## **Electronic Supplementary Information**

of

## Evaluation of intracellular distribution of folate functionalized silica nanoparticles using fluorescence and hyperspectral enhanced dark field microscopy

Luminita Claudia Miclea<sup>a</sup>, Mona Mihailescu<sup>\*,b</sup>, Nicolae Tarba<sup>c</sup>, Ana-Maria Brezoiu<sup>d</sup>, Ana Maria Sandu<sup>e</sup>, Raul-Augustin Mitran<sup>f</sup>, Daniela Berger<sup>d</sup>, Cristian Matei<sup>d</sup>, Mihaela Georgeta Moisescu<sup>\*,a</sup>, Tudor Savopol<sup>a</sup>

<sup>d</sup>Department of Inorganic Chemistry, Physical-Chemistry & Electrochemistry, Faculty of Chemical Engineering and Biotechnologies, University "Politehnica" of Bucharest, 1-7 Polizu st., 11061, Bucharest, Romania

<sup>e</sup>CAMPUS Research Center, University "Politehnica" of Bucharest, 313 Splaiul Independentei, Bucharest, 060042, Romania <sup>f</sup>"Ilie Murgulescu" Institute of Physical-Chemistry, Romanian Academy, 202 Splaiul Indepedenței, Bucharest, 060021,

<sup>&</sup>lt;sup>a</sup>Biophysics and Cellular Biotechnology Department, Excellence Center for Research in Biophysics and Cellular Biotechnology, Faculty of Medicine, University of Medicine and Pharmacy Carol Davila, 8 Eroii Sanitari Blvd., Bucharest, 050474, Romania

<sup>&</sup>lt;sup>b</sup>Digital Holography Imaging and Processing Laboratory, Fundamental Sciences Applied in Engineering Research Center, Faculty of Applied Sciences, University "Politehnica" of Bucharest, 313 Splaiul Independentei, Bucharest, 060042, Romania<sup>c</sup>Physics Department, Faculty of Applied Sciences, Doctoral School of Automatic Control and Computers, University "Politehnica" of Bucharest, 313 Splaiul Independentei, Bucharest, 060042, Romania

Romania

<sup>\*</sup> Mihaela Georgeta Moisescu (<u>mihaela.moisescu@umfcd.ro</u>), and Mona Mihailescu (<u>mona.mihailescu@upb.ro</u>) are equally sharing the corresponding author position. Refer to Mihaela Georgeta Moisescu for biology-related items and Mona Mihailescu for microscopy and software items.



**Fig. S1** Example of fluorescence images of control Caco-2 cell (slices extracted from Z-stack), for nucleus (A) and cytoskeletal actin (B). Images were acquired using enhanced dark field microscopy (ob. 60× with immersion). Cells were double stained with DAPI for the nucleus (digitally colored in blue) and with Alexa Fluor<sup>™</sup> 488 Phalloidin for cytoskeletal actin (digitally colored in green). The image resulting from superposing the digitally colored images was obtained using ImageJ software (C).



*Fig. S2* Nitrogen adsorption-desorption isotherms of SBA-15 and SBA-NH-folate carriers, as well as the corresponding Irinotecan-loaded carrier (A) and the pore size values distribution curves for mesoporous silica-type supports (B).



Fig. S3 Wide angle X-ray diffraction patterns for Irinotecan-loaded samples and folate-functionalized supports.



Fig. S4 SEM images for mesoporous silica particles of SBA-15 (A) and SBA-NH-folate (B).



**Fig. S5** Hyperspectral images of nanoparticles suspended in buffered saline solution. Nanoparticles appear as bright dots on a dark background (objective 60× with immersion).