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Supporting Information for
Plasmonic Control of Drug Release Efficiency in Agarose Gel Loaded with Gold-Nanoparticle Assemblies

1 NPs synthesis

Gold nanoparticles (Au NPs) were synthesized based on the Turkevich method, as modified in the seeded-growth one-pot strategy developed by Silvestri and co-workers [1]. The seed solution (composed by 7.5 mL of HAuCl₄ 10 mM, 9 mL of Na₃Ct 2% w/w and 420 μL of AgNO₃ 0.1 % w/w) was prepared and stirred at room temperature for 6 minutes and then blended to 250 mL of milliQ water at 100 °C. The nucleation took place immediately, but in order to achieve a complete reduction of the gold precursors and a good homogenization of the crystals, the reaction mixture was kept at 100 °C and maintained under stirring at 750 rpm for 1 hour. Afterwards, the obtained seeds solution was cooled down to room temperature and 5 mL of glycerol were added, leaving the system stir for 15 minutes more. In the meanwhile, the growing solution (composed by 7.5 mL of HAuCl₄ 10 mM, 10 mL of Na₃Ct 1% w/w and 426 μL of AgNO₃ 0.1% w/w) was prepared and stirred at room temperature for 6 minutes, before being added to the system, followed immediately by 8 mL of an aqueous solution of hydroquinone 1% w/w. The system was stirred for 70 minutes, obtaining in the end a colloidal solution of spherical Au NPs dispersed in sodium citrate that acts as temporary stabilizing agent. PEGylated Au NPs were synthesized adding a basified aqueous solution of the desired thiolated PEGs (30 mg, 1:1 HS-PEG5000-OMe:HS-PEG5000-COOH, 5 mL of milliQ water and 100 μL of NaOH 1M) to the colloidal solution of Au-NPs. The system was stirred for at least 48 hours at room temperature in order to complete the functionalization process. The coated NPs were then purified and concentrated using Amicon centrifugal filter units (5 mL of NPs solution at 6000 rpm for 5 minutes) and named Au-PEG NPs. The resulting NPs were fully characterized by UV–vis absorption, dynamic light scattering (DLS) and transmission electron microscopy (TEM). Static absorption spectra were obtained using a Jasco® V-570 UV-VIS-NIR spectrophotometer. The samples were measured at room temperature in a 1-cm optical path plastic cuvette, in which a gel of 3-mm-thickness was fixed with water. Same samples conditions were used in the ultrafast pump-probe measurements. Final particle size was determined by DLS (Malvern, Zetanano ZS, US); analyses
were performed in triplicate and reported data show the average of three runs where the standard deviation was always below 5%. Size and morphology of produced NPs were also confirmed by TEM (using an EFTEM Leo 912AB, at 80 kV, by Karl Zeiss, Jena, Germany). Samples were prepared placing 5 mL drop of NP dispersion on a Formvar/carbon-coated copper grid and dried overnight. Digital images were acquired by a charge-coupled device (CCD; Esi Vision Proscan camera).

2 Hydrogel synthesis and characterization techniques

Polymeric solution was achieved by mixing polymer powders of agarose and carbomer 974P in phosphate buffer saline (PBS) solution in batch condition, adding a mixture of cross-linking agents made of propylene glycol and glycerol along with NaOH 1 N (reaction pH was kept neutral) [2]. The gelation onset was achieved by means of electromagnetic stimulation (500 W irradiated power) heating in ratio of 1 min per 10 mL of polymeric solution at 80°C. Microwave-enhanced chemistry is based on the efficient heating of materials by “microwave dielectric heating” effects. During hydrogels (HGs) cooling phase, slightly above 37 °C, HGs at sol state were mixed with Au NPs and Au-PEG NPs solutions (3 mL of NPs dispersion/mL of hydrogel): this procedure allows solute loading still during sol state, i.e. before sol/gel transition. The loaded HG-NPs systems were cast in standard plastic 48-well cell culture plates, 0.5 mL/well (diameter = 1.1 cm). Gelation studies were performed using an inverter tube test compared with blank HGs.

