

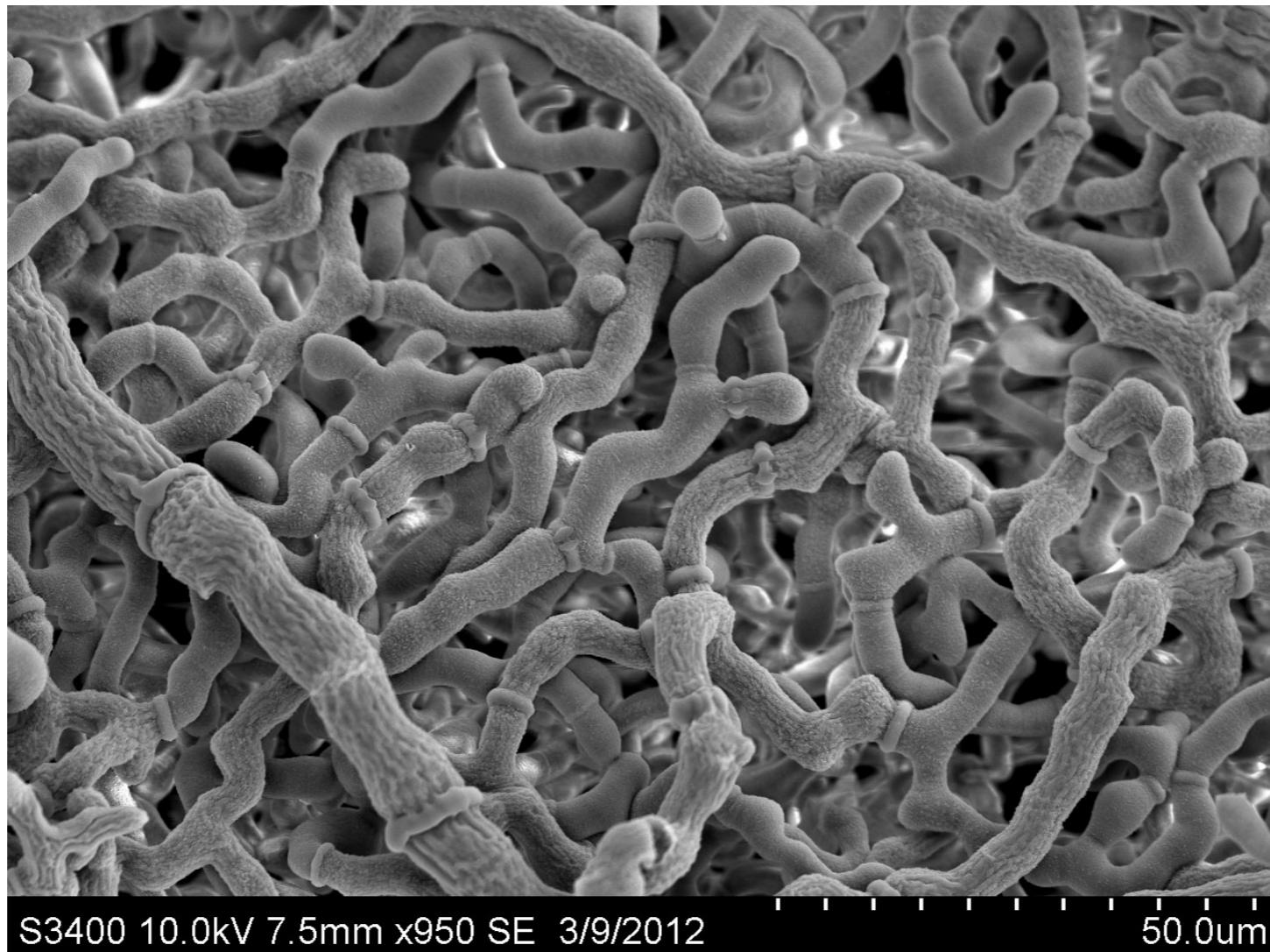
Genetic interactions: epistasis



George Beadle



Ed Tatum

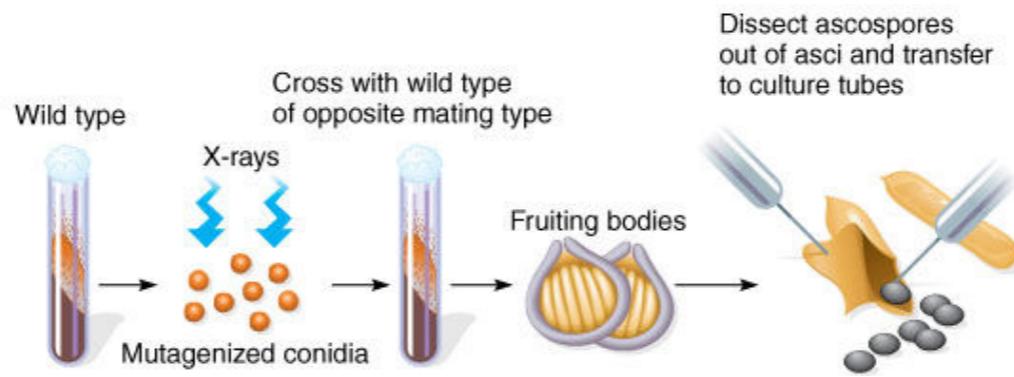


S3400 10.0kV 7.5mm x950 SE 3/9/2012

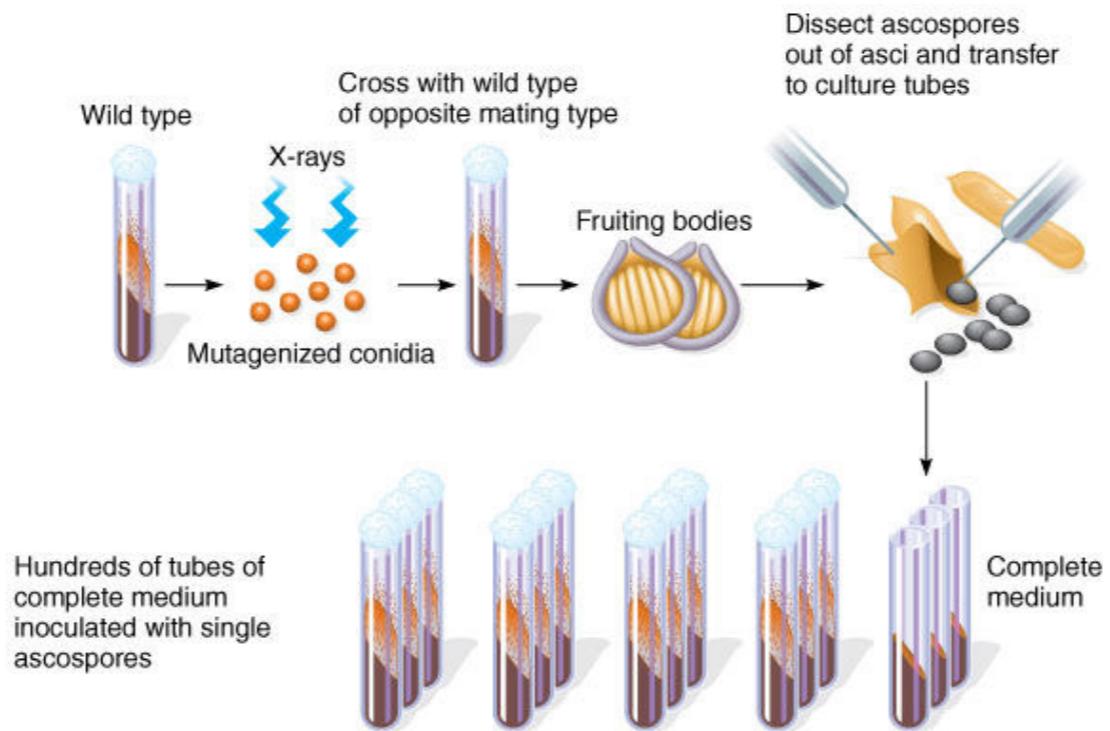
50.0μm

Neurospora crassa

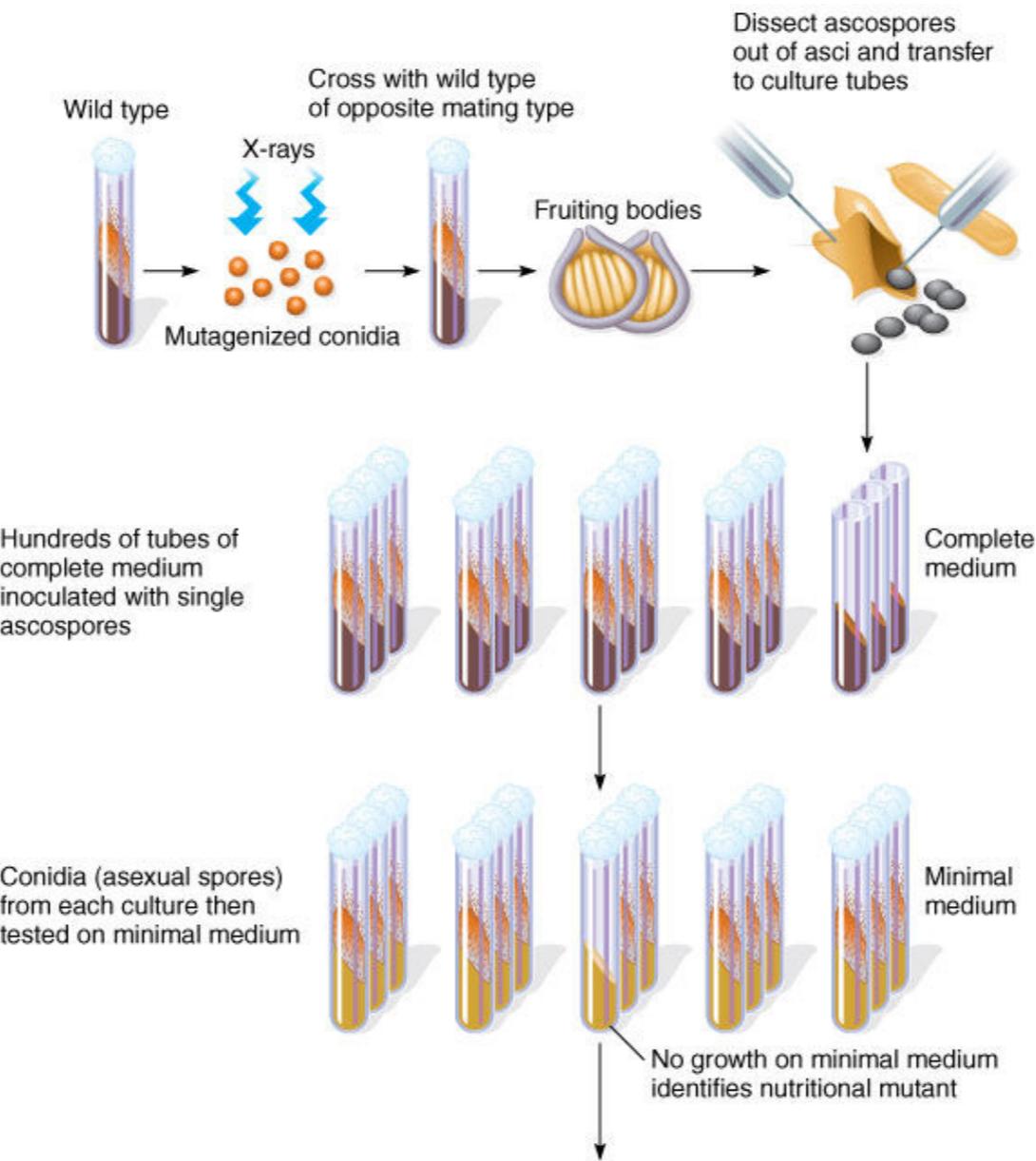
The Beadle- Tatum Experiment



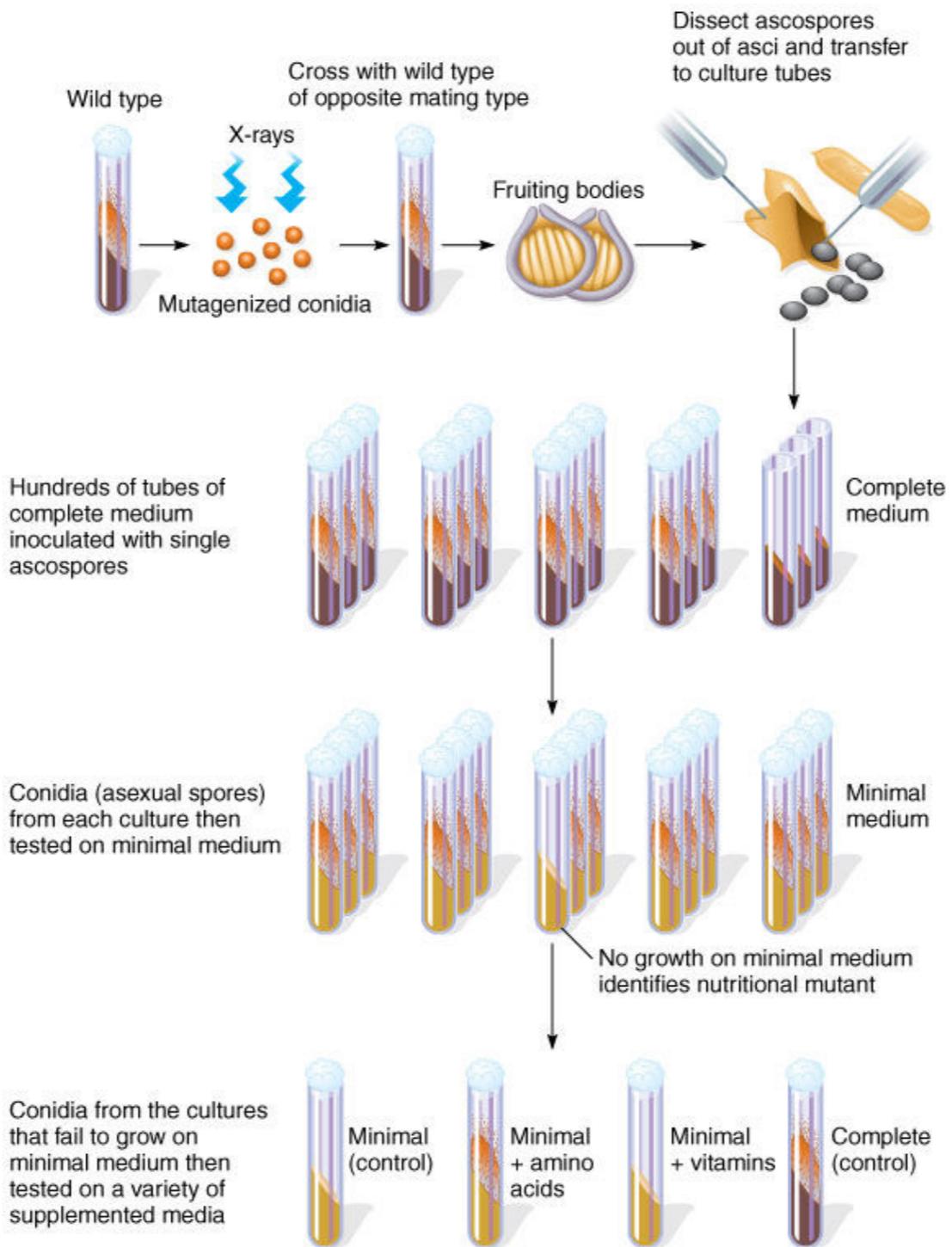
The Beadle- Tatum Experiment



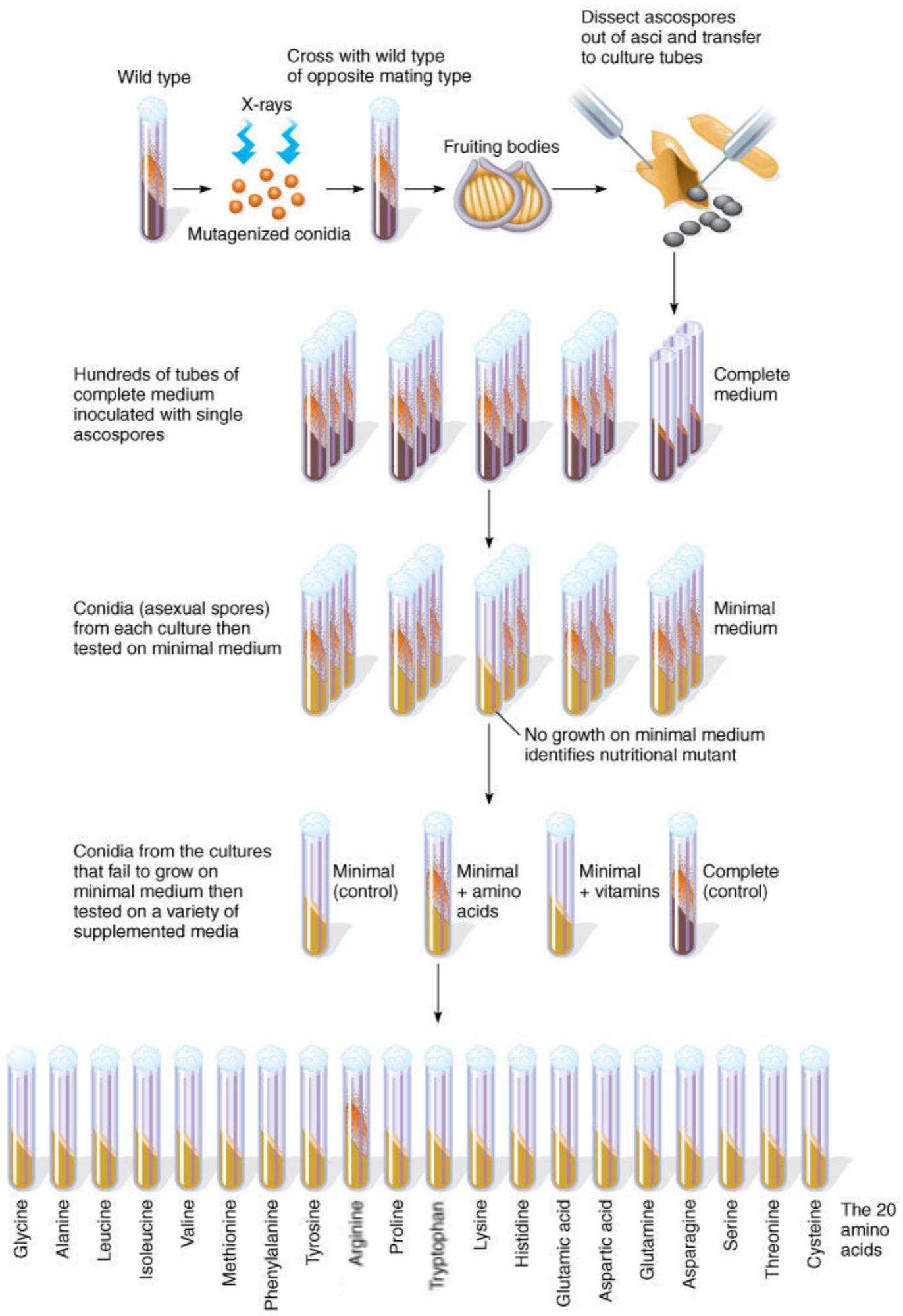
The Beadle- Tatum Experiment



The Beadle- Tatum Experiment

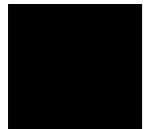


The Beadle- Tatum Experiment



Arginine mutant complementation experiment

	<i>arg-a</i>	<i>arg-b</i>	<i>arg-c</i>	<i>arg-d</i>	<i>arg-e</i>	<i>arg-f</i>	<i>arg-g</i>	<i>arg-h</i>	<i>arg-i</i>
<i>arg-a</i>									
<i>arg-b</i>									
<i>arg-c</i>									
<i>arg-d</i>									
<i>arg-e</i>									
<i>arg-f</i>									
<i>arg-g</i>									
<i>arg-h</i>									
<i>arg-i</i>									



