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Cancer Cell Killing Effect of Silver Colloidal Nanoparticles and $Ag@TiO_2$ Colloidal Nanoclusters on HeLa Cells

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[Abstract] Inorganic metal nanoparticles such as Ag have the size-encouraged electronic and optical properties, which are widely used in biosensing, catalysis, biological labeling and data storage. The photoinduced Ag colloidal nanoparticles convert their sizes under the visible light and provide the photothermal heat, which kills the cancer cells. A distinct cell killing effect was observed by this procedure. Very little photothermal cell destruction was observed in the absence of the silver colloidal nanoparticles.

The photocatalytic cancer cell killing activities of the $Ag@TiO_2$ core-shell composite colloidal nanoclusters have been investigated on malignant cell (HeLa) killing under the UV-visible light irradiation. The photocatalytic cell killing activities of the $Ag@TiO_2$ core-shell composite nanoclusters have been found to vary with the Ag and TiO_2 molar ratio concentrations. The direct involvement of the metal particles in mediating electron transfer from the photoexcited TiO_2 under the band gap excitation is considered to carry out the photocatalytic reaction on cells. The charge separation and interfacial charge-transfer promote the photocatalytic cancer cell killing (Fig.1) more than the TiO_2 semiconductor alone. It was found that the $Ag@TiO_2$ nanocluster under the low concentrations kill more malignant (HeLa) cells by 80% than TiO_2 nanoparticles alone under the same concentrations.

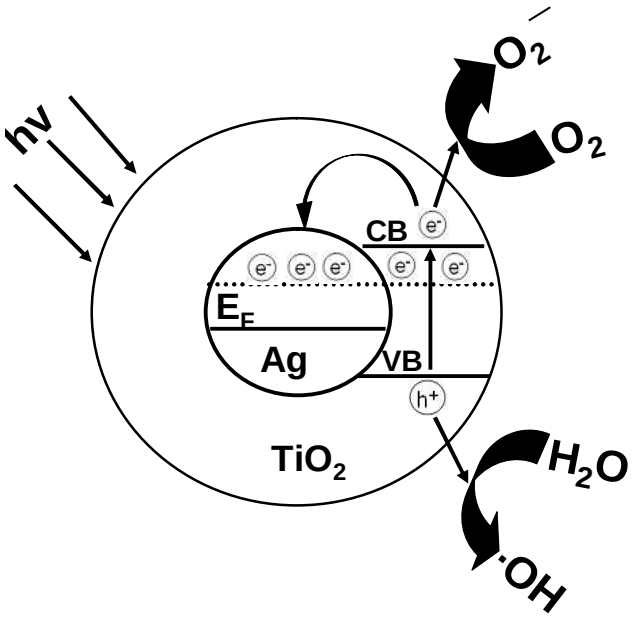

Fig. 1 Proposed mechanism for the interfacial charge transfer of $Ag@TiO_2$

[Experimental] The Ag colloidal nanoparticles were prepared by the laser ablation of metal targets in liquids (water) environment (AgL) and also citrate reduction from $AgNO_3$ salt (AgC). UV-visible absorption spectrometry, transmission electron microscopy (TEM), scanning electron microscopy (SEM), X-ray photoelectron spectroscopy (XPS) and atomic force microscopy (AFM) were used for the characterization of nanoparticles. Cells were cultured in 60 mm Petri dishes and the effect was observed by treatment of Ag colloidal nanoparticles under 400-600 nm visible light irradiation (10 min).

The $Ag@TiO_2$ core-shell nanocluster was prepared by citrate reduction from $AgNO_3$ salt and titanium (IV) (triethanolaminato)-isopropoxide ($N((CH_2)_2O)_3TiOCH(CH_3)_2$) (TTEAIP) (in 80 wt % 2-propanol solution) in the millipore-distilled water with vigorous stirring at 60 °C constant

temperature. Five different suspensions of $Ag@TiO_2$ nanoclusters were prepared by keeping the $AgNO_3$ concentration constant at 1mM while the TTEAIP concentrations are varied at 1,3,5,7 and 9 mM. The [Ag] : [TiO_2] ratios of these five $Ag@TiO_2$ suspensions were 1:1, 1:3, 1:5, 1:7 and 1:9. All the concentrations are based on molar concentrations. Cells were cultured in 60 mm Petri dishes and the effect was observed by treatment of $Ag@TiO_2$ with 350-600 nm UV-visible light irradiation (5 min).

