Supporting Information

Rapid and sensitive colorimetric visualization of phthalates using UTP-modified gold nanoparticles cross-linked by copper (Ⅱ)

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Experimental Section

Reagents and materials. Hydrogen tetrachloroaurate (III) dehydrate ($HAuCl_4$) was purchased from Sinopharm Chemical Reagent Company (Shanghai, China). KCl, $BaCl_2$, NaCl, $MnCl_2$, $ZnCl_2$, $FeCl_3$, $MgCl_2$, $CaCl_2$ and $CuCl_2$ were purchased from Lingfeng Fine Chemical Co., Ltd. (Shanghai, China). Sodium oxalate, sodium citrate, ethyl benzoate and phthalate acid were purchased from Sinopharm Chemical Reagent Company (Shanghai, China). The four kinds of mononucleotide, i.e., 2'-deoxyadenosine triphosphate (dATP), 2'-deoxyguanosine triphosphate (dGTP), 2'-deoxycytidine triphosphate (dCTP), and 2'-deoxythymidine triphosphate (dTTP) were purchased from TaKaRa Biotechnology (Dalian) Co., Ltd. Adenosine 5'-triphosphate (ATP), guanosine 5'-triphosphate (GTP), cytidine 5'-triphosphate (CTP) and uridine 5'-triphosphate (UTP) were purchased from Sigma-Aldrich (St. Louis, MO). Dimethyl phthalate (DMP), di (2-ethylhexyl) phthalate (DEHP) and di (n-octyl) phthalate (DNOP) were ordered form Alfa Aesar China (Tianjin) Co., Ltd. All chemicals used in this work were of analytical reagent and obtained from commercial sources and directly used without additional purification. Some drinks were used in the real sample assay including green tea (Unity, Shanghai Bo Honey Food Co., Ltd.), carbonated drink (7UP, Shanghai Pepsi-Cola Beverage Co., Ltd.), juice drink (Hui Le Duo, Zhejiang Hui Mei Food and Beverage Co., Ltd.), and vegetable protein drink (Nong Fu Guo Yuan, ZheJiang Nong Fu Shan Quan Qian Dao Hu Co., Ltd.). 10×$NaNO_3$-MOPS buffer (500 mM $NaNO_3$ and 200 mM 3-(4-morpholinyl)-1-propanesulfonic acid, pH 7.0) was prepared using metal-free reagents in distilled water purified by a Milli-Q water purification system (Millipore Corp., Bedford, MA) with an electrical resistance of 18.2 MΩ. The experiments were conducted at room temperature.

Instrumentation. The ultraviolet visible (UV-vis) absorption spectrum was recorded with a microplate reader (BioTek Instruments, Winooski, VT, USA) with wavelength ranging from 400

nm to 900 nm using a transparent 384-well microplate (Greiner, Germany). Transmission electron microscope (TEM) measurements were performed on Jeol JEM-1230 instrument operated at an accelerating voltage of 80 kV. Samples for TEM studies were prepared by placing a drop of AuNPs solution (or AuNPs and copper and/or phthalate mixture) on a copper grid. The films on the TEM grids were allowed to dry for 2 min following that the extra solution was removed using a blotting paper. Photographs were taken with Nikon D3100 digital camera (Tokyo, Japan).

Preparation of Citrate-Stabilized Au Nanoparticles (AuNPs). All glassware was thoroughly cleaned overnight with freshly prepared 3:1 HCl/HNO_3 (*aqua regia*) and rinsed thoroughly with Mill-Q water prior to use. AuNPs were prepared according to the reported method.[1] Briefly, 100 mL of $HAuCl_4$ (0.01%) was added to a 250 mL round bottle and then boiled. Under rapid stirring, 3.5 mL of trisodium citrate (1%) was added and further rapidly stirred for 15 min. After stirred for 30 min, the solution was then gradually cooled to room temperature, and was filtered by 0.22 μm filter paper, which was stored in the refrigerator (4℃) before further use.

Surface Modification of Au Nanoparticles with Mononucleotides. For the surface modification of AuNPs with mononucleotides, an aliquot of 30 μL (2.0 mM) UTP, ATP, GTP, CTP, dATPs, dTTPs, dGTPs or dCTPs was added to 970 μL of AuNPs colloidal solutions. After incubating for 3 h at 4 ℃, the resulting mixture was subject to centrifugation at the speed of 12000 rpm for 30 min. The supernatant fluid was removed, while the AuNPs precipitate was dissolved with 200 μL Mili-Q H_2O. These mononucleotides-modified AuNPs solutions were stored in the refrigerator (4℃) for further use.

