Supplementary methods

Cellular proliferation

Cells were labelled with the CellTrace™ CFSE Cell Proliferation Kit (Thermo Fisher Scientific) and cellular fluorescence of 10 000 cells analysed at 24h, 48h and 72h on the FACSCalibur [1].

Cell cycle

Cells ($5x10^5$) were fixed in ethanol overnight at -20^{o}C and labelled with propidium iodide (40µg/ml Sigma-Aldrich), containing 3.8mM sodium citrate (VWR International, Leicestershire, UK), 1.5U of RNase Cocktail Enzyme Mix (Applied Biosystems, Thermo Fisher Scientific), in PBS for 3h at 4^{o}C [1]. Labelled cells (100 000) were analysed on the FACSCalibur.

Telomere repeats

DNA was extracted using the DNeasy Blood & Tissue Kit (Qiagen, UK). The quantity and quality of extracted DNA was assessed using the[2]) and the size and fragmentation by electrophoresis (Agilent Technologies 2100 bioanalyzer). The relative telomere length measured by quantitative PCR (qPCR) for telomere repeats relative to a single-copy gene, 36B4 [3]. The difference between the Ct values of the telomere reaction (T) and single copy gene (S) (T/S ratio) was used to generate the average telomere length per genome.

Migration

Migration was determined up to 72h as previously described [1]. Migration Index (MI) = total migrated area relative to the size of the spheroid core at 0h [1].

MRP1 activity

MRP1 dependent efflux activity was measured using the calcein-F efflux assay [4].

Effect of doxorubicin

Cells were treated with increasing concentrations of the MRP1 substrate doxorubicin (3.5-224nM) for 48h and viable cell number counted using the trypan blue exclusion assay (Vi-cell, Beckman Coulter) [5].

References

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