Electronic Supplementary Material (ESI) for Biomaterials Science.

Supporting Information

Multicomponent 5-fluorouracil loaded PAMAM Stabilized-Silver Nanocomposites Synergistically Induce Apoptosis in Human Cancer Cells

Ishita Matai[a], *Abhay Sachdev*[a] *and P.Gopinath**[a,b]

[a]*Nanobiotechnology Laboratory, Centre for Nanotechnology,*

[b]*Department of Biotechnology,*

Indian Institute of Technology Roorkee, Roorkee, Uttarakhand-247667, India.

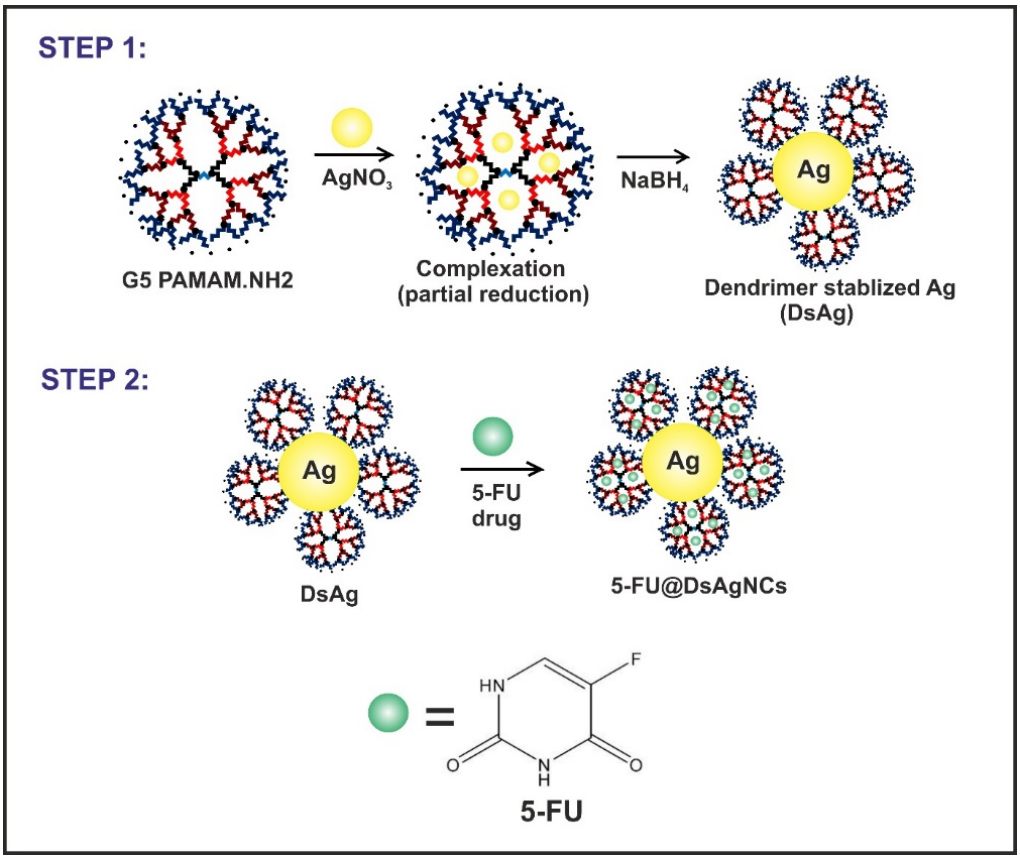
Fax: +91-1332-273560;

Tel: 91-1332-285650;

