

S. 1. Representative clonogenic plates of SiHa and SiHa/RR upon challenging with 0, 2, 4, 6, 8 Gy of radiation followed by incubation for 3 weeks. Plated colonies were fixed in methanol: acetic acid followed by crystal violet staining. Images were captured in Gel Doc (BioRad, Chemidoc XRS+)

A

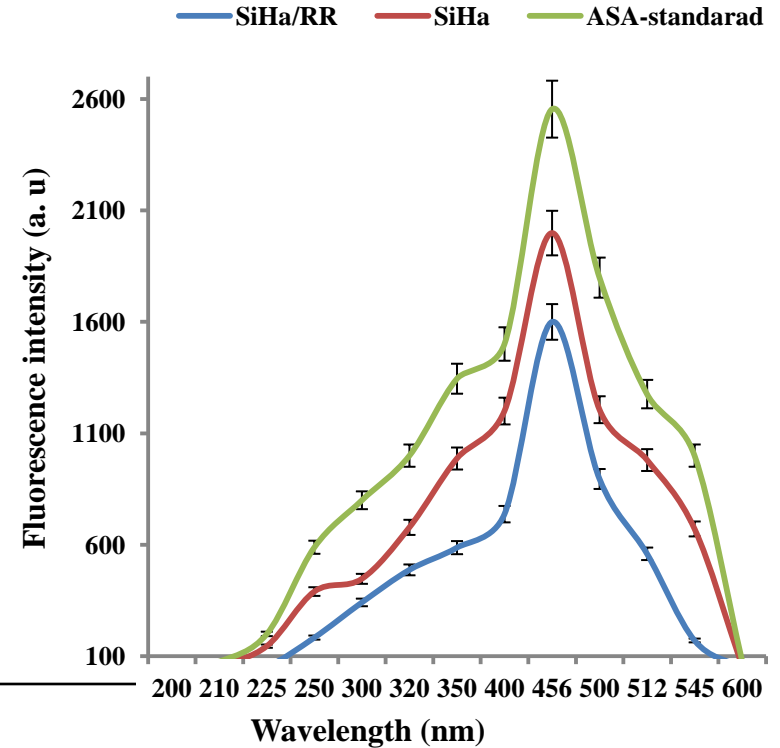
Cell Line	D ₀ (Gy)	D ₁ (Gy)	SF ₂
SiHa	4 ± 0.5	2 ± 0.5	0.6 ± 0.02
SiHa/RR	5.5 ± 0.5	3 ± 0.8	0.71 ± 0.18

A. Calculation of D₀, D₁, SF₂ values of SiHa and SiHa/RR upon exposure to 0, 2, 4, 6, 8 Gy of radiation has been represented in a tabulated form. [D₀ and D₁ values are the radiation doses at which the survival is reduced to 37% and 90% respectively compared to non-irradiated cells. SF₂ = Survival fraction of cells at 2 Gy radiation].

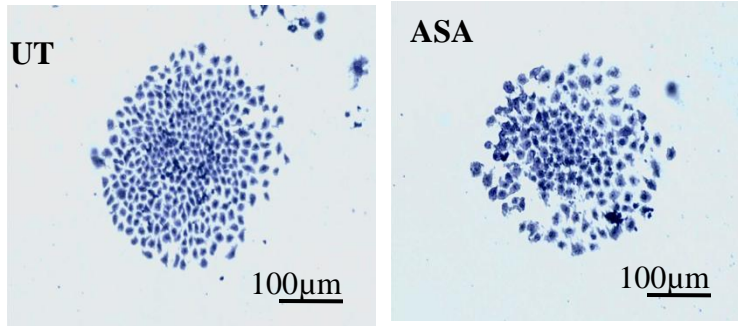
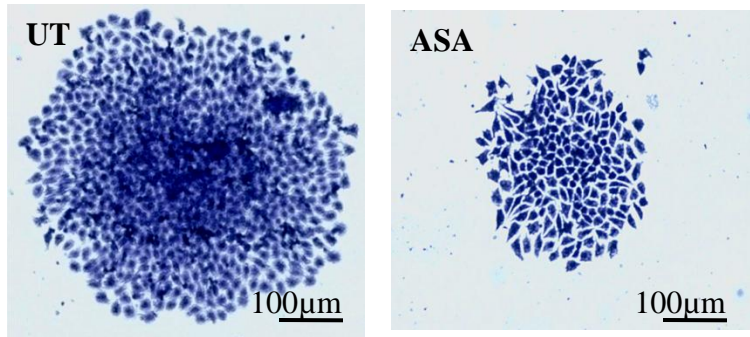
B

