

Internal Seminar

Developing tools for investigating DNA damage response (DDR) dynamics in living cells

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To maintain genomic integrity, cells have specialised DNA repair mechanisms that are essential for proper cellular and organismal function. Defects in these repair mechanisms are associated with the development of many cancers, neurodegenerative diseases and aging. In-depth molecular, biochemical and genetic analyses have defined the molecular framework involved in cellular DNA-repair pathways, however only recently cell-biological approaches have revealed important roles for the spatial and temporal organization of the chromatin and DNA-repair machinery during the recognition of DNA damage and the assembly of repair complexes. The temporal sequence of events at a repair site still remains somewhat elusive and new assays are needed to follow repair dynamics in live cells. In this talk, I will describe our recent attempts towards understanding the dynamics of DNA damage response in living cells. This is an ongoing study, and I will describe my efforts towards generating stable cell-lines expressing both chromatin and repair markers with fluorescently tagged fusion proteins. Furthermore, I will describe a novel method to study DDR in live cells using nano antibodies called Chromobodies. I will also talk about the advantages of Chromobodies above the earlier developed scratch labelling assay in studying DDR in live cells. Immunofluorescence is used to confirm the nature of observed foci and quantify the damage response. In the days to come we plan to combine Chromobodies or Scratch labelling assay with fluorescently tagged chromatin protein to follow the dynamics of chromatin, replication and repair foci in the same cells for the first time, to elucidate the sequence of events at a site of damage.

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2:00 PM (Tea/Coffee at 1:45 PM)

Seminar Hall, TCIS