The polymers used in this work were: derivatives of polyethylene glycol 5000, in details HS-PEG5000-OMe, HS-PEG5000-COOH (MW = 5 kDa, by Rapp Polymer GmbH, Tuebingen - Germany, employed as received and stored at -20 °C under dry argon atmosphere); carbomer 974P (MW = 1 MDa, by Fagron, The Netherlands) and agarose (MW = 200 kDa, by Invitrogen Corp., USA). All other chemicals were purchased from Merck (previously Sigma Aldrich Chemie GmbH, Deisenhofen, Germany). The materials were used as received, without further purifications. Solvents were of analytical grade. The glassware employed for the synthesis was cleaned using aqua regia, a 3:1 mixture of HCl and HNO₃. All the aqueous solutions were prepared using milliQ water, an ultrapure deionized water.

2.1 Gelation studies

Gelation of HG-NPs was assessed using the inverted tube test comparing with neat HG. Two ml microcentrifuge tubes (Fisher Scientific, Ottawa, ON, CA) were filled with 900 μl PBS and equilibrated to 37 °C. 100 μl of the polymer solution was injected into the bottom of the tube and incubated at 37 °C. At 2, 5, 10, 15, and 20 min intervals, tubes were inverted to observe if the gel flowed. The time at which the gel did not flow was recorded as the gelation time.

2.2 Rheology

Rheological analyses were performed, on HG-NPs systems, at 37 °C, using a Rheometric Scientific ARES (TA Instruments, New Castle, DE, USA) equipped with parallel plates of 30 mm diameter with a a 4-mm gap. Oscillatory responses, dynamic frequency sweep tests (G’, elastic modulus, and G’”, loss/viscous modulus) were determined at low strain values over the frequency range 0.1–500 rad/s. Dynamic Strain Sweep tests were also performed at frequency of 20 rad s⁻¹ over the strain range of 0.01–100%.

2.3 Swelling behavior

To assess swelling kinetics, solid samples were freeze-dried, weighed (W₀) and poured in excess of PBS to achieve complete swelling at 37 °C in 5% CO₂ atmosphere. The swelling kinetics was measured gravimetrically: samples were removed from PBS at regular time points, they were wiped with moistened filter paper in order to remove the excess of PBS and then weighed (Wₜ). Swelling ratio is defined as follows: (Wₜ - W₀) / W₀.
2.4 Mathematical modelling for the estimation of drug diffusivity

Drug diffusivity \( (D) \) of sodium fluorescein (SF) and fluorescein-dextran 70 kDa (DEX) were evaluated with a mathematical model based on mass balances, i.e. on fundamental conservation laws. Diffusion is described through the second Fick law with a 1-dimensional model in a cylindrical geometry, as presented in Eq. (1). Here radius \( (r) \) is the characteristic dimension for the investigated transport phenomenon. Therefore, the increase of mean drug concentration is triggered by the material flux, which takes place at the PBS/hydrogel surface. Eqs. (5) and (6) represent the boundary conditions for the left and the right border, respectively. The first one implies profile symmetry at the center (that is, with respect to the cylindrical axis), while the second one represents the equivalence between the material diffusive fluxes at the PBS/hydrogel surface. The entire set of equations of the model reads then as follows:

\[
\frac{\partial C_G}{\partial t} = D \frac{1}{r^2} \frac{\partial}{\partial r} \left( r^2 \frac{\partial C_G}{\partial r} \right), \quad (1)
\]

\[
V_s \frac{\partial C_G}{\partial t} = k_C S_{exc} (C_G - C_S). \quad (2)
\]

\[
C_s(t = 0) = 0, \quad (3)
\]

\[
C_C(t = 0) = C_{G,0} = \frac{m_{G,0}}{V_G}, \quad (4)
\]

\[
\left. \frac{\partial C_G}{\partial r} \right|_{r=0} = 0, \quad (5)
\]

\[
-D \left. \frac{\partial C_G}{\partial r} \right|_{r=R} = k_C (C_G - C_S). \quad (6)
\]

The two mass balance equations (Eqs. (1) and (2) with Eqs. (3) and (4) as initial conditions) involve the mean drug concentration within the hydrogel \( (C_G) \), the mean drug concentration in the outer solution \( (C_S) \), the volume of the solution \( (V_s) \), the hydrogel volume \( (V_G) \), the drug mass present inside the matrix \( (m_{G,0}) \) and the exchange interfacial surface \( (S_{exc}) \), i.e. the boundary surface between gel and surrounding solution (which can be here considered to be the side surface only, as a simplification). Finally, \( D \) represents the diffusion coefficient and \( k_C \) the mass transfer coefficient. The latter is computed through Sherwood number (Sh) obtained by means of penetration theory expressed according to Eq. (7):

\[
Sh = \frac{8}{\pi} = \frac{k_C \cdot 2r}{D}. \quad (7)
\]

Solution of Eqs. (1)-(2) was calculated numerically assuming gel dimensions to remain constant, the latter being a reasonably valid assumption as degradation phenomena occur at a much slower rate than delivery.

