Gold nanostars co-coated with the Cu(II) complex of a tetraaza macrocyclic ligand

Electronic Supporting Information

1. Materials and Syntheses

- *Materials*. Ethylbromoacetate (97%), potassium carbonate (99%), diethyl ether (99.8%), ethylenediamine (99%), dichloromethane (99.6%), sodium triacetoxyborohydride (97%), sodium borohydride (98%), propionaldehyde (97%), poly(ethylene glycol) methyl ether thiol (PEG-SH, average mw 2000), formic acid (96%), formaldehyde (37% wt in H_2O), sodium hydroxide(97%), propionaldehyde (97%), trifluoroacetic acid (99%), triethylsilane (99%), N,N'-disuccinimidyl carbonate (95%), lipoic acid (99%), trietylamine (99%), magnesium sulfate (97%), sodium bicarbonate (99.5%), sodium trifluoromethanesulfonate (98%), zinc trifluoromethanesulfonate (98%), copper trifluoromethanesulfonate (98%), iron(II) trifluoromethanesulfonate (85%), perchloric acid 70%, dichloroethane (99%), pentane (99%), ethanol (99.8%), 1,4-dioxane (99%), acetonitrile (99.9%), t-butanol (99.7%), silver nitrate (99.8%), tetrachlorauric acid (30% weight in diluted HCl), laurylsulphobetaine (97%), were purchased from Sigma Aldrich.

- *Compound **1*** (1,4,7-trimethyl-1,4,7,10-tetraazacyclododecane) **1** was prepared in two steps starting from Monobenzyl-N-cyclen. First, 1-benzyl-4,7,10-trimethyl cyclen was prepared. 10g of Monobenzyl-N-cyclen (0,0381mol, purchased from CheMatech, Dijon, France) were dissolved in 40mL of formic acid. Then, 35mL of formaldehyde were added slowly. At the end of the addition, some drops of water were added. The mixture was heated at reflux overnight. The solvent was evaporated and a solution of NaOH 13M was added until pH=12 was obtained. After extraction with DCM, the organic phase was dried over $MgSO_4$ and filtered on celite. After evaporation of the solvent, 13g of light yellow oil were obtained. 150mL of pentane were added and the mixture was stirred overnight. The mixture was filtered on celite and 10.3g of light yellow oil were obtained. The residue was purified by chromatography on alumina column (DMC:pentane 80:20, then DCM:pentane 50:50) to give 1-benzyl-4,7,10-trimethyl cyclen as a yellow oil. (4.67g, 0.0178mol, yield = 46.7%) **^{1}H NMR** (300 MHz, $CDCl_3$, 300K). δ (ppm): 2.24 (m, 9H, CH_3), 2.60 (m, 16H, CH_2), 3.56 (s, 2H, CH_2 benzyl), 7.29 (m, 5H, Ar). In the second step, 0.8g of Pd/C were added to a solution of 4.67g (0.0178mol) of 1-benzyl-4,7,10-trimethyl cyclen in 50mL of ethanol. The mixture was stirred in an hydrogen atmosphere at room temperature for 3 days. The mixture was filtered on celite and the solvent was evaporated to obtain **1** as a yellow oil (3.44g, 0.0161mol, yield = 90.4%).

^{1}H NMR (300 MHz, $CDCl_3$, 300K). δ (ppm): 2.14 (s, 3H, CH_3), 2.35 (s, 6H, CH_3), 2.49 (s, 8H, CH_2), 2.72 (m, 8H, CH_2). **MALDI-TOF** : 214.8 $(M+H)^+$

- *Compound* **2** (Ethyl 2-(4,7,10-trimethyl-1,4,7,10-tetraazacyclododecan-1-yl)acetate). 1.09g (0.00645mol) of ethylbromoacetate were dissolved in 100mL of CH_3CN and slowly added to a stirred mixture of **1** (1.38g, 0.00645mol) in CH_3CN (300mL) and K_2CO_3 (1.78g, 0.0129mol). The mixture was stirred overnight at room temperature. After filtration on celite, the solvent was evaporated. The residue was taken up in diethylether (200mL) and the mixture was stirred overnight. After filtration, the solvent was evaporated and **2** was obtained as a yellow oil (830mg, 0.00277mol, yield = 42.9%). **^{1}H NMR** (300 MHz, $CDCl_3$, 300K). δ (ppm): 1.20 (t, 3H, CH_3, J= 7.2Hz), 2.17 (9H, CH_3), 2.47 (12H, CH_2), 2.77 (m, 4H, CH_2), 3.35 (s, 2H, CH_2CO), 4.08 (q, 2H, OCH_2, J= 7.2Hz).

- *Compound* ***3*** (*N*-(2-Aminoethyl)-2-(4,7,10-trimethyl-1,4,7,10-tetraazacyclododecan-1-yl)acetamide). 1.66g of ethylendiamine (0.0277mol, 10eqv) were added to 830mg of **2** (0.00277mol). The mixture was stirred at room temperature for 3 days. Ethylenediamine was evaporated to obtain a yellow oil. The oil was taken up in diethylether. Solid impurities were removed by filtration and the solvent was evaporated to give **3** as a yellow oil (594mg, 0.00189mol, yield = 68.2%). **^{1}H NMR** (300 MHz, $CDCl_3$, 300K). δ (ppm): 1.65 (9H, CH_3), 1.93 (m, 4H, CH_2), 2.10 (m, 12H, CH_2), 2.31 (t, 2H, CH_2NH_2, J=6.0Hz), 2.63 (s, 2H, CH_2CO), 2.84 (q, 2H, CH_2NH, J=6.0Hz), 8.81 (1H, NHCO).

