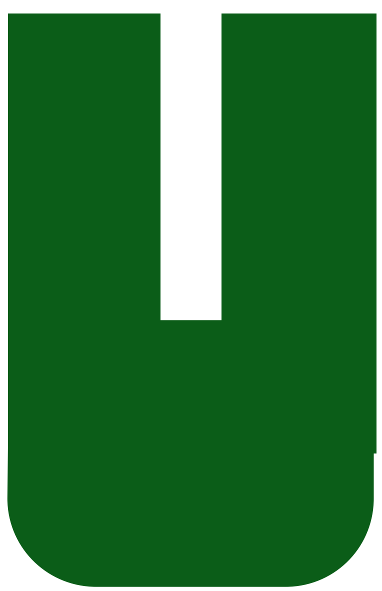

Wild-type

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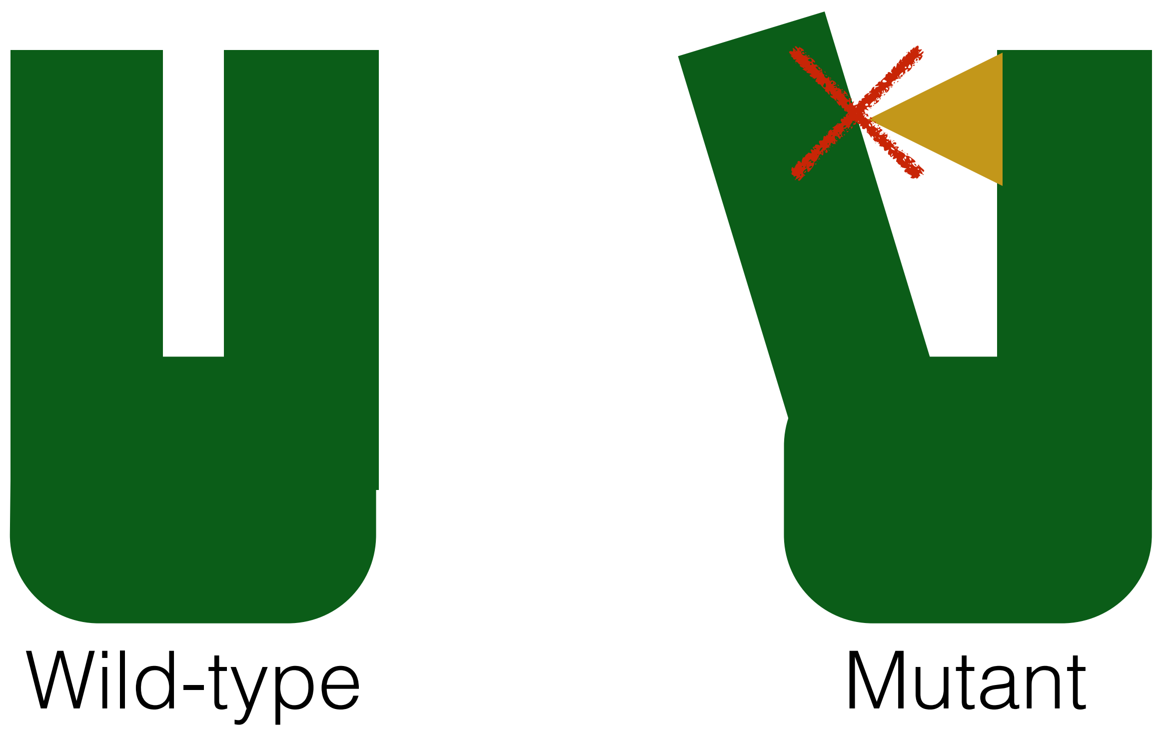

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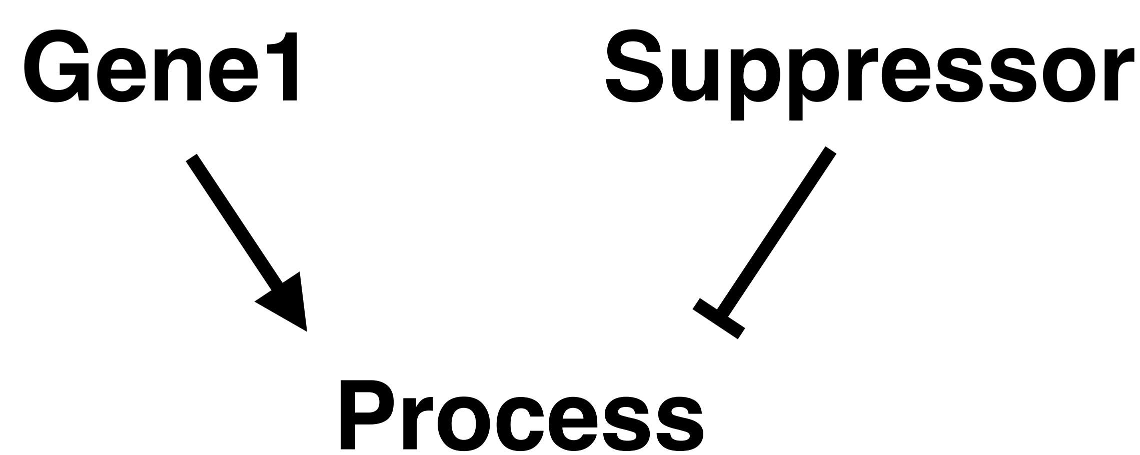

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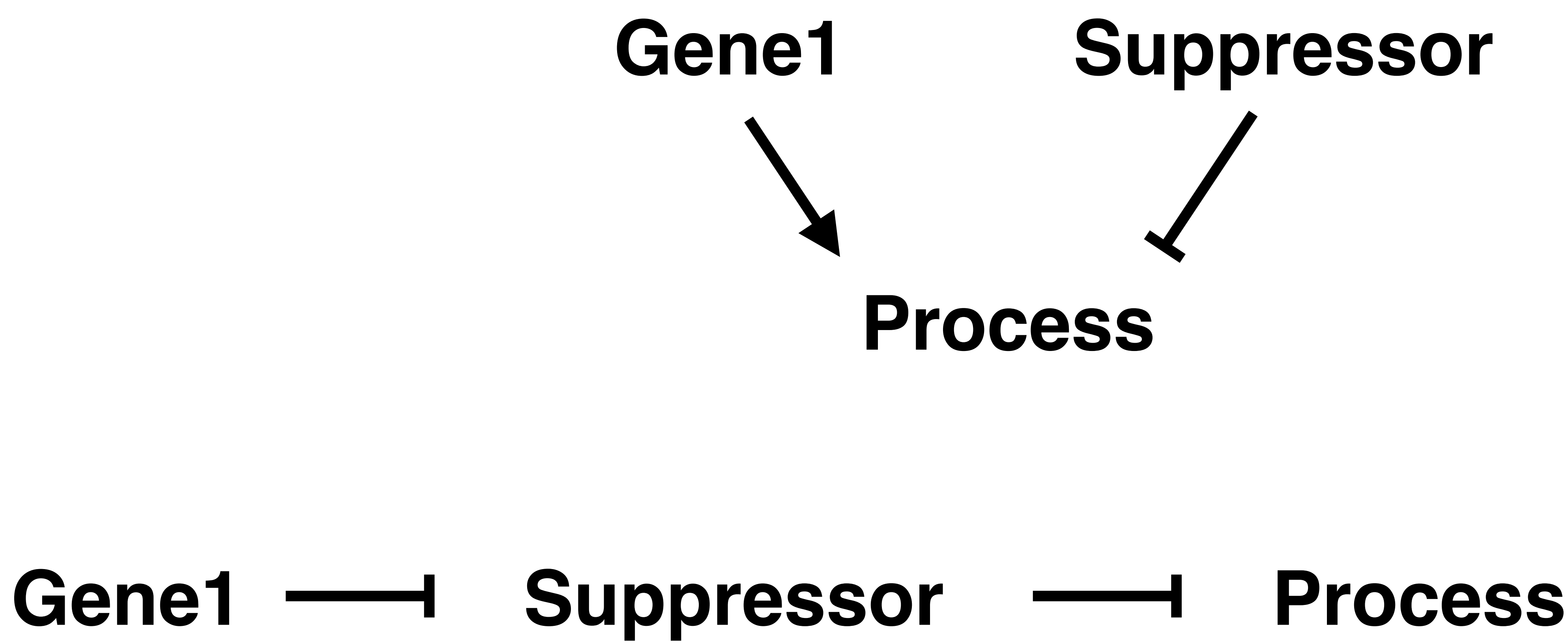

