

## Fabrication of Porous Scaffolds for Bone Tissue Engineering Using a 3-D Robotic System: Comparison with Conventional Scaffolds Fabricated by Particulate Leaching

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### 1 Introduction

The 3-D scaffolds are necessary for the cells to proliferate and maintain their differentiation. These scaffolds should be made by porous structure which helps cells proliferate well when scaffolds are implanted to a body. There are several conventional methods to fabricate 3-D scaffolds such as particulate leaching, freeze drying, electro-spinning [1-3]. Even though, they can control the pore size and porosity, there is limitation such as penetration of cells. Recent various methods for 3-D scaffolds are introduced to overcome this limitation [4]. This study aims at fabrication 3-D scaffolds using modified RP (rapid-prototype) system and comparing conventional 3-D scaffolds fabricated by particulate leaching.

### 2 Materials and Methods

**Fabrication of two-types scaffolds.** The novel scaffolds were fabricated using modified RP (rapid-prototype) system. PCL (poly ε-caprolactone, Sigma Aldrich, Mw 65000, USA) were dissolved into 10 ml of chloroform by stirring for several hours (~4 hrs) and inserted a syringe type injector including this PCL solution in RP (rapid-prototype) system. The conventional PCL 3-D scaffolds were fabricated by GS (gas foaming/salt leaching)

process (pore size: ~250 μm).

**Mechanical test.** The compressive mechanical properties of the each scaffolds were evaluated ( $n=4$ ) with a Micro-load System (Microload system, R&B Inc, Korea) at room temperature. The displacement rate was set to be 0.5 mm/min. The compressive modulus was determined from the initial porous response region from 0 to 30% strain.

**Seeding cell and MTT assay.** MG-63 osteoblast-like cells (American Type Culture Collection, No. CRL-1427, USA) were used for experiments on scaffolds. The culture medium was Dulbecco's Modified Eagle Medium (Gibco, USA) with 10% fetal bovine serum (FBS) and 1% Penicillin/Streptomycin (Hyclone, USA). The cells were seeded on scaffolds at  $10^6$  cells/ml. To determine the cell viability and proliferation on the scaffolds, we carried out a MTT assay (Cell Proliferation Kit I, Boehringer Mannheim, Mannheim, Germany). The surface and cross-sectional morphologies of scaffolds were observed by a FE-SEM (Hitachi Ltd., S-4300SE, 5kV, JAPAN).

### 3 Results and Discussion

Porous scaffolds were successfully fabricated through RP process and GS process (Fig. 1). The scaffolds fabricated by RP process were found to be stronger than scaffolds fabricated by conventional GS process (Fig. 2). The MTT assay for the scaffolds made by GS showed that the proliferation rate was decreased along the time. However, in the scaffolds fabricated by RP process cells were steadily proliferated (Fig. 3). It suggested that cells could not penetrate into scaffolds made by GS,

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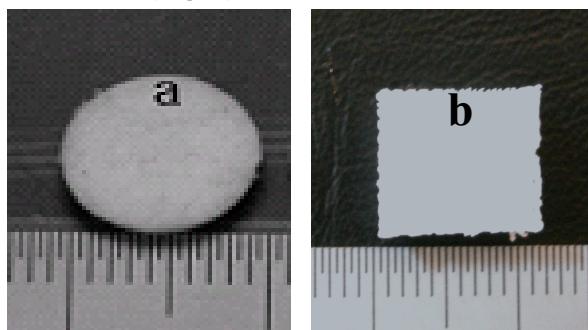
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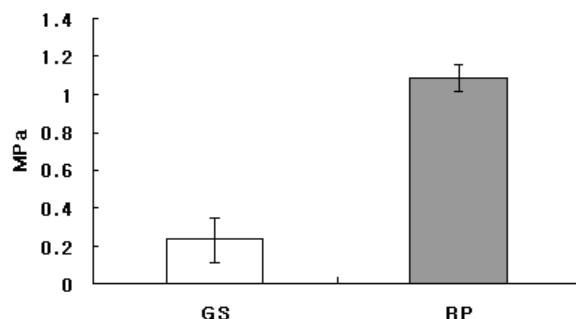
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while they could penetrate into the scaffolds made by RP. This was confirmed through the SEM observations (**Fig. 4**).

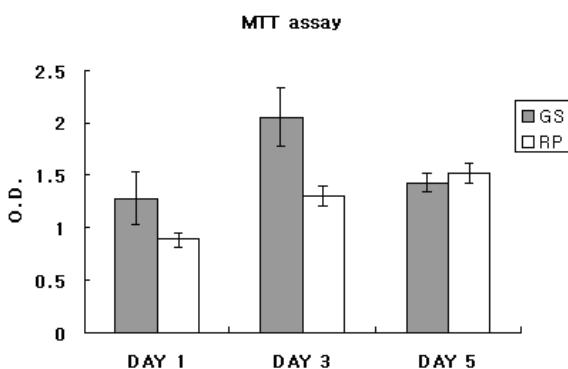


**Figure 1 :** Macroscopic images of PCL scaffolds.

(a: GS process, b: RP process).



**Figure 2 :** Compressive modulus of two type scaffolds ( $n = 4$ ).

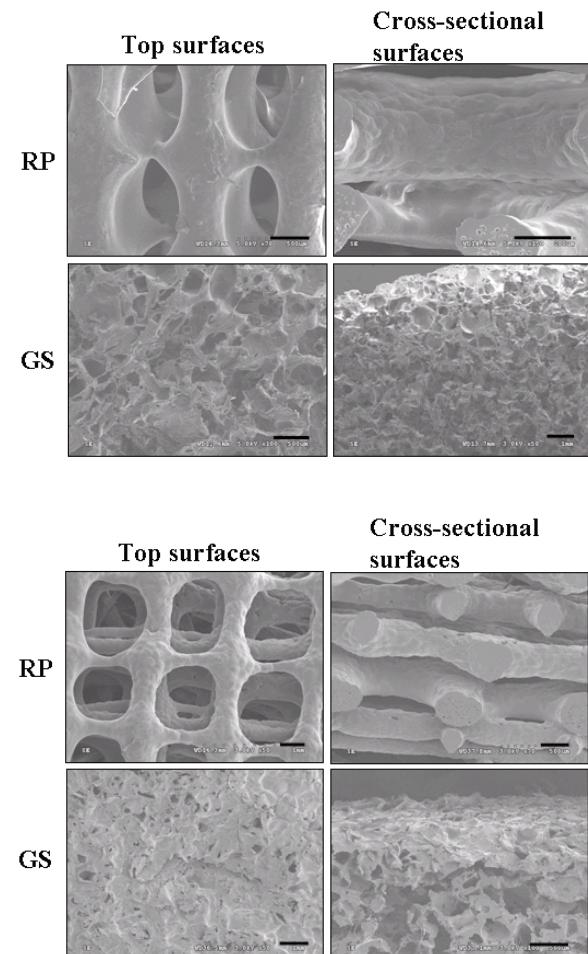


**Figure 3 :** MTT assay ( $n=5$ ).

#### 4 Conclusion

From the study, the scaffolds fabricated by RP process offer more capability for cells to penetrate

into the scaffolds than scaffolds by GS process. Therefore RP method is more suitable for scaffolds of bone regeneration than conventional GS process scaffolds. However, further study should be carried out such as long term experiments through the animal studies.



**Figure 4 :** SEM images. **Top**, day 1, **bottom**, day 5. Scale bar = 200  $\mu\text{m}$ .

#### References

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