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### Longevity and Aging Mediated by FOXO and the Gene Methuselah

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# Longevity and aging mediated by FOXO and the gene methuselah

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## Background

- Forkhead box (FOXO) proteins have been found to be involved in many processes involving longevity of cells (Jiramongkol & Lam, 2020).
- Transcriptional regulation of growth and longevity has been exhibited and studied with the invertebrate *Drosophila melanogaster* (Martins, Lithgow, & Link 2016).
- The gene, *methuselah* within the genome of *D. Melanogaster*, has been known to be involved in embryo and larvae development (Arujo et. al 2020).
- The gene is a G protein-coupled receptor that can be regulated by FOXO proteins (Araujo et. al 2020).
- This present study is addressing the effects of FOXO gene knockout on longevity and cancer cell proliferation

Table 1. Primers chosen for FOXO expression experiments

Oligo Sequence description	Primer
ACCAAAGTGGGCCAACGTCTTTC	dm methuselah F
CACTGTTGTTACCTCCTCACCT	dm methuselah R

## Thesis

- Hypothesis: If FOXO is removed from the regulation process, then the methuselah gene will increase in transcription thus longevity and stress resistance will increase.
- Purpose: Try to knock down FOXO in which will allow gene expression, thus foresee an increase in longevity and stress resistance.

## Methods

- Step 1: First, a sequence of the gene methuselah was isolated, and then two primers were selected.
- Step 2: The cells went through the process of transformation and plasmid formation.
- Step 3: The cells went through a series of RNA knockdown steps that included in vitro transcription, annealing dsRNA, and prep for cells.

## Results

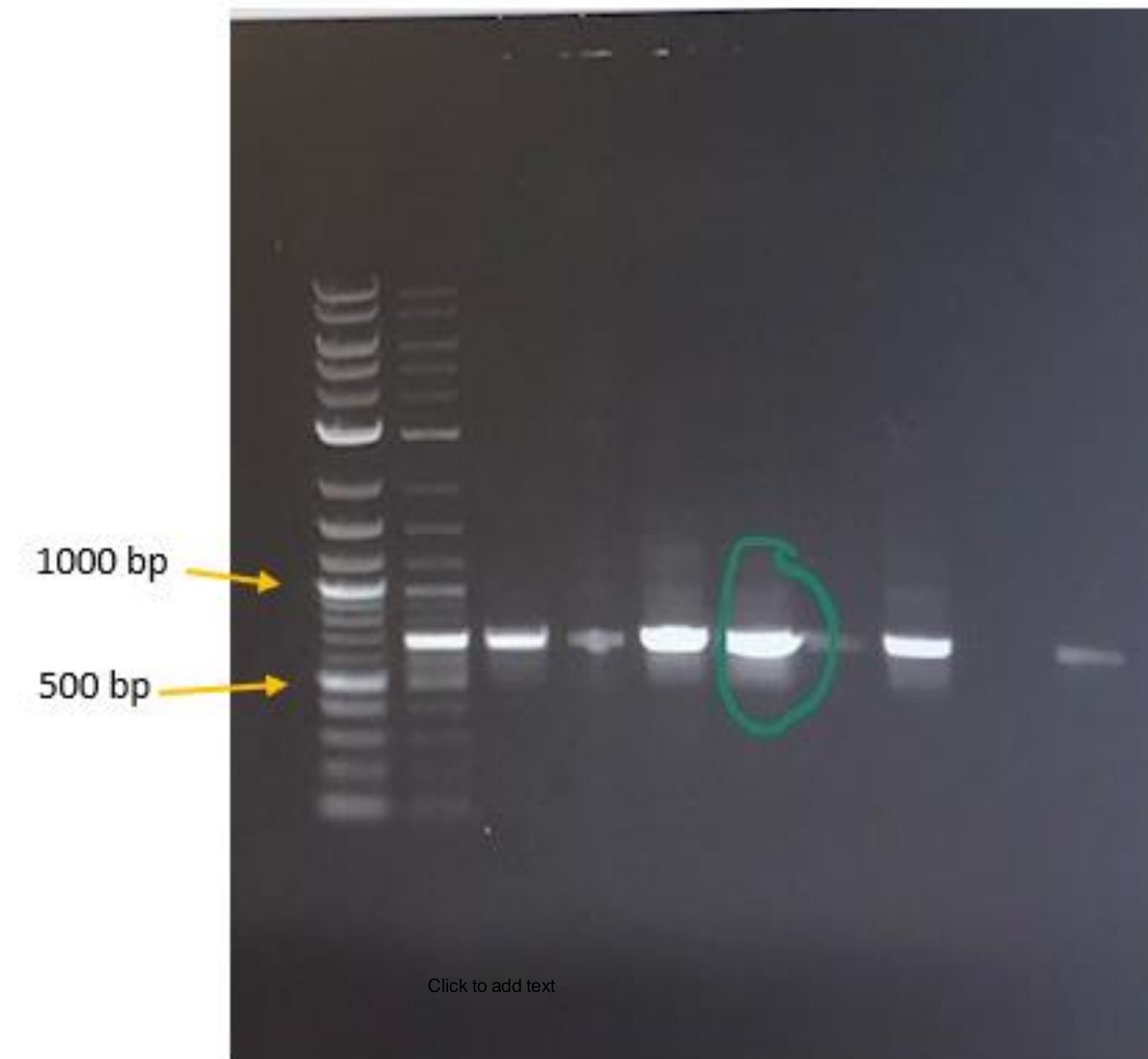


Figure 1: Gel Electrophoresis under UV light shows the DNA sample against a DNA ladder of 1-Kb increments. Our DNA sample, circled in green, sits at 700 base pairs