Three general types of suppressor mutants

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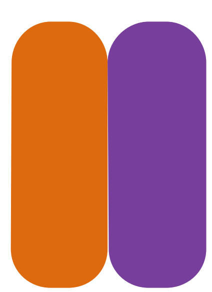

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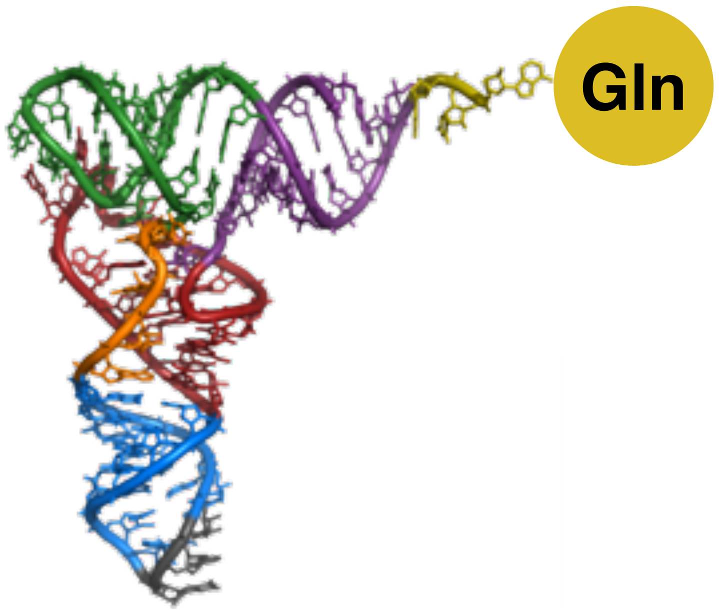
    4. Informational suppressor
       (gene-nonspecific, allele-specific)

Informational suppressors are allele-specific but gene-nonspecific



Anticodon	GUC
Codon	CAG
Encodes	Glutamine

Informational suppressors are allele-specific but gene-nonspecific

Mutate tRNA^{Gln} gene →

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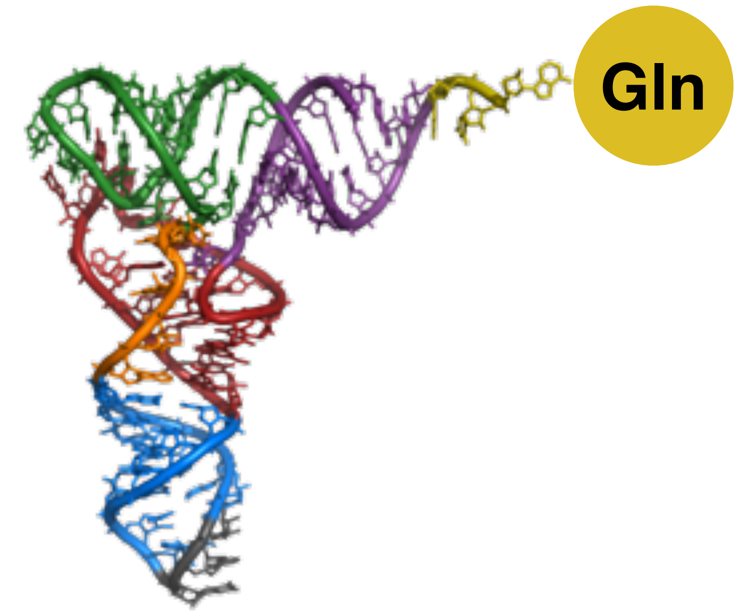


Anticodon	GUC	Anticodon	AUC
Codon	CAG	Codon	UAG
Encodes	Glutamine	Encodes	STOP

Informational suppressors are allele-specific but gene-nonspecific

Mutate tRNA^{Gln} gene →

Anticodon	GUC		Anticodon	AUC
Codon	CAG		Codon	UAG
Encodes	Glutamine		Encodes	STOP

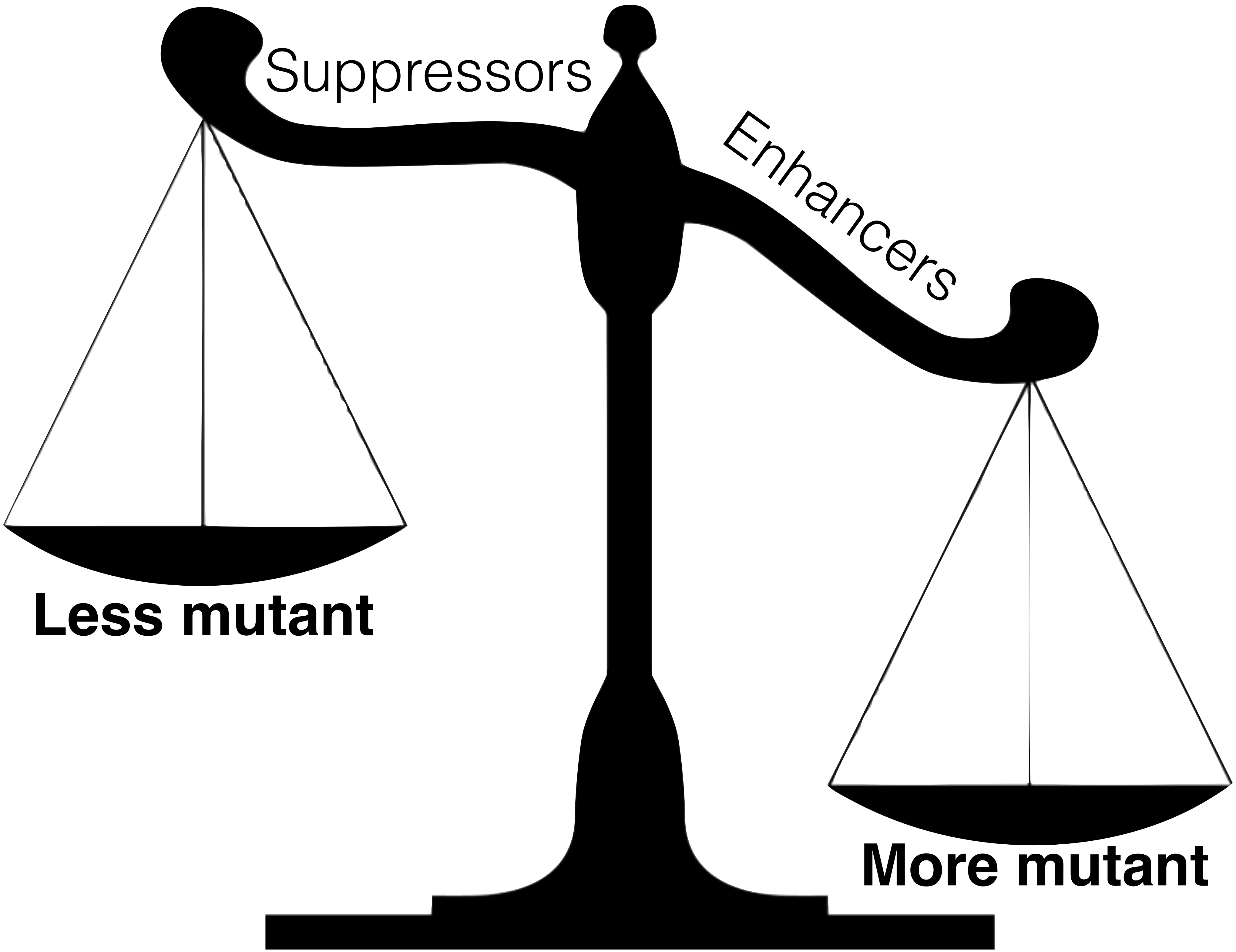
Many amber (UAG) stop codons will be read through

