Assessing DNA Repair Capacity of Hydrogen Peroxide-Induced Oxidative Damage Using *In Vitro* Comet Assay in 3T3 Cells

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OUR AIM

…to examine **DNA repair capacity** as a time interval in response to oxidative DNA damage induced by **H_2O_2** in the **3T3 cell line**

…to contribute to the **standardization of the *in vitro* comet assay** method

METHODS

1. Cell culture & DNA-damaging agent
2. Change the medium
3. DNA Repair
4. Embedding the cells in agarose
5. Lysis of cells
6. Washing
7. Alkaline treatment
8. Electrophoresis
9. Neutralization
10. Staining & Visualization

Maximum **DNA damage** caused by **H_2O_2** was determined **50 µM** at **30 minutes**.

DNA repair in 3T3 cells begins significantly from the **6th hour** following oxidative damage induced by **hydrogen peroxide**.

Take a picture to read more information!

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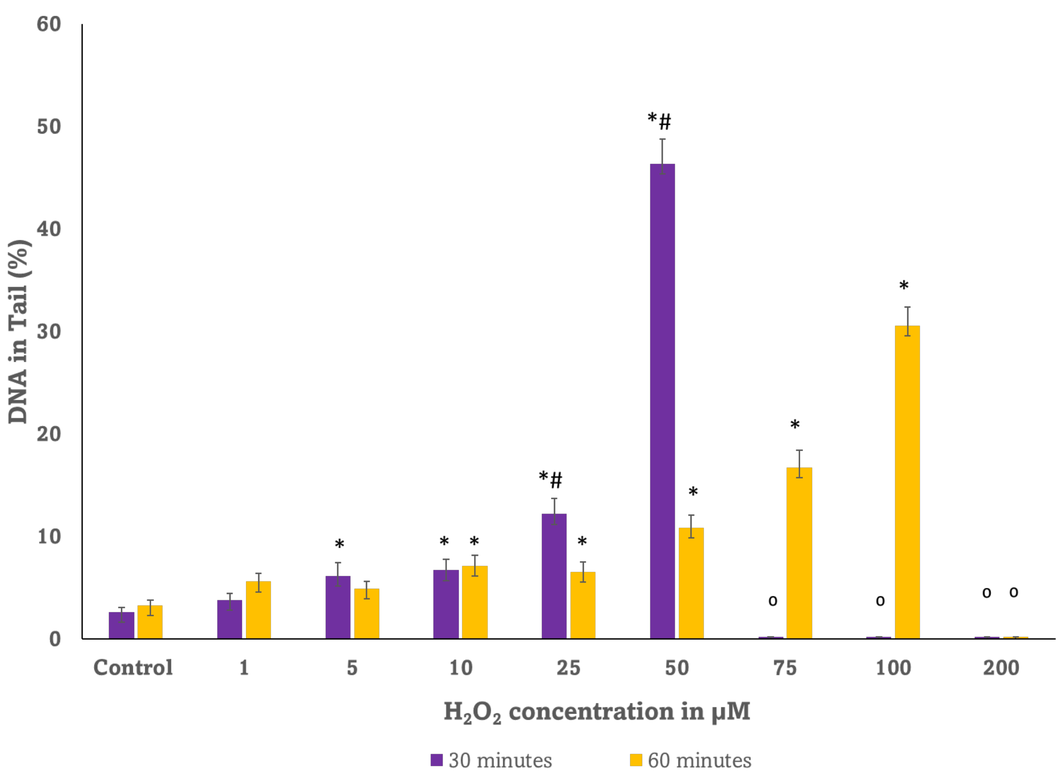

Figure 1. The mean tail intensity (n=100) ± SEM in 3T3 cells following exposures to H_2O_2 for 30 and 60 minutes.; *p<0.05, comparing the control and 30-minute results; *p<0.05, comparing the control and 60-minute results; #p<0.05, compared to results at 30 and 60 minutes (One-Way ANOVA). º Ghost cells

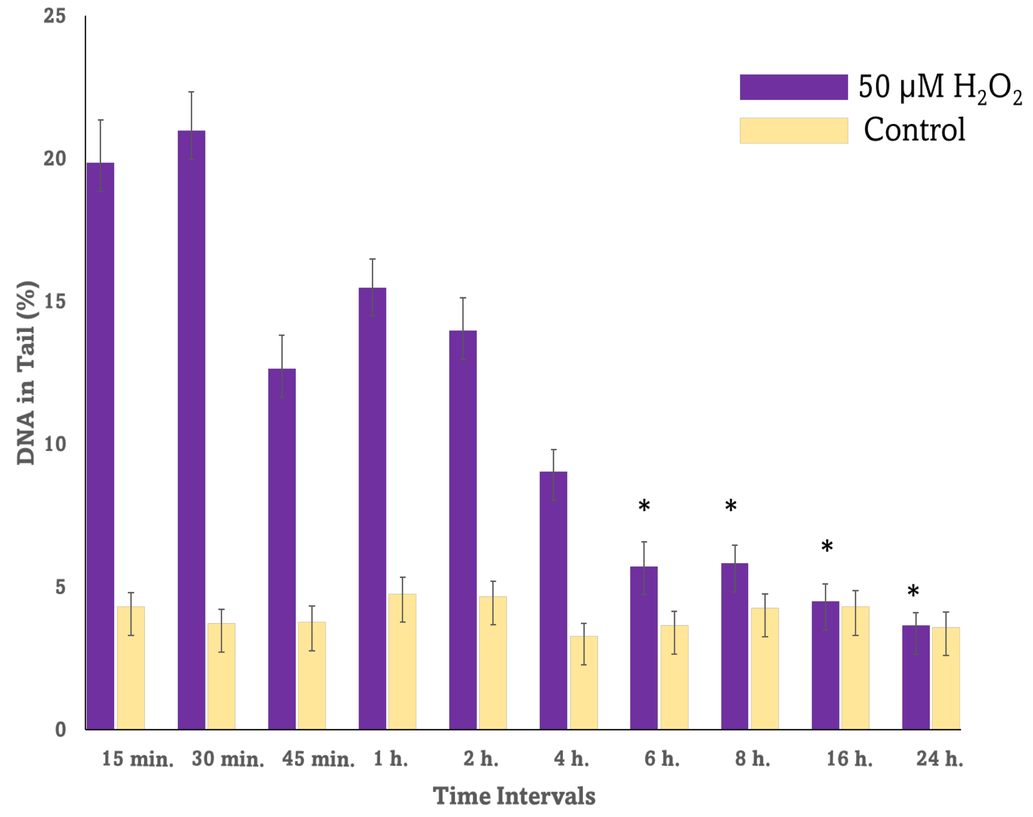

Figure 2. DNA strand breaks induced by 50 µM H_2O_2 over time in the alkaline comet assay. *p<0.05 vs control at the same point

DISCUSSION

The results of this study may contribute to valuable insights for future investigations, such as human biomonitoring studies, which are aimed at evaluating intra- and inter-individual variations in DNA repair capacity. Furthermore, the outcomes can serve as a meaningful tool to help researchers who **aim to standardize the in vitro alkaline comet assay**.

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