Induction of oxidative stress in 3T3-L1 differentiated adipocyte cells

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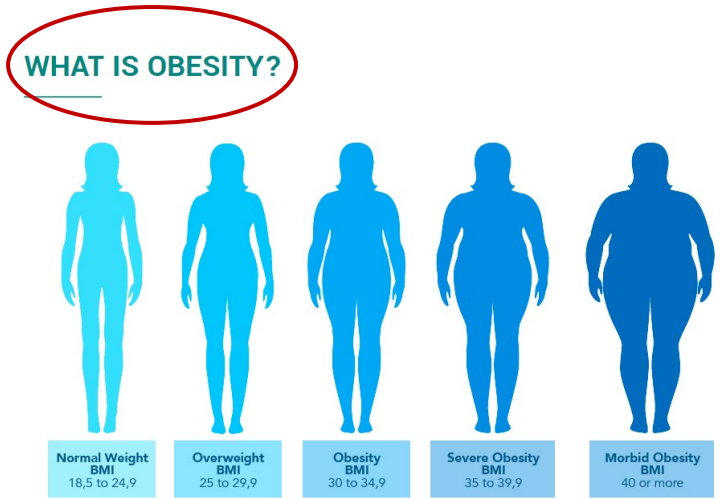
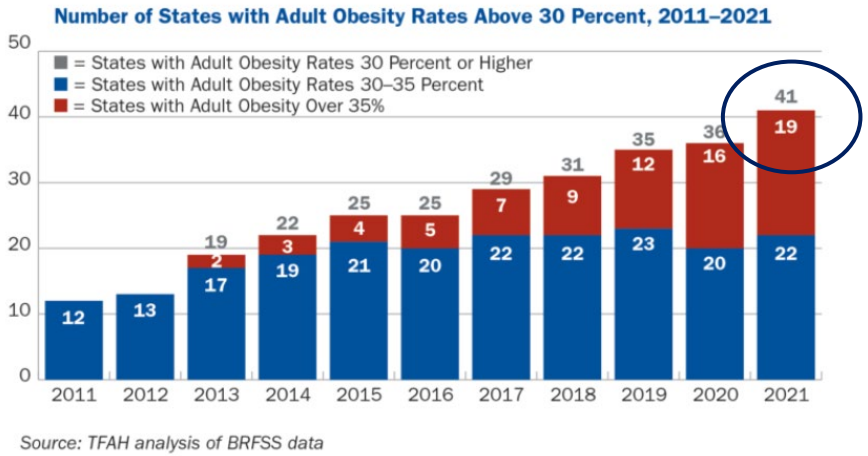
Background

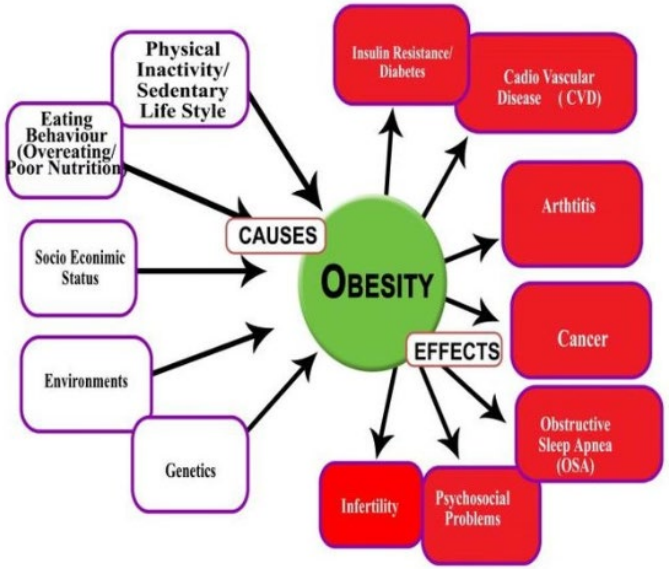
OBESITY: Prevalence, Cause, and Adverse effect.



Obesity is defined as abnormal or excessive fat accumulation.

