Students' Annual Seminar

Monitoring global changes in chromatin compaction states upon localized DNA damage with tools of fluorescence anisotropy

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DNA in cells is subject to continuous damage from endogenous and exogenous sources. Failure to repair such damage can cause mutations that give rise to diseases like cancers and neurodegenerative diseases. DNA is compactly packed in the form of chromatin inside the nucleus. The compact chromatin structure acts as a barrier for efficient repair of DNA, but also can make DNA more refractory to damage. Chromatin has to be remodelled transiently for DNA damage responses (DDR) to work efficiently. We use fluorescence anisotropy imaging of histone H2B-EGFP to image the compaction state of the chromatin. H2B-EGFP anisotropy maps can be used to map out the euchromatin or heterochromatin like regions in live and fixed cells. We induce local double strand breaks with laser micro irradiation and observe the dynamics of chromatin compaction states in live cells for 2 h after damage, and then later correlate it to markers of damage using immunofluorescence. Compact nodes of chromatin nodes were formed, and overall there was a global compaction of chromatin upon local irradiation. We also observed the dynamics of endogenous repair factors using chromo bodies for PARP1 and PCNA, which were immediately recruited to the site of damage. PARP1 showed a more transient recruitment than PCNA. Together these studies are yielding insight into changes in chromatin compaction states and associated responses upon local induction of clustered double strand breaks.

Friday, Apr 12th 2019 2:30 PM (Tea/Coffee at 1:30 PM) Seminar Hall, TIFR-H