[Results and Discussion]

UV-visible absorption spectra of Ag and $Ag@TiO_2$ core-shell nanocluster suspensions show the strong absorption in the visible region with plasmonic peaks [Fig. 2 (A) and (B)]. The plasmonic peaks in [Fig. 2 (B)] indicate that the Ag core is not oxidized, because the absence of plasmonic peak in absorption spectra is associated with oxidation of Ag nanoparticles [1].

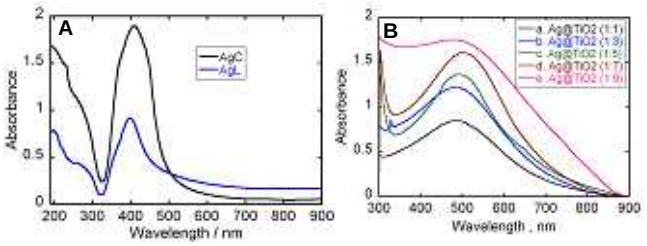

Fig. 2. Absorption spectra of (A) Ag (B)$Ag@TiO_2$ core-shell nanoclusters

The TEM image shows that the size and shape of Ag and $Ag@TiO_2$ core-shell nanoparticles are mostly spherical and symmetrical [Fig. 3]. The round dark structure of TiO_2 shell around the Ag core clearly shows that the core is uniformly covered by the shell [Fig. 3 (D)].

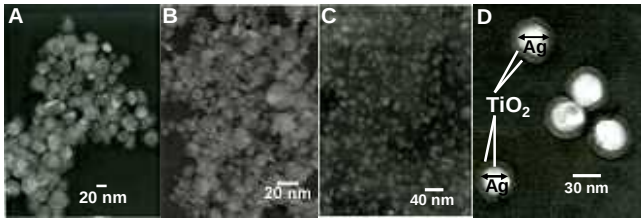

Fig. 3. TEM image of (A) Ag C (B) AgL (C) $Ag@TiO_2$ (D) $Ag@TiO_2$

To test the cell viability, the cell dishes treated with the different amounts of Ag-colloidal nanoparticles (0.1 ml, 0.5 ml, 1 ml, 2 ml and 3 ml) and one dish not treated (controlled dish) were incubated. The cell dishes treated by nanoparticles solution were irradiated by visible light for 10 min. Maximum 98% cells were killed rapidly [Fig.4 (A)]. This indicates that Ag nanoparticles are cytotoxic and are able to kill the cancer cells under visible light irradiation.

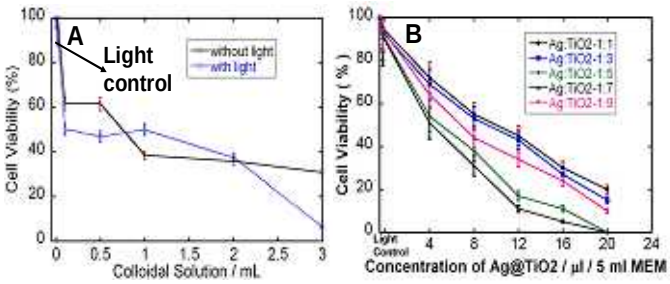

Fig. 4. Surviving viability of HeLa cells (A) Ag treated nanoparticles (B) $Ag@TiO_2$ treated core-shell nanoclusters

For the $Ag@TiO_2$, the cell dishes were incubated with different amounts of colloidal nanocluster solutions (4 µl, 8 µl, 12 µl, 16µl and 20 µl) and another dish without nanocluster solution (controlled dish) were incubated [Fig.4 (B)]. Under 5 min light irradiation, 100% cell was killed in the presence of 20 µl $Ag@TiO_2$ nanoclusters [Fig. 4 (B)].

Reference

[**1**] Awazu K., Fujimaki M., Rockstuhl C., Tominaga J.,Murakami H.,Ohki Y., Yoshida N. and Watanabe T., J. Am. Chem. Soc., **130**, 1676-1680 (2008).