E-mail: pgopifnt@iitr.ernet.in, genegopi@gmail.com

Gene	Primers
Beta-actin	Forward: 5’ CTGTCTGGCGGCACCACCAT 3’ Reverse : 5’ GCAACTAAGTCATAGTCCGC 3’
p53	Forward: 5’ TGGCCCCTCCTCAGCATCTTAT 3’ Reverse : 5’ GTTGGGCAGTGCTCGCTTAGTG 3’
Caspase 3	Forward : 5’ TTCAGAGGGGATCGTTGTAGAAGTC 3’ Reverse : 5’ CAAGCTTGTCGGCATACTGTTTCAG 3’
C-myc	Forward : 5’ CCAGGACTGTATGTGGAGCG 3’ Reverse : 5’ CTTGAGGACCAGTGGGCTGT 3’
Bax	Forward : 5’ AAGCTGAGCGAGTGTCTCAAGCGC 3’ Reverse : 5’ TCCCGCCACAAAGATGGTCACG 3’
Bad	Forward : 5’ CCTTTAAGAAGGGACTTCCTCGCC 3’ Reverse : 5’ ACTTCCGATGGGACCAAGCCTTCC 3’
Bcl-xl	Forward : 5’ ATGGCAGCAGTAAAGCAAGC 3’ Reverse : 5’ CGGAAGAGTTCATTCACTACCTGT 3’
Bcl-2	Forward : 5’ TCCGCATCAGGAAGGCTAGA 3’ Reverse : 5’ AGGACCAGGCCTCCAAGCT 3’

Table S1. Forward and reverse primer sequences used in the gene expression studies.



Scheme S1. Schematic description of formation of dendrimer stabilized AgNPs (STEP 1) and 5-FU@DsAgNCs (STEP 2).

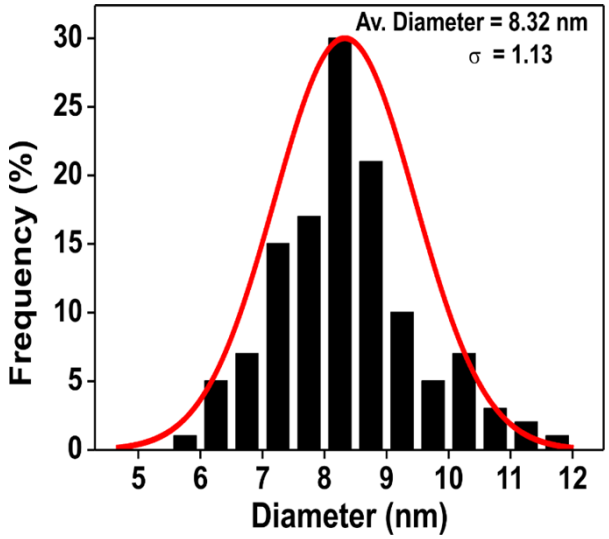


Figure S1. Size distribution histogram of DsAgNPs.

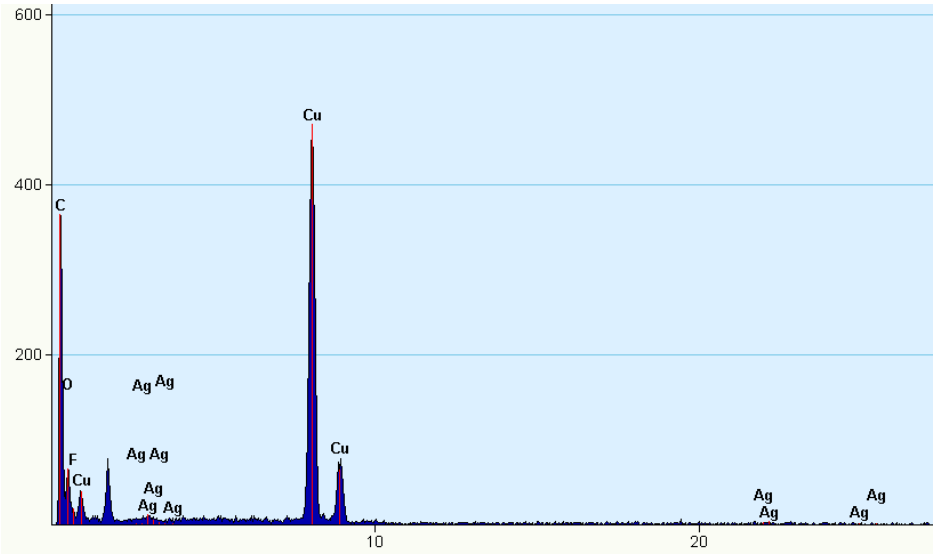


Figure S2. Energy dispersive spectra (EDS) of 5-FU@DsAgNCs. Existence of Ag and F (of 5-FU) elements indicate successful formation of 5-FU@DsAgNCs.