- *Activated lipoic acid*. 3g of N,N'-Disuccinimidyl carbonate (0.0118mol, 1.5eqv) were added to a solution of 1.6g of lipoic acid (0.00787mol) and 2.4g of triethylamine (0.0236mol, 3eqv) in 80mL of CH_3CN. The mixture was stirred overnight at room temperature. The solvent was evaporated at temperature lower then 30°C. 100mL of a solution of $NaHCO_3$ 5% were added and the compound was extracted with ethyl acetate. The organic phase was dried over $MgSO_4$ and the solvent was evaporated to give the activated lipoic acid as a yellow solid (2.49g, 0.0082mol, yield=100%). **^{1}H NMR** (300 MHz, $CDCl_3$, 300K) δ (ppm): 3.58 (m, 1H), 3.16 (m, 2H), 2.84 (s, 4H), 2.63 (t,2H), 2.48 (m, 1H), 1.94 (m, 1H), 1.47-1.86 (m, 6H).

- *Gold nanostars*. The synthesis was carried out as described in references 6 and 7, main text. The reactants concentrations used for this paper are described here in detail. The seed solution was prepared in a 20mL vial. 5mL of $HAuCl_4$ 5×10^{-4}M in water were added to 5mL of an aqueous solution of LSB 0.2M. The mixture was gently hand-shaken and a pale yellow colour was obtained. Then, 0.6mL of a previously ice-cooled solution of $NaBH_4$ 0.01M in water were added. The mixture was gently hand-shaken and a reddish-brown colour appeared. The seed solution was kept in ice and used within few hours. The growth solution was prepared in a 20mL vial, 180µL of

$AgNO_3$ 0.004M (7.0×10^{-5}M in the final growth solution) in water, 5.0 mL of $HAuCl_4$ 0.001M in water were added in this order to 5mL of an aqueous solution of LSB 0.2M. Then, 95μL of an aqueous solution of ascorbic acid 0.0788M were added (7.3×10^{-4}M in the final growth solution). The solution, after gentle mixing, became colourless. Soon after, 12μL of the seed solution were added. The solution was gently hand-shaken and a grey colour appeared and quickly changed to blue and became more intense. The samples were allowed to equilibrate for 1h at room temperature, then were ultracentrifugated (13000 rpm, 20 min), the supernatant discarded and the pellet of GNS redissolved in 10.0 mL bidistilled water).

Instrumentation. Phototothermal experiments were run on solutions using a multimode AlGaAs laser diode (model L808P200, by Thorlabs GmbH), characterized by a maximum output power of 1000 mW, and emitting light at the wavelength of about 808 nm, and reading the temperature with a FLIR E40 thermocamera.

Absorbance spectra of solutions were recorded with a Varian Cary 50 spectrophotometer in the 200–1100 nm range. TEM images were taken with Jeol JEM-1200 EX II instrument on 10 μL colloidal solution drops, deposited on Nickel grids (300 mesh) covered with a Parlodion membrane. The ^{1}H NMR spectra were recorded at room temperature on a Bruker Avance II 300 (300 MHz) or on a Bruker Avance DRX 600 (600 MHz) spectrometer at the "Welience, Pôle Chimie Moléculaire de l'Université de Bourgogne (WPCM)". Chemical shifts (^{1}H NMR spectra) are expressed in ppm relative to chloroform (7.26 ppm).

Mass spectra were obtained on a Bruker Daltonics Ultraflex II spectrometer in the MALDI/TOF reflectron mode using dithranol as a matrix or on a LTQ Orbitrap XL (Thermo Scientific) instrument in ESI mode. Differential scanning calorimetric measurements have been performed in the Q2000 equipment by TA Instuments by heating about 3 mg of each sample in an Al open crucible under nitrogen flow at 5°C/min from 0°C to 250 °C for pure PEG (melting point at 53 °C) and pegylated GNS and up to 450 °C for the [Cu**L1**]$(CF_3SO_3)_2$ and the $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ ($CF_3SO_3^-$ salt).

Two photon excitation was primed by a Ti:Sapph laser (Tsunami, Spectra Physics, CA) tuned at 800 nm, pulsed at 80 MHz (200 fs pulse width on the microscope stage). The solutions were hosted in a 8 well glass slides (Lab-Tek II chamber slide system, Nunc, USA) set on the sample stage of a Nikon TE300 inverted microscope coupled to the Ti:Sapph laser. The fluorescence output was detected through a 670 nm beam splitter (Chroma, Inc., Brattelboro, VT) with no further emission filtering. The side (C) port of the microscope was coupled to a custom cross-correlation detector with two SPAD modules (SPCM-AQR15 Perkin- Elmer, DE) for Fluorescence Cross-Correlation

measurements. The TPL emission spectra were recorded by means of a CCD (DV420A-BV, Andor, IRL)-based spectrometer (MS125, Lot-Oriel,UK), connected to the front port of the microscope.

2. Yield of GNS

Five 10mL samples of gold nanostars were prepared as described in the main text and after the functionalization with PEG-SH underwent 4 cycles of ultracentrifugation (13000rpm, 25')/redissolution of the pellet in water (10mL). The samples were oxidized with 2mL of aqua regia, diluted 1:20 with bidistilled water. The content of gold was measured by ICP-OES. The average yield of Au is 75(5)%.

*3. UV-Vis spectra after addition of [Cu**L1**]$^{2+}$ to a solution of non-pegilated GNS*