Cell Line	ASA Dose (μM)				
	IC ₁₀	IC ₂₀	IC ₃₀	IC ₅₀	IC ₉₀
SiHa	1 ± 0.5	5 ± 0.5	7.5 ± 0.18	20 ± 3.5	200 ± 2.5
SiHa/RR	2.5 ± 0.5	7.5 ± 1.5	17 ± 2.5	40 ± 5.5	1000 ± 0.5

B. Tabulated representation of calculated IC₁₀, IC₂₀, IC₃₀, IC₅₀ and IC₉₀ values of ASA for SiHa and SiHa/RR cells based on the data obtained from MTT assay.

S. 2.**C**

C. SiHa and SiHa/RR cells were exposed to 5μM of ASA and its intracellular uptake was determined by fluorescence intensity of ASA at a range of wavelength from 220nm to 600 nm. Maximum intensity of ASA was observed at 460nm. SiHa and SiHa/RR treated with ASA also showed highest intensity at 460nm.

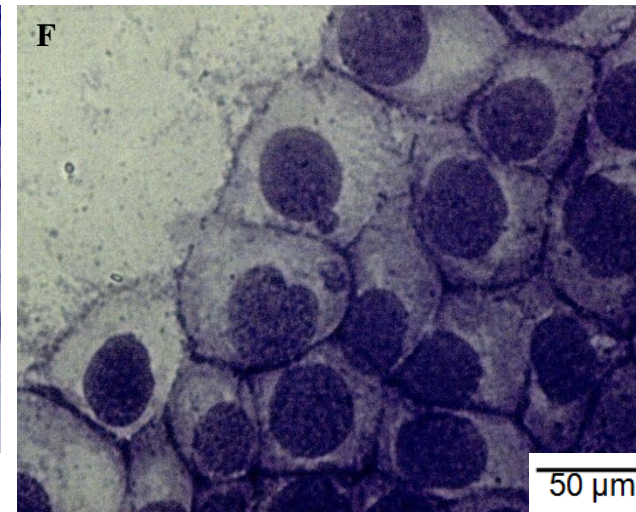
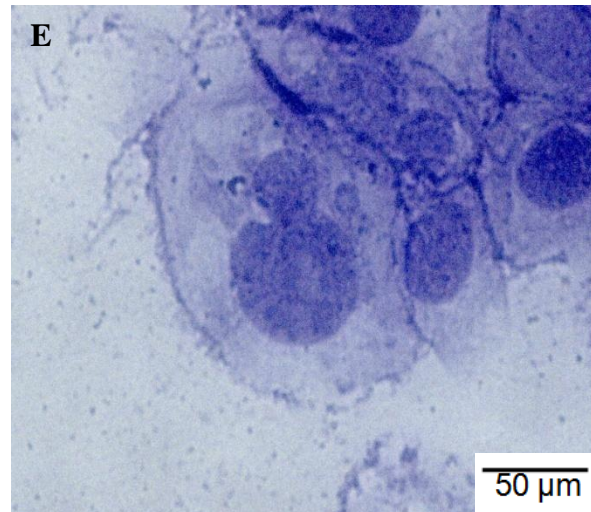
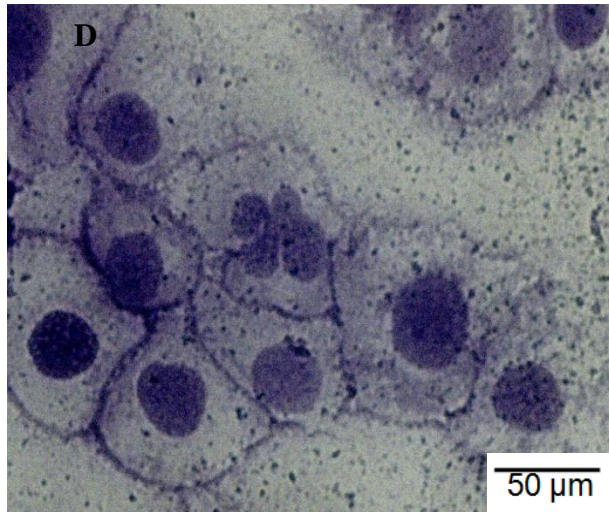
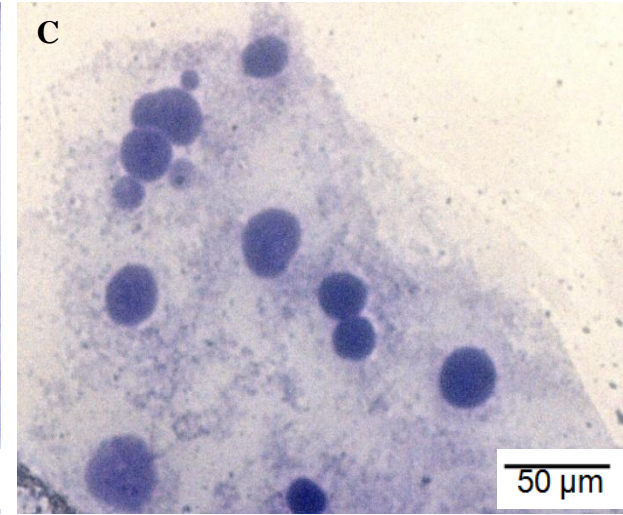
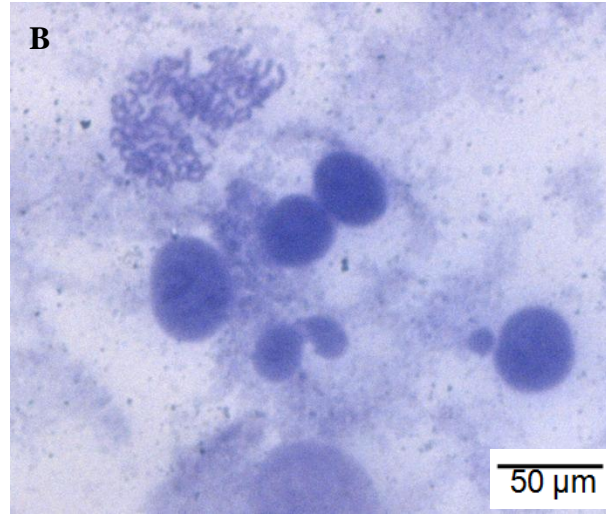
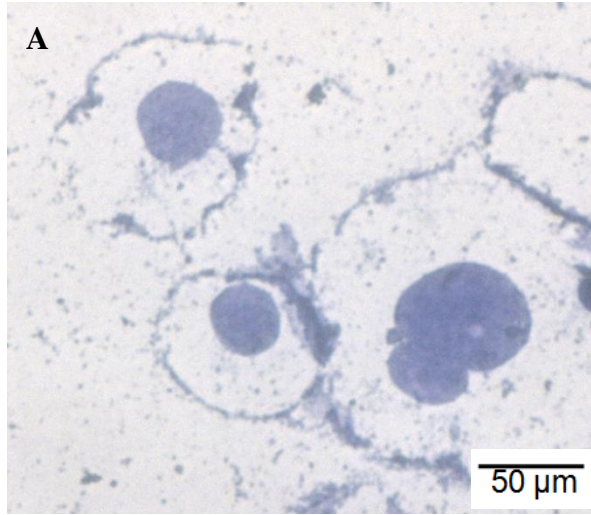
A**SiHa****SiHa/RR**

