

Preparation of Well-Defined Chitosan Nanofibers by Atom Transfer Radical Polymerization

Nilhan Kayaman-Apohan, Burcu Oktay
Marmara University, Department of Chemistry
34722 Goztepe-Istanbul, Turkey
napohan@marmara.edu.tr; brcoktay@hotmail.com

Serap Erdem-Kuruca
Istanbul University Istanbul Medical Faculty, Department of Physiology
34390 Capa-Istanbul, Turkey
sererdem@yahoo.com

Extended Abstract

Electrospinning is the most widely used technique for the development of nanofibrous scaffolds. The three dimensional (3D) biodegradable scaffolds designed using nanofibers serve as an excellent framework for cell adhesion, proliferation, and differentiation because of their high surface area to volume ratio and porosity (Vasita and Katti, 2006). An ideal tissue engineering scaffold should mimic both the form and functionality of the native extra cellular matrix (ECM) (Oktay et al, 2015a).

3D nanofibers were prepared by using electrospinning process. Firstly, the amino groups of chitosan chemo-selectively protected with a simple and convenient procedure (Nishimura et al., 1991) and then chitosan reacted with sodium azide to give a chitosan azide (Zampano et al., 2010). The protecting groups were removed backbone before electrospinning of chitosan azide. To improve the biocompatibility, a well-defined polymer brushes were produced on nanofiber. For this, we grafted 2-methacryloyloxyethyl phosphorylcholine (MPC) polymers onto the chitosan nanofiber because of its structure similar to the phospholipid bilayer of a cell membrane. Phosphorylcholine containing polymers are amongst the most biocompatible polymer known, which are used a high level of biocompatibility and resistance to protein adsorption is required (Chen et al., 2009).

In general, there are two methods for modification of the polymer surface. The first method involves a surface absorption and the second is grafting of polymer molecule onto the surface (Kyomoto et al., 2008). Grafting polymerization is performed the following methods: (1) surface initiated graft polymerization, termed as the “grafting from”, (2) attachment of the polymer to the surface, termed as the “grafting to”. However, the grafting to method is not uniform. For this, we synthesized propargyl 2-bromoisobutyrate as a bifunctional initiator for both click reaction and atom transfer radical polymerization (ATRP). ATRP is one of the CRP techniques, which allow for both control over molar mass and for more complex molecular architectures (Oktay et al, 2015b). The azido groups on the chitosan nanofiber and alkyne groups of propargyl 2-bromoisobutyrate capped by click cyclization reaction. Thereafter, poly(MPC) grafted nanofiber produced by SI-ATRP of MPC onto the initiator immobilized chitosan nanofiber.

The structure and the morphology of the chitosan nanofibers were characterized by Scanning electron microscopy (SEM) and Fourier transform infrared spectroscopy (FT-IR) technique. The nanofibers with a narrow size distribution were obtained. The nanofibers have an average diameter of about 40–100 nm. In vitro attachment and growth of 3T3 mouse fibroblasts on well-defined chitosan nanofibers were also investigated. Cell attachment, proliferation and MTT cytotoxicity assays indicated good cell viability throughout the culture time, which was also confirmed by SEM analysis. Nanofiber matrix was completely covered by 3T3 fibroblast cells and cell proliferation significantly increased after incubation.

The cell morphology gradually changed to a more flattened morphology. High density and high mobility of free end groups of the well-ordered poly(MPC) brushes considered cell growth. Addition, penetration of 3T3 fibroblast cells to the scaffolds were revealed with laser scanning confocal microscopy.

- Chen, X., McRae, S., Parekar, S., & Emrick, T. (2009). Polymeric Phosphorylcholine-Camptothecin Conjugates Prepared by Controlled Free Radical Polymerization and Click Chemistry. *Bioconjugate Chem*, 20, 2331–2341.
- Kyomoto, M., Moro, T., Iwasaki, Y., Miyaji, F., Kawaguchi, H., Takatori, Y., Nakamura, K., & Ishihara, K. (2008). Superlubricious Surface Mimicking Articular Cartilage Bygrafting Poly(2-Methacryloyloxyethyl Phosphorylcholine) On Orthopaedic Metal Bearings. *Journal of Biomedical Materials Research Part A*, 91, 730-741.
- Nishimura, S., Kohgo, O., & Kurita, K. (1991). Chemospecific Manipulations of a Rigid Polysaccharide: Syntheses of Novel Chitosan Derivatives with Excellent Solubility in Common Organic Solvents by Regioselective Chemical Modifications. *Macromolecules*, 24, 4745-4748.
- Oktay, B., Kayaman-Apohan, N., Erdem-Kuruca, S., & Süleymanoğlu, M. (2015a). Fabrication Of Collagen Immobilized Electrospun Poly (Vinyl Alcohol) Scaffolds. *Polymers for Advanced Technologies*, DOI:10.1002/pat.3512.
- Oktay, B., Demir, S., & Kayaman-Apohan, N. (2015b). Immobilization Of A-Amylase Onto Poly(Glycidyl Methacrylate) Grafted Electrospun Fibers By ATRP. *Materials Science and Engineering C*, 50, 386-393.
- Vasita, R., & Katti, D.S. (2006). Nanofibers And Their Applications In Tissue Engineering. *International Journal of Nanomedicine*, 1, 15-30.
- Zampano, D., Bertoldo, M., & Ciardelli, F. (2010). Defined Chitosan-Based Networks By C-6-Azide-Alkyne “Click” Reaction. *Reactive And Functional Polymers*, 70, 272-281.