Gelatin Methacrylate Hydrogels with Dielectrophoretically Aligned Carbon Nanotube for Muscle Tissue Engineering

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Abstract—This study uses dielectrophoresis (DEP) to align carbon nanotubes (CNTs) within hydrogels that were made from gelatin methacrylate (GelMA). The resulting gels were highly conductive compared with GelMA hydrogels with random or without CNTs. The proposed DEP procedure is rapid, simple, and inexpensive. GelMA hydrogels were used to fabricate skeletal muscle tissues indicating that GelMA hydrogels with aligned CNTs were more efficient in generating muscle tissues compared with other controls, particularly after applying electrical stimulation (ES).

I. INTRODUCTION

Construction of muscle tissues requires cells to seed, proliferate, and differentiate within a scaffold. Currently used scaffolds often are not conductive; therefore, they may not be functional to regulate the behavior of electrophysiologically active cells, such as muscle or cardiac cells [1]. We previously demonstrated that GelMA hydrogels provide a suitable environment for C2C12 myoblasts to proliferate and differentiate into myotubes [2,3]. However, they were not conductive and mechanically strong, which limited their performance to differentiate muscle cells upon applying ES. In this study, we introduce a simple technique to dielectrophoretically align CNTs within GelMA hydrogels to increase their electrical conductivity and mechanical strength. GelMA prepolymer has low viscosity and ionic content, the characteristics required for an appropriate medium in DEP. Therefore, DEP was successfully applied to obtain aligned CNTs within GelMA hydrogels. The GelMA hydrogels with aligned CNTs were then examined in fabricating muscle tissues.

II. EXPERIMENTAL SECTION

GelMA hydrogel was synthesized as described elsewhere [4]. Briefly, it was made of gelatin and methacrylic anhydride and was crosslinked using UV. An AC electric field (voltage 20 Vpp and frequency 2 MHz) was applied through interdigitated array of Pt (IDA-Pt) electrodes to dielectrophoretically align CNTs within GelMA prepolymer, which was subsequently crosslinked to preserve the CNT alignment (Fig. 1A). C2C12 myoblasts were loaded on the microgrooved GelMA hydrogels and after 1 day they underwent the differentiation process for 9 days of culture. C2C12 myotubes were electrically stimulated (voltage 8 V, frequency 1 Hz, duration 10 ms) for 2 continuous days starting at day 8 of culture.

III. RESULTS AND DISCUSSION

CNTs aligned along with the electric field direction generated using IDA-Pt electrodes. Therefore, they could increase the conductivity of hydrogels. C2C12 myoblasts were proliferated and differentiated on three GelMA hydrogels, i.e., pristine GelMA, GelMA with aligned CNTs and GelMA with random CNTs. C2C12 myotubes were stained against myosin heavy chain to indicate the myoblast differentiation (Fig. 1B). Figure 2 shows the gene expression results for C2C12 myotubes cultured on these hydrogels. ES significantly enhanced the genes related to maturation and contractility of muscle myofibers on the aligned CNTs in GelMA hydrogels compared with the other hydrogel controls.

In summary, DEP was employed to align CNTs in GelMA hydrogel and tune their conductivity and mechanical properties for muscle tissue engineering.

REFERENCES