3 Hydrogel characterization

Hydrogels loaded with Au NPs were prepared through chemical cross-linking of the two polymers, agarose (AGR in Figure S1) and carbomer 974P (CBM in Figure S1), by microwave-assisted free radical polymerization. Heating to 80 °C leads to a higher macromer mobility, and thus enhances short-range interconnections among functional groups of the polymers. Esterification, the main reaction, takes place between carbomer carboxyl groups and the agarose hydroxyl groups, producing local networks also known as microgels. As polycondensation proceeds, system viscosity increases continuously, decreasing the probability of interaction between macromer reactive sites. Nevertheless, functional groups
close to each other still react efficiently due to a slower mobility, but longer available reaction time. This physico-
chemical condition results in a “welding” between microgels surfaces, giving rise to the final three-dimensional macro-
structure (Figure S1). The gelling system was homogenized together with Au NPs or Au-PEG NPs solution above the
sol/gel transition temperature and then cooled down to 37 °C (gelling temperature). The obtained three-dimensional
network is nanostructured and exhibits anionic nature due to the high presence of carboxylate groups and to the choice
of the solvent. In fact, PBS salts freely solvated in solution increase the presence of electrolytes within the system and
cause salt carboxylates formation. The building blocks, or subunits, of macromolecules form a stable structure mostly
made up of C–C bonds, as already discussed in previous works [2]. The chemical nature of the network was confirmed
by FT-IR analysis, as shown in Figure S2 (red line): the spectrum shows a broad peak of around 3450 cm⁻¹ which is due
to the stretching vibration of O-H bonds (labeled as 1), while peaks around 2960 cm⁻¹ are due to the C-H stretch (labeled
as 2). The formation of ester bonds is visible by peaks corresponding to symmetric (labeled as 3, around 1560 cm⁻¹) and
asymmetric (labeled as 4, around 1420 cm⁻¹) CO₂ stretches. The presence of Au NPs and the ability of the system to
entrain them, avoiding their release, was studied by placing the resulting material in excess of PBS. UV measurements
showed no release of Au NPs in the outer medium (data not shown) and FT-IR analysis revealed the presence of NPs
(Figure S2, black line). In particular, the plot regards Au-PEG NPs since Au on its own does not have relevant IR peaks.
The newly visible peaks are therefore related to the PEG capping and are: 1100 cm⁻¹ (labeled as 5, C-O-C stretching)
and 700-900 cm⁻¹ (N-H wagging).

Figure S1. Scheme of Au NPs and Au-PEG NPs inclusion in agarose-carbomer based hydrogels.

Figure S2. FT-IR of neat hydrogel (red line) and hydrogel loaded with Au-PEG NPs (black line).

As said, gelation was assessed using the inverted tube test. All the samples gelled in less than 5 min without any
noticeable differences between them. The gelation phenomenon for these systems is rapid, underlining their suitability
for biomedical applications. The ability to absorb and retain a large amount of water (swelling equilibrium ratio) is one
of the most important features of 3D polymeric networks, such as hydrogel systems, and is evaluated as swelling ratio.
At 37 °C, typical for biomedical applications, all samples exhibited fast swelling kinetics and they reached swelling equilibrium within the first 2 hours (Figure S3). No differences are visible between Au NPs loaded hydrogels (black dots) and neat ones (red dots). Moreover, Au-PEG NPs present similar values to Au NPs and dots are perfectly overlapped (not shown for image clarity).

Therefore we conclude that Au NPs loading within hydrogel 3D network do not significantly affect the physical properties of the network. Regarding the rheology (Figure S4), the storage modulus (\(G'\)) was found to be approximately one order of magnitude higher than the loss modulus (\(G''\)) in both samples, indicating an elastic rather than viscous material. Both moduli were essentially independent of frequency. In detail, \(G'\) values observed for neat hydrogels and Au-NPs hydrogel were 3750 Pa and 4200 Pa, respectively (Figure S4A). The Au-NPs loaded hydrogel presented stiffer and more elastic properties than the neat hydrogel. This behavior can be attributed to the interactions between the polymeric network and Au NPs in accordance with previous studies [3]. Nevertheless, Au NPs loading does not irreversibly change the rheological properties due to the absence of high steric hindrance.

