Noble Metal Nanocrystals:
Synthesis, Optical Properties, and Biological Applications

Nanocrystals with a size in the range of 1-100 nm (1 nm=10^{-9} m) are an important class of materials that are having a major impact in a diverse range of applications, including chemical and biological detection, *in-vivo* drug delivery, catalysis, optics, and high density data storage.

The primary goal of this thesis research was to synthesize new types of metal nanocrystals, evaluate their fundamental optical properties, and develop biological applications based upon properties that offer advantages in terms of sensitivity, selectivity, and multiplexing capabilities. Noble metal nanocrystals are particularly important because of their chemical stability and fascinating optical properties that can be tailored through control over particle size, shape, composition, and morphology.

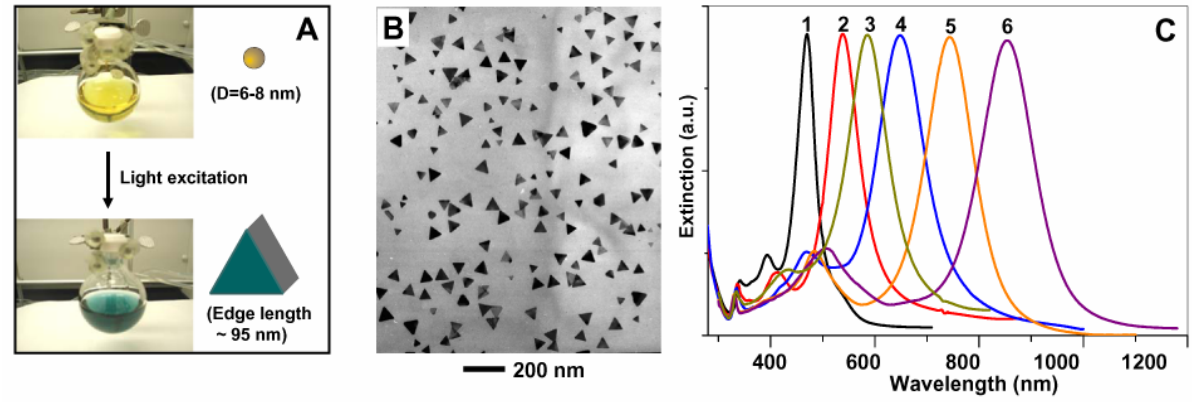

Fig 1. Light-induced conversion of colloidal silver nanospheres into nanoprisms. **(**A) The color change during the conversion process. (B) TEM image of the final nanoprisms (average edge length: 95 ± 11 nm. (C) The optical spectra for six different sized nanoprisms (edge length: 38 ± 7 nm, 50 ± 7 nm, 62 ± 9 nm, 72 ± 8 nm, 95 ± 11 nm, and 120 ± 14 nm for 1-6, respectively) prepared by varying excitation wavelength.

The first part of the thesis work presents a photochemical method for converting spherical Ag nanoparticles into triangular nanoprisms in the form of colloidal suspensions (Fig 1A-B). It is well-known that the size and shape of nanoparticles provides important control over many of their physical and chemical properties, including light absorption, photoluminescence, and catalytic activity. Although colloid chemists have gained excellent control over spherical particle size for several compositions (e.g. Au, Ag), the challenge of synthetically controlling particle shape has been met with limited success. To realize the full potential of these novel materials, the development of bulk solution synthetic methods that offer shape and size control is of paramount importance. Our work has demonstrated a remarkably efficient light-induced method for synthesizing large quantities of Ag nanoprisms of desired sizes (edge length) in high yield in the form of colloidal suspensions. This photo-mediated route has led to a colloid with unusual optical properties that directly relate to particle shape and size (Fig 1C). Theoretical calculations (in collaboration with Prof. *George C. Schatz*) allowed for the assignment of the nanoprism plasmon bands and the identification of two distinct quadrupole plasmon resonances. Importantly, the nanoprism edge length (thickness remains constant) can be controlled over the 30-120 nm range by excitation wavelength. Potential applications of these novel nanoparticles include biological labeling, high-density recording media, optical enhancers, and nano-pigments, etc. This work

provides the first demonstration of the use of plasmon excitation as a chemical tool to provide control over nanoparticle growth and, ultimately, particle size and shape.

