

**UNDERGRADUATE SUMMER VACATION SCHOLARSHIP AWARDS – FINAL SUMMARY REPORT FORM 2016/17**

*NB: This report will be posted on the Society's website therefore authors should NOT include sensitive material or data that they do not want disclosed at this time.*

**Name of student:**

Ms. Lindsey Watters

**Name of supervisor(s):**

Dr. Denis Barry

**Project Title: (no more than 220 characters)**

Examining the cellular and lineage significance of ketone supplementation on CNS neural precursors during development

**Project aims: (no more than 700 words)**

**1) Develop a stable cell culture model.**

Frozen SH-SY5Y cells will be purchased from Sigma and grown under normal cell culture conditions in T25 flasks with DMEM-F12 medium. These will then be divided into 3 groups; the control group, a test group, and a frozen group to be used in case of experimental cell loss. Cells will then be treated with ketone containing glucose free medium, glucose free medium and regular feeding medium to assess macroscopic changes in their morphology and cell cycle properties, over a period of 14 days. Cellular changes will be assessed by immunocytochemistry at 5 DIV, 7 DIV and 14 DIV.

**2) Assess the lineage potentials of cellular integrity and cell proliferation properties of SH-SY5Y cells treated with ketone medium and compare with no glucose controls and regular feeding medium**

Cytoskeletal changes will be assessed using immunocytochemistry involving the antibodies beta-III tubulin and actin, while cell division will be measured using the proliferation marker ki-67.

**3) Determine if KD during embryological development exerts an effect on cell lineage and phenotype through analysis of all data.**

Cell will be differentiated into TH expressing neurons and the effects of ketone treatment on this process will be analysed.

**Project Outcomes and Experience Gained by the Student (no more than 700 words)**

Outcomes:

1. Successful growth and stabilisation of the SHSY5Y cell line in the lab.
2. Successful growth of these cells in 1mM and 5mM beta-hydroxybutyric acid (a ketone body) cell culture medium in the absence of glucose inducing ketosis in SHSY5Y cells over a 10 day period.
3. Comparisons made with no glucose control and regular feeding medium.
4. Preliminary findings that cell number is decreased, cell proliferation is slowed and cell morphology is altered in cells treated with ketogenic medium.

The student plans to continue the research in collaboration with other researchers throughout the next several months as part of her final year project to further assess and quantify the varying cellular alterations between SH-SY5Y cells grown in normal medium versus ketogenic medium.

**Experience Gained by the Student:**

1. An in-depth knowledge of neurophysiology and neuronal cell biology.
2. Proficiency in cell culture and sterility techniques.
3. Microscopy.
4. Understanding of how a lab is managed, how to write protocols, and how to carry out a research project in an organised and efficient manner.

**Please state which Society Winter or Summer Meeting the student is intending to present his/her poster at:**

Winter Meeting 2017, Dundee, Scotland.

**Proposed Poster Submission Details (within 12 months of the completion of the project) for an AS Winter/ Summer Meeting – (no more than 300 words)**

The student plans to register and submit a poster for the 2017 Winter Meeting in Dundee. This will include an explanation of the work carried out thus far with images of cells in regular versus ketogenic medium as well as a brief summary of future directions for the research.

**Brief Resume of your Project's outcomes: (no more than 200-250 words).**

*The title of your project and a brief 200-250 word description of the proposed/completed project. The description should include sufficient detail to be of general interest to a broad readership including scientists and non-specialists. Please also try to include 1-2 graphical images (minimum 75dpi). NB: Authors should NOT include sensitive material or data that they do not want disclosed at this time.*

**PROJECT TITLE: Examining the cellular and lineage significance of ketone supplement on CNS precursors during development.**

The overall aim of this research, in combination with other experiments being carried out elsewhere, is to determine if a ketogenic diet affects pregnancy and/or foetal CNS development in cell culture models and mice.

