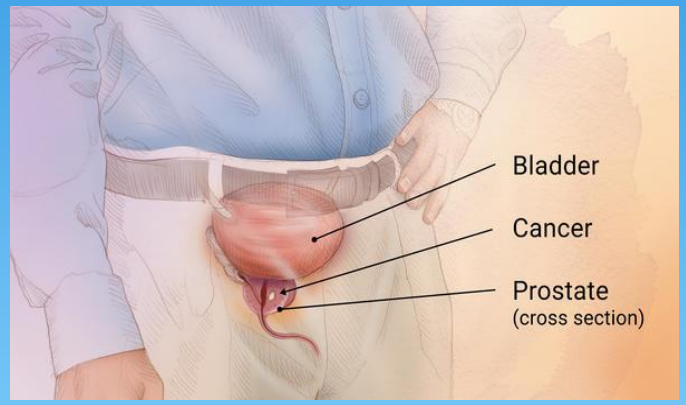
# Impact of vitamin C on genistein-induced cell death in prostate cancer

1st Electronic Conference on Molecular Science

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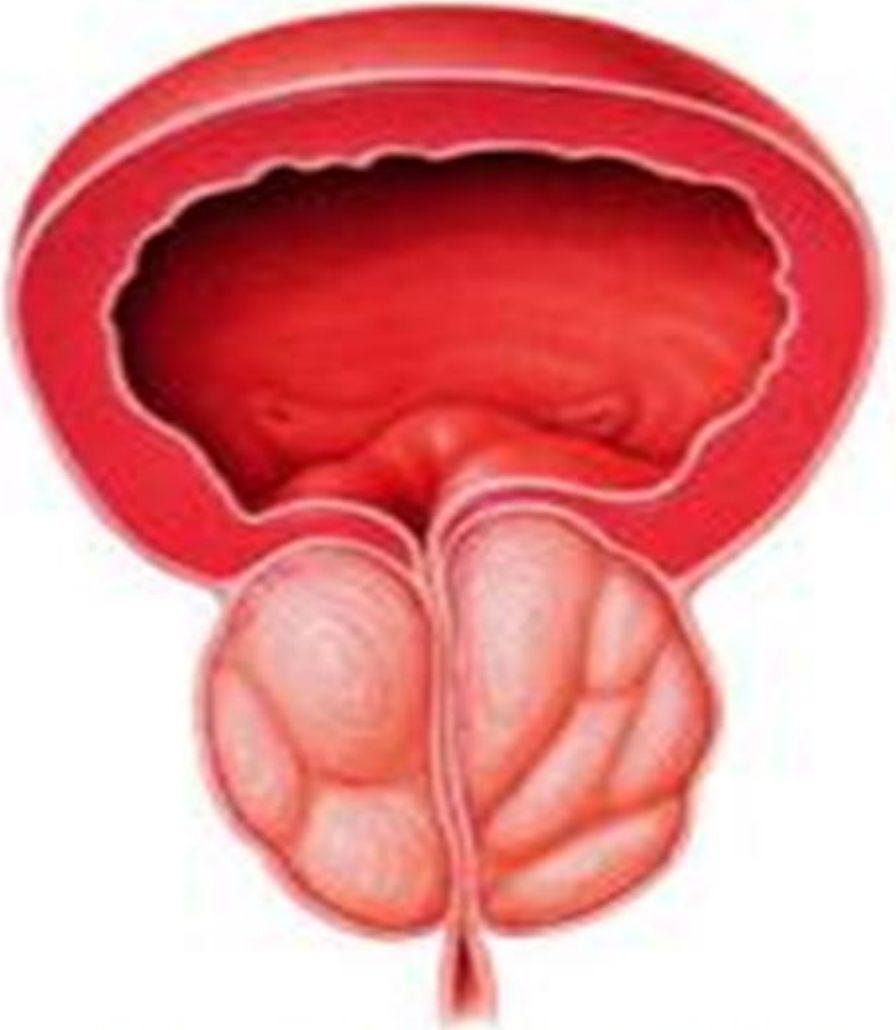
09/20/2015

