

Nanohydroxyapatite (nHAp) doped with iron oxide nanoparticles (IO), miR-21 and miR-124 under magnetic field conditions modulates osteoblast viability, reduces inflammation and inhibits the growth of osteoclast – A novel concept for osteoporosis treatment: Part 1.

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Rok wydania

2021

CzasopismoInternational Journal of
NanomedicineNumer woluminu

Volume 16

Strony

3429-3456

DOI

10.2147/ijn.s303412

Kolekcja

Naukowa

Streszczenie

Purpose: Osteoporosis results in a severe decrease in the life quality of many people worldwide. The latest data shows that the number of osteoporotic fractures is becoming an increasing international health service problem. Therefore, a new kind of controllable treatment methods for osteoporotic fractures is extensively desired. For that reason, we have manufactured and evaluated nanohydroxyapatite (nHAp)-based composite co-doped with iron oxide (IO) nanoparticles. The biomaterial was used as a matrix for the controlled delivery of miR-21-5p and miR-124-3p, which have a proven impact on bone cell metabolism.

Methods: The nanocomposite $\text{Ca}_5(\text{PO}_4)_3\text{OH}/\text{Fe}_3\text{O}_4$ (later called nHAp/IO) was obtained by the wet chemistry method and functionalised with microRNAs (nHAp/IO@miR-21/124). Its physicochemical characterization was performed using XRPD, FT-IR, SEM-EDS and HRTEM and SAED methods. The modulatory effect of the composite was tested in vitro using murine pre-osteoblasts MC3T3-E1 and pre-osteoclasts 4B12. Moreover, the anti-inflammatory effects of biomaterial were analysed using a model of LPS-treated murine macrophages RAW 264.7. We have analysed the cells' viability, mitochondria membrane potential and oxidative stress under magnetic field (MF+) and without (MF-). Moreover, the results were supplemented with RT-qPCR and Western blot assays to evaluate the expression profile for master regulators of bone metabolism.

Results: The results indicated pro-osteogenic effects of nHAp/IO@miR-21/124 composite enhanced by exposure to MF. The enhanced osteogenesis guided by nHAp/IO@miR-21/124 presence was associated with increased metabolism of progenitor cells and activation of osteogenic markers (*Runx-2*,

Język

Angielski

Typ publikacji

Artykuł

Opn, *Coll-1*). Simultaneously, nanocomposite decreased metabolism and differentiation of pre-osteoclastic 4B12 cells accompanied by reduced expression of *Cal* and *Ctsk*. Obtained composite regulated viability of bone progenitor cells and showed immunomodulatory properties inhibiting the expression of inflammatory markers, ie, *TNF- α* , *iNOs* or *IL-1 β* , in LPS-stimulated RAW 264.7 cells.

Conclusion: We have described for the first time a new concept of osteoporosis treatment based on nHAp/IO@miR-21/124 application. Obtained results indicated that fabricated nanocomposite might impact proper regeneration of osteoporotic bone, restoring the balance between osteoblasts and osteoclast.

Słowa kluczowe

nanocomposites, hydroxyapatite, osteoblasts, osteoclasts, osteoporosis

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Adres publiczny

<http://dx.doi.org/10.2147/ijn.s303412>

Strona internetowa wydawcy

<http://www.dovepress.com/international-journal-of-nanomedicine-journal>