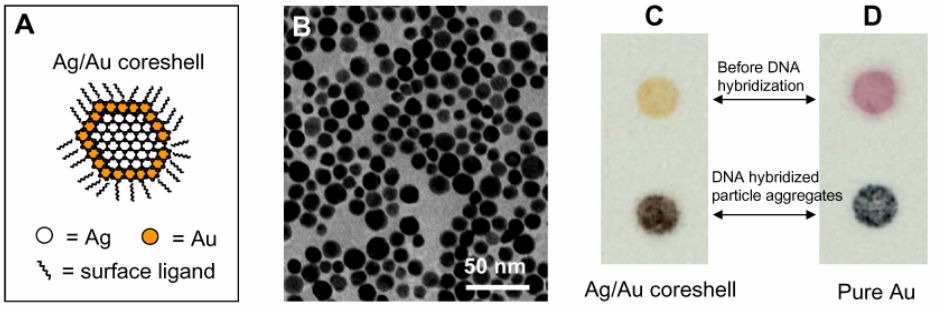

Fig 2. Ag/Au coreshell nanocrystals. (A) Scheme; (B) TEM image; (C) A colorimetric DNA detection method based on this novel type of nanoparticles; (D) A colorimetric response of pure Au nanoparticles.

The second part of the thesis work focuses on the development of nanoparticle-based multiplexed DNA detection systems. DNA detection is particularly important for medical, forensic, environmental, and warfare applications. The central technique is the labeling strategies, as DNA itself does not have intrinsic properties that can be used for differentiation. Conventionally, organic fluorescent dyes are widely used to label DNA targets. However, the inherent broad melting properties of DNA greatly limit the selectivity and sensitivity of assays. A promising method is using nanoparticles to replace fluorescent tags. Previous work had demonstrated that gold nanoparticles functionalized with oligonucleotides offer advantages of extraordinarily detection sensitivity and specificity (or selectivity). However, a limitation of this approach is that it is inherently a one-color system based upon Au nanoparticles. The flexibility and applicability of all DNA detection systems benefit from access to multiple types of labels with addressable and individually discernable labeling information. In this work, we introduced two strategies for highly selective and sensitive detection of DNA in a multiplexed format. The first strategy is to exploit the rich optical properties of metal nanoparticles. For example, silver nanoparticles exhibit a surface plasmon band at ~ 400 nm— a spectral regime that is distinct from that of Au (~ 520 nm). However, colloidal Ag nanoparticles exhibit poor chemical stability; no successful routes had been developed for creating stable oligonucleotide conjugates with Ag nanoparticles. Herein, we used a coreshell strategy and developed a low temperature synthetic method for generating coreshell particles consisting of a core of Ag and a monolayer shell of Au (Fig 2A-B). This new type of coreshell particle yields a novel nanoparticle composition with the optical properties of silver but the chemical stability of gold. These coreshell particles can be readily functionalized with oligonucleotides using the proven preparatory methods for pure gold particles. We have used this novel nanocrystal composition to access a new colorimetric detection system that is distinct from the pure gold system (Fig 2C-D). This coreshell approach could be generalized to prepare other particles such as Cu/Au to create multicolor nanoparticles for multiplexed DNA detection. In contrast, our second strategy for multiplexed detection, utilizes only one type of particles (e.g. Au) as carriers that are functionalized with different Raman-dye labeled oligonucleotides (Fig 3A). The individual Raman spectroscopic fingerprints (with narrow bands) of the particle probes, which can be identified after silver enhancing via surface-enhanced Raman scattering (SERS) spectroscopy, are used to label and identify different DNA targets (Fig 3B-C). Since the SERS-active substrate in this strategy is generated immediately prior to the detection event, a large and reproducible Raman scattering signal can be obtained. Massively parallel labeling can further be achieved by choice of different Raman dyes and dye mixing to generate new Raman spectroscopic fingerprints.