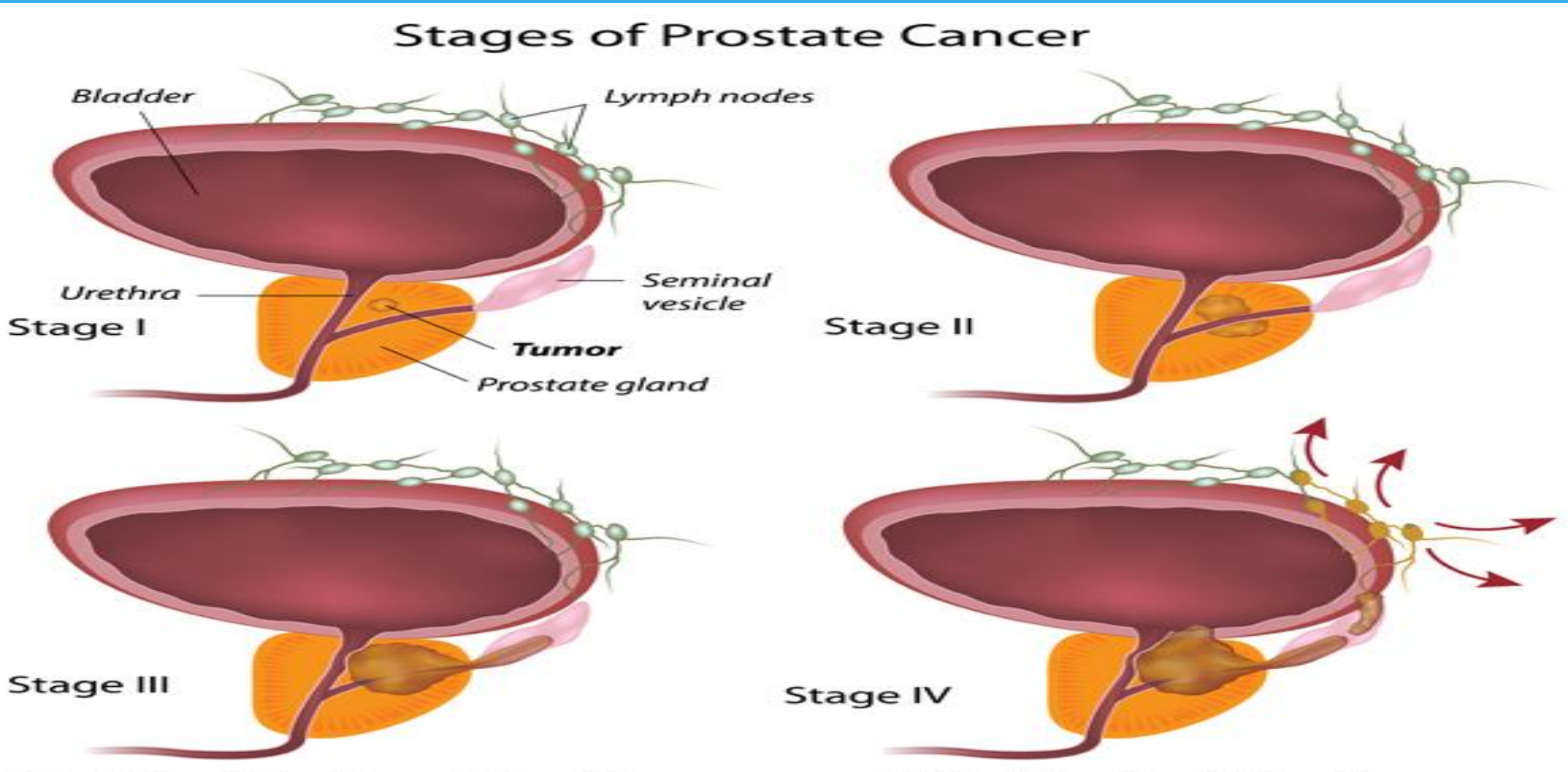
# Introduction

Normal Prostate



Enlarged Prostate

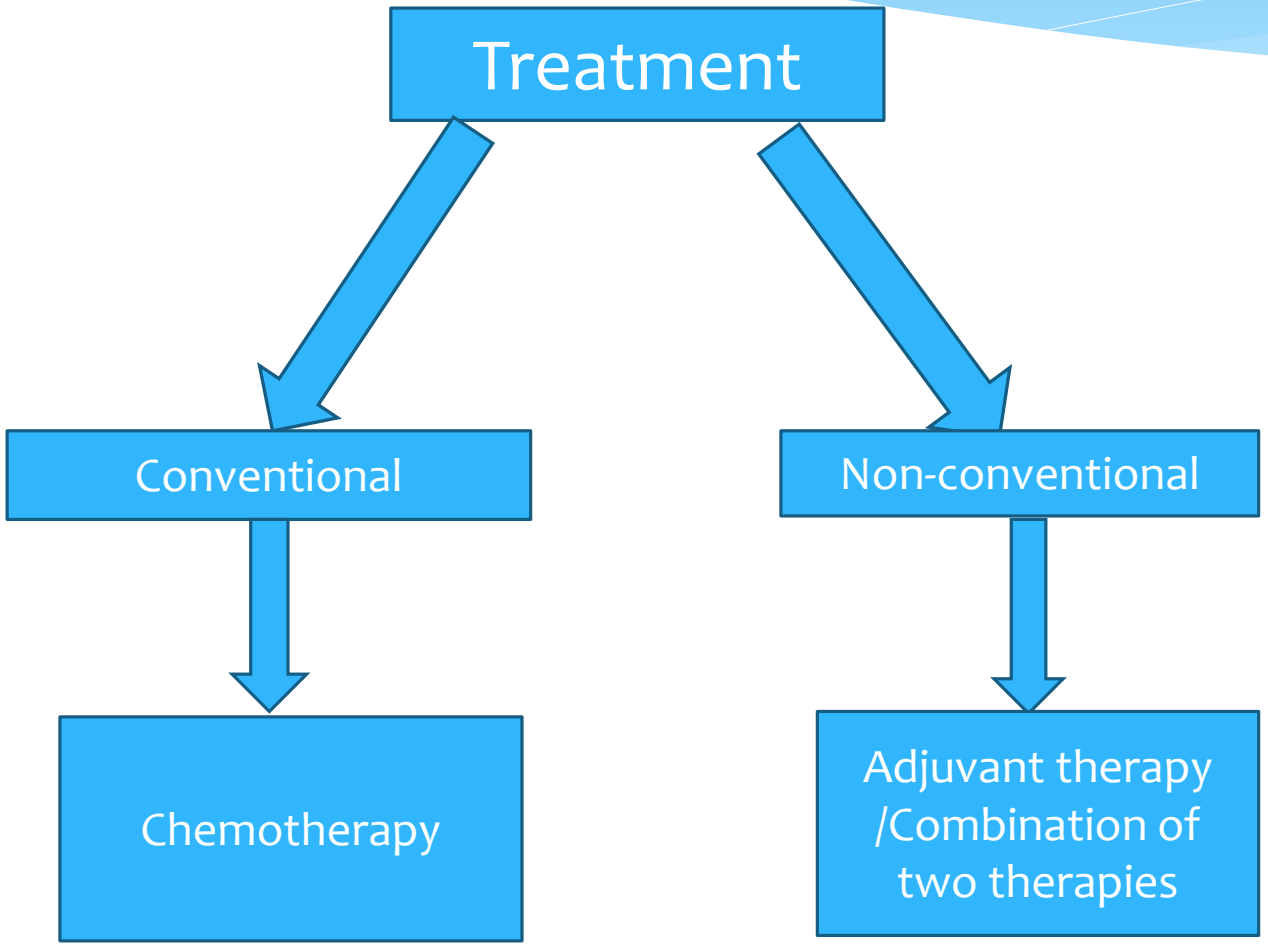
# Stages of Prostate Cancer



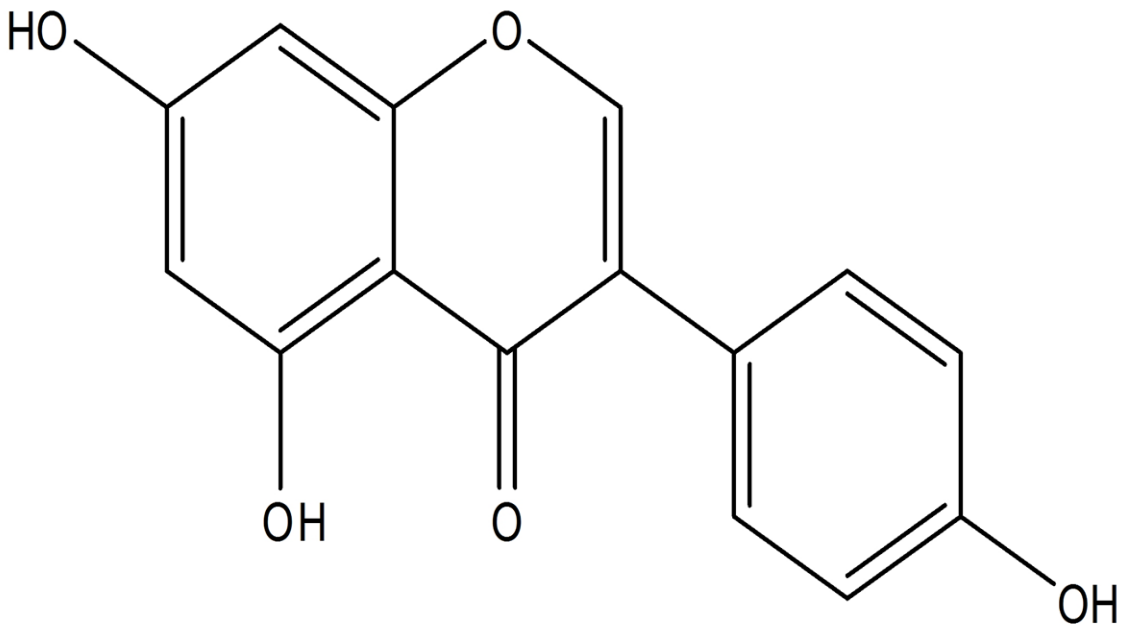
# Key statistics of prostate cancer

* Prostate cancer(PCa) is the most common cancer in American men.
* Estimated 220,800 new cases and 27,540 cancer-related deaths in 2015.
* About 1 in every 7 men will be diagnosed with PCa during a lifetime.
* About 6 cases in 10 are diagnosed in men aged 65 or older.
* The average age at the time of diagnosis is about 66.

# Treatment options of prostate cancer



# Adjuvant therapy / combination of two therapies


5,7-Dihydroxy-3-(4-hydroxy-phenyl)-chromen-4-one
(**genistein**, isoflavone)

# Objective

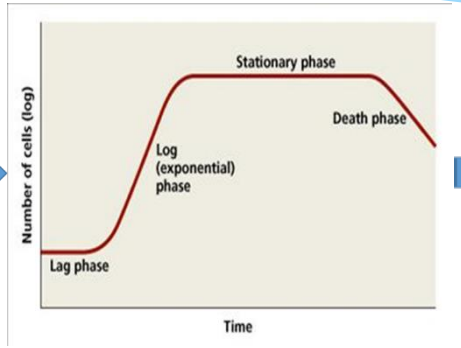
The Purpose of this study was to:

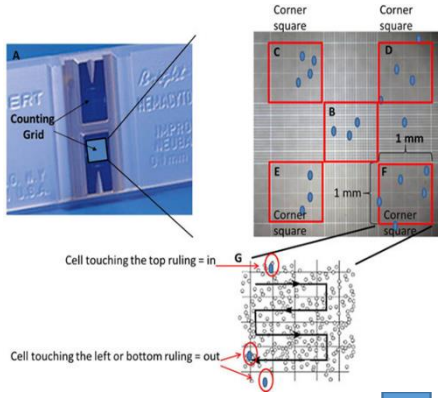
Investigate and  determine the potential therapeutic additive effect of genistein and vitamin C on prostate cancer

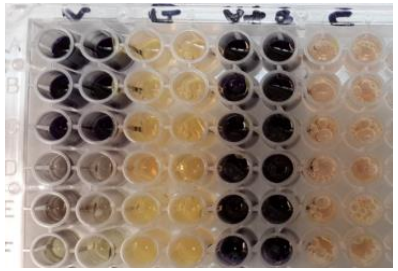
# Null Hypothesis

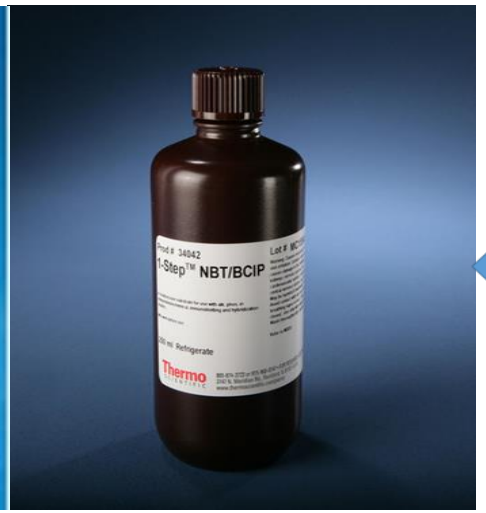
Combine treatment of genistein and vitamin C will not cause more cells to die by apoptosis than genistein only.

