Gold nanorod photoluminescence : applications to imaging and temperature sensing

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CONCLUSIONS & OUTLOOK

6.1. GENERAL CONCLUSIONS

THIS thesis is a collection of heterogeneous results that range from etching of single gold nanorods to studying their anti-Stokes luminescence. Gold nanoparticles have been in the spotlight for almost two decades because of their optical properties[1]. They are ideal candidates for labelling[2] applications and also as biosensors[3]. Many properties of the nanoparticles have been already characterized but there is still a large number of them that needs to be addressed; this thesis provides several illustrations of this need.

Wet chemical synthesis of nanorods yields a high degree of heterogeneity between individual particles[4]. This was already observed in our group when measuring the quantum yield (QY) of particles with different aspect ratios[5]. The values differ by almost one order of magnitude between particles that, up to experimental accuracy, should have been identical. In every chapter of this thesis single-particle results have always been complemented with statistics.

Chapter 2 shows that the mean behavior of single particles is different from what is observed in bulk suspension. Chapters 3 and 4 focus on the anti-Stokes luminescence, a phenomenon greatly overlooked in the past decade. Chapter 5 on plasmon width is again an example of the heterogeneity observed at single-particle level. Experiments similar to these need proper statistics to be complete.

The four chapters of this thesis are but a proof that there is still room for investigation at single-particle level. Many intriguing phenomena can still be left to discover.

6.2. OUTLOOK

Every chapter includes a conclusion regarding the content of the chapter itself. This section on the other hand aims at pointing out what are the different possibilities that every chapter opens for future research.

6.2.1. CYANIDE ETCHING

Chapter 2 shows that is possible to change the shape of gold nanoparticles once immobilized on a glass coverslip. We employed cyanide etching because of its well understood chemistry with gold but the methodology is not limited to it; other reactions are possible alternatives. Moreover we have shown that it is possible to monitor the changes of shape by studying the evolution of the plasmon resonance and therefore the experiments can be performed under an optical microscope.

Other works have focused into the possibility of using the plasmon resonance shift as a detector of minute concentrations of cyanide[6]. At a single particle level we can easily detect μM concentrations and nM should be reachable without changes to the setup. Lowering the concentrations keeping reasonable measurement times reduces to improving the detection of the plasmon shift. However gold nanoparticles are completely etched away after being exposed to cyanide ions for enough time. This would make samples non reusable.

Another interesting opportunity is the ability to change the spacing between particles with sub nanometer accuracy[7]. Gold nanorods are becoming promising nano antennas, and dimers of particles have a much stronger near field. However, controlling the spacing between particles is a major challenge. The results shown in chapter 2 can be extended to dimers, where slow etching of the surface of the particles can be used for tuning the distance between them.

6.2.2. BACKGROUND SUPPRESSION

Chapter 3 shows that it is possible to image gold nanorods in high background conditions by detecting their anti-Stokes emission. The chapter focuses into imaging under living cells but the technique is not limited to biological applications. High background conditions can include working with fluorescent molecules in solution, for example to study enhanced FCS[8]. Background suppression is not the only advantage of anti-Stokes imaging.

A common problem in colocalization studies is the correction for chromatic aberrations and misalignment of different beams. If one desires to colocalize a gold nanorod and a fluorescent dye with absorption in the same spectral region, the anti-Stokes emission provides a way to achieve it with only one excitation laser and one detection path. Employing a single laser beam rules out the possibility of a misalignment of the excitation path; the detection of both channels (anti-Stokes for the rods and Stokes for the dye) can be concentrated over a short spectral range, thus minimizing chromatic aberrations.

Colocalizing gold nanorods and fluorescently labelled proteins can give insight into the different processes that mediate the uptake of gold nanorods[2]. It can also be useful for characterizing the targeting of proteins in living cells. A gold nanorod can be functionalized to bind to specific proteins[9]; the binding efficiency and specificity, however, are difficult to determine *in vivo* if there is a high background signal.

Anti-Stokes detection is not limited to imaging. If used as labels, nanorods can be used for tracking[10] specific proteins for extended periods of time. In living cells, regulatory mechanisms depend on free diffusion and active transport[11]. Tracking of functionalized single nanoparticles can provide important insight into mechanisms that are active over different timescales[12].

6.2.3. TEMPERATURE SENSING WITH ANTI-STOKES LUMINESCENCE

Chapter 4 shows that anti-Stokes luminescence from single nanoparticles can be used for nano-thermometry. This novel result opens many possibilities in the fields of photothermal therapy[13] and nano fabrication[14]. For over 20 years gold nanoparticles have been studied as possible candidates for treating cancer[15]. A large community is focused into using nanoparticles to locally increase the temperature of malignant cells, preserving the healthy ones.

After decades of research, however, there is little insight into the temperature that the nanoparticles have to reach to induce cellular death[16]. The conclusions of chapter 4 clearly show that the technique developed is ready to be implemented in biologically relevant conditions. Being able to actively control and monitor the temperature of nanoparticles in or around cells has never been done before and can yield important answers to the mechanisms that induce cell death.

Moreover the method described in chapter 4 can be used to measure the temperature of nanoparticles in various situations. For instance the characterization of optically trapped nanoparticles normally relies on assumptions of the temperature[17]. Nano bubble generation[18], polymerization at the nanoscale[19], controlled chemical reactions[20], photothermal detection[21] are some of the fields where actually measuring the temperature of the nanoparticles instead of estimating it can provide insight into new phenomena.

An important task for future work should be to characterize different particle geometries. Gold shells[22], bipyramids[23], even spheres of different diameters can be better suited for temperature sensing. Different plasmon resonances and different quantum yields can make other particles better anti-Stokes emitters.

Acquiring spectra as was done in chapter 4 is a slow process; it can take several minutes to obtain a proper signal-to-noise ratio. There is a possibility to shorten the acquisition times by studying the ratio of anti-Stokes to Stokes emission[24]. In principle the Stokes emission is constant with temperature and depends only on the laser power; the anti-Stokes however will be brighter for higher temperatures. Already in figure 4.2 it is possible to observe that the ratio of both types of emission can be easily reproduced by numerical calculations. Preliminary calculations show that the ratio of anti-Stokes to Stokes changes with temperatures, but experimental data is missing.

6.2.4. PLASMON DAMPING

Chapter 5 shows the relation between the plasmon damping rate and the temperature of the medium surrounding the nanoparticles. The main idea of the chapter was to explore the possibility of using the broadening of the plasmon resonance as an alternative thermometry strategy. Anti-Stokes luminescence has the advantage of not needing a calibration, but the higher laser powers employed induce a temperature rise that can be much higher than the temperature to be detected.

Chapter 5 shows that, on average, the linear relationship between the plasmon full width at half maximum and temperature agrees with the expected value from bulk gold. However there is a big heterogeneity between nanoparticles, not all of them have the same broadening rate. This explains why the broadening was not observed in bulk suspension and also sets a limit to the applicability of this method for temperature measurements.

Since every nanoparticle behaves in slightly a different way when increasing temperature, one needs to build proper statistics to determine how much the temperature of the sample increased or decreased. The statistics can be built either by studying several individual nanoparticles, as was done in this thesis, or by placing a bigger number of particles in the focal spot, as done with quantum dots[25]. The heterogeneity, however, can pose a limit to the applicability of the method. It is possible that other geometries such as bipyramids exhibit a more homogeneous behavior.

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