The dynamic strain sweep tests (Figure S4B) showed that the behavior of both neat hydrogel and Au-NPs loaded hydrogel is dominated by the elastic modulus at low strain values. Increasing the strain values, the elastic structure of the network breaks down and the elastic modulus rapidly decreases. The crossover strain (\(\gamma_c\)) can be evaluated as the value at which the contribution of the material damping tan(\(\delta\)) is predominant with respect to \(G'\). At low values of strain, the two \(G'\) trends indicate the presence of a closely-packed polymeric network and the behaviors of tan(\(\delta\)) are very similar, thus indicating very similar liquid-like behavior. As before, Au-PEG NPs present similar values to Au NPs and, since dots are overlapping, data are not presented for image clarity.
4 Modelling the optical response of Au NPs

4.1 Quasi-static model for static response

As mentioned in the Methods section, the static optical behavior of the experimental samples has been modelled using a quasi-static approximation. In this framework, absorption, scattering and subsequently extinction cross-sections of the considered nanostructures can be computed starting from their polarizability, α, and exploited to determine the sample absorbance as detailed in the main text.

Regarding isolated NPs (Au-PEG NPs), we assumed for α the isotropic polarizability of a metal nanosphere of radius R and permittivity ε embedded in a homogeneous medium with permittivity ε_m (see e.g. Ref. [4]), reading α = 4πR³(ε − ε_m)/(ε + 2ε_m). For the simulation of non-PEGylated NPs assemblies, we used NP dimers. We followed the approach detailed in Ref. [5], according to which the longitudinal α_|| and transversal α_⊥ components of the polarizability tensor for a nanosphere dimer can be written as a function of the polarizability α of the individual nanospheres and their center-to-center distance d:

\[
\alpha_{||} = \frac{2\alpha + 2\alpha^2/(2\pi d^3)}{1 - \alpha^2/(2\pi d^3)^2}, \tag{8}
\]

\[
\alpha_{\perp} = \frac{2\alpha - 2\alpha^2/(4\pi d^3)}{1 - \alpha^2/(4\pi d^3)^2}. \tag{9}
\]

Moreover, to take into account the random orientation of the dimers in the ensemble of non-PEGylated NPs, in the calculation of the absorption and scattering cross-sections for the tensorial polarizability, we assumed an effective polarization of the incoming light defining an angle of 70° (fitted on the experimental absorbance of the sample) with respect to the longitudinal axis of the dimer [6].

In the simulations we set R = 10 nm for the isolated NPs and R = 7.5 nm for the radius of the nanospheres in the NP dimer whose center-to-center distance was fitted on the experimental absorbance of the sample, providing d = 11.7 nm. For the gold permittivity, ε, we assumed a Drude-Lorentz model fitted on Johnson and Christy experimental data [7], whereas for the permittivity of the environment (the HG), we assumed ε_m = 1.823 (very close to the permittivity of pure water, 1.77).

Thus, with the expression of the static polarizability of the nanostructure at hand, cross-sections upon quasi-static approximation can be straightforwardly computed and the overall sample absorbance readily determined.
4.2 Dynamical model of ultrafast photoexcitation

The ultrafast photoinduced transfer of energy among the inner energetic degrees of freedom of plasmonic NPs has been modelled according to the well-established 3TM [8, 9] introduced in the main text [Eqs. (1)-(3)].

Regarding the parameters used in our simulations, we assumed, in agreement with [6], $a = 12.9$ THz, $b = 0.33$ THz, $G = 2.2 \times 10^{16}$ W m$^{-3}$ K$^{-1}$, $\gamma = 68$ J m$^{-3}$ K$^{-2}$, $C_i = 2.5 \times 10^5$ J m$^{-3}$ K$^{-1}$ and $G_a$ was fitted on the cooling dynamics retrieved by thermal simulations (see below), providing $G_a = 2.19 \times 10^{16}$ W m$^{-3}$ K$^{-1}$.

