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Effects of Raspberry Ketone on Lipid Metabolism

People consume fat burners in an effort to lose fat, however, these compounds may have additional negative effects on the body. An example of a fat burner is raspberry ketone; it has been shown to break down fat in adipose cells. It is possible that raspberry ketone and other fat burners may breakdown lipids that are important in maintaining proper cell function, i.e. phospholipids. The raspberry ketone mechanism of action is not fully understood and understudied. Therefore, we aimed to study the effects of raspberry ketone on essential lipids in the cell by assessing gene expression of important lipases using the HL-60 (Human Leukemia) cell line. We hypothesized that the cells treated with raspberry ketone will exhibit an increase in gene expression of lipase, which may lead to a decrease in essential lipids in the cell. The project was initiated by establishing a cell line of HL-60 cells. Then, a pre-treatment cell count was done using trypan blue to assess the density and percent viability of the cells. This was followed by treatment with 50µM, 20µM, and 10µM of raspberry ketone. A post-treatment cell count was then taken and no changes in cell density or cell viability were detected at these concentrations. In order to measure changes in gene expression of hormone sensitive lipase, and phospholipase A2, RNA was extracted from the cells, converted into cDNA, and RT-PCR was performed. Cells treated with 250µM, 500µM and 1000µM of raspberry ketone are currently being assessed for cell density, cell viability, and gene expression as described above.

Effects of Raspberry Ketone on Lipid Metabolism

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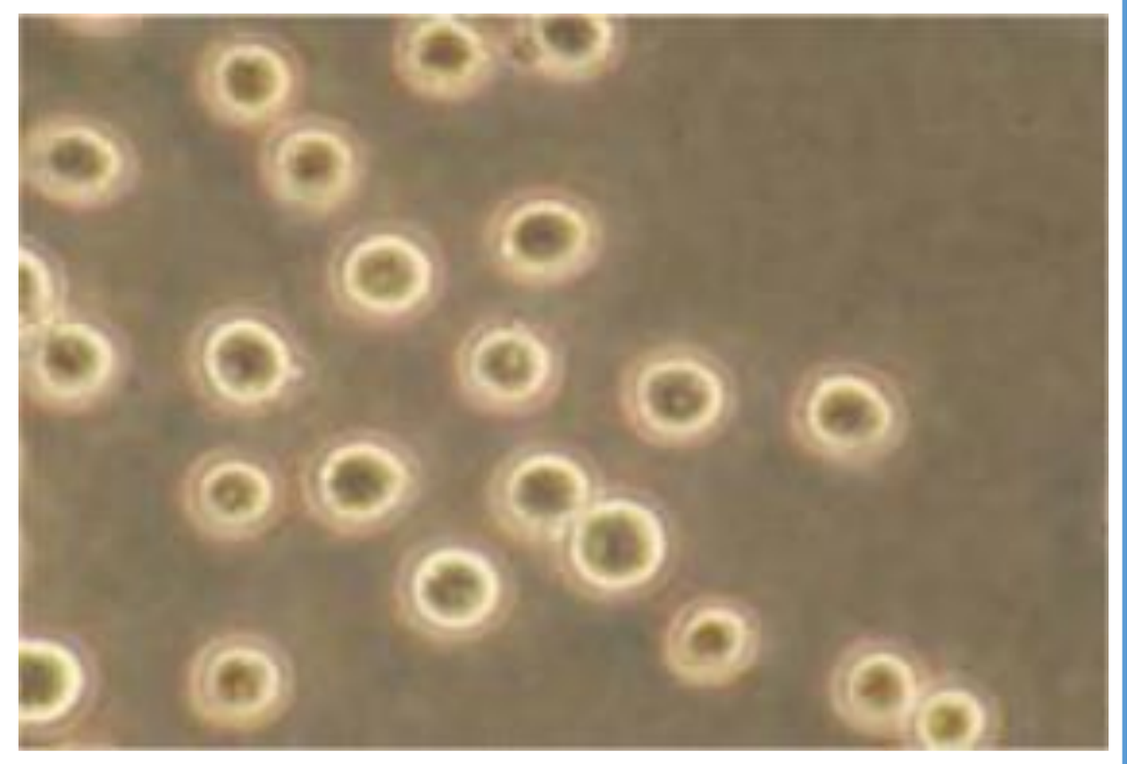
Beta Beta Beta National Biological Honor Society