Growth = wild type



No growth = mutant

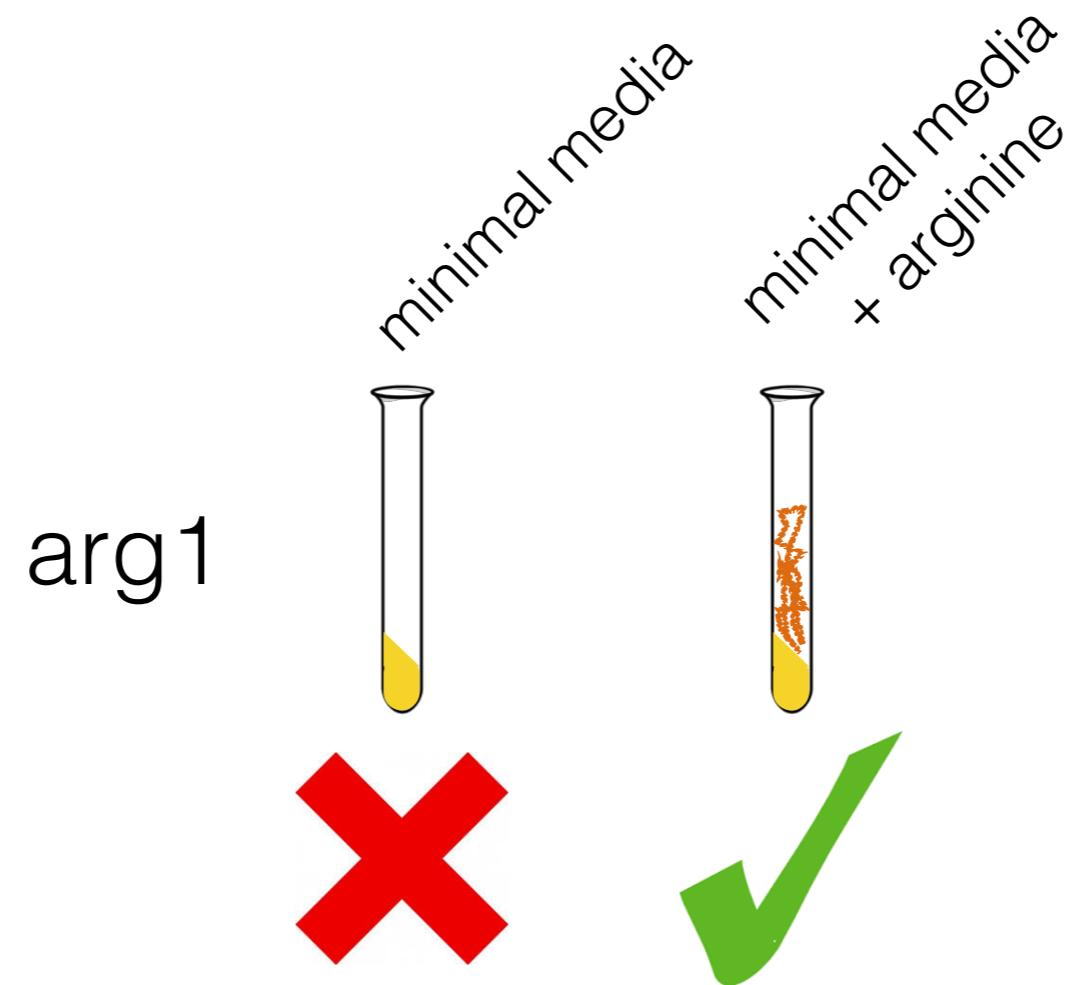
Arginine mutant complementation experiment

Three genes

arg1 = [a, d, f, g]

arg2 = [b, c]

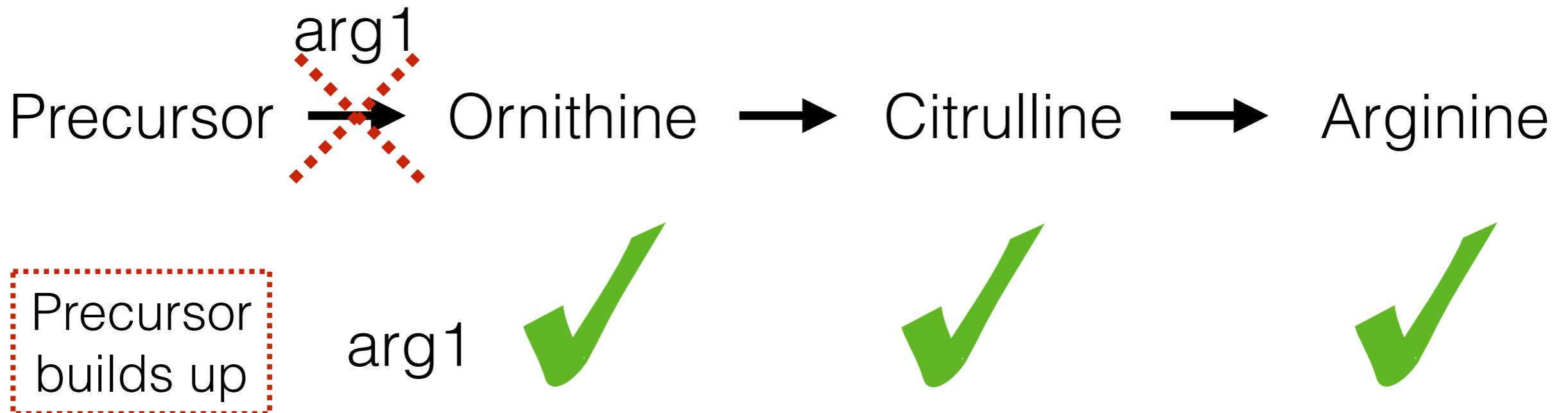
arg3 = [e, h, i]



One gene - one enzyme hypothesis

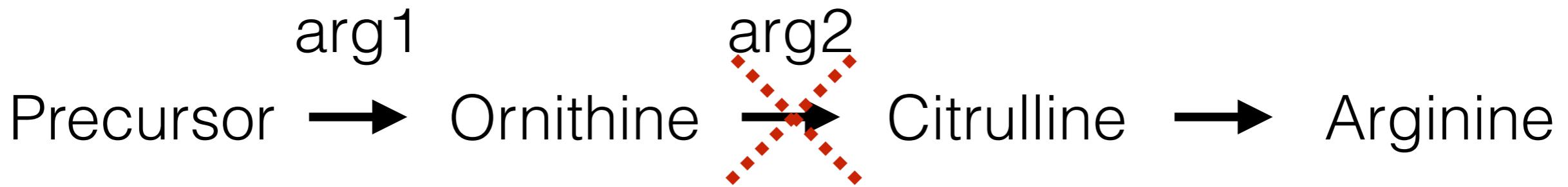
Precursor → Ornithine → Citrulline → Arginine

One gene - one enzyme hypothesis



Mutants accumulate precursor for previous step

One gene - one enzyme hypothesis

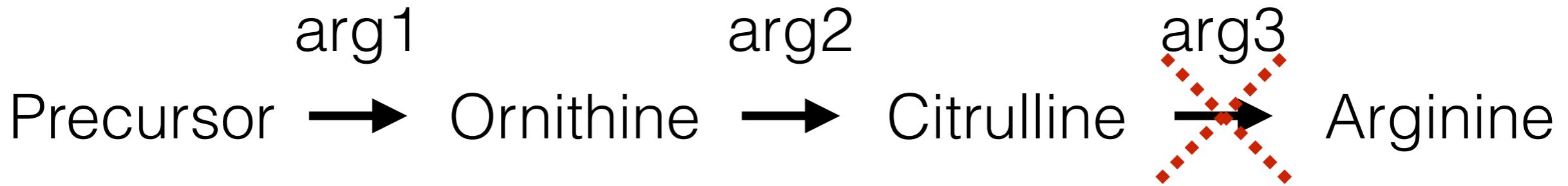


Precursor
builds up



Mutants accumulate precursor for previous step

One gene - one enzyme hypothesis



Precursor builds up

Ornithine builds up

Citrulline builds up



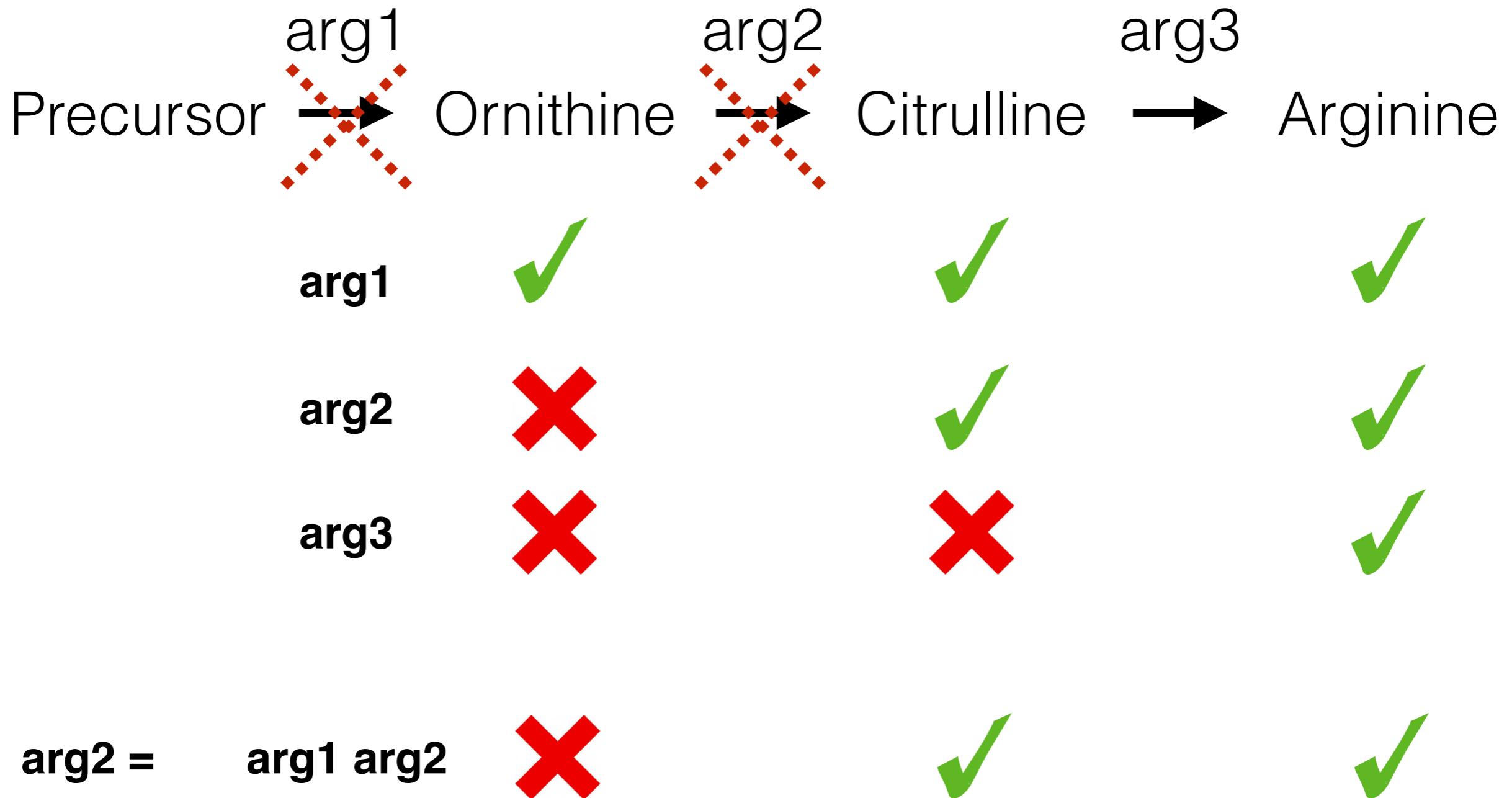
Mutants accumulate precursor for previous step

