Effect of Connexin Gap Junction Expression on Skeletal Muscle Function

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Gap junctions are intercellular conduits which allow the direct transfer of small molecules such as metabolites and ions from the cytoplasm of one cell to another, enabling cellular communication and synchronized activity. A single gap junction is comprised of subunits called connexins, which are found in many forms and vary by molecular weight. While connexin gap junctions play an important role in the electrical coupling of cardiac muscle, mature skeletal muscle does not express connexin, and thus each muscle cell must be independently stimulated to contract.

In this study, we utilize C2C12 myoblasts transfected with connexin 37 (Cx37), connexin 40 (Cx40), or connexin 43 (Cx43) under the muscle creatine kinase (MCK) promoter to study the effect of connexin expression on skeletal muscle function. We confirm and quantify connexin expression throughout differentiation through immunofluorescent imaging as well as RT-qPCR analysis of RNA expression. We demonstrate no effect of connexin expression on cell viability, but a change in myotube morphology, with all connexin-expressing cell lines exhibiting increased myotube length upon differentiation. We also examine the expression profile of early, mid, and late-stage myogenic markers throughout differentiation, with connexin-expressing C2C12s exhibiting a relatively early increase in the expression of late-stage myogenic markers. We also analyze the impact of connexin expression on action potential propagation and functional performance of skeletal muscle. Ultimately, we aim to tissue engineer electrically coupled skeletal muscle bioactuators to better enable motor neuron innervation and control of skeletal muscle force production.