CHROMOSOMAL ABERRATIONS INDUCED BY IONISING RADIATION AND MITOMYCIN C FOR FANCONI ANAEMIA DIAGNOSIS

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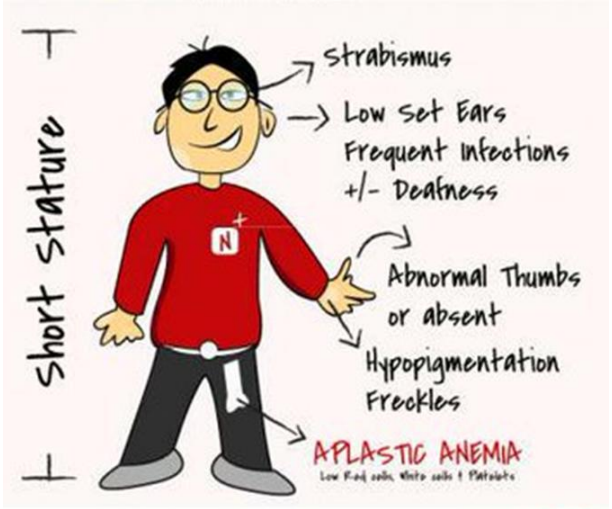
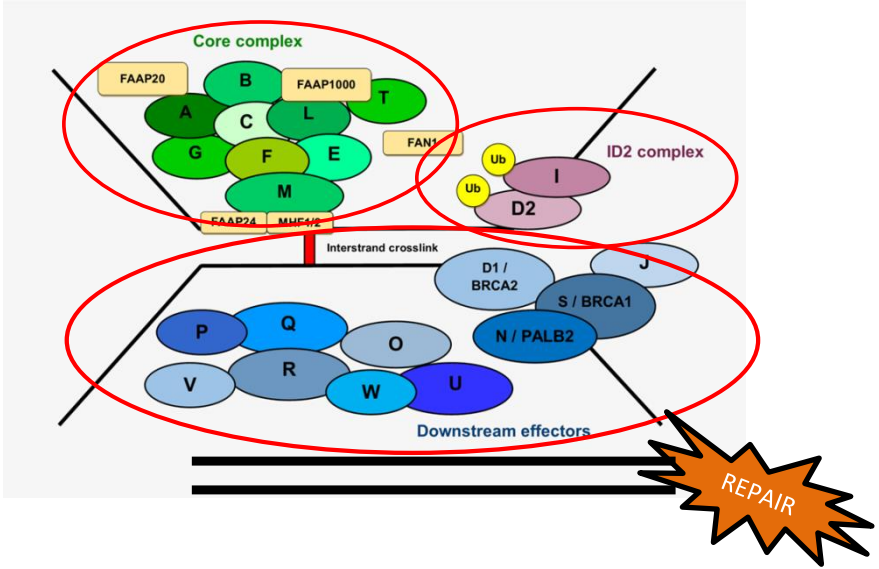
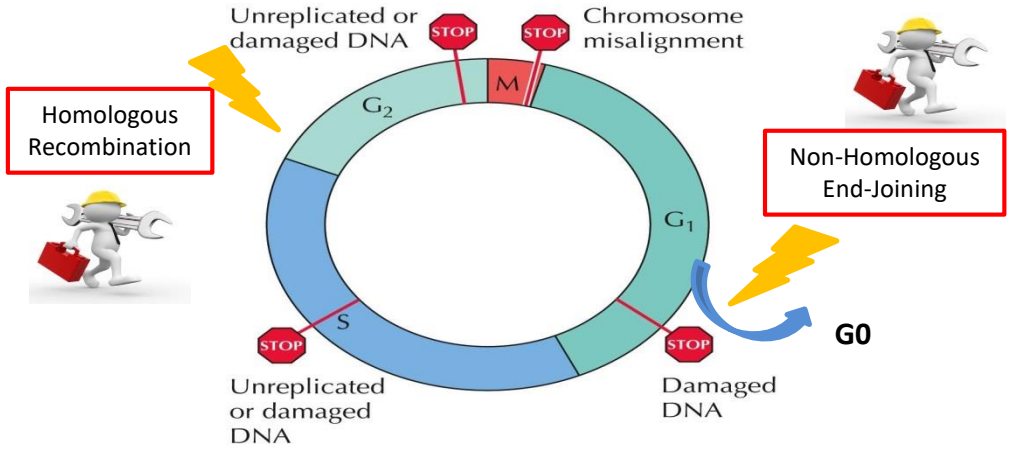




- Fanconi Anaemia patients treated with radiotherapy have shown increased clinical radiosensitivity
- Radiosensitivity is defined as the susceptibility of cells to the damaging effects of ionising radiation

Investigate the chromosomal instability and radiosensitivity of Fanconi Anaemia patients by using 3 chromosomal aberration assays:

G0, S/G2 CBMN assay and Mitomycin C assay



G0 MN

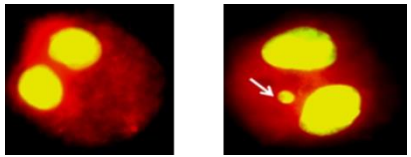
- *In vitro* irradiation of cultures to 2 and 4 Gy
- Immediate after irradiation, cell division stimulated by phytohaemagglutinin (PHA)
- Addition of Cytochalasin B (Cyto B) 23 hrs after irradiation blocks cytokinesis