The driving term in Eq. (2) from the main text, expressing the electromagnetic power density transferred from the ultrafast pump pulse to the metallic structure, is calculated as:

$$p_a(t) = \frac{2 \sigma_a^{eff}(\lambda_p)F}{\pi V \tau_p} \exp \left( -\frac{2t^2}{\tau_p^2} \right),$$  

where

$$
\sigma_a^{eff}(\lambda) = 1 - \exp \left[ -\sigma_a(\lambda)N_pL \right] N_pL. 
$$  

In equations above, $V$ is the volume of the NP (being either the single nanosphere or the nanospheres dimer), $F = 0.17$ mJ/cm$^2$ is the measured pump fluence, $\lambda_p = 400$ nm is the pump wavelength, and $\tau_p = 200$ fs the pump pulse half width at 1/e$^2$ power (fitted on the rising time of the pump-probe data).

Concerning instead the mechanical oscillation model [10] used to account for the dynamical expansion of NPs in the photoexcitation process, a key quantity in the modelling is $\omega_a$, the mechanical mode resonant frequency, its expression depending on the structure under investigation. Indeed, in the case of isolated nanospheres, this was estimated as $\omega_a = 2\pi v_a/(2R)$, where $v_a = 3240$ m/s is the speed of sound in gold [10]. On the other hand, for the nanosphere dimer, we followed the approach reported in Ref. [11] for the mechanical oscillation of weakly coupled nanoparticles. For the longitudinal stretching ($A_{1g}$) mode, the dimer can be modelled as a mass-spring system, with reduced mass $M_{red} = M/2$ ($M$ being the mass of the individual nanospheres) and spring elastic constant $k_{sp} = Y_{Au}A_{sp}/L_{sp}$, with $Y_{Au} = 78 \times 10^9$ GPa the Young modulus of bulk gold and $A_{sp}$ and $L_{sp}$ the cross-sectional area and length of the spring, respectively. The latter two parameters have been estimated as, respectively, the peak cross-sectional area of the interception between the two nanospheres ($A_{sp} = \pi r_{sp}^2$, with $r_{sp} = R \sin(\cos(d/(2R)))$) and the interpenetration depth ($L_{sp} = 2R - d$). The resonance frequency of the stretching mode of the dimer is accordingly estimated as $\omega_a = \eta \sqrt{k_{sp}/M_{red}}$ with $\eta$ a fitting parameter of the order of 1 (we found good agreement with the measured dynamics after taking $\eta = 2.8$).

4.3 Nonlinear model for dynamic response

In order to describe the impact of the dynamical evolution of the internal plasmonic degrees of freedom on the metallic structure optical response, the solution of the 3TM [Eqs. (1)-(3) from the main text] is the starting point to compute the corresponding Au permittivity variation in time. We followed a well-established [9] approach in which each of the plasmonic degrees of freedom leads to a complex permittivity variation $\Delta \varepsilon(\lambda, t)$, whose expressions stem from the physical processes accounted for in our model. Both the quantities $N(t)$ and $\Theta(t)$ cause a modulation in the interband transitions for gold, due to the smearing of the Fermi distribution of electrons. In turn, this change in the Fermi distribution leads to a different absorption profile of the probe beam, which is modelled by a purely imaginary permittivity variation. The relative real permittivity variation is then retrieved by Kramers-Kronig relations. The third temperature $\Theta(t)$ leads to a variation of the Drude permittivity of free electrons in gold, that follows from a linear increase of electron-phonon scattering rate with respect to the lattice temperature. These three effects combined define the total transient complex permittivity $\varepsilon_{trans}(\lambda, t) = \varepsilon(\lambda) + \Delta \varepsilon_{trans}(\lambda, t)$. Refer to [9] and references therein for further details.

Regarding instead the nonlinear and dynamical effect due to NPs expansion, described according to Ref. [10], having modelled the deformed NPs as ellipsoids, and considering the permittivity changes calculated in the 3TM, the new time dependent NP polarizability to be employed in the dimer polarizability formulas is computed as the mean of the polarizability components of the nanoellipsoids, reading [4]

$$
\alpha_i(\lambda, t) = 4\pi R_x(t)R_y(t)R_z(t) \frac{\varepsilon_{trans}(\lambda, t) - \varepsilon_m}{3L_i \varepsilon_{trans}(\lambda, t) + 3(1 - L_i)\varepsilon_m}, \quad i = x, y, z
$$  

$$
\varepsilon_{trans}(\lambda, t) = \varepsilon(\lambda) + \Delta \varepsilon_{trans}(\lambda, t).
$$
where we assumed the modified structure semi-axes $R_x(t) = R + s(t)$, $R_y(t) = R_x(t)$, and $L_s$ are geometrical factors, taking into account depolarization effects [4]. In particular, for NPs dimers, the longitudinal and transversal components of the time-dependent polarizability are updated in time with the same rationale mentioned in the steady-state regime, considering a stretched center-to-center distance increased by $2s(t)$.

