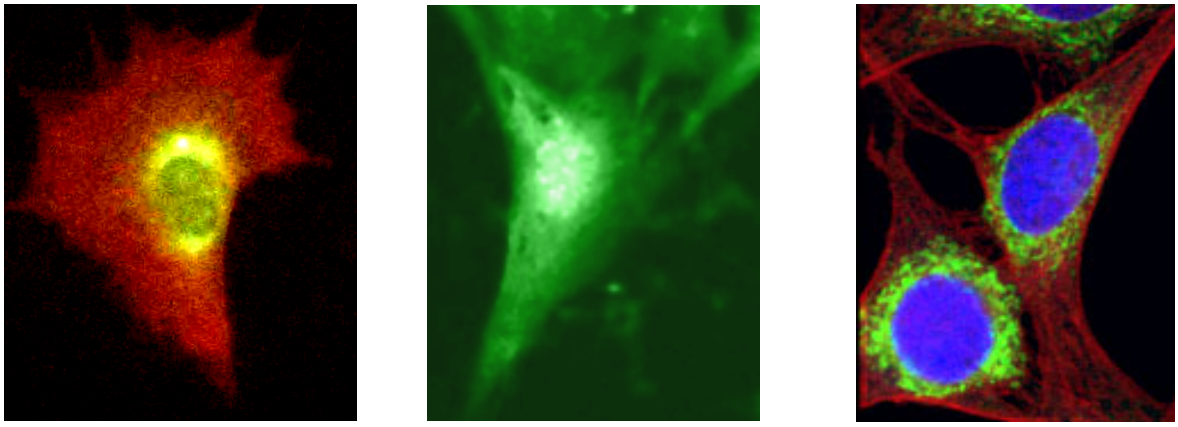
Electronic Supplementary Material (ESI) for Nanoscale

Supplementary Information

Nanostructured silicon nitride thin films for label-free multicolor luminescent cell imaging

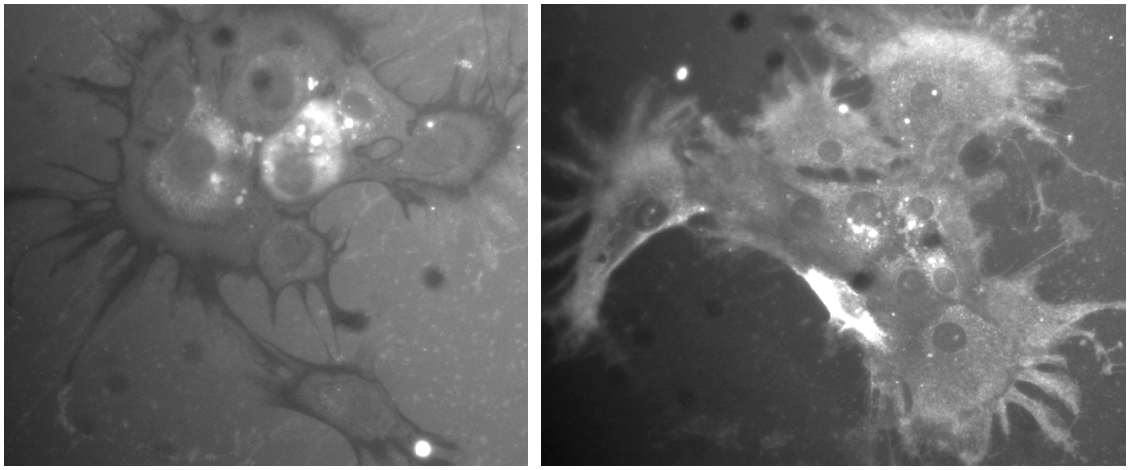
Figure S1 shows comparison of the label-free fluorescent image of a single 3T3 cell obtained by means of the SiNx/glass substrates reported in our manuscript (see Figure S1-a shown below) correlates very well with images of the 3T3 cells grown on traditional glass substrates and labelled with either SiC quantum dots (Figure S1-b) [1] or conventional dyes (Figure S1-c). Thus, one can conclude that the new imaging method reported in our manuscript is evident to allow visualisation of the cell compartments.



(a) **(b)** **(c)**

Figure S1. (a) Fluorescent image of 3T3 cells as shown in Figure 1-e) of manuscript; (b) uorescent image of 3T3 cells labelled with green emitting SiC quantum dots characterized by eferential nuclear localization [1]; (c) fluorescent labelling of 3T3 cells with conventional dyes he photo is taken from the following web site: http://www.cellsignal.com/products/5051.html).