A. Representative images showing average colony size of SiHa and SiHa/RR formed after 3 weeks of cell seeding. Crystal violet stained colonies were observed under inverted microscope (Olympus); at 40X magnification in untreated and ASA treated SiHa (top panel) and SiHa/RR (bottom panel) respectively.

B

Cell Line		D_0 (Gy)	D_1 (Gy)	SF_2
SiHa	+RT	4 ± 0.5	2 ± 0.5	0.6 ± 0.02
	RT+ASA	2 ± 0.2	1 ± 0.1	0.36 ± 0.06
SiHa/RR	+RT	5.5 ± 0.5	3 ± 0.8	0.71 ± 0.18
	RT+ASA	3.96 ± 0.04	1.8 ± 0.1	0.5 ± 0.09

B. Effect of ASA treatment in improving radiosensitivity in SiHa and SiHa/RR cells were observed upon challenging cells with, 2, 4, 6, 8 Gy of radiation in the presence and absence of ASA treatment. The Surviving Fraction values (D_0 , D_1 and SF_2) were quantitated and depicted in tabulated form.



SiHa/RR

S. 4 Representative images of SiHa/RR showing features of ASA induced mitotic catastrophe like **A.** giant cells with nuclear bridge, **B.** micronuclei with chromosomal arrest, **C.** simultaneous polynucleation and micronucleation, **D.** quadrinucleated cells, **E and F.** giant cells with micronucleation (Panel 2 and 3).