

Cell uptake

Part Numbers:

CU-

Introduction

These nanoparticles uptake readily into cells.

Materials

Cell uptake polymer coated Nanopartz Gold Nanorods and Spherical Gold Nanoparticles (SGNPs).

J774a.1 murine macrophages were purchased from the American Type Culture Collection (ATCC® TIB-67™, Manassas, VA, USA) and seeded on plastic culture flasks in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum, 1% l-glutamine and 1% penicillin–streptomycin solution. Cells were kept and left to grow under standard culture conditions (37 °C, 5% CO₂).

Procedure

A. Protocol

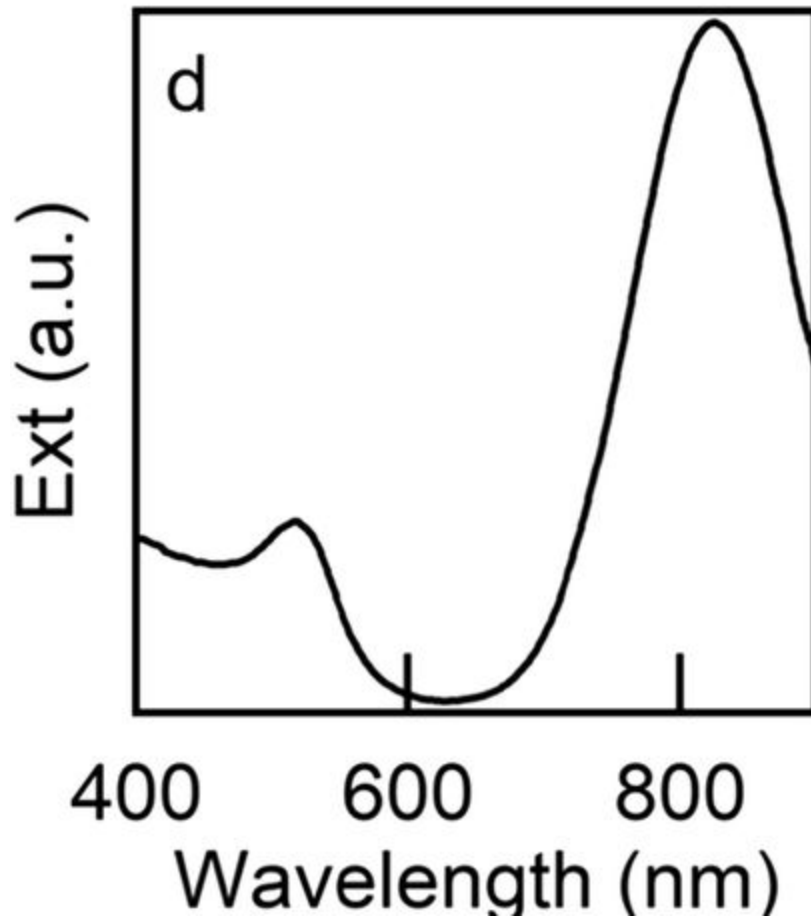
The accumulation of gold nanoparticles in macrophages was quantified by an optical analysis. 5×10^5 J774a.1 cells were plated in Petri dishes and exposed to 50 nM Au cell uptake coated gold nanoparticles from 1 to 48 h, in serum-free medium (SFM). At the end of these treatments, particles were accurately removed and macrophages were sequentially fixed with a solution of 3.6% PFA in PBS for 10 min at room temperature, washed with PBS, harvested, centrifuged for 5 min at 1000 rpm, re-suspended in 120 μ l PBS in a quartz micro-cuvette and directed to an optical inspection with a V-560 spectrophotometer from Jasco.

In order to assess the effect of exocytic release on the retention of the passenger particles subsequent to their internalization, macrophages were treated with 50nM Au gold nanoparticles for 24 h and then cultured in fresh SFM from 24 to 48 h, fixed and prepared for the optical analysis as is mentioned above.

