Aberrant DNA damage response and receptor tyrosine kinase signaling in non-small cell lung cancer tumor initiating cells

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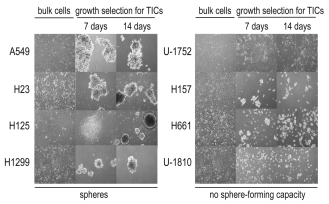
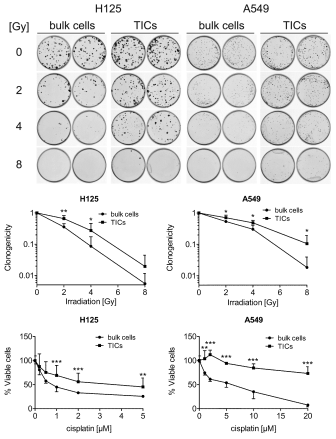
Aim

- Non-small cell lung cancer (NSCLC) is characterized by a chemo- and radiotherapy refractory phenotype.
- Here we aimed to elucidate if chemo- and radiotherapy resistance in NSCLC is attributed to a TICs phenotype and delineate components involved.

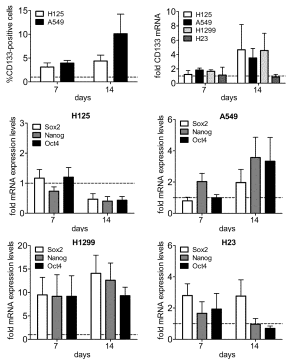
TICs can be isolated from NSCLC cell lines



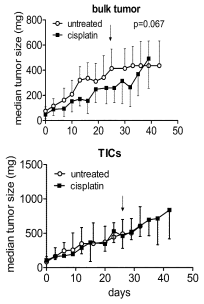
NSCLC TICs show resistance to IR and cisplatin



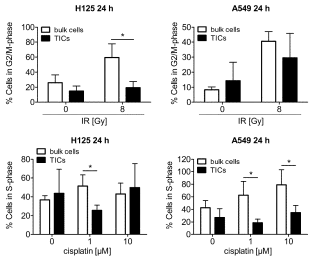
NSCLC TICs show increased expression of CD133 and some but not all stem cells markers



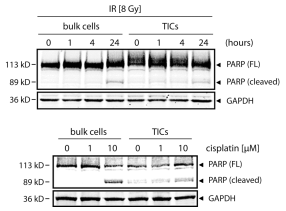
H125 TICs display a cisplatin resistant phenotype *in vivo* in mouse xenografts



NSCLC TICs show impaired G2/M or S-phase arrest after IR and cisplatin treatment



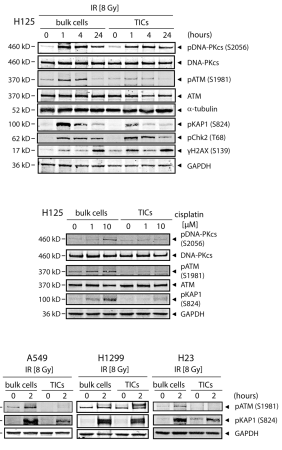
NSCLC TICs display reduced PARP cleavage after IR and cisplatin treatment



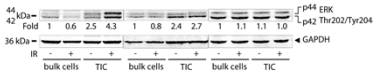
Results and conclusions

- CD133-positive NSCLC cancer stem cells or tumor initiating cells (TICs) showed increased survival and reduced apoptotic response after irradiation (IR) and cisplatin compared to bulk cells.
- A cisplatin-refractory phenotype of TICs was confirmed *in vivo* in a mouse xenograft model.
- TICs displayed less pronounced G2 cell cycle arrest or S-phase block after IR or cisplatin, respectively.
- TICs exhibited increased γH2AX at baseline, and diminished DNA damage-induced phosphorylation of DNA-PK, ATM, KAP-1 and H2AX.
- Analysis of phosphorylated receptor tyrosine kinases displayed lower IGF1R phosphorylation and expression and a higher degree of ERK phosphorylation in TICs as compared to bulk cells.
- MEK inhibition reduced TIC viability alone and decreased clonogenicity upon IR suggesting that MEK and downstream signaling impart on TIC radiation response.
- **We demonstrate that reduced apoptotic response, less pronounced cell cycle arrest, altered DNA repair and receptor tyrosine kinase signaling in NSCLC TICs are possible factors contributing to their therapy resistance.**

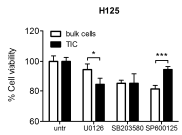
DDR and DNA repair proteins show sub-optimal activation in NSCLC TICs



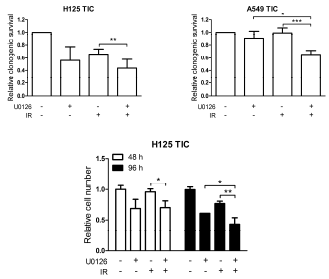
Increased ERK phosphorylation in NSCLC TIC



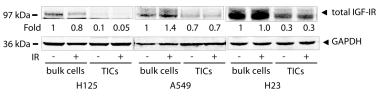
MEK and p38 inhibitors reduce cell viability of TICs



MEK inhibitor in combination with IR reduce cell number and clonogenic survival in NSCLC TICs



NSCLC TICs show reduced IGF1R expression



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