

Noble Metals on the Nanoscale: Optical and Photothermal Properties and Some Applications in Imaging, Sensing, Biology, and Medicine

PRASHANT K. JAIN, XIAOHUA HUANG, IVAN H. EL-SAYED,
AND MOSTAFA A. EL-SAYED*

[†]Laser Dynamics Laboratory, School of Chemistry and Biochemistry,
Georgia Institute of Technology, Atlanta, Georgia 30332, and

[‡]Department of Otolaryngology-Head and Neck Surgery, Comprehensive
Cancer Center, University of California at San Francisco,
San Francisco, California 94143

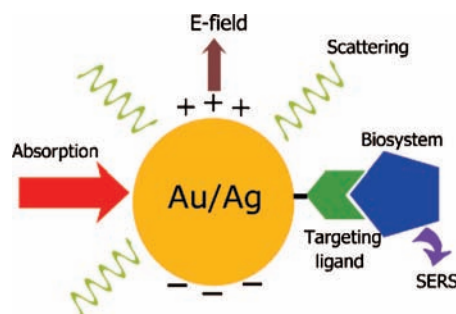
RECEIVED ON DECEMBER 17, 2007

CONSPECTUS

Noble metal nanostructures attract much interest because of their unique properties, including large optical field enhancements resulting in the strong scattering and absorption of light. The enhancement in the optical and photothermal properties of noble metal nanoparticles arises from resonant oscillation of their free electrons in the presence of light, also known as localized surface plasmon resonance (LSPR). The plasmon resonance can either radiate light (Mie scattering), a process that finds great utility in optical and imaging fields, or be rapidly converted to heat (absorption); the latter mechanism of dissipation has opened up applications in several new areas.

The ability to integrate metal nanoparticles into biological systems has had greatest impact in biology and biomedicine. In this Account, we discuss the plasmonic properties of gold and silver nanostructures and present examples of how they are being utilized for biodiagnostics, biophysical studies, and medical therapy. For instance, taking advantage of the strong LSPR scattering of gold nanoparticles conjugated with specific targeting molecules allows the molecule-specific imaging and diagnosis of diseases such as cancer. We emphasize in particular how the unique tunability of the plasmon resonance properties of metal nanoparticles through variation of their size, shape, composition, and medium allows chemists to design nanostructures geared for specific bio-applications. We discuss some interesting nanostructure geometries, including nanorods, nanoshells, and nanoparticle pairs, that exhibit dramatically enhanced and tunable plasmon resonances, making them highly suitable for bio-applications. Tuning the nanostructure shape (e.g., nanoprisms, nanorods, or nanoshells) is another means of enhancing the sensitivity of the LSPR to the nanoparticle environment and, thereby, designing effective biosensing agents. Metal nanoparticle pairs or assemblies display distance-dependent plasmon resonances as a result of field coupling. A universal scaling model, relating the plasmon resonance frequency to the interparticle distance in terms of the particle size, becomes potentially useful for measuring nanoscale distances (and their changes) in biological systems. The strong plasmon absorption and photothermal conversion of gold nanoparticles has been exploited in cancer therapy through the selective localized photothermal heating of cancer cells. For nanorods or nanoshells, the LSPR can be tuned to the near-infrared region, making it possible to perform *in vivo* imaging and therapy.

The examples of the applications of noble metal nanostructures provided herein can be readily generalized to other areas of biology and medicine because plasmonic nanomaterials exhibit great range, versatility, and systematic tunability of their optical attributes.



Introduction

The fields that have recently been greatly impacted by the advancement in nanostructured materials¹ are biology, biophysics, and medicine. The nanobiology toolkit has been greatly enhanced by noble metal

nanostructures,² which have proven to be highly versatile and tunable materials for a range of bioapplications including biophysical studies,^{3,4} biological sensing,^{5,6} imaging,⁷⁻⁹ medical diagnostics,¹⁰ and cancer therapy.¹¹⁻¹⁷ In this Account, we describe the inter-

esting optical properties of noble metal nanostructures and discuss recent research advances in their bioapplications.

Plasmon Resonance in Noble Metal Nanostructures.

The interesting optical attributes of metal nanoparticles, as reflected in their bright intense colors, are due to their unique interaction with light. In the presence of the oscillating electromagnetic field of the light, the free electrons of the metal nanoparticle undergo a collective coherent oscillation with respect to the positive metallic lattice.^{18–21} This process is resonant at a particular frequency of the light and is termed the localized surface plasmon resonance (LSPR) oscillation. This electronic oscillation can be simply visualized as a photon confined to the small size of the nanostructure, constituting an intense electric field around the particle. The surface plasmon oscillation decays by radiating its energy resulting in light scattering or decays nonradiatively as a result of conversion of absorbed light to heat.²² The electric field intensity and the scattering and absorption cross-sections are all strongly enhanced at the LSPR frequency,²³ which for gold, silver, and copper lies in the visible region.^{20,21} Since copper is easily oxidized, gold and silver nanostructures are most attractive for optical applications.

Nanosized dimensions of the nanoparticle probes make it easy to incorporate them into biological systems, which are on the same size scale.⁹ Metal nanoparticles can be conjugated with small molecule or biomolecular targeting or recognition ligands²⁴ for achieving molecular specificity. While the use of photoabsorbing and fluorescent dyes as biological labels and stains has been common over the last few decades, metal nanoparticles are fast replacing them as optical labels and probes.²⁵ Because of surface plasmon enhancement, optical cross-sections of metal nanoparticles (10–100 nm) are 5 orders of magnitude or more larger than those of dye molecules.¹⁸ Each metal nanoparticle can be considered an optical probe equivalent to up to a million dye molecules. This provides a large margin for enhancing the probing sensitivity. Unlike dyes, metal nanoparticles are photostable and do not undergo photobleaching, allowing higher light excitation energies and longer probing times.⁴ There is a range of enhanced radiative and nonradiative attributes associated with the LSPR. The optical probing strategy can thus be chosen depending on the specific biological application. Different strategies may also be combined. Another unique property of LSPR is that it can be tuned by changing the nanostructure size, shape, composition, or environment,^{18,19,26,27} (Figure 1) in order to suit the bioapplication.