Epistasis: the effect of one gene is dependent on another gene



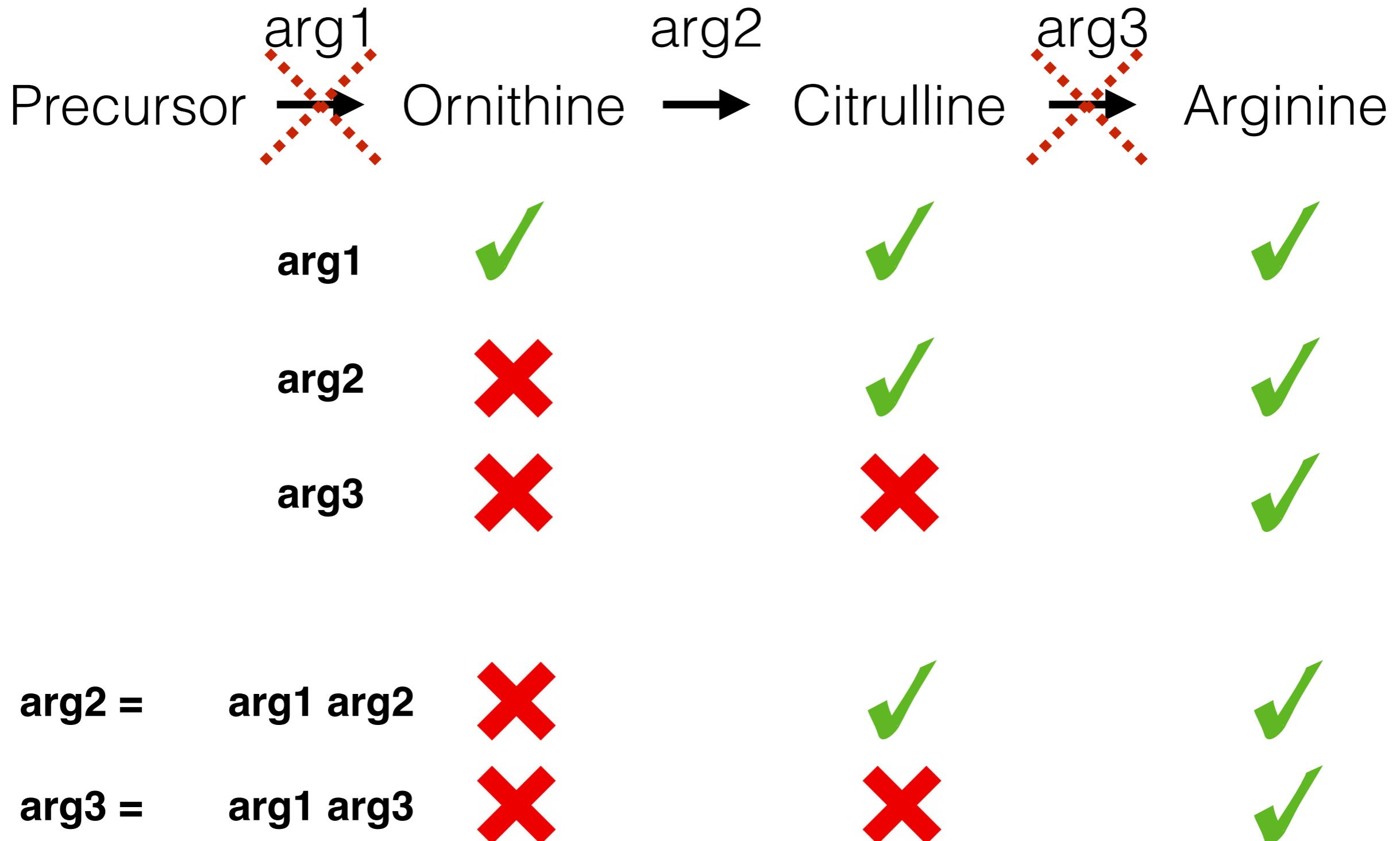
William Bateson

Biochemical epistasis



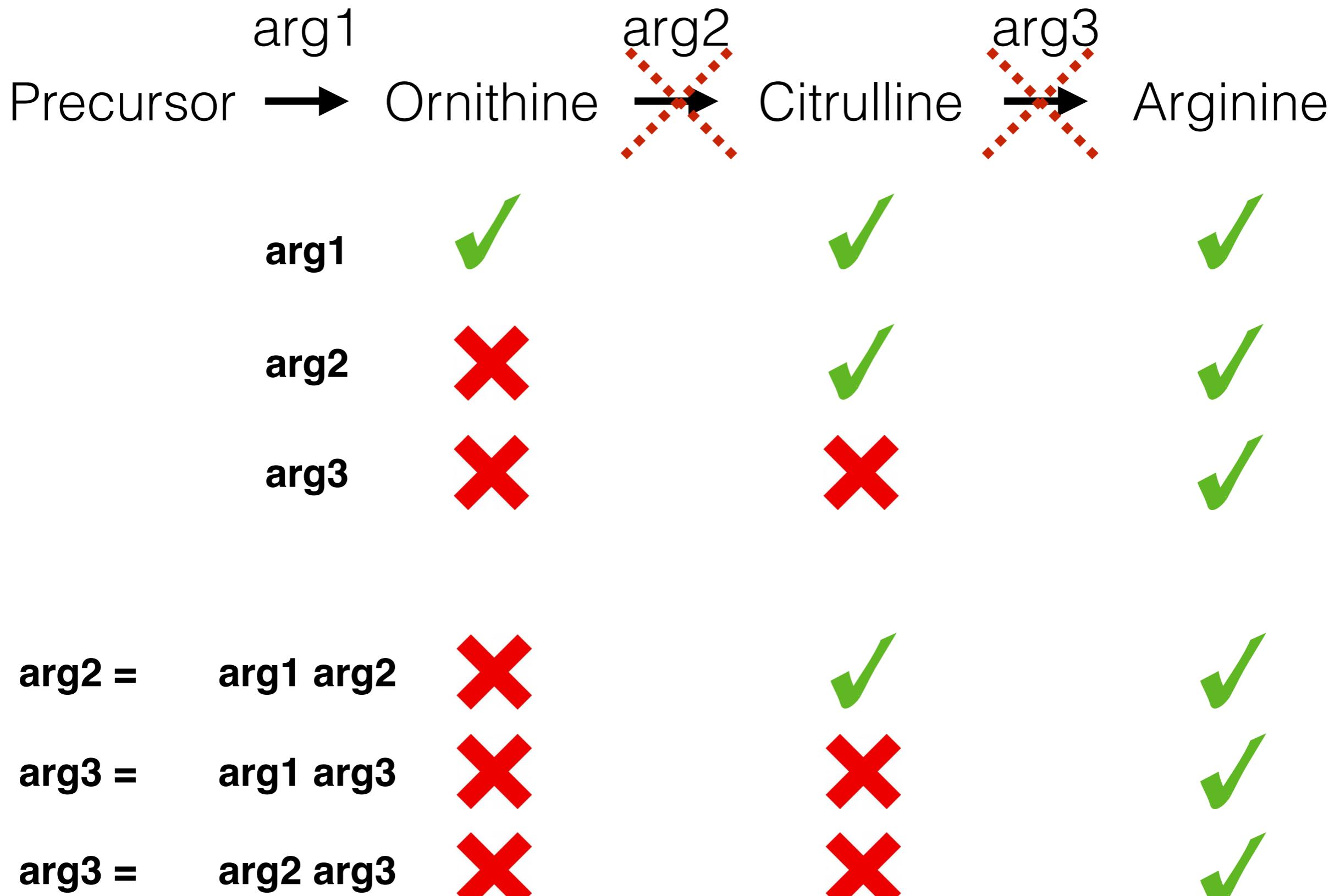
arg2 is epistatic to arg1, arg2 is downstream (or in parallel) to arg1

Biochemical epistasis



arg3 is epistatic to arg1, arg3 is downstream (or in parallel) to arg1

Biochemical epistasis



arg3 is epistatic to arg2, arg3 is downstream (or in parallel) to arg2

Approach to understanding biochemical epistasis



1. Single mutants fail in a step in a biosynthetic pathway
2. Double mutants fail in two steps. The phenotype dictates the most downstream gene in the pathway.
3. What will the single and double mutants accumulate?
4. Pathways can be branched

Epistasis - one mutant phenotype masks another



(A)



(B)

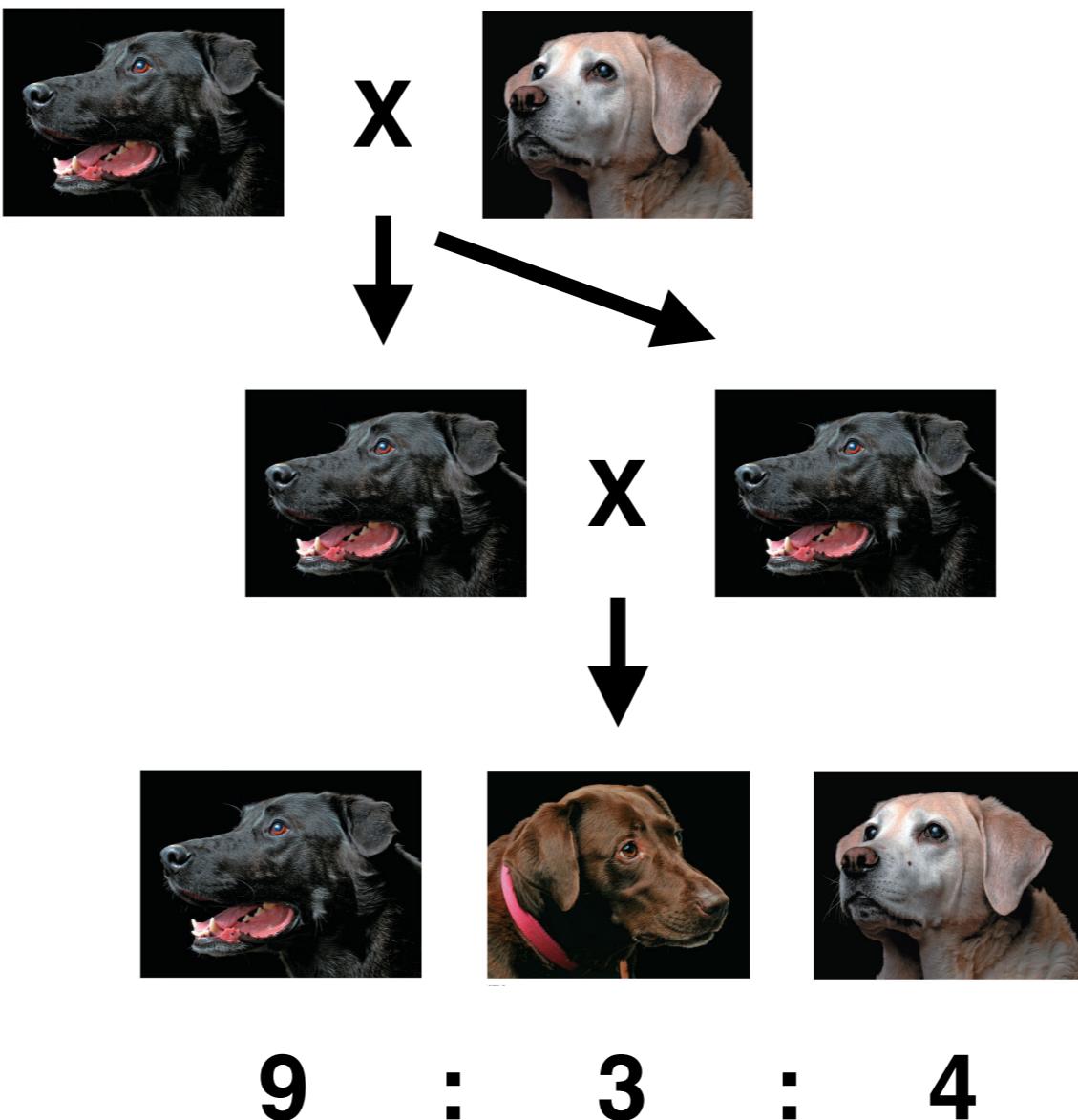


(C)

