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Fast, Efficient, and Stable Conjugation of Multiple DNA Strands on Colloidal Quantum Dots

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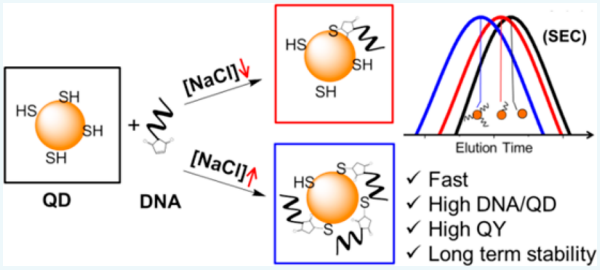
Supporting Information

ABSTRACT: A novel method for covalent conjugation of DNA to polymer coated quantum dots (QDs) is investigated in detail. This method is fast and efficient: up to 12 DNA strands can be covalently conjugated per QD in optimized reaction conditions. The QD-DNA conjugates can be purified using size exclusion chromatography and the QDs retain high quantum yield and excellent stability after DNA coupling. We explored single-stranded and double-stranded DNA coupling, as well as various lengths. We show that the DNA coupling is most efficient for short (15 mer) single-stranded DNA. The DNA coupling has been performed on QDs emitting at four different wavelengths, as well as on gold nanoparticles, suggesting that this technique can be generalized to a wide range of nanoparticles.



INTRODUCTION

Several exceptional properties such as broad absorption, size-tunable emission, high photostability, and remarkable TEM contrast position semiconductor nanoparticles or quantum dots (QDs) as desirable candidates for applications in biology.[1−3] For most of these applications, QDs need to be functionalized with biomolecules such as peptides, proteins, and DNA.[4−6] Particularly, QD functionalized with DNA (QD-DNA) have been extensively used for siRNA,[7] FISH,[8−10] and DNA mismatch detection and femtomolar detection of nucleic acids.[11,12] QD-DNA have also been used for exploring bimolecular FRET,[13−15] and in photovoltaics.[16,17] Facile chemical modification[18] and predictable base pairing properties of DNA enable QD-DNA conjugates to be further utilized to control QD valency and form heteromolecular structures with precise control.[19] These applications have fostered interest in investigating novel methods for conjugation of DNA to QDs with high yields and reproducibility.

Conjugation reactions with DNA and other biomolecules are carried out in aqueous solution. However, immediately after synthesis, QDs are covered with aliphatic organic ligands which render them insoluble in water. Therefore, before conjugation with DNA, QDs need to be first made water-soluble. There are primarily two approaches to transfer QDs from organic solvents to aqueous solution. The first approach involves encapsulation of QDs inside a phospholipid layer.[20,21] QDs thus obtained have substantially increased hydrodynamic diameter, and limited stability in complex biological media.[22] Another approach involves ligand exchange of QDs with hydrophilic small molecules or polymers.[23−28] These polymers generally bear functional groups like thiolates or pyridines[29] with affinity toward the cations on surface of QDs, and other functional groups that impart water solubility. QDs coated with polymers bearing multidentate ligands often possess greater colloidal stability along with smaller size. Also, polymers can be customized to bear additional functional groups, useful for functionalization of QDs with biomolecules such as DNA.[30,31]

Several methods to conjugate DNA to QDs have been explored previously. For example, amine functionalized DNA (DNA-NH_2) has been coupled to carboxyl group on a polymer-coated QDs by reaction with 1-ethyl-3-(3-(dimethylamino)-propyl)carbodiimide hydrochloride (EDC) and *N*-hydroxysuccinimide (NHS).[30,32] Another method to generate QD-DNA conjugates involves ligand exchange of QDs first with mercaptopropionic acid (MPA) followed by displacement of MPA by thiol-functionalized DNA (DNA-SH) directly,[33−35] or mediated by a linker.[36] Alternately, QDs have been functionalized with streptavidin first, followed by addition of a biotinylated DNA.[7,15] Nevertheless, each strategy has its own limitations. EDC-NHS coupling suffers from low conjugation efficiencies due to hydrolysis of NHS esters. Ligand exchange with thiol-labeled DNA is limited because the surface bound

