mel-28 Genetic Interactions mutants in *C. elegans* worms

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Introduction

This experiment was performed to study the effects of disrupting different genes via RNA interference on wild-type and *mel-28* mutant *C. elegans*. RNA interference is the process of depleting RNA to stop the production of a protein from a certain gene.

- Central Dogma:

  DNA → RNA → Protein

Introduction Cont.

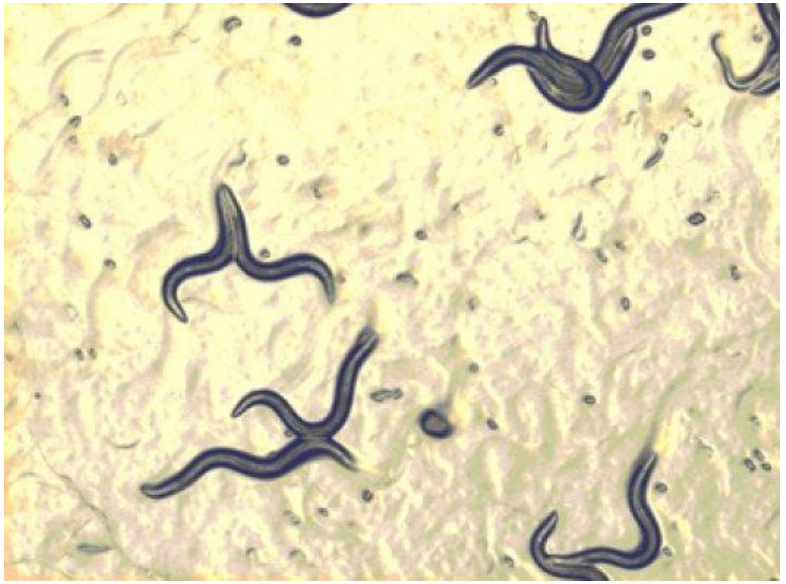
Why *C. elegans*?

- Small
- Cheap to raise
- Transparent
- Fast reproduction rate
- Large egg size

N2 (wild-type) and *mel-28* characteristics

N2 strain

- Appearance is normal
- Self fertilizes
- Lays eggs that hatch



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mel-28 mutants

- Appearance is normal
- Self fertilizes
- Lay eggs that don't hatch

Hypothesis/Predictions

Some of these genes interact with *mel-28* and redundantly contribute to similar processes to *mel-28*. If the hypothesis is true, then the RNAi depletion of these genes will cause a novel phenotype in *mel-28* mutants (and not the wild-type N2 animals).

Procedure

Monday: Three *mel-28* heterozygotes were moved to one plate, and wild-type worms (N2) were moved to another. These were left to lay eggs.

Tuesday: The parents were removed from plates.

Wednesday: Eggs were left to grow and reproduce.

Thursday: Five *mel-28* mutants and five N2 worms were chosen from each plate and individualized.

Friday: Observations were recorded regarding the appearance of the worm and the number of offspring.

Results

- Each plate was scored for number of eggs and larvae:

