



Enhanced adhesion of osteoblastic cells on polystyrene films by independent control of surface topography and wettability

Seung Yun Yang ^{a,1}, Eung-Sam Kim ^{b,1}, Gumhye Jeon ^a, Kwan Yong Choi ^{b,c,*}, Jin Kon Kim ^{a,*}

^a National Creative Research Center for Block Copolymer Self-Assembly, Departments of Environmental Science & Engineering and Chemical Engineering, Pohang University of Science and Technology, Pohang, 790-784, South Korea

^b School of Interdisciplinary Bioscience and Bioengineering, Pohang University of Science and Technology, Pohang, 790-784, South Korea

^c Department of Life Science, Division of Molecular and Life Science, Pohang University of Science and Technology, Pohang, 790-784, South Korea

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ABSTRACT

We independently controlled surface topography and wettability of polystyrene (PS) films by CF₄ and oxygen plasma treatments, respectively, to evaluate the adhesion and proliferation of human fetal osteoblastic (hFOB) cells on the films. Among the CF₄ plasma-treated PS films with the average surface roughness ranging from 0.9 to 70 nm, the highest adhesion of hFOB cells was observed on a PS film with roughness of ~11 nm. When this film was additionally treated by oxygen plasma to provide a hydrophilic surface with a contact angle less than 10°, the proliferation of bone-forming cell was further enhanced. Thus, the plasma-based independent modification of PS film into an optimum nanotexture for human osteoblast cells could be applied to materials used in bone tissue engineering.

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1. Introduction

A compatible interaction between a target cell and the surface of solid biomaterials is a pivotal factor for the seamless integration of artificial implants into tissues [1–3]. Surface characteristics in nanoscale, for instance, topography or chemical and physical properties, are crucial in directing cellular behaviors such as adhesion, proliferation, and differentiation in the cell–surface interface in the bone [4–9]. Especially, since bone-forming osteoblastic cells can recognize the nanotextured resorption pit produced by the osteoclast and form new osteoid during the bone remodeling process [10], the proper modification of the solid surfaces is essential for cell survival on foreign substrates as well as subsequent cellular proliferation and the deposition of calcium-containing mineral for the bone-forming process. Chemical oxidation [11], electron-beam lithography [12], colloidal particle adsorption [13], addition of carbon nanotube [14,15],

plasma treatment [16], and self-organizing [17] or self-assembling techniques [18] were introduced to tailor the surface characteristics in nanoscale in order to enhance the selective adhesion of osteoblast and to retard the attachment of fibroblast, which can inhibit the adhesion of osteoblast on the surfaces.

Two important parameters of the surface characteristics should be considered. First, nanoscale topography greatly influences cellular behavior due to its bio-mimicry of in vivo tissue environment at the molecular level [19]. Second, the wettability of a substrate surface plays a critical role in the adhesion of osteoblastic cells. Surface wettability has been modulated by self-assembled monolayer or plasma treatment [20–22]. Previous results showed that a modestly hydrophilic surface with water contact angle (θ) less than 60° exhibited an enhanced adhesion of cells [5]. Although many research groups have investigated the effect of either topography or wettability of a substrate surface on the adhesion of osteoblast cells [23], those methods have shown a limited range of controllability of these two parameters. Moreover, these two parameters could not be changed independently because of the experimental limitations [18,24]. For instance, an introduction of functional groups on a surface by utilizing self-assembled monolayer (SAM) is a suitable way to change the wettability of the substrate, while the roughness is maintained. However, in this case, it is not easy to investigate the effect of wettability on the cellular response in a wide range of wettability. This is because SAM-modified surfaces show only a limited range of wettability, though various molecules with end groups such as CF₃, NH₂, or CH₃ could be grafted on the substrate. Alternatively, plasma

Abbreviations: MW, molecular weight; PS, polystyrene; RMS, root mean square; AFM, atomic force microscope; XPS, X-ray photoelectron spectroscopy; hFOB, human fetal osteoblast; SEM, scanning electron microscope; SD, standard deviation.

* Correspondence to: K.Y. Choi, School of Interdisciplinary Bioscience and Bioengineering, Pohang University of Science and Technology, Pohang, 790-784, South Korea. Tel.: +82 54 279 2295; fax: +82 54 279 8290.

** Corresponding author. Tel.: +82 54 279 2276; fax: +82 54 279 8298.

E-mail addresses: kchoi@postech.ac.kr (K.Y. Choi), jkkim@postech.ac.kr (J.K. Kim).

¹ These authors contributed equally to this work.

treatment has been widely used for surface modifications due to its convenience and cost-effectiveness. But, it is mainly used for control of the wettability of a substrate surface, not for surface roughness.

Recently, Dowling et al. investigated the effects of the surface roughness and the wettability of polystyrene films on the adhesion of human osteosarcomas cells [9]. They changed the surface roughness by grinding the PS films with silicon carbide papers having different grit sizes, while the wettability was controlled by the deposition of poly(dimethyl siloxane) (PDMS) from its liquid precursor under He/O₂ plasma treatment and a fluorination through CF₄ plasma treatment. However, because the roughness of the surface was changed by grinding the PS films, the surface roughness could not be controlled within nanometer scales. Furthermore, PDMS coating or a relatively long etching of CF₄ plasma employed to control the surface wettability could greatly affect the surface roughness.