Introduction

- People consume fat burners to lose the fat present in the abdomen with disregard to the ultimate effects on essential fats in the body.
- Raspberry ketone (RK), [4-(4-hydroxyphenyl) butan-2-one] is an aromatic compound extracted from red raspberries and has been used in cosmetics.
  - Decrease percentage of adipose tissue.
  - Inhibits fat accumulation.
  - Elevates expression of the lipase to boost lipid metabolism.
- Cell membrane is composed mostly of phospholipids.
- Lipase enzymes are the responsible components in hydrolyzing their specified lipids in the body, including phospholipids. Two types of lipases:
  - Hormone sensitive lipase, hydrolyze esters during lipogenesis.
  - Phospholipase A2, cleaves phospholipids.
- **Purpose:** investigate whether raspberry ketone is capable of increasing the metabolism of essential lipids by assessing the expression of lipases in the cell.
- **Hypothesis:** cells treated with raspberry ketone will display increased gene expression of lipases.

Methodology

- HL-60 (Human acute myeloid leukemia cells) cell line culture was established.



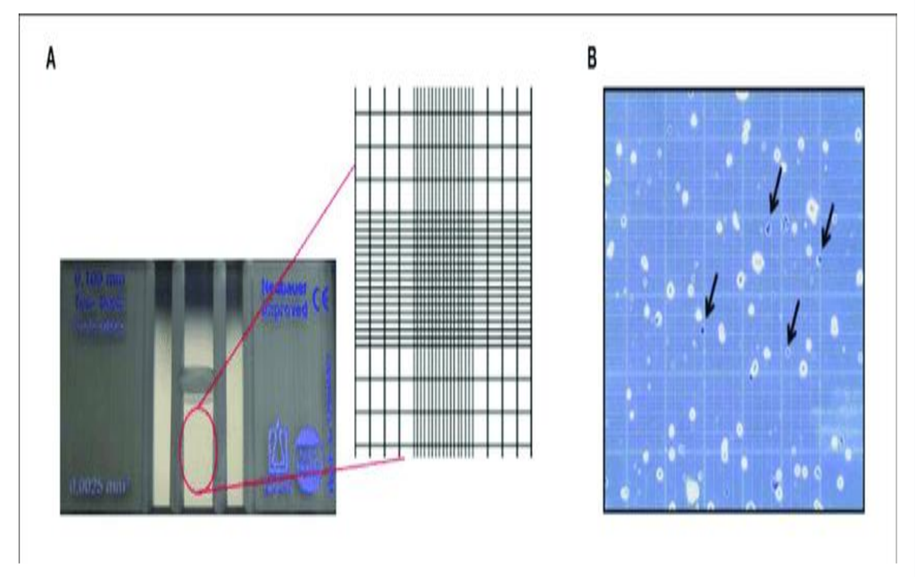
Methodology

Pre-treatment cell count and RNA extraction → Treatment day one
- Untreated, and 0.25% ethanol control.
- 250 µM, 500 µM, and1000 µM.

Media exchange on day three, containing same amount of Raspberry Ketone → Treatment day seven

Post-treatment cell count and RNA extraction → RT-PCR reaction was done
- convert RNA → cDNA

PCR reaction using three primers:
- Actin → Reference gene
- Hormone sensitive lipase (LIPE)
- Phospholipase A2 (PLA2G5)



Ct values were obtained → Data analysis was performed including Student T-test.

RNA Concentration		
Flask	Concentration (µg/µl)	A260/280
Pre-Treatment	0.231	1.95
Untreated Control	0.676	2
Untreated Control	0.561	1.95
ETOH Control	0.596	1.79
ETOH Control	0.515	1.973
250 µM of RK	0.348	1.879
250 µM of RK	0.188	1.929
500 µM of RK	0.2	1.953
500 µM of RK	0.209	1.921
1000 µM of RK	0.12	1.993
1000 µM of RK	0.149	1.7

Table 1- RNA concentration. Representing the purity and amount of RNA extracted for each flask.

Results

The effects of Raspberry Ketone on cell density		
Flask	Pre-Treatment Cell Density	Post-Treatment Cell Density
Untreated Control	1980000	1645000
Untreated Control	1955000	2115000
ETOH Control	745000	290000
ETOH Control	1210000	300000
250 µM of RK	990000	355000
250 µM of RK	1680000	360000
500 µM of RK	1280000	155000
500 µM of RK	570000	315000
1000 µM of RK	430000	210000
1000 µM of RK	1235000	220000

Table 2- The effects of Raspberry Ketone on cell density. Represent the experimental groups according to flask. As shown, a reduction in the cell density of post-treatment groups, focusing on the raspberry ketone administered groups, suggesting more cell death due to the presence of raspberry ketone treatment. Cell density represents the concentration of viable cells per ml.