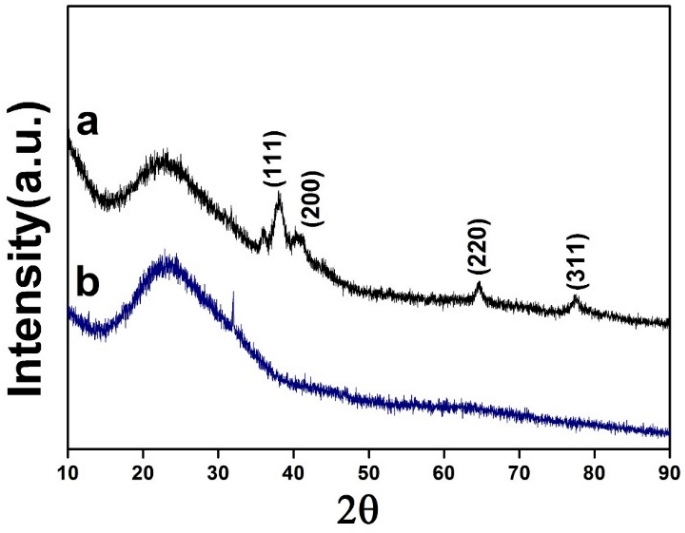


Figure S3. XRD spectra of a) 5-FU@DsAgNCs and b) blank G5 PAMAM.NH_2 dendrimer. Crystallographic phases (111), (200), (220) and (311) in (a) match closely with JCPDS card# 04-0783 corresponding to face centered cubic structure of Ag. Presence of no separate peaks related to 5-FU indicate its complete encapsulation.

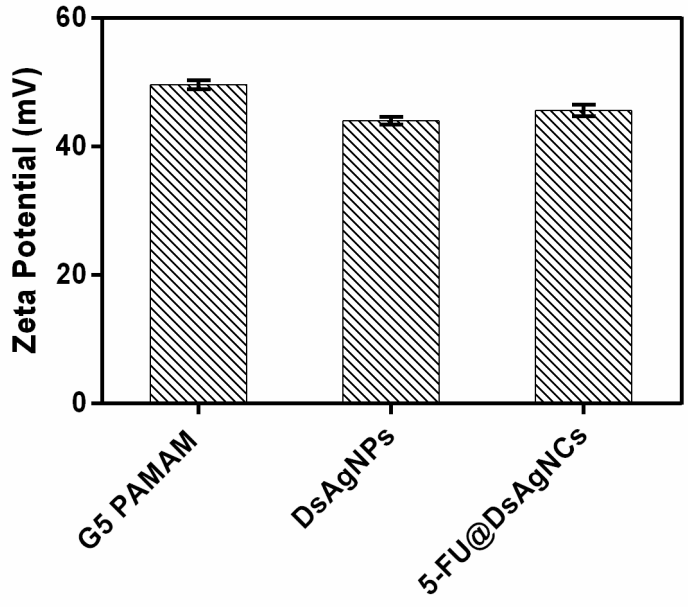


Figure S4. Zeta potential (ζ) measurements of G5 PAMAM, DsAgNPs and 5-FU@DsAgNCs.

