Exercise in forward genetics part I

Regulation of lifespan in the Nematode *C. elegans*

The Nematode *C. elegans* is an excellent model to identify and study genes determining the lifespan of animals. There exist many different *C. elegans* strains, which are either short lived with an average lifespan of only 10 days or long lived with an average lifespan of up to 30 days.

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1. You want to use these long and short lived strains to identify *quantitative trait loci* (QTLs) regulating the lifespan of *C. elegans*. Describe in a few sentences the experiments you need to perform to find QTLs determining the lifespan of *C. elegans* and to identify the *individual genes* responsible for the differences in lifespan.
2. Through this QTL analysis, you have identified five genes called *age-1* to *age-5*. Using reverse genetics, you have generated *loss-of-function* ("knock-out") mutations in each of the five *age* genes in the standard *C. elegans* Bristol background. You have measured the lifespan of the individual *age* mutants. These are the results:

genotype	**average lifespan** **± standard deviation (days)**
+/+ (wild-type *C.e.* Bristol)	18 ±2
age- 1(lf)/+	17 ±3
age- 1(lf)/age- 1(lf)	29 ±3
age- 2(lf)/+	19 ±2
age- 2(lf)/age- 2(lf)	27 ±5
age- 3(lf)/+	18 ±3
age- 3(lf)/age-3(lf)	12 ±3
age- 4(lf)/+	18 ±2
age- 4(lf)/age- 4(lf)	27 ±4
age- 5(lf)/+	20 ±3
age- 5(lf)/age- 5(lf)	11 ±2

(a) Which of these genes five inhibit ageing and which genes promote ageing?

You then perform epistasis analysis by measuring the lifespan of all the double mutant combinations. These are the results: (The numbers indicate the average life span ±std.dev. of the respective double mutants.)

there are several solutions (3) to this ex. Presenting all solutions will give extra points.

gene1/gene2	*age-1(lf)/age-1(lf)*	*age-2(lf)/age-2(lf)*	*age-3(lf)/age-3(lf)*	*age-4(lf)/age-4(lf)*	*age-5(lf)/age-5(lf)*
age-1(lf)/age-1(lf)		28±4	11±3	30±4	19±3
age-2(lf)/age-2(lf)	28±4		13±4	29±3	18±2
age-3(lf)/age-3(lf)	11±3	13±4		10±4	11±3
age-4(lf)/age-4(lf)	30±4	29±3	10±4		28±3
age-5(lf)/age-5(lf)	19±3	18±2	12±2	28±3	

(b) Draw a genetic model based the results of the epistasis analysis. Indicate the sign (positive or negative) of each genetic interaction (for example use $\rightarrow$ for positive and $\dashv$ for negative regulation). Be sure to include the outcome (aging) at the end of your pathway. Discuss the different scenarios that are possible based on the available data.

(c) Design a synthetic forward screen to identify additional genes (i.e. *age-6*, *age-7* etc) regulating the lifespan of *C. elegans*. What phenotype can you select for to identify new regulators of lifespan?

c) first: look for genes that are epistatic to age-3, since age-3 is epistatic to all known genes in the exercise. a double mutant is long-lived.