Supplementary material

1 Experimental section

1.1 Instruments

AuNP samples were dropped on carbon-coated copper grids and dried at room temperature. The particle size distribution was determined using ImageJ software. Transmission electron microscopy (TEM) analysis was performed using a JEM-1230 instrument (JEOL, Tokyo, Japan). Fourier transform infrared (FTIR) spectra were obtained using a Spectrum 100 FTIR spectrometer (PerkinElmer, Shelton, USA). The zeta potentials of the particles were determined using a Zetasizer Nano ZS (Malvern Instruments, Worcestershire, UK).

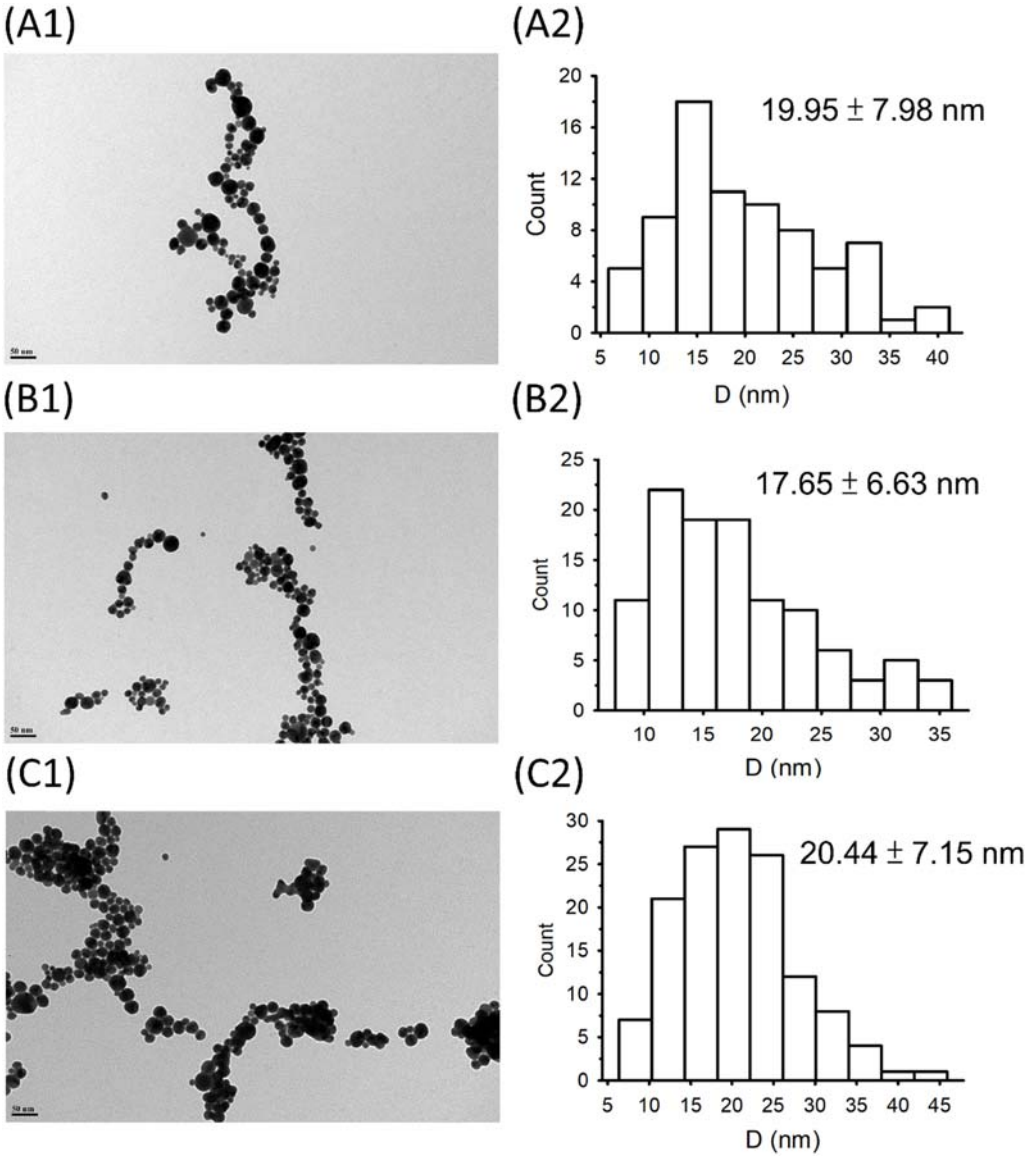

Figure S1. : TEM images and size distribution histograms of AuNPs prepared under different condition: - aptamer / - serotonin (A1 and A2), + aptamer / - serotonin (B1 and B2), and + aptamer / + serotonin (C1 and C2). The TEM images were analyzed using the ImageJ software. The average diameter and standard deviation of (A2) 76, (B2) 105 and (C2) 133 analyzed gold particles were determined to be 19.95 ± 7.98, 17.65 ± 6.63, and 20.44 ± 7.15 nm, respectively.

