Supplementary Information

Targeted multifunctional tannic acid nanoparticles

J. R. Aguilera,^{a,†} V. Venegas,^{a,b,†} J. M. Oliva,^a M. J. Sayagués,^c M. de Miguel,^d J. A. Sánchez-Alcázar,^{a,e} M. Arévalo-Rodríguez^f and A. P. Zaderenko^{a,*}

^a Departamento de Sistemas Físicos, Químicos y Naturales. Universidad Pablo de Olavide, ES-41013-Seville, Spain. E-mail: apzadpar@upo.es; Tel: +34 954977365; Fax: + 34 954 977305.

^b Centro de Investigación Cooperativa en Biociencias, ES-48160-Bizkaia, Spain.

^c Instituto de Ciencia de Materiales de Sevilla, ES-41092-Seville, Spain.

^d Departamento de Citología e Histología Normal y Patológica. Universidad de Sevilla, ES-41009-Seville, Spain.

^e Centro Andaluz de Biología del Desarrollo, ES-41013-Seville, Spain.

^f Biomedal S.L, ES-41092-Seville, Spain.

* Author to whom correspondences should be addressed

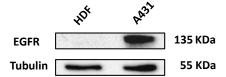


Figure S1. Expression of EGFR in HDF and A-431 cells determined by Western blot. For Western blot assay 80% confluence cells were washed two times with phosphate-buffered saline and scraped into mammalian protein extraction reagent. Cells were lysed 15 minutes on ice, and insoluble materials were removed by centrifugation. Total protein concentrations in cell lysates were determined by Bradford method. A 50-µg sample was separated by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) and electrotransferred to an Immobilon membrane (Amersham Pharmacia, Buckinghamshire, UK). After overnight blocking with block buffer at 4°C, membrane was incubated for 1 hour with anti-EGFR antibody (1:1000; Santa Cruz Biotechnology), rinsed 3 × 15 minutes by trisbuffered saline tween (TBST), and then incubated with horseradish peroxidase-conjugated secondary antibody (1:2500) for 1 hour and rinsed 6 × 5 minutes by TBST. Immunolabeled proteins were detected by using a chemioluminiscence method (Immun-Star HRP substrate kit, Bio-Rad, Hercules, CA, USA). Loading control was tubulin.

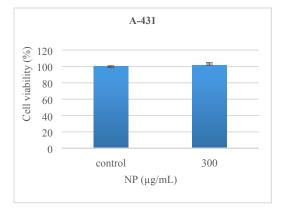


Figure S2. Cell viability of A-431 cell cultures (control) and A-431 cell cultures inoculated with 300 μg/ml of PIlabeled TA nanoparticles conjugated to anti-EGFR (TAN[PI]-Ab), after 4 hours treatment. Cell viability was evaluated by MTT assay, according to the kit specifications, and expressed as a relative percentage compared to untreated cells. All results are expressed as mean ± standard deviation.