Thermosensitive liposome delivery to the brain after FUS-induced blood-brain barrier (BBB) opening for the treatment of glioblastoma.

Paul Cressey[1], Chris Payne[2], Antonios Poulopoulos[2], Maya Thanou[1*]
King's College London, Institute of Pharmaceutical Sciences[1], Surgical & Intervention Engineering[2]

Glioblastoma is an extremely aggressive brain tumour and currently has limited effective treatment options, with median survival of only 14 months.[1] This low survival is attributed to the blood brain barrier (BBB) especially in early-stage glioblastoma.[2] In this study, we formulated imageable drug loaded thermosensitive liposomes (iTSL-S_M/C_C, fig 1A) and tested their efficacy against U87 *in vitro* and *in vivo* (subcutaneous). In addition, we performed focused ultrasound (FUS)-induced BBB opening to deliver the iTSLs into non-tumour bearing mice. Liposome uptake was determined and quantified *in vivo* and *ex vivo* using MRI and near-infrared fluorescence (NIRF) respectively.

FUS-induced BBB opening was performed in mice either with or without administration of iTSLs. Mice were also administered iTSLs without BBB opening. FUS parameters: Pressure = 450 kPa, cycles = 500, PRF = 5, pulses = 600. SonoVue microbubbles (3 ml/kg) were administered i.v. simultaneously to sonication. iTSL's (7 ml/kg) were administered i.v. 10 minutes after sonication. T_1-weighted MRI was performed in a 9.4-T magnet 2 and 4 hours after BBB opening, to allow time for iTSLs to accumulate in the brain for optimal T_1 contrast. NIRF imaging was also performed on perfused and excised mouse brains after the final time point. In addition, the ability of iTSL-S_M/C_C to supress tumour growth was investigated with single treatments against U87-MG subcutaneous xenograft tumours (10 mg/kg SN38, 42 ^oC, 10 min).

iTSLs were successfully formulated with both gadolinium and NIRF lipid conjugates. iTSLs were also loaded with SN-38 (1.11 mg/ml) and carboplatin (0.84 mg/ml) (iTSL-S_M/C_C). iTSL-S_M/C_C (10mg/kg SN-38) treatment caused an increase in survival when administered alone and a significant effect when combined with FUS (42 °C, 10 min). MRI and NIRF imaging confirmed localized increased uptake of iTSLs where FUS-induced BBB opening was performed (Figure. 1. C, yellow arrows), compared to the contralateral control side. Localised uptake was also confirmed compared to mice without BBB opening. These results suggest that MRI/NIRF-tagged drug loaded liposomes are suitable drug carriers for the treatment of glioblastoma using FUS-induced BBB opening.

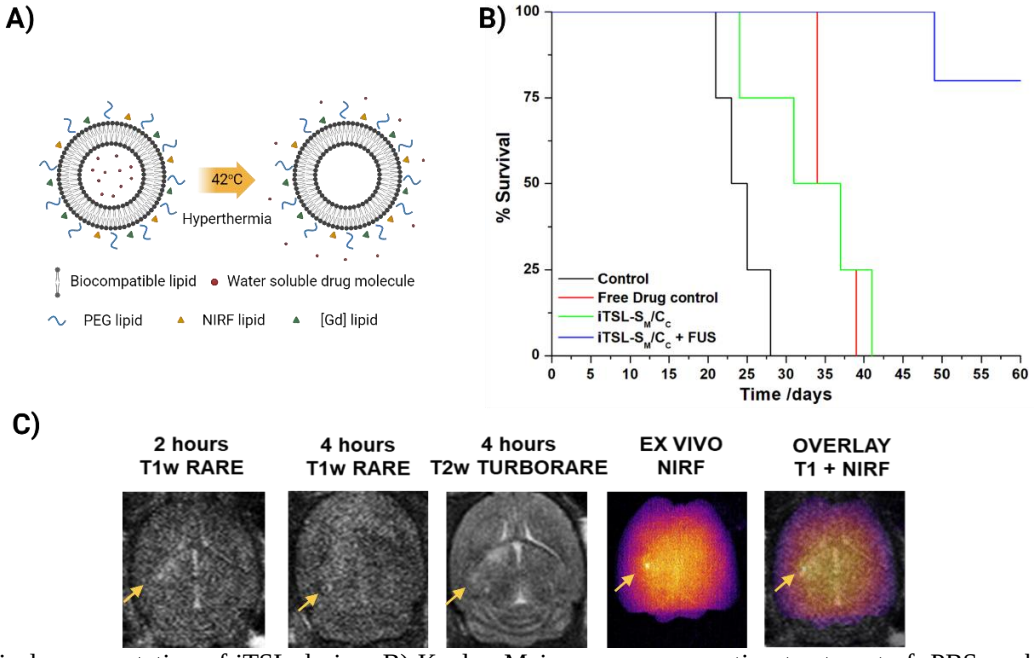

Fig.1 A) Graphical representation of iTSL design. B) Keplan-Meier curve representing treatment of: PBS, carboplatin + irinotecan (10 mg/kg), iTSL-S_M/C_C (10 mg/kg SN-39) and iTSL-S_M/C_C +FUS (10 mg/kg SN-38, 42 °C, 10 min) against U87-MG subcutaneous xenograft tumours. C) Example T1w and T2w MRI images of non-tumour bearing mouse after BBBO. Overlay between ex-vivo NIRF image and 2h T1w RARE highlights co-localisation of delivery.

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References: 1. S. Mohammed, *et al.*, *Reports of Practical Oncology and Radiotherapy*, 2022, **27**, 1026–1036. 2. J. N. Sarkaria, *et al.*, *Neuro Oncol*, 2018, **20**, 184–191.