HYDROGEL MICRO-PATTERN ON NANOPOROUS MEMBRANE FOR MANIPULATION OF CELL-CELL INTERACTION

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ABSTRACT
Combination of nanoporous membrane and hydrogel micro-patterning technique is suitable for biomimetic microenvironment in tissue engineering. To make hydrogel micropattern on nanoporous membrane with immobilization of cells and growth factors, the nanoporous membrane was fabricated by electrospinning method and micro-patterning was generated from concave micromold, respectively. Encapsulated cells were observed for cell-cells interaction on hydrogel patterned nanoporous membrane. Hydrogel micropatterned nanoporous membrane could be applied to 3D biomimetic co-culture system for cell-cell interaction study.

KEYWORDS: hydrogel, cell-cell interaction, nanoporous membrane, micropattern, micromold

INTRODUCTION
Micro and nano patterns are already an introduced technique for cell behavior study. Micro-patterns are an enabling tool as a biomimetic microenvironment in cell-cell interaction study [1]. Recent researches in combination of biocompatible hydrogel and micro-patterning technique showed much attention of encapsulation and delivery of drugs and cells using a micro-mold system [2]. However, the conventional of patterning system are generated on surface treated slide glass that is not suitable for supporting of biomimetic microenvironments to cells. The 2D micro patterning is simple but has limited application due to surface condition of substrate. In order to overcome 2D microenvironment, electrospinning method was used for making nanoporous membrane that similar to extra cellular matrix (ECM) and it makes biomimetic environment [3]. Here, we presented the hydrogel 3D micro patterning on electrospun nanoporous membrane for cell-cell interaction study with encapsulation and release control of micro-particles, growth factors, and cells.

Figure 1: Schematic diagram of hydrogel micro patterning process on nanoporous membrane

EXPERIMENTAL

Fabrication of 3D hydrogel structure was performed using a hemispheric polydimethylsiloxane (PDMS)-based micro mold and UV-curable poly (ethylene glycol) diacrylate (PEGDA) solution onto polyethersulfone (PES) electrospun nanoporous membrane with UV source. Figure 1 indicated the process of fabrication hydrogel structure on the nanoporous membrane. During fabrication of 3D patterning process, micro-particles, growth factors and cells were encapsulated for manipulation and leading of cell behaviors. Immobilized growth factor of release control was generated by degradation of PEGDA and concentration of growth factor on process. In order to observe the cell-cell interaction, the HepG2 cell and CCD-986sk fibroblast cells were co-cultured. Also, to estimate the cell-cell behaviors, cells and growth factors, which were immobilized in PEGDA micro-pattern onto micro-patterned nanoporous membrane with continuous flow condition. Cell behaviors were observed by fluorescence microscopy and confocal laser microscopy (CLM).

RESULTS AND DISCUSSION

Convex structured PEGDA hydrogel patterns were induced on the nanoporous membrane by UV exposure. Figure 2 is optical and fluorescence images that patterned hydrogel on the membrane. For indicated the encapsulation of hydrogel, we used Rhodamine 6G, BSA-FITC and 10 um red fluorescence silica beads. Figure 3 shows that their confocal images that indicated encapsulation were successful and dye and beads encapsulated hydrogel was formed 3D convex structure.

To evaluate the mass transport, dye solution was released from hydrogel structure by the diffusion through the membrane and degradation of hydrogel. Figure 4 is each encapsulated cell, cell cultured on
patterned membrane, localization of cells for cell-cell interaction. It was dyed by DAPI and Live and dead kit. It is shows that two kinds of cells were localized completely.

**CONCLUSION**
Here, we demonstrated the manipulation of cell-cell interaction by cell localization into hydrogel micropattern on nanoporous membrane. This method could be used for the investigation of cell-cell interaction and cell behavior. Moreover, this technology will be able to use for enabling tool of cells in 3D environments.

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**REFERENCES**

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