# Materials and methods

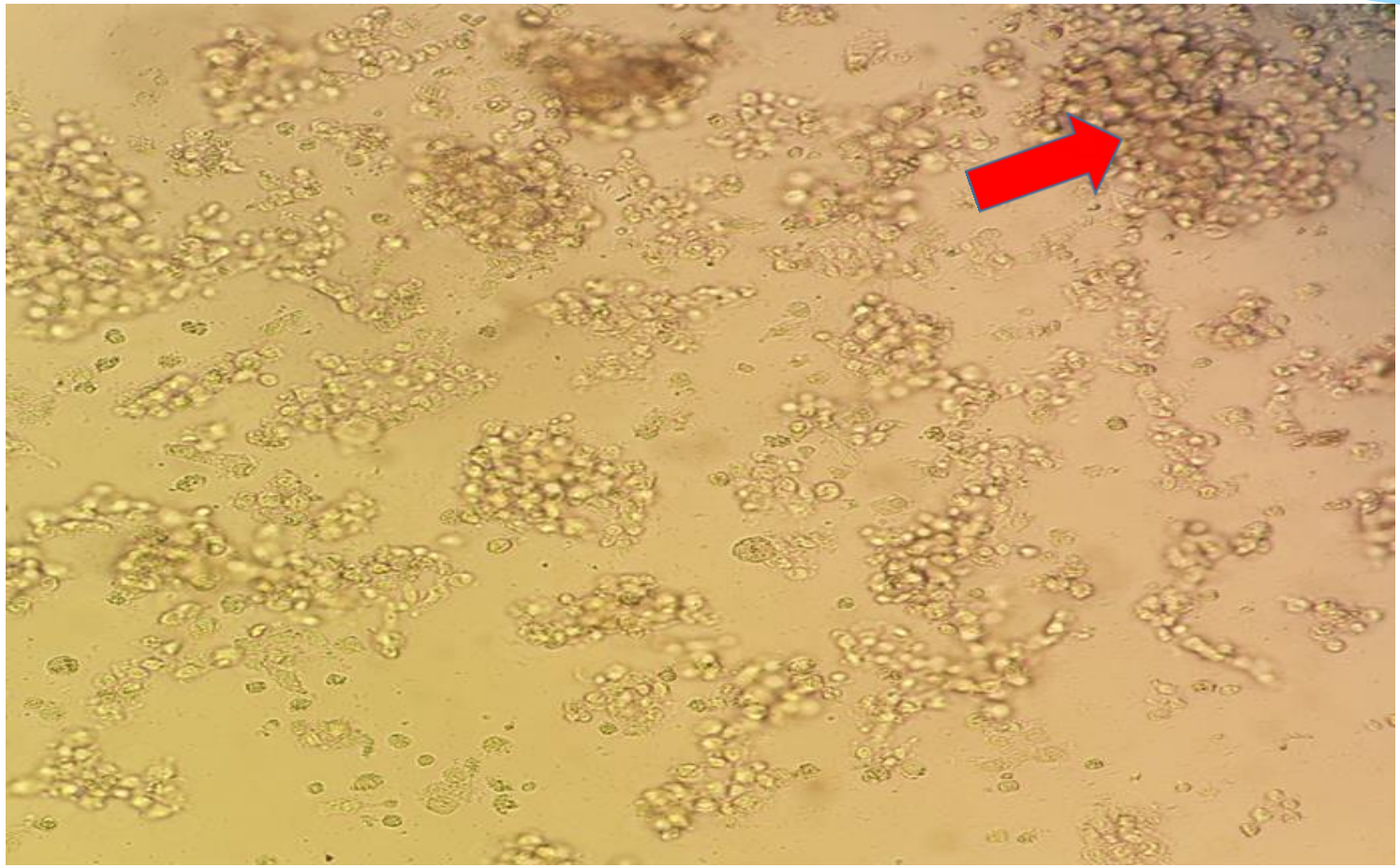



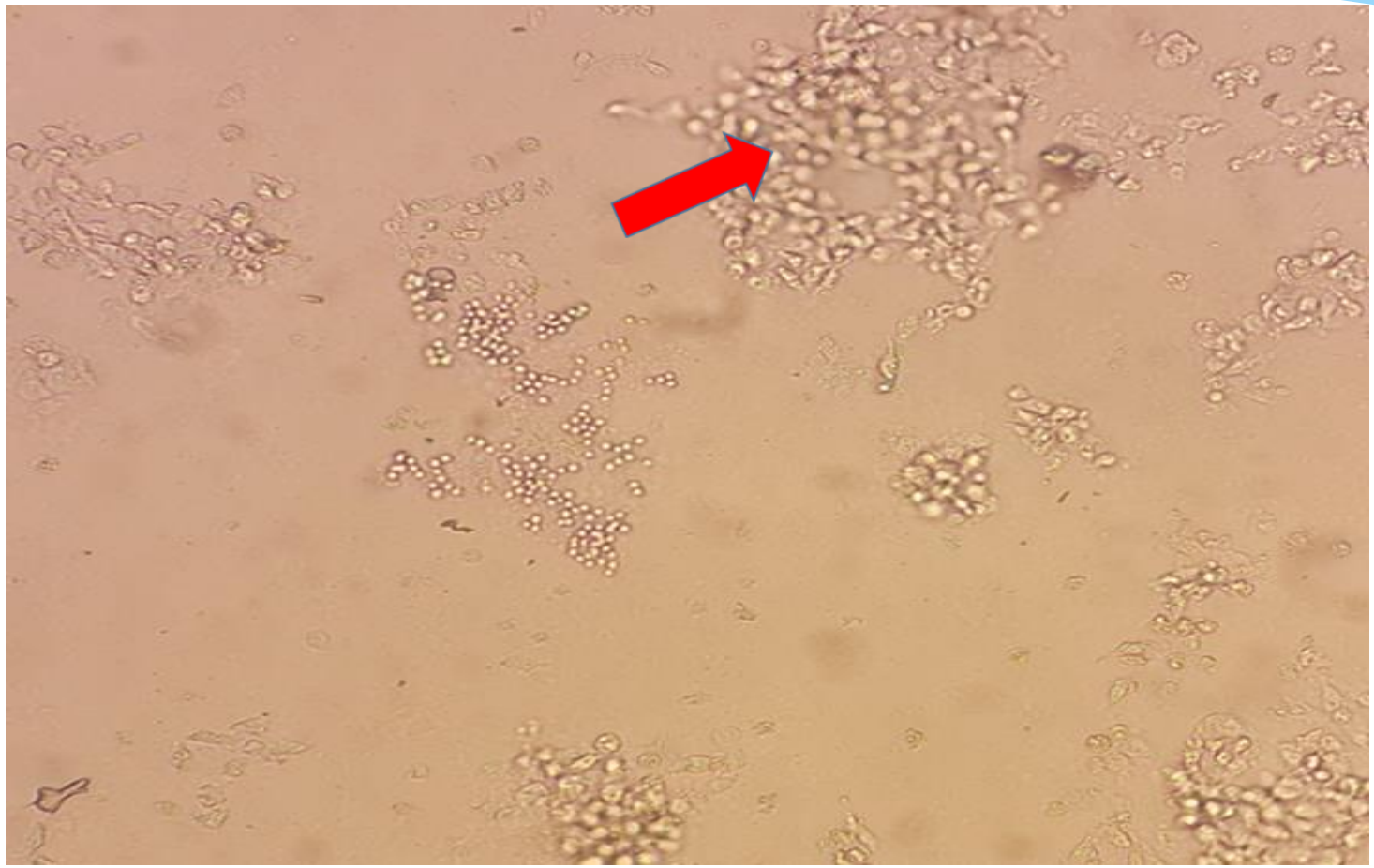


# Pictures of cancer cells after treatment



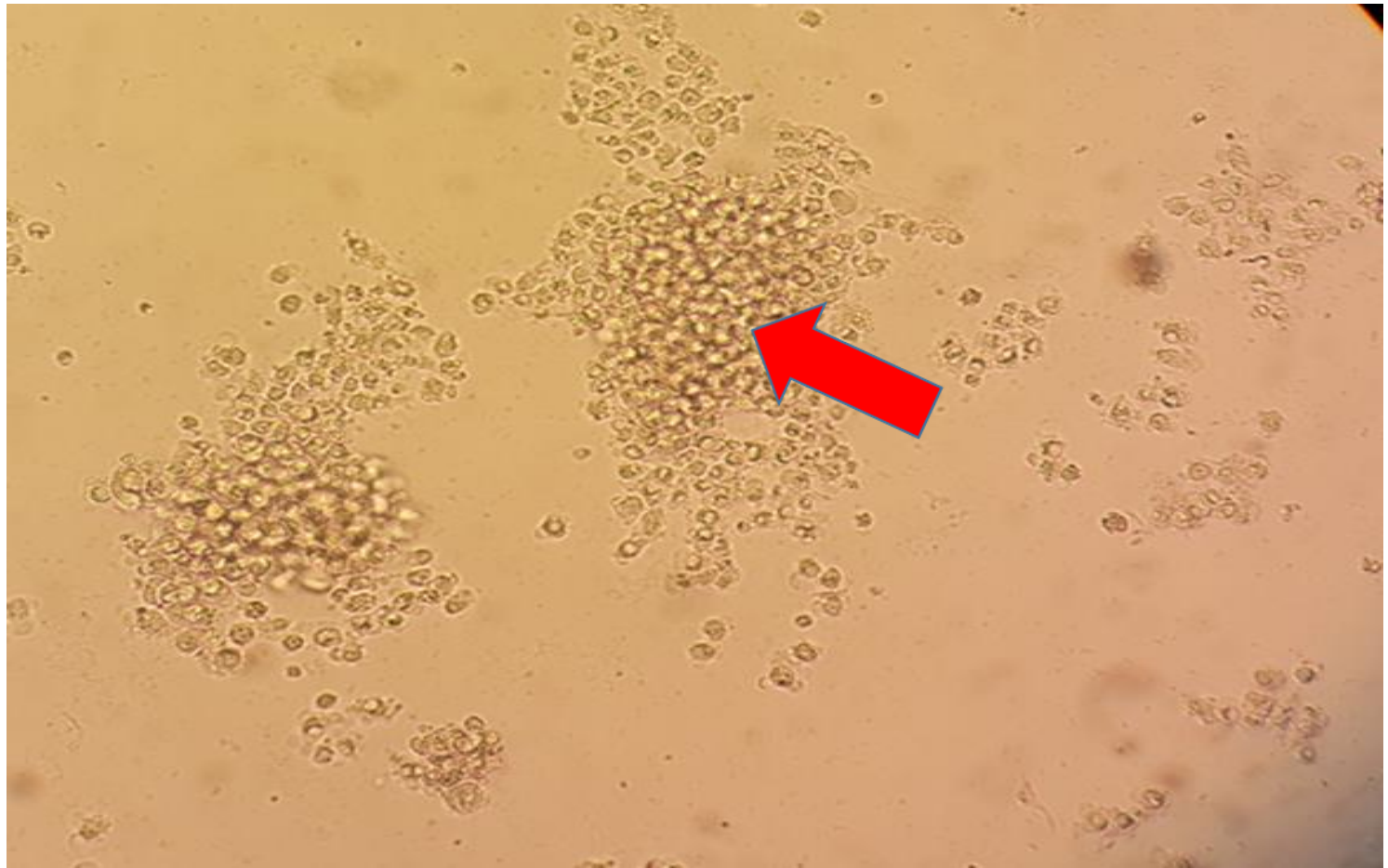
24hrs 40uM
vitamin C

# Pictures of cancer cells after treatment



24hrs 40uM
genistein

# Pictures of cancer cells after treatment



24hrs (40uM vitamin C + 40uM genistein