Nonsense suppression as therapy

PTC THERAPEUTICS

- Cystic Fibrosis (CFTR)
- Duchenne muscular dystrophy (dystrophin)
- Beta thalassaemia (beta-globin)
- Hurler syndrome (alpha-L iduronidase)
- Ullrich disease (collagen type VI)

Identification of mutants that modify an existing mutant phenotype



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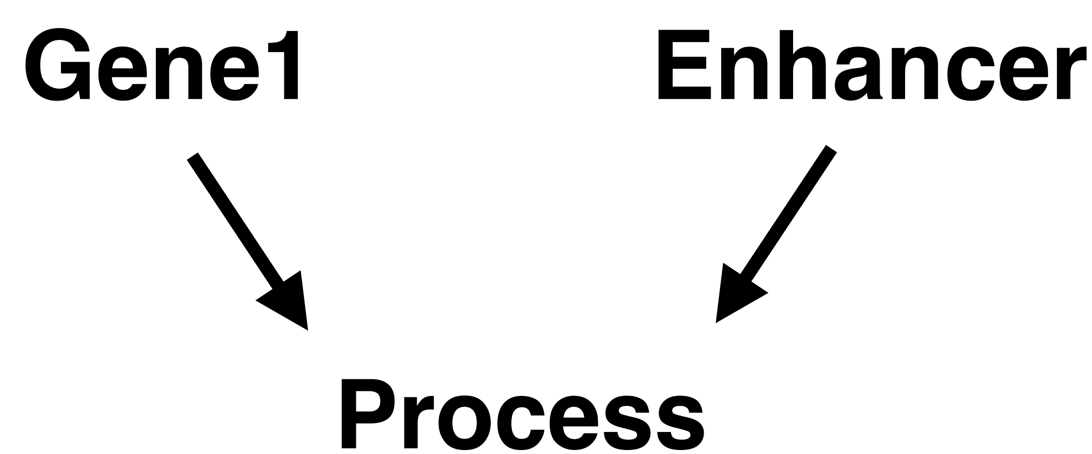

Identification of mutants that modify an existing mutant phenotype

Enhancers make the mutant phenotype worse.

The normal function of the enhancer gene acts with the same effect as the original gene.

Gene1 **Enhancer**

Process

Why do we care?

Enhancers act in redundant processes

1. Gene families (redundancy by similarity of function)

Gene1

Enhancers act in redundant processes

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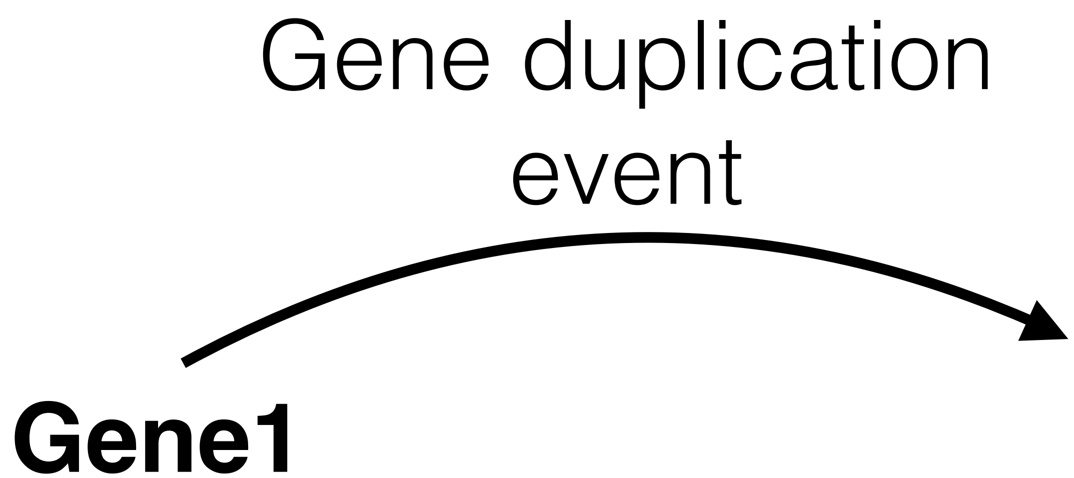

Enhancers act in redundant processes

1. Gene families (redundancy by similarity of function)

Gene duplication
event

Gene1

Gene2

Gene1

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Gene duplication event

Gene1 → **Gene2**

~~**Gene1**~~

Enhancers act in redundant processes

1. Gene families (redundancy by similarity of function)

Gene duplication event

Gene1 **Gene2**

~~**Gene1**~~ Slightly mutant

Enhancers act in redundant processes

1. Gene families (redundancy by similarity of function)

Gene duplication
event

Gene1 → **~~Gene2~~**

~~Gene1~~ Slightly mutant

Enhancers act in redundant processes

1. Gene families (redundancy by similarity of function)

Gene duplication event

Gene1

~~**Gene2**~~ More mutant

~~**Gene1**~~ Slightly mutant

Enhancers act in redundant processes

1. Gene families (redundancy by similarity of function)

2. Parallel pathways (redundancy by similarity of process)

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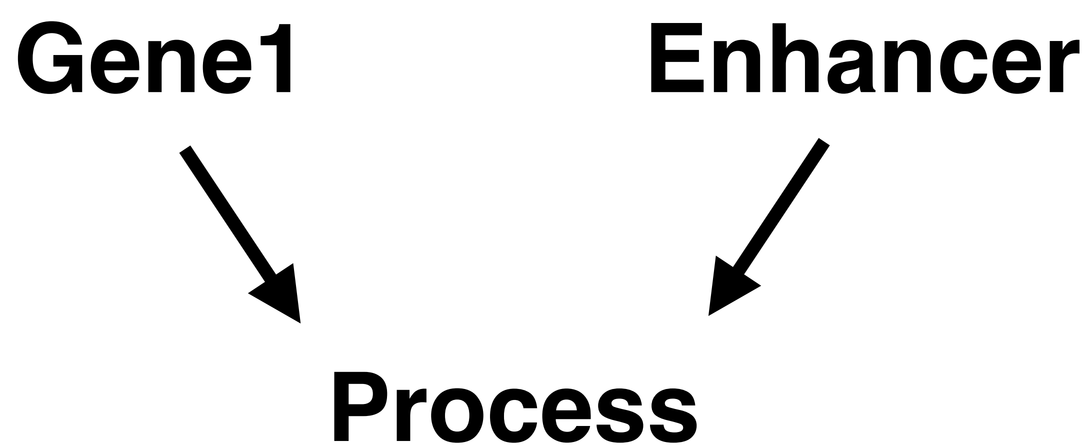

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$$\frac{\text{Gene1} \qquad +}{+ \qquad \text{Gene2}}$$

