# Nonsense mutations in the prionogenic domain of the yeast SUP35 gene induce prion conversion of the Sup35 protein

P-02.4-16

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Sporadic forms of prion diseases can be considered as a result of violation of folding of corresponding prionogenic proteins. These disorders, which are traditionally considered accidental, can also result from rare and difficult to track events - in particular, certain mutations in the genes of these proteins that could occur in individual somatic cells.

In this paper, we simulated the early stages of the process described above using the prionogenic protein Sup35 of the yeast Saccharomyces cerevisiae. It is known that the emergence of the prion state of proteins requires the overproduction of these proteins. We searched for mutant variants of Sup35 protein inducing prionogenesis at the normal expression level. Using error-prone PCR, we mutagenized the prionogenic N-domain of the SUP35 gene and selected two mutant SUP35 alleles, which, being located on a low-copy plasmid, caused a significant increase in de novo formation of the [PSI+] prion. Nonsense mutations were present in both mutant alleles leading to the synthesis of shortened Sup35 fragments of 98 and 108 amino acid residues (a.a.), respectively. Then we obtained a set of 12 alleles that encode Sup35 N-terminal fragments 19 to 240 a.a. long. It turned out that the fragments of 75 to 112 a.a. led to a great increase of prionogenesis, while fragments shorter than 73 a.a. or longer than 123 a.a., as well as wild type Sup35 protein, did not promote prion formation. This process depended on the presence of the [PIN+] prion. Importantly, the [PSI+] variants selected in the study were able to be maintained in the cells after loss of the mutant SUP35 allele. We also discovered that SUP35 mRNA could undergo splicing into transcript encoding the shortened Sup35 isoform, which was also highly prionogenic. Our data evidence that truncated forms of amyloidogenic proteins might initiate the spontaneous formation of amyloids, which can then spread via prion-like mechanism, resulting in sporadic amyloid disease.