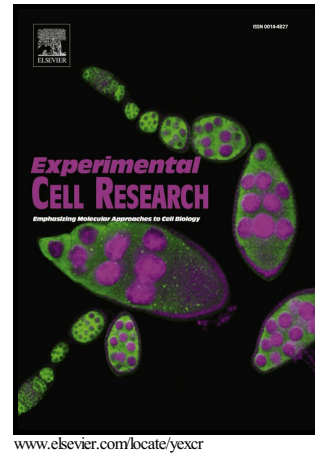


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Three-dimensional contractile muscle tissue consisting of human skeletal myocyte cell line

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## Abstract

This paper describes a method to construct three-dimensional (3D) contractile human skeletal muscle tissues from a cell line. The 3D tissue was fabricated as a fiber-based structure and cultured for two weeks under tension by anchoring its both ends. While myotubes from the immortalized human skeletal myocytes used in this study never contracted in the conventional two-dimensional (2D) monolayer culture, myotubes in the 3D tissue showed spontaneous contraction at a high frequency and also reacted to the electrical stimulation. Immunofluorescence revealed that the myotubes in the 3D tissues had sarcomeres and expressed ryanodine receptor (RyR) and sarco/endoplasmic reticulum  $\text{Ca}^{2+}$ -ATPase (SERCA). In addition, intracellular calcium oscillations in the myotubes in the 3D tissue were observed. These results indicated that the 3D culture enabled the myocyte cell line to reach a more highly matured state compared to 2D culture. Since contraction is the most significant feature of skeletal muscle, we believe that our 3D human muscle tissue with the contractile ability would be a useful tool for both basic biology research and drug discovery as one of the muscle-on-chips.

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