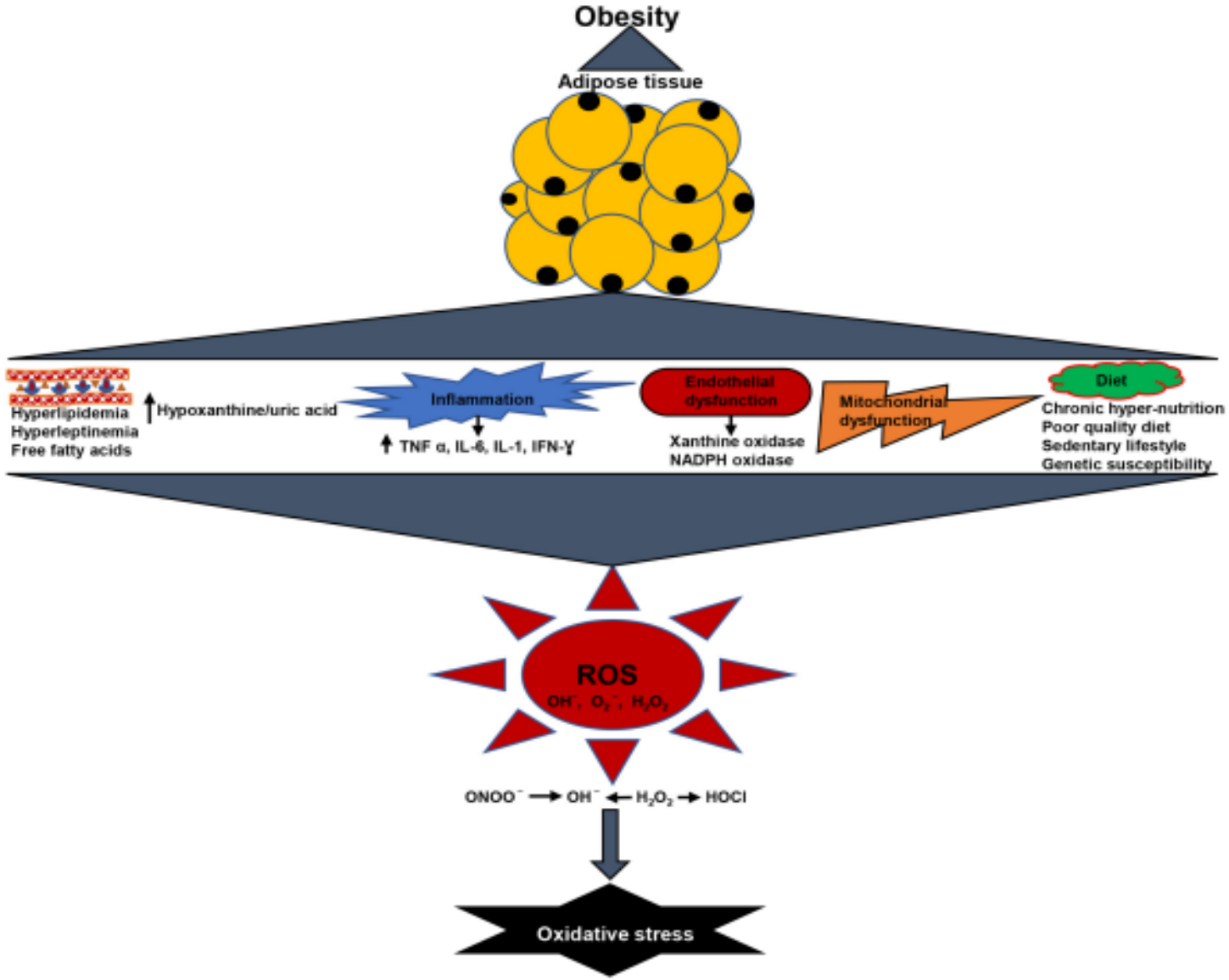


As of 2021, according to National Health and Nutrition Examination Survey in US shown that 41.9 percent of adults have obesity.