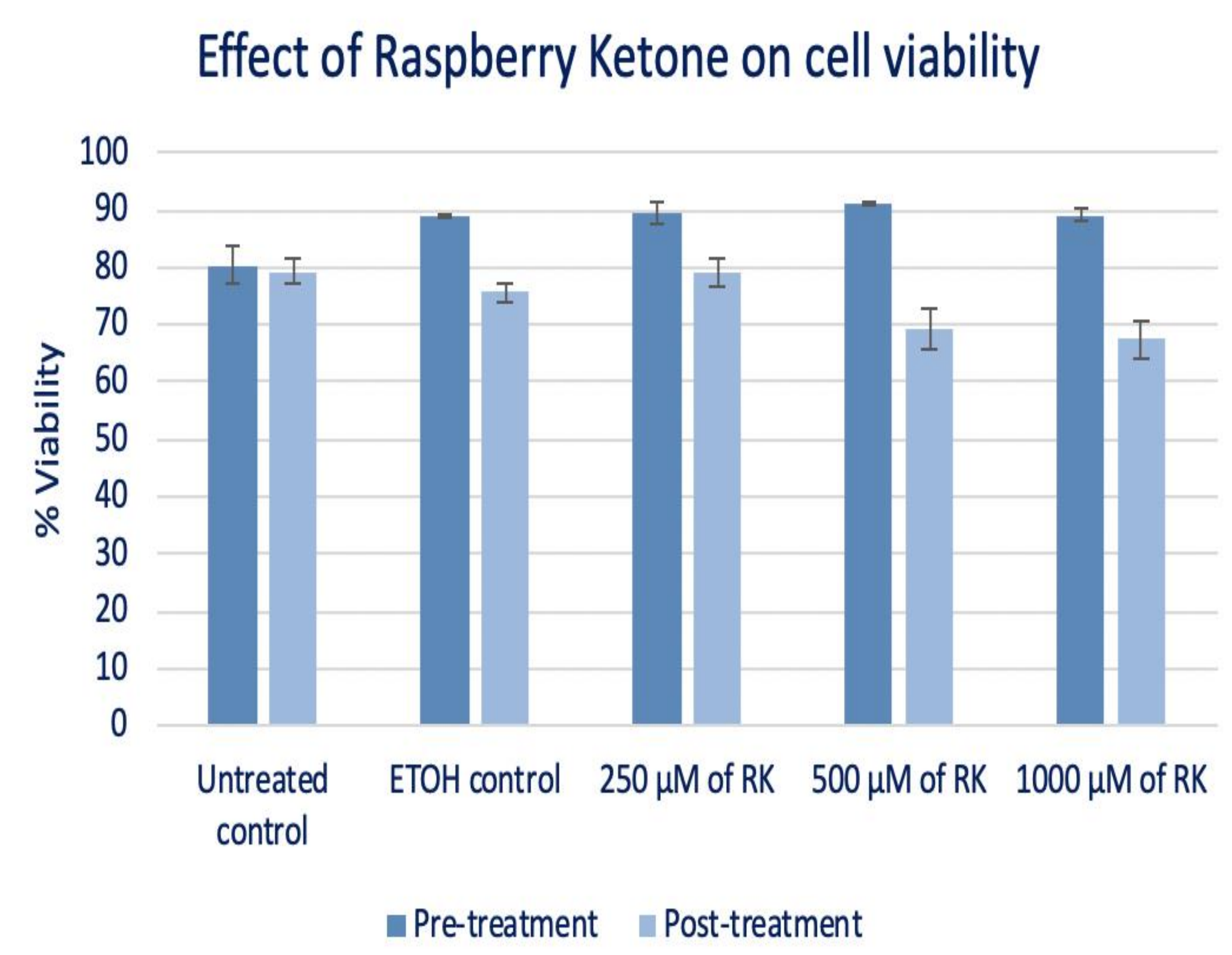


Figure 1- Effects of Raspberry Ketone on cell viability. As represented, a trend was observed when comparing groups of different raspberry ketone concentration and controls to one another. The graph display cell viability mean, and the standard error of the mean (SEM).

