RNA polymerase II drives FUS exclusion from damaged chromatin

Sylvia Varhoshkova[1], *Stoyno Stoynov*[1]

[1]*Institute of Molecular Biology, Bulgarian Academy of Sciences, Acad. G. Bonchev Str. Bl.21, 1113 Sofia, Bulgaria*

sylviamarinova@bio21.bas.bg

FUS is an RNA-binding protein that accumulates at DNA damage sites in a PAR-dependent manner following irradiation. Interestingly, FUS is progressively excluded from regions of complex DNA damage, though the mechanism and functional relevance of this exclusion have remained unclear. Here, we demonstrate that inhibition of transcription-associated cyclin-dependent kinases (CDK7, CDK9, CDK12/13) or targeted depletion of RNA polymerase II subunit RPB1 prevents this exclusion. Notably, RPB1 itself is excluded with kinetics similar to FUS. Additionally, FUS mutants with impaired phase separation fail to be excluded, implicating liquid-liquid phase separation (LLPS) in the process. Importantly, FUS exclusion facilitates efficient 53BP1 recruitment to DNA lesions *in vivo*. There findings suggest a mechanism in which RNAPII removal from complex DNA lesions leads to FUS exclusion from damaged chromatin to support coordinated DNA repair.