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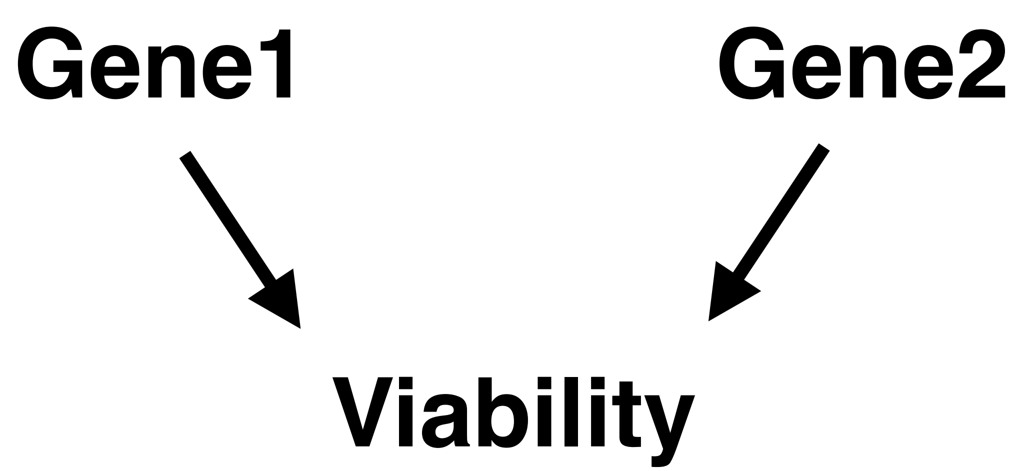
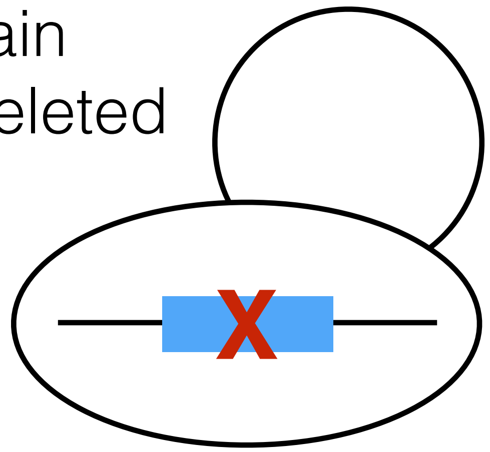


Synthetic lethal screens in yeast

Query strain
with geneX deleted
(MATa)



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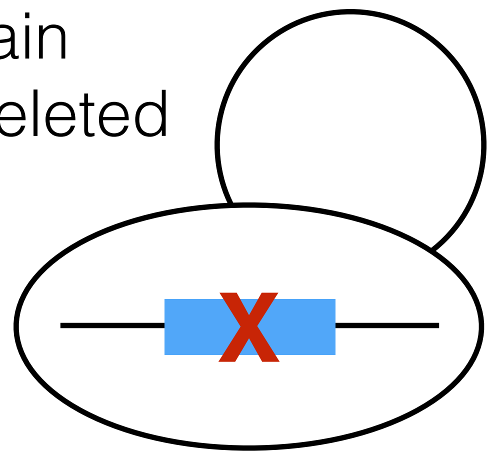
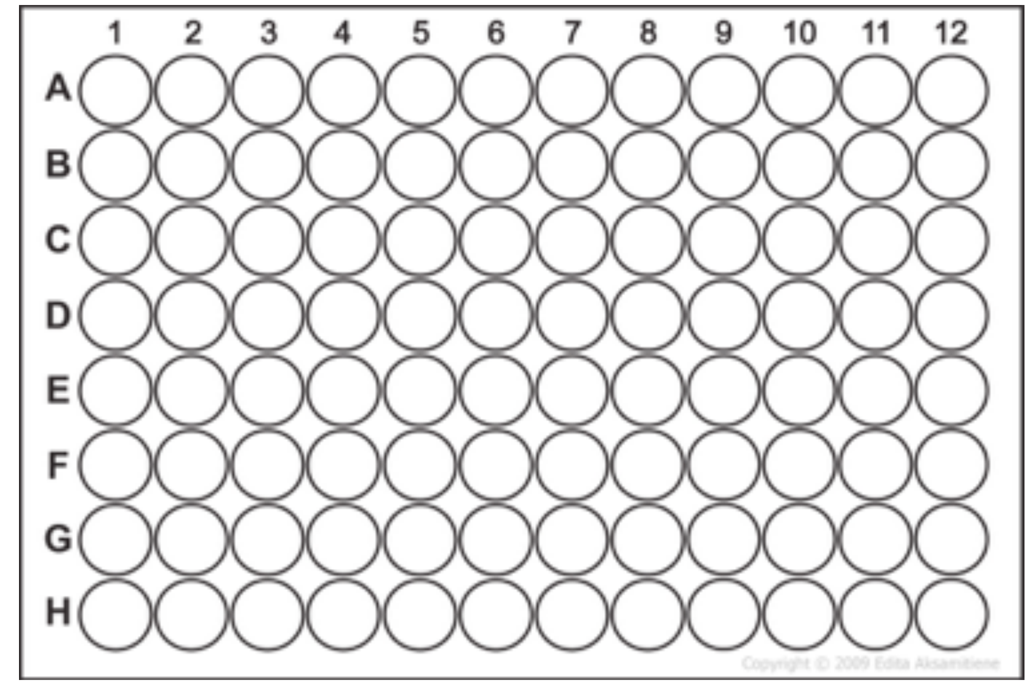


X



96 deletion collection strains
(MATα)

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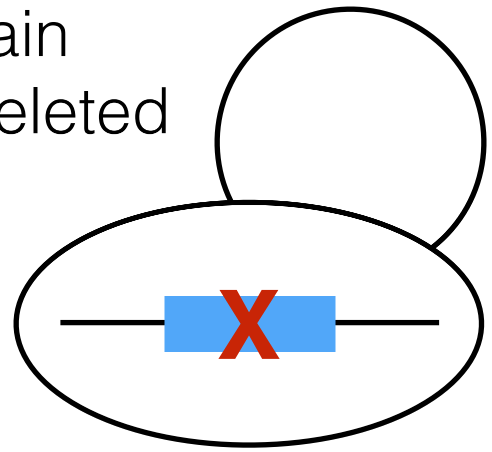
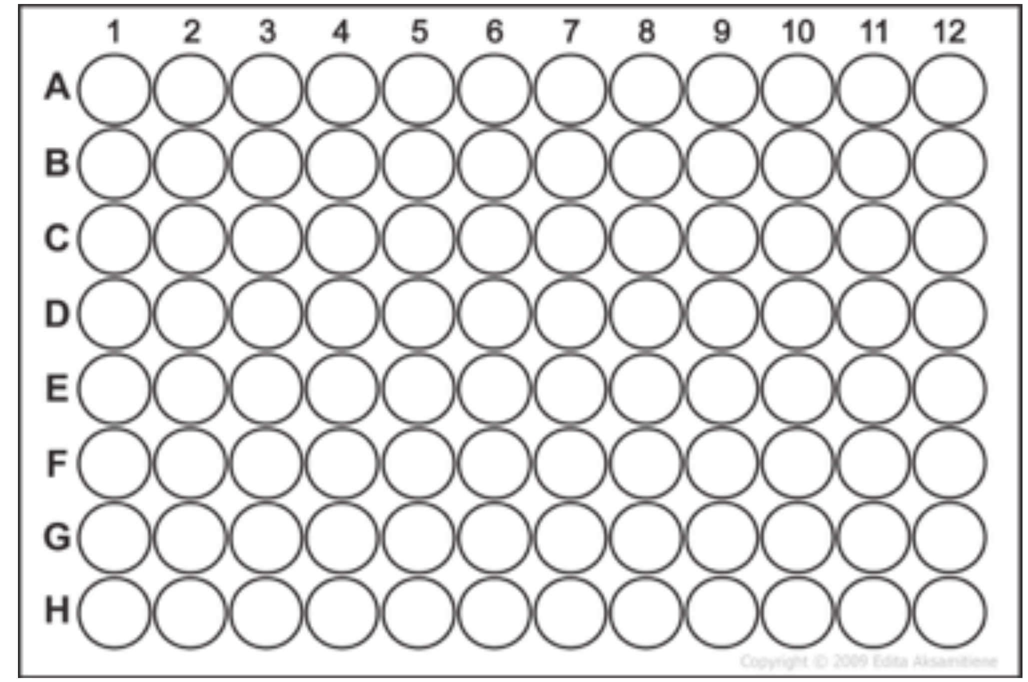


