The effects of mineral microparticles on dental cell differentiation

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Introduction

With the growth of regenerative medicine, the use of natural tooth fillers that can stimulate host tissue growth has become more popular. Recent research delves into effects of adding apatite minerals to osteogenic cells to stimulate bone growth.

Materials and Methods

Cells studied: bone forming cells (7F2 Osteoblasts), bone marrow stem cells (BMSCs), and dental pulp stem cells (DPSCs).

Standard growth media: AlphaMEM with 15% FBS and 5% pen/strep.

Mineral microparticles: Hydroxyapatite (HA) and Fluorapatite (FA).

Determined total protein content by running bicinchoninic acid (BCA) assays.

Determined ALP specific activity by running alkaline phosphatase assays (ALP). ALP is an early marker of mineral cell differentiation.

Use Alizarin Red staining to find areas of calcification.

Used antibody stains to determine the presence of osteocalcin and collagen I.

Examined cell-particle interactions.

Results

HA (hydroxyapatite) cells had higher levels of osteocalcin and collagen I in the first five days of culture than did both FA (fluorapatite) and control cells. DPSCs responded best to being cultured with microparticles (Figure 1). Cells cultured with HA had higher levels of ALP in the first 5 days than did the others, but after 5 days the presence of either type of microparticle hindered ALP specific activity.

Cells tended to congregate in large clusters around HA particles (Figure 2), but in smaller groups around small clusters of FA particles. It was not uncommon to see one or two cells next to a FA particle.

Discussion and Conclusions

Cells reacted better to being cultured with HA particles than FA particles (Fig. 2).

The data showed that HA stimulated protein and ALP levels in the first 5 days, then inhibited levels after. HA cell cultures showed higher levels of ALP specific activity (Fig. 1), osteocalcin, and collagen I than FA.

Though the antibody stain results were scattered, they still followed the trend of HA cells having higher levels of protein.

Overall, the results show a lower differentiation potential for cells that were exposed to mineral microparticles. This was an unexpected result and contradicts data shown in the literature. While it is known that the chemical compositions of the particles have an effect of cell differentiation potential, the effect of the size of the particles is not clear. Our particles were on a much larger scale than in previously performed experiments which may have affected cell differentiation.

Current and Future Work

Currently performing polymerase chain reaction assays for dentin sialophosphoprotein (DSPP) and secreted phosphoprotein (SPP) recognition to further quantify cell differentiation.

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Selected References