S/G2 MN

- Assay initiated by addition of PHA prior to irradiation
- *In vitro* irradiation of cultures to 2 and 4 Gy, 72 hrs post stimulation
- Addition of Cyto B immediately after irradiation

MMC MN

- Similar to G0 MN assay
- DNA damage induced by addition of MMC: 0.02 µg/ml and 0.1 µg/ml
- Cultures stimulated with PHA and 23 hrs later, Cyto B blocks cytokinesis



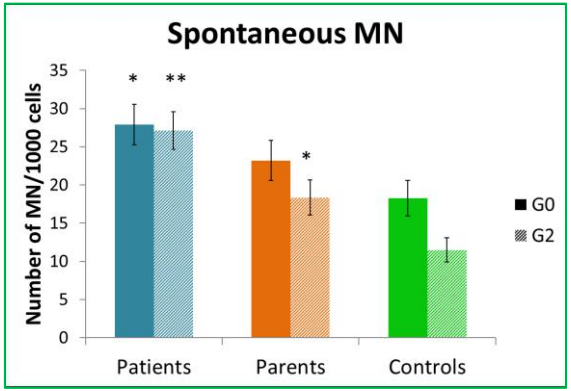
FA patients: n = 14

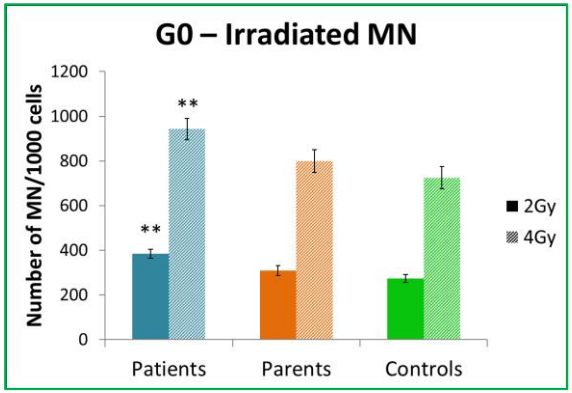
FA parents: n = 14

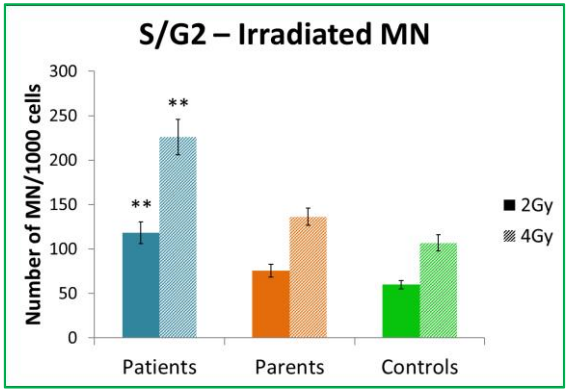
Controls: n = 14

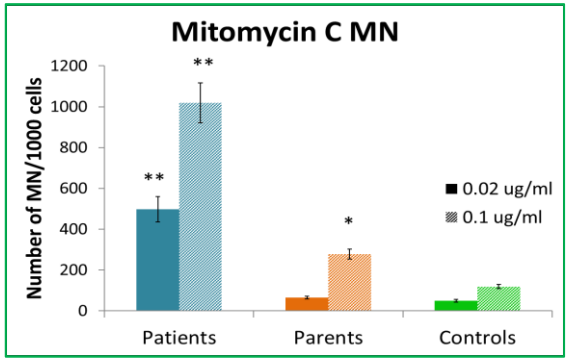
*Significantly different from controls

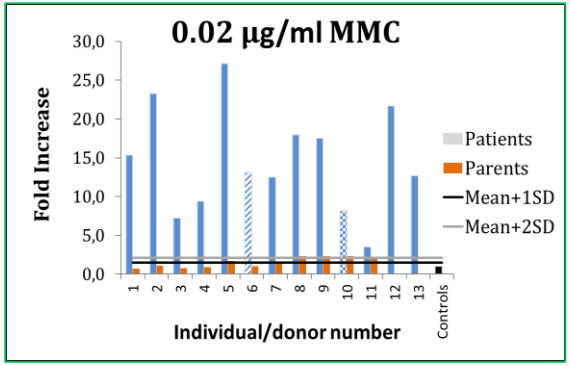
**Significantly different from parents and controls

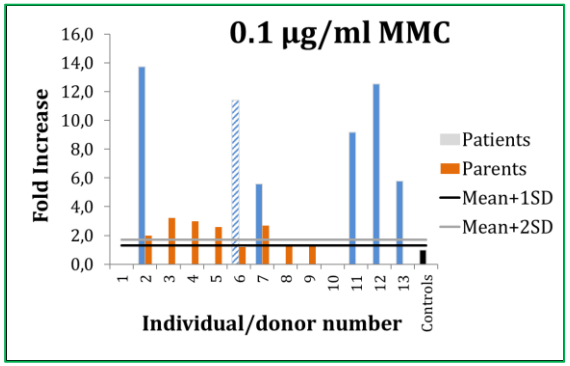












Conclusion

- FA patients can be distinguished from healthy controls with all 3 MN assays
- MMC MN assay: Identification of heterozygotes from controls is possible using the highest MMC concentration
- Advantages of the MN assay:
  - Time effective
  - Less labour intense
  - Ease of analysis
  - Low cost
- Radiation-induced MN data is important information for further treatment/screening
- Further validation of these functional MN assays on larger patient cohort is ongoing