Colorimetric Assay. An aliquot of 50 μL mixed solution containing 10 μL 4 μM Cu^{2+}, 10 μL various concentration of phthalate dissolved in ethanol, 10 μL 10×$NaNO_3$-MOPS buffer (500 mM $NaNO_3$ and 200 mM 3-(4-morpholinyl)-1-propanesulfonic acid, pH 7.0) and 20 μL Mili-Q H_2O was placed in the wells of a transparent 384-well microtiter plate. Then, 50 μL mononucleotides-modified AuNPs was added to the corresponding wells. Subsequently, the resulting solutions were incubated for 5 min before measuring their extinction spectra. The experiments of optimization of sensing conditions were carried out under identical conditions. All experiments were repeated three times.

Real Sample Assay. For the assay of real sample including tea drinks, carbonated drinks, juice drinks and vegetable protein drinks, some pretreatment should be conducted. Due to the extremely

low pH of certain juice drinks, the pH value of samples under test should be measured and adjusted to nearly neutral (ca. 7) using 10 M NaOH. The sarcocarp in certain drinks was centrifuged to precipitate (4000 rpm, 5 min). Then the supernatant was collected for phthalate-tainted and further colorimetric assay using the method mentioned above.

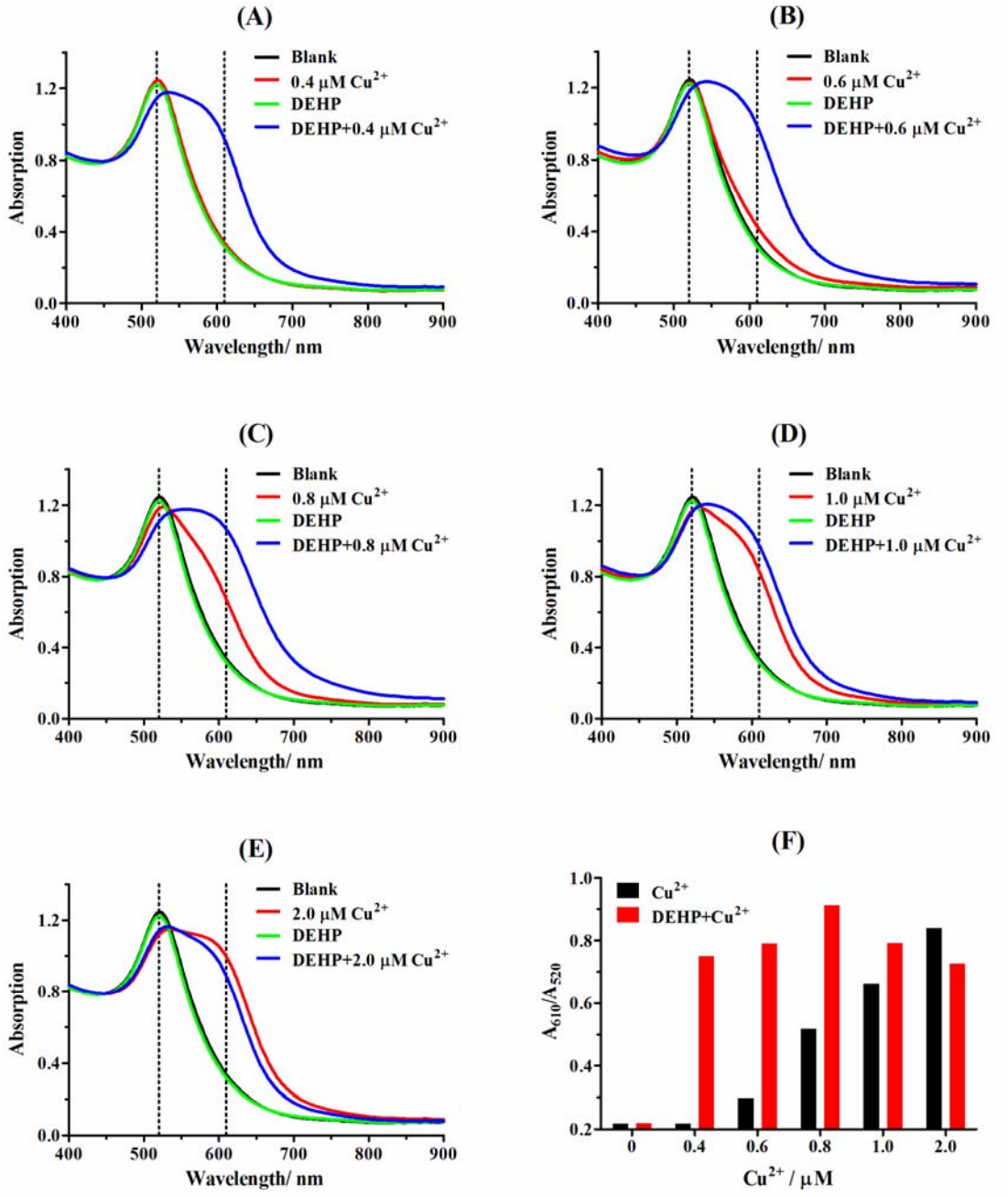

Figure S1. UV-vis absorbance of U-AuNPs in solution upon addition of 10 g/L DEHP in the presence of (A) 0.4 μM, (B) 0.6 μM, (C) 0.8 μM, (D) 1 μM, and (E) 2 μM Cu^{2+} acting as a cross-linker. (F) Bars represent the absorption ration A_{610}/A_{520} of U-AuNPs in solution upon addition of 10 g/L DEHP in the presence of various concentration of Cu^{2+} mentioned above.

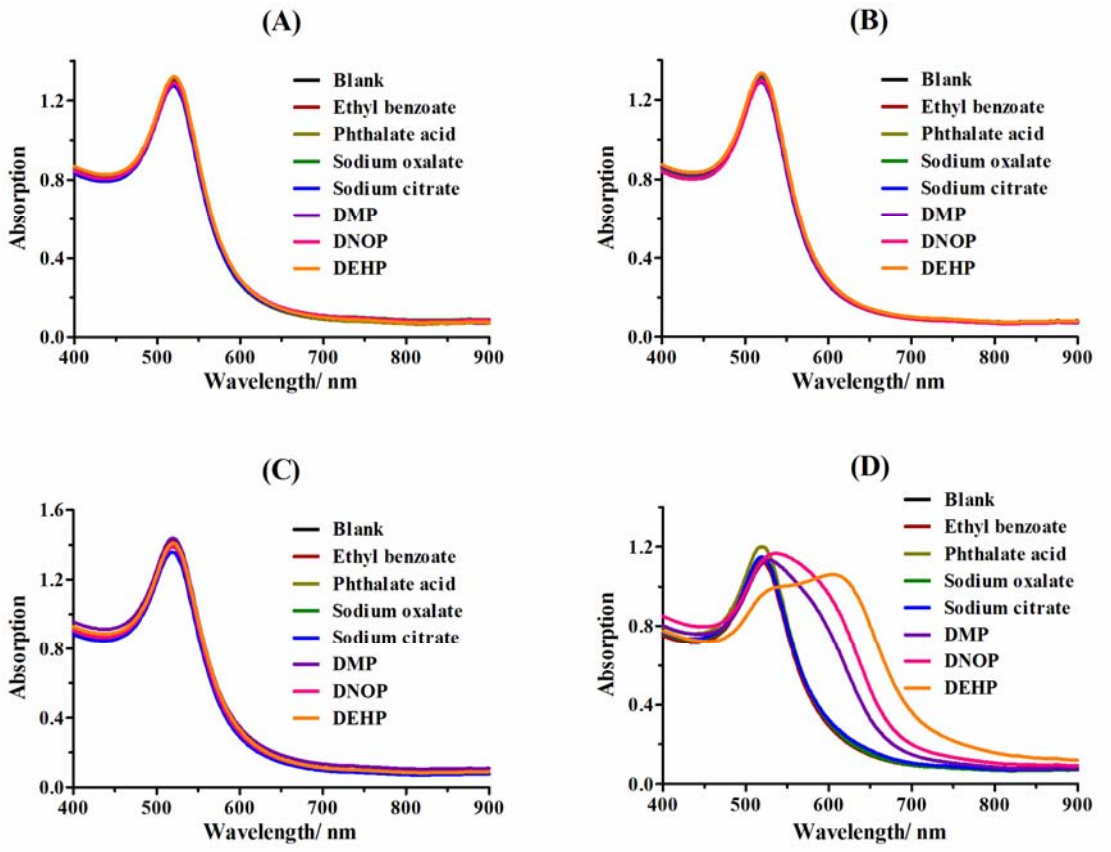

Figure S2. UV-vis absorbance of (A) ATP-modified AuNPs, (B) CTP-modified AuNPs, (C) GTP-modified AuNPs, and (D) UTP-modified AuNPs in solution upon addition of various phthalates including 10 g/L of DMP, DNOP and DEHP and certain competing stimulus including 10 g/L ethyl benzoate, phthalate acid, sodium oxalate and sodium citrate.