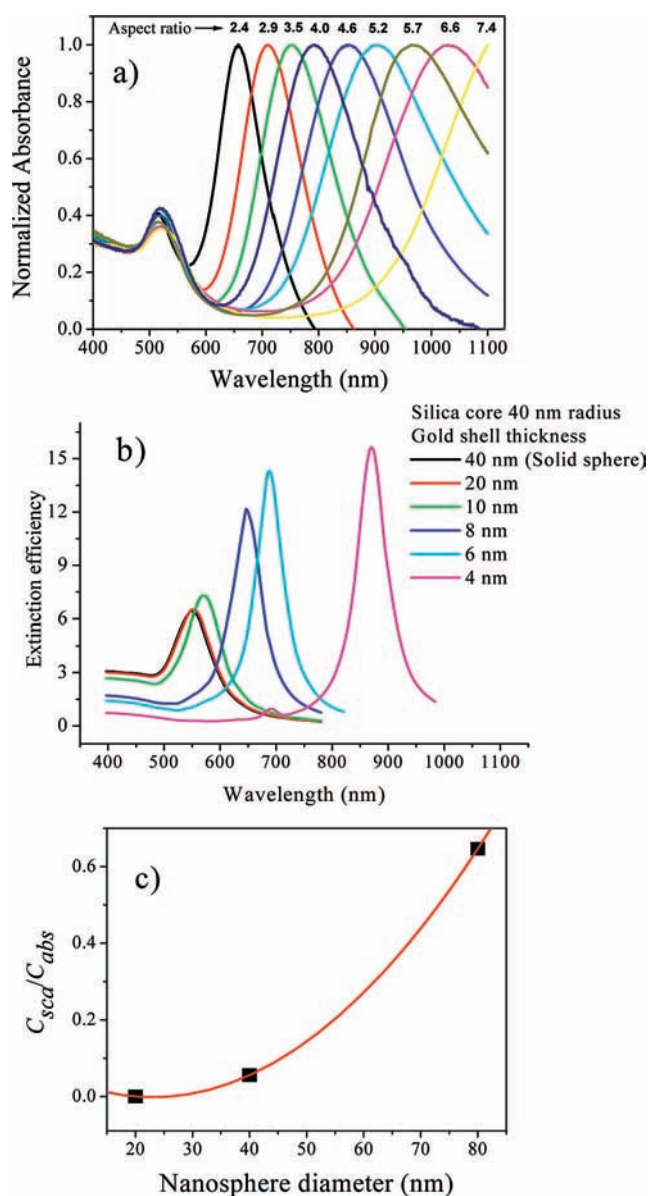


FIGURE 1. Size, shape, and composition tunability of the plasmon resonance of noble metal nanostructures. (a) Tuning the LSPR frequency of the gold nanorod long-axis mode by synthetically controlling aspect ratio. Reprinted with permission from ref 12. Copyright 2006 American Chemical Society. (b) Silica core–gold nanoshells show plasmon resonance frequency tunable from the visible to the NIR by changing the shell thickness relative to the core size. Reprinted with permission from ref 34. Copyright 2007 American Chemical Society. (c) Increase in the plasmon scattering to absorption ratio by increase in particle volume in gold nanospheres. Reprinted with permission from ref 18. Copyright 2006 American Chemical Society.

Tunable Radiative Properties

Gold nanospheres in the 10-nm size range have a strong absorption maximum around 520 nm in water due to their LSPR. This occurs around 390 nm for silver nanospheres.¹⁹ With increase in the nanosphere size, there is some LSPR red

shift due to electromagnetic retardation in larger particles.¹⁹ For example, LSPR for 40-nm gold nanospheres is at ~ 530 nm.²⁸ However, size tunability of the nanosphere LSPR is quite limited. In many biological applications, especially *in vivo* studies, it is desirable to work in the near-infrared (NIR) region of the spectrum, especially 650–900 nm, due to the high transmission of tissue, blood, and water in this window.²⁹ Alternatively, one may need to tune the LSPR depending on the availability of a suitable laser or to resonantly enhance an optical process. One way of tuning the LSPR is by changing the particle shape from spherical to rod-shaped. Rod-shaped nanoparticles have two resonances: one due to plasmon oscillation along the nanorod short axis and another due to plasmon oscillation along the long axis,^{30,31} which depends strongly on the nanorod aspect ratio, that is, length-to-width ratio. When the nanorod aspect ratio is increased, the long-axis LSPR wavelength position red-shifts from the visible to the NIR and also progressively increases in oscillator strength.³¹ Gold nanorods of controlled aspect ratio from 2 to 7 (Figure 1a)¹² can be easily made by the seeded growth method.^{32,33}

The second method of LSPR tuning is by using a metal nanoshell.²⁷ Starting from a solid spherical particle, as we decrease shell thickness, the LSPR red-shifts from the visible to the NIR (Figure 1b)^{27,34} due to increased coupling between the inner and outer shell surface plasmons.^{34,35} We recently showed that the LSPR frequency decreases near-exponentially with decrease in the shell thickness-to-core radius ratio,³⁴ with a trend that is universal and independent of the nanoshell size, core material, shell metal, or surrounding medium, thus making it easy to design nanoshells with a desired optical resonance. The Halas group first realized the nanoshell structure via formation of a thin gold shell on the surface of silica nanoparticles.²⁷ Hollow nanocages and nanocubes synthesized by galvanic replacement of silver nanoparticles by gold atoms show similar visible–NIR tunability by control of the wall thickness and void size of the particle.^{36,37}

Biological Imaging. Due to the strongly enhanced LSPR, noble metal nanoparticles scatter light very strongly at the LSPR frequency, making them very promising for optical imaging and labeling of biological systems.^{7,8} While scattering and absorption are competing processes, the relative contribution of scattering increases rapidly with increase in the nanostructure volume (Figure 1c).¹⁸ While the scattering from a 10-nm gold nanoparticle is negligible, an 80-nm gold nanoparticle offers scattering 5 orders of magnitude larger than the typical emission from a dye.¹⁸ Such highly enhanced cross-sections offer sensitive and highly contrasted imaging^{7,38}

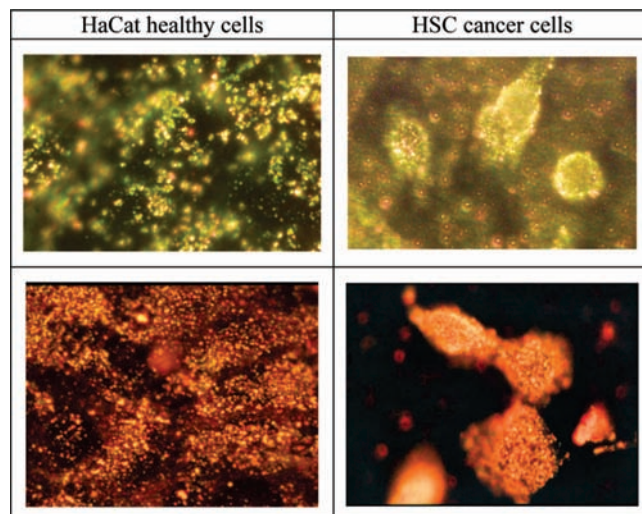


FIGURE 2. Molecular-specific imaging of cancer using gold nanoparticle/anti-EGFR conjugates. Dark-field microscopy shows (right) HSC cancer cells clearly defined by the strong LSPR scattering from (top) gold nanospheres and (bottom) gold nanorods bound specifically to the cancer cell surface, whereas (left) HaCat healthy cells have (top) gold nanospheres and (bottom) gold nanorods randomly dispersed without specific binding. The scattering color (LSPR frequency) of the nanospheres and nanorods can be clearly distinguished. Reprinted with permission from ref 8, Copyright 2005 American Chemical Society, and ref 12, Copyright 2006 American Chemical Society.

