

## Supporting Information

### Ovarian Spheroid Based Tumor Model Represents Vascularized Tumor and Enables the Investigation of Nanomedicine Therapeutics

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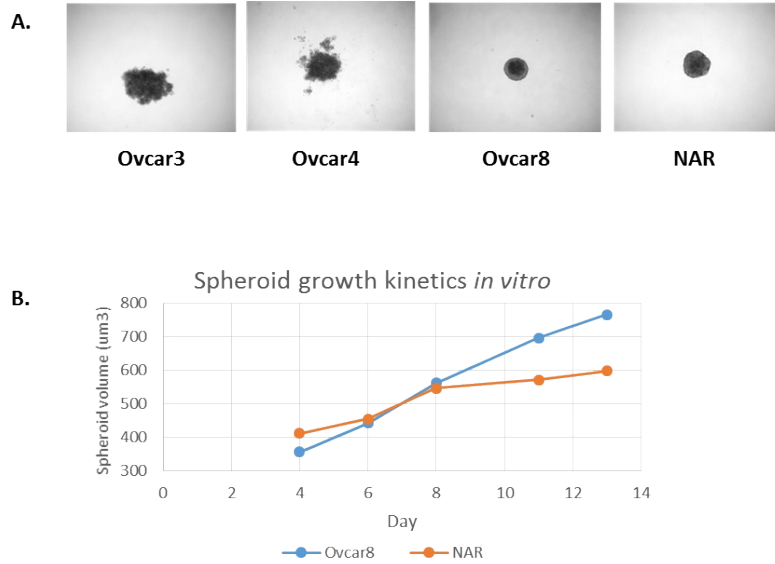
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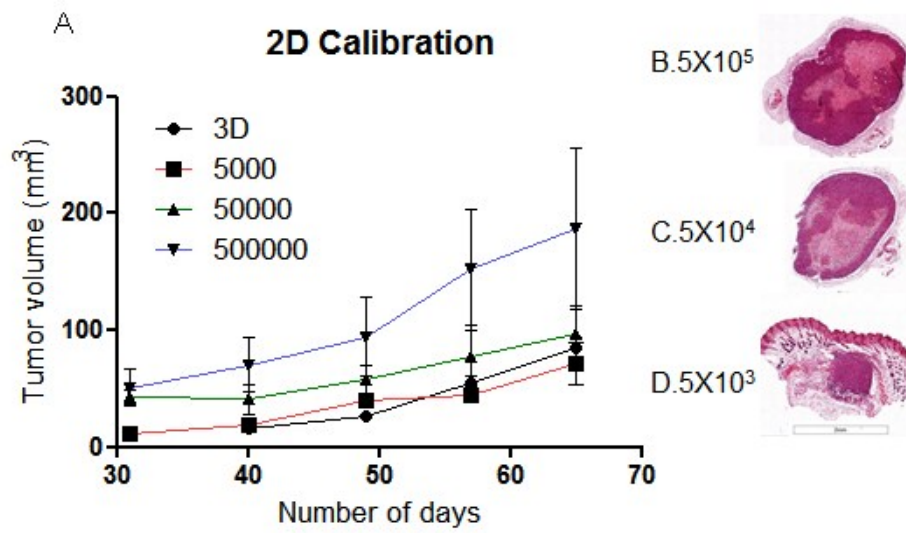
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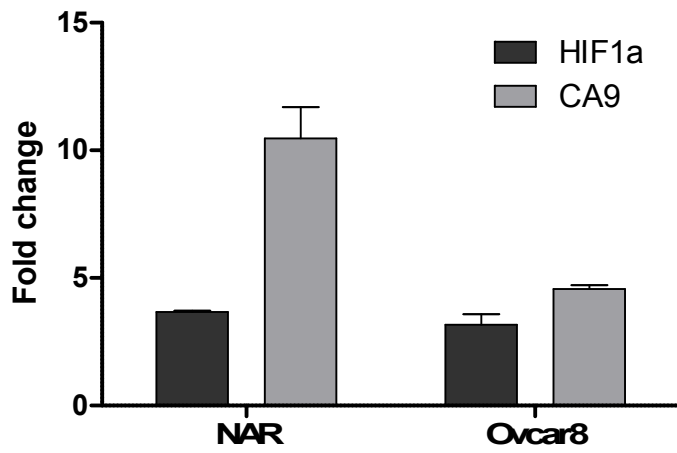
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**Figure S1. Characterization of Spheroids in vitro.** **A.** 2000 cells were seeded in ultra low attachment 96 –well plate to screen for their ability to form spheroid and observed under microscope after 4 days. Ovc3 and Ovc4 formed loose aggregates whereas spheroids of Ovc8 and NAR appeared round and robust (Phase-contrast Microscope Zeiss Axiovert 200/M). **B.** Growth kinetics of Ovc8 and NAR spheroids formed post-seeding 2000 cells per well on day 0 (n=15, 3 independent experiments). Ovc8 spheroids exhibited higher growth kinetics than NAR in terms of spheroid volume.