This modified expression for the polarizability, both in the case of isolated NPs and NPs dimers, is then used to compute the dynamical structure cross-sections and corresponding $\Delta T/T(\lambda, t)$ as outlined in the Methods section.

### 4.4 Thermal modelling of nanostructures

By referring to the heat transfer problem solved to investigate the thermal behaviour of the sample and introduced in the Methods section of the main text [refer to Eq. (9)], in the simulations Au has been described [12, 13] by a $\rho_1 = 19320$ kg/m$^3$, $C_p = 129$ J/(kg·K), $\kappa = 317$ W/(m·K). In water instead $\rho_2 = 1000$ kg/m$^3$, $C_p = 4186$ J/(kg·K), $\kappa = 0.6$ W/(m·K) [14]. Regarding thermal sources, the heat dissipation term has been defined in Au domains and written according to the case under consideration, being the pulsed or the CW regime. In particular, considering that the rise of temperature ($\Theta$) is driven by an increase of Au electronic temperature and therefore governed by electron-phonon scattering events, to simulate the behaviour of an isolated NP upon pulsed illumination, we set $Q_{np} = P_0 V_{Au} \exp[-t/\tau_{eph}]$, where $P_0$ is the estimated power exciting the structure, $V_{Au}$ the Au volume and $\tau_{eph}$ the electron-phonon scattering time rate, which was set equal to 3 ps (thus corresponding to a full electron-phonon thermalization in less than 10 ps, in agreement with Ref. [15]). For the NxN dimer array (DA) simulations, $Q_{np}^{DA}$ has an analogous expression, where $P_0$ and $V_{Au}$ are consistently adjusted to the Au dimers geometry and a factor $2N^2$ is included to account for the total number of particles dissipating heat. On the other hand, in CW illumination conditions, heat sources have been written as $Q_{cw}^{DA} = P_{in}/V_{Au}$, with $P_{in} = 30$ W (corresponding to an incident intensity of $I_0 = P_{in}/\sigma_\lambda \approx 10^7$ W/cm$^2$ with $\sigma_\lambda$ the absorption cross-section of the individual NP) as an exemplary value for the isolated sphere, and $Q_{cw}^{DA} = 2N^2 P_{in}/V_{Au}$. Both time-dependent and stationary analyses have been solved by defining infinite elements (IEs) domains surrounding the physical domain and by defining boundary conditions so to fix the temperature beyond such IEs to its equilibrium value.

### 5 Thermal response of a Au NPs assembly

#### 5.1 Effects of the internal configuration of the assembly

In presenting the main results from the numerical simulations of the thermal behaviour of the system under both ultrafast and continuous wave excitation (refer to Fig. 3 and corresponding comments in the main text), we limited our discussion to one assembly configuration, namely a regular square 6x6 array. Evidently, such geometry does not extinguish the infinite configurational possibilities of an actual aggregated sample. An exhaustive investigation of the role of NPs arrangement within the assembly would in fact deserve a dedicated study, requiring stochastic simulations to explore several possible realizations of the assembly structure, exceeding the scope of the present work. Nevertheless, the simple 2D arrangement we considered is expected to thoroughly account for the general trend of the heat dynamics in the structures we are interested in. Thus, to prove the robustness of the results of the thermal simulations upon variations of the internal arrangement of NPs within the assembly, we performed the same numerical investigation as the one of Fig. 3, for different array geometries.

The main results of our computations are reported in Fig. S6, where data, both in time domain (upon impulsive excitation, mimicking the ultrafast pump-probe experimental conditions) and in the stationary regime, i.e. upon continuous wave excitation, are shown.
Figure S6. (A-C) Temperature dynamics, computed from simulations, modelling the thermal response of a 6x6 array of Au NP dimers upon impulsive (ultrafast pump-probe experiment-like) excitation for different arrangements of NPs within the assembly. In particular, (A) an array of dimers with disordered displacement with respect to a square lattice, (B) a square lattice of dimers with alternatively rotated axis, and (C) a regular configuration of two interleaved arrays of spheres (refer to each panel insets). Red and green curves are the temperature increase in two exemplary points of the domain (highlighted with same colours in the insets), whereas the blue curve is the average temperature increase within the surrounding water domain (10 times magnified for clarity). (D) Temperature increase in the same spots and for the same internal arrangements as (A)-(C), upon continuous wave excitation for an exemplary input laser intensity $I_0 = 10^5$ W/cm$^2$.