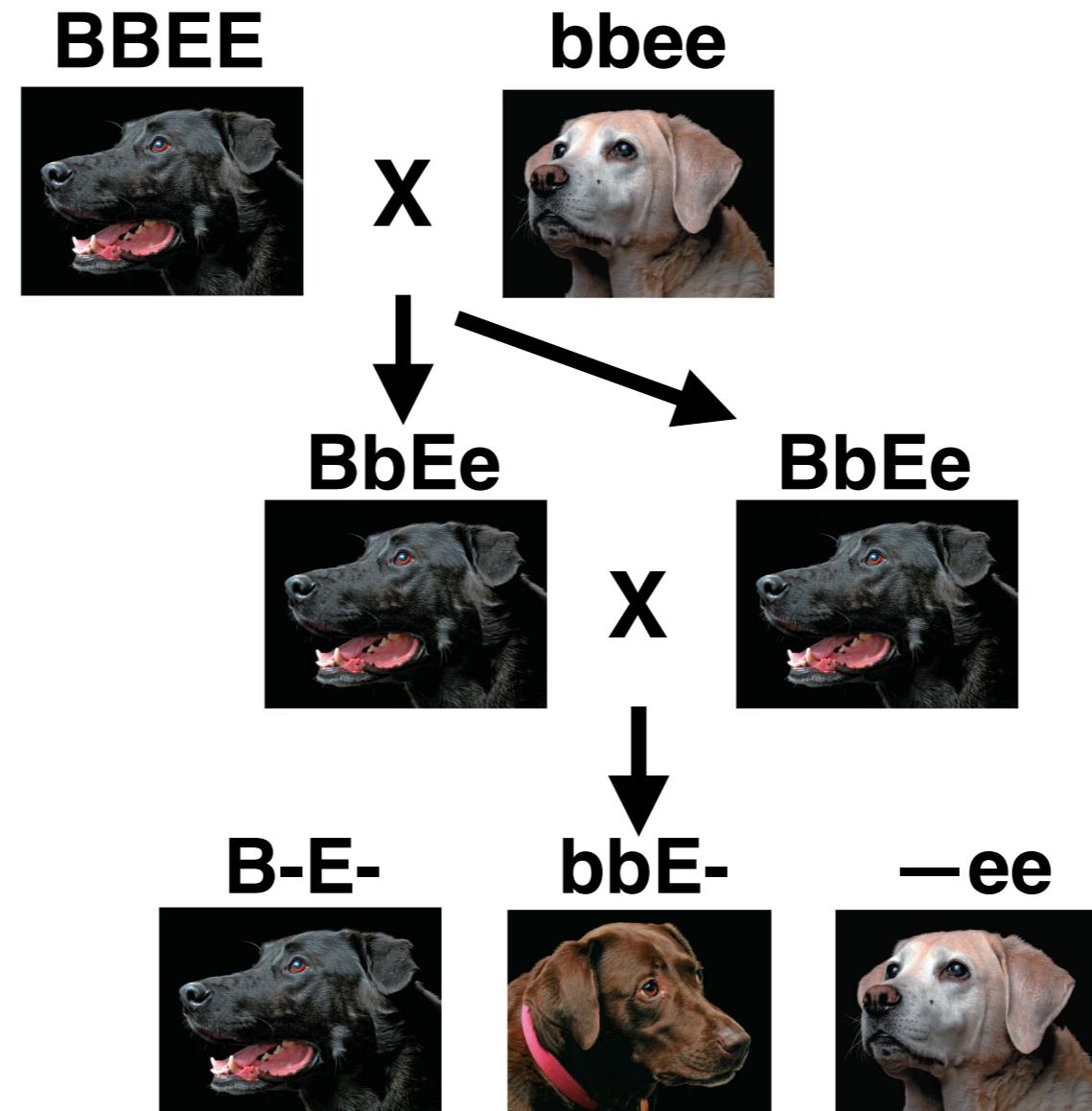
LIFE 8e, Figure 10.14

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Epistasis - one mutant phenotype masks another



