Cerebrospinal Fluid Research

BioMed Central

Oral presentation

Open Access

The CSF of normal H-Tx rats promotes neuronal differentiation from neurospheres but CSF of hydrocephalic H-Tx rats does not

César González*, Karin Vío, Rosa I Muñoz and Esteban M Rodríguez

Address: Instituto de Histología y Patología, Universidad Austral de Chile, Valdivia, Chile

Email: César González* - cesargonzalez@uach.cl

* Corresponding author

from 50th Annual Meeting of the Society for Research into Hydrocephalus and Spina Bifida
Cambridge, UK. 30 August – 2 September 2006

Published: 21 December 2006

Cerebrospinal Fluid Research 2006, **3**(Suppl 1):S10 doi:10.1186/1743-8454-3-S1-S10

Background

There is evidence that in both animal models and humans, hydrocephalus affects neuronal migration and maturation. In HTx rats evidence points to a critical role of CSF in maintaining the normal proliferation of the stem cells in the developing cortex. Furthermore, such an activity is affected by CSF of HTx rats with early-onset hydrocephalus. The source and nature of these signals in not known. However, the presence in the CSF of hydrocephalic HTx rats and *hyh* mice of abnormal forms of the proteins secreted by the subcommissural organ points to them as good candidates for conveying signals to the developing brain cortex. The present study was designed to investigate the effect of CSF on neurospheres obtained from neural stem cells of normal H-Tx rats.

Materials and methods

Neurospheres were obtained by dissecting the lateral walls of lateral ventricles of normal PN7 H-Tx rats. The tissue was mechanically dissociated to obtain a single-cell suspension. The cells were plated at 20 viable cells/μl in serum-free medium supplemented with N2 and EGF (20 ng/ml). CSF samples were collected from normal and hydrocephalic H-Tx rats and from hydrocephalic and non-hydrocephalic human patients. After 4 days *in vitro*, neurospheres were treated with CSF (1:10) and further cultured for 1–2 days. Neurospheres were analysed by immunocytochemistry, transmission and scanning electron microscopy and immunoblotting.

Results

After 3–4 days *in vitro*, clonally-derived neurospheres were seen. Neurospheres were formed by undifferentiated cells and a small number of immature neurons, astrocytes and ependymal cells. The cells were joined together by adherent junctions. After addition of normal and hydrocephalic CSF, neurospheres disassembled and cells started to migrate and grow. Neurons and astrocytes became more differentiated and readily distinguishable. Normal CSF displayed a twofold higher neurite extension promoting activity than hydrocephalic CSF. Western blots of both types of CSF, using a set of antibodies including those against the subcommissural organ secretory proteins, revealed qualitative and quantitative differences. Preliminary findings indicate that CSF of human hydrocephalic patients has an effect similar to that of hydrocephalic H-Tx rats. Studies are in progress to identify the CSF-polypeptides responsible for (i) disassembling of neurospheres and (ii) stimulating and/or interfering with neuronal differentiation.

Conclusion

(i) The CSF of normal H-Tx rats promotes neuronal and glial differentiation from neurospheres; (ii) The CSF from hydrocephalic H-Tx rats interferes with neuronal differentiation.

Acknowledgements

Supported by Grant 1030265 from Fondecyt, Chile