**Efficient gene silencing in brain tumors with hydrophobically modified siRNAs**

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**Supporting Information**

**Fig. S1.** Neurosphere viability following Chol-hsiRNA treatment. GBM8 neurospheres were treated with 0-5 µM Chol-hsiRNA for 72 h. Cell viability was measured via alamarBlue cell viability assay and presented as percent of untreated control (n = 3 biological replicates, mean +/- SD, see Supplemental Methods).

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**Table S1.**

**Table S2.**

**Supplemental Methods**

**alamarBlue® cellular viability assay**

GBM8 cell viability following Chol-hsiRNA administration was assessed by the alamarBlue® (Life Technologies) assay, a fluorescence-based protocol that measures metabolism in proliferating cells. GBM8 neurospheres were treated with 0–5 μM Chol-hsiRNA for 72 h in a 96-well plate. Cells were then incubated for 0–2.5 h with fresh medium containing 10% alamarBlue. Absorbance at 570 and 600 nm was measured with a Tecan Infinite M200 microplate reader after 2 h. Cellular viability was calculated according to the manufacturer's protocol and normalized to an untreated control (n = 3 biological replicates, mean ± SD).