Source : Google

Pathway of oxidative stress generation in obesity



Free fatty acids (FFA) elevate the production of superoxide

Obesity induces the production of common inflammatory cytokine mediators

Increased muscle activity in obese individuals can result in an excess of free radicals

Aim & Hypothesis

AIM

To identify increase Oxidative Stress (OS) in 3T3-L1 differentiated adipocytes via the hydrogen peroxide treatment

HYPOTHESIS

The production of Reactive Oxygen Species (ROS) can induce increase in Oxidative Stress (OS), which may give insight into the metabolic condition of an obesity-related disease in an obese individual

Methodology

Differentiation of 3T3-L1 MBX clone to adipocyte cells

3T3-L1 Mouse embryonic fibroblast adipose clone

Cells seeded

Differentiated media (Day 0)

Insulin media (Day 3)

Growth media (Day 5)

Maintained with growth media

Fully differentiated adipocyte cells (Day 10)

Oxidative stress was induced in differentiated adipocytes

Fully differentiated adipocyte cells

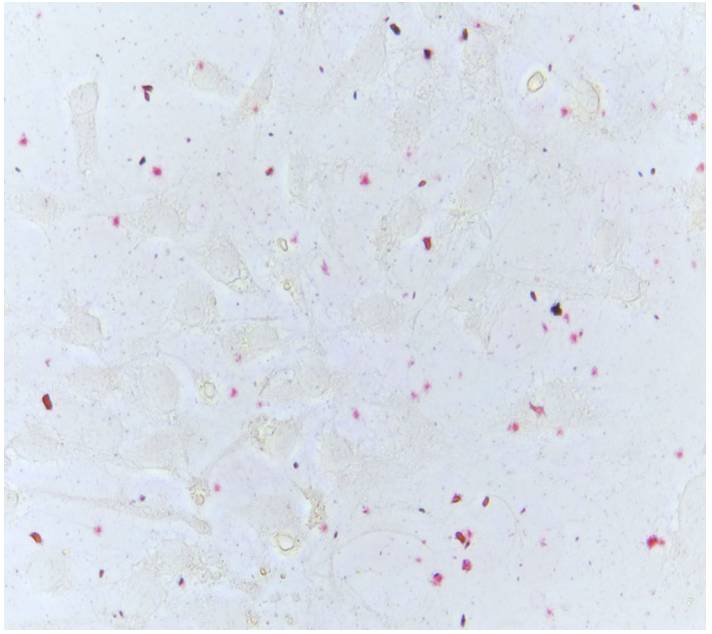
The cells were treated with fresh media containing diff conc. of H202 (0µM, 100µM, 300µM, and 500µM) and incubated for 12, and 24 hours.