# MTT Assay result

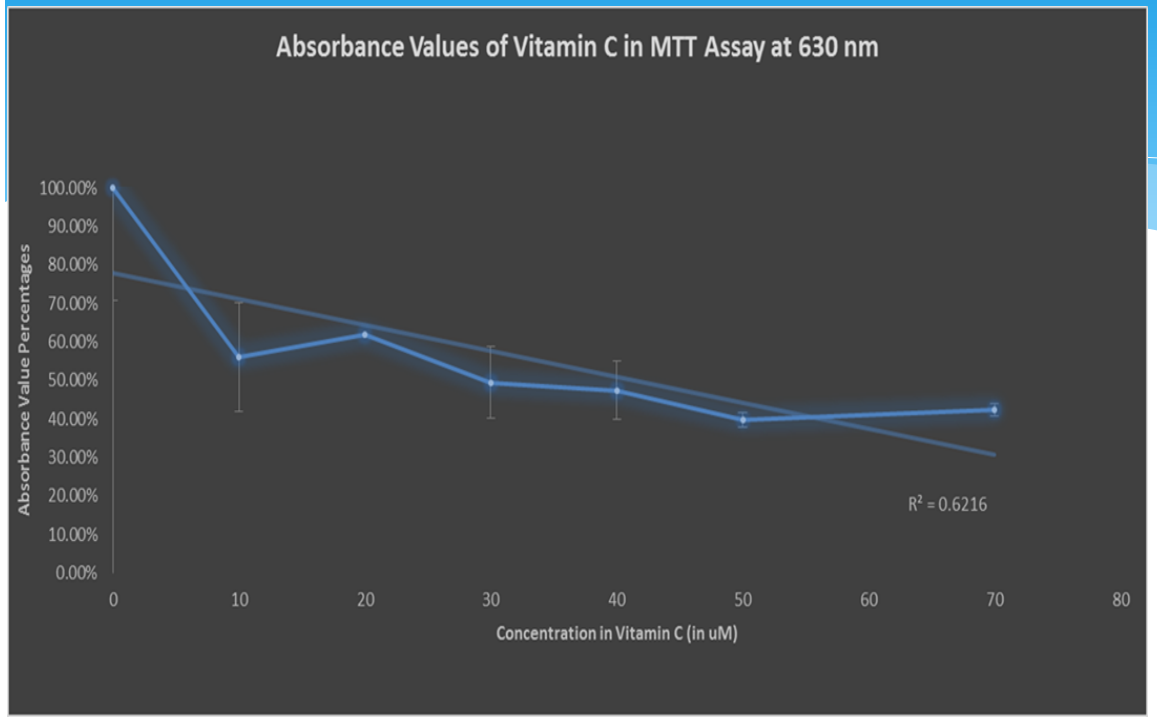



Fig A

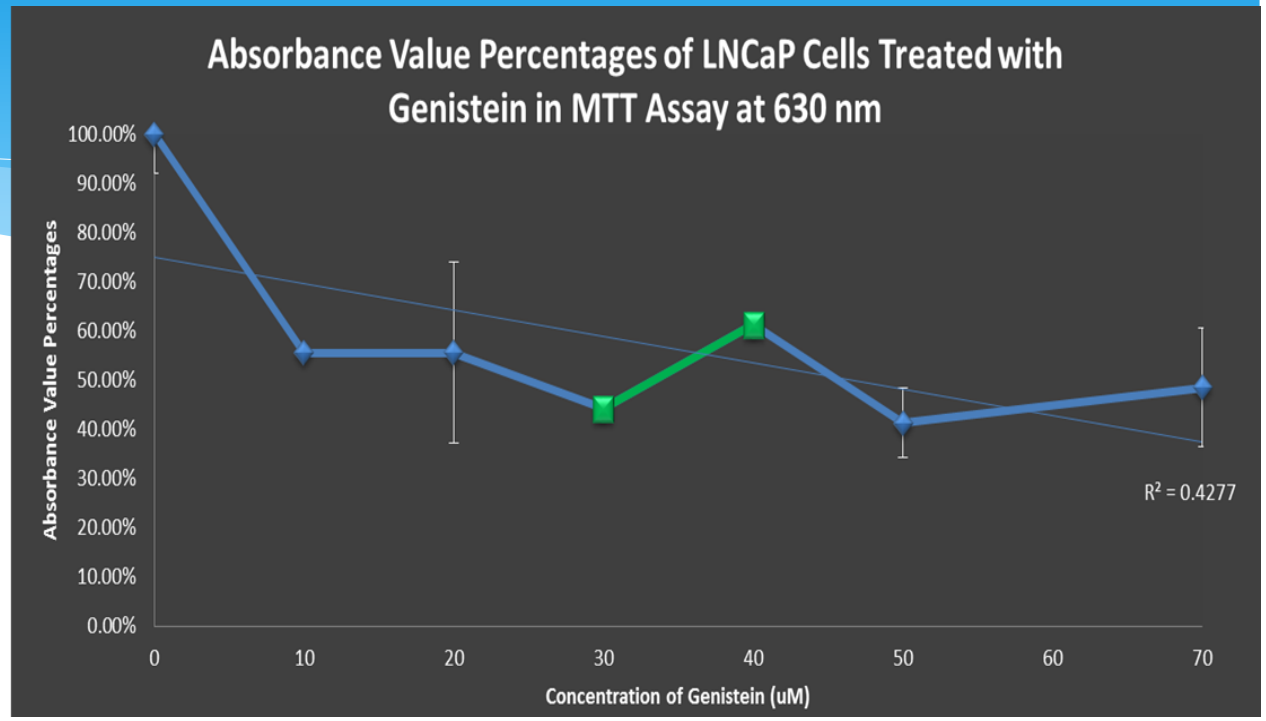
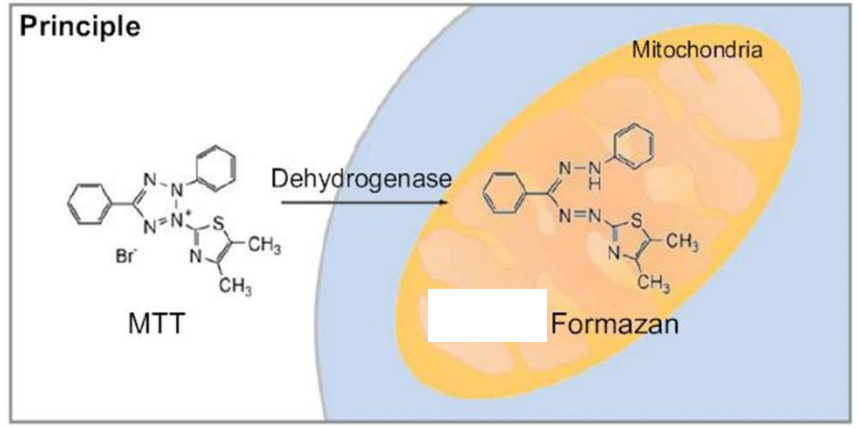



Fig B

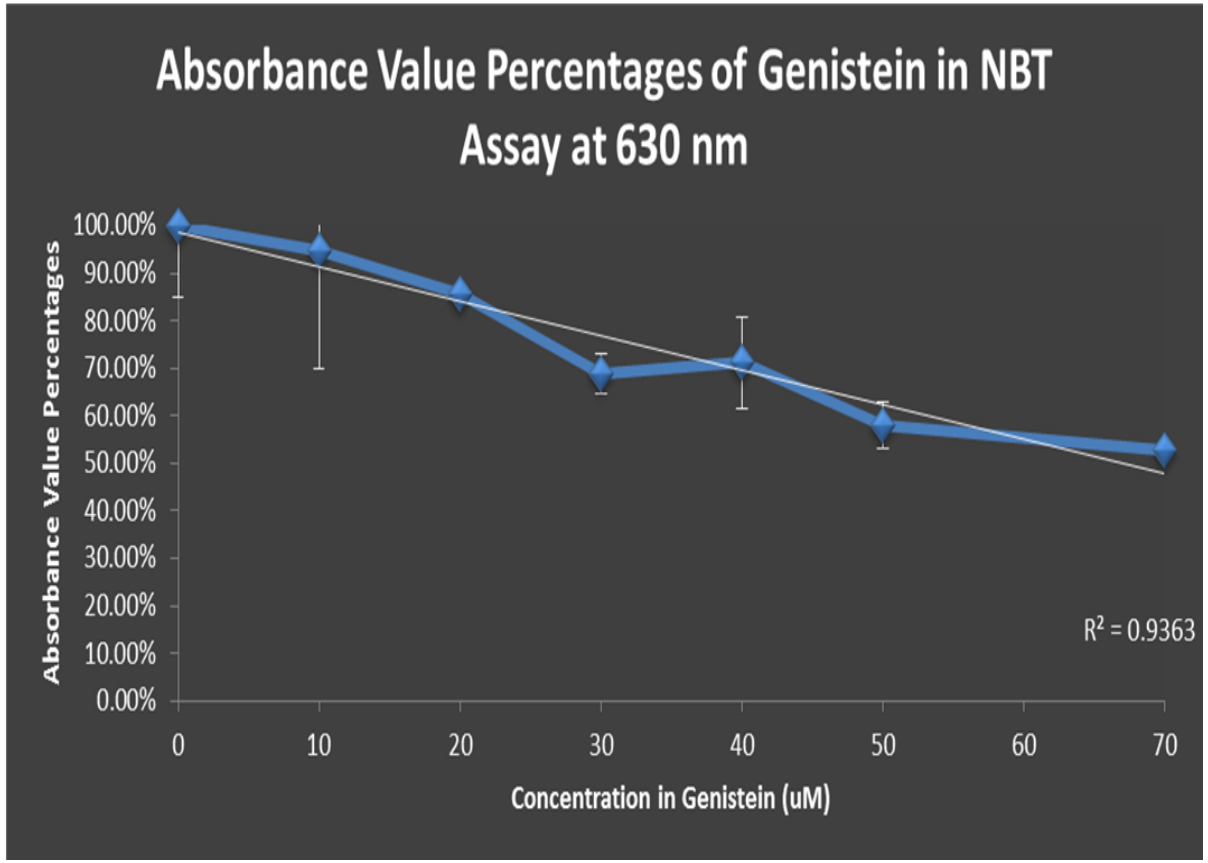
IC-50 of vitamin C = 40uM

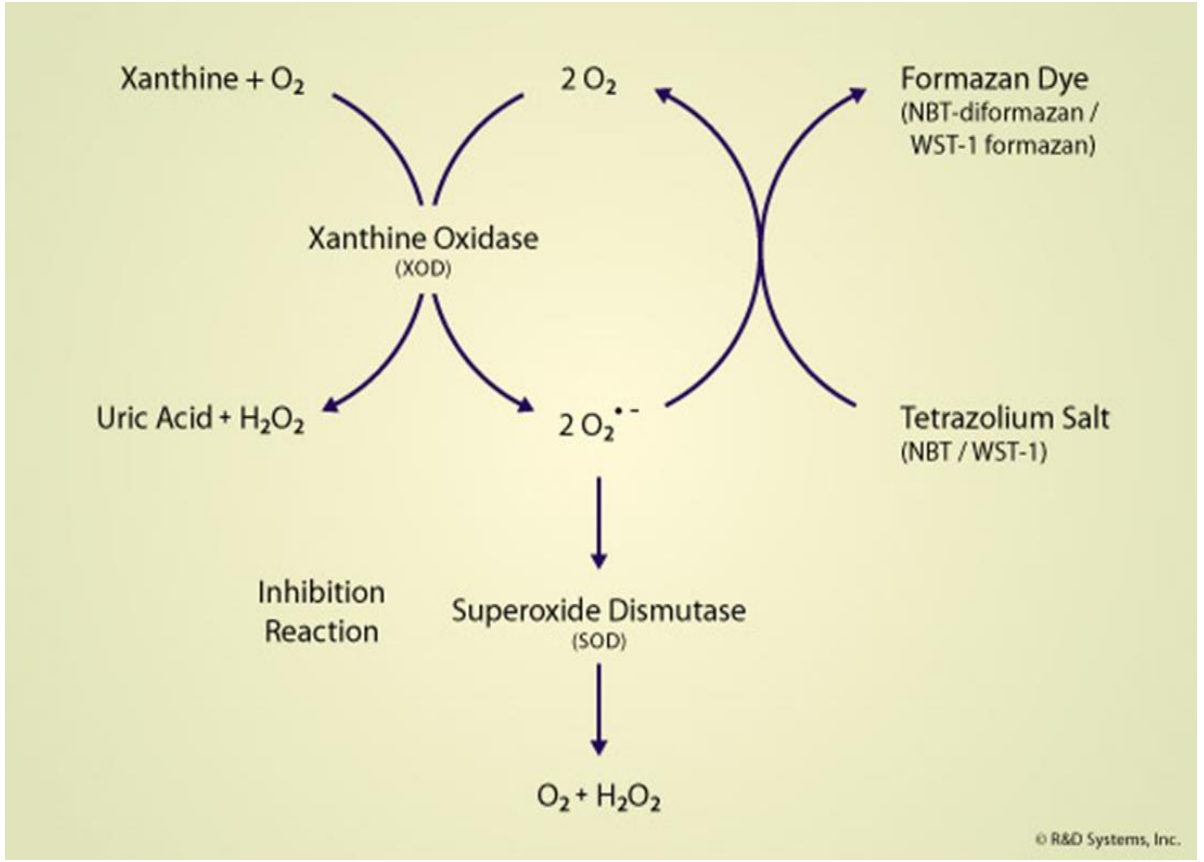
IC-50 of genistein = 28uM

Higher concentration of vitamin C is needed to kill 50% of LNCaP cells.