allowing use of the much simpler but powerful dark-field microscopy.⁸

While most imaging techniques require sophisticated and expensive lasers, optical components, and detectors and complex image processing, dark-field imaging^{8,25,39} using gold nanoparticles requires a simple optical microscope equipped with a dark field condenser. The latter ensures that the excitation light is incident at high angles such that only light scattered by the sample is collected by the microscope objective. The gold nanoparticles are excited by a broad white-light source, but only light frequencies corresponding to the LSPR are strongly scattered. The nanoparticles are seen as bright spots with a color corresponding to the LSPR frequency on a dark background. In fact, due to the high scattering cross-section, individual nanoparticles can also be imaged.³⁹ The dark-field microscopy technique can be utilized very effectively for the molecular-specific imaging of biomolecules by integrating the gold nanoparticles with specific targeting molecules. As an example, El-Sayed et al. diagnosed cancer (Figure 2)⁸ by imaging the cancer biomarker epidermal growth factor receptor (EGFR), present in significantly higher amounts on cancer cells.⁷ Gold nanospheres conjugated to anti-EGFR antibodies specifically target the cancer cells as shown by the dark-field imaging. The cancer cell surface was defined by strong LSPR

scattering from gold nanoparticles bound specifically to the EGFR on the cancer cells. Thus, cancer cells could be easily identified from the healthy cells, in which case the gold nanoparticles were dispersed randomly due to nonspecific binding. This diagnostic approach is quite general since gold nanoparticles can be conjugated to a range of proteins, antibodies, and small molecules.²⁴ The targeting ligands can be chosen depending on the disease biomarkers to be targeted. Dark-field imaging is also possible in the NIR region by the use of gold nanorods (Figure 2).¹²

Optical Sensing. An interesting attribute of the surface plasmon oscillation is that its resonance frequency depends on the dielectric constant, that is, refractive index (RI), of the medium surrounding the nanoparticle.^{6,40,41} With increase in medium RI, the LSPR red-shifts. When this shift is followed using absorption spectroscopy (for colloidal nanoparticles) or scattering spectroscopy (for nanoparticles deposited on a substrate), changes in the nanoparticle environment can be sensed. In order to sense chemical/biological species, the nanoparticles are conjugated with recognition molecules, which specifically bind the target analyte, while appropriate surface capping is required to minimize nonspecific binding. The binding of the target molecule to the recognition molecules causes a plasmon band shift due to a local RI change, serving as an optical sensing tool.⁶ To achieve sensitivities down to few molecules bound per nanoparticle, the plasmon band shift is desired to be as high as possible in response to small RI changes. Nanostructure geometry can be readily tuned to enhance plasmonic sensitivity, defined as the plasmon shift per refractive index unit (RIU) change.^{40,42,43} Gold nanospheres of 30-nm size offer a modest sensitivity of ~ 70 nm/RIU.⁴³ When the particle volume is increased, sensitivity increases by a limited extent, possibly due to increase in radiative damping and retardation.⁴³ However, more molecules would be required to effectively change the RI around a larger nanoparticle. In addition, the plasmon band broadens due to damping and retardation, reducing the sensitivity of determining band shifts.

Nanoparticle shape offers a handle for tuning sensitivity without changing particle volume. Interestingly, nanostructure geometries that offer plasmon resonance tunability also offer high plasmon sensitivity,⁴³ for example, sharp surface curvatures and tips (nanoprisms⁶ or nanorods⁴⁰) and junctions (metal nanoshells⁴³ or particle dimers⁴⁴). Silver nanoprisms (100-nm width, 50-nm height), due to their high sensitivity (200 nm/RIU),⁴⁵ have been used to sense streptavidin⁶ and Alzheimer's disease markers⁴⁶ and have also been used in single-particle assays.⁴⁷

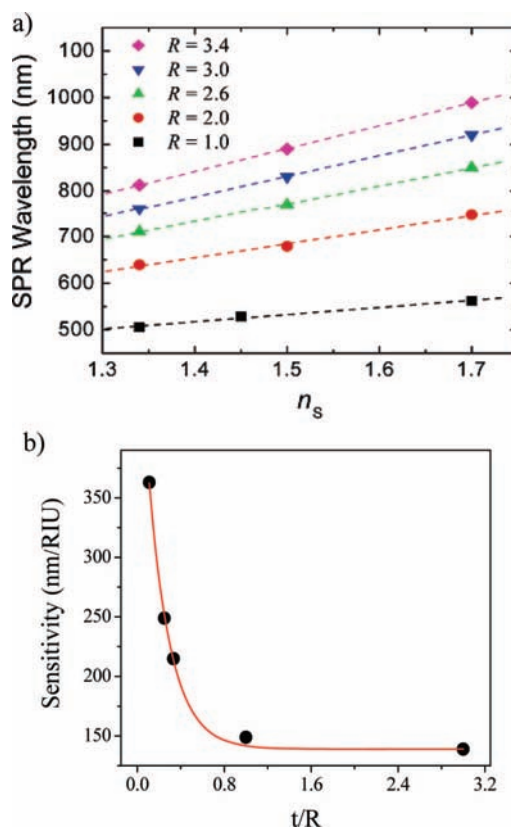


FIGURE 3. Enhancing the sensitivity of plasmonic nanostructures: (a) By increasing the gold nanorod aspect ratio, sensitivity of long-axis LSPR to medium RI changes can be increased. Reprinted with permission from ref 40, Copyright 2006 American Chemical Society. (b) The plasmon sensitivity of silica core–gold nanoshells can be increased near-exponentially by decreasing nanoshell thickness (t) relative to the core radius (R). Reprinted with permission from ref 43, Copyright 2007 American Chemical Society.