In particular, three different realizations of the internal arrangement of NPs for the 6x6 assembly discussed in Fig. 3F of the main text have been studied: (i) a 6x6 array of aligned dimers with disordered displacement with respect to the square lattice positions; (ii) a regular square lattice of dimers with alternatively rotated axis; (iii) a regular configuration of two interleaved arrays of NPs, built by disjointing dimers within the array. The dynamics of the temperature increase across the structure under a pulse regime of excitation (Figs. S6A-6C) as well as the variation in temperature in the CW thermal loading regime (Fig. S6D) are shown.

Results of numerical simulations clearly show the marginal role of the internal configuration of NPs in the assembly. Indeed, at least for the 2D closely packed arrangement under consideration and by considering a uniform thermal loading (which is reasonable at the pump wavelength considered in our experiments, far enough from resonant features in the structure absorbance spectrum), the thermal response of the system turns out to be essentially insensitive to a change in the internal configuration of the assembly. Rather, the key factor which is expected to dominate the thermal behavior of the system is the degree of assembling (as further discussed below), which does not vary in the three configurations here detailed.

5.2 Effect of the degree of assembling

While, as proven above, the thermal response of a NPs assembly should only slightly be affected by their internal arrangement, a major role may be played by the degree of nanoassembling. To explore the cumulative heating effect induced in a closely packed arrangement, we performed numerical computations in the continuous wave excitation regime. Indeed, as outlined in the main text, superior nanoheating performances from nanoassemblies compared to isolated NPs require excitations with pulse duration longer than the characteristic time of heat diffusion across the entire assembly ($\sim \mu$s). For this reason, an investigation in the ultrafast regime would provide comparable results for differently degrees of assembling.

The main results of the thermal simulations are shown in Fig. S7, where the temperature increase in the CW heat loading regime is compared in the case of two structures: (i) a sparse regular 5x5 array of NPs dimers surrounded by aqueous environment and well separated to each other (centre-to-centre distance equal to 125 nm, Fig. S7A); (ii) the same 5x5 square array of dimers, but assembled in a closely-packed arrangement (centre-to-centre distance five times smaller than the previous one, Fig. S7B). The latter configuration, more ‘condensed’ than the former, is the most similar to the one considered in the simulations presented in Fig. 3 and Fig. S6 in terms of degree of assembling. For a fair comparison, the number of NPs (twice 25, i.e. the number of dimers) and the surrounding water volume have been considered constant in comparing the two configurations.
Figure S7. (A) Numerical domain used for simulations, consisting of a regular 5x5 array (referred to as sparse) of Au NPs dimers embedded in water. The centre-to-centre distance between dimers (i.e. the lattice constant) is set to 125 nm. (B) Same as (A), for a centre-to-centre distance equal to 25 nm, five times smaller. Both the total number of dimers in the condensed array and the volume of surrounding water have been kept constant with respect to the sparse configuration. (C) Temperature increase of the two analysed configurations upon continuous wave excitation for an exemplary input laser intensity $I_0 = 10^5$ W/cm$^2$. Red and green traces refer to the temperature increase evaluated in the spots highlighted in the insets of (A) and (B), respectively, whereas the blue curve is the average temperature increase in the water volume.

Results of the predicted temperature increase in the two cases (Fig. S7C) show in a clear-cut manner to what extent a condensed array, corresponding to a higher degree of assembling, is able to outperform a sparse configuration. Indeed, while average temperature in water (blue curve) is marginally affected by the assembly structure under the same thermal loading of the individual NPs (which has been considered the same in the two cases), temperature within the metallic dimer (red curve) and close to the external edge of the assembly (in green) experience a dramatic increase in the closely packed assembly if compared to the sparse array. Our results, in accord with Refs. [16, 17], point in favor of the significant contribution in the heating process related to cumulative effects induced by the nanoassembling.

Bibliography

