

Supplementary Data

Supplementary figures:

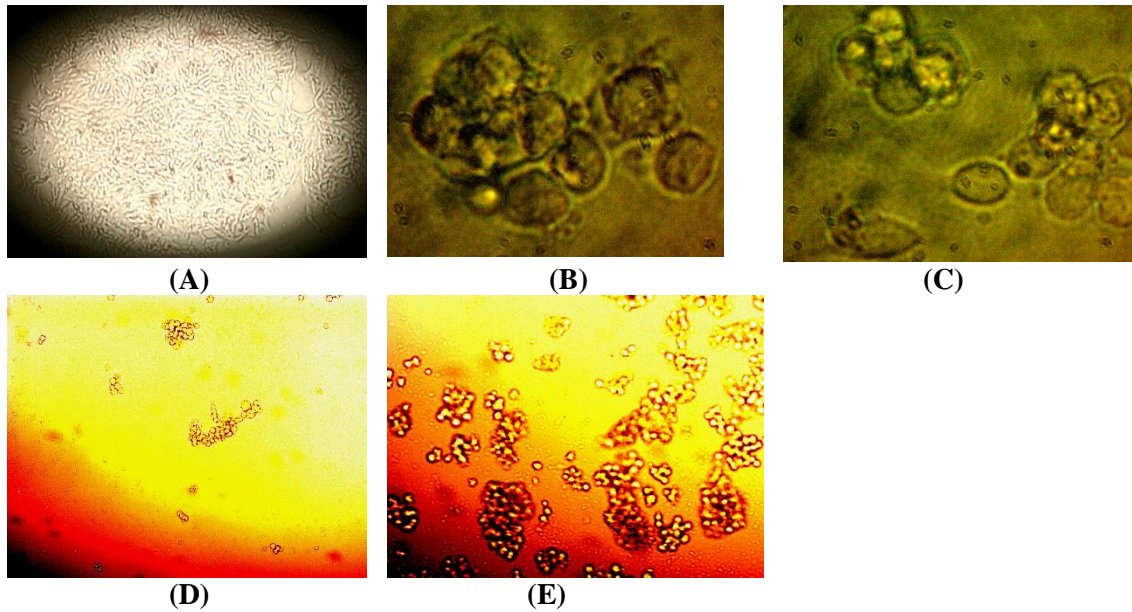
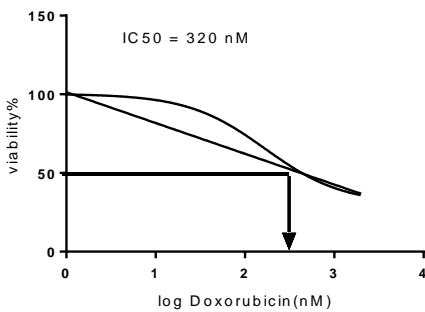


Figure 1 3D Morphological Characteristics of Mammospheres generated from MDA-MB231 Cell line

A) MDA-MB231 cell line, captured by confocal light microscope, lens 10X.

B, C) MDA-MB231 mammospheres, captured by confocal light microscope, lens 40X.

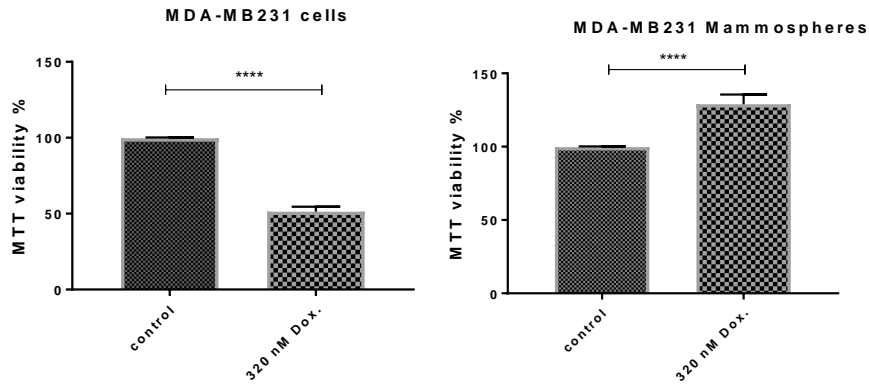
D, E) MDA-MB231 mammospheres captured by confocal light microscope, lens 20X.



(A)

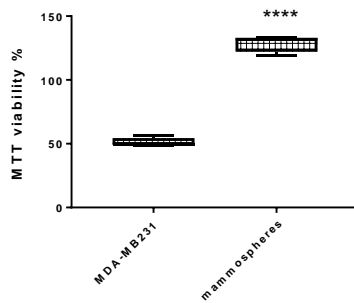


(B)



(C)

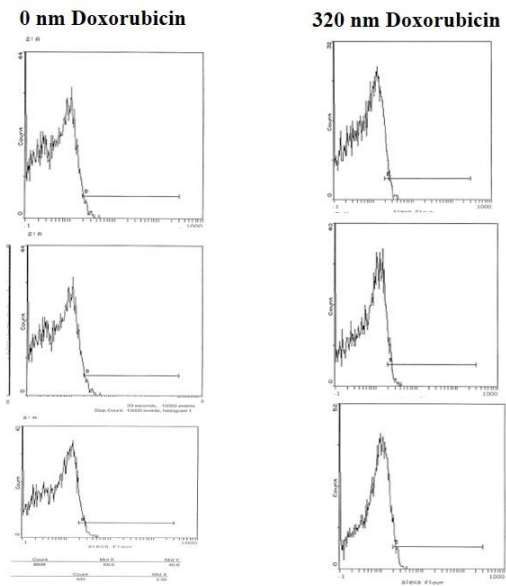
(D)



(E)

Figure 2 Comparing the Chemosensitivity Response to Doxorubicin in MDA-MB231 Cell line and Mammospheres.