Gold nanorods are also highly suited for plasmon sensing.⁴⁰ The nanorod longitudinal LSPR has considerably higher sensitivity compared with the nanosphere resonance, increasing almost quadratically (Figure 3a) with increase in aspect ratio (i.e., surface curvature).⁴⁰ The long-axis of gold nanorods of aspect ratio 3 shows 6 times higher sensitivity compared with nanospheres.⁴⁸ Yu et al. have utilized colloidal gold nanorods functionalized at the ends with Fab segments of IgG for effective detection of anti-IgG.⁴⁹ The shape tunability of the plasmon resonance also becomes useful for simultaneous multiplexed sensing of different biomolecules in the same solution by using a mixture of nanorods of significantly different aspect ratio possessing well-separated resonance maxima.⁴⁹

The metal nanoshell also offers enhanced sensitivity. With decreasing shell thickness (relative to core size), there is near-exponential increase in the plasmon sensitivity, for example, from 129 to 363 nm/RIU with variation in shell thickness from 40 nm (sphere) to 4 nm (Figure 3b).⁴³ In thinner shells, plas-

monic coupling is stronger and the sensitivity of the junction or hybridized system is higher. Furthermore, we have seen, in agreement with the Xia group, that hollow gold nanoshells, which offer two surfaces where the medium RI change is experienced, show even higher sensitivity enhancement, 6 times that of a nanosphere.^{43,50}

Another type of plasmonic sensing takes advantage of the LSPR dependence on the proximity of other nanoparticles.^{5,51} When nanospheres assemble, the LSPR red-shifts due to favorable coupling between the plasmon oscillations/fields of the interacting particles.^{5,52} The red shift depends on the interparticle distances (particle density) in the assembly. By using appropriately functionalized particles that assemble in the presence of the bioanalyte, the assembly induced shift or color change has enabled sensing of DNA, antibody–antigen interactions, and disease biomarkers.¹⁰

Plasmon Ruler for Probing Biological Distances. Since the coupling-induced red shift depends on the distance between the interacting particles,^{44,53} a metal particle pair becomes very attractive as a distance probe similar in concept to the fluorescence resonance energy transfer (FRET)-based distance probes.⁵⁴ FRET probing involves a pair of dyes, in which energy is transferred from the excited donor to the acceptor, the efficiency of which depends on the interdy distance as per Forster theory. While FRET has become a valuable tool in probing distances and dynamic distance changes in biomolecules, there is increasing interest in metal particle pair rulers due to their stronger optical signals and lack of photobleaching.³⁹ Sönnichsen et al. first demonstrated the plasmon ruler concept,³⁹ by showing that the formation of a gold nanoparticle dimer via streptavidin–biotin binding resulted in a plasmon shift of 23 nm, observed by scattering spectroscopy of single particle pairs. The dimerization is clearly identified by strong increase in scattering and change in the color of the scattered light. Particle pair plasmon rulers linked by ss-DNA have also shown to be able to detect DNA hybridization,³⁹ DNA bending, and cleavage³ by means of distance change-induced plasmon shift or scattering change.

The distance dependence of plasmon coupling has been systematically calibrated. Spectroscopy of electron-beam lithography-fabricated gold nanodisc pairs in conjunction with electrodynamic simulations has shown that the fractional plasmon red shift ($\Delta\lambda/\lambda_0$) falls as a function of interparticle gap s scaled by the particle size D with a near-exponential decay rate ($\tau \sim 0.2$) universally independent of nanoparticle size, shape, metal, or medium.⁴⁴ We present a simulated “plasmon ruler equation” based on the universal scaling behavior:

TABLE 1. Application of Universal Scaling Model to the Plasmon Ruler^a

DNA spacer length (base pairs)	observed $\Delta\lambda/\lambda_0$ (%)	experimental separation (nm)	calculated separation from equation (nm)
250	0.0015	93	91
110	0.16	45	46
67	0.66	31	32
40	1.62	22	23
20	3.14	15	17
10	4.38	11	14

^a Comparison of experimental interparticle separations of gold nanoparticle pairs linked by DNA spacers (Reinhard et al.)⁵⁵ with those calculated using our “plasmon ruler equation”.⁴⁴ Reprinted with permission from ref 44, Copyright 2007 American Chemical Society.

$$\Delta\lambda/\lambda_0 \approx 0.18 \exp(-s/0.23D) \quad (1)$$

which gives a measure of the interparticle separation from an observed plasmon shift (Table 1), in good agreement with the experiments of Reinhard et al. on gold nanoparticle plasmon rulers assembled by DNA linkers of different lengths.⁵⁵ This makes the equation useful in the determination of intersite distances in biological systems using the plasmon ruler.⁴⁴ The universal scaling model becomes a quantitative guide for the design of plasmon rulers. While the FRET ruler has a maximum range of 10 nm due to the steep $1/R^6$ dependence between two molecular species, the plasmon coupling distance range is much larger, that is, 70 nm for a dimer of 40-nm gold nanoparticles,⁴ and can be increased directly by increasing the particle dimension.⁴⁴

Molecular Diagnostics using Plasmon-Enhanced Raman Spectroscopy. High polarizable nanostructures such as nanorods^{56,57} and nanoshells⁵⁸ support strong electric fields, which have the ability to strongly enhance (10^5 – 10^6 times)⁵⁹ the Raman scattering of molecules in the vicinity of the nanostructure, resulting in surface-enhanced Raman scattering (SERS). In addition to shape, another enhancement in SERS occurs from assembly of nanoparticles since interparticle junctions support intense fields due to favorable plasmon coupling.^{60,61} Our group recently utilized this effect for the molecular diagnosis of cancer (Figure 4), by showing that gold nanorods conjugated with anti-EGFR antibodies assembled onto the surface of cancer cells due to specific binding, resulting in the observation of highly enhanced, sharp, and polarized SERS of the nanorod capping and bridging molecules, anti-EGFR, and EGFR receptors or other molecules on the cell surface.⁵⁷ The cancer cells were clearly distinguished from normal cells, which showed weak or no SERS.

Nonradiative Properties

In addition to giving rise to light scattering, the plasmon oscillations relax nonradiatively via collisions of electron–electron

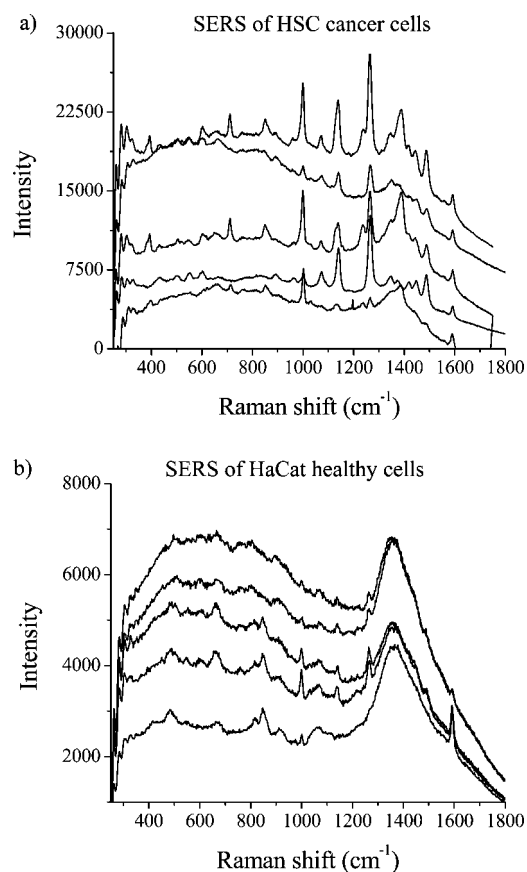


FIGURE 4. Molecular diagnostics of cancer using gold nanorod SERS.⁵⁷ (a) HSC cancer cells bind and assemble the gold nanorods conjugated with anti-EGFR antibodies and as a result exhibit enhanced, sharp, and polarized SERS spectra; (b) HaCat healthy cells, which do not specifically bind the gold nanorod bioconjugates, do not show well-resolved SERS spectra. Reprinted with permission from ref 57. Copyright 2007 American Chemical Society.

