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Developing a Thermo-Sensitive Conductive Stretchable Film to Allow Cell Sheet Harvest after Mechanical and Electrical Treatments

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Abstract : Depositing conductive polypyrrole (PPy) onto elastic polydimethylsiloxane (PDMS) substrate can obtain a highly stretchable conductive film, which can be used to construct a bioreactor to cyclically stretch and electrically stimulate surface cells. However, how to completely harvest these stimulated muscle tissue to repair damaged muscle is a challenge. To address this concern, N-isopropylacrylamide (NIPAAm), a monomer of temperature-sensitive polymer, was added during the polymerization of pyrrole on PDMS so that the resulting P(Py-co-NIPAAm)/PDMS should own both conductivity and thermo-sensitivity. Therefore, cells after stimulation can be completely harvested as cell sheets by reducing temperature. Mouse skeletal myoblast, C2C12 cells, were applied to examine our hypothesis. In electrical stimulation, C2C12 cells on P(Py-co-NIPAAm)/PDMS demonstrated the best myo-differentiation under the electric field of 1 V/cm. Regarding cyclic stretching, the strain equal to or higher than 9% can highly align C2C12 perpendicular to the stretching direction. The Western blotting experiments demonstrated that the cell sheets harvested by cooling reserved more extracellular matrix (ECM) than cells collected by the traditional trypsin digestion method. Immunostaining of myosin heavy chain protein (MHC) indicated that both mechanical and electrical stimuli effectively increased the number of myotubes and the differentiation ratio, and the myotubes can be aligned by cyclic stretching. Stimulated cell sheets can be harvested by cooling, and the alignment of myotubes was still maintained. These results suggested that the deposition of P(Py-co-NIPAAm) on PDMS can be applied to harvest intact cell sheets after cyclic stretching and electrical stimulation, which increased the feasibility of bioreactor for the application of tissue engineering and regenerative medicine.

Keywords : bioreactor, cell sheet, conductive polymer, cyclic stretching, electrical stimulation, muscle tissue engineering, myogenesis, thermosensitive hydrophobicity
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