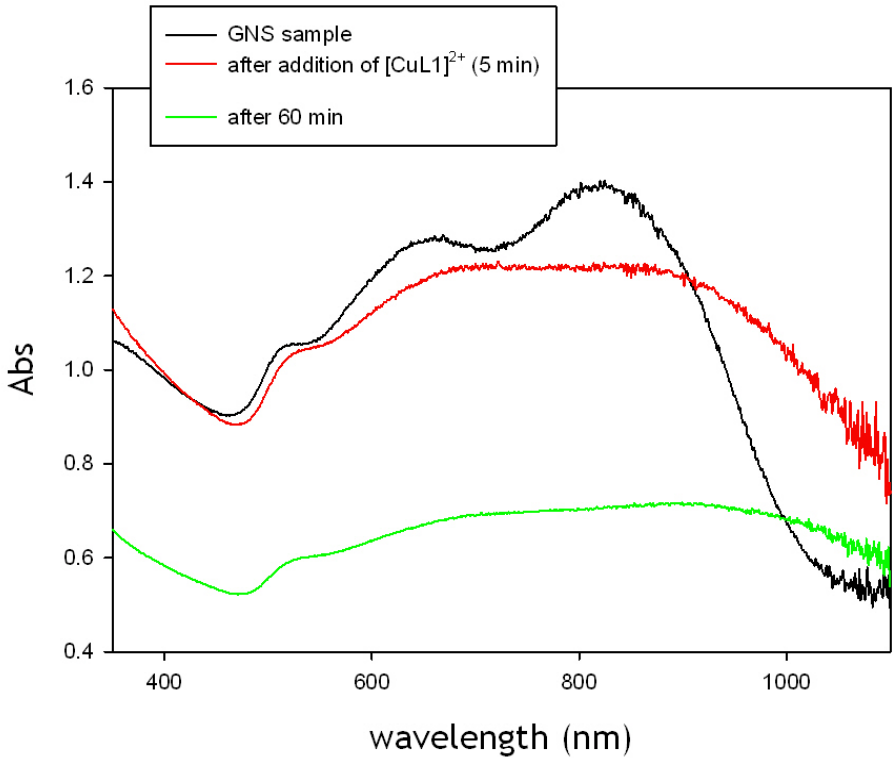

Figure S1 – GNS spectra before and after addition of [Cu**L1**]$^{2+}$

*4. TEM images of pegilated GNS and of [Cu$_n$(**L1**@GNS)]$^{2n+}$.*

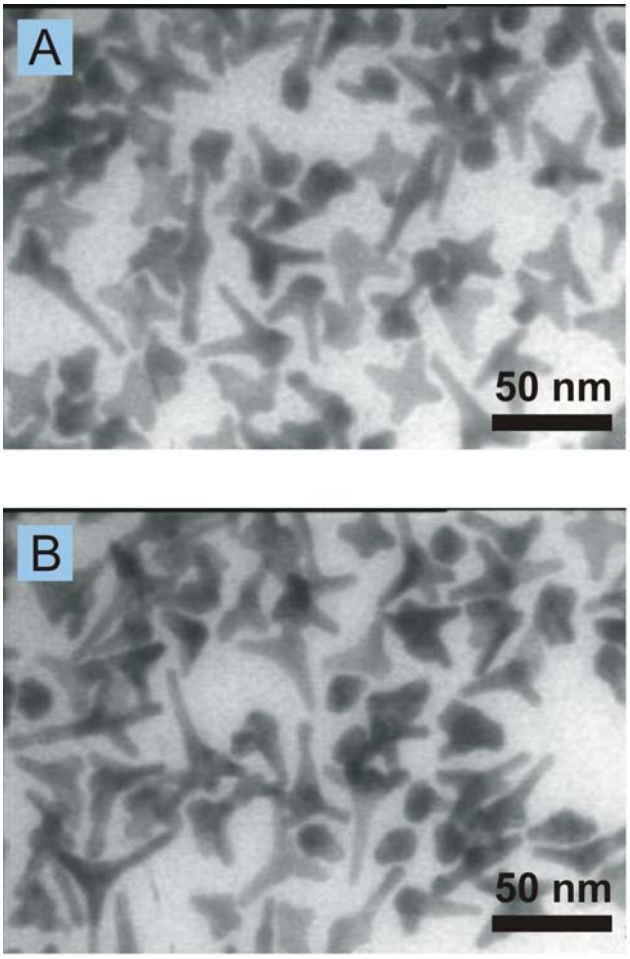

Figure S2 - Images are taken before (A) and after (B) treatment with 10^{-3} M [Cu**L1**]$^{2+}$

5. Morphological analysis and calculation of the average mass of a single GNS.

i) Transmission Electron Microscopy (TEM). A sample of gold nanostars was further diluted 20 times with bidistilled water. 10μL of the diluted sample were deposited on a copper grid (300 mesh) covered with a Parlodion membrane and allowed to dry in a dessicator. Images were taken using a JEOL JEM-1200 EX II 140 instrument. The average length of a branch, the width at the base, the diameter of the core of the nanoparticles were measured for both regular and penta-twinned nano-objects analyzing the images through the software Image J. The Aspect Ratio of the branches is calculated as the ratio between the average length of a branch and the average base width.