(intraband- or interband-type excitations), electron–lattice phonon, and electron–surface types, leading to absorption of light by the nanoparticle.^{21,26,62} Ultrafast laser spectroscopic studies at low laser excitation energies^{21,26,62} have established that the photoexcitation of the metal nanoparticle free electrons is followed by their cooling back to equilibrium by energy exchange with lattice phonons at the rate of ~ 1 ps for gold, heating up the nanoparticle. At very fast rates of energy deposition relative to lattice cooling, the photothermal heating can result in the ablation of the nanoparticle⁶³ (useful in nanomotors⁶⁴) or in desorption of surface-capping,⁶⁵ or it may melt or reshape the nanoparticle (~ 30 ps).^{66,67} At slower rates, the lattice cools via phonon–phonon processes (~ 100 ps) leading to heating of the medium surrounding the nanoparticle, which can be used for photothermal cancer therapy.^{11–16}

Photothermal Cancer Therapy. Metal nanoparticles serve as “light-activated nanoscopic heaters” useful for biomedicine, especially the selective laser photothermolysis of

cancer cells.^{11–16} Gold nanoparticles conjugated to antibodies can be selectively targeted to cancer cells without significant binding to healthy cells.^{13,68} Irradiation of the cancer cells selectively labeled with the nanoparticles with a laser of frequency overlapping with the LSPR absorption maximum of the nanoparticles results in selective heating and destruction of cancer cells at much lower laser powers than those required to destroy healthy cells to which nanoparticles do not bind specifically. Gold nanoparticles (10–50 nm) offer 5 or more orders of magnitude larger absorption coefficients compared with conventional dyes; thus much lower laser energies can be used to achieve cell destruction, making therapy minimally invasive.^{13,68} Further, the therapy can be combined with dark-field imaging, which is a significant advantage over other nanostructures such as quantum dots or carbon nanotubes, which can achieve only one of the two, imaging or therapy.

While the use of visible light resonant gold nanospheres can be useful for external skin/surface cancer treatments, for tumors within bodily tissue, it becomes necessary to use NIR light in the biological window. One method demonstrated by the El-Sayed group involves the use of gold nanorods of aspect ratio 3.9, which have longitudinal LSPR around 800 nm overlapping with a NIR Ti:sapphire laser.¹² NIR laser irradiation of cancer cells labeled with gold nanorod/anti-EGFR conjugates resulted in selective destruction of the cancer cells. NIR imaging and therapy have also been achieved with NIR-resonant gold nanoshells^{11,15} and nanocages.³⁷ The assembly of metal nanoparticle clusters on cells also offers NIR resonance due to the coupling-induced red shift, which is useful for photothermal therapy.⁶⁹ Recently, our group demonstrated a novel way to achieve NIR therapy using gold nanospheres by utilizing their two-photon absorption of 800-nm laser light.⁷⁰

The approach of using noble metal nanoparticles for selective targeting, molecular imaging, and selective therapy is general and versatile. Cancer is only one example. The use of plasmonic nanoparticles can be extended to other biological applications, for example, destruction of viruses or bacteria⁷¹ or controlled localized denaturation or cleavage of proteins and nucleic acids, potentially useful for diagnostic or therapeutic goals. Chemists have now established excellent control over the surface chemistry, biofunctionalization, and optical properties of metal nanoparticles aimed at targeting desired biological systems.

M.A.E. would like to thank S. Link, B. Nikoobakht, S. Eustis, K. S. Lee, W. Qian, W. Huang, C. Tabor, and several other members of his group that greatly contributed to the development of the nanoscience and nanotechnology program since mov-

ing to Georgia Tech. We thank the Department of Energy, Grant DE-FG02-97 ER 14799, for funding.

BIOGRAPHICAL INFORMATION

Prashant K. Jain, currently a postdoctoral fellow at Harvard University, received his B. Tech from UICT, Bombay, India, in 2003 and Ph.D from Georgia Tech in 2008. Prashant performed his doctoral research under Prof. Mostafa El-Sayed on radiative, nonradiative, and near-field coupling properties of plasmonic nanostructures, with implications for biomedicine and nanophotonics. He has published 15 papers and served as a reviewer for several nanoscience and physical chemistry journals. His academic honors include UICT Merit prizes, study-abroad fellowships from Lotus Trust and University of Mumbai, election to Sigma Xi full membership, American Chemical Society PHYS Division Outstanding Graduate Poster Award in 2006, best Chemistry graduate student award at Georgia Tech in 2006, recognition from College of Sciences in 2007, Materials Research Society Graduate Student Gold Award in 2007, Atlanta Area Chemical Physics Award in 2008, and Miller Fellowship of UC Berkeley for 2008–2011.

Xiaohua Huang was born in Chongqing, P.R. China. She received her B.S. from Jilin University in 1996, M.S. in Physical Chemistry from Peking University in 2001, and Ph.D. in Analytical Chemistry from Georgia Tech in 2006. Dr. Huang performed her doctoral research under Prof. Mostafa El-Sayed on cancer targeting, imaging, diagnosis, and photothermal therapy using gold nanoparticles. She is currently a postdoctoral fellow in El-Sayed's group. She has published 13 papers in the field, including 2 invited review articles. She received the AACR-Women in Cancer Research Brigid G. Leventhal Scholar Award in Cancer Research in 2007.

Ivan H. El-Sayed is a head and neck surgeon and assistant professor in otolaryngology at UC San Francisco School of Medicine. He earned a medical degree at Boston University School of Medicine in 1996 and completed a residency in otolaryngology there. He completed a fellowship at UC San Francisco in head and neck oncology. El-Sayed is certified by the American Board of Otolaryngology and is a Fellow of the American College of Surgeons. He specializes in the treatment of benign and malignant lesions of the head and neck, paranasal sinuses, and skull base. His clinical research interests include application of endoscopic technology toward skull base surgery. He is also investigating potential diagnostic and therapeutic use of lasers and nanoparticles in the management of cancer.