Results Continued

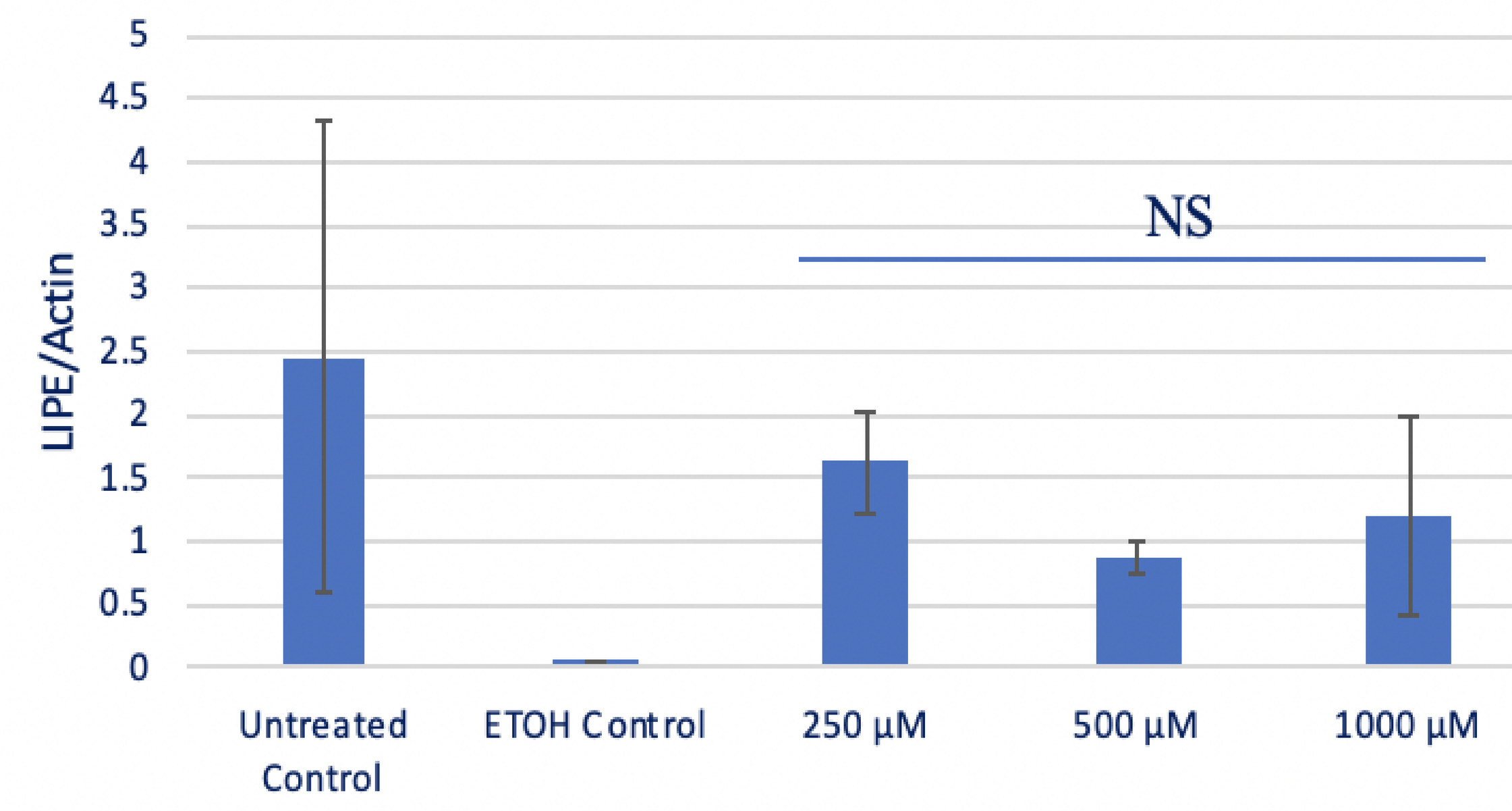


Figure 2- Hormone Sensitive lipase. The graph display the mean of groups and standard error of mean (SEM). A decrease in the experimental groups suggests that raspberry ketone did not upregulate its activity. P value 250 µM, 0.77752441. P value 500 µM, 0.57292856. P value 1000 µM, 0.71638974. Student T-test, two tail, alpha < 0.05.

