**Anat Cell Biol 2020; 53(3): 292-300**

# Role of cerebrospinal fluid in differentiation of human dental pulp stem cells into neuron-like cells

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https://doi.org/10.5115/acb.19.241

Abstract

Human dental pulp stem cells (hDPSCs) could be differentiated into neuron like-cells under particular microenvironments. It has been reported that a wide range of factors, presented in cerebrospinal fluid (CSF), playing part in neuronal differentiation during embryonic stages, we herein introduce a novel culture media complex to differentiate hDPSCs into neuron-like cells. The hDPSCs were initially isolated and characterized. The CSF was prepared from the Cisterna magna of 19-day-old Wistar rat embryos, embryonic cerebrospinal fluid (E-CSF). The hDPSCs were treated by 5% E-CSF for 2 days, then neurospheres were cultured in DMEM/F12 supplemented with $10^{-6}$ µm retinoic acid (RA), glial-derived neurotrophic factor and brain-derived neurotrophic factor for 6 days. The cells which were cultured in basic culture medium were considered as control group. Morphology of differentiated cells as well as process elongation were examined by an inverted microscope. In addition, the neural differentiation markers (Nestin and MAP2) were studied employing immunocytochemistry. Neuronal-like processes appeared 8 days after treatment. Neural progenitor marker (Nestin) and a mature neural marker (MAP2) were expressed in treated group. Moreover Nissl bodies were found in the cytoplasm of treated group. Taking these together, we have designed a simple protocol for generating neuron-like cells using CSF from the hDPSCs, applicable for cell therapy in several neurodegenerative disorders including Alzheimer's disease.

**Keywords**: Human dental pulp stem cells, Cerebrospinal fluid, Cell transdifferentiation, Alzheimer