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Counteracting the Effect of Bacterial Lipopolysaccharide on Dental Pulp Stem-Cells

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Objectives Drug-loaded non-resorbable polymeric nanoparticles (NPs) are proposed as an adjunctive treatment for pulp regenerative strategies. The present *in vitro* investigation aimed to evaluate the effect of NPs loaded with tideglusib (TDg-NPs) on the viability, morphology, migration, differentiation and mineralization potential of human dental pulp stem cells (hDPSCs) in the presence of bacterial lipopolysaccharide endotoxin (LPS).

Methods Cell viability, proliferation, and differentiation were assessed using a MTT-based assay; cell migration evaluation, cell cytoskeleton staining analysis, Alizarin Red S staining and expression of the odontogenic related genes by a real-time quantitative polymerase chain reaction (RT-qPCR) were also performed. Cells were tested with and without previous stimulation with LPS at different time points. One-way ANOVA and Tukey's test were employed for statistical analysis ($p<0.05$).

Results Adequate cell viability was encountered in all groups and at every tested time point (24, 48, 72 and 168 h), without significant differences among the tested groups ($p>0.05$). The analysis of cell cytoskeleton showed some nuclear alteration in cultures with undoped NPs after LPS stimulation. These cells exhibited an in blue diffuse and multifocal appearance. Some nuclei looked fragmented and condensed. hDPSCs after LPS stimulation but in the presence of TDg-NPs evidenced less nuclei changes. LPS induced down-regulation of alkaline phosphatase, osteonectin and collagen1 gene markers, after 21d. LPS significantly half-reduced the cells production of calcium deposits in all groups ($p<0.05$), except in the group in which hDPSCs were cultured with TDg-NPs where the decrease was only about 10%.

Conclusions LPS induced two effects in cultured hDPSCs, *i)* lower mineral deposition and *ii)* lower F-actin fibers content together with cytoskeletal disorganization and nuclei alterations. These effects were counteracted by TDg-NPs. Grant PID2020-114694RB-I00 and Grant PID2020–115887GB-I00 funded by MCIN/AEI 10.13039/501100011033.