Table S1: Working ranges and optimal values of parameters for the colorimetric detection of serotonin

Parameter	Working range	Optimal value
Concentration of aptamer (nM)	15–30	20
Concentration of gold ion (μM)	167–667	333
Reaction temperature (°C)	25–44	30
Concentration of ascorbic acid (mM)	1–7.5	2.5
Concentration of NaCl (mM)	24–36	28
Time after adding with NaCl (min)	0–15	5

1.2 Specificity and real samples tests

For the specificity test, three interfering analogs (epinephrine, dopamine, and acetylcholine) were used at a concentration of 10 ng/mL and exposed to the same conditions as those described for serotonin analysis. To evaluate the application of this method in human samples, sterile-filtered male plasma samples were purchased from Sigma-Aldrich (St. Louis, MO, USA).

1.3 Fluorescent analysis of serotonin

One microliter of 2 μM anti-serotonin aptamer and 1 μL of 2 μg/mL serotonin were added to 1X TAE buffer. Subsequently,

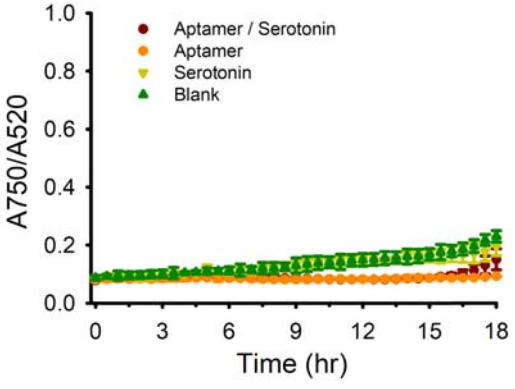

Figure S2: Time-absorbance ratio curve of the AuNPs synthesized *in situ* in the presence of aptamer/serotonin, aptamer, serotonin, and blank within 18 h. The final concentrations of aptamer, and serotonin were 20 nM, and 10 ng/mL, respectively.

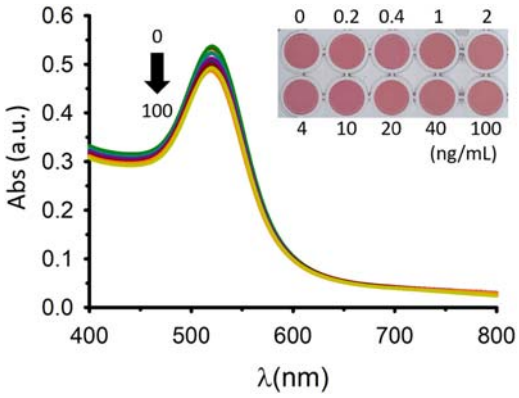

Figure S3: Absorption spectra of AuNP solutions at various serotonin concentrations before adding NaCl. The inset shows an image of an AuNP solution containing different concentrations of serotonin.

80 μL of reaction solution was incubated at 37°C for 30 min. Prior to measuring the fluorescence, 20 μL of an OliGreen reagent with 200-fold dilution was added to the solutions, and the total volume of the 100 μL mixture was equilibrated for 10 min. Finally, 1 μL of 40 mM $HAuCl_4$ was added to the mixture and gently shaken for 1 min. The fluorescence spectra were collected.

1.4 Data analysis

Each experiment was performed in triplicate. A four-parameter logistic equation was used to analyze the calibration curves calculated using the Sigma Plot software [S1]:

$$Y = B + \frac{(A - B)}{1 + \left(\frac{X}{C}\right)^{D}}$$

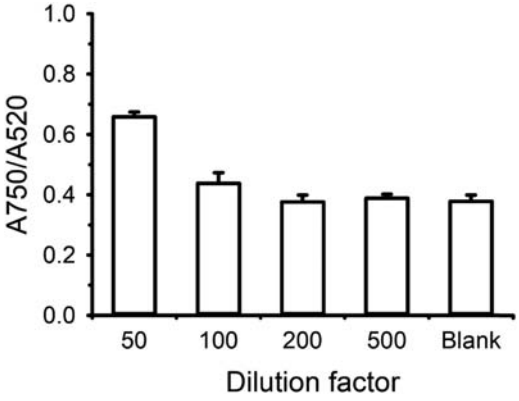

Figure S4: Effect of different dilutions (50-, 100-, 200-, and 500-fold) of serum samples on the spectral ratio of AuNPs without serotonin.

where Y is the absorption ratio ($A750/A520$), X is the concentration of serotonin, A is the highest value of $A750/A520$, B is the lowest value of $A750/A520$, C represents the concentration with the half-maximal absorption ratio, and D represents the slope of the inflection point (Table S1) (Figures S1–S4).

Reference

[1] Sun Y, Xia Y. Gold and silver nanoparticles: A class of chromophores with colors tunable in the range from 400 to 750 nm. Analyst. 2003;128(6):686–91.