Epistasis - one mutant phenotype masks another



B = black

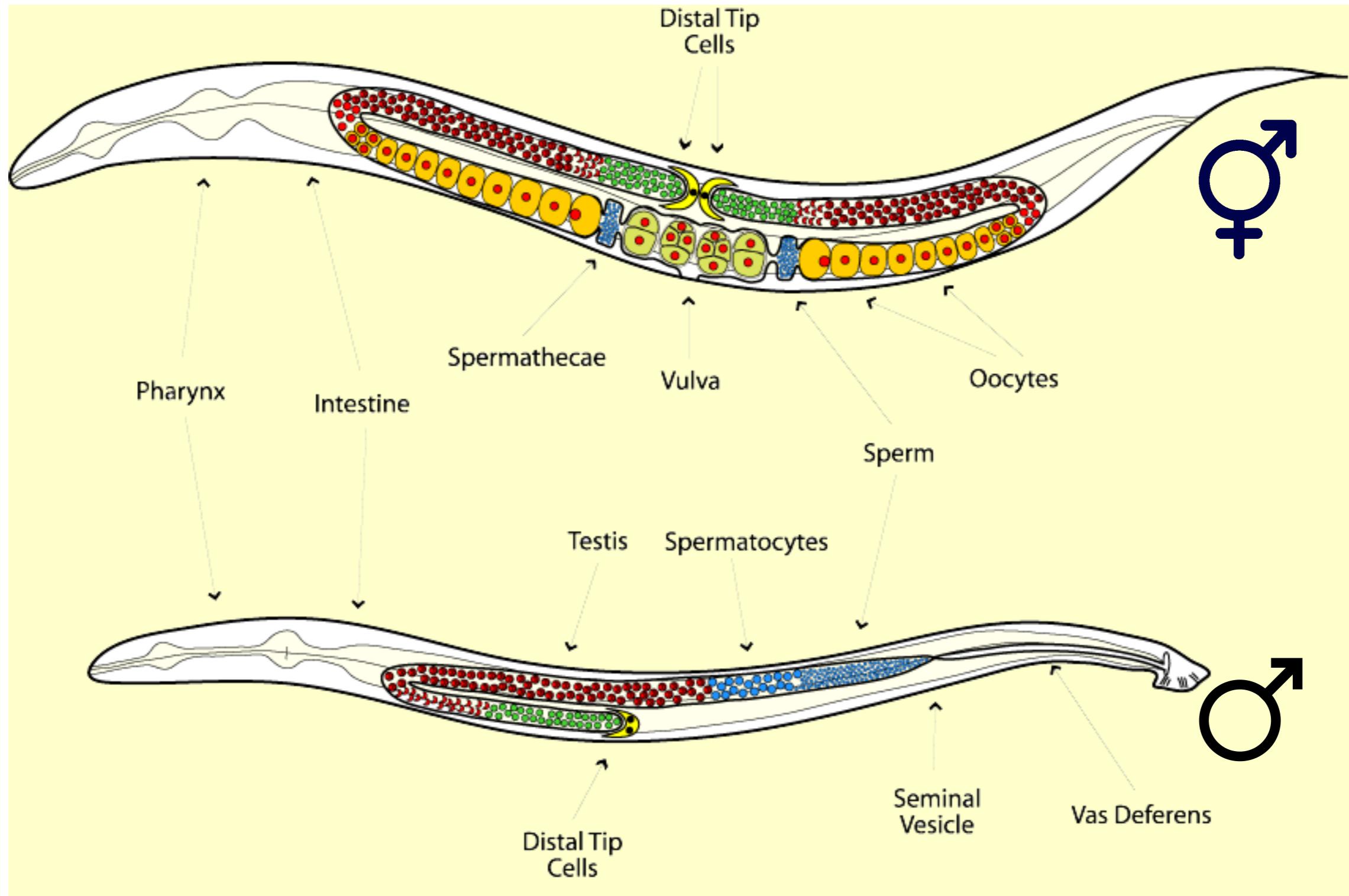
b = brown

E = color

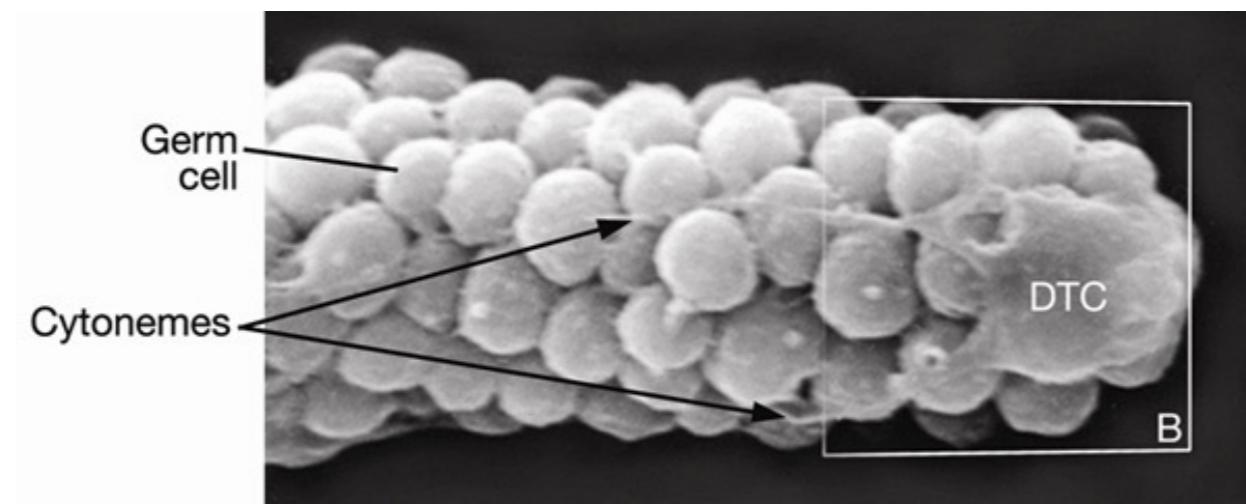
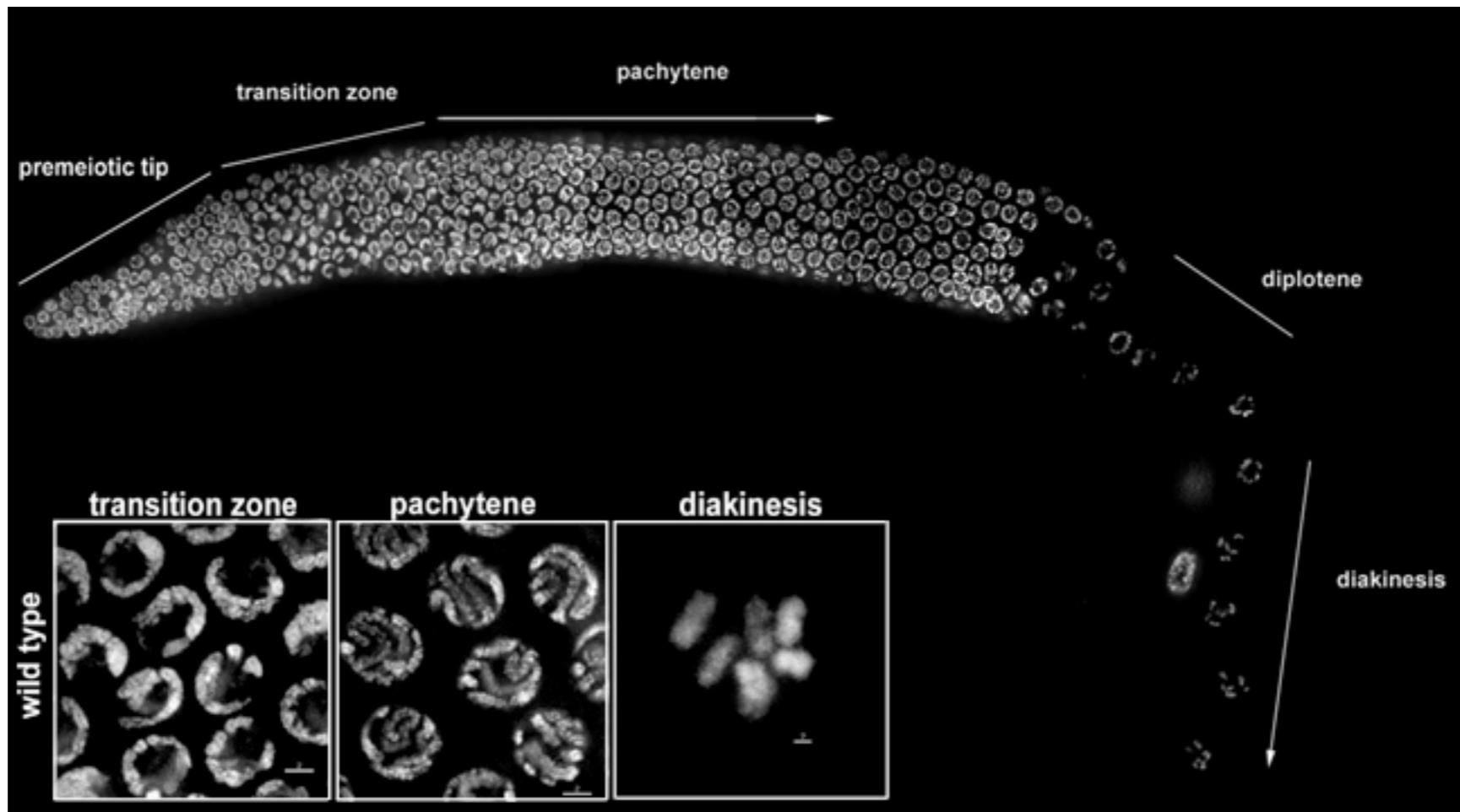
e = no color

9 : 3 : 4

The *C. elegans* germline



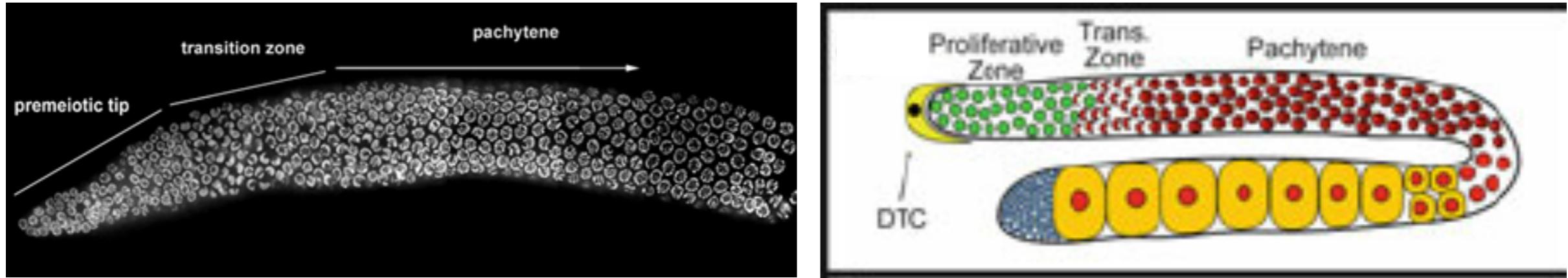
The *C. elegans* germline





Judith Kimble

C. elegans germline mutants

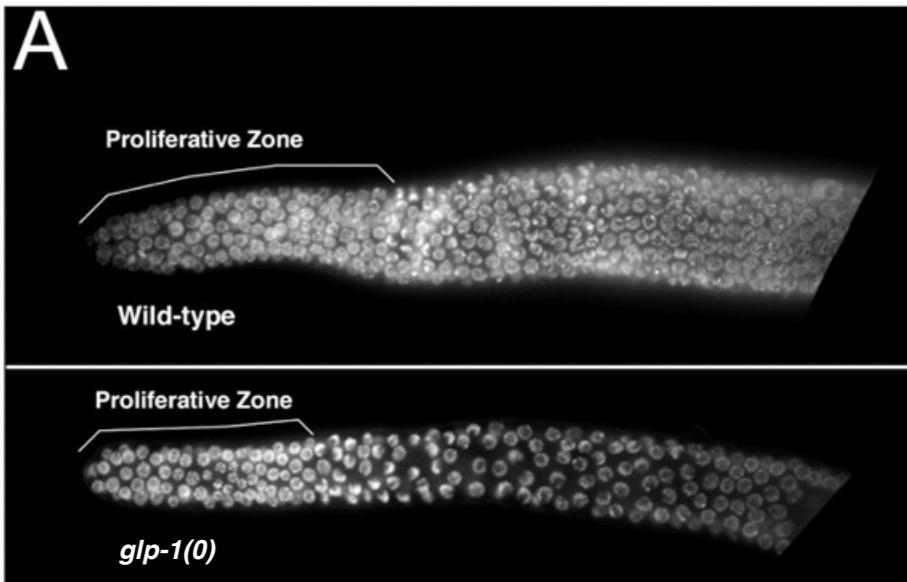


glp-1(0) = all meiotic germ cells

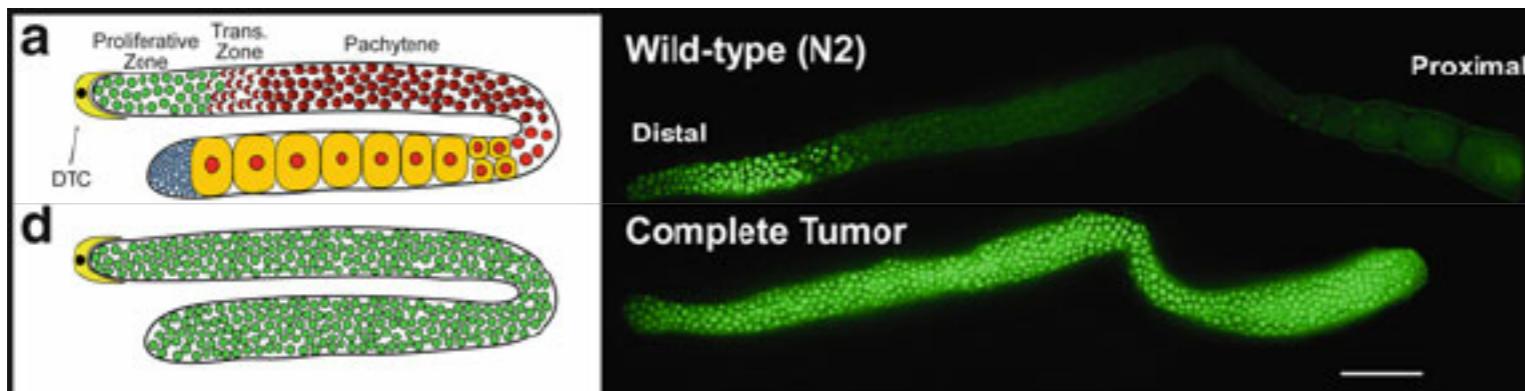
glp-1(gf) = all mitotic germ cells

glp-1 → **germ cell proliferation**

C. elegans germline mutants



glp-1(0) =
more meiosis, less mitosis



glp-1(gf) =
less meiosis,
more mitosis

glp-1 → germ cell proliferation

C. elegans germline mutants

Mutant	Phenotype
<i>glp-1(0)</i>	meiotic cells
<i>glp-1(gf)</i>	mitotic cells
<i>lag-2(0)</i>	meiotic cells
<i>fbf-1(0)</i>	meiotic cells
<i>gld-1(0)</i>	mitotic cells

***glp-1* → GSC prolif.**

***lag-2* → GSC prolif.**

***fbf-1* → GSC prolif.**

***gld-1* → GSC prolif.**

How do these genes work together?

Mutant	Phenotype
<i>glp-1(0)</i>	meiotic cells
<i>glp-1(gf)</i>	mitotic cells
<i>lag-2(0)</i>	meiotic cells
<i>fbf-1(0)</i>	meiotic cells
<i>gld-1(0)</i>	mitotic cells
<i>glp-1(0); lag-2(0)</i>	meiotic cells

You can only do epistasis tests with mutants that have different phenotypes

How do these genes work together?