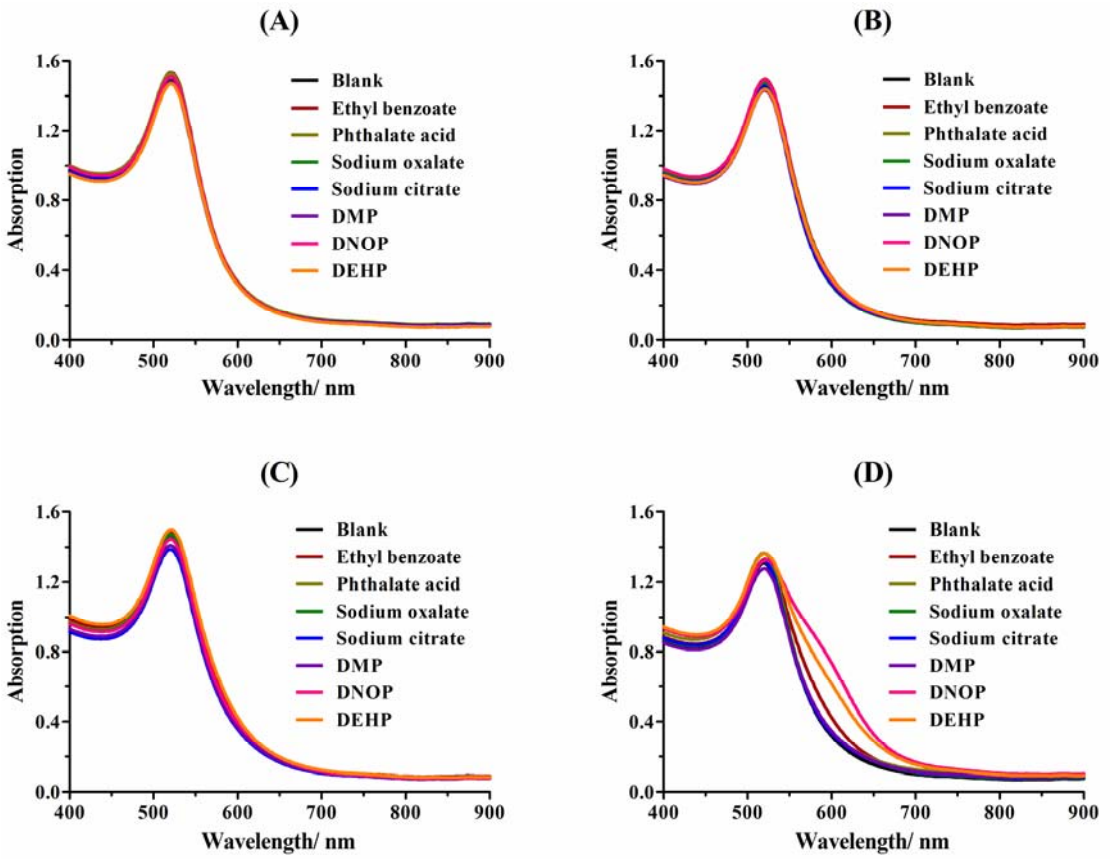

Figure S3. UV-vis absorbance of (A) dATP-modified AuNPs, (B) dCTP-modified AuNPs, (C) dGTP-modified AuNPs, and (D) dTTP-modified AuNPs in solution upon addition of various phthalates including 10 g/L of DMP, DNOP and DEHP and certain competing stimulus including 10 g/L ethyl benzoate, phthalate acid, sodium oxalate and sodium citrate.