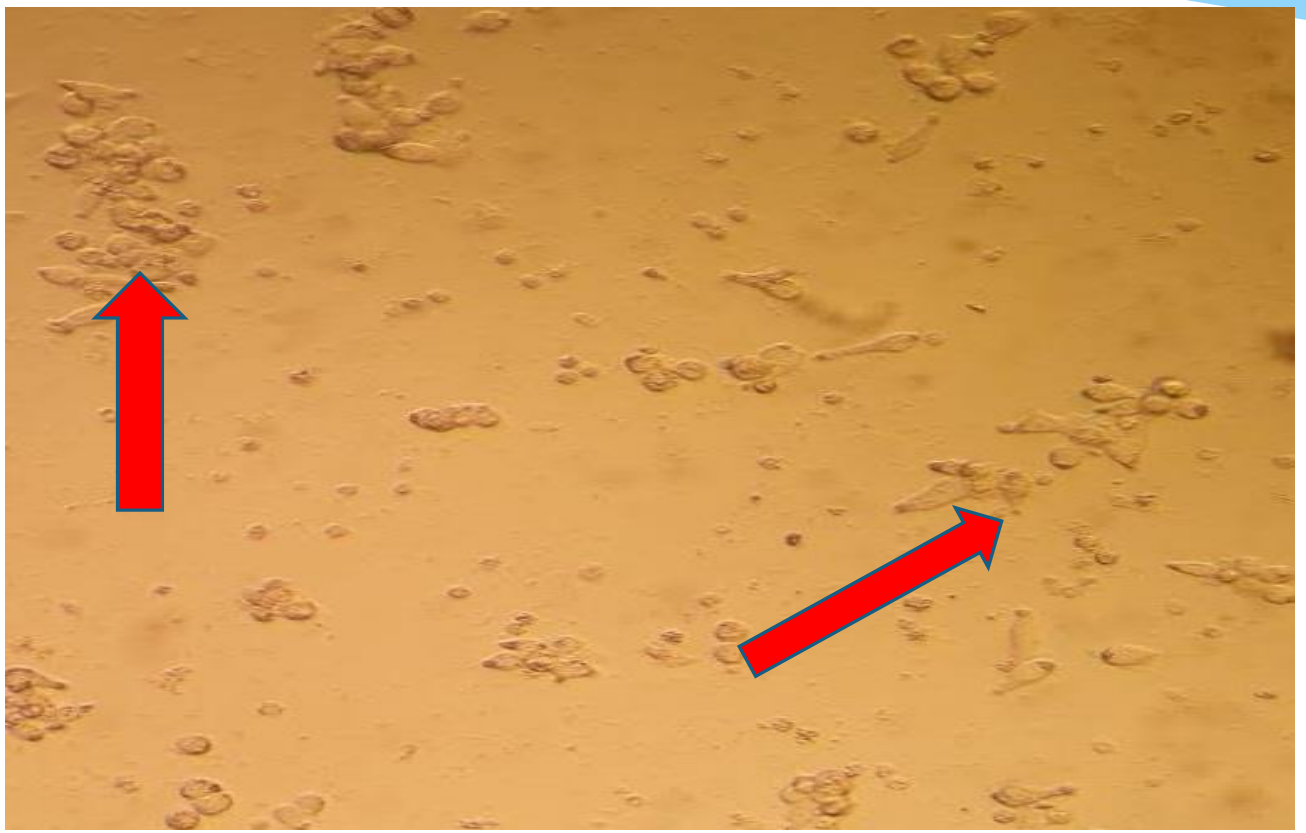


# NBT Assay result

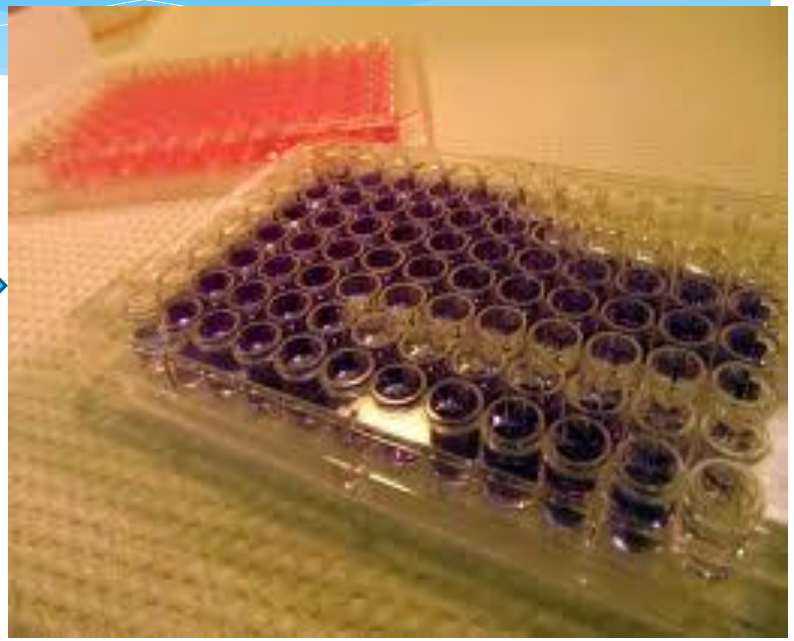


# Picture of LNCaP cells few minutes before treatment

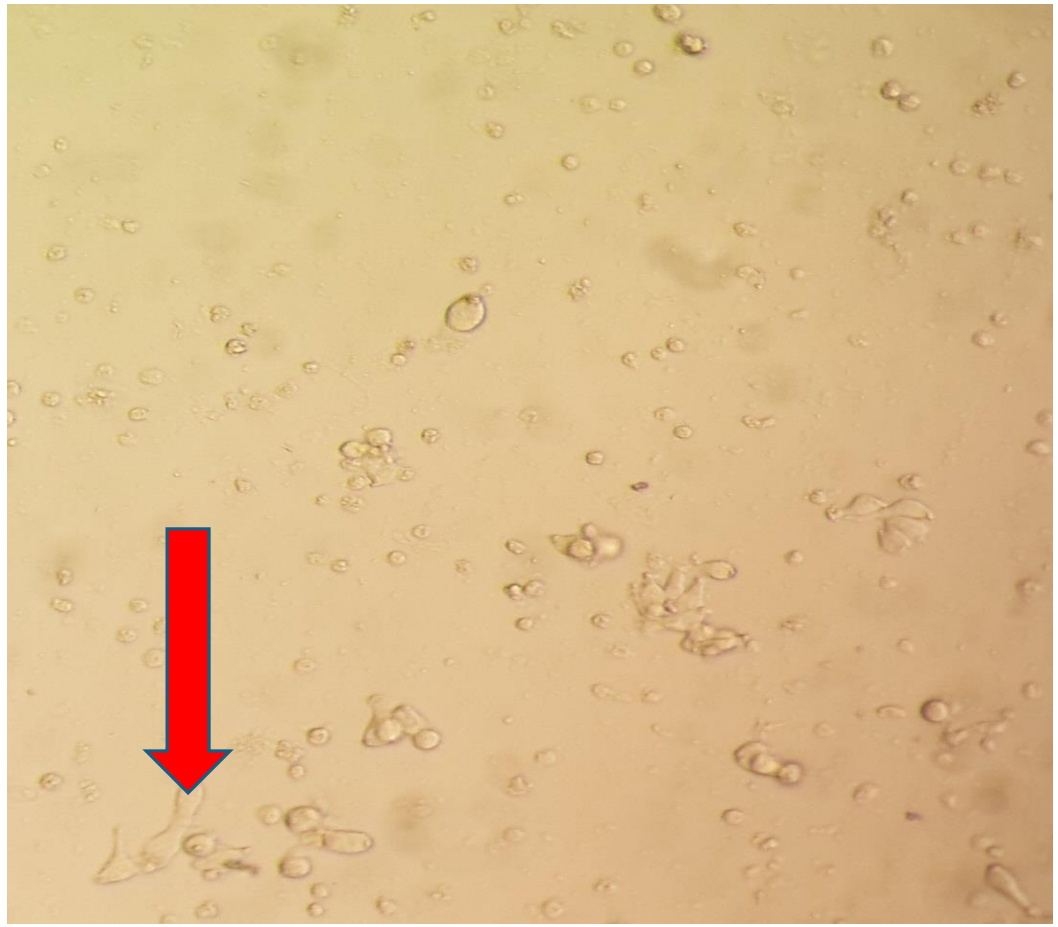


4th day few minutes before treatment with drug

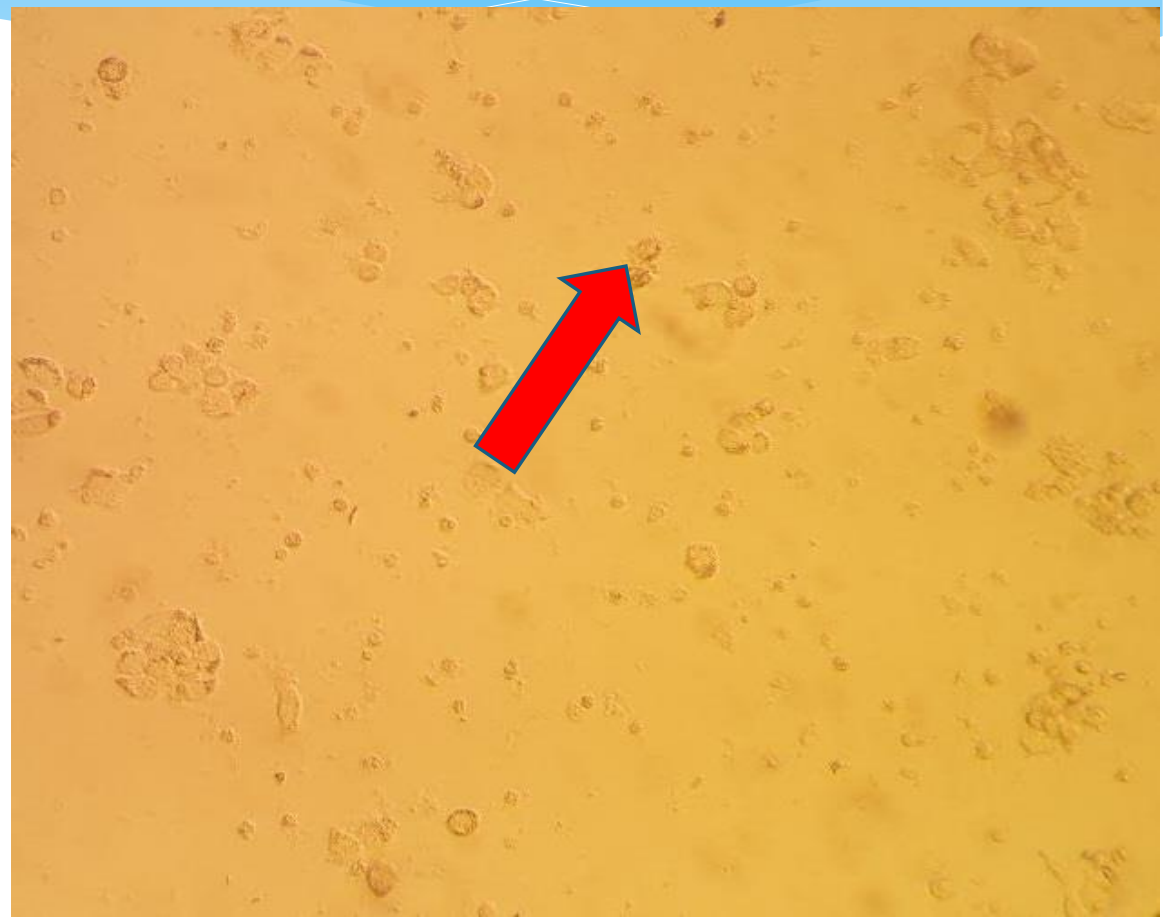


Cells were seeded into the 96-well MTP at 100ul on the 3rd day, cells treated when they reached 80% of confluence on 4th day and MTT assay performed on the 5th day .