Oxidative stress induced in differentiated adipocyte cells

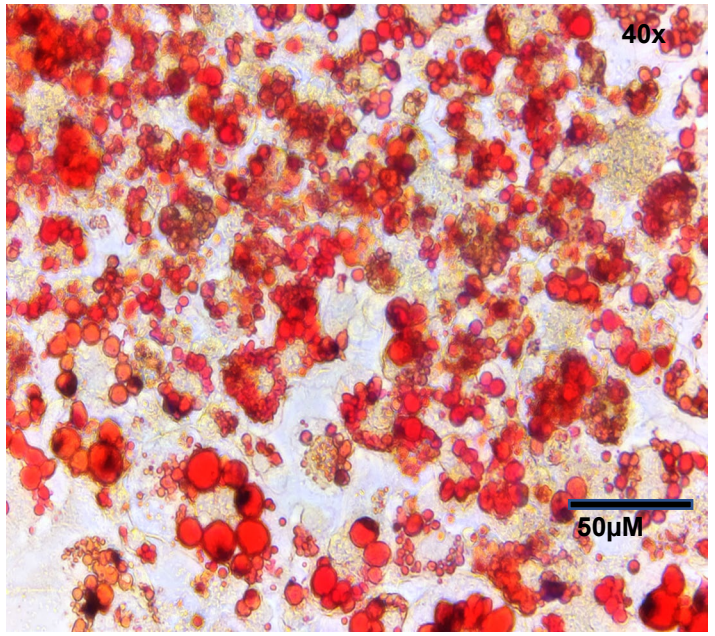
Oxidative stress assays

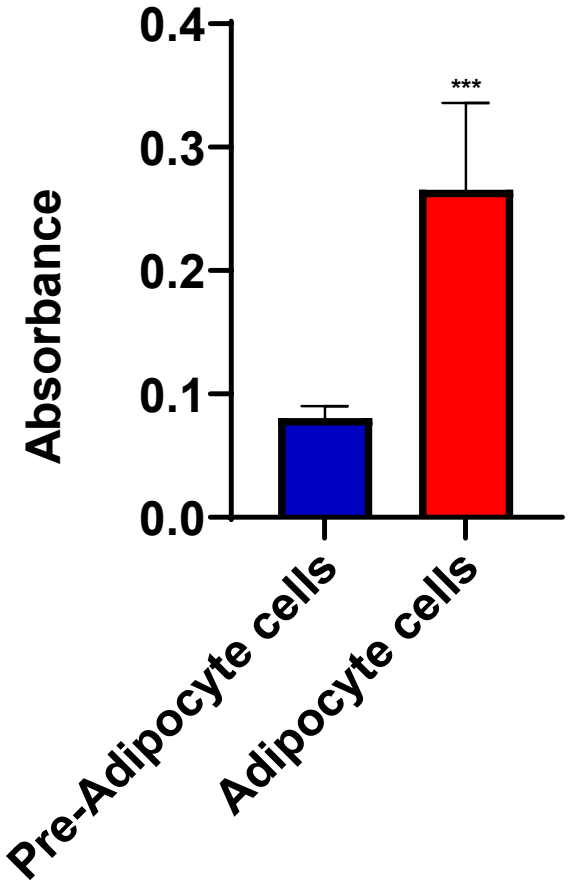
Obesity Adipocyte cells Model

Pre-Adipocyte cells



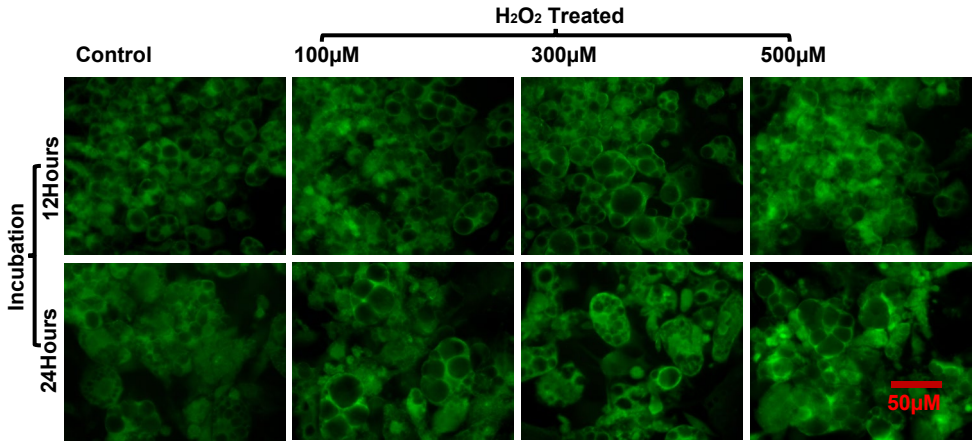
Differentiated Adipocyte cells