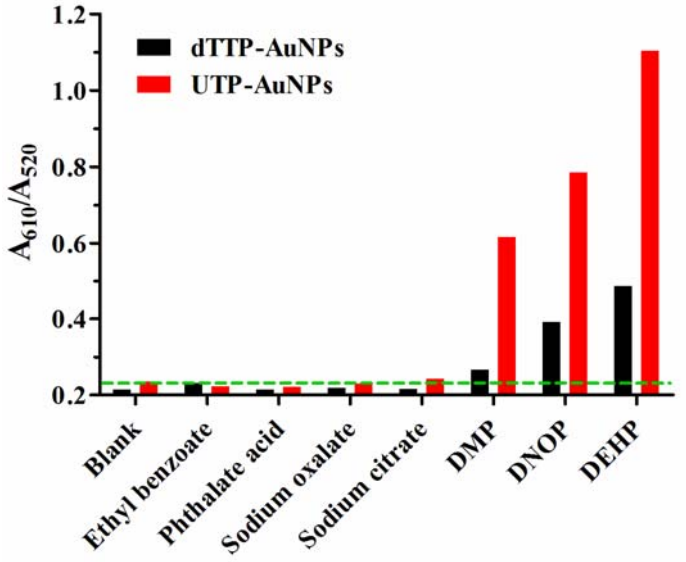

Figure S4. Bars represent the absorption ratio A_{610}/A_{520} of dTTP-modified AuNPs and UTP-modified AuNPs in solution upon addition of various phthalates including 10 g/L of DMP, DNOP and DEHP and certain competing stimulus including 10 g/L ethyl benzoate, phthalate acid, sodium oxalate and sodium citrate.

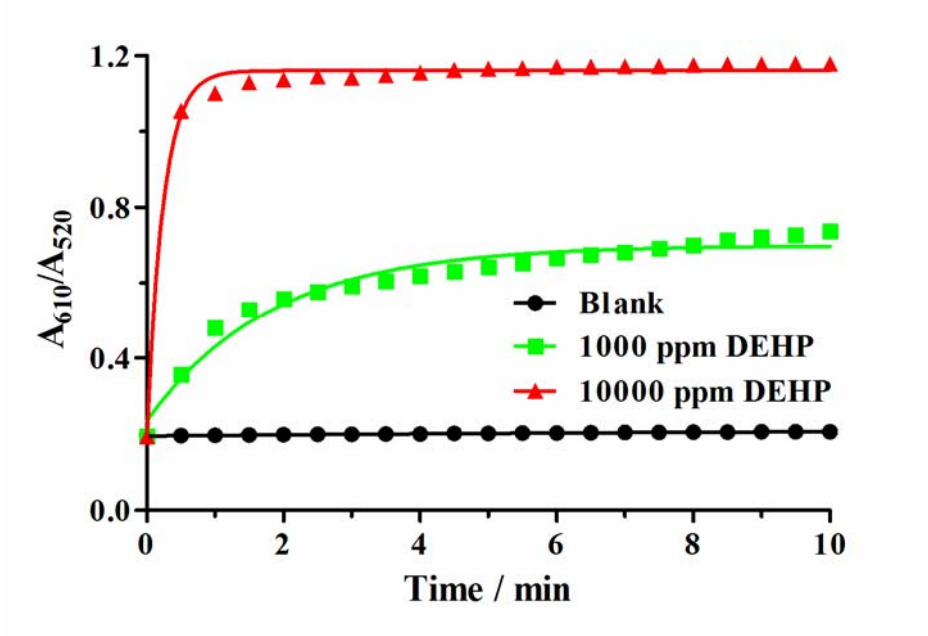

Figure S5. Time-coure behavior of the absorption ratio A_{610}/A_{520} of UTP-modified AuNPs responding to DEHP in the presence of Cu^{2+}.