Thus, to understand the effect of surface roughness and wettability on cell adhesion or proliferation, a facile method is needed to control independently one parameter without affecting another one, while the uniformity of surface roughness and wettability should be

maintained on a large area. Also, each modification process should be simple and be finished in a short time.

Among various biomaterials, polymers have been widely used in bone tissue engineering due to facile fabrication of complex shape in addition to cost-effectiveness [25,26]. Especially, polystyrene (PS) has received great attention as a coating material for surface modification of a bone implant because it can be coated on a target substrate by simple methods such as drop-casting [27]. In addition, PS surface is easily functionalized with hydroxyl, aldehyde, carboxyl or amine groups to increase the surface wettability and to introduce negative (or positive) charge for better cell adhesion [28,29].

In this work, we present a plasma-based method to independently control the surface topography and wettability of PS films. Through this work, we can understand the effect of surface characteristics on the cellular behaviors of human osteoblastic cells on polystyrene films. CF₄ and O₂ plasma were shortly treated to control surface roughness and wettability, respectively. Surface topography of the PS films was fine-tuned in nanoscale by CF₄ plasma, whereas their wettability was independently modulated from a hydrophobic

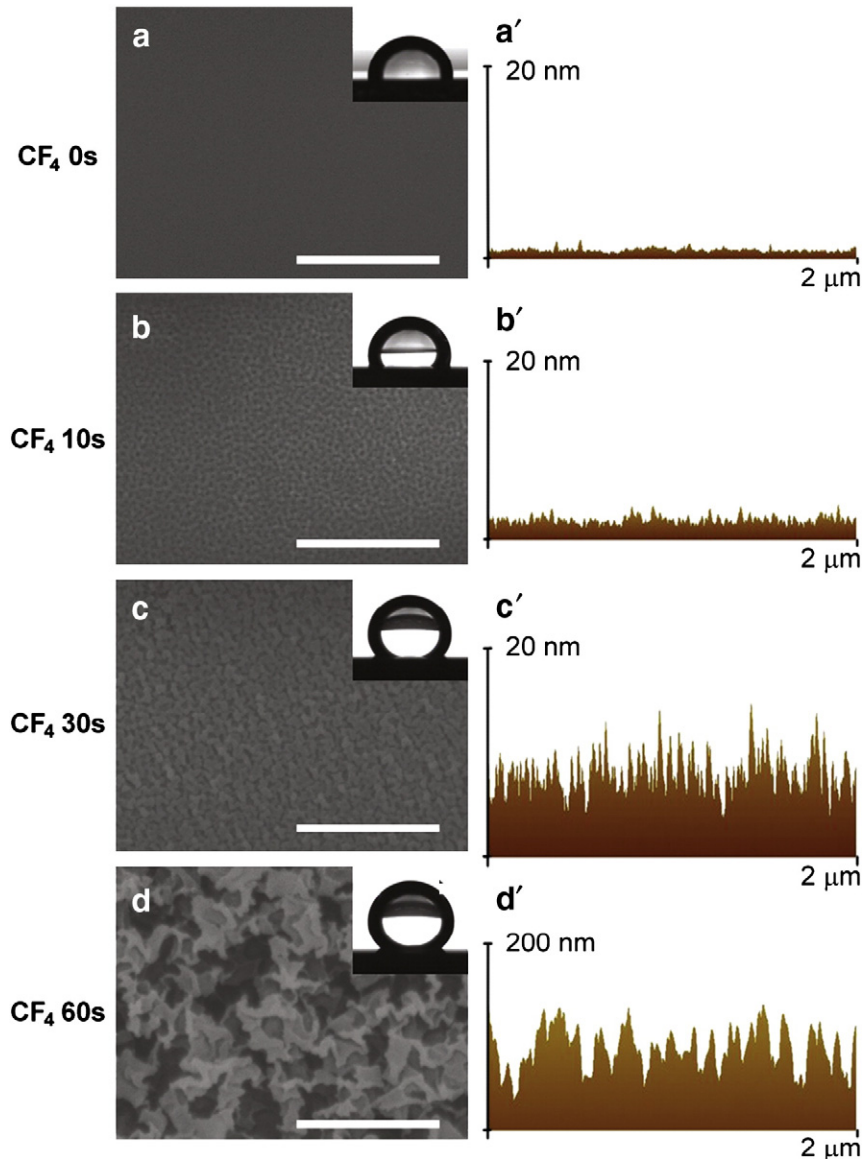


Fig. 1. FE-SEM (a–d) images and AFM (a'–d') profiles for the nanotextured PS films treated by CF₄ plasma for various times. Surface of the PS films became roughened with increasing CF₄ plasma treatment. At a given treatment time, rather uniform surface structure is seen over the entire PS surface. A water droplet on the nanotextured PS surfaces (a–d) was displayed in the inset of each image. (Scale bar: 300 nm).

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