Mostafa A. El-Sayed received his B.Sc. in Egypt and his Ph.D. from Florida State University working with Michael Kasha. After postdoctoral work at Yale, Harvard, and Caltech, he joined UCLA in 1961. In 1994, he became Julius Brown Chair and Director of the Laser Dynamics Lab at Georgia Tech, and later Regents Professor. His research interests include the use of steady-state and ultrafast laser spectroscopy to understand relaxation, transport, and conversion of energy in molecules, solids, photosynthetic systems, and nanostructures. The cur-

rent focus of his lab is on the properties of semiconductor and metal nanoparticles and their applications in nanocatalysis, nanophotonics, and nanomedicine. El-Sayed has over 500 refereed publications in these areas. He served as the Chief Editor of the *Journal of Physical Chemistry* from 1980–2004 and U.S. editor of the *International Reviews in Physical Chemistry*. El-Sayed is an elected member of the National Academy of Science and Fellow of the American Academy of Arts and Sciences (AAAS) and APS. He has received several national awards including the Fresenius, Tolman, Richard's medal, numerous ACS local section awards, and the ACS-APS Langmuir Award in Chemical Physics. He also received the 1990 King Faisal International Prize in Science, an Honorary Ph.D. from the Hebrew University, and the Georgia Tech "Class of 1943" Distinguished Professor Award. He was a Miller Visiting Professor at UC Berkeley during Spring 2007.

FOOTNOTES

*Corresponding author. E-mail: melsayed@gatech.edu. Tel: 404-894-0292. Fax: 404-894-0294.

REFERENCES

- Burda, C.; Chen, X.; Narayanan, R.; El-Sayed, M. A. Chemistry and Properties of Nanocrystals of Different Shapes. *Chem. Rev.* **2005**, *105*, 1025–1102.
- El-Sayed, M. A. Some Interesting Properties of Metals Confined in Time and Nanometer Space of Different Shapes. *Acc. Chem. Res.* **2001**, *34*, 257–264.
- Reinhard, B.; Sheikholeslami, S.; Mastroianni, A.; Alivisatos, A. P.; Liphardt, J. Use of Plasmon Coupling to Reveal the Dynamics of DNA Bending and Cleavage by Single EcoRV Restriction Enzymes. *Proc. Natl. Acad. Sci. U.S.A.* **2007**, *104*, 2667–2672.
- Sönnichsen, C.; Reinhard, B. M.; Liphardt, J.; Alivisatos, A. P. A Molecular Ruler Based on Plasmon Coupling of Single Gold and Silver Nanoparticles. *Nat. Biotechnol.* **2005**, *23*, 741–745.
- Elghanian, R.; Storhoff, J. J.; Mucic, R. C.; Letsinger, R. L.; Mirkin, C. A. Selective Colorimetric Detection of Polynucleotides Based on the Distance-Dependent Optical Properties of Gold Nanoparticles. *Science* **1997**, *277*, 1078–1080.
- Haes, A. J.; Van Duyne, R. P. A Nanoscale Optical Biosensor: Sensitivity and Selectivity of an Approach Based on the Localized Surface Plasmon Resonance Spectroscopy of Triangular Silver Nanoparticles. *J. Am. Chem. Soc.* **2002**, *124*, 10596–10604.
- Sokolov, K.; Follen, M.; Aaron, J.; Pavlova, I.; Malpica, A.; Lotan, R.; Richards-Kortum, R. Real-Time Vital Optical Imaging of Precancer Using Anti-Epidermal Growth Factor Receptor Antibodies Conjugated to Gold Nanoparticles. *Cancer Res.* **2003**, *63*, 1999–2004.
- El-Sayed, I. H.; Huang, X.; El-Sayed, M. A. Surface Plasmon Resonance Scattering and Absorption of Anti-EGFR Antibody Conjugated Gold Nanoparticles in Cancer Diagnostics: Applications in Oral Cancer. *Nano Lett.* **2005**, *5*, 829–834.
- Alivisatos, A. P. The Use of Nanocrystals in Biological Detection. *Nat. Biotechnol.* **2004**, *22*, 47–52.
- Rosi, N. L.; Mirkin, C. A. Nanostructures in Biodiagnostics. *Chem. Rev.* **2005**, *105*, 1547–1562.
- Hirsch, L. R.; Stafford, R. J.; Bankson, J. A.; Sershen, S. R.; Rivera, B.; Price, R. E.; Hazle, J. D.; Halas, N. J.; West, J. L. Nanoshell-Assisted Tumor Ablation Using near Infrared Light under Magnetic Resonance Guidance. *Proc. Natl. Acad. Sci. U.S.A.* **2003**, *100*, 13549–13554.
- Huang, X.; El-Sayed, I. H.; Qian, W.; El-Sayed, M. A. Cancer Cell Imaging and Photothermal Therapy in the Near-Infrared Region by Using Gold Nanorods. *J. Am. Chem. Soc.* **2006**, *128*, 2115–2120.
- El-Sayed, I. H.; Huang, X.; El-Sayed, M. A. Selective Laser Photo-Thermal Therapy of Epithelial Carcinoma Using Anti-EGFR Antibody Conjugated Gold Nanoparticles. *Cancer Lett.* **2006**, *239*, 129–135.
- O'Neal, D. P.; Hirsch, L. R.; Halas, N. J.; Payne, J. D.; West, J. L. Photo-thermal Tumor Ablation in Mice Using near Infrared-Absorbing Nanoparticles. *Cancer Lett.* **2004**, *209*, 171–176.
- Loo, C., A.; Lowery, A.; Halas, N.; West, J.; Drezek, R. Immunotargeted Nanoshells for Integrated Cancer Imaging and Therapy. *Nano Lett.* **2005**, *5*, 709–711.