**Figure S2.** Optimization of 2D model. **A.** Growth kinetics H&E sections of *in vivo* tumor injected with **B.**  $5 \times 10^5$  cells (Day 35),  $5 \times 10^4$  cells (Day 58),  $5 \times 10^3$  cells (Day 58),  $n=3$ , Scale 2mm.



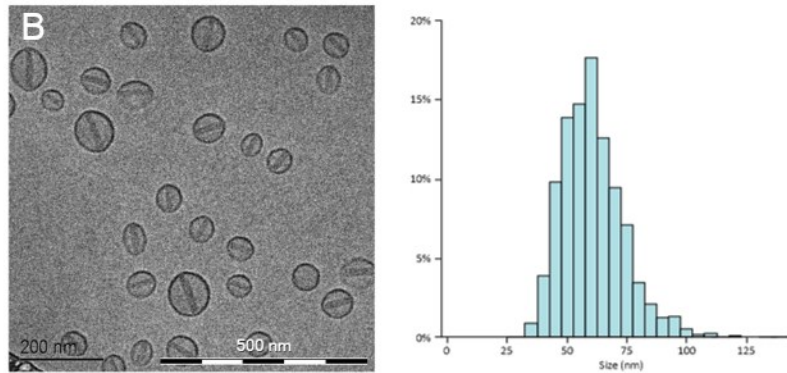
**Figure S3.** Relative Quantification of genes in 3D spheroids (day 4) as compared to cell lines grown as 2-dimension culture. Both the hypoxia-related genes CA9 and HIF1 $\alpha$  are expressed 3-10 fold more when grown as 3D spheroids.

## Doxil® composition

Total Lipids	15.6 mg/mL
Lipids Ratio	Cholesterol 18.8 % (w/w) HSPC 60.4% (w/w) MPEG-DSPE 20.8% (w/w)
Encapsulation	94.2%
Drug-to-Lipid Ratio	0.13 mg/mg

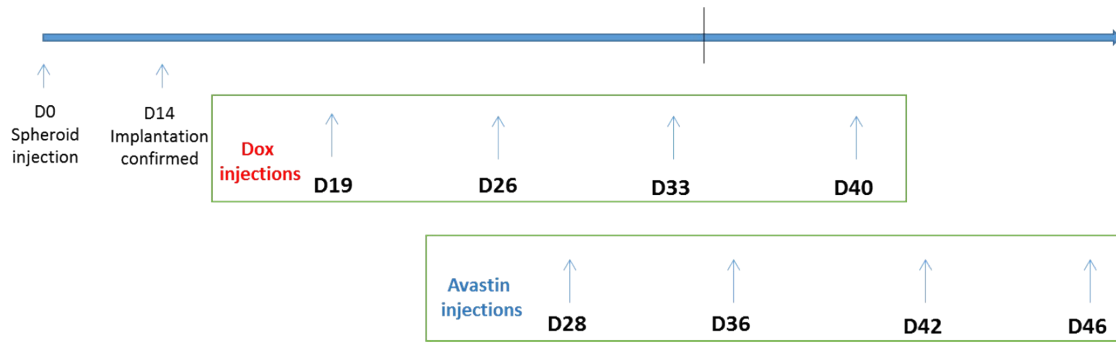
**A**

Z-Average (d), (nm)	PDI	ζ-Potential (mV)
81	0.06	-31



No. of Images	No. of Particles	Mean Size (nm)	Unilamellar Liposomes (%)	Elongatedness (Aspect Ratio)
16	1260	61 ± 13	99	1.17 ± 0.14

**Figure S4. Characterization of Doxil: A.** Physicochemical Characterization, **B.** Cryo-Transmission Electron Micrograph (TEM) Analysis



**Figure S5.** Scheme of experiment and treatment with following groups- 1. Control, 2. S-Dox, 3. Doxil, 4. Avastin, 5. Avastin and S-Dox, 6. Avastin and Doxil.