However, strong cell adhesion on SiNx/glass substrates is necessary condition for contrast image appearance, it was ensured in our experiments by necessary time (about 5 days) taken by the cell culture to create a reliable interface with the SiNx surface. Indeed, as one can see in Figure S2, systematic appearance of a dark shade around the grown cells after only 3 days of the cell incubation indicates on the still bad adhesion of the cell periphery. In contrast, the cell images obtained after 5 days of the incubation appear to be completely bright which means that the cell adhesion is sufficiently good to visualize the cells. Therefore, in all our experiments, the cells were grown at least 5 days before acquisition of their images.



(a) **(b)**

Figure S2. Fluorescent images (in black-and-white) of the cells: (a) after 3 days cubation period; (b) after 5 days incubation period.

Figure S3 shows influence of the SiNx layer thickness (D) on the cell imaging quality.

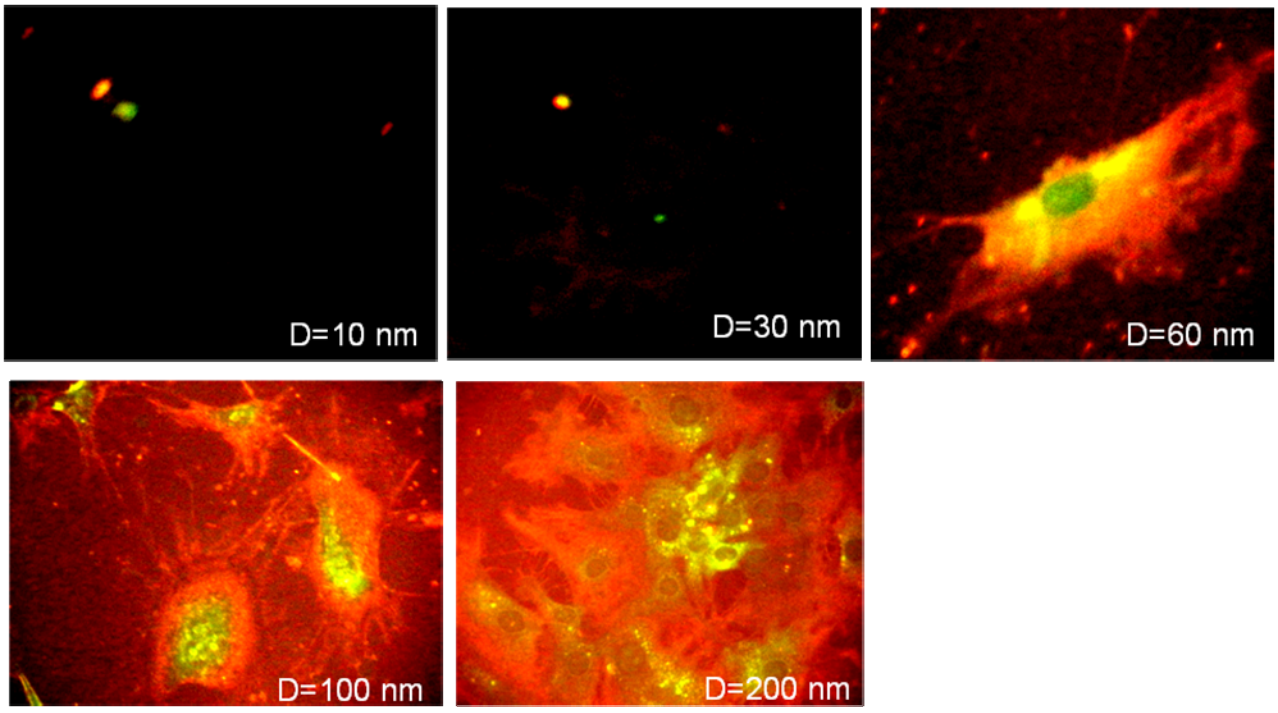


Figure S3. Dependence of the cell imaging on thickness (D) of the SiNx thin films.

One can see that starting from D=60 nm intensity of the cell fluorescent imaging is quite similar and stable. It means that the critical thickness of the SiNx layer is necessary to ensure a detectible fluorescent signal.

Supporting Information References:

[1] J.Botsoa, V.Lysenko, A.Géloën, O.Marty, J.M. Bluet, G. Guillot, Application of 3C-SiC quantum dots for living cell imaging. *Appl Phys Lett* **2008**, 92, 173902.