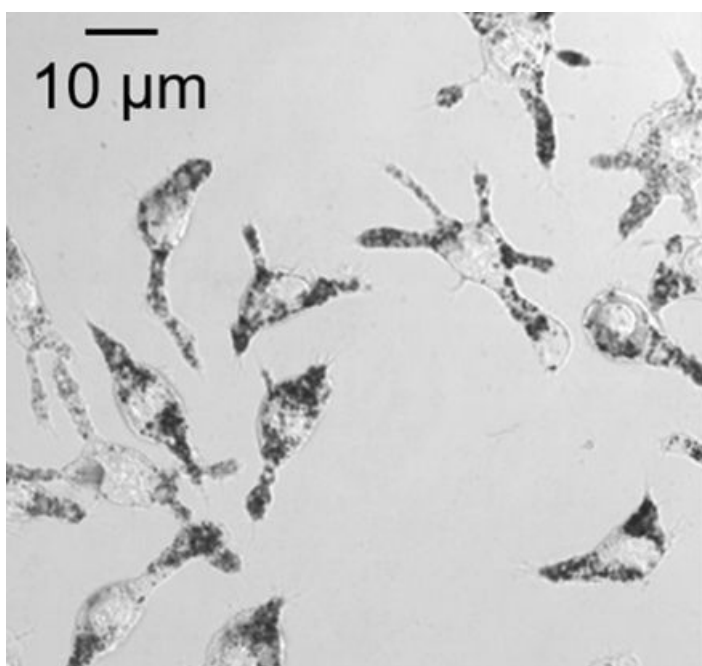
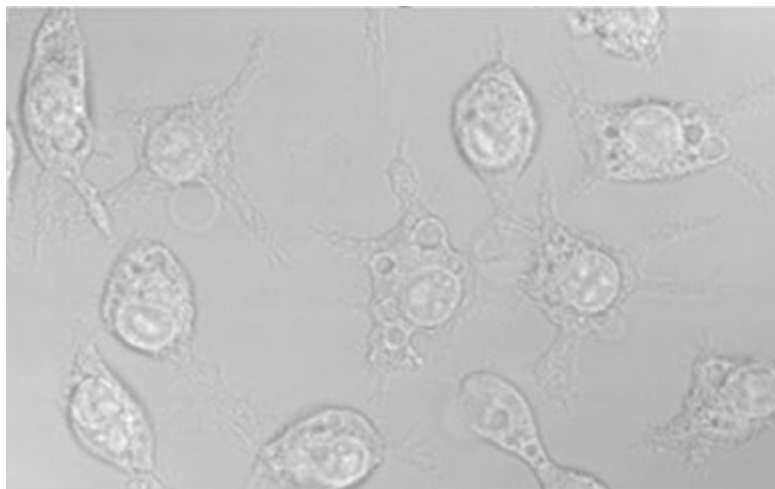
In order to quantify the amount of particles taken up per cell, the spectra of optical extinction were modeled as a linear combination of separate contributions from the macrophages and the gold nanoparticles. In particular, the former was recovered from an empirical measurement of a standard population of macrophages. The latter was devised as a numerical approximation of the plasmonic band from an ensemble of gold

nanoparticles, which was described as an integral over Gans lineshapes , by using the dielectric function of gold by Etchegoin et al. and was also subject to an empirical calibration. With these calibrations, the analysis of the experimental spectra returns a number density of cells in cm^{-3} and a density of gold nanoparticles in ppm or $\mu\text{g cm}^{-3}$, which are then combined to achieve a mass of gold per cell.

Typical Results



Typical UV VIS for 10x 808 cell uptake polymer nanorods. Refer to COA that comes with product for exact UV VIS.



Representative images of untreated controls and macrophages exposed to 50nM Au cell uptake polymer-coated gold nanoparticles for 24 h, from a confocal microscope (TCS SP8, Leica Microsystems, Heidelberg, Germany) operated in transmission mode at 63× magnification