Les Algorithmes
rue Jean Sapidus
Parc d'Innovation
67400 ILLKIRCH - France
Tel : +33 (0)3 88 65 16 06
Fax : +33 (0)3 88 65 16 22

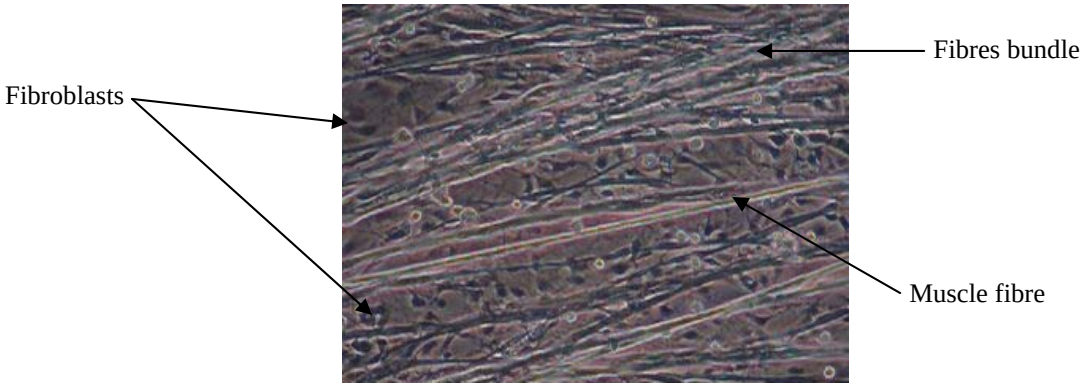
RAT NERVE-HUMAN MUSCLE COCULTURE : A FUNCTIONAL MODEL OF THE MOTOR UNIT

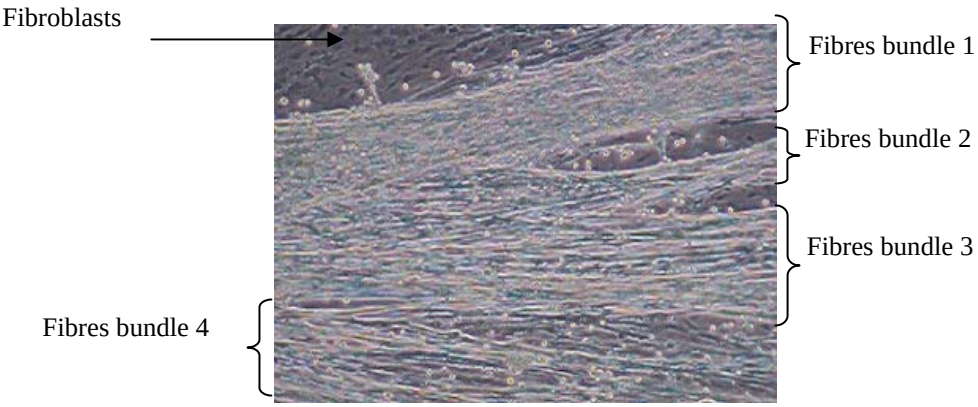
1. Introduction

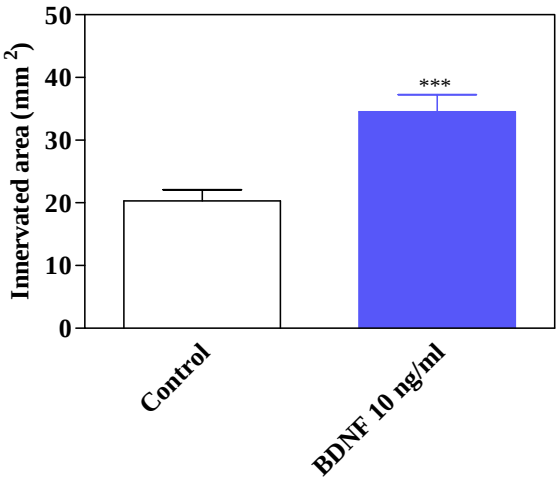
The motor unit is an intricate pluricellular structure in which motor neurons and muscle fibers are dependant on each other. Its formation and maintenance depend on continuous trophic, electrical and mechanical cross-talk between muscle and motor nerve.
The ability of compounds to improve spinal motor neurons functionality can be assess by determining their ability to innervate and to induce muscle innervation in a co-culture of rat spinal motor neurons with human muscle.

2. Compound testing

Muscle derived cells, mainly myoblastes and fibroblasts are cultured in monolayers. Immediately after myoblaste fusion, whole transverse slices of 13-day-old rat embryos spinal cords with dorsal root ganglia attached are placed on the muscle monolayer. After 24h neuritis are observed growing out of the spinal cord explants. They make contacts with myotubes and induce the first contractions. Quickly thereafter, innervated muscle fibres located in proximity to the spinal cord explants, are virtually continuously contracting. Innervated fibres are morphologically and spatially distinct from the non-innervated ones and could easily be distinguished from them (Askanas et al., 1987).
The ability of a molecule to promote spinal motor neuron survival, neurites outgrowth and/or neurons functionality can be evaluated by measuring neurite sprouting (number and length), innervation rate (number of spinal cord-muscle couple which lead to an innervation), innervation area (surface covered by innervated muscle fibers per spinal cord explant), ACh receptor aggregation as well as several other parameters depending upon the mode of action of the compound (Braun et al., 1996).







BDNF at 10 ng/ml increases the innervate rate from 20% under control condition to 25% with BDNF and augments the surface of skeletal muscle innervated by an efficient rat spinal cord-human muscle coupling.

3. References.

Askanas V., Kwan H., Alvarez R.B., Engel W.K., Kobayashi T., Martinuzzi A. and Hawkins E.F. (1987). De novo neuromuscular junction formation on human muscle fibres cultured in monolayer and innervated by foetal rat spinal cord: ultrastructural and ultrastructural-cytochemical studies. J Neurocytol., 523-537

Braun S., Croizat B., Lagrange M.-C., Warter J.-M. and Poindron P. (1996). Neurotrophins increase motoneurons' ability to innervate skeletal muscle fibers in rat spinal cord--human muscle co-cultures. J. Neurol. Sci., 136: 17-23.