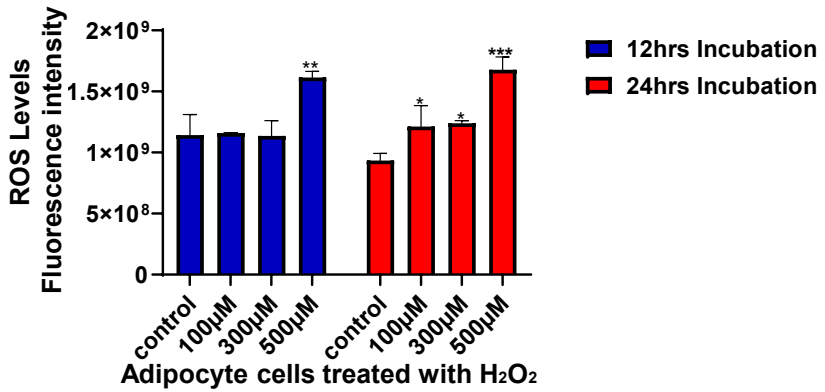




Significant level: *** p<0.001

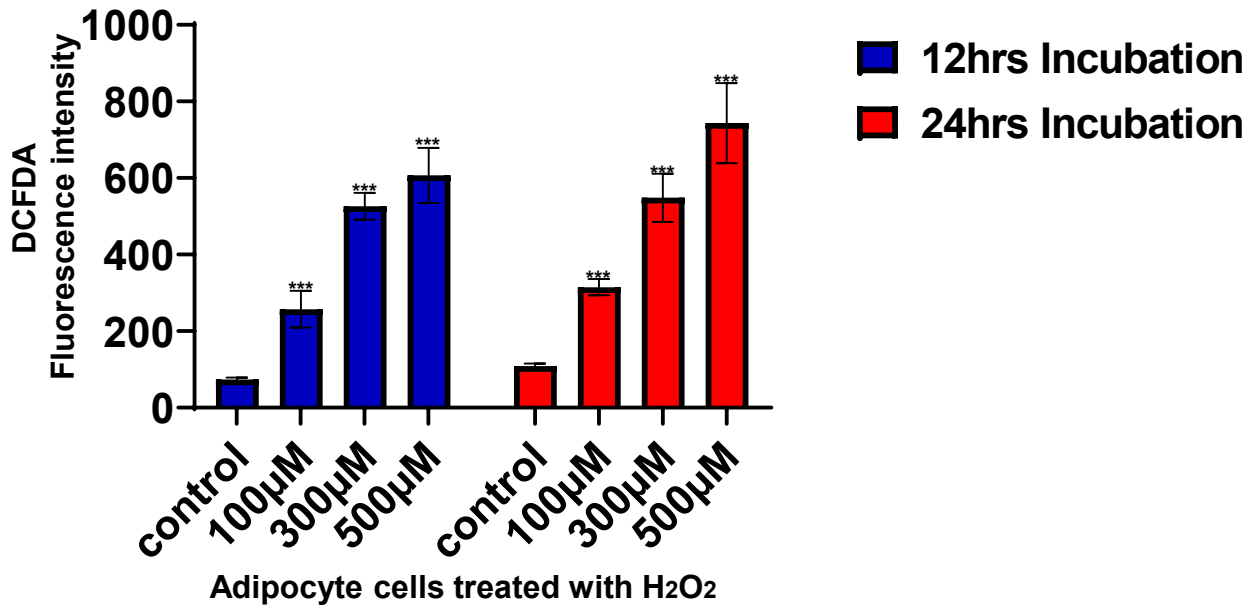
Obesity model - 3T3-L1 adipocytes ROS generation





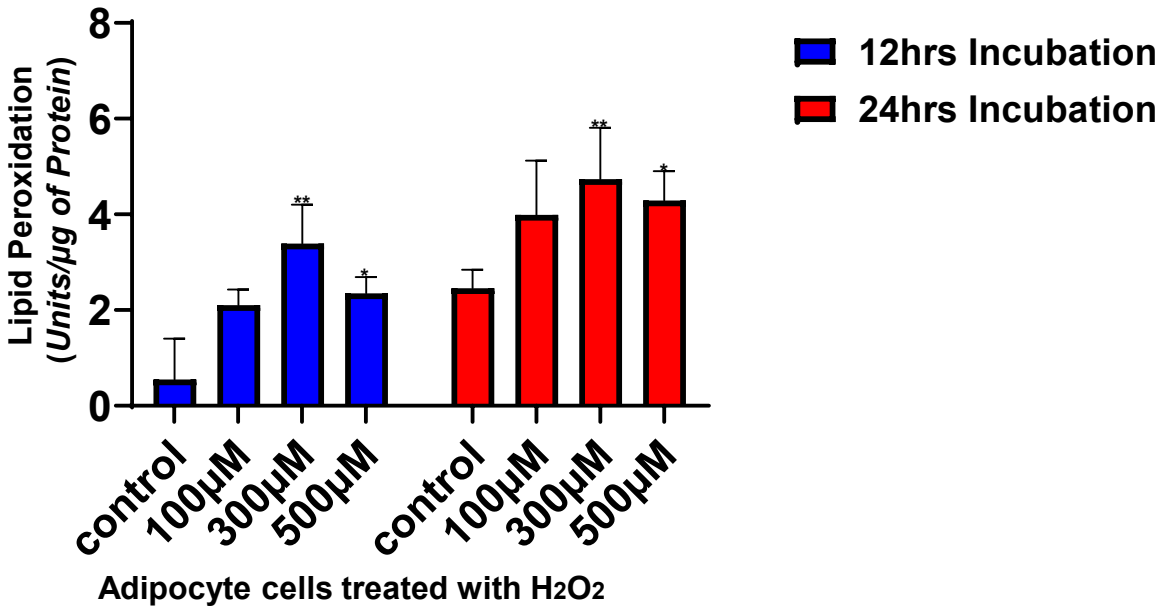
Significant level: * p<0.05; ** p<0.01; *** p<0.001