X

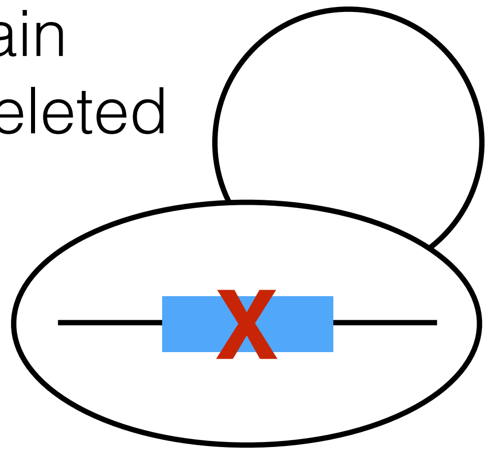


96 deletion collection strains
(MATα)

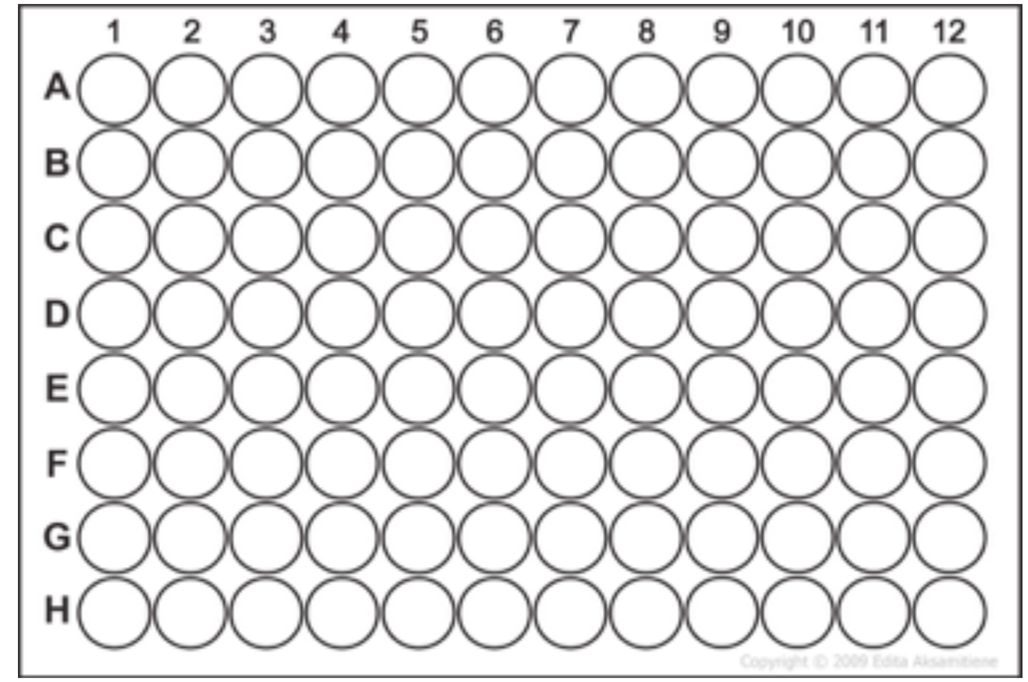
Mating and
sporulation of diploids

Synthetic lethal screens in yeast

Query strain
with geneX deleted
(MATa)



X

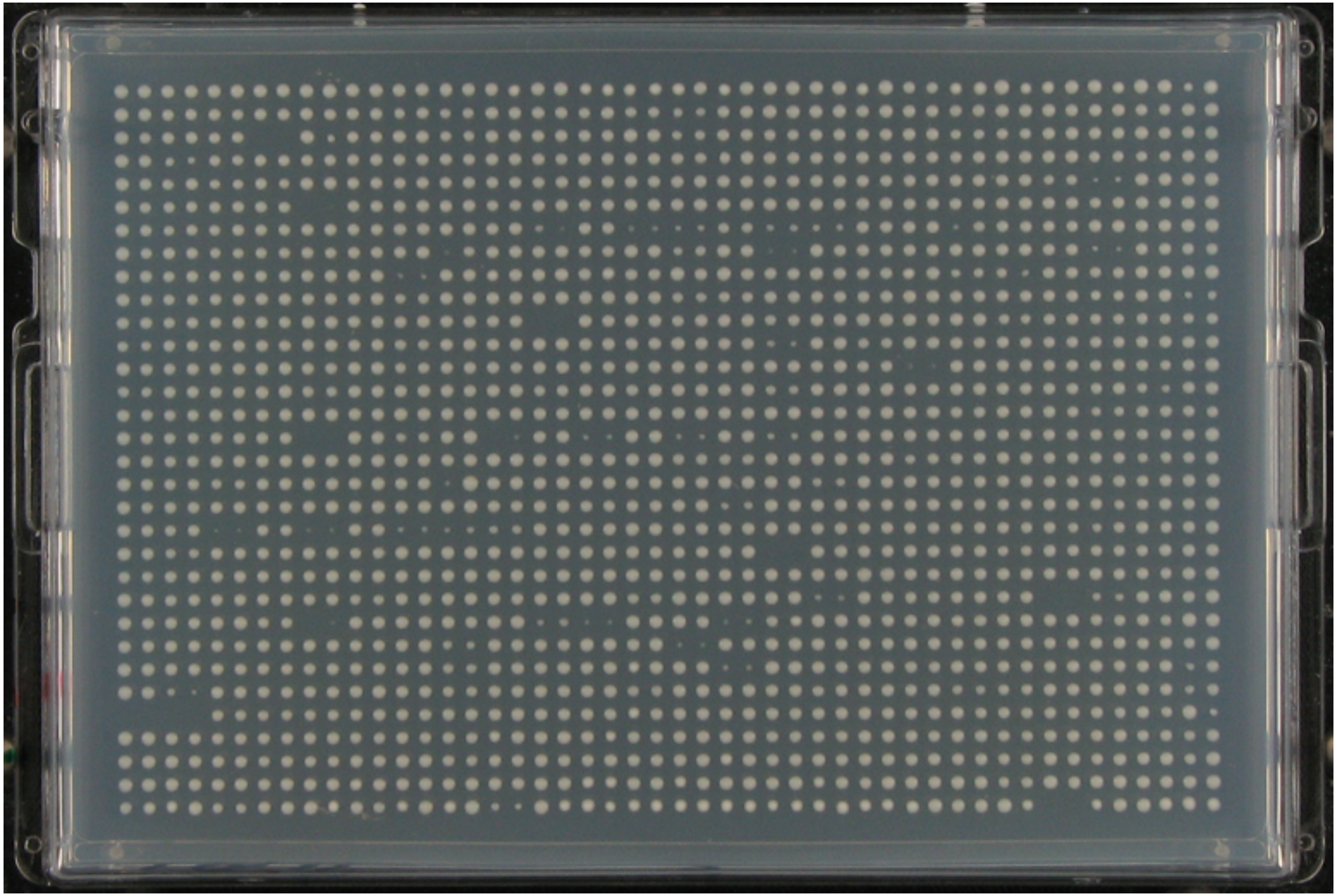


96 deletion collection strains
(MATα)