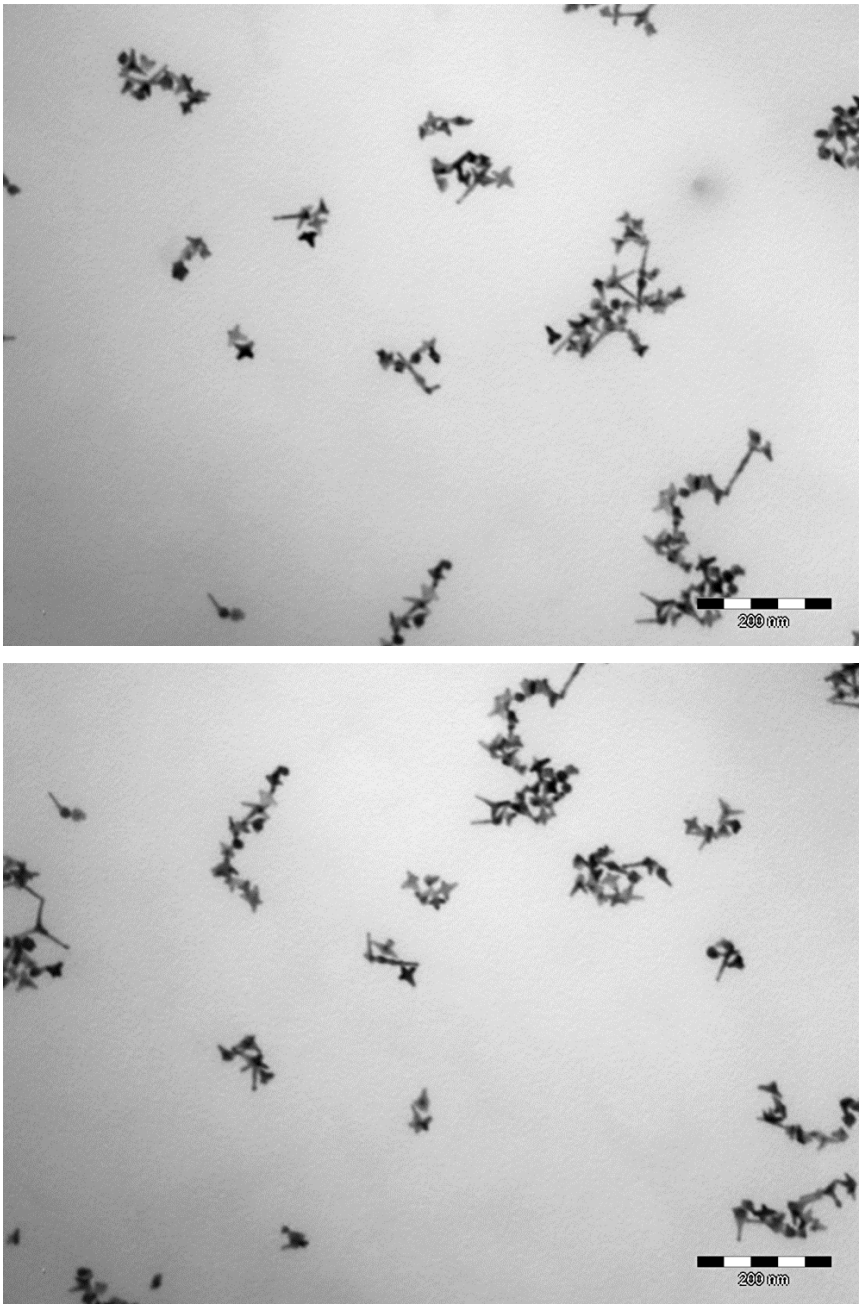

Figure S3 – Representative TEM images used for morphological analysis

Regular gold nanostars			
Length of the branch	***Width at the base***	***AR***	***Diameter of the core***
12(3)nm	***10(2)***	***1.2***	***11(2)***

Irregular gold nanostars			
Length of the branch	***Width at the base***	***AR***	***Diameter of the core***
20(9)nm	***11(2)***	***1.8***	***11(2)***

ii) Volume and mass calculation. A geometrical model was used to calculate the mass of a single object (both regular and irregular nanostars). For pentatwinned nanoparticles the average number of the formed branches was also calculated and it is equal to 3. For regular monocristalline nanostars the number of the branches was taken as six (four on the XY plane and two on the Z axis see ref 9b anc main text). The volume of an object was calculated as the sum of the volume of a central spherical core and of the volume of three or six (irregular and regular, respectively) conical branches.

- Regular nanostars.

The core was assumed to be a sphere with ray r_1=5.5nm. $V_{core} = \frac{4}{3}\pi r_1^3$ = 696nm^3

The branches were assumed to be conical with height h=12nm, ray of the base r_2=5nm. $V_{branch} = \frac{\pi r_2^2 h}{3}$ = 314nm^3

$V_{regular\ nanostar} = V_{core} + 6\ V_{branch}$ = 2580nm^3

The mass is calculated considering the density of gold (d=19.3g/mL). $M_{regular\ nanostar}$ = **5×10^{-17}g**

- Pentatwinned nanostars.

The core was assumed to be a sphere with ray r_1=5.5nm. $V_{core} = \frac{4}{3}\pi r_1^3$ = 696nm^3

The branches were assumed to be conical with height h=20nm, ray of the base r_2=5.5nm. $V_{branch} = \frac{\pi r_2^2 h}{3}$ = 633nm^3

$V_{regular\ nanostar} = V_{core} + 3\ V_{branch}$ = 2595nm^3

The mass is calculated considering the density of gold (d=19.3g/mL). $M_{regular\ nanostar}$ = **5×10^{-17}g**

iii) calculation of the GNS concentration in the colloidal solutions.

In both cases, the average mass of a single GNS is 5×10^{-17} g. The concentration of $HAuCl_4$ in the growth solution is 5×10^{-4} M and as the yield of conversion is 75 %, the concentration of gold transformed into GNS is 3.75×10^{-4} M. This corresponds to 0.07386 g Au/l.

Dividing by the mass of a single GNS we obtain 1.477×10^{17} GNS/l. Dividing by the Avogadro number, **the concentration of GNS in mol/l is obtained (2.45×10^{-9} M)**

6. Differential scanning calorimetry.

DSC has been carried out on pegilated GNS and on pegilated GNS after the exchange reaction with 10^{-4} M $[Cu\mathbf{L1}]^{2+}$ ($CF_3SO_3^-$ anion). This has been done preparing a large solution (200 mL) of pegilated GNS, and dividing it in two parts. The first unverwent two ultracentrifugation/redissolution cycles, and the obtained pellet was dried in a nitrogen flux and

under vacuum, and finally analyzed by DSC. The second was treated with 10^{-4} M [Cu**L1**]$(CF_3SO_3)_2$ for 1 hour at 60 °C, then the solution unverwent two ultracentrifugation/redissolution cycles and the obtained pellet was dried and analyzed by DSC. By this way, the quantity of grafted PEG in the second solution was identical to that of the first, before adding $[Cu\mathbf{L1}]^{2+}$.
Moreover, pure PEG-SH and [Cu**L1**]$(CF_3SO_3)_2$ were also indagated by DSC.