# Pictures of LNCaP cells 24hrs after treatment

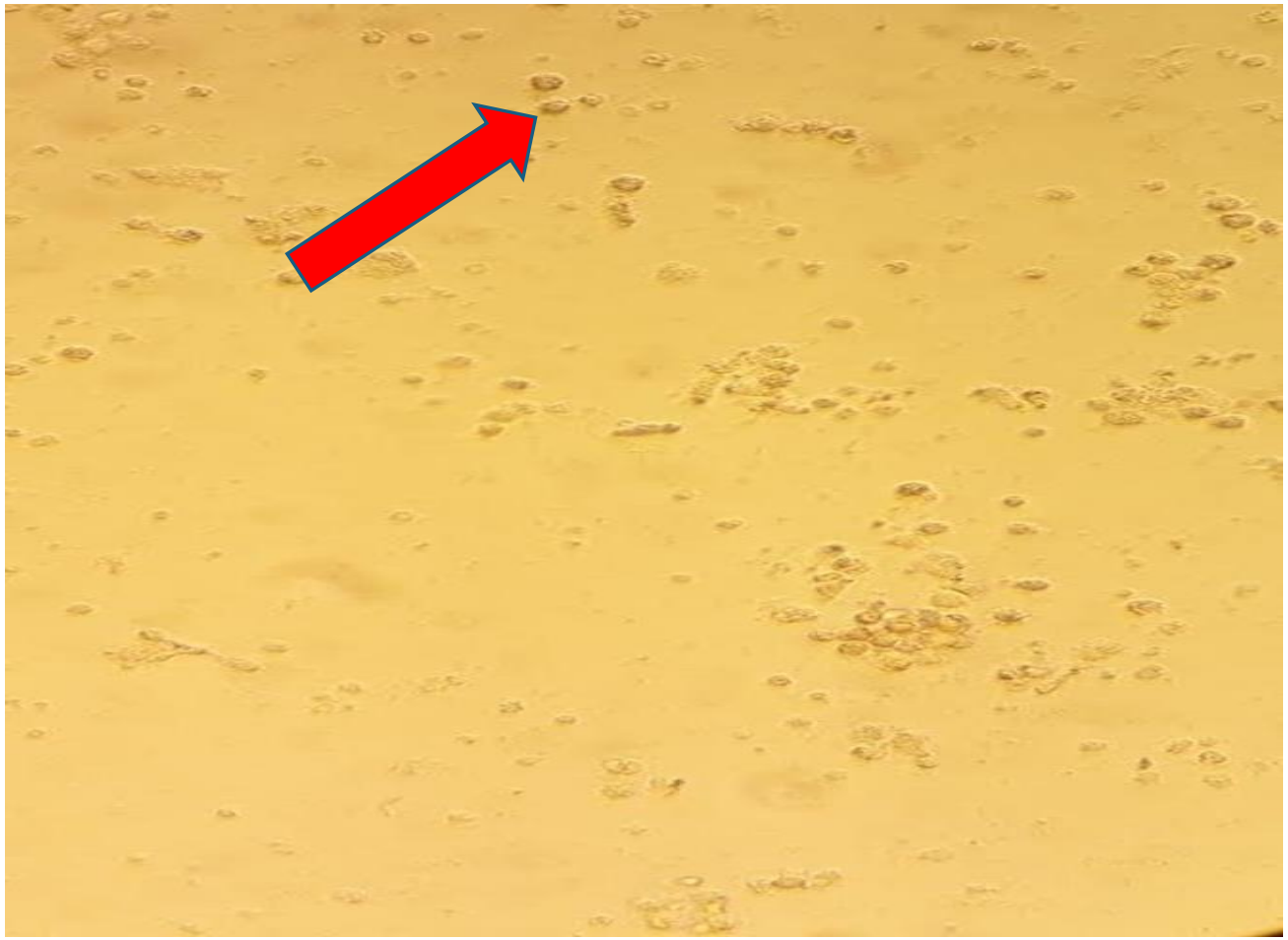


Control



Vitamin C at 40uM concentration

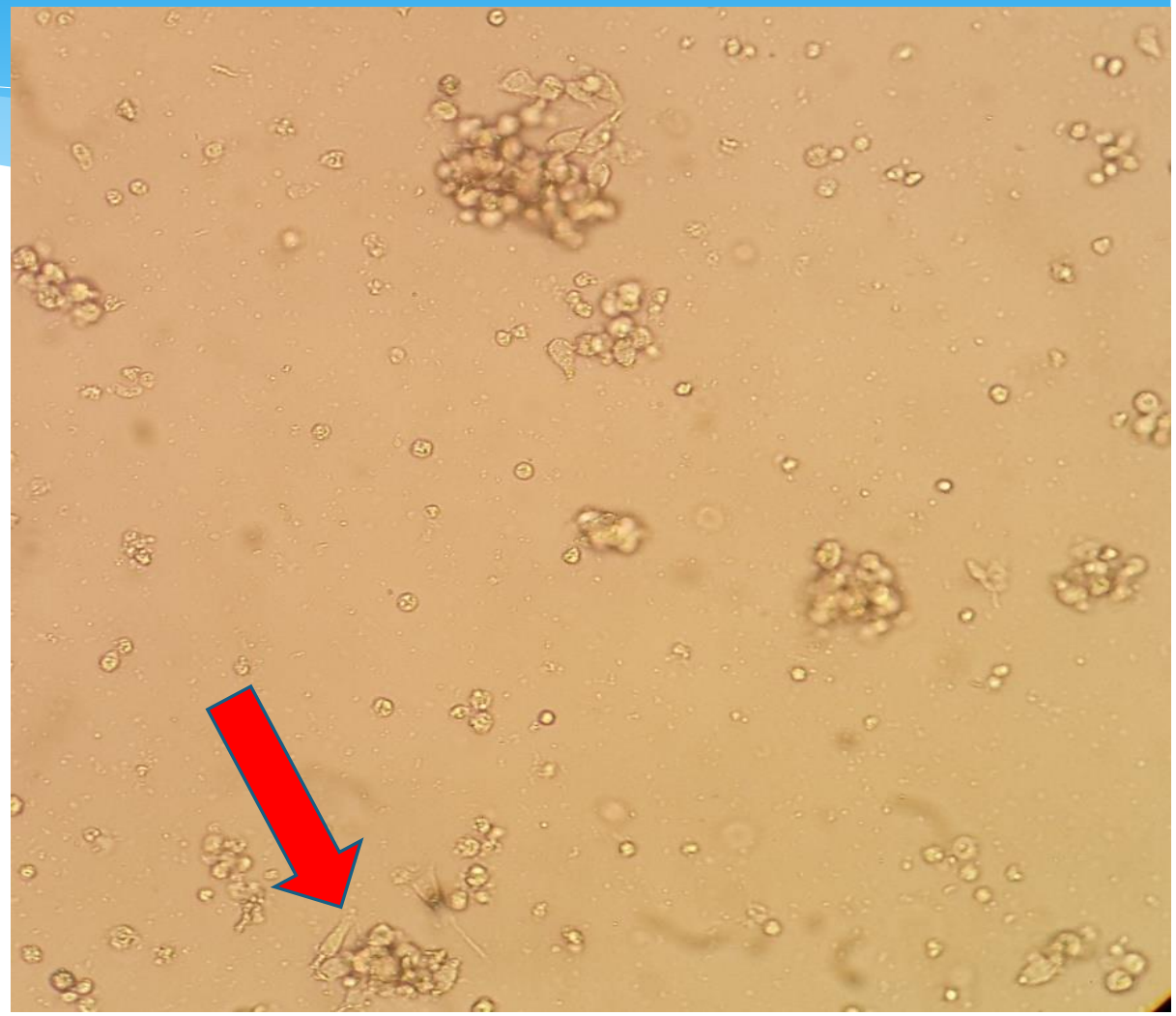
# Pictures of LNCaP cells 24hrs after treatment



