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**Development of a micro-electrode array-cantilever system for the detection of functional neuromuscular junctions formed between human stem cell-derived motoneurons and human skeletal muscle *in vitro***

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The ability to efficiently monitor the functionality of neuromuscular junctions (NMJs) *in vitro* is essential to understanding the mechanisms that lead to their disruption during the early onset of neurodegenerative diseases. Functional *in vitro* NMJ systems with human cells instead of animal cells eliminate the issue of species variability in the development of new therapeutics. In this study we report a compartmentalized co-culture system that monitors the functionality of neuromuscular interactions by recording myotube contraction in response to isolated electrical stimulation of motoneurons. Motoneuron cell bodies are located on microelectrodes arrays (MEAs) in a motoneuron chamber while their axons extend through micro fluidic tunnels to a distal chamber containing skeletal muscle on surface-modified micro fabricated silicon cantilevers. While the MEAs stimulate and/or record the activity of motoneurons, cantilever deflection monitored by laser from underneath allows for measurement of the force and frequency of myotube contraction. Two-chambered micro fluidic devices have been used extensively to study neuronal function, axonal biology and synapses. However, this is the first time that this platform has been coupled with two functional Bio-MEM (biological microelectromechanical system) devices namely, MEAs and cantilevers. This elegant system allows non-invasive high-throughput interrogation of the function of NMJs, providing a valuable test bed for the etiological study and drug development of relevant neurodegenerative diseases.

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