Mutant	Phenotype
<i>glp-1(0)</i>	meiotic cells
<i>glp-1(gf)</i>	mitotic cells
<i>lag-2(0)</i>	meiotic cells
<i>fbf-1(0)</i>	meiotic cells
<i>gld-1(0)</i>	mitotic cells
<i>glp-1(gf); lag-2(0)</i>	mitotic cells

***glp-1* → GSC prolif.**

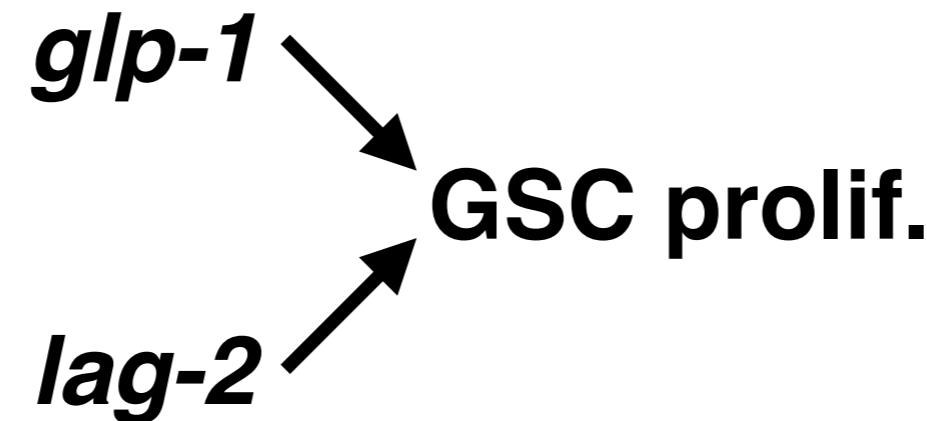
***lag-2* → GSC prolif.**

Which phenotype
is epistatic?

***lag-2* → *glp-1* → GSC prolif.**

Parallel gene action can NEVER be formally excluded by phenotype alone

***lag-2* → *glp-1* → GSC prolif.**



Null alleles have to be used for this reason.

Approach to understanding regulatory epistasis

1. Decide what is the output phenotype; keep it consistent
2. Look at single mutants and make a model with output
3. Look at double mutants and make a model with output and respect to single mutant models - epistatic gene acts downstream
4. Remember parallel but don't assume it is always parallel (*i.e.* make linear models for regulatory epistasis)
5. Remember two negatives make a positive

How do these genes work together?

Mutant	Phenotype
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<i>glp-1(gf)</i>	mitotic cells
<i>lag-2(0)</i>	meiotic cells
<i>fbf-1(0)</i>	meiotic cells
<i>gld-1(0)</i>	mitotic cells
<i>glp-1(gf); lag-2(0)</i>	mitotic cells
<i>glp-1(gf); fbf-1(0)</i>	meiotic cells
<i>glp-1(0); gld-1(0)</i>	mitotic cells
<i>fbf-1(0); gld-1(0)</i>	mitotic cells
<i>lag-2(0); fbf-1(0)</i>	meiotic cells
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<i>gld-1(0)</i>	mitotic cells
<i>glp-1(gf); lag-2(0)</i>	mitotic cells
<i>glp-1(gf); fbf-1(0)</i>	meiotic cells
<i>glp-1(0); gld-1(0)</i>	mitotic cells
<i>fbf-1(0); gld-1(0)</i>	mitotic cells
<i>lag-2(0); fbf-1(0)</i>	meiotic cells
<i>lag-2(0); gld-1(0)</i>	mitotic cells

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***fbf-1* → GSC prolif.**
***gld-1* → GSC prolif.**

***lag-2* → *glp-1* → *fbf-1* → GSC prolif.**

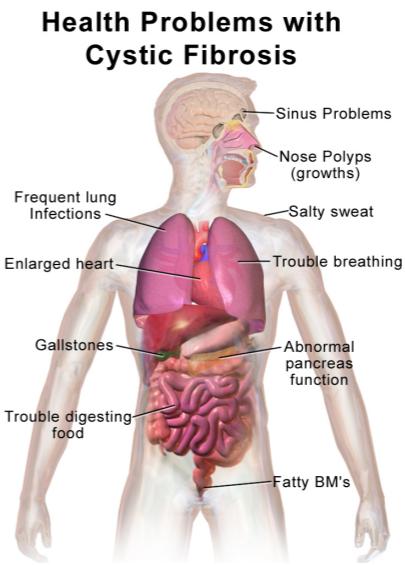
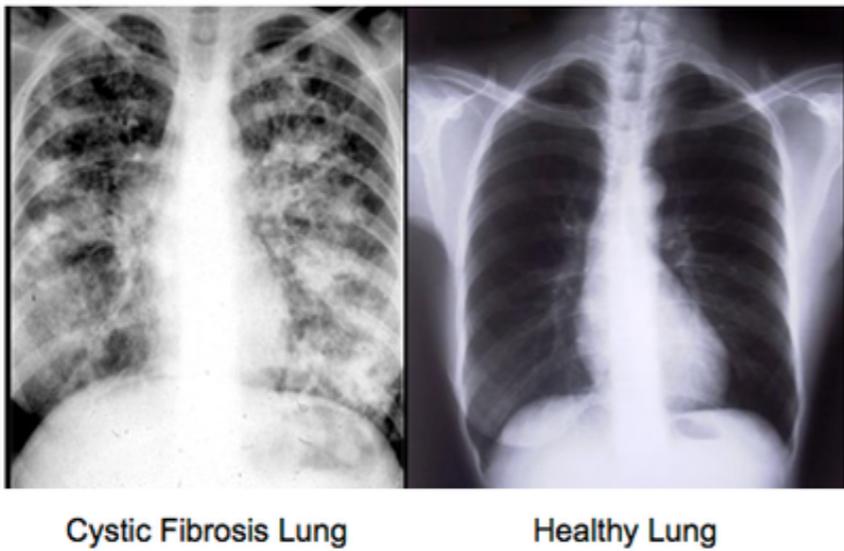
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<i>gld-1(0)</i>	mitotic cells
<i>glp-1(gf); lag-2(0)</i>	mitotic cells
<i>glp-1(gf); fbf-1(0)</i>	meiotic cells
<i>glp-1(0); gld-1(0)</i>	mitotic cells
<i>fbf-1(0); gld-1(0)</i>	mitotic cells
<i>lag-2(0); fbf-1(0)</i>	meiotic cells
<i>lag-2(0); gld-1(0)</i>	mitotic cells

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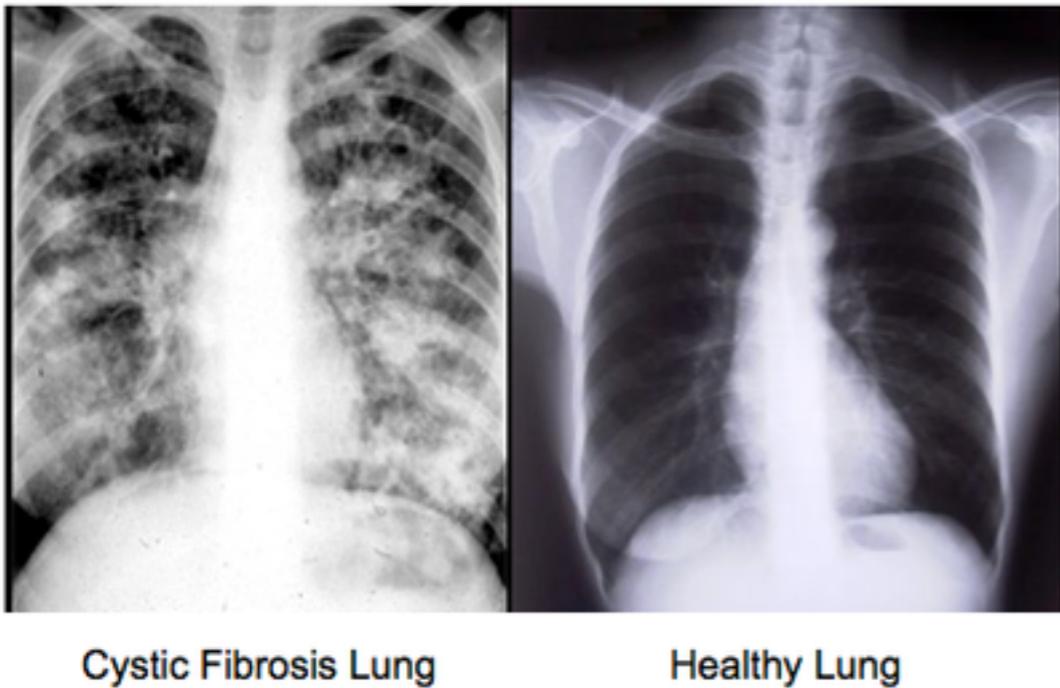
***lag-2* → *glp-1* → *fbf-1* → *gld-1* → GSC prolif.**

What about cystic fibrosis and today's topic?

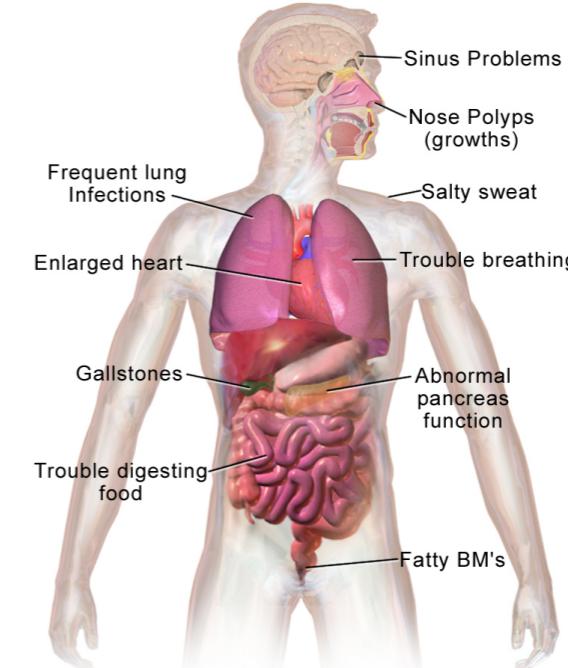


1. Autosomal recessive disorder
2. Not caused by chromosomal aberrations or meiotic NDJ
3. Mapped to chromosome 7
4. Mutations in CF gene are null or hypomorphs
5. Compound heterozygosity (failure to complement) is common

What about cystic fibrosis and today's topic?

