Province de Liège
Enseignement

HAUTE ECOLE DE LA PROVINCE DE LIEGE

Cellular and tissular distribution of EFHC1, a protein mutated in juvenile myoclonic epilepsy (JME)

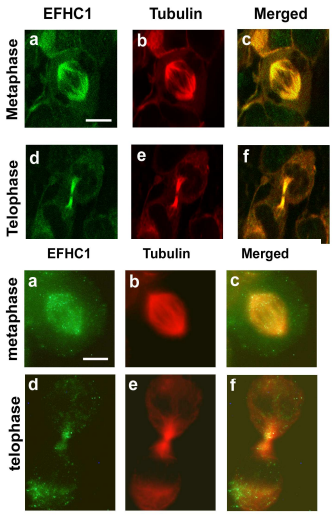
[1]C. Léon, [1]L. de Nijs, [1,4]G. Chanas, [2]T. Ikeda, [3]A.V. Delgado-Escueta, [1]L.F. Bettendorff*, [1]T. Grisar and [1]B. Lakaye

INTRODUCTION

A novel gene, *EFHC1*, mutated in juvenile myoclonic epilepsy (JME) encodes a protein with three DM10 domains of unknown function and one putative EF-hand motif. Our aim is to characterize the basic functions of EFHC1.

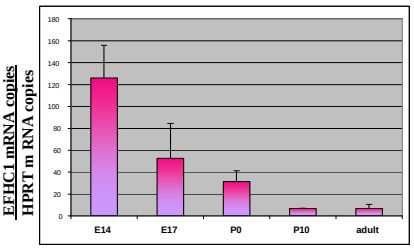
We recently demonstrated an association of EFHC1 with the centrosome and the mitotic spindle in different cell lines (de Nijs *et al.*, 2006), suggesting a potential implication of this protein in cell division. The aim of this work is to determine the neuroanatomical pattern of EFHC1 expression during mouse brain development.

EFHC1 associates with the mitotic spindle in nerve cells

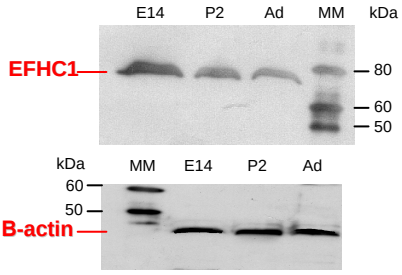


Multipotent neural stem cells (A) and neuroblastoma cells (B) were immunostained with anti-EFHC1 antibody (in green) and anti-alpha-tubulin antibody (in red). Two mitotic phases are shown: metaphase (a,b,c) and telophase (d,e,f).

EFHC1 expression is higher in mouse brain during development



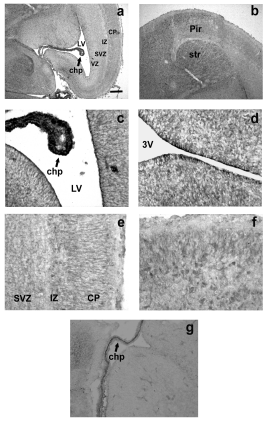
Quantitative RT-PCR analysis was performed on mRNA from mouse brains at different ages: E14, E17, P0, P10 and adult.



Western blot on mouse brains lysates at different ages: E14, P2, Ad. The membrane in A was revealed with anti-EFHC1 antibody and in B using anti-β-actin antibody as control.

EFHC1 distribution in embryonic mouse brain

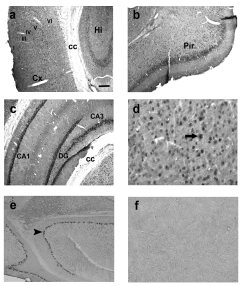
In the brain at E16, EFHC1 immunostaining was observed in neocortex (a), piriform cortex and striatum (b). The protein was also strongly expressed at the border of the cerebral ventricles as well as in the choroids plexus (c,d). At the subcellular level, the signal is localized mainly in the cytoplasm (e) but, in some cells in the choroids plexus (c), the piriform cortex (f) and the deep layers of the cortical plate (e), the signal appeared also in the nucleus.



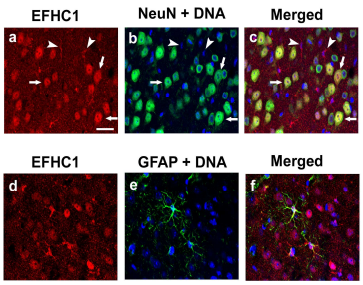
Coronal sections of embryonic mouse brain at E16 were stained with anti-EFHC1 antibody.

EFHC1 distribution in adult mouse brain

In adult, the staining was present in all cortical layers (a) however it was more intense in pyramidal cells in cortical layer V but also in the piriform cortex (b) and in the hippocampus (CA and DG) (c). The signal was particularly concentrated in the nucleus (d). EFHC1 stained also very nicely Purkinje cells of adult cerebellum (e).



EFHC1 is expressed in both neurons and astrocytes



Double immunofluorescent labeling adult moude cortex with anti-EFHC1 (in red) and anti-NeuN (b, c, in green) or anti-GFAP (e, f, in green) antibodies. DNA was stained with TOPRO-3 (in blue).

CONCLUSION

Our results suggest that EFHC1 is a protein which expression levels, distribution and subcellular localization are developmentally regulated in brain. These finding strongly suggest that EFHC1 may contribute to the development of the central nervous system.

AFFILIATIONS

[1]*Center for Cellular and Molecular Neurobiology, University of Liège, Liège, Belgium*
[2]*Dept. of Biol. Sci., University of Tokyo, Tokyo, Japan*
[3]*UCLA, Los Angeles, California, USA*
[4]*Haute Ecole de la Province de Liège, Belgium*

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