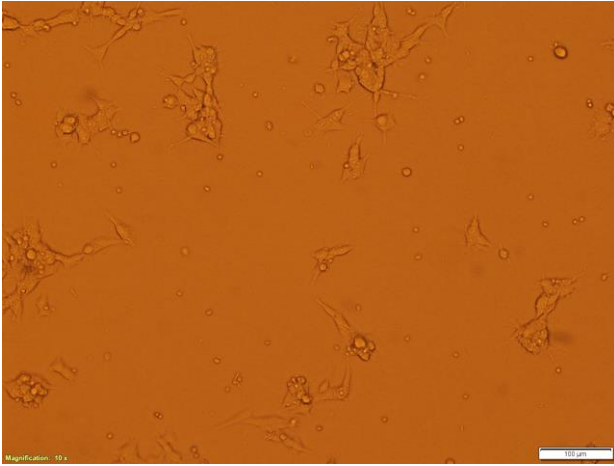
**Objectives:**

- 1) Develop a stable cell culture model.
- 2) Assess the lineage potentials of cellular integrity and cell proliferation properties of SH-SY5Y cells treated with ketone medium and compare with no glucose controls and regular feeding medium.
- 3) Determine if KD during embryological development exerts an effect on cell lineage and phenotype through analysis of all data.

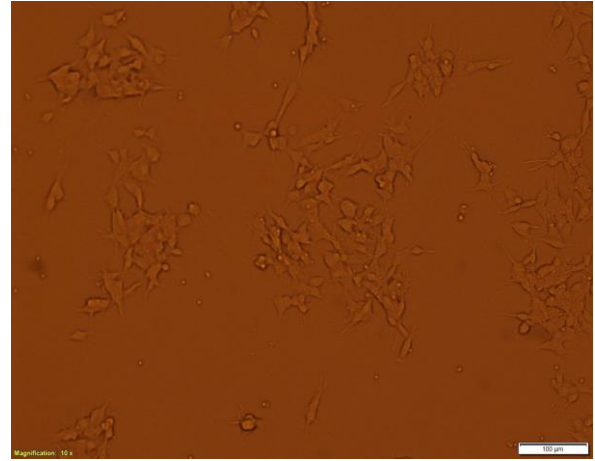
**Outcomes:**

1. Successful growth and stabilisation of the SHSY5Y cell line in the lab.
2. Successful growth of these cells in 1mM and 5mM beta-hydroxybutyric acid (a ketone body) cell culture medium in the absence of glucose inducing ketosis in SHSY5Y cells over a 10 day period.
3. Comparisons made with no glucose control and regular feeding medium.

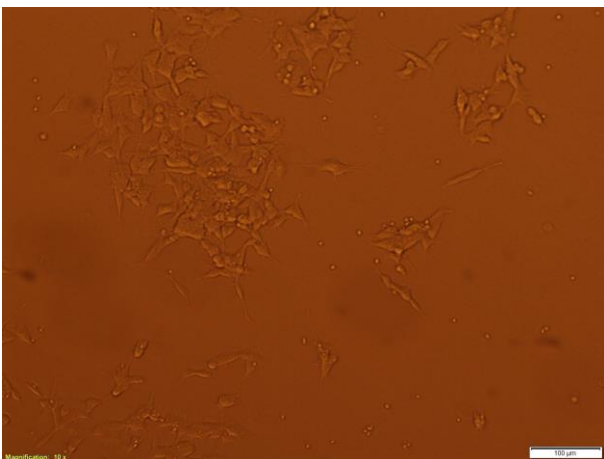
4. Preliminary findings that cell number is decreased, cell proliferation is slowed and cell morphology is altered in cells treated with ketogenic medium.



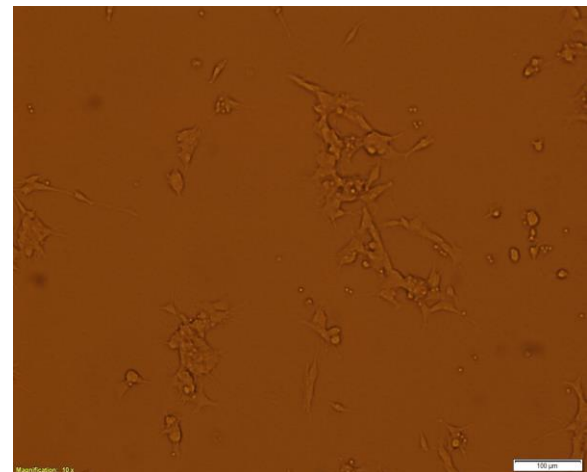
1mM Ketone Media



5 mM Ketone Media



No-glucose Media



Standard DMEM-F12 Media with Glucose

These findings are significant in the context of future research underlying the cellular effects of ketone treatment and its role in cell differentiation. This work also highlights the need for a greater understanding of the effects of the ketone diet on brain development and will complement new animal based research to be undertaken in the near future.

*Signature of student.....Lindsey B. Watters.....Date.....9/11/17*

*Signature of supervisor.....Dr. Denis Barry*

*Date 10/11/17*

