

Electronic Supplementary Information for

Ultra-small Platinum Nanoparticles on Gold Nanorods Induced Intracellular ROS Fluctuation to Drive Megakaryocytic Differentiation of Leukemia Cells

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Experimental information

Synthesis of Au@Pt

Au@Pt were synthesized based on AuNRs, typically, AuNRs solution (1 mL) was mixed with K₂PtCl₄ aqueous solution (75 μ L, 2 mM). Then, ascorbic acid (15 μ L, 0.1 M) was added and the total solution volume was diluted to 2 mL. The mixture was incubated in a 30 °C water bath for 1 h and centrifuged once (12000 rpm, 10 min) after adding 1 mL cetyltrimethylammonium bromide (0.1 M). The precipitate of AuNRs and Au@Pt were dispersed in 0.5 mL PSS aqueous solution (2 mg mL⁻¹ containing 6 mM NaCl), and the solution was incubated overnight. Finally, they were centrifuged once (12000 rpm, 10 min), and re-dispersed in sterile water.

Cell Culture

Human chronic myelocytic leukaemia cell line (K562) was purchased from the Cell Resource Center of Chinese Academy of Medical Sciences & Peking Union Medical College (Beijing, China). The cells were cultured in RPMI 1640 medium (HyClone), and supplemented with 10% fetal bovine serum (FBS, Gibco), streptomycin (100 μ g mL⁻¹), and penicillin (100 U mL⁻¹). All cells were incubated at 37 °C with 5% CO₂.

Isolation of bone marrow mononuclear cells (Clinical sample collection)

This study was performed in accordance with the Declaration of Helsinki and was approved by the Peking Union Medical College Hospital ethics committee, and written informed consent was obtained from all subjects. The samples were first diluted with PBS, and then added to an equal-volume human lymphocyte separation solution (Dakewe Biotech Co. Ltd.) for centrifugation. Bone marrow mononuclear cells (BM-MNCs) in the middle layer were collected and resuspended in the culture medium. Cell were grown at a density of 1 \times 10⁵ cells well⁻¹ on 24-well cell culture plate, and exposed to various concentrations of Au@Pt nanoparticles for 72 h at 37 °C for cell differentiation detection.

Western blot

Proteins (30 μ g lane⁻¹) were resolved by means of SDS-PAGE and electro transferred onto polyvinylidene fluoride (PVDF) membranes (Millipore, Bedford,

Massachusetts). The membranes were blocked using TBST containing 5% skim milk (and, if necessary, the membranes were stripped), and then incubated overnight at 4 °C with primary antibodies (Cell Signaling Technology, all diluted 1:1000 in blocking buffer). The membranes were next washed in TBST and then incubated for 2 h at room temperature with horseradish peroxidase conjugated secondary IgGs (1:10000).

Quantitate the concentration of Au@Pt according to extinction spectroscopy

The nanoparticle concentration of Au@Pt is estimated based on the concentration of AuNRs. Generally, Inductively Coupled Plasma-Mass Spectrometry (ICP-MS) is employed to determine the total concentration of Au atoms in a given AuNRs suspension. According to the molar volume of a gold atom ($10.2 \text{ cm}^3/\text{mol}$), we obtain the total volume (V) of all AuNRs in the suspension. From the TEM images, the diameters and lengths of AuNRs can be measured and then calculate the mean volume of a single AuNRs (V_0) by assuming a cylindrical shape with two half-sphere end-caps. The AuNRs molar concentration in the suspension can be estimated as V/AV_0 , where A is Avogadro's constant. The spectrum of Au@Pt at the same particle concentration can be obtained during the process of syntheses. Therefore, the particle concentration of Au@Pt can be estimated according to the Lambert-Beer law that the concentration is proportional to the extinction value of spectrum.