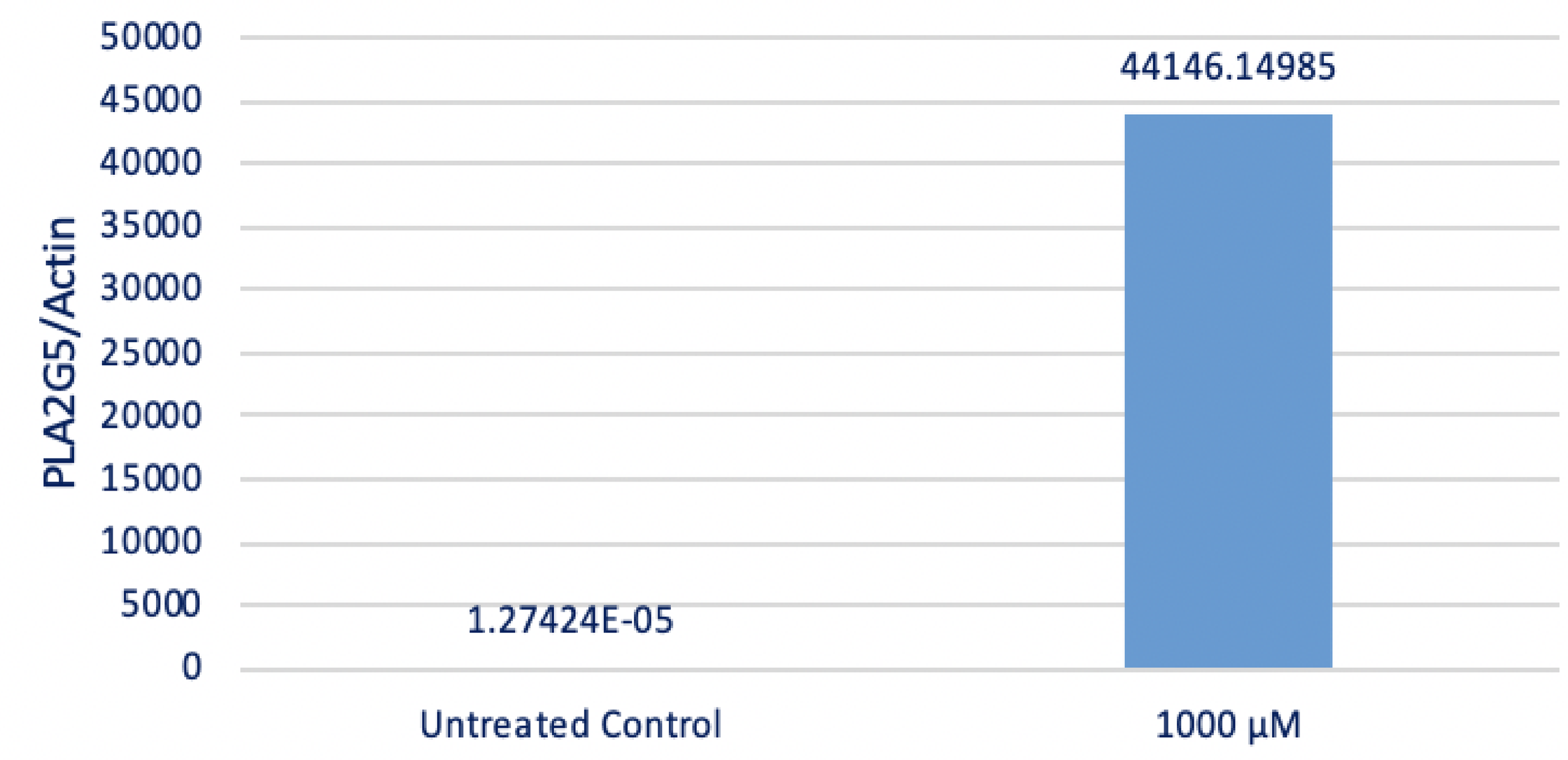


Figure 3- Phospholipase A2. A noticeable increase with the highest concentration. Neither the mean nor standard error were shown due to the presence of one group only.

Conclusion

Results demonstrated a comparable decrease in cells viability in experimental treated groups but not untreated control. Tying to increase in the 1000 µM phospholipase A2, suggesting that cell death was caused due to the breakdown of the phospholipid bilayer. No significant effect on hormone sensitive lipase.

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