University of CINCINNATI

The Affect of Mutations Within the RGL2 Gene has on the Development of the Heart

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Background Information

- RASopathesis are developmental disorders that occur due to germline mutations in the Ras/MAPK pathway.
- This pathway is responsible for the development of organisms from the very beginning of cell development as well as gene construction.
- The RGL2 gene is a gene responsible for many functions within development and might include aspects of the heart.
- The purpose of this research is to find how the RGL2 gene is expressed and how mutations in these expressions can affect the heart.

Research Question and Hypothesis

- RGL2 has an impact on the development of the human heart.
- Will a mutation in RGL2 affect the heart?

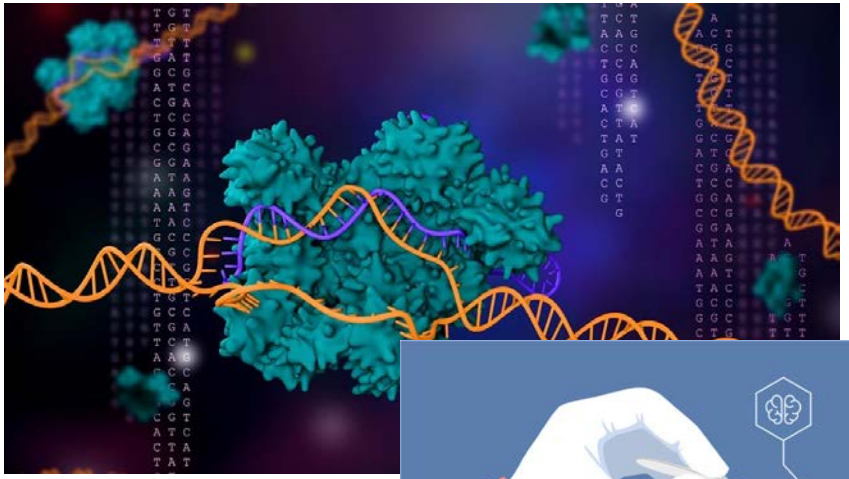
Lets first talk about our lovely helpers!

Zebrafish

- Share 70% of genes with humans
- 2 chambered heart
- Perfect model organisms
  - Short lifespan
  - Lay many offspring
  - Grow very fast
  - Embryos are transparent

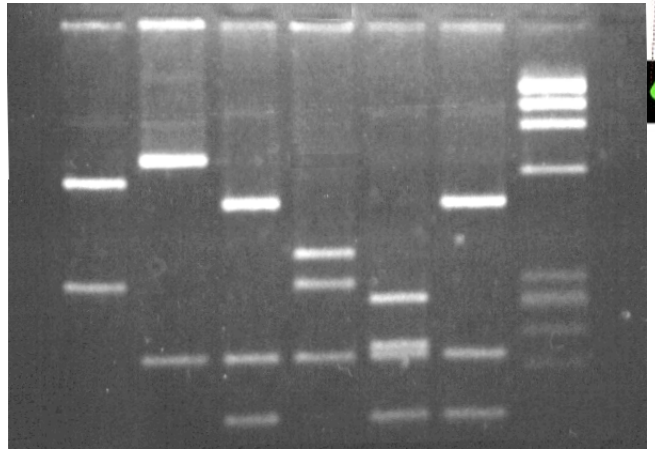
Methodology

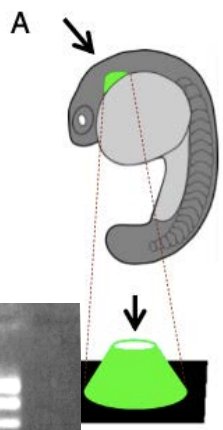
- Step 1
- Use CRISPR to create RGL2 mutated fish
  - This allows us to observe RGL2 by viewing the phenotypes (physical attributes) developed by the mutation.

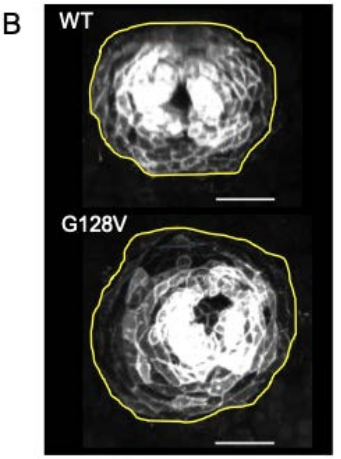


Methodology

- Step 2
- Understand the expression of the RGL2 gene.
  - By viewing the phenotypes caused by mutating the RGL2 gene we can compare these to healthy fish containing the gene to view the effect it has on heart development.
  - Use Gel electrophoresis and DNA prepping to view makeup and function.

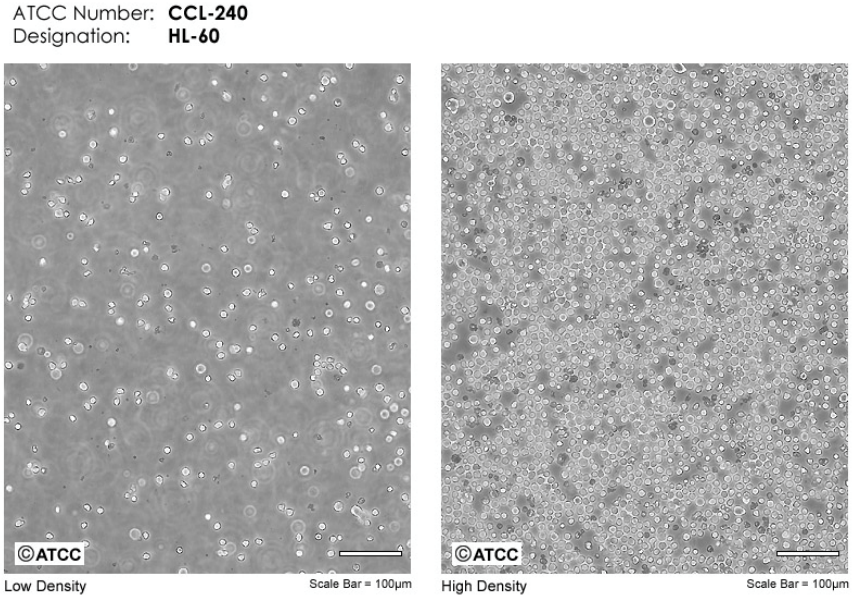






Methodology

- Step 3
- Observe RGL2 in human genes
  - Use an HL-60 cell line to observe the gene
  - Use CRISPR to isolate and manipulate RGL2
  - Observe the RGL2 expression



Results and Role

- Some results showed that RGL2 mutant fish showed a lack in development of some areas of the heart as well as thinner heart walls.
- RGL2 also showed signs of interacting with genes pertinent to heart development in early zebrafish embryos.
- In using this information we hope to find a way to stop the cause of mutation within this gene and ways to fix it.
- My role: run ISH (In situ Hybridization), run cells lines, and view RGL2 expression

Thank You!