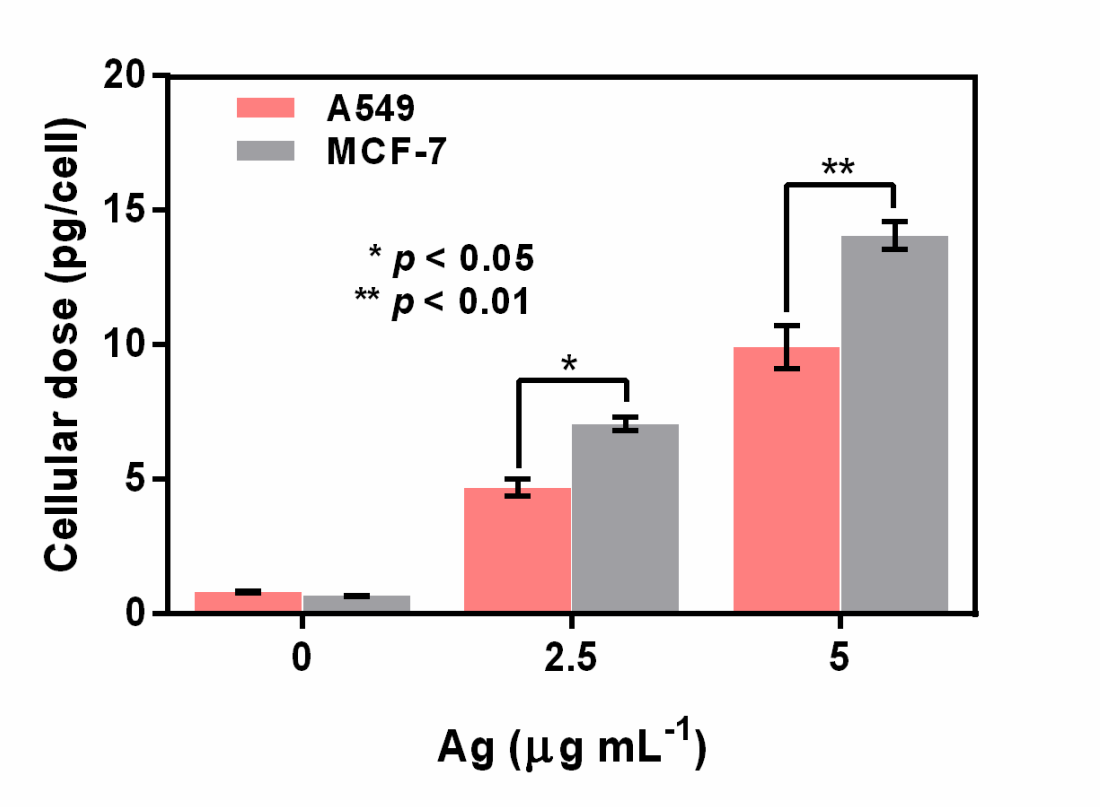


Figure S5. Cellular uptake of Ag in A549 and MCF-7 cells treated with different concentrations of 5-FU@DsAgNCs after 3 h. Data is represented as mean ± S.E.M (*n* =2).