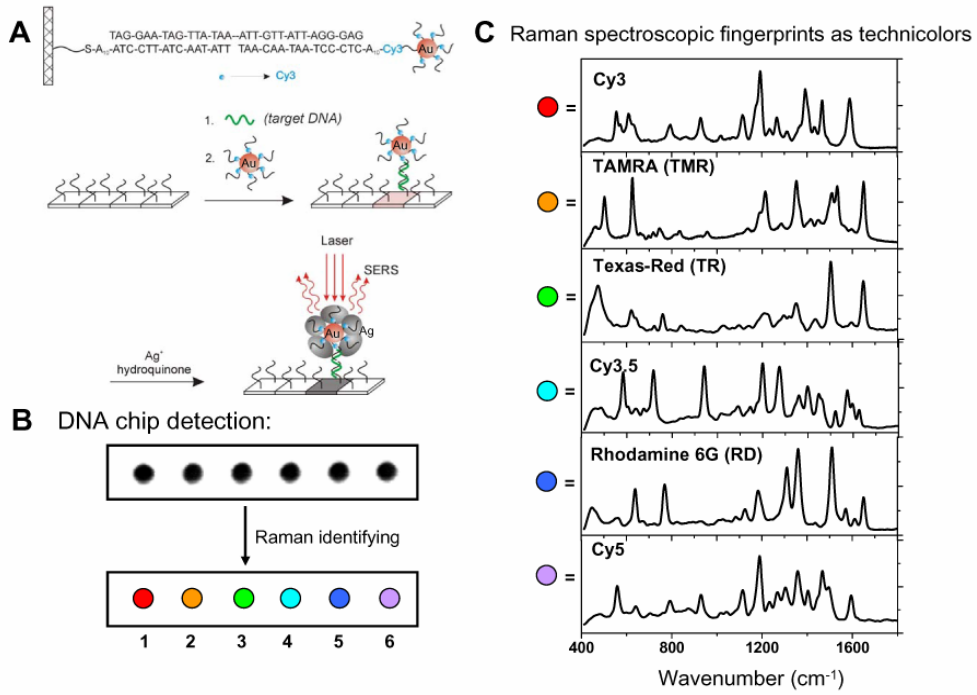

Fig 3. The Raman-based method for multiplexed DNA detection. (A) Scheme; (B) A proof-of-concept test to distinguish six dissimilar DNA targets in one solution with six Raman labeled nanoparticle probes with their color-coded spectra shown in (C). The targets were correctly read-out after Raman decoding.

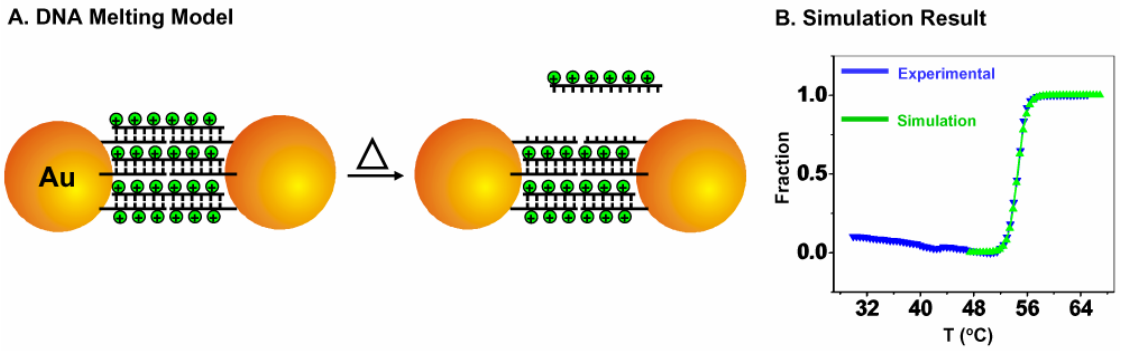

Fig 4. The cooperative DNA melting mechanism in the presence of nanoparticles. (A) Scheme (for clarity, only one pair of particles is drawn); (B) A comparison of experimental and theoretical melting curves.

Finally, we systematically investigated what controls the melting properties of DNA-linked nanoparticle assemblies. This work was intended to gain insight into the mechanism why oligouncleotide-functionalized nanoparticle probes can significantly increase DNA detection selectivity and sensitivity as compared with many conventional assays that rely on molecular fluorophores. To elucidate this mechanism is also essential for developing other high selectivity DNA detection systems and for constructing materials from these novel nanoparticle materials. In this work, the experimental data, coupled with theoretical modeling (in collaboration with Prof. *George C. Schatz*), identified a cooperative DNA melting mechanism that attributes the sharp melting to two key factors: the presence of multiple DNA linkers between nanoparticles (hence, high local salt concentration) and a decrease in the melting temperature as DNA duplexes melt due to a concomitant reduction in local salt concentration (Fig 4). This cooperative melting effect originates from short-range duplex-to-duplex interactions and is independent of DNA sequences and nanoparticle type. Indeed, it is this sharp melting transition that leads to the ultrahigh selectivity observed in assays based upon these types of nanoparticle probes.