Interaction of AGN-1 protein, which binds to protein phosphatase 6, with tau alternative splicing variants mRNAs

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We cloned AGN-1 that interacted with protein phosphatase 6 as a molecule highly expressed in nephritic rat kidney. Our previous studies also revealed that AGN-1 was co-localized with U1snRNP and splicing factor SC35 participating in the synthesis of tau alternative splicing variants, 4R-tau and 3R-tau. In addition, we showed that AGN-1-siRNA treatment altered the expression ratio of 4R-tau/3R-tau mRNA in Neuro2a cell and that AGN-1 may localize with 4R-tau and 3R-tau mRNAs in U1 spliceosome. In this study, we investigated an interaction of AGN-1 protein with tau alternative splicing variants mRNAs. By using nuclear and cytosol fractions prepared from Neuro2a cells, RNA immunoprecipitation analysis was carried out with anti-AGN-1 antibody, followed by RT-PCR to detect 4R-tau and 3R-tau mRNAs bound to AGN-1protein. RNA immunoprecipitation with nuclear fraction showed PCR fragments derived from 4R-tau and 3R-tau mRNAs, accompanied with an equal binding level of 4R-tau and 3R-tau mRNAs. However, in cytosol fraction, 4R-tau mRNA amount bound to AGN-1 protein was higher than 3R-tau mRNA. This result suggests that AGN-1 protein interacts with 4R-tau and 3R-tau mRNAs, and may involve in the differential regulation of a metabolism of 4R-tau and 3R-tau mRNAs.