Homework 6

1. Please perform a statistical analysis of the data provided for your group

2. Make conclusions if these differences can be considered significant or not

3. Suggest optimisations if any are required

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D. melanogaster

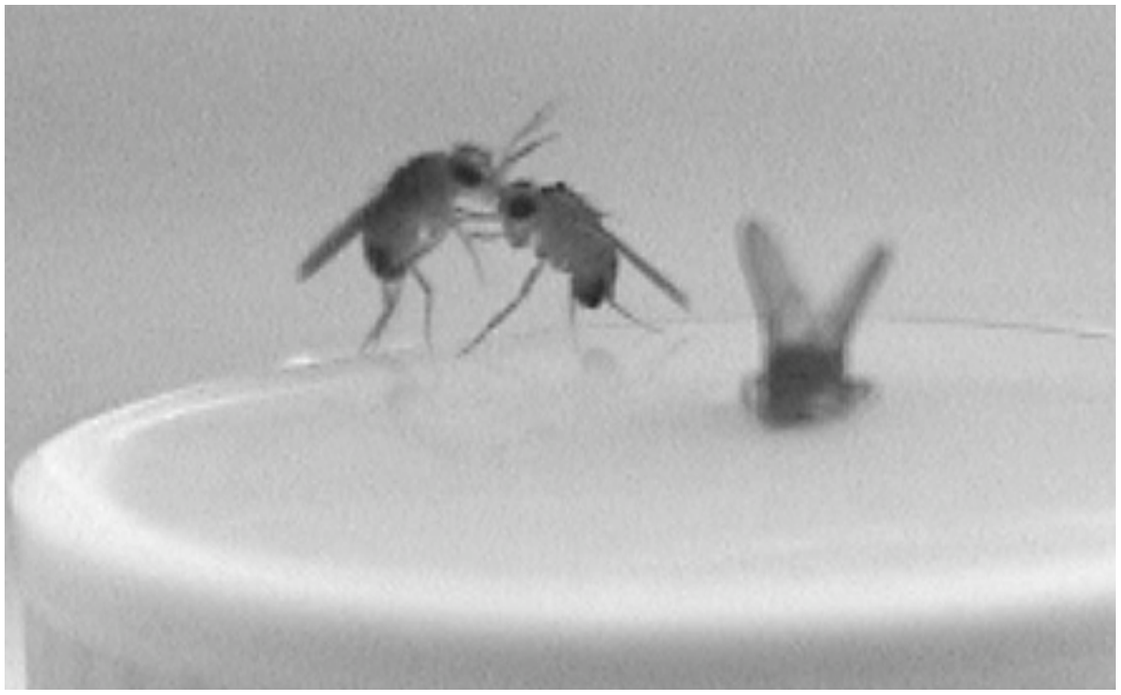
S. lycopersicum

NB!! None of the following experiments are recommended for scientific or any other purposes!

Group 1

Continuing your research on Drosophila melanogaster

You want to know if aggressive flies produce more saliva than non aggressive flies



1. You found a non aggressive mutant flies who supposedly have much better manners than the wild type flies

2. You established an excellent protocol to piss off the flies a little bit and to piss them off a lot

3. You are detecting the salivating efficiency by measuring expression levels of Lysozyme X gene

4. You want to see if the mutants lose their manners under pressure and how much you need to annoy them to bring them to the wild type condition

Group 1

1. You found reference genes during the previous seminar
2. Ran the experiment using 3 flies of corresponding genotype for each treatment (3 biological replicates)
3. Found M-value and selected 2 best out of your reference candidates
4. Performed a ΔΔCt calculation using Pfaffl method

And here are your normalised expression data

Genotype	How much are they pissed off	Target (Gene)	Biological replicate #	Normalized Expression
wild type	not at all	Lysozyme X	1	0,99468
wild type	not at all	Lysozyme X	2	1
wild type	not at all	Lysozyme X	3	1,28214
wild type	a little bit	Lysozyme X	1	1,2409
wild type	a little bit	Lysozyme X	2	2,0399
wild type	a little bit	Lysozyme X	3	1,5992
wild type	a lot	Lysozyme X	1	4,3716
wild type	a lot	Lysozyme X	2	4,9418
wild type	a lot	Lysozyme X	3	4,8015
extra-polite mutant	not at all	Lysozyme X	1	0,0999
extra-polite mutant	not at all	Lysozyme X	2	0,03752
extra-polite mutant	not at all	Lysozyme X	3	0,03719
extra-polite mutant	a little bit	Lysozyme X	1	0,71059
extra-polite mutant	a little bit	Lysozyme X	2	0,541
extra-polite mutant	a little bit	Lysozyme X	3	0,80314
extra-polite mutant	a lot	Lysozyme X	1	2,0025
extra-polite mutant	a lot	Lysozyme X	2	1,2063
extra-polite mutant	a lot	Lysozyme X	3	1,8471

Group 2

Continuing your research on Drosophila melanogaster

You want to know if ɣ-irradiated flies develop strong biceps

1. You noticed that after a mild dose of ɣ-irradtiation flies act very tough and develop a taste for body-building activities

