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REPEAT EXPANSION DISORDERS

New evidence for RNA gelation in repeat expansion disorders

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Sol–gel phase transitions in nucleotide repeat-containing RNA could contribute to the pathogenesis of certain neurological disorders, a study published in *Nature* suggests. Nucleotide repeat expansions have been identified as causative factors in conditions including amyotrophic lateral sclerosis (ALS), muscular dystrophy and Huntington disease, but the mechanisms through which these repeats lead to neurotoxicity have not been fully elucidated.

In most repeat expansion disorders, the repeat-containing RNA forms punctate aggregates, termed RNA foci, in the nuclei of cells. The new research, by Ankur Jain and Ronald Vale, was prompted by previous demonstrations that purified proteins can undergo phase separation *in vitro* to form granules. "As a fun side project, we decided to test if nucleic acids phase separate," explains Vale. "Initial experiments with DNA worked really well, and allowed us to carefully map out how nucleic acids undergo liquid–liquid phase separation."

The researchers showed that RNA molecules containing 47 CAG or CUG repeats formed spherical clusters *in vitro*, whereas scrambled RNA molecules of equivalent length and base composition remained dispersed in solution. The RNA within the clusters was immobile, indicating that the molecules had become cross-linked to form a gel. CAG-repeat RNAs were found to exhibit similar phase-separation behaviour in a live-cell reporter assay.

To further explore the relevance of RNA gelation to neurological disease, Jain and Vale examined the properties of RNA molecules containing GGGGCC repeats. Expansion of this hexanucleotide repeat in the *C9orf72* gene has been strongly linked to familial forms of ALS and frontotemporal dementia.

As expected, RNA molecules containing expanded GGGGCC repeats underwent gelation, and the propensity of these molecules to form RNA foci increased in parallel with the number of repeats. Interestingly, the repeat length at which foci were observed in the majority of cells (16–29 repeats) was comparable to the estimated threshold number of repeats for disease onset (>20) in *C9orf72* expansion carriers.

"The key finding is that multivalent base-pairing interactions in nucleic acids can lead to aggregation of RNA, which results in the formation of RNA foci," concludes Vale. "Aggregation of proteins is a well-accepted cause of many neurodegenerative disorders, and our work suggests that aggregation of RNA might also be a culprit in neurodegeneration in patients with nucleotide repeat expansions."

The researchers are exploring the therapeutic potential of drugs that disrupt RNA–RNA base pairing. They found that the chemotherapy drug doxorubicin could break up RNA foci, but this agent targets all RNA and DNA in the cell, and has an unfavourable adverse effect profile. Therefore, the search continues for drugs that specifically promote disaggregation of disease-related RNA repeats.

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ORIGINAL ARTICLE Jain, A. & Vale, R. D. RNA phase transitions in repeat expansion disorders. *Nature* http://dx.doi.org/10.1038/nature22386 (2017)