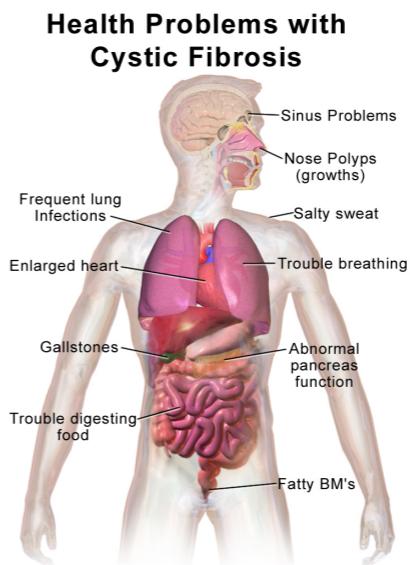
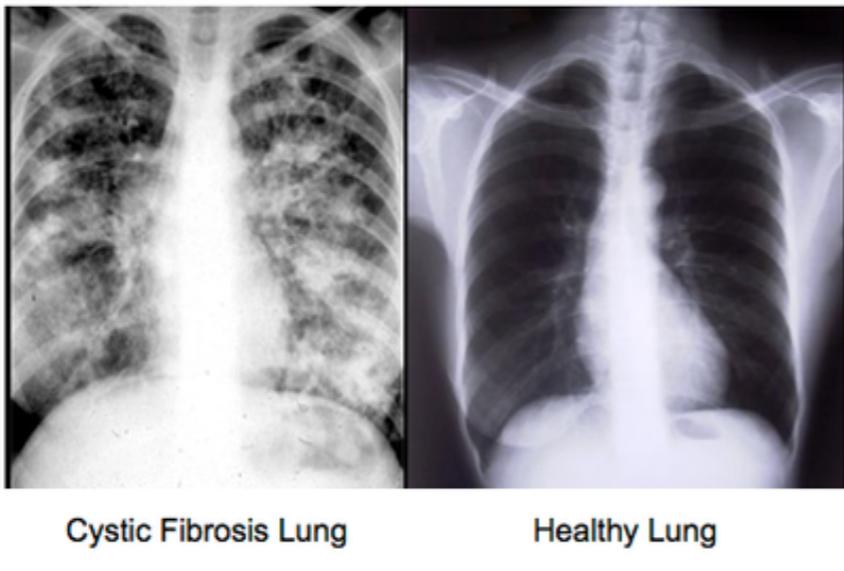


Health Problems with Cystic Fibrosis

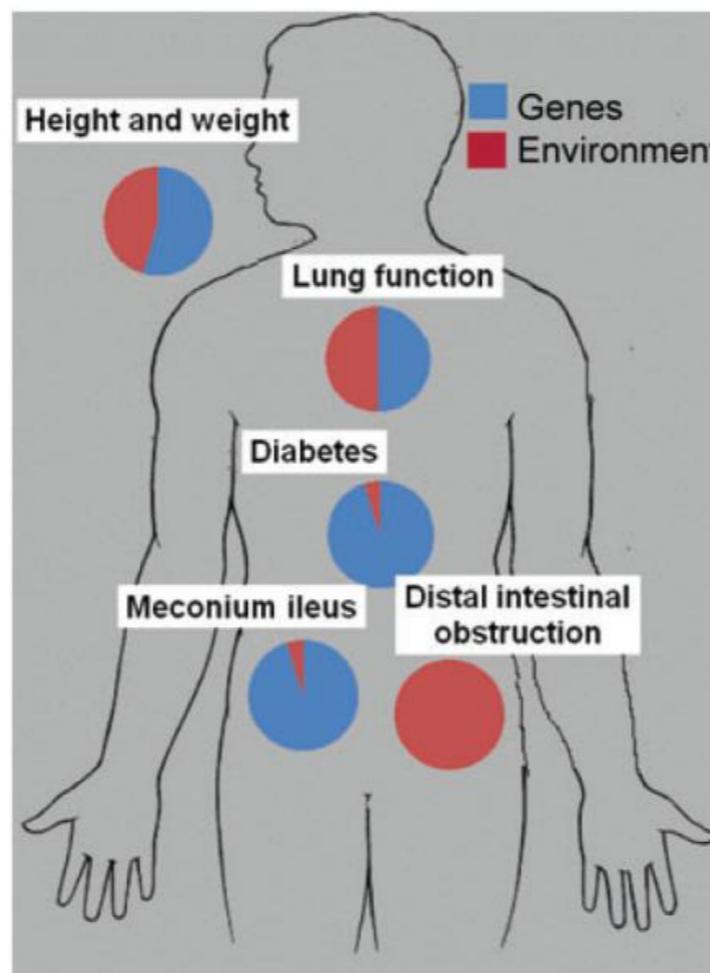


What data do we need to understand genetic interactions with the CF disease phenotype?

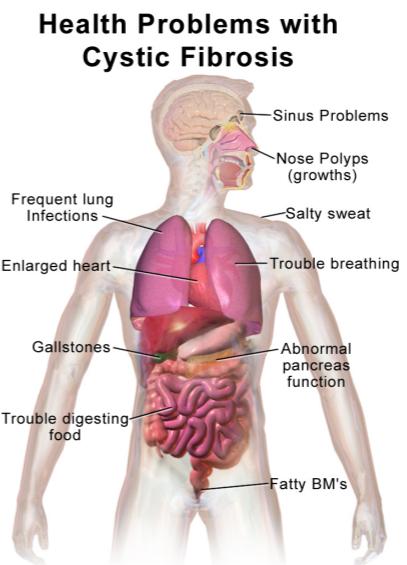
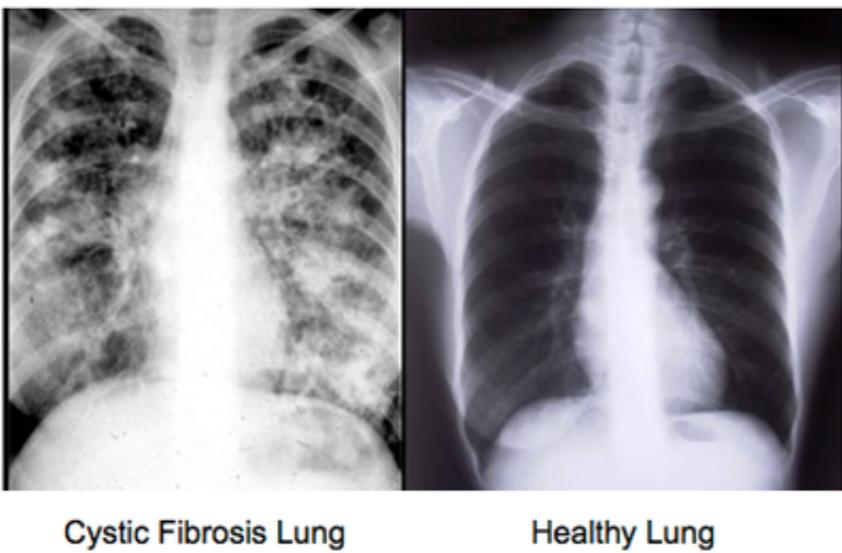
What about cystic fibrosis and today's topic?



Genetic modifiers are enhancers of CF



What about cystic fibrosis and today's topic?



Gene	Pulmonary Function (FEV ₁)	<i>P. aeruginosa</i>	Intestinal Obstruction	Diabetes	Liver Disease
		Acquisition/Colonization			
<i>ADIPOR2</i>			Possible effect ⁸⁷		
<i>EDNRA</i>	Probable effect ⁷⁹				
<i>IFRD1</i>	Possible effect ⁷⁴				
<i>IL8</i>	Possible effect ⁷⁵				
<i>MBL2</i>	Probable effect ^{55, 62, 64, 70, 94}	Probable effect ^{60, 70, 94}			
<i>MSRA</i>			Probable effect ⁸⁶		
<i>SERPINA1</i>	No effect ^{57, 95, 98}	Likely no effect ^{97, 99, 100}		Possible effect ⁴⁹	
<i>TCF7L2</i>				Probable effect ³⁷	
<i>TGFB1</i>	Probable effect ^{57, 59, 69, 70, 73, 78}	No effect ^{70, 72, 73, 78, 101}		Likely no effect ^{49, 101}	

Probable effect: Association observed in ≥ 3 independent populations with ≥ 1000 participants in aggregate.

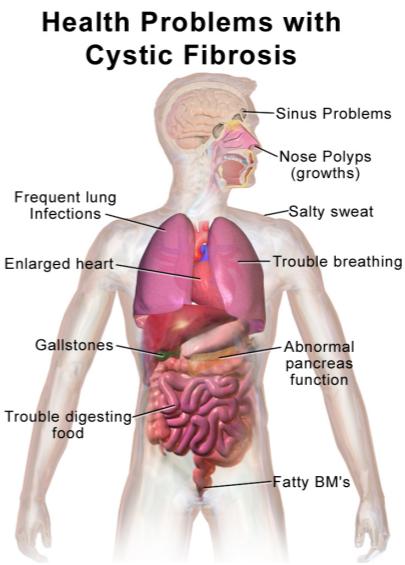
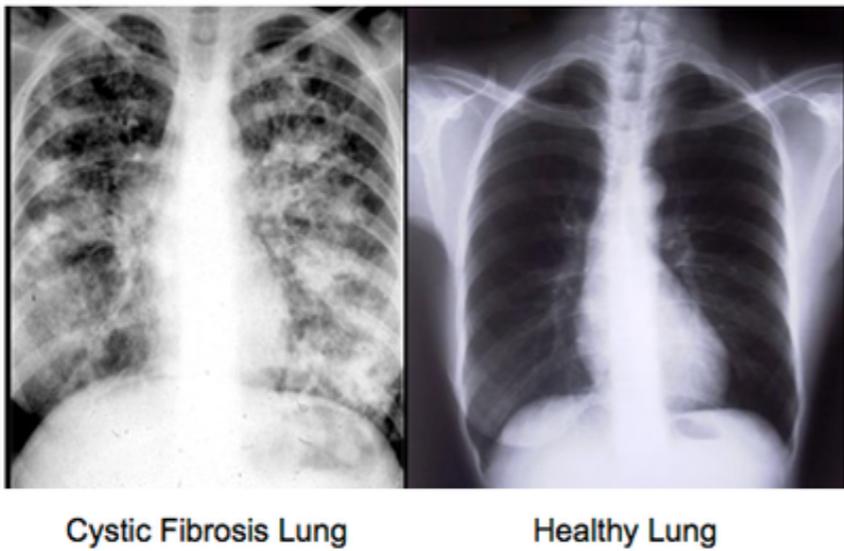
Possible effect: Association observed in ≥ 2 independent populations with ≥ 500 participants in aggregate.

Likely no effect: No association observed in ≥ 2 independent populations with ≥ 500 participants in aggregate.

No effect: No association observed in ≥ 3 more independent populations with ≥ 1000 participants in aggregate.

N.B.: Some studies include replication populations, which are treated as separate independent populations.

What about cystic fibrosis and today's topic?



1. Autosomal recessive disorder
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6. Enhancers of CF disease phenotype are known