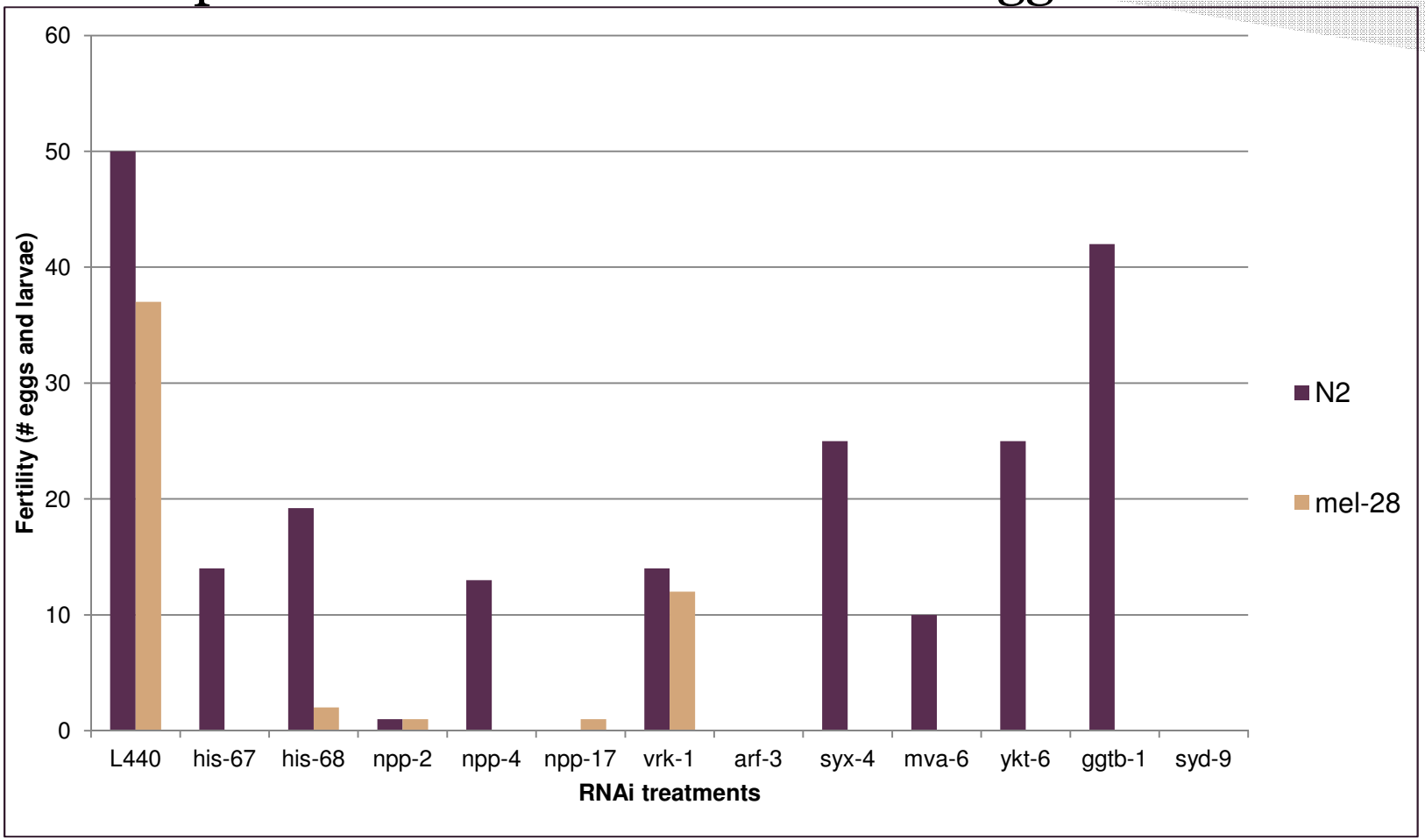


Figure 1: Fertility of N2 and mel-28 worms when treated with different RNAi treatment.

Results Cont.

- Each plate was also scored based on health of Mother.

Table 1: Health of Mother in N2 and mel-28 worms		
RNAi	N2	mel-28
L440	5/5 healthy	3/3 sick/dead
his-67	5/5 healthy	4/4 okay
his-68	5/5 alive	5/5 alive, 50% sick
npp-2	5/5 alive but sick	5/5 alive with pvl
npp-4	5/5 healthy	5/5 alive, 3/5 sick
npp-17	4/5 alive	5/5 dead, no tracks
vrk-1	4/5 alive	5/5 alive, 3/5 sick
arf-3	2/5 alive	3/5 alive
syx-4	5/5 alive but sick	5/5 alive and sick
mva-6	5/5 healthy	N/A (all
ykt-6	5/5 healthy	4/5 healthy, 1 died young
ggtb-1	3/5 alive, 2/5 bag of worms	N/A
syd-9	2/5 produced larvae	2/5 healthy, 3/5 sick/dead

Conclusion

We were then able to see if there is any *mel-28* genetic interactions with any of the listed genes.

