Drug screening to reduce genome instability in Fanconi Anemia

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The only treatment of bone marrow failure (BMF) in Fanconi Anemia (FA) patients is hematopoietic stem cell (HSC) transplantation, which has a high survival rate but also important limitations including lack of available donor in many families and further increase in cancer risk in post-transplanted patients. In addition, while curative for BMF, HSC transplantation will not prevent solid tumours. In order to find new candidate drugs for preventing bone marrow failure and cancer in FA patients, we have adapted and scaled down the flow-cytometry micronucleus (MN) assay to 96 multiwell plates in order to screen large number of drugs in a short period of time. MN are biomarkers of chromosome fragility in FA as they derive from acentric chromosome fragments that are left behind in anaphase and appear in the cytoplasm of daughter cells as small nuclei. In addition, flow cytometry data allowed to check cell cycle progression which is also altered in FA cells. We initially selected 10 antioxidants and p38 inhibitors previously described in the literature as potentially beneficial for FA cells, including our previous studies in FA IPS cells (Liu et al., Nature Com 2014). We tested them for their ability to suppress spontaneous and DEB-induced chromosomal instability in FANCA-deficient cells. Our results show that antioxidants NAC, resveratrol and quercetin and anti-inflammatory drugs danazol and dasatinib reduced chromosome fragility to basal levels while also reducing G2/M arrest. Quercetin and danazol were the most effective. On the other hand, antioxidants tempol and α-lipoic acid did not exert a positive effect on chromosome fragility, nor did aldehyde chelant cysteamine, adh2 inductor alda-1 or anti-inflammatory drug doramapimod. We also tested 84 antioxidant compounds from a redox library and observed a beneficial effect of 22 additional drugs for their ability to reduce genomic instability in DEB, formaldehyde and/or acetaldehyde-treated FANCA-deficient cells. Drugs that reduced the damage caused by at least two chromosome instability inducers were tested for their ability to improve the spontaneous chromosome fragility and we are now assaying the selected drugs *in vivo* in FANCA KO mice, using the *in vivo* flow cytometry MN assay and checking for beneficial effects in terms of hematopoietic stem cell performance.