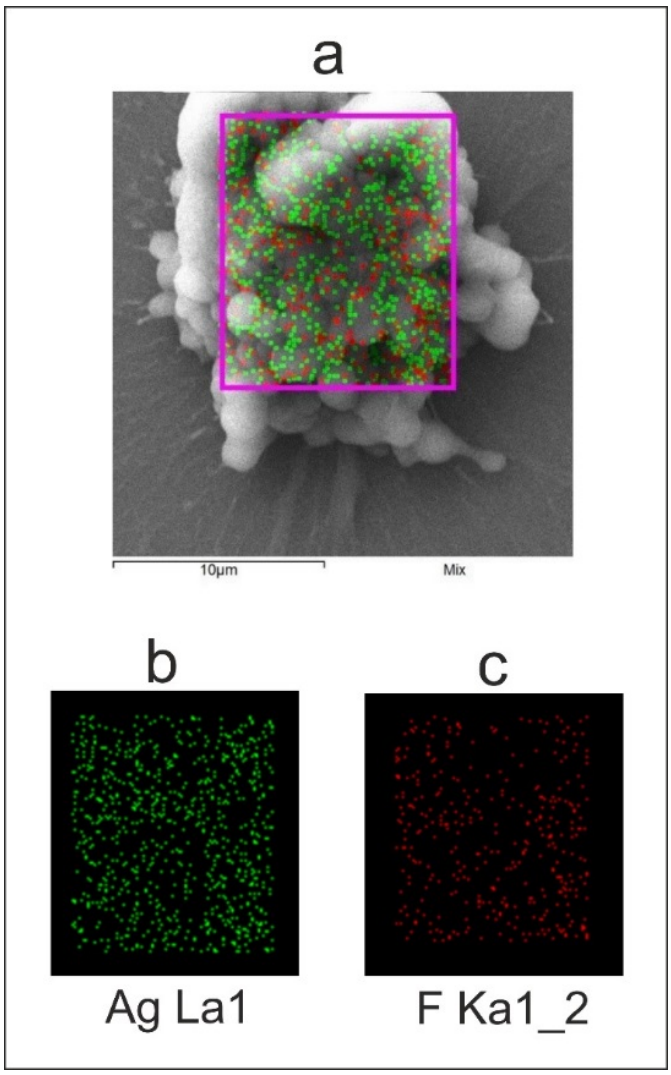


Figure S6. FE-SEM image of 5-FU@DsAgNC treated MCF-7 cell and color coded SEM/EDX dot maps. a) Overlay SEM image depicting elemental distributions in 5-FU@DsAgNC treated MCF-7 cell, b-c) Individual elemental distribution (green for silver and red for fluorine).

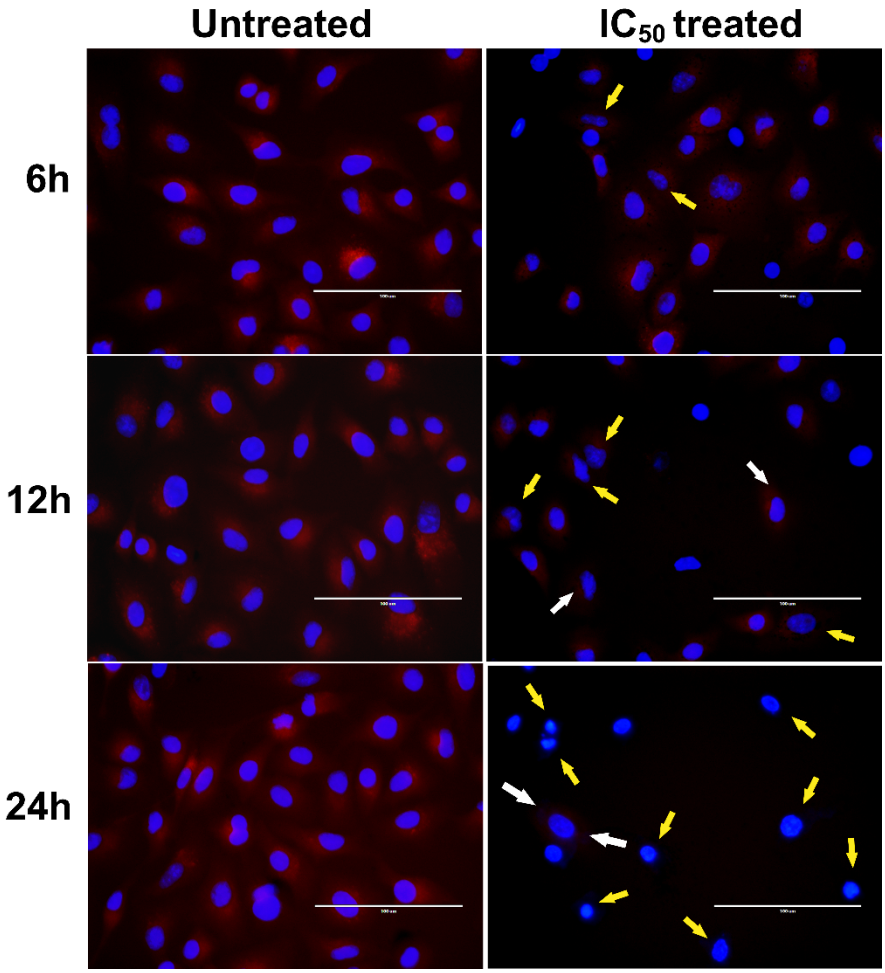


Figure S7. Time-dependent overlay images of untreated and 5-FU@DsAgNC (IC_{50}) treated A549 cells stained with Hoechst 33342 (blue) and rhodamine B (red). Yellow arrows indicate chromatin condensation (dark spots) and white arrows point towards cytoskeleton compaction. Scale bar: 100 µm.