Table 2: RNAi treatment and interpretation of genetic interactions		
Process the genes are involved	**RNAi**	**Genetic Interaction (Y or N)**
	L440	Negative control
	Mel-28	Positive control
Chromatin	His-67	Yes
	His-68	Yes
Nuclear envelope	Npp-2	Yes
	Npp-4	Yes
	Npp-17	Yes
	Vrk-1	No
Vesicle Fusion	Arf-3	No
	Syx-4	Yes
	Mva-6	Yes
	Ykt-6	Yes
	Ggbt-1	Yes
	Syd-9	No

***Refer to Figure 1 for evidence of genetic interaction**

Conclusion Cont.

In conclusion, the hypothesis was supported by the results of the experiment. The RNA interference affected the fertility of the mel-28 worms. Most of the mel-28 worms showed signs of sickness, death, or sterility. On the other hand the N2 worms were more fertile and healthier.

Embryogenesis of *mel-28* and N2 worms

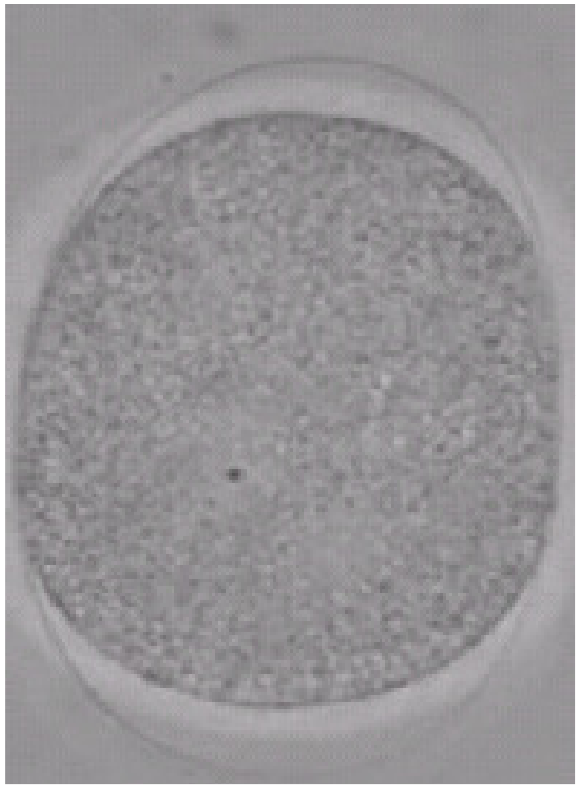
- We obtained images of embryo development in wild-type (N2) worms and in *mel-28* mutants to see the difference.
- After taking pictures of development every 30 seconds we were able to see clear differences between the two strains of worms.

