Combine methylation sensitive electrophoresis and suppression subtractive hybridization to identify differential methyla

張文彥、李泰林

E-mail: 9417461@mail.dyu.edu.tw

ABSTRACT

ABSTRACT Imprinting gene phenomenon may be taking place in fertilized period, according the allele to producing different epigenetic marks in this period, utilize these mark to distinguish paternal or maternal allele, have already known that DNA methylation may influence the nuclear chromatin structure or been involved in a certain protein and nucleic acid combination. And the nucleic acid methylation is one kind of the epigenetic mechanism, more evidences present that the DNA methylation controlled the expression of some basic genes and tissue specifical genes.Igf2 and Snrpn are expressed from the paternal genome while H19,Igf2r and Mash2 are expressed from the maternal genome, at the time tracking the cell expression position of chimeria mouse. We combine methylation sensitive electrophoresis and suppression subtractive hybridization to identify differential methylation fragments between cortex and hypothalamus. Take the advantage of methylation sensitive restriction enzyme digestion and gel electrophoresis and suppression subtractive hybridization, we isolated a differential methylation DNA fragment between cortex and hypothalamus. The methylated site located in intron two of Zswim6.The differential methylation was confirmed by southern hybridization and methylation sensitive PCR. The expression level of Zswim6 mRNA is the highest in liver among 8 tissues, also the DNA methylation at the intron. Another short form of swim6 is also examination, the expression level is similar among tissues. Putting together, we propose the differential methylation make the post transcriptional machinery select different exon among tissues. Key Words : DNA methylation, Imprinted gene, methylation sensitive, suppression subtractive hybridization, methylation sensitive restriction enzyme, swim6

Keywords : methylation

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