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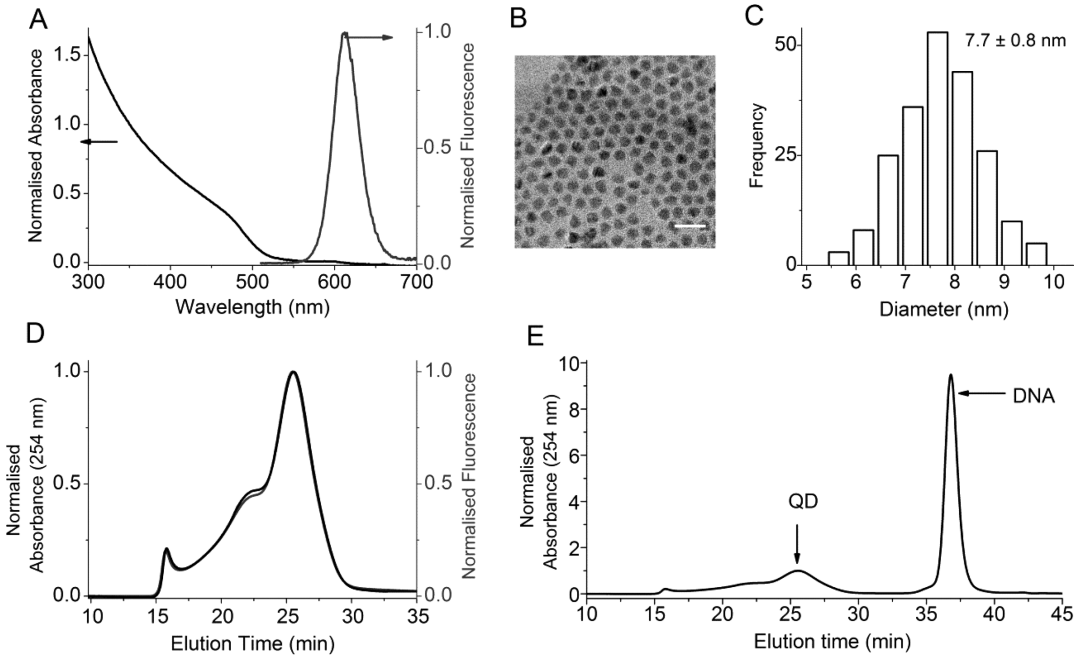

Figure 1. Characterization of QDs. (A) UV−vis absorption (black) and fluorescence emission (gray) spectra of QD in hexane. (B) TEM image of QDs. (C) QDs are 7.7 ± 0.8 nm in diameter. (D) Chromatogram showing elution of polymer capped QDs monitored by absorbance at 254 nm (black) and by fluorescence with λ_{exc} = 350 nm, λ_{em} = 610 nm (gray). (E) Chromatogram of mixture of DNA and QDs (DNA/QD = 40/1) monitored by absorbance at 254 nm. (D) and (E) were obtained from size exclusion column in PBS (1×) pH 7.4 at 20 °C.

thiols can be easily oxidized by light or oxygen. Finally, the biotin−streptavidin strategy increases the size of QDs substantially, thereby limiting its applicability. Also, conjugation of DNA on the QD surface is limited by the amount of streptavidin available on the QD. Along with these limitations specific to chemical reactions, purification of QD-DNA conjugates is often tricky. The most conventional method is gel purification and centrifugation, which both are labor intensive and tend to decrease yields.[12,37,38]