Embryogenesis of Mel-28 and N2 worms

1 Cell Stage

N2

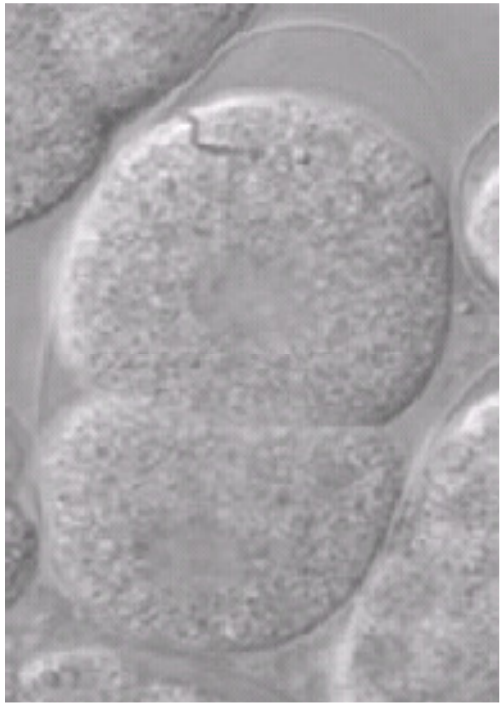
mel-28



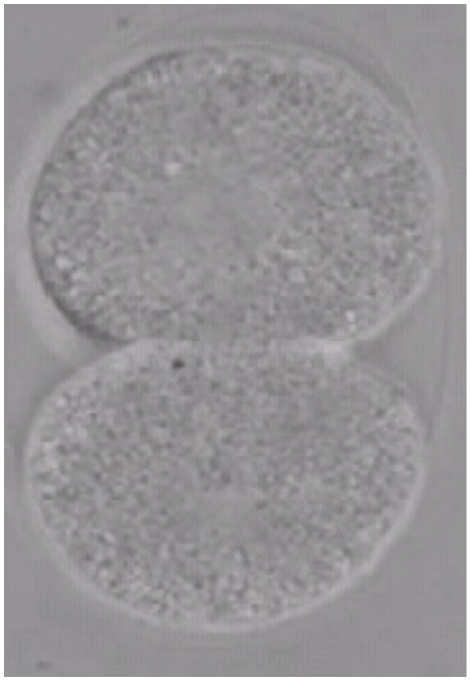
Embryogenesis of Mel-28 and N2 worms

2 Cell Stage

N2



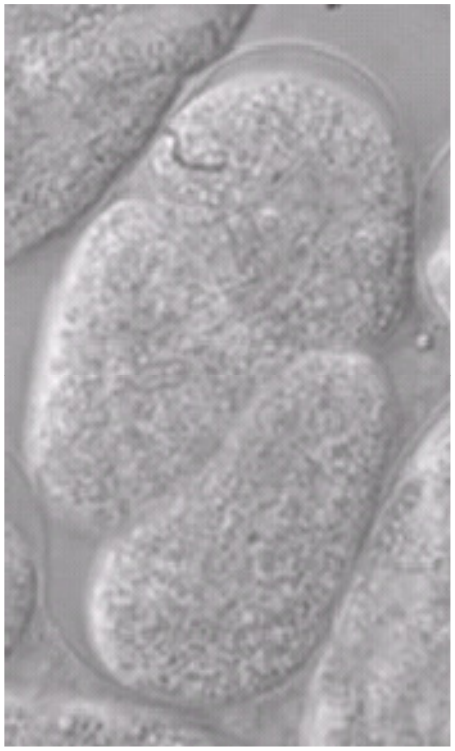
mel-28



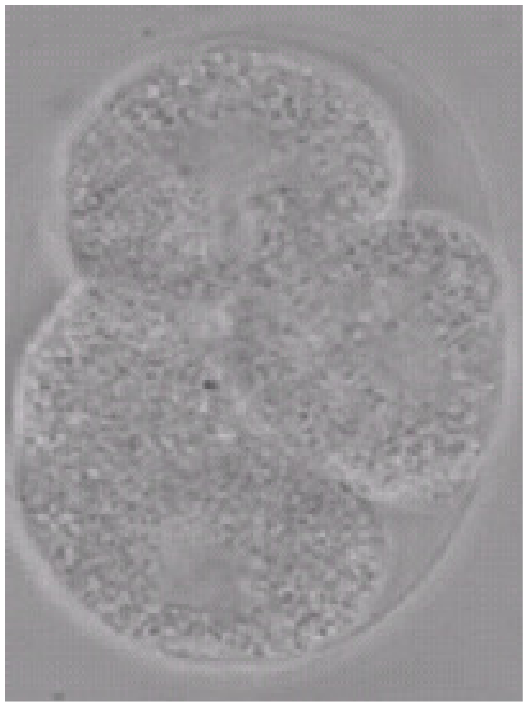
Embryogenesis of Mel-28 and N2 worms

3 Cell Stage

N2



mel-28



Embryogenesis of Mel-28 and N2 worms

4 Cell Stage

N2

mel-28

Acknowledgements

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