Gen at 30uM + Vitamin C at 40uM

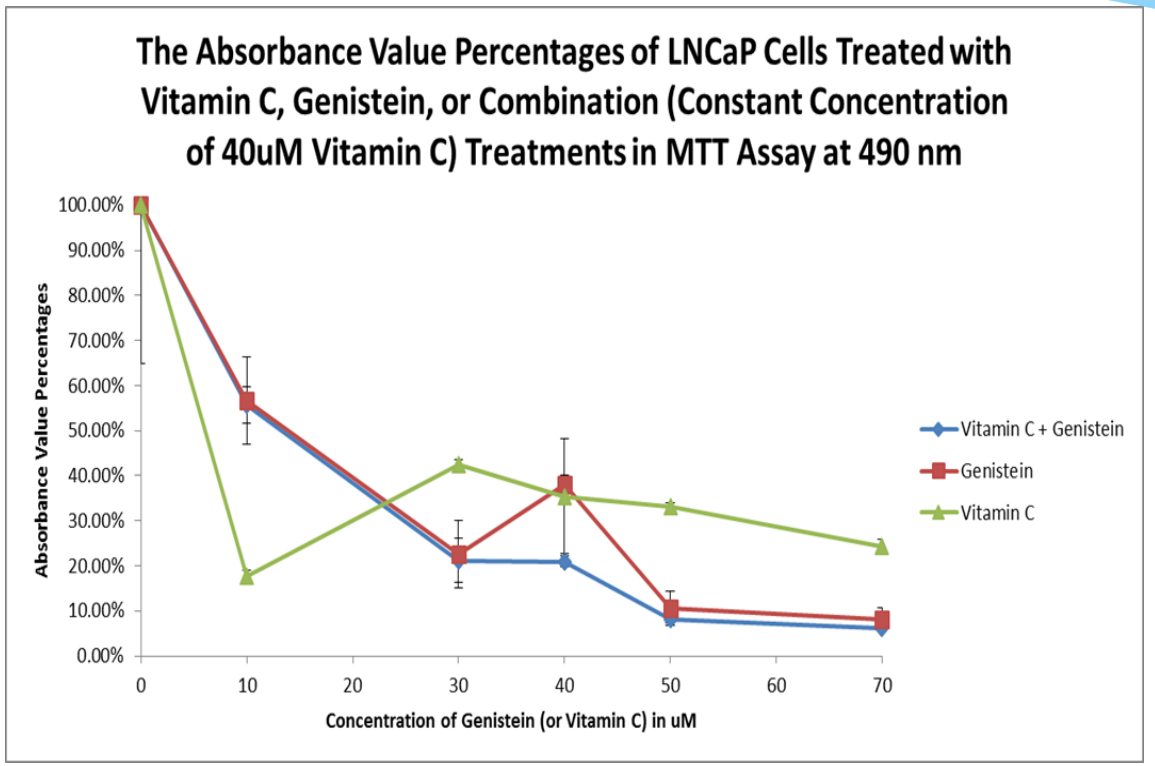
# Third set of result

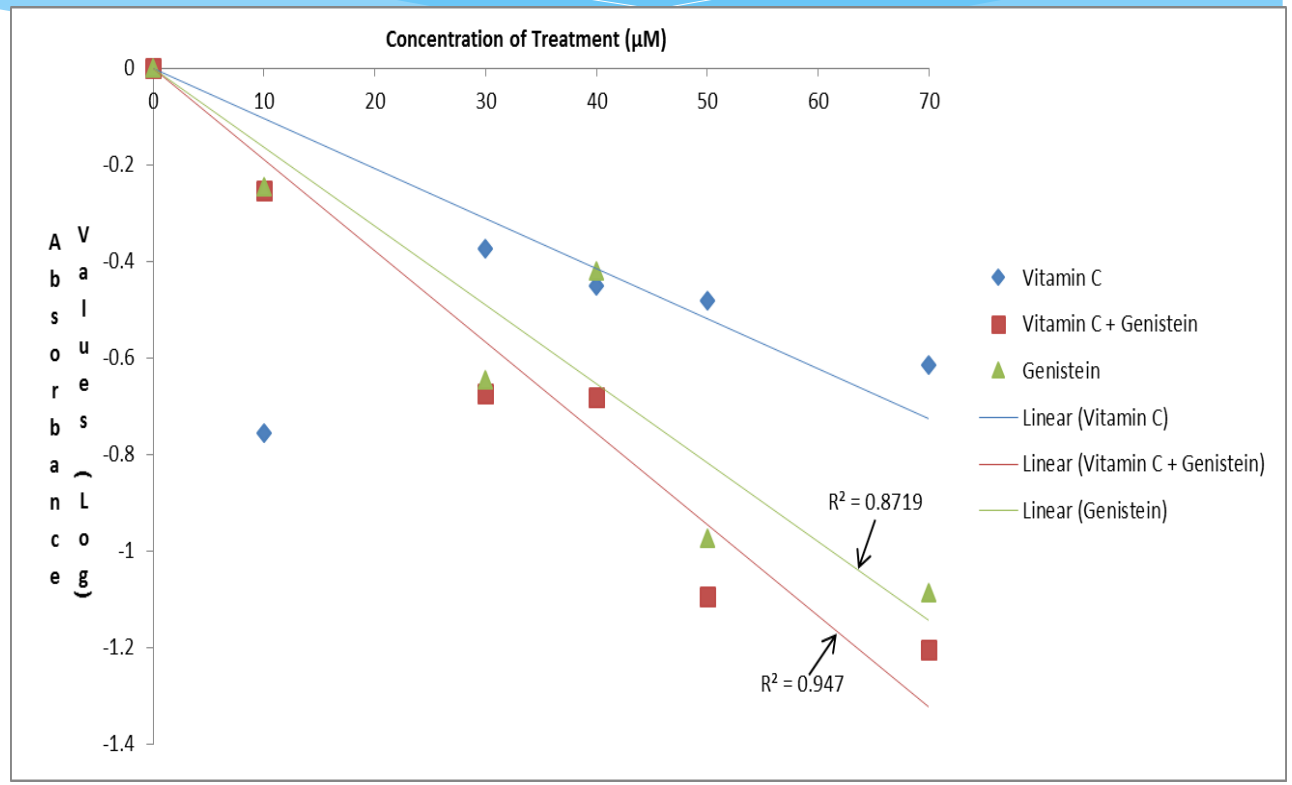
1st day of cell culture



4th day of cell culture

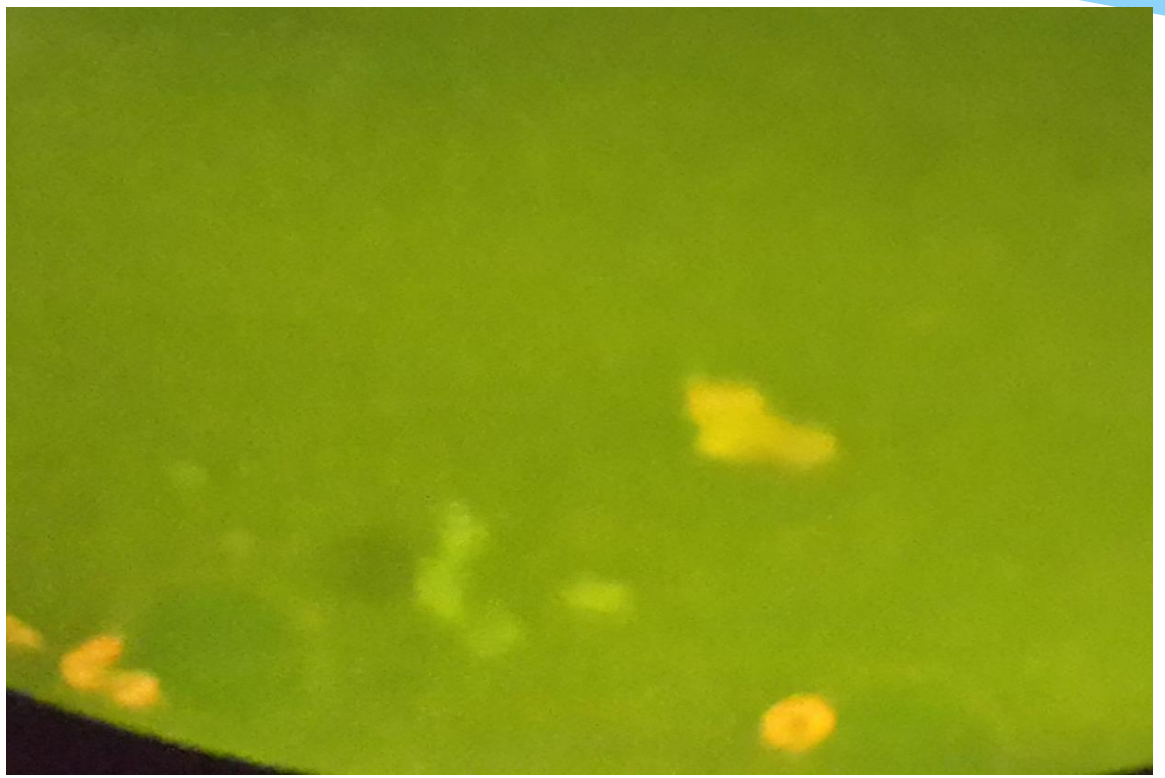
# MTT assay



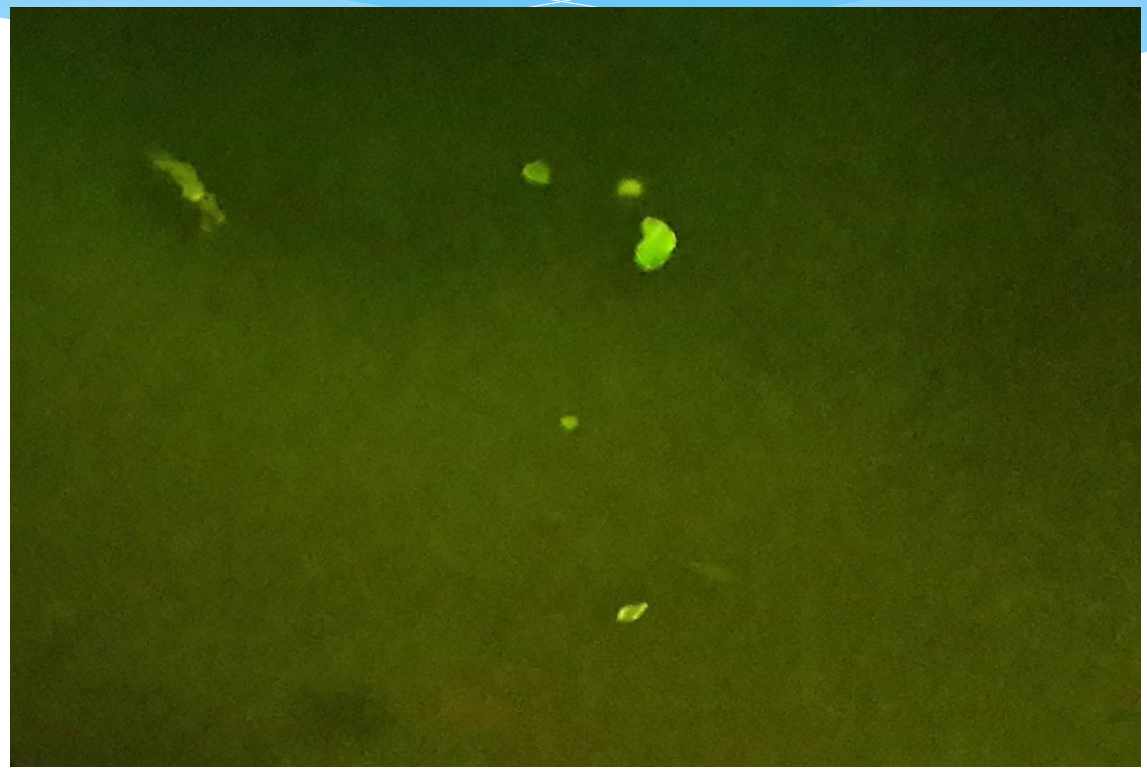




Level of significance = 0.05 and p value of t-test =0.11

# Fluorescence assay of combination treatment showing qualitative result

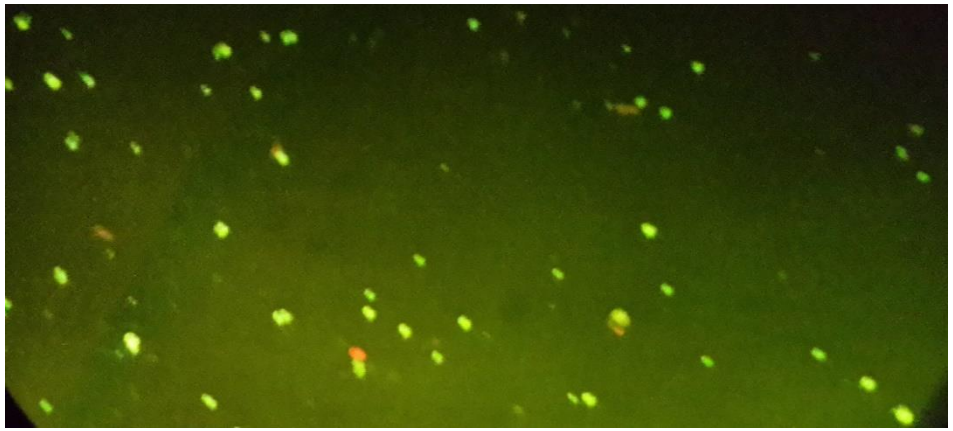


80uM genistein  + 70uM vitamin C

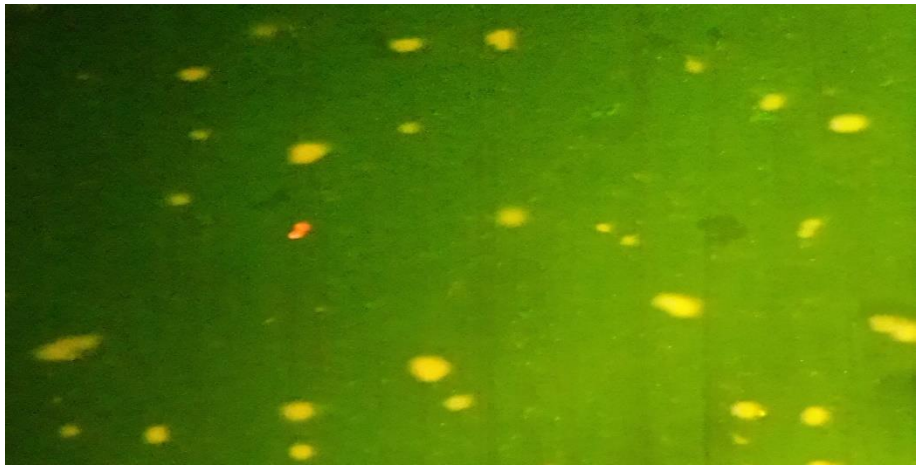


50uM genistein + 70uM vitamin C

# Quantitative result of florescence assay



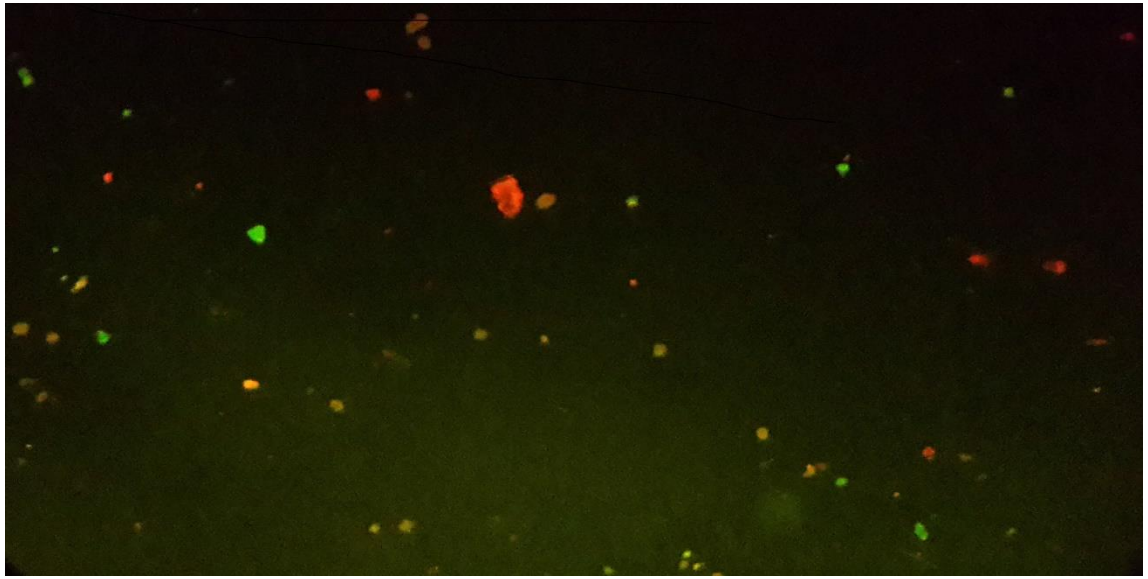
Genistein 10uM + vitamin C 40uM



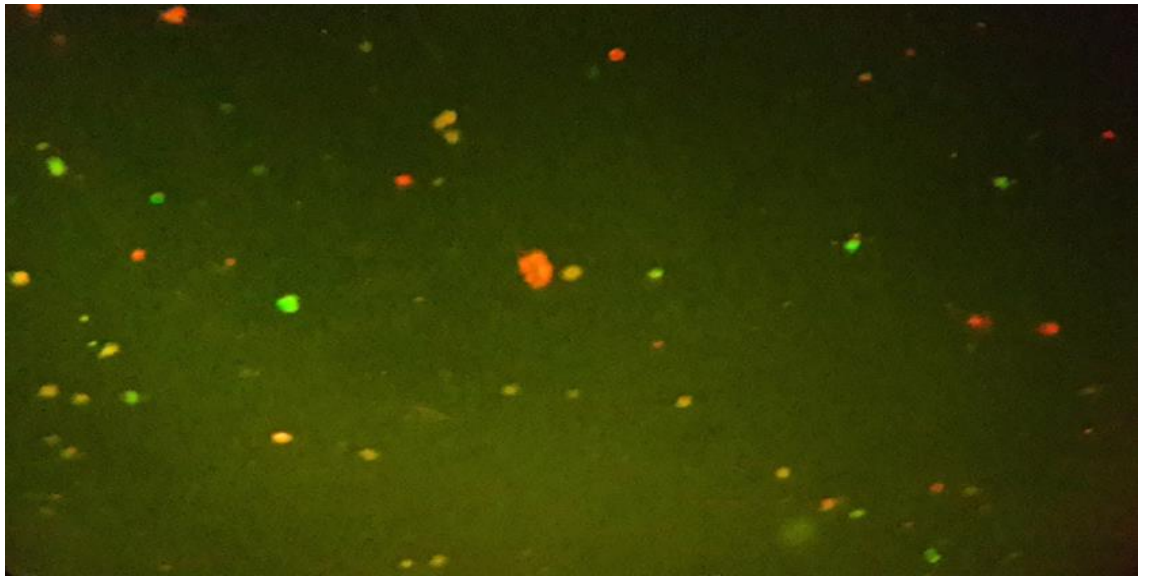
Genistein 70uM + vitamin C 40uM

Control

# Quantitative result of florescence assay

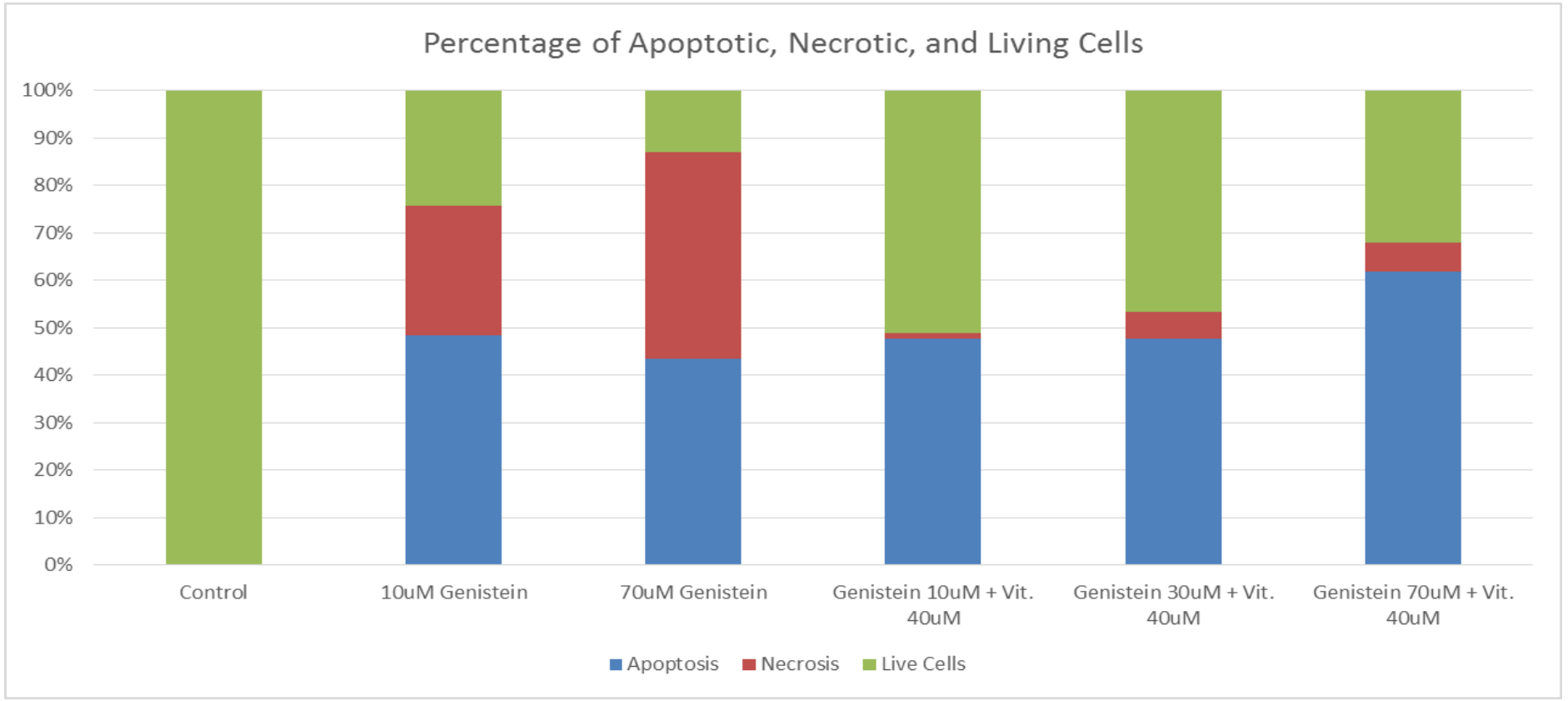


10uM genistein single treatment



70uM genistein single reatment

# Percentage of Apoptosis, Necrotic and Living cells in the treatment groups




P value =0.0003

Level of significance = 0.05

# Ratio of Apoptosis to Necrosis in each treatment

| Treatment | % Apoptosis | % Necrosis | % Apoptosis/ %Necrosis |
|---|---|---|---|
| Control | 0 | 0 | 0 |
| 10uM of genistein | 48 | 17 | 2.8= 3 |