Mating and
sporulation of diploids

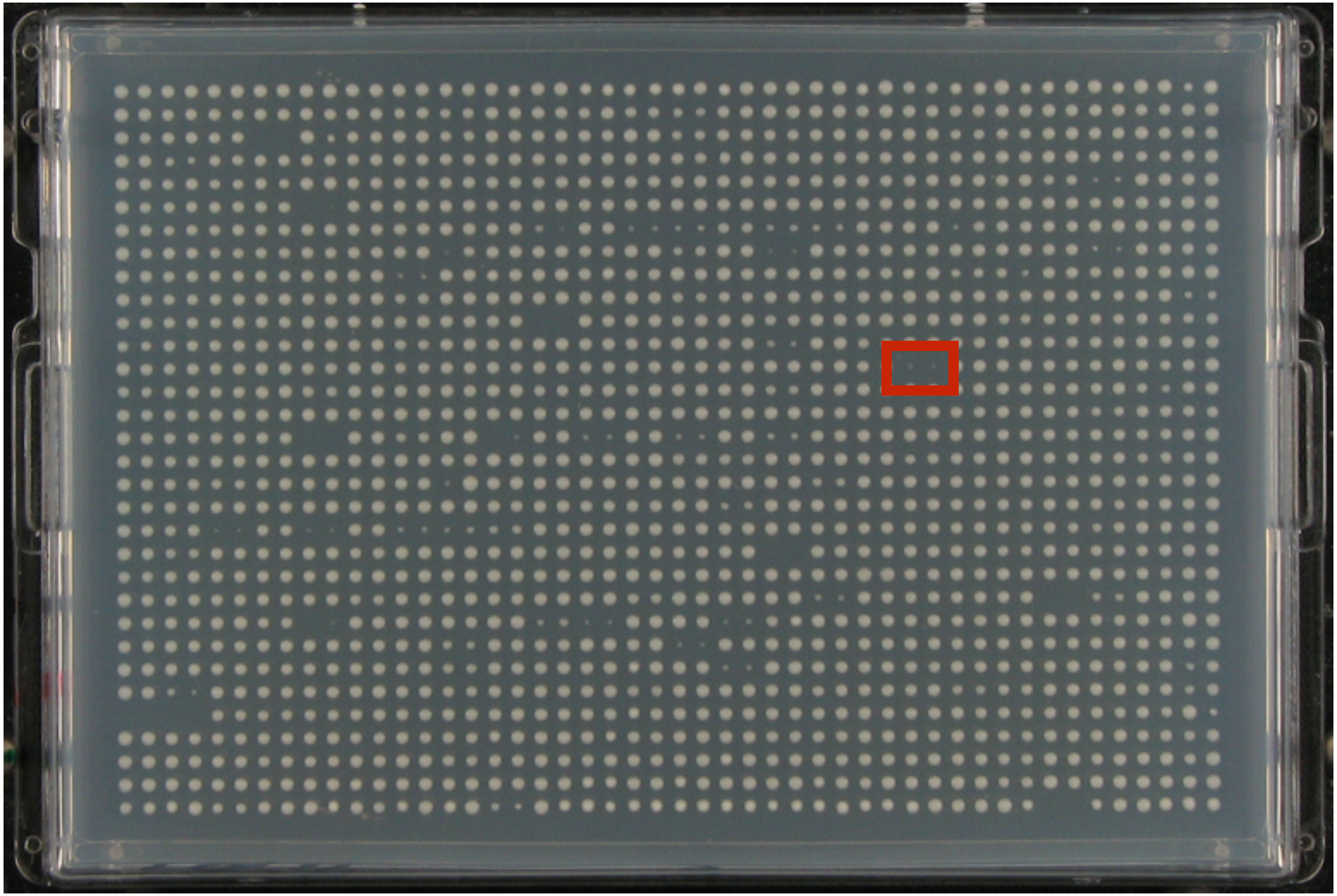
Selection of double mutants

Synthetic lethal screens in yeast



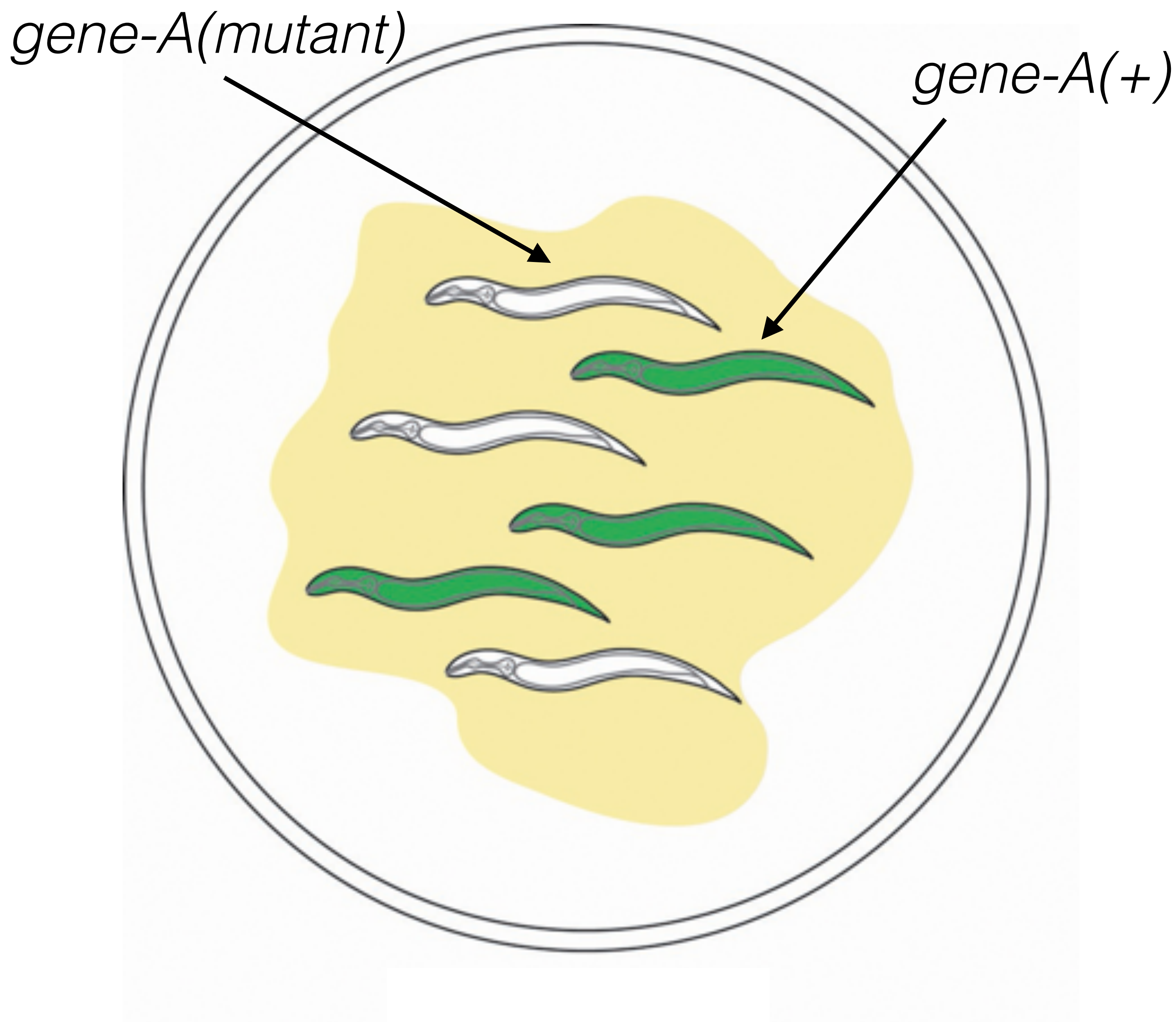
Two 96-well plates arrayed in duplicate
384 total wells

