CALIFORNIA STATE SCIENCE FAIR
2003 PROJECT SUMMARY

Name(s)
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Project Number
S1404

Project Title
Regulation of Receptor Expression by Thyroid Hormone and Methoprene Acid

Abstract

Objectives/Goals
The purpose of this experiment was to determine the affects of Methoprene acid exposure on 3T3 cells. The objective is to expose the two hormones, T3 and Methoprene Acid, to the 3T3 cells (Mouse fibroblasts) and determine if the receptors are actually expressed, and if the two hormones alter the expression of these receptors.

Methods/Materials
Cell Culture-3T3 cells were grown in culture medium with serum at 370C. Cells were removed from the flask with trypsin and counted with a hemacytometer. The same number of cells were plated into each well of a 24-well tissue culture plate. After 24 hours, the test solutions were added to each well (10-7 M MA and 10-4 M T3.
cDNA Synthesis:
1. Detach cells and wash once with cold PBS buffer.
2. Count the number of cells using a hemacytometer.
3. Add ice-cold Cell Lysis II Buffer, mix, and incubate for ten minutes
4. Add DNase
5. Incubate at for 15 minutes.
6. Inactivate the DNase
7. Heat Denaturation of the RNA template
8. Reverse Transcribe RNA
a. Assemble mixture in microfuge tube then mix gently
b. Heat for 3 Minutes
c. Place reaction on ice for 1 min; centrifuge briefly and place back on ice.
d. Add the remaining RT reagents, then mix gently and
e. Incubate for 15-60 minutes
f. Incubate for 10 min to inactivate the reverse transcriptase.
g. Store reaction at -20(C

PCR Amplification and Gel Electrophoresis

Results
Unexposed 3T3 cells express RXRg at detectable levels by using PCR. Unexposed 3T3 cells do not

Summary Statement
the regulation of receptor expression by two hormones: Thyroid Hormone and Methoprene Acid

Help Received
Father helped teach techniques, Dr's in laboratory helped teach techniques and lab manners.