- A) MTT assay for determining the inhibitory concentration (IC₅₀) of Doxorubicin in MDA-MB231 Cells. IC₅₀ was calculated after plotting log of the concentrations on the x-axis while the viability percentage was plotted on the Y-axis, data was fit with nonlinear regression Curve fit.
- B) MTT assay for examining the viability in mammospheres post-treatment with step-wise concentrations of Doxorubicin 250-2000 nM (not all doses are shown), data was fit using linear regression Analysis.
- C) Compared viability of non-treated and 320 nM Doxorubicin treated MDA-MB231 cells, P values unpaired student's t-test.
- D) Compared viability of non-treated and 320 nM Doxorubicin treated MDA-MB231 mammospheres, P values unpaired student's t-test.
- E) Viability in MDA-MB231 cells post-treatment with 320 nM Doxorubicin is compared to the viability of mammospheres treated with the same dose of the drug P values unpaired student's t-test.



(A)

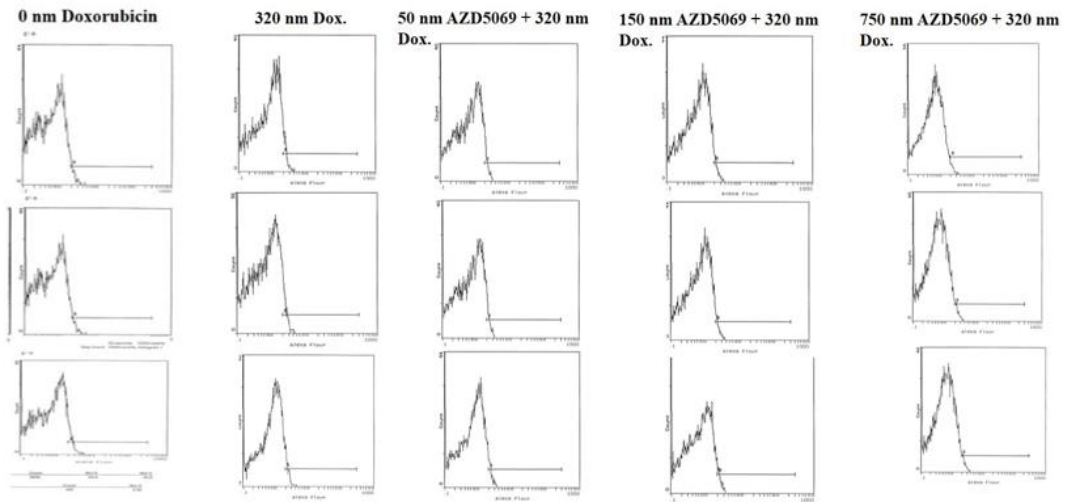
0 nm Doxorubicin	Hist	Region ID	%	Count	Mnl X
	2	B B	3.74	363	2.50

(B)

320 nm Doxorubicin	Hist	Region ID	%	Count	Mnl X
	2	B B	5.23	430	2.40

Figure 3 Doxorubicin Induces Higher CXCR2 Expression level in MDA-MB-231 Mammospheres.(flow cytometric charts)

- A) Flow Charts for CXCR2 expression in mammospheres.
- B) CXCR2 percentages obtained from flow cytometric analysis.

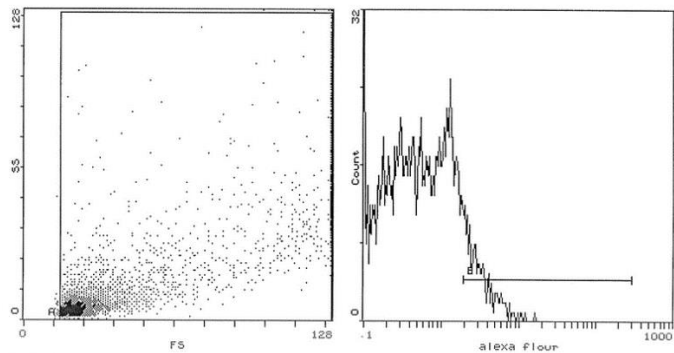


Treatment	Hist	Region ID	%	Count	Mnl X
0 nm Doxorubicin	2	B B	3.74	363	2.50
0 nm AZD5069+ 320 nm Dox.	2	B B	4.73	408	2.39
50 nm AZD5069+ 320 nm Dox.	2	B B	3.16	273	2.61
150 nm AZD5069+ 320 nm Dox.	2	B B	2.95	258	2.38
750 nm AZD5069+ 320 nm Dox.	2	B B	1.59	148	2.46

Figure 4 AZD5069 inhibits Doxorubicin mediated CXCR2 overexpression and restores primary levels of the receptor

- (A) Flow Charts for CXCR2 expression in mammospheres.
- (B) CXCR2 percentages obtained from flow cytometric analysis.

(A)



(B)

Hist	Region ID	%	Count	Mnl X
2	B B	9.56	755	3.96

Figure 5 CXCR2 Expression level in MDA-MB-231 Cells

- A) Flow Charts for CXCR2 expression in MDA-MB-231 Cells.
- B) CXCR2 percentages obtained from flow cytometric analysis.