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PhD thesis title - The effect of ALS-associated FUS mutations on U7 snRNP activity and the expression of core canonical histone genes in neuronal cells

Amyotrophic lateral sclerosis (ALS) is a neurodegenerative disorder which involves the progressive loss of upper as well as the lower motor neurons of the nervous system. ALS is usually categorized as sporadic ALS (sALS) which constitutes for about 90 to 95% of the cases and autosomal dominant familial ALS (fALS) comprising of the remaining 5 to 10% cases. Different genes involving varied mutations have been discovered to be involved in fALS with mutations in *FUS* gene being one of them. FUS is a ubiquitously expressed predominantly nuclear protein involved in DNA repair and transcription regulation, RNA splicing and export to cytoplasm.

The ALS-linked FUS mutations (ALS-FUS) are mainly observed in the conserved C terminus region and are associated with mislocalization and cytoplasmic inclusion formation. We have previously shown that the FUS protein interacts with U7 snRNP and is involved in the regulation of transcription and 3' end processing efficiency of the replication-dependent histone (RDH) genes.

Based on the available data and our previous observation, the aim of the thesis was to analyze the effect of ALS-FUS mutations on U7 snRNP activity and the efficiency of transcription and processing of core canonical histone pre-mRNAs as the molecular mechanism underlying ALS. From the data obtained from our experimental results, it shows that ALS-FUS mutations along with itself mislocalize U7 snRNP in the cytoplasmic aggregates of cellular models and rat primary neurons. This cytoplasmic entrapment of ALS-FUS along with U7 snRNP has a significant impact on the transcriptional activity and aberrant 3' end processing of RDH pre-mRNAs. In proliferating neuroblastoma cells transfected with ALS-FUS mutants we observed inhibition of RDH gene transcription and impairment of the 3' end pre-mRNA maturation. At the same time, in terminally differentiated cells we observed no obvious impairment in the 3' end maturation but significant downregulation of transcript level due to inhibited transcription.

The obtained results indicate U7 snRNP is one of the snRNP whose activity is affected by ALS-FUS mutations resulting in a range of complications from inefficient splicing to disturbed RDH transcripts processing. Undoubtedly, there are even more parameters which are yet to be explored. But summarizing the available data, increased DNA damage and disrupted RDH pre-mRNA processing caused due to ALS-FUS mutations can cause genomic instability and may be the molecular mechanisms underlying in ALS.