Improved Adhesion and Growth of Human Osteoblast-like MG 63 Cells on Biomaterials Modified with Carbon Nanoparticles

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In recent years, carbon nanoparticles, such as fullerenes, nanotubes and nanodiamonds, are considered as promising materials for biomedical applications, such as drug or gene delivery, photodynamic therapy against tumors and infectious agents, quenching oxygen radicals, biosensor technology or simulation of cellular components, such as membrane pores or ion channels [1-4]. Despite of these exciting perspectives, relatively little is known about the influence of carbon nanoparticles present on the biomaterial surface on the adhesion and growth of cells. Therefore, in the first set of experiments, a layer of fullerenes C_{60} was deposited on carbon fibre-reinforced carbon composites (CFRC), i.e. materials promising for hard tissue surgery [5], and seeded with human osteoblast-like MG 63 cells. On day 2 after seeding, the cells adhered to these surfaces in lower numbers (19222 ± 664 cells/cm^2) than on the control uncoated material and tissue culture polystyrene (44286 ± 4155 cells/cm^2 and 67276 ± 7287 cells/cm^2, respectively), which was probably due to a relatively high hydrophobicity of fullerenes. On the other hand, the spreading area of cells on the fullerene-coated samples amounted to 3182 ± 670 um^2, while on both control surfaces, it was only 1888 ± 400 and 1300 ± 102 um^2. These cells also assembled numerous dot-like vinculin-containing focal adhesion plaques and a rich fine mesh-like beta-actin cytoskeleton. Similar results were obtained in the second set of experiments, performed on a terpolymer of polypropylene, polytetrafluorethylene and polyvinyldifluoride mixed with 4 wt.% of single- or multi-walled carbon nanotubes (SWCNT or MWCNT, respectively). The MG 63 cells on these materials were well spread, polygonal and contained distinct beta-actin filament bundles, whereas most cells on the pure terpolymer were round and clustered into aggregates. Enzyme-linked immunosorbent assay (ELISA) revealed that the cells on the material with SWCNT contained in average a higher concentration of vinculin and talin, i.e. components of focal adhesion plaques (by 90 and 40%, respectively), in comparison with cells on the pure terpolymer. In addition, the cells on the samples with MWCNT were more active in proliferation. Although on day 1 after seeding, the initial cell population density was similar on both materials with and without nanotubes (40340 ± 5765 cells/cm^2 and 35489 ± 5833 cells/cm^2, respectively), on day 7, the cells on the MWCNT-modified terpolymer reached 4.5 times higher density (228029 ± 10050 cells/cm^2) than on the unmodified samples (50300 ± 5400 cells/cm^2). The improved adhesion and growth of MG 63 cells could be explained by the nanostructure of the material surface, which resembled the architecture of the natural extracellular matrix (ECM) and probably allowed adsorption of cell adhesion-mediating ECM molecules (mainly vitronectin, fibronectin) in spatial conformation optimal for their binding by cell adhesion receptors [6].

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[1] P. Kohli, C.R. Martin, *Curr. Pharm. Biotechnol.* **6**, 35-47 (2005).
[2] G. Gruner, *Anal. Bioanal. Chem.* **384**, 322-335 (2006).
[3] C.C Harrell, P. Kohli, Z. Siwy, C.R Martin, *J. Am. Chem. Soc.* **126**, 15646-15647 (2004).
[4] B.J. Hinds, N. Chopra, T. Rantell, R. Andrews, V. Gavalas, L.G. Bachas, *Science* **303**, 62-65 (2004).
[5] L. Bacakova, V. Stary, O. Kofronova, V. Lisa, *J. Biomed. Mater. Res.* **54**, 567-78 (2001).
[6] D.C. Miller, K.M. Haberstroh, T.J. Webster, *J. Biomed. Mater. Res.A*, **73**, 476-84 (2005).