Synthetic lethal screens in yeast



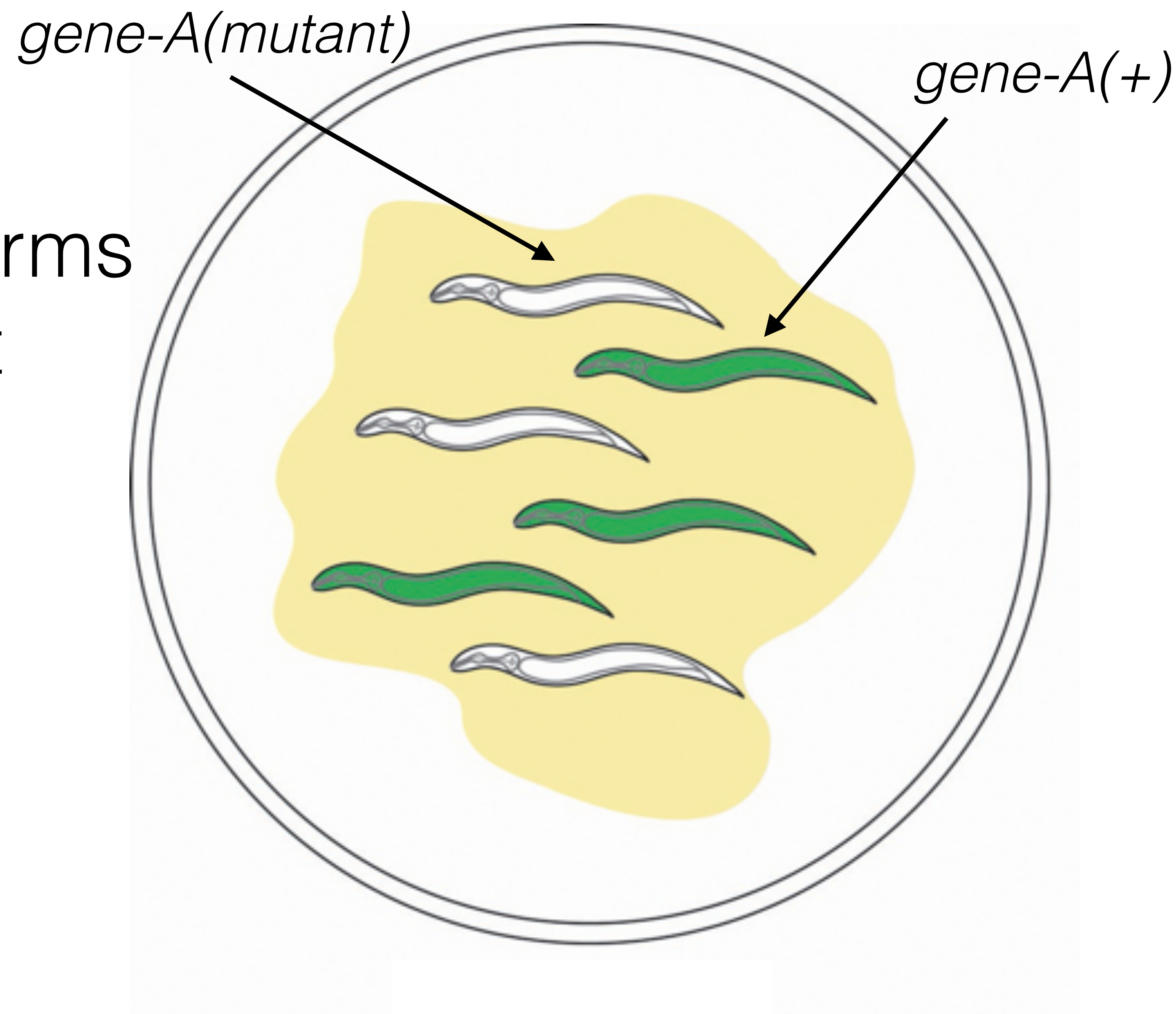
Two 96-well plates arrayed in duplicate
384 total wells

Synthetic lethal screens in *C. elegans*



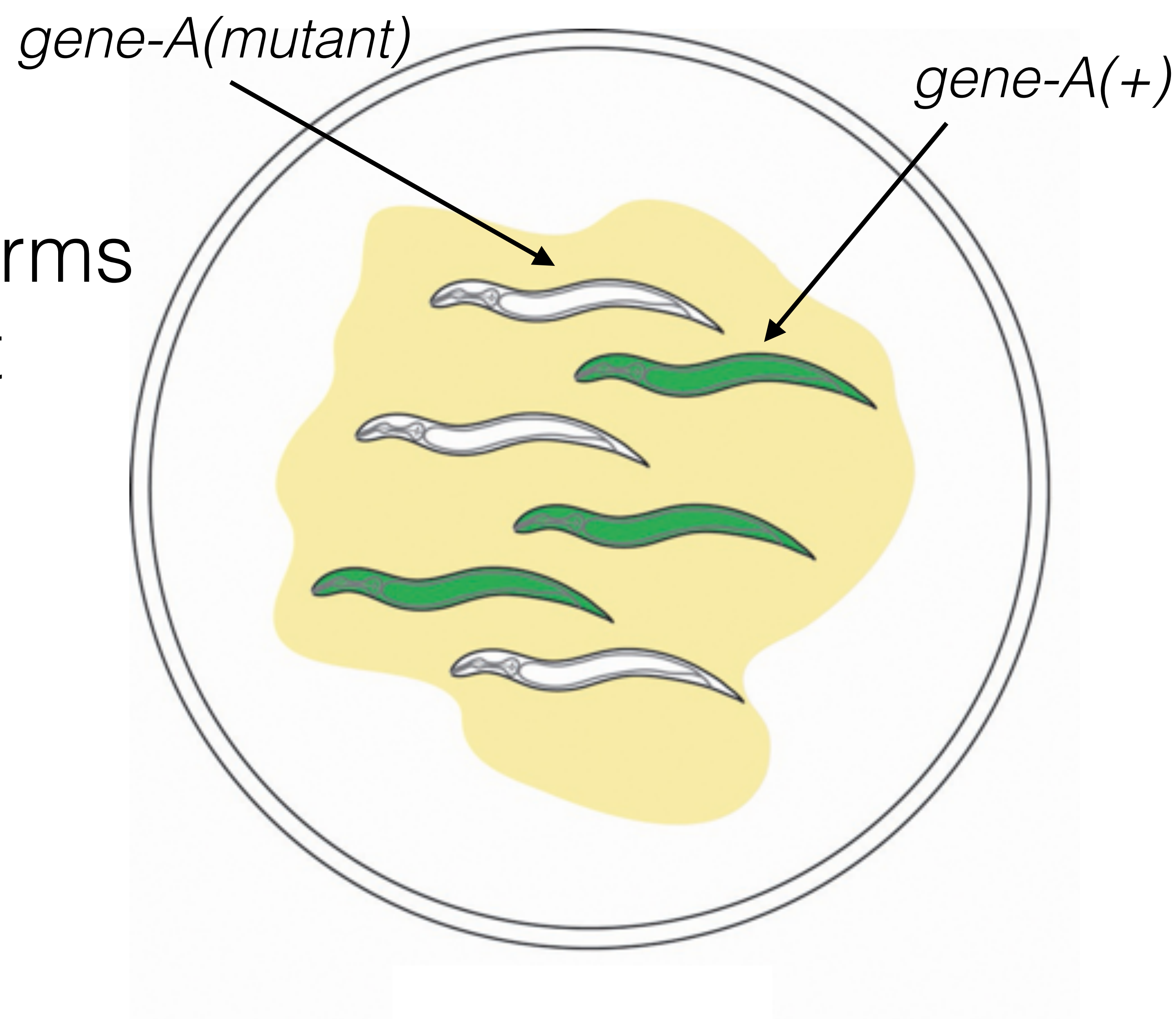
Synthetic lethal screens in *C. elegans*