2. You established an excellent protocol to irradiate the flies just a little bit and a bit more

3. You are going to detect the muscle growth by measuring expression levels of tropomyosin and actin genes

4. You want to see if both genes undergo the same changes in expression upon the two treatments you learned to do. And which of the treatments has the best effect on what gene

Group 2

1. You found reference genes during the previous seminar
2. Ran the experiment using 12 flies of corresponding genotype for each treatment (4 per a biological replicate)
3. Found M-stability value and selected 2 best out of your reference candidates
4. Performed a ΔΔCt calculation using Pfaffl method

And here are your normalised expression data

Irradiation level	Target (Gene)	Biological replicate #	Normalized Expression
None	tropomyosin	1	0,99468
None	tropomyosin	2	1
None	tropomyosin	3	1,28214
a little bit	tropomyosin	1	1,2409
a little bit	tropomyosin	2	2,0399
a little bit	tropomyosin	3	1,5992
a little bit more	tropomyosin	1	4,3716
a little bit more	tropomyosin	2	4,9418
a little bit more	tropomyosin	3	4,8015
None	actin	1	0,0999
None	actin	2	0,03752
None	actin	3	0,03719
a little bit	actin	1	0,71059
a little bit	actin	2	0,541
a little bit	actin	3	0,80314
a little bit more	actin	1	2,0025
a little bit more	actin	2	1,2063
a little bit more	actin	3	1,8471

Group 3

Continuing the research for your underground tomato fight club in Vietnam

You are interested in tomatoes which get sweeter after a fight

1. You noticed that after a proper fight some cultivars of tomato become extremely sweet and you want to figure out how it works and then sell the know-how to Heinz

2. You organised six tomato battles: three for professionals and three for beginners

3. For both fights you used two different cultivars: Money Maker and ToughOne

4. You are measuring expression levels of aldolase B gene in both cultivars before and after the fight

5. You want to see if there is a significant change in the aldolase expression in any cultivar and what kind of fight is stimulates it best

Group 3

1. You found reference genes during the previous seminar
2. Ran the experiment using 12 tons of each cultivar for each battle
3. Found M-stability value and selected 2 best out of your reference candidates
4. Performed a ΔΔCt calculation using Pfaffl method

And here are your normalised expression data

Cultivar	Battle	Target (Gene)	Normalized Expression before the battle	Normalized Expression after the battle
MoneyMaker	battle 1 for profies	Aldolase	0,3268	0,4268
MoneyMaker	battle 2 for profies	Aldolase	0,4758	1
MoneyMaker	battle 3 for profies	Aldolase	0.2365	1,28214
MoneyMaker	battle 1 for beginners	Aldolase	0.4592	1.8912
MoneyMaker	battle 2 for beginners	Aldolase	0.2873	1.9321
MoneyMaker	battle 3 for beginners	Aldolase	0.39002	0.9931
ToughOne	battle 1 for profies	Aldolase	0.11934	3.08121
ToughOne	battle 2 for profies	Aldolase	0.7648	2.00321
ToughOne	battle 3 for profies	Aldolase	0.1327	1.8421
ToughOne	battle 1 for beginners	Aldolase	0.7642	3.2109
ToughOne	battle 2 for beginners	Aldolase	0.1773	0.9212
ToughOne	battle 3 for beginners	Aldolase	0.4094	0.74123

Group 4

Continuing the research for your tomato plantation

1. You noticed that upon a very annoying viroid PTSVd infection, production of flavonoids in tomatoes goes up and they taste terrible, but can be stored forever. You have a brilliant marketing idea to rebrand the infected tomatoes as antioxidant enriched veggies and sell them in ICA. But you need a scientific proof for it.

2. You are growing two cultivars MoneyMaker and Rutgers on your plantation. At least half of your plantation is heavily infected, one forth is a little bit infected and some plants are still resisting the epidemic

3. You are going to detect the flavonoid production by measuring expression levels of 3-O-gluosyltransferase

4. You want to find out which cultivar has higher expression of a flavonoid 3 O-glucosyltransferase gene and what level of infection stimulates it the best.

Group 4

1. You found reference genes during the previous seminar

2. Ran the experiment using 12 fruits from 3 different infected plants as one biological replicate and 1 fruit from 2 non-infected plants as one biological replicate

3. Found M-stability value and selected 2 best out of your reference candidates

4. Performed a ΔΔCt calculation using Pfaffl method

And here are your normalised expression data

Cultivar	Biological replicate	PTSVd Infection level	Normalized Expression of Glucosyltransferase
MoneyMaker	1	None	0,3268
MoneyMaker	2	None	0,4758
MoneyMaker	3	None	0.2365
MoneyMaker	1	a little bit	0,4268
MoneyMaker	2	a little bit	1
MoneyMaker	3	a little bit	1,28214
MoneyMaker	1	a lot	1.8912
MoneyMaker	2	a lot	1.9321
MoneyMaker	3	a lot	0.9931
Rutgers	1	None	0.4592
Rutgers	2	None	0.2873
Rutgers	3	None	0.39002
Rutgers	1	a little bit	3.08121
Rutgers	2	a little bit	2.00321
Rutgers	3	a little bit	1.8421
Rutgers	1	a lot	3.2109
Rutgers	2	a lot	0.9212
Rutgers	3	a lot	0.74123