Obesity model - 3T3-L1 adipocytes ROS generation



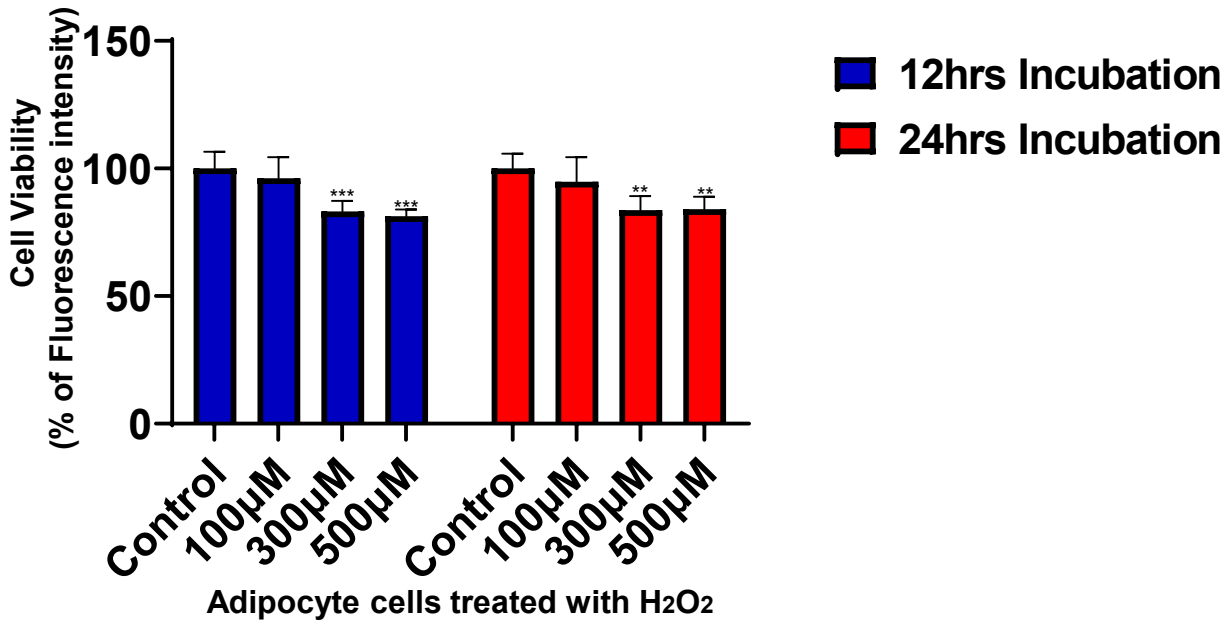
Significant level: *** p<0.001

Obesity model - 3T3-L1 adipocytes Lipid Peroxidation



Significant level: * p<0.05; ** p<0.01

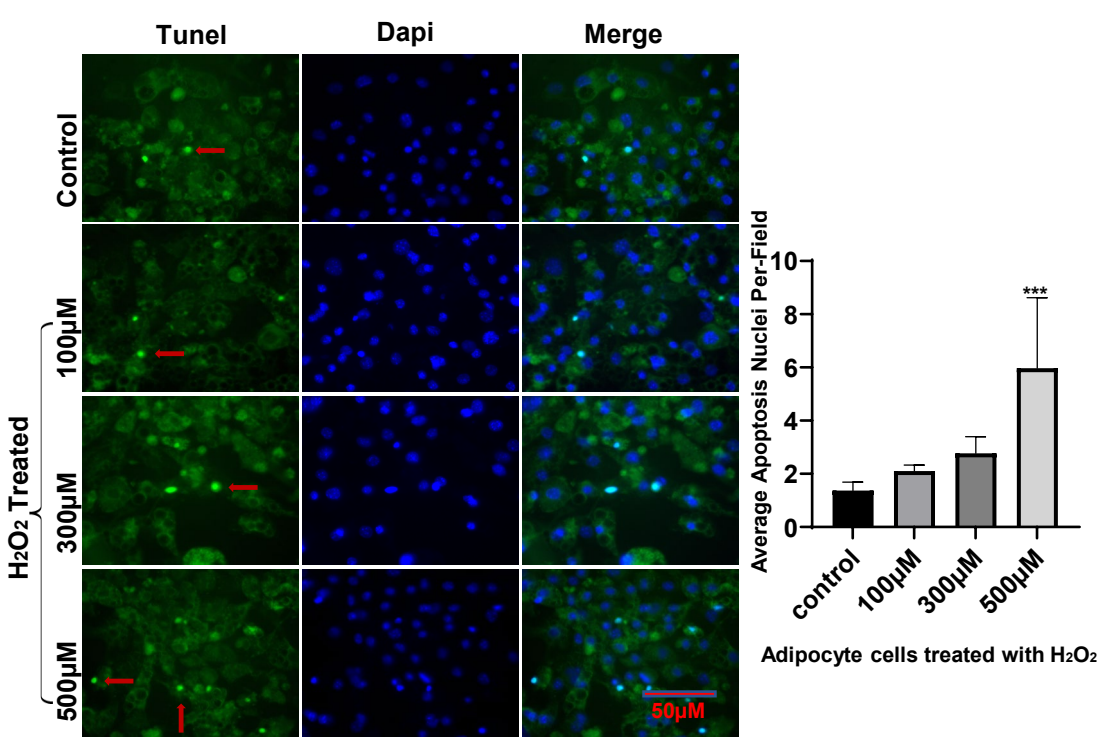
Obesity model - 3T3-L1 adipocytes Cell viability



