Can We Cure Galactosemia ?

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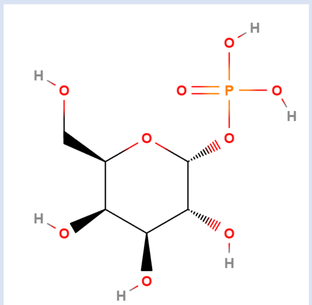
Galactosemia

- A rare genetic condition that causes a loss of function in the galactose-1-phosphate uridylyl transferase enzyme
- This causes a large build-up of galactose-1-phosphate
- Galactose-1-phosphate is produced by galactokinase 1 (GALK1)
- Therefore, a proposed treatment for galactosemia is the inhibition of GALK1



Galactose-1-phosphate molecular structure

Aims and Objectives

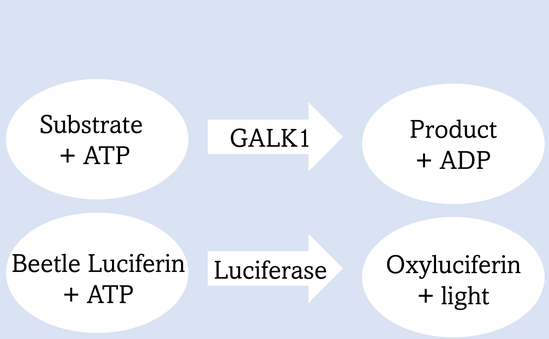
Aims:

- Attempt to show inhibition of GALK1
- Achieve a better understanding of research
- Learn new biochemical research techniques

Objectives:

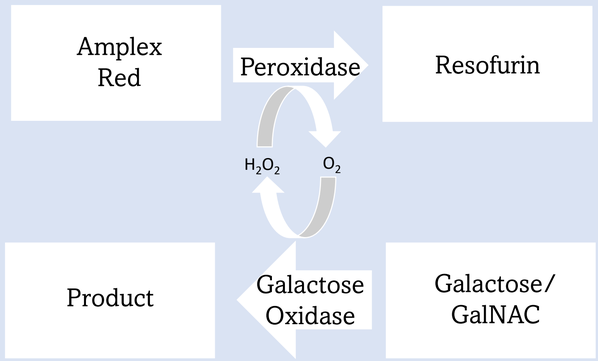
- Perform assays and experiments to achieve the half maximal inhibitory concentration (IC50)
- Engage and participate in activities associated with research
- Learn from the researchers and students who provide me information and advice

Primary Assays



Kinase Glo assay:

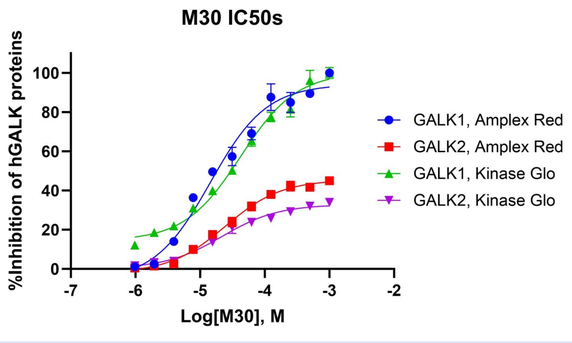
Remaining ATP from the initial reaction is used in producing oxyluciferin by the luciferase enzyme. Oxyluciferin produces luminescence.



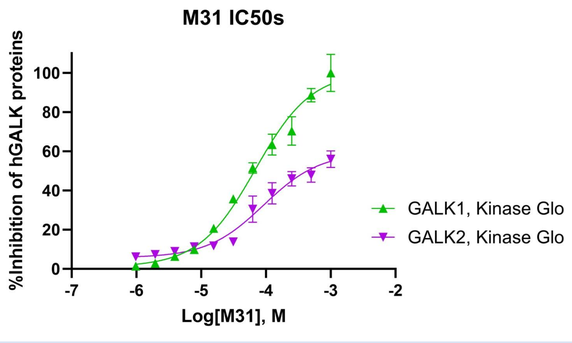
Amplex Red assay:

Firstly, the galactose remaining after the reaction is acted on by galactose oxidase. Then, the hydrogen peroxide (H_2O_2) is used to convert Amplex Red into Resorufin. Resorufin produces fluorescence.

Figures



IC50 – the half maximal inhibitory concentration.
IC50 from Amplex Red = 14 μM
IC50 from Kinase Glo = 42 μM.



IC50 from Kinase Glo = 69 μM.

Results

- Firstly, the IC50 values show the concentration of the inhibitor compound required to inhibit the GALK1 enzyme to 50%.
- Additionally, the lower the IC50 the more potent the inhibitor compound
- Therefore, from the figures, I can deduce that the M30 inhibitor compound is more potent than the M31
- However, due to an error I didn't achieve a result for the Amplex Red for both GALK1 and GALK2.
- This means that the IC50 for M31 could change dependent on the use of Amplex Red

Conclusion

- I can conclude, through the use of assays and experiments, that I have obtained values that indicate binding and inhibition of the GALK1 enzyme
- Additionally, I have engaged in weekly meetings related to scientific studies and experiments surrounding other areas of research
- I have learnt and experienced new techniques within a lab environment
- Finally, this experience has influenced me to contemplate a future within biochemical research

References

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3. Mackinnon SR, Krojer T, Foster WR, Diaz-Saez L, Tang M, Huber KVM, et al. Fragment Screening Reveals Starting Points for Rational Design of Galactokinase 1 Inhibitors to Treat Classic Galactosemia. ACS Chemical Biology. 2021 Mar 16;16(4):586–95

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