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# Plasma Treatment in Conjunction with EGM-2 Medium Can Enhance Endothelial and Osteogenic Marker Expressions of Bone Marrow MSCs

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**Abstract :** For many tissue engineering applications, an important goal is to create functional tissues in-vitro, and such tissues to be viable, they have to be vascularized. Endothelial cells (EC) and endothelial progenitor cells (EPC) are promising candidates for vascularization. However, both of them have limited expansion capacity and autologous cells currently do not exist for either ECs or EPCs. Therefore, we use bone marrow mesenchymal stem cells (MSC) as a source material for ECs. Growth supplements are commonly used to induce MSC differentiation, and further improvements in differentiation conditions can be made by modifying the cell's growth environment. An example is pre-treatment of the growth dish with gas plasma, in order to modify the surface functional groups of the material that the cells are seeded on. In this work, we compare the effects of different gas plasmas on the growth and differentiation of MSCs. We treat the dish with different plasmas (CO2, N2, and O2) and then induce MSC differentiation with endothelial growth medium-2 (EGM-2). We find that EGM-2 by itself upregulates EC marker CD31 mRNA expression, but not VEGFR2, CD34, or vWF. However, these additional EC marker expressions were increased for cells seeded on plasma treated substrates. Specifically, for EC markers, we found that N2 plasma treatment upregulated CD31 and VEGFR-2 mRNA expressions; CO2 plasma treatment upregulated CD34 and vWF mRNA expressions. The osteogenic markers ALP and osteopontin mRNA expressions were markedly enhanced on all plasma-treated dishes. We also found that plasma treatment in conjunction with EGM-2 growth medium can enhance MSCs differentiation into endothelial-like cells and osteogenic-like cells. Our work shows that the effect of the growth medium (EGM-2) on MSCs differentiation is influenced by the plasma modified surface chemistry of the substrate. In conclusion, plasma surface modification can enhance EGM-2 effectiveness and induced both endothelial and osteogenic differentiation. Our findings provide a method to enhance EGM-2 based cell differentiation, with consequences for tissue engineering and stem cell biology applications.

**Keywords :** endothelial differentiation, EGM-2, osteogenesis, plasma treatment, surface modification
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