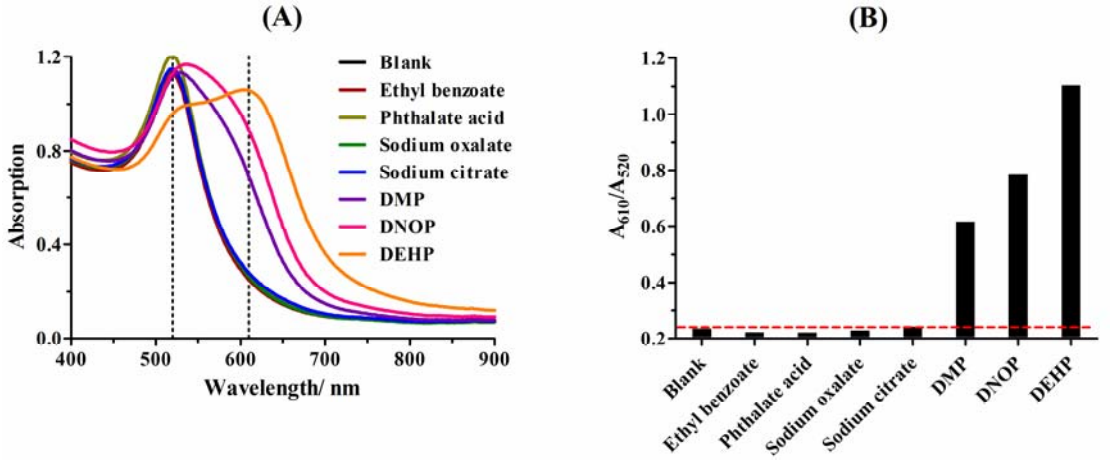

Figure S6. (A) UV-vis absorbance of U-AuNPs in solution upon addition of various phthalates including 10 g/L of DMP, DNOP and DEHP and certain competing stimulus including 10 g/L ethyl benzoate, phthalate acid, sodium oxalate and sodium citrate. (B) Plot of absorption ratios A_{610}/A_{520} corresponding to analytes mentioned above.

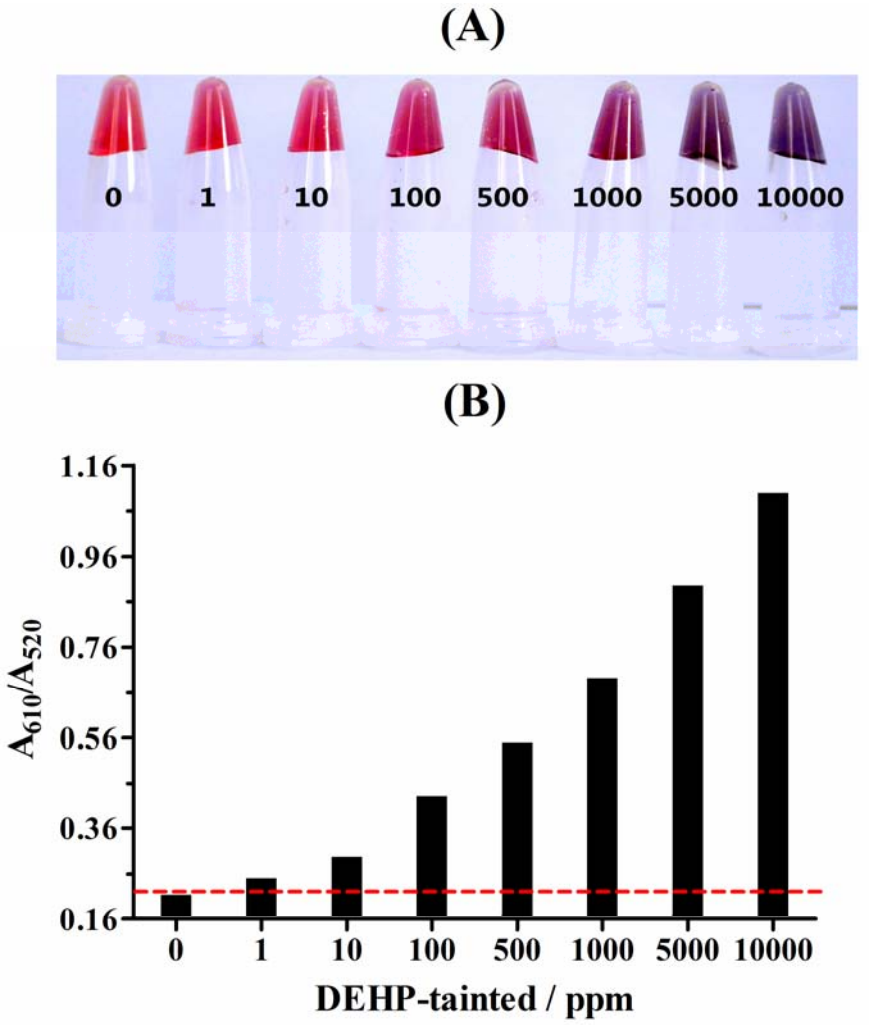

Figure S7. (A) Visual color change of the U-AuNPs sensor system without any addition and with the addition of raw samples tainted with 1, 10, 100, 500, 1000, 5000 and 10000 ppm DEHP. (B) The corresponding absorption ratio A_{610}/A_{520}.

Reference:

1. K. C. Grabar, R. G. Freeman, M. B. Hommer, M. J. Natan, *Anal. Chem*. 1995, **67**, 735.