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Proteomics study of CSF composition in the developing H-Tx rat

M Nabiyouni*, C Storey, J Owen-Lynch and J Miyan

Address: Biomolecular Sciences, UMIST, Manchester, UK

Email: M Nabiyouni* - M.Nabiyouni@stud.umist.ac.uk

* Corresponding author

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Previous work of our group has shown that CSF from affected H-Tx rat fetuses aged E20 and E21 has an inhibitory effect on cortical cell proliferation in culture but that this effect does not occur with normal CSF. The inhibitory effects are removed by heating the CSF suggesting that the active component(s) is a protein(s).

These data led us to the use of a proteomics approach to investigate the protein profile of CSF and compare this between normal and affected CSF from fetuses at late stages of gestation. We compared CSF collected from the lateral ventricles of affected fetuses at E17-E21 to CSF collected from the cisterna magna of normal animals of the same ages. 2D gel results showed significant differences between normal and affected CSF at the different ages. MALDI-TOF Mass spectroscopy was used to identify some of the different proteins and Liquid Chromatography-Mass Spectroscopy was used to confirm the data.

Interesting components, such as a reelin precursor, NGF precursor, alpha-feto protein and apolipoprotein where found to change with age and whether collected from normal or affected animals. Reelin, NGF and alpha-feto protein are all known to have an important role in cortical development. Reelin precursor was present in CSF from normal animals but was absent in H-Tx fetuses aged E21 and P0 (the day of birth). NGF was present in both animal groups but in reduced amounts in the H-Tx rats on Day E21. Alpha-feto protein was present in both and in the same amount. With increasing age the total amount of protein decreases in the CSF of both normal and affected fetuses.

These findings strengthen the hypothesis that CSF has got an important role in normal brain development and in the abnormal development associated with early-onset hydrocephalus.

Proteomics provides an important method for the identification of CSF proteins particularly where the amount of CSF available is small; all other methods require much larger amounts which is a major restriction for the present study. LC-MS is a new technique that identifies large numbers of proteins in very small samples and this will aid the identification, purification and analysis of candidate proteins involved in normal and abnormal development.