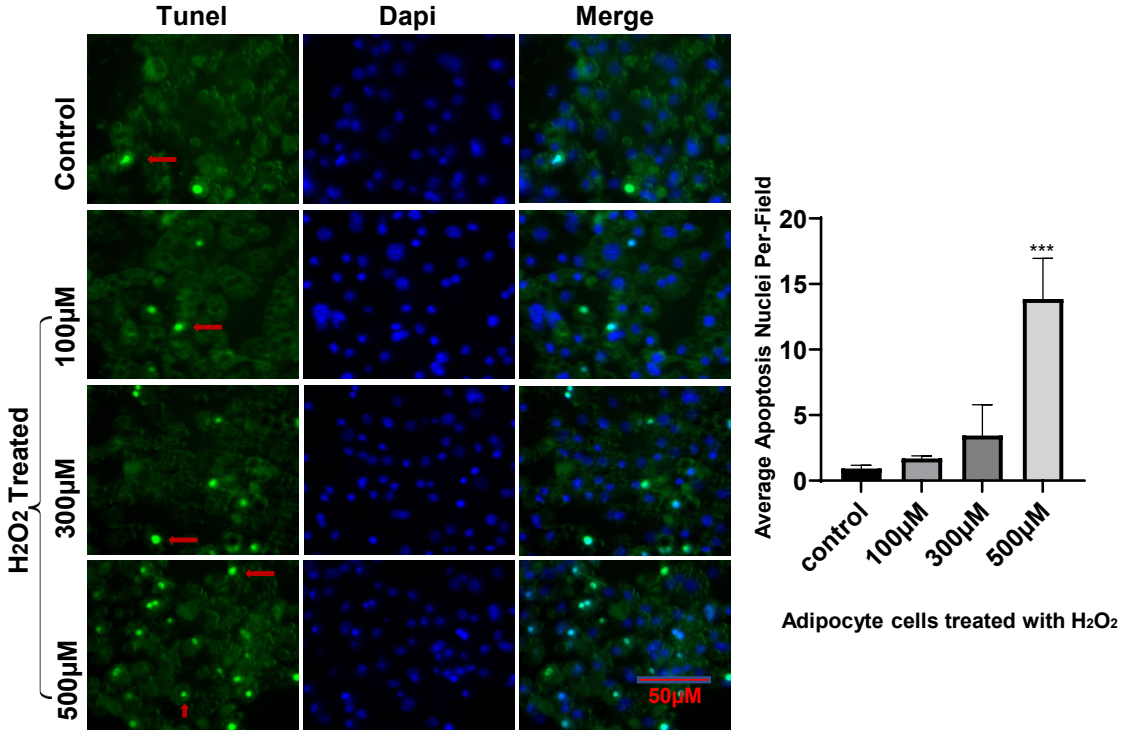
Significant level: ** p<0.01; *** p<0.001

