

Experiment Plans Version #2: Ashwini Suriyaprakash

Rapamycin amount should be same when it is alone and when it is in cocktail

Genotype	No tumor (Controls)
EGT (no tumor w/ MRP)	<u>Experiment 1</u> : With no chemo drugs
UAS-EGT x UAS-RNAi-MRP (no tumor w/o MRP)	<u>Experiment 2</u> : With no chemo drugs

Genotype	5-day tumor
<p>UAS-RAF:EGT (tumor stem cell flies w/ MRP) Different reds = different batches</p>	<p><u>Experiment 3</u>: With no drugs <u>Experiment 4</u>: With drug rapamycin <u>Experiment 5</u>: With drug cocktail (rapamycin and metformin in 1:10 ratio)</p>
<p>UAS-RAF:EGT x UAS-RNAi-MRP (tumor stem cell flies w/o MRP) Different greens = different batches</p>	<p><u>Experiment 6</u>: With no drugs <u>Experiment 7</u>: With drug rapamycin <u>Experiment 8</u>: With drug cocktail (rapamycin and metformin in 1:10 ratio)</p>

Week 4:

Wednesday, July 24

- Put UAS-RAF:EGT x UAS-RNAi-MRP at 29C (for Experiments 6-8, Batch 1)
- Put UAS-EGT x UAS-RNAi-MRP at 29C (for Experiment 2, Batch 1)

Thursday, July 25

- Label flies at 29C with set and date (organize them with drug group, etc.)
 - 3 labeled vials: no drug, Drug A, Drug B (UAS-RAF:EGT x UAS-RNAi-MRP at 29C (for Experiments 6-8, Batch 1))
 - 1 labeled vial: UAS-EGT x UAS-RNAi-MRP at 29C (for Experiment 2, Batch 2)
- Put UAS-RAF:EGT flies at 29C (for Experiments 3-5, Batch 1)
- Flip F1 flies from UAS-RAF:EGT x UAS-RNAi-MRP cross

Friday, July 26

- Label flies at 29C with set and date (organize them with drug group, etc.)
 - 3 labeled vials: no drug, Drug A, Drug B (UAS-RAF:EGT flies at 29C (for Experiments 3-5, Batch 1))
- Put UAS-RAF:EGT x UAS-RNAi-MRP at 29C (for Experiments 6-8, Batch 2)
- Flip F1 flies from UAS-RAF:EGT x UAS-RNAi-MRP cross
- Request drugs rapamycin and rapamycin:metformin (1:10) cocktail for Saturday, Sunday, and Monday
- Request RAF EGT flies to be put at 29C on Saturday

Saturday, July 27

- Add drugs to food for UAS-RAF:EGT x UAS-RNAi-MRP (for Experiments 7 and 8, Batch 1)
- Put UAS-RAF:EGT flies at 29C (for Experiments 3-5, Batch 2)

Sunday, July 28

- Add drugs to food for UAS-RAF:EGT flies (for Experiments 4 and 5, Batch 1)

Week 5:

Monday, July 29 (seeing 5-day tumor without MRP)

- Add drugs to food for UAS-RAF:EGT x UAS-RNAi-MRP (for Experiments 6-8, Batch 2)
- Dissect UAS-RAF:EGT x UAS-RNAi-MRP (for Experiments 6-8, Batch 1): no drugs, Drug A, Drug B
- Flip F1 flies from UAS-RAF:EGT x UAS-RNAi-MRP cross
- Request drugs rapamycin and rapamycin:metformin for Tuesday

Tuesday, July 30 (seeing 5-day tumor with MRP)

- Add drugs to food for UAS-RAF:EGT flies (for Experiments 3-5, Batch 2)
- Dissect UAS-RAF:EGT flies (for Experiments 3-5, Batch 1): no drugs, Drug A, and Drug B
- Flip F1 flies from UAS-RAF:EGT x UAS-RNAi-MRP cross

Wednesday, July 31 (seeing 5-day tumor without MRP)

- Dissect UAS-RAF:EGT x UAS-RNAi-MRP at 29C (for Experiments 6-8, Batch 2): no drugs, Drug A, Drug B
- Flip F1 flies from UAS-RAF:EGT x UAS-RNAi-MRP cross
- Put more flies at 29C (if dissections don't go well)

Thursday, August 1 (seeing 5-day tumor with MRP)

- Dissect UAS-RAF:EGT flies (for Experiments 3-5, Batch 2): no drugs, Drug A, and Drug B
- Request EGT flies

Friday, August 2

- Dissect UAS-EGT x UAS-RNAi-MRP and image on mitt romney + confocal (for Experiment 2, Batch 1)
- Dissect EGT flies and image