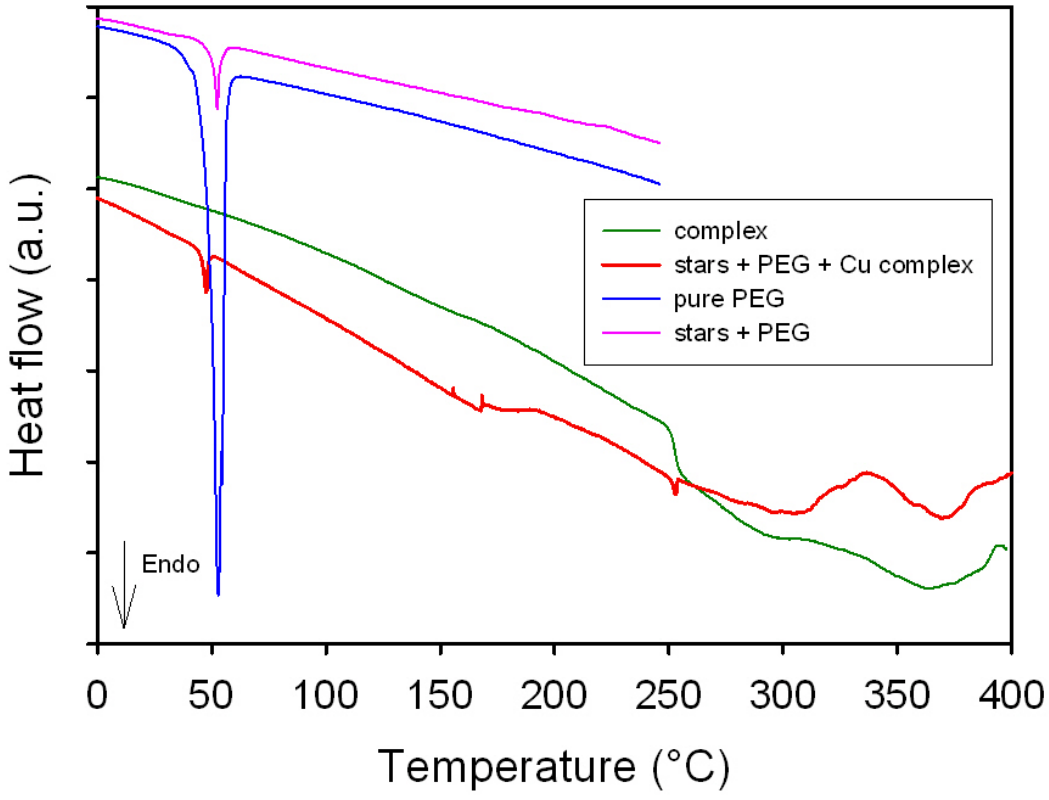

Figure S4 – DSC analysis

DSC measurements have been performed in the Q2000 equipment by TA Instruments by heating about 3 mg of each sample in an Al open crucible under nitrogen flow at 5°C/min. Pure PEG-SH (MW 2000) and GNS coated with PEG-SH have been heated up to 250 °C (melting of pure PEG is at about 53 °C – peak temperature). [Cu**L1**]$(CF_3SO_3)_2$ and $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ ($CF_3SO_3^-$ salt) have been heated up to 450 °C.

In the calorimetric profiles of the two Au stars samples (red and purple profiles, see Figure), the endothermic peak due to the melting of PEG-SH is evident. By comparing the melting enthalpy of the pure PEG-SH (155.9 J/g, blue profile) with the values measured in pegilated GNS (7.67 J/g) and $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ ($CF_3SO_3^-$ salt) (5.91 J/g) it is possible to estimate the mass % amount of PEG-SH coating the stars themselves, i.e. **4.92 %** and **3.79 %** respectively.

By comparing the calorimetric profiles of the $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ ($CF_3SO_3^-$ salt) with the trace of the pure Cu complex, it is possible to clearly recognize the presence of the complex itself on the GNS, thanks to the similarity of the measures starting from around 220 °C. However the quantification is impossible. This is due to the fact that the pure Cu complex, i.e $[Cu\mathbf{L1}](CF_3SO_3)_2$ was obtained only in a colloidal form. It was precipitated as an oil from its tert-butanol solution using diethyl ether, and the oil was dried under vacuum (oil pump) but not obtaining a crystalline powder. Its calorimetric profile shows a glass transition at 250 °C and the subsequent decomposition at temperatures higher than 350 °C. On the contrary, in the $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ sample, the complex is probably present in crystalline form, melting at 250 °C (see the small endo peak on the calorimetric profile, red line, close to the glass transition of the complex) and subsequently decomposing.

7. Calculation of % (weight) and number of $[Cu\mathbf{L1}]^{2+}$ and PEG-SH in pegilated GNS and in $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$.

a) Pegilated GNS. DSC analysis (SI 4) allows to determine that in GNS coated with PEG-SH (mw 2000) the latter is 4.92 % weight.

100 g coated GNS = 4.92g PEG-SH + 95.08 g Au*

In 100 g: 2.46×10^{-3} mol PEG-SH.

A single GNS has an average mass of 5.0×10^{-17}g (vide supra). In 100g there are 1.90×10^{18}GNS units, i.e. (dividing by the Avogadro's number) 3.16×10^{-6} mol GNS.

Dividing the mol of PEG-SH in 100g product by the mol of GNS in 100 g product we obtain the average number of PEG-SH on a single GNS:

PEG-SH 779/GNS

b) $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$. From DSC analysis, we determined that in 100 g of the product PEG-SH is 3.79g (the co-coated GNS used for this determination were prepared using PEG-SH coated GNS and displacing PEG-SH with 10^{-4} M preformed $[Cu\mathbf{L1}]^{2+}$.

As we use $Cu(CF_3SO_3)_2$ as the Cu(II) salt to form $[Cu\mathbf{L1}]^{2+}$, we assume that the counter anions are trifluoromethanesulfonate.