| 70uM of genistein | 43 | 40 | 1.1= 1 |
| 10uM of genistein + 40uM of vitamin C | 48 | 2 | 24.8 =25 |
| 30uM of genistein + 40uM of vitamin C | 48 | 4 | 12 |
| 70uM of genistein + 40uM of vitamin C | 62 | 6 | 10.3= 10 |

# Conclusion

* P value of chi-square test = 0.0003 and the level of significance is 0.05
* Reject the null hypothesis
* Fluorescence assay shows different number of apoptotic death at different combination concentrations.
* Decrease in absorbance reflects a decreased level of intracellular ROS
* The order of effectiveness of treatment is (Gen 10uM + Vit C 40uM) > ( Gen 30uM + Vit C 40uM) >(Gen 70uM + Vit C 40uM) > > 10uM genistein only > 70uM genistein only.

# References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* American Cancer Society http://www.cancer.org/cancer/prostatecancer/detailedguide/prostate-cancer-key-statistics; 2015.
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* YangY, Karakhanova S,Werner J, Bazhin AV(2013) Reactive oxygen species in cancer biology and anticancer therapy. Curr med chem factor-1/chemokine (C-X-C motif) ligand 12 stimulates human hepatoma cell growth,migration, and invasion. Mol Cancer Res 5(1):21–33.

# Acknowledgement

Department of Biological Sciences of Florida Atlantic University


THANK
YOU!