- 16 Jain, P. K.; El-Sayed, I. H.; El-Sayed, M. A. Au Nanoparticles Target Cancer. *Nano Today* **2007**, *2*, 18–29.
- 17 Huang, X.; Jain, P. K.; El-Sayed, I. H.; El-Sayed, M. A. Gold Nanoparticles: Interesting Optical Properties and Recent Applications in Cancer Diagnostics and Therapy. *Nanomedicine* **2007**, *2*, 681–693.
- 18 Jain, P. K.; Lee, K. S.; El-Sayed, I. H.; El-Sayed, M. A. Calculated Absorption and Scattering Properties of Gold Nanoparticles of Different Size, Shape, and Composition: Applications in Biological Imaging and Biomedicine. *J. Phys. Chem. B* **2006**, *110*, 7238–7248.
- 19 Kelly, K. L.; Coronado, E.; Zhao, L. L.; Schatz, G. C. The Optical Properties of Metal Nanoparticles: The Influence of Size, Shape, and Dielectric Environment. *J. Phys. Chem. B* **2003**, *107*, 668–677.
- 20 Kreibig, U.; Vollmer, M., *Optical Properties of Metal Clusters*; Springer: Berlin, 1995; Vol. 25.
- 21 Link, S.; El-Sayed, M. A. Optical Properties and Ultrafast Dynamics of Metallic Nanocrystals. *Annu. Rev. Phys. Chem.* **2003**, *54*, 331–366.
- 22 Jain, P. K.; Huang, X.; El-Sayed, I. H.; El-Sayed, M. A. Review of Some Interesting Surface Plasmon Resonance-Enhanced Properties of Noble Metal Nanoparticles and Their Applications to Biosystems. *Plasmonics* **2007**, *2*, 107–118.
- 23 Eustis, S.; El-Sayed, M. A. Why Gold Nanoparticles Are More Precious than Pretty Gold: Noble Metal Surface Plasmon Resonance and Its Enhancement of the Radiative and Nonradiative Properties of Nanocrystals of Different Shapes. *Chem. Soc. Rev.* **2006**, *35*, 209–217.
- 24 Katz, E.; Willner, I. Nanobiotechnology: Integrated Nanoparticle-Biomolecule Hybrid Systems: Synthesis, Properties, And Applications. *Angew. Chem., Int. Ed.* **2004**, *43*, 6042–6108.
- 25 Yguerabide, J.; Yguerabide, E. E. Light-Scattering Submicroscopic Particles as Highly Fluorescent Analogs and Their Use as Tracer Labels in Clinical and Biological Applications I. and II. *Anal. Biochem.* **1998**, *262*, 137–176.
- 26 Link, S.; El-Sayed, M. A. Shape and Size Dependence of Radiative, Non-Radiative and Photothermal Properties of Gold Nanocrystals. *Int. Rev. Phys. Chem.* **2000**, *19*, 409–453.
- 27 Oldenburg, S. J.; Averitt, R. D.; Westcott, S. L.; Halas, N. J. Nanoengineering of Optical Resonances. *Chem. Phys. Lett.* **1998**, *28*, 243–247.
- 28 Link, S.; El-Sayed, M. A. Size and Temperature Dependence of the Plasmon Absorption of Colloidal Gold Nanoparticles. *J. Phys. Chem. B* **1999**, *103*, 4212–4217.
- 29 Weissleder, R. A Clearer Vision for in Vivo Imaging. *Nat. Biotechnol.* **2001**, *19*, 316–317.
- 30 Murphy, C. J.; Sau, T. K.; Gole, A. M.; Orendorff, C. J.; Gao, J.; Gou, L.; Hunyadi, S. E.; Li, T. Anisotropic Metal Nanoparticles: Synthesis, Assembly, and Optical Applications. *J. Phys. Chem. B* **2005**, *109*, 13857–13870.
- 31 Link, S.; Mohamed, M. B.; El-Sayed, M. A. Simulation of the Optical Absorption Spectra of Gold Nanorods as a Function of Their Aspect Ratio and the Effect of the Medium Dielectric Constant. *J. Phys. Chem. B* **1999**, *103*, 3073–3077. Erratum: *J. Phys. Chem. B* **2005**, *109*, 10531–10532.
- 32 Jana, N. R.; Gearheart, L.; Murphy, C. J. Seed-Mediated Growth Approach for Shape-Controlled Synthesis of Spheroidal and Rod-like Gold Nanoparticles Using a Surfactant Template. *Adv. Mater.* **2001**, *13*, 1389–1393.
- 33 Nikoobakht, B.; El-Sayed, M. A. Preparation and Growth Mechanism of Gold Nanorods (NRs) Using Seed-Mediated Growth Method. *Chem. Mater.* **2003**, *15*, 1957–1962.
- 34 Jain, P. K.; El-Sayed, M. A. Universal Scaling of Plasmon Coupling in Metal Nanostructures: Extension from Particle Pairs to Nanoshells. *Nano Lett.* **2007**, *7*, 2854–2858.
- 35 Prodan, E.; Radloff, C.; Halas, N. J.; Nordlander, P. A Hybridization Model for the Plasmon Response of Complex Nanostructures. *Science* **2003**, *302*, 419–422.
- 36 Sun, Y.; Mayers, B. T.; Xia, Y. Template-Engaged Replacement Reaction: A One-Step Approach to the Large-Scale Synthesis of Metal Nanostructures with Hollow Interiors. *Nano Lett.* **2002**, *2*, 481–485.
- 37 Hu, M.; Chen, J.; Li, Z.-Y.; Au, L.; Hartland, G. V.; Li, X.; Marquez, M.; Xia, Y. Gold Nanostructures: Engineering their Plasmonic Properties for Biomedical Applications. *Chem. Soc. Rev.* **2006**, *35*, 1084–1094.
- 38 Yiwen, Wang; Xie, X.; Wang, X.; Ku, G.; Gill, K. L.; O'Neal, D. P.; Stoica, G.; Wang, L. V. Photoacoustic Tomography of a Nanoshell Contrast Agent in the in Vivo Rat Brain. *Nano Lett.* **2004**, *4*, 1689–1692.
- 39 Sönnichsen, C.; Franzl, T.; Wilk, T.; von Plessen, G.; Feldmann, J.; Wilson, O.; Mulvaney, P. Drastic Reduction of Plasmon Damping in Gold Nanorods. *Phys. Rev. Lett.* **2002**, *88*, 077402.
- 40 Lee, K.-S.; El-Sayed, M. A. Gold and Silver Nanoparticles in Sensing and Imaging: Sensitivity of Plasmon Response to Size, Shape, and Metal Composition. *J. Phys. Chem. B* **2006**, *110*, 19220–19225.
- 41 Underwood, S.; Mulvaney, P. Effect of the Solution Refractive Index on the Color of Gold Colloids. *Langmuir* **1994**, *10*, 3427–3430.
- 42 Haes, A. J.; Zou, S.; Schatz, G. C.; Van Duyne, R. P. Nanoscale Optical Biosensor: Short Range Distance Dependence of the Localized Surface Plasmon Resonance of Noble Metal Nanoparticles. *J. Phys. Chem. B* **2004**, *108*, 6961–6968.
- 43 Jain, P. K.; El-Sayed, M. A. Surface Plasmon Resonance Sensitivity of Metal Nanostructures: Physical Basis and Universal Scaling in Metal Nanoshells. *J. Phys. Chem. C* **2007**, *111*, 17451–17454.
- 44 Jain, P. K.; Huang, W.; El-Sayed, M. A. On the Universal Scaling Behavior of the Distance Decay of Plasmon Coupling in Metal Nanoparticle Pairs. *Nano Lett.* **2007**, *7*, 2080–2088.
- 45 Malinsky, M. D.; Kelly, K. L.; Schatz, G. C.; Van Duyne, R. P. Chain Length Dependence and Sensing Capabilities of the Localized Surface Plasmon Resonance of Silver Nanoparticles Chemically Modified with Alkanethiol Self-Assembled Monolayers. *J. Am. Chem. Soc.* **2001**, *123*, 1471.
- 46 Haes, A. J.; Hall, W. P.; Chang, L.; Klein, W. L.; Van Duyne, R. P. A Localized Surface Plasmon Resonance Biosensor: First Steps toward an Assay for Alzheimer's Disease. *Nano Lett.* **2004**, *4*, 1029–1034.
- 47 McFarland, A. D.; Van Duyne, R. P. Single Silver Nanoparticles as Real-Time Optical Sensors with Zeptomole Sensitivity. *Nano Lett.* **2003**, *3*, 1057.
- 48 Perez-Juste, J.; Pastoriza-Santos, I.; Liz-Marzán, L. M.; Mulvaney, P. Gold Nanorods: Synthesis, Characterization and Applications. *Coord. Chem. Rev.* **2005**, *249*, 1870–1901.
- 49 Yu, C.; Irudayaraj, J. Multiplex Biosensor Using Gold Nanorods. *Anal. Chem.* **2007**, *79*, 572–579.
- 50 Sun, Y.; Xia, Y. Increased Sensitivity of Surface Plasmon Resonance of Gold Nanoshells Compared to That of Gold Solid Colloids in Response to Environmental Changes. *Anal. Chem.* **2002**, *74*, 5297–5305.
- 51 Jain, P. K.; Eustis, S.; El-Sayed, M. A. Plasmon Coupling in Nanorod Assemblies: Optical Absorption, Discrete Dipole Approximation Simulation, and Exciton-Coupling Model. *J. Phys. Chem. B* **2006**, *110*, 18243–18253.
- 52 Jain, P. K.; Qian, W.; El-Sayed, M. A. Ultrafast Electron Relaxation Dynamics in Coupled Metal Nanoparticles in Aggregates. *J. Phys. Chem. B* **2006**, *110*, 136–142.
- 53 Su, K. H.; Wei, Q.-H.; Zhang, X.; Mock, J. J.; Smith, D. R.; Schultz, S. Interparticle Coupling Effects on Plasmon Resonances of Nanogold Particles. *Nano Lett.* **2003**, *3*, 1087–1090.
- 54 Deniz, A. A.; Dahan, M.; Grunwell, J. R.; Ha, T.; Faulhaber, A. E.; Chemla, D. S.; Weiss, S.; Schultz, P. G. Observation of Förster Distance Dependence and Subpopulations Single-Pair Fluorescence Resonance Energy Transfer on Freely Diffusing Molecules. *Proc. Nat. Acad. Sci. U.S.A.* **2007**, *96*, 3670–3675.
- 55 Reinhard, B. M.; Sui, M.; Agarwal, H.; Alivasatos, A. P.; Liphardt, J. Calibration of Dynamic Molecular Rulers Based on Plasmon Coupling between Gold Nanoparticles. *Nano Lett.* **2005**, *5*, 2246–2252.
- 56 Nikoobakht, B.; Wang, J.; El-Sayed, M. A. Surface-Enhanced Raman Scattering of Molecules Adsorbed on Gold Nanorods: Off-Surface Plasmon Resonance Condition. *Chem. Phys. Lett.* **2002**, *366*, 17–23.
- 57 Huang, X.; El-Sayed, I. H.; Qian, W.; El-Sayed, M. A. Cancer Cells Assemble and Align Gold Nanorods Conjugated to Antibodies to Produce Highly Enhanced, Sharp, and Polarized Surface Raman Spectra: A Potential Cancer Diagnostic Marker. *Nano Lett.* **2007**, *7*, 1591–1597.
- 58 Jackson, J. B.; Halas, N. J. Surface-Enhanced Raman Scattering on Tunable Plasmonic Nanoparticle Substrates. *Proc. Natl. Acad. Sci. U.S.A.* **2004**, *101*, 17930–17935.
- 59 Schatz, G. C. Theoretical Studies of Surface Enhanced Raman Scattering. *Acc. Chem. Res.* **1984**, *17*, 370–376.
- 60 Michaels, A. M.; Jiang, J.; Brus, L. Ag Nanocrystal Junctions as the Site for Surface-Enhanced Raman Scattering of Single Rhodamine 6G Molecules. *J. Phys. Chem. B* **2000**, *104*, 11965–11971.
- 61 Nikoobakht, B.; El-Sayed, M. A. Surface-Enhanced Raman Scattering Studies on Aggregated Gold Nanorods. *J. Phys. Chem. A* **2003**, *107*, 3372–3378.
- 62 Link, S.; El-Sayed, M. A. Spectral Properties and Relaxation Dynamics of Surface Plasmon Electronic Oscillations in Gold and Silver Nanodots and Nanorods. *J. Phys. Chem. B* **1999**, *103*, 8410–8426.
- 63 Tabor, C.; Qian, W.; El-Sayed, M. A. Dependence of the Threshold Energy of Femtosecond Laser Ejection of Gold Nanoprisms from Quartz Substrates on the Nanoparticle Environment. *J. Phys. Chem. C* **2007**, *111*, 8934–8941.
- 64 Huang, W.; Qian, W.; El-Sayed, M. A. Gold Nanoparticles Propulsion from Surface Fueled by Absorption of Femtosecond Laser Pulse at Their Surface Plasmon Resonance. *J. Am. Chem. Soc.* **2006**, *128*, 13330–13331.