Obesity model – H_2O_2 induced apoptosis in 3T3-L1 adipocytes

12 hours incubation



24 hours incubation

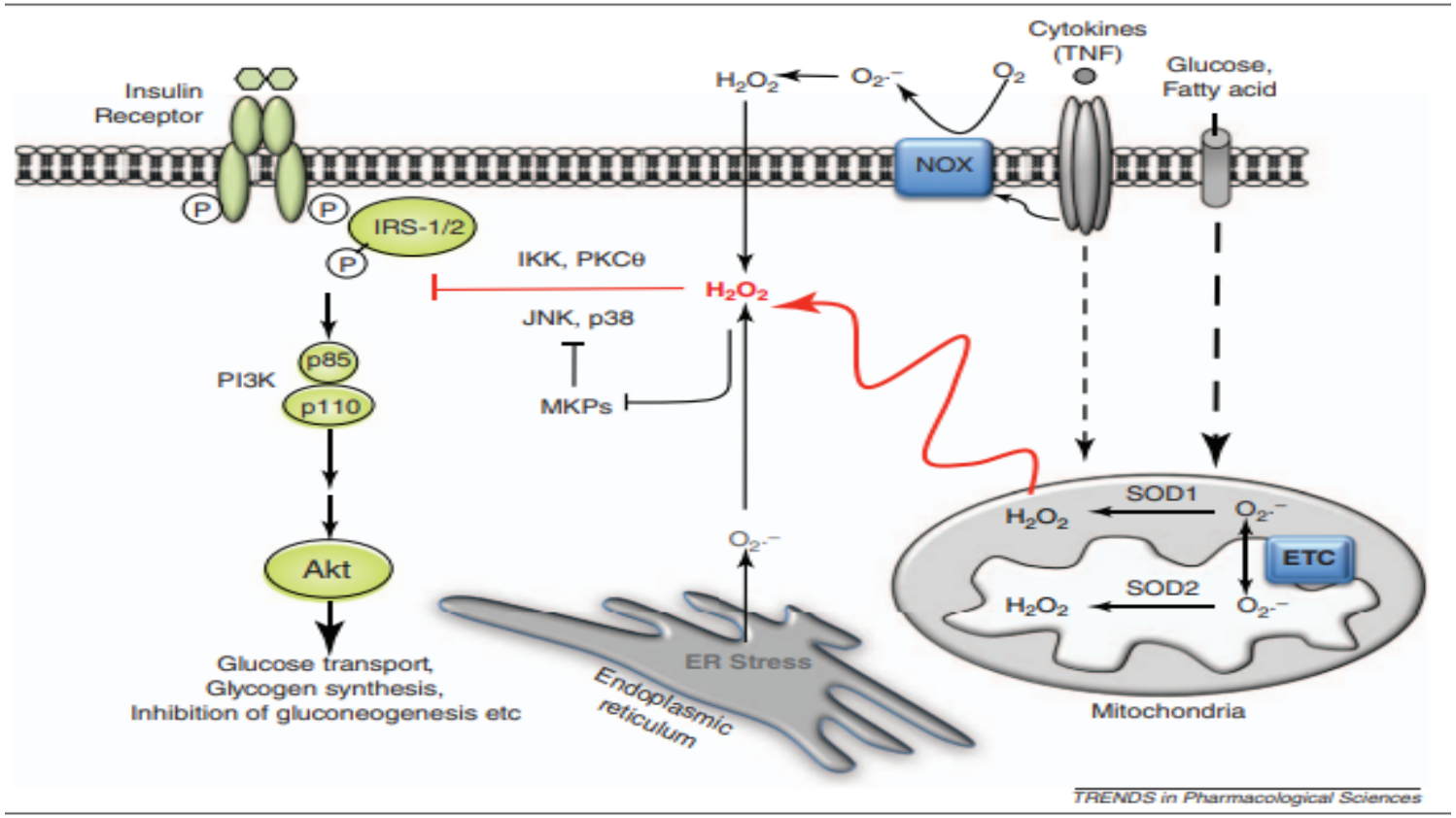


Significant level: *** p<0.001

Conclusion

In differentiated adipocytes cells; 300 µM, and 500 µM of H_2O_2 can produce substantial OS without altering the cell morphology

Oxidative Stress influences metabolic enzyme activity, transcription factor, and gene expression that can lead to type 2 diabetes and other obesity-related diseases



Future Direction

To identify the cellular Volatile Organic Compounds, release due to the increase in Oxidative Stress in differentiated 3T3-L1 adipocytes obese model which can serve as biomarker for obesity-related diseases

Questions?