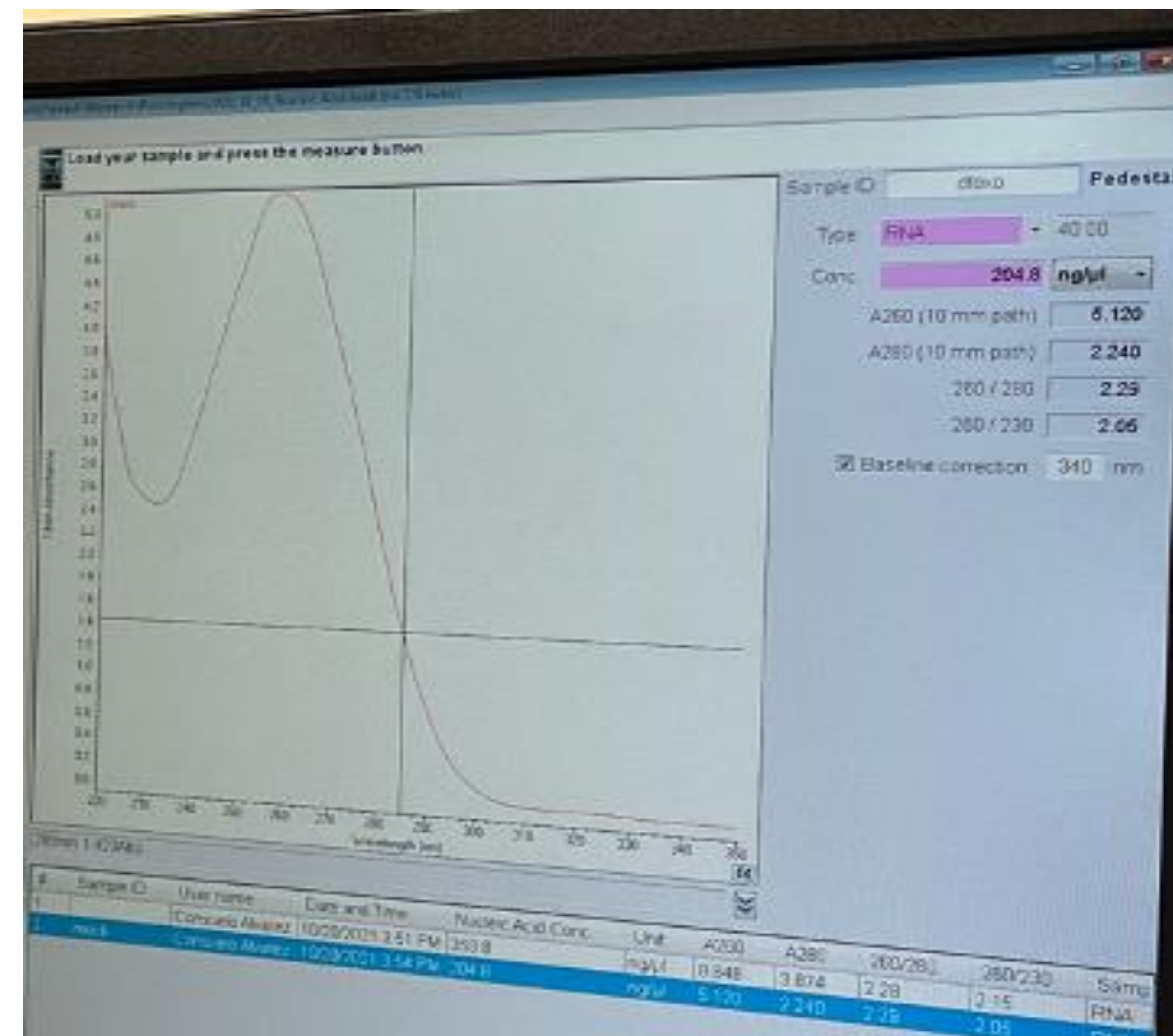


Figure 2: dFOXO RNA concentration shown via spec to be 204.8 ng/μl. A peak is shown at A260 which is shown to be 5.12 and a dip at A280 shown to be 2.24

## Results



Figure 3: The relative gene expression of the target gene and dFOXO gene as a fraction of the mock. Mock gene expression for both was one. Gene expression of the Target gene with dFOXO dsRNA was 1.66 and 0.47 for the dFOXO gene.

## Conclusions

- Based on the data, there was an increase in target gene with the FOXO being knockdown. The Methuselah gene codes for family B G protein-coupled receptor(GPCR), which is known to be associated with longevity and stress Resistance. With the increase in transcription, we can assume that the drosophila cells will live longer than their normal life.

## Future Directions

- In future research we can see if the drosophila cells will have an increase in cell growth/ tumors with FOXO being knock down because of the increase longevity.

## References

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