- 65 Jain, P. K.; Qian, W.; El-Sayed, M. A. Ultrafast Cooling of Photoexcited Electrons in Gold Nanoparticle-Thiolated DNA Conjugates Involves the Dissociation of the Gold-Thiol Bond. *J. Am. Chem. Soc.* **2006**, *128*, 2426–2433.
- 66 Link, S.; Burda, C.; Nikoobakht, B.; El-Sayed, M. A. How Long Does It Take to Melt a Gold Nanorod? A Femtosecond Pump-Probe Absorption Spectroscopic Study. *Chem. Phys. Lett.* **1999**, *315*, 12–18.
- 67 Huang, W.; Qian, W.; El-Sayed, M. A. Photothermal Reshaping of Prismatic Au Nanoparticles in Periodic Monolayer Arrays by Femtosecond Laser Pulses. *J. Appl. Phys.* **2005**, *98*, 114301/1–114301/8.
- 68 Huang, X.; Jain, P. K.; El-Sayed, I. H.; El-Sayed, M. A. Determination of the Minimum Temperature Required for Selective Photothermal Destruction of Cancer Cells with the Use of Immunotargeted Gold Nanoparticles. *Photochem. Photobiol.* **2006**, *82*, 412–417.
- 69 Zharov, V. P.; Galitovskaya, E. N.; Johnson, C.; Kelly, T. Synergistic Enhancement of Selective Nanophotothermolysis with Gold Nanoclusters: Potential for Cancer Therapy. *Lasers Surg. Med.* **2005**, *37*, 219–226.
- 70 Huang, X.; Qian, W.; El-Sayed, I. H.; El-Sayed, M. A. The Potential Use of the Enhanced Nonlinear Properties of Gold Nanospheres in Photothermal Cancer Therapy. *Lasers Surg. Med.* **2007**, *39*, 747–753.
- 71 Zharov, V. P.; Mercer, K. E.; Galitovskaya, E. N.; Smeltzer, M. S. Photothermal Nanotherapeutics and Nanodiagnostics for Selective Killing of Bacteria Targeted with Gold Nanoparticles. *Biophys. J.* **2006**, *90*, 619–627.