Non-green worms
are mutant



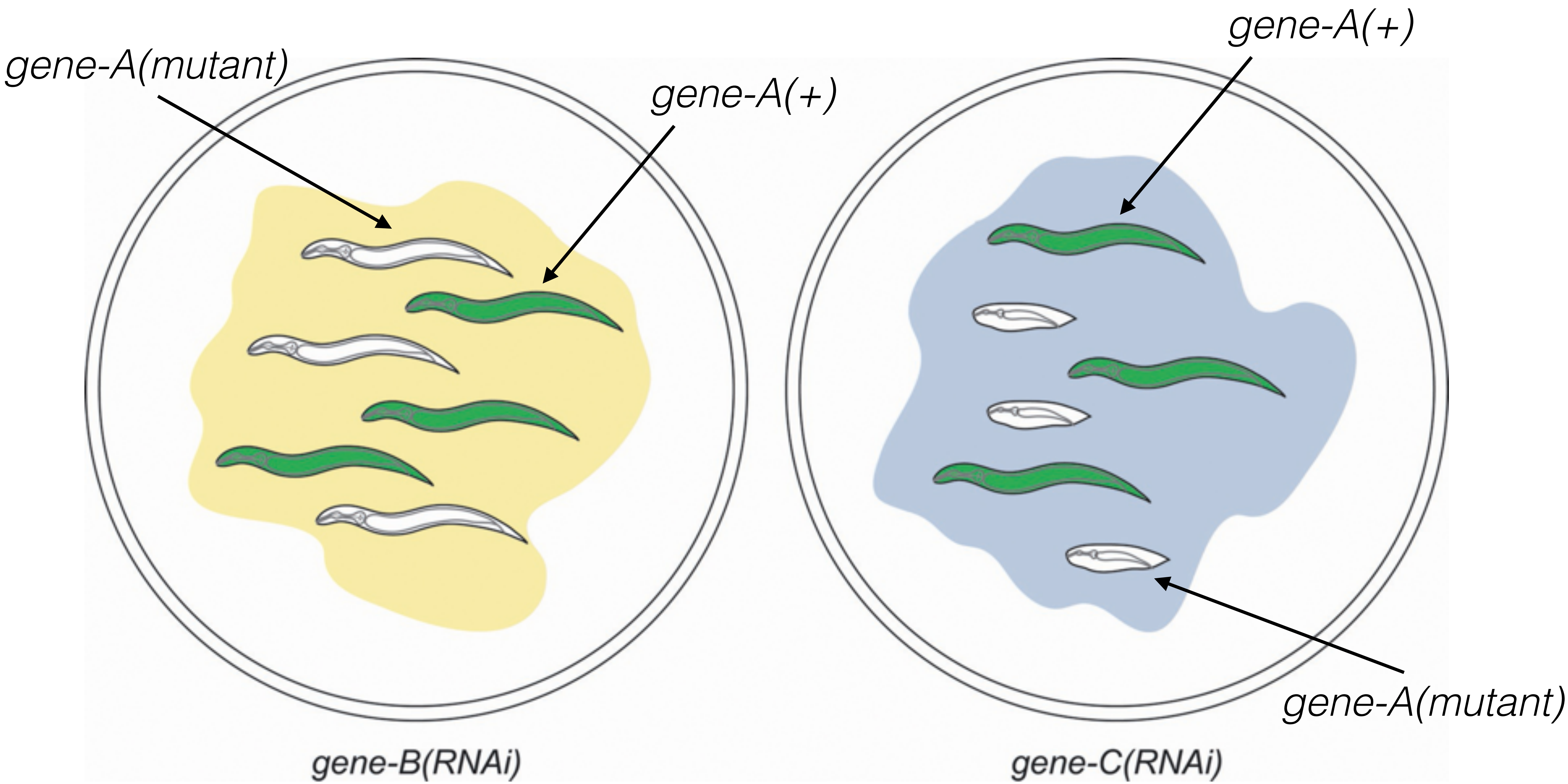
Synthetic lethal screens in *C. elegans*

Non-green worms are mutant

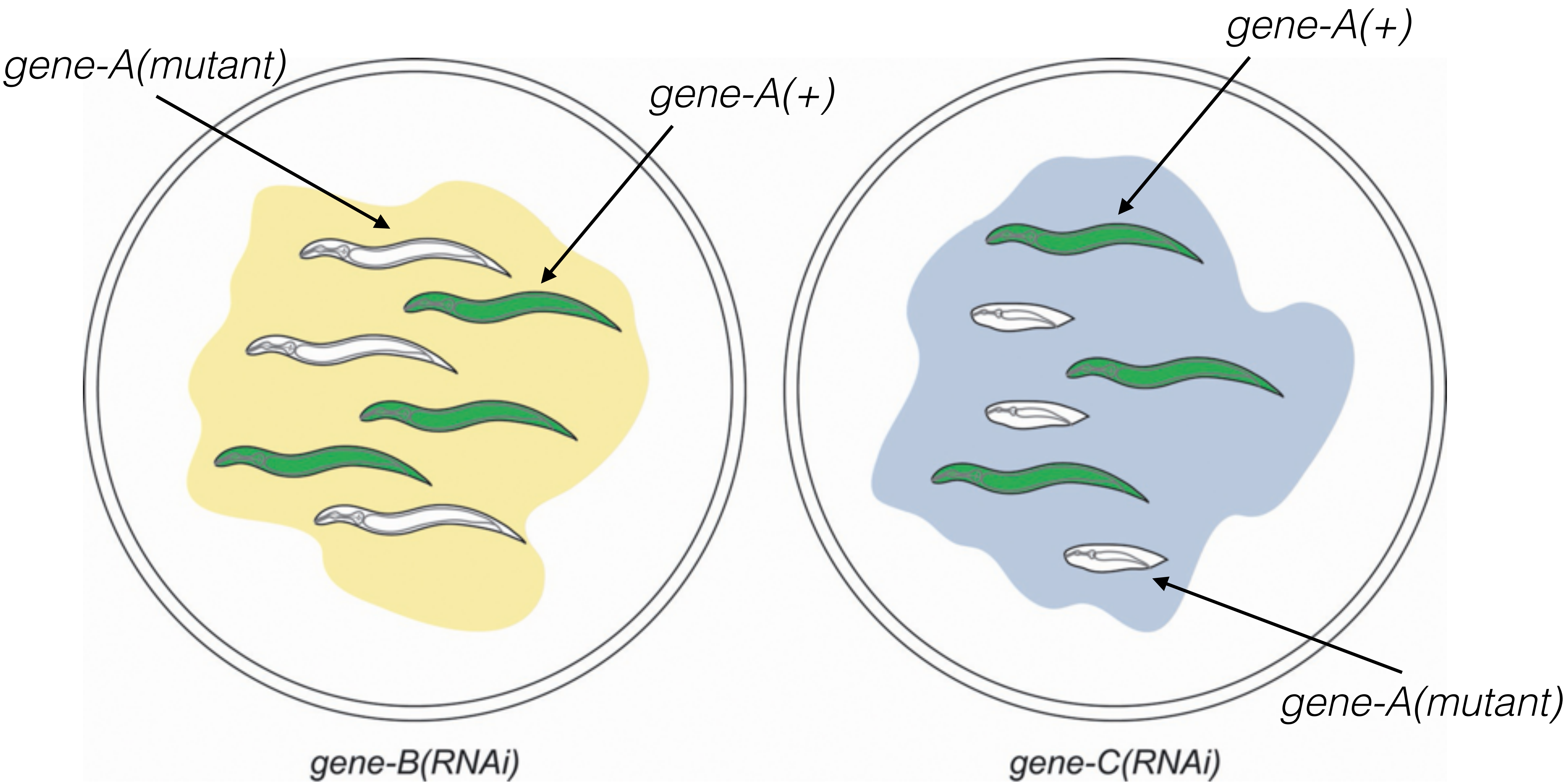


Nearly every gene in the worm can be inactivated by RNA interference

Synthetic lethal screens in *C. elegans*



Synthetic lethal screens in *C. elegans*



Loss of *gene-A* and *gene-C* is synthetic lethal