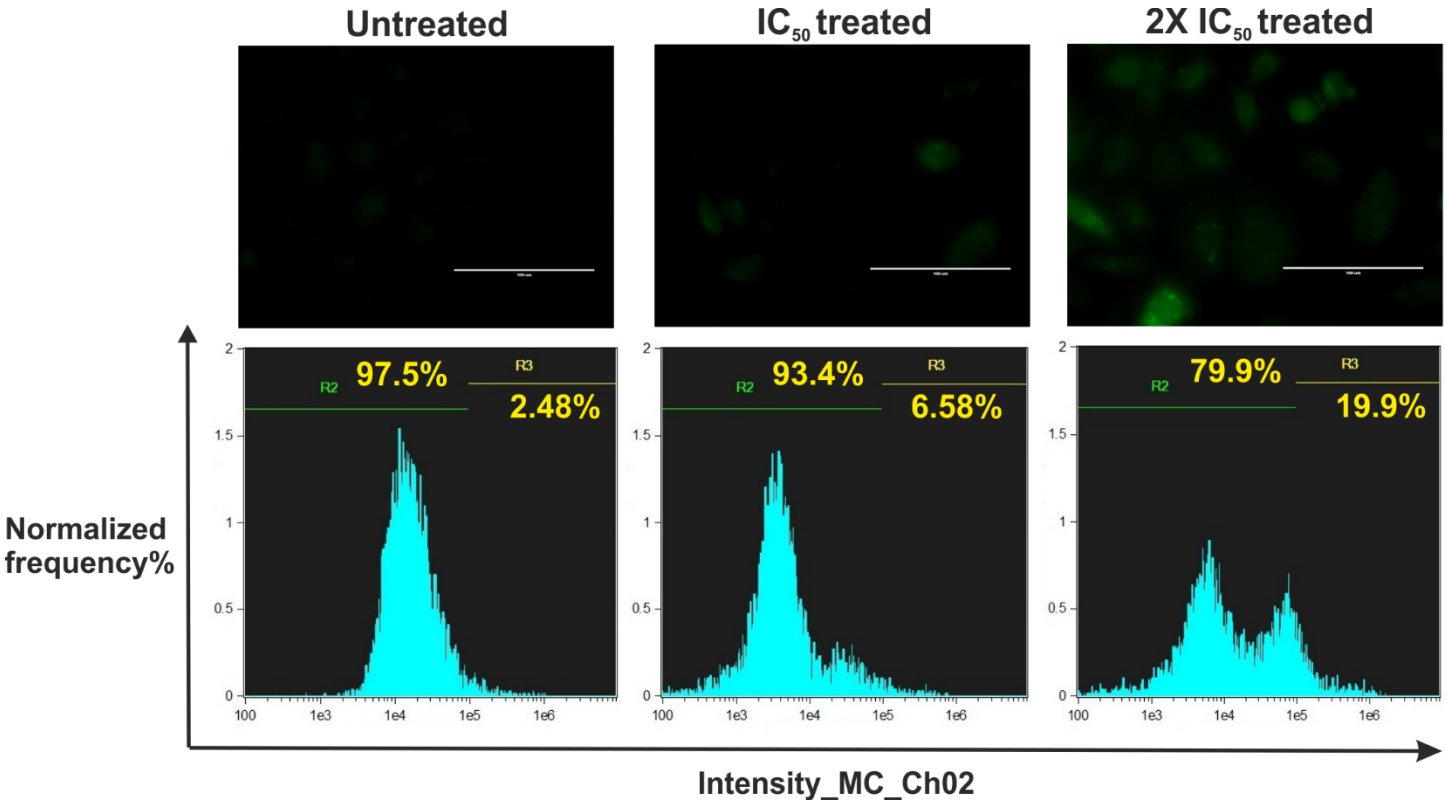


Figure S8. Microscopic and flow cytometric analysis of ROS production in 5-FU@DsAgNCs treated A549 cells. Upper panel: Green DCF fluorescence indicates intracellular ROS generation. Scale bar: 100 µm. Lower panel: Corresponding flow cytometric quantitation.