Molecular weight of $[Cu\mathbf{L1}](CF_3SO_3)_2$ = 863.7

PA Au = 196.96

Calling X the mol of $[Cu\mathbf{L1}](CF_3SO_3)_2$ and Y the mol of Au, we write

100g of $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ = 3.79g PEG-SH + Xmol×863.7g/mol + Ymol ×196.96g/mol

The ratio Y/X is obtained from an independent experiment, i.e. ICP-OES for the same type of preparation. For $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ prepared by displacement on pegilated GNS with 10^{-4} M $[Cu\mathbf{L1}]^{2+}$ we obtained Cu = 1.58×10^{-6} M. In the GNS solutions, Au concentration is 3.75×10^{-4}M. This allows to calculate Y/X as 3.75×10^{-4}M/1.58×10^{-6}M = 237.3

We can now write

100g of $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ = 3.79g PEG-SH + Xmol×863.7g/mol + 237.3Xmol×196.96g/mol

X = 2.021×10^{-3} mol $\Rightarrow$ 1.75 g of $[Cu\mathbf{L1}](CF_3SO_3)_2$ in 100 g product.

Y = 237.3×2.021×10^{-3}mol = 0.479 mol Au $\Rightarrow$ 94.46 g Au in 100 g product

Summary of % weight in $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$:

Au	94.46%
PEG-SH	3.79% (1.89×10^{-3} mol)
$[Cu\mathbf{L1}]^{2+}$	1.75% (2.02×10^{-3} mol)

From mass of Au in 100g and a mass of a single GNS we can obtain the number of GNS in the same quantity and in turn the mol of GNS in 100 g product: 3.14×10^{-6} mol.

PEG-SH	602/GNS
$[Cu\mathbf{L1}]^{2+}$	645/GNS

* In the used synthetic conditions 6-8 % Ag is included in the GNS crystals (see refs 9b,c main text). The consequent slight weight variation is neglected here, considering for sake of simplicity that GNS are made of Au only.

*8. pH-metric titration of the Cu^{2+} complex of **L2**.*

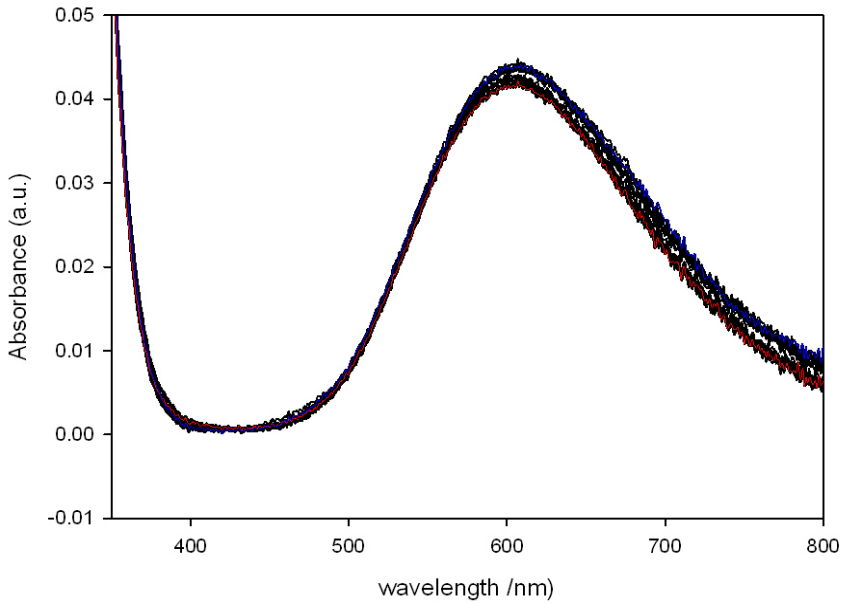

Figure S5 - Series of spectra obtained in the pH metric titration from pH 3.9 (red, [Cu**L2**$(H_2O)]^{2+}$), to pH 10.5 (blue, [Cu**L2**$(OH)]^{+}$)

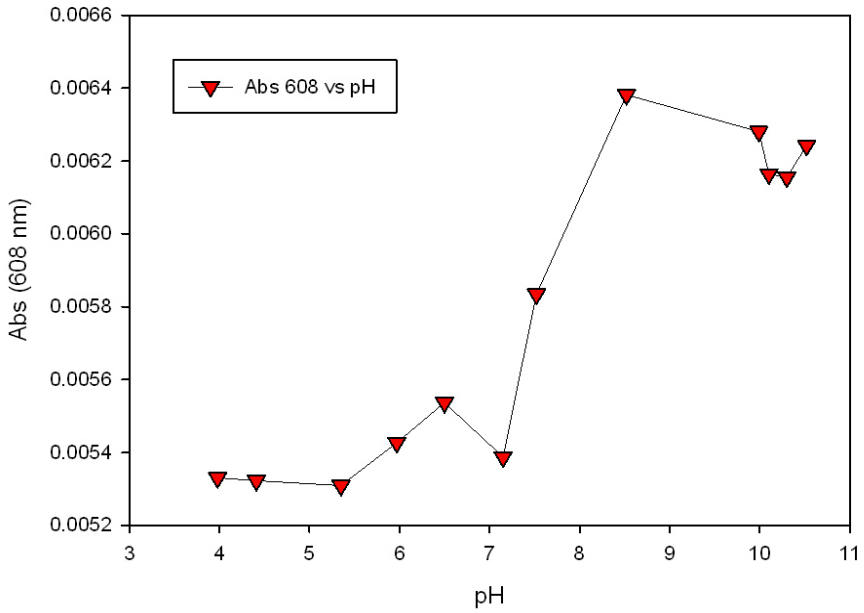

Figure S6 - Plot of Absorbance at 608 nm vs pH, data taken from the previous figure (the line is not a fit but a simple connection to guide the eye)

