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# Isolation of cDNA clones of the rat mRNAs expressed preferentially in the prenatal stages of brain development.

Usui H, Ichikawa T, Miyazaki Y, Nagai S, Kumanishi T.

Department of Molecular Neuropathology, Niigata University, Japan.

## Abstract

For better understanding of the molecular mechanisms underlying the developmental processes of the mammalian brain, we isolated rat fetal brain-enriched (FBE) cDNA clones, whose corresponding mRNAs were expressed at least 5-fold more in the fetal brain than in the adult brain. Our modified differential screening procedure, which utilized a two-vector (pT7T3D and pBluescript) system and showed low background levels of colony hybridization for screening, efficiently identified 64 candidate FBE clones from a small number (475) of colonies in the fetal brain cDNA library. After subsequent second screening of the candidate FBE clones by Northern blot analysis, we successfully isolated 22 distinct FBE clones. The nucleotide sequence analysis of the 22 FBE clones revealed that 13 of them had no significant matches to the sequences reported in the databases, whereas 9 of them matched previously reported sequences (alpha-tubulin M alpha 1, beta-tubulin M beta 5, thymosin-beta 10, stathmin, beta-tubulin M beta 2, alpha-internexin, ferritin Lg chain, **neuronatin** and amphoterin), most of which have been shown to be down-regulated during brain development. We also found that the Northern blot analysis in the second screening could be replaced by cDNA library DNA-Southern blot analysis, in most clones corresponding to relatively abundantly expressed mRNAs. Thus, once the cDNA library is constructed, clone selection will be possible in such clones without the use of additional RNA or Northern blot in screening, allowing the analysis of small brain regions of interest.

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