ABSTRACT

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Title of diploma thesis: Oxidative stress in human fibroblasts after exposure to titanium and graphene nanoparticles

Nanoparticles, or particles which have at least one dimension in the size range of 1-100 nm in size, have been studied extensively. Especially in recent years, the number of publications dealing with the toxic effects of nanoparticles has increased rapidly. One of the main mechanisms involved in the toxic effect of nanoparticles is the formation of reactive oxygen species and the associated oxidative stress.

This thesis aimed to determine the degree of oxidative stress in human fibroblasts after exposure to titanium and graphene nanoparticles, which correlates closely with the experimentally measured level of reduced glutathione in the affected cells. For this purpose, two optimized methods were used, namely the determination of glutathione by monochlorobimane and the determination of total and oxidized glutathione by enzymatic recycling reaction.

One part of the cells was treated with two forms of unmodified graphene nanoplates of different lateral sizes (80-300 and 250-400 nm) for 24 and 48 hours. For the tested graphene nanoparticles, the dependence of oxidative stress level on particle size, concentration, and incubation time was demonstrated.

The second part of the cells was affected by two forms of titanium anatase/rutile nanoparticles of similar size (~27 and 20-30 nm) and one form of rutile nanoparticles (4-6 nm) for 24 and 48 hours. No statistically significant dependence on the degree of oxidative stress was observed for the tested titanium nanoparticles. After 48 hours, an increase in the level of reduced glutathione was detected.

Based on the experimentally obtained data from three independent measurements, statistical analysis was performed, and the two optimized methods were compared. A significant correlation was found between the methods used, which gave comparable values of reduced glutathione concentrations in the samples.

Key words:

Nanoparticles, TiO_2, graphene, oxidative stress, glutathione, enzymatic recycling method, monochlorobimane