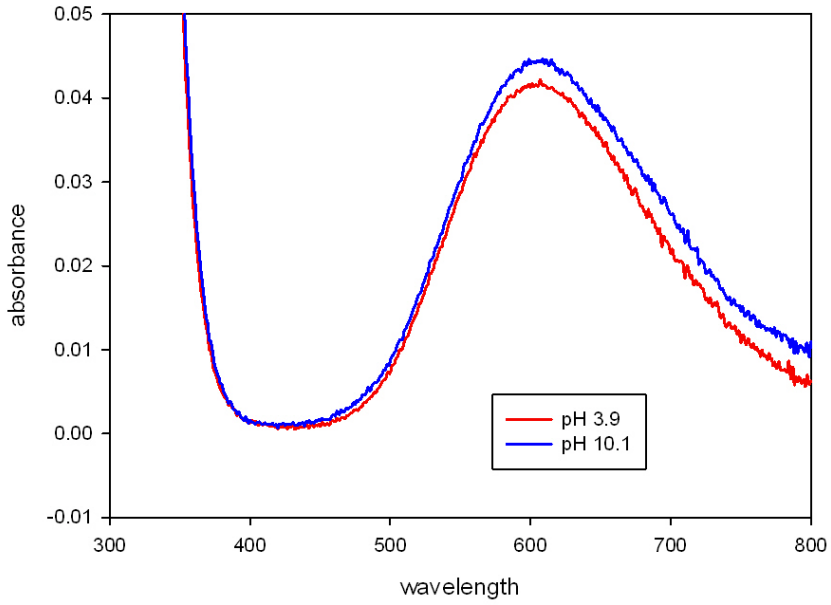

Figure S7 - Spectra at pH 3.9 (red) and 10.1 (blue), evidencing the changes in the absorption band

*9. Solution absorption spectra after the addition of void **L1** to pegilated GNS.*

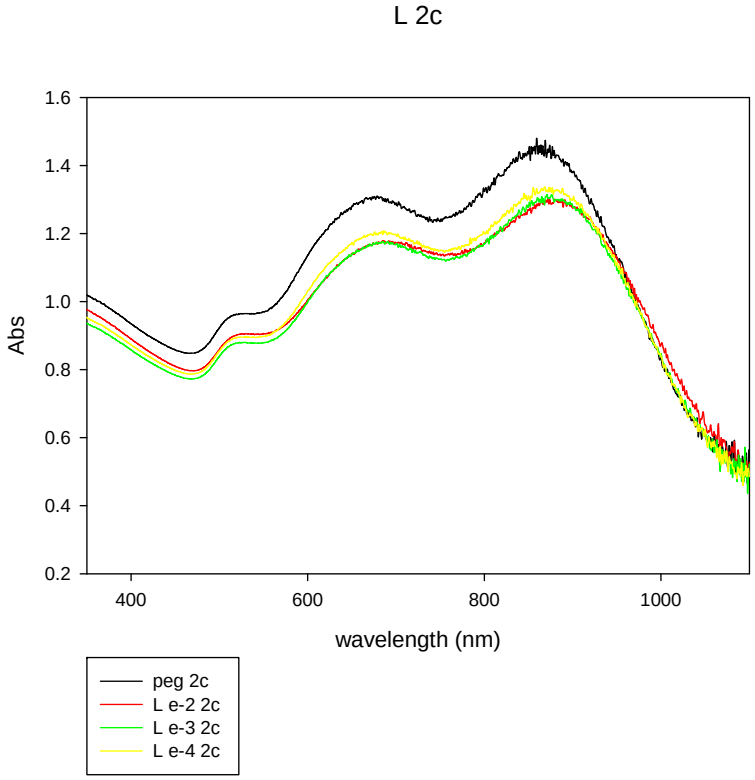

Figure S8 - Black: spectrum of the pegilated GNS solution; yellow: after addition of 10^{-4} M **L1**; green: after addition of 10^{-3} M **L1**; red: after addition of 10^{-2} M **L1**.

*10. Z potential vs pH titration on **L1**@GNS*

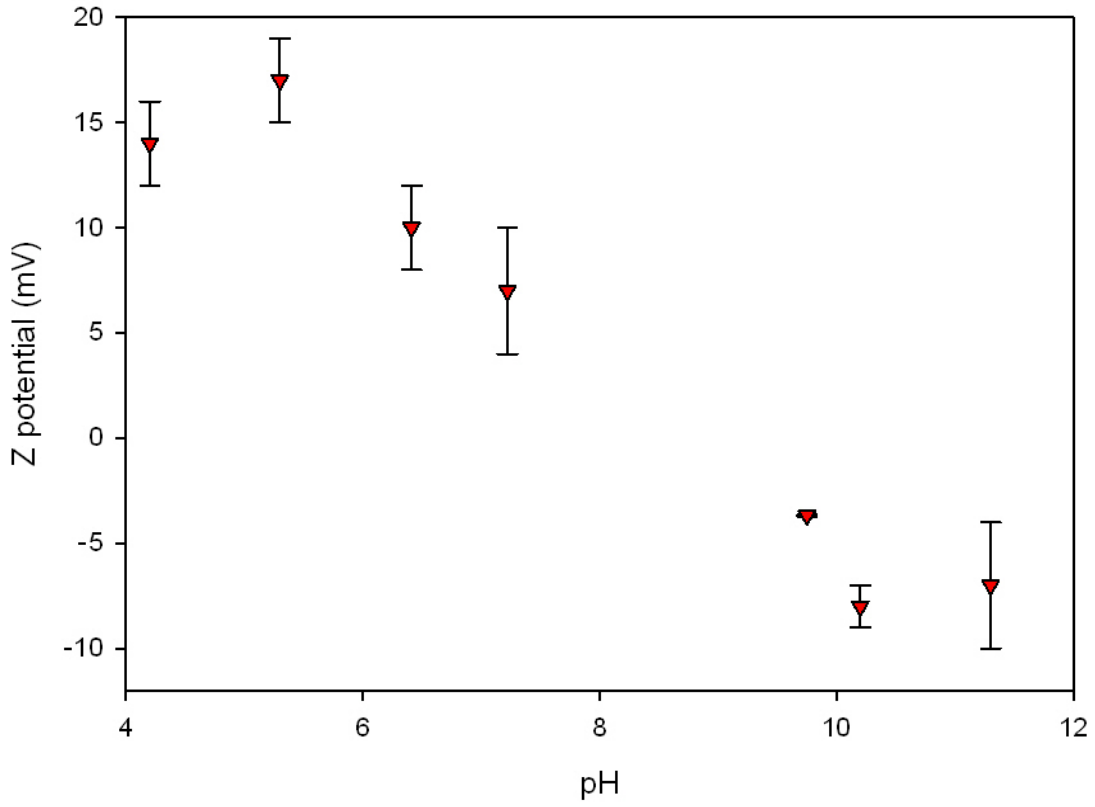

Figure S9 - Titration carried out on a 10 ml solution of **L1**@GNS, prepared with ligand exchange (**L1** = 10^{-4} M); pH was changed by microadditions of standard NaOH and $HClO_4$ (10^{-1} M)

11. TPL emission spectra and brightness test on pegilated GNS and of $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$.

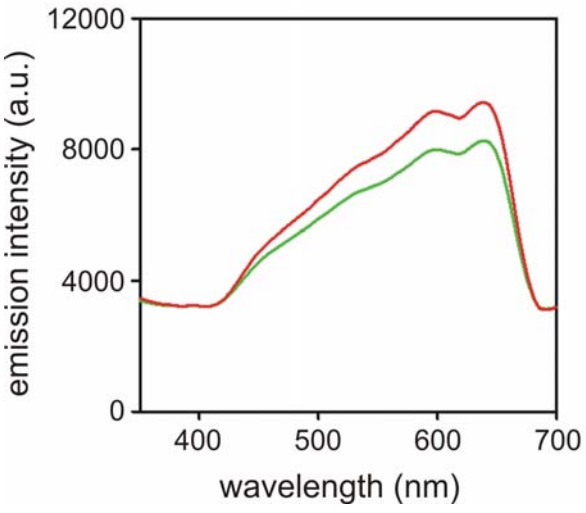

Figure S10 - TPL emission spectra of pegilated GNS (green line) and of $[Cu_n\mathbf{L1}@GNS]^{2n+}$ (red line), using a 800 nm excitation source (the intensity drop at longer wavelength is due to a cut off filter applied at 670 nm).

Brightness test. As it is described in the main text, half of a bulk (20 ml) pegilated GNS solution was treated with 10^{-4} M $[Cu\mathbf{L1}]^{2+}$ at 60 °C, then the brightness was tested. Excitation wavelength = 800 nm, filter 560/40 nm. Power = 22 mW.

Reference: Rhodamine 6G in EtOH. Fit with a single diffusive component: Diff.Coeff=300 $\mu m^2/s$

Pegilated GNS and $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ show much slower diffusion and a rapid exponential decay a small lag times. The latter is due to rotational diffusion and has been fit with two exponential decays.

The overall rate of the suspension was very similar for the two species. However it must be taken into account that the concentration also was different ($C_{GNS} > C_{[Cun(\mathbf{L1}@GNS)]2n+}$) and that the diffusion coefficient also was different ($D_{GNS} \approx 2\ D_{[Cun(\mathbf{L1}@GNS)]2n+}$). This implies that the Cu treated GNSs are electrostatically aggregated during such experiment (aggregation number ≈ 6). NB: "aggregation" does not mean at all that GNS undergo and Ostwald ripening process, as it can be sharply stated from their constant NIR absorption spectrum. When rescaling for this aggregation effect the brightness (number fo counts per second and per object) is very similar for the pegilated GNS and $[Cu_n(\mathbf{L1}@GNS)]^{2n+}$ (see Table).

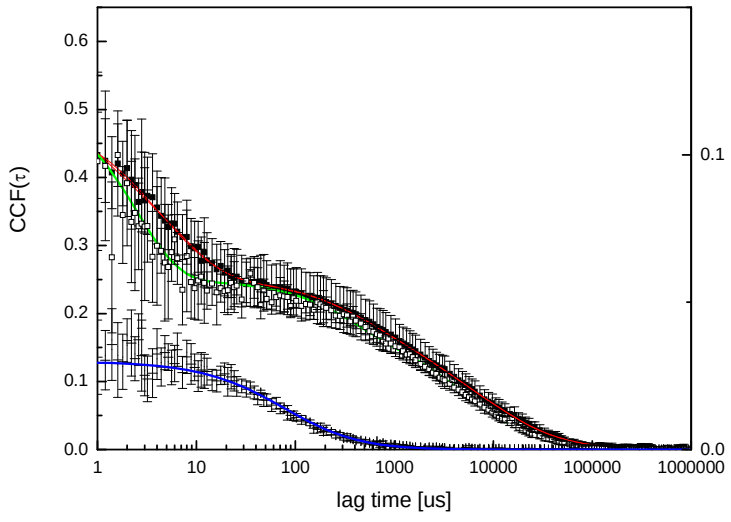

Figure S11 - Cross-correlation functions for Rh6G (blue), GNS-PEG (green and open squares) and GNS-PEG-Cu (red and closed squares). The solid lines are best fit functions. Power = 22 mW.

Table: summary of the best fit results

	w0	0.542	um
	Power =	22	mW
	Rh6G	GNS (pegilated)	$[Cu_n(\mathbf{L1}@GNS)]^{2n+}$
<C>[nM]	21	9.6	1.5
Diff[um^2/s]	300	6.2	3.4
Radius[nm]	0.707376165	34.22788	62.41554397
Brightness [kHz/molecule]	3.4	8.2	59(8.3)*

*: 59 kHz is the brightness measured for the single diffusing object in the GNS-PEG-Cu suspension. However the diffusion is slower and the aggregation number is about 6. When rescaling to the single object (59/6) we get 8.3, very similar to the case o GNS-PEG nanoparticle.
Notice that pegilated has an average Hydrodynamic Radius = 34 nm