In this paper, we discuss a novel method to conjugate DNA onto polymer-coated QDs. The polymer used to cover the QDs displays both anchoring (thiol-functionalized monomer) and water-solubilizing (zwitterion-based monomer) groups. These QDs have excellent stability in aqueous solutions of varied dilutions and ionic strength, without compromising the quantum yield.[27] In this work we choose to use some of the thiols present on this zwitterionic polymer to conjugate DNA-NH_2 on QDs. Utilizing the thiols eliminates the need for incorporation of any additional functional groups specifically for conjugation, thereby simplifying coupling of biomolecules on QDs.[39] The QD-DNA conjugates have been purified by size exclusion chromatography (SEC). We have characterized the coupling reaction exhaustively and find that the strategy is fast, efficient, robust, and reproducible. Covalent conjugation of DNA on polymer coated QD does not affect the colloidal stability and photophysical properties of QDs. The conjugated DNA remains available for hybridization and does not leach out of the polymer surface in dilute conditions over days. Additionally the strategy is also generalizable to a broad spectrum of nanoparticles. This approach overcomes several limitations of yield, purification, and colloidal stability reported previously and strongly positions our QD-DNA conjugates for applications at the interface of material science and biology.

RESULTS

2.1. Charaterization of QDs. QDs in hexane emitting at 610 nm (called QDs hereafter, unless specified otherwise) were obtained from Nexdot (www.nexdot.fr). These QDs have a broad absorption range (200−600 nm) with primary excitonic peak at 595 nm and emission maximum at 610 nm with full width of half-maxima (fwhm) around 33 nm (Figure 1A). The QDs are 7.7 ± 0.8 nm in diameter (Figure. 1B) and are capped by zwitterionic polymer as described previously.[27] The polymer capped QDs were then characterized by SEC (Figure 1D). These QDs have identical elution profiles in both absorbance and fluorescence modes of detection. This confirms that all QDs remain fluorescent when they pass through the SEC column. These QDs elute between 15 and 30 min, with a maximum concentration at 25.0 ± 0.1 min. Free ssDNA (DNA_a) elutes around 36.0 ± 0.3 min (Figure. 1E). This difference in retention time of QDs and DNA ensures that QD-DNA conjugates can be efficiently separated from free (unconjugated) DNA using SEC.

2.2. Synthesis and Purification of QD-DNA Conjugates. *2.2.1. Conjugation of Amine-Labeled DNA to QD.* In order to couple the DNA-NH_2 with the thiols on the QDs, a heterobifunctional linker, sulfosuccinimidyl-4-(*N*-maleimidomethyl) cyclohexane-1-carboxylate (sSMCC) was used. Typically, the DNA-NH_2 (ss DNA_a, Table SI1) was reacted with the NHS of sSMCC (sSMCC/DNA = 25/1) and then purified by precipitation to obtain DNA-maleimide. Recovery of DNA was estimated to be >95%, as monitored by absorbance spectroscopy before and after ethanol precipitation. In parallel, water-solubilized QDs were reduced with tris(2-carboxyethyl)-phosphine hydrochloride (TCEP) in order to regenerate free thiols (QD-SH) on the polymer for reaction with DNA-maleimide. Subsequently TCEP was removed from reduced QDs (QD-SH) by SEC. QD-SH were recovered from SEC with yield of 90% or above, as measured by absorbance

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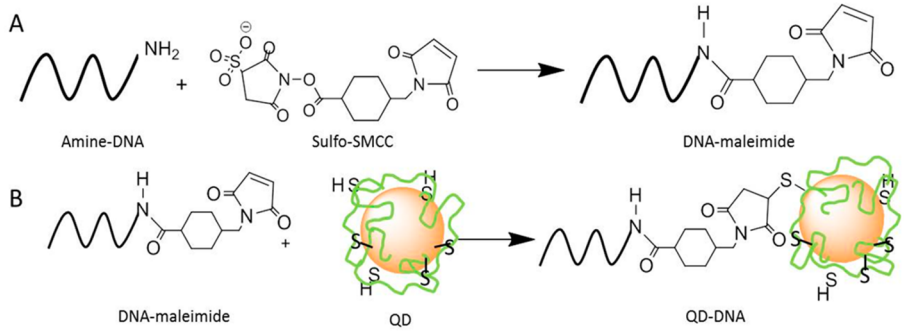
Scheme 1. General Scheme for Conjugation of NH_2-DNA to Thiols on QD[a]



[a](A) Reaction of DNA-NH_2 to NHS of sSMCC to form DNA-maleimide. (B) Reaction of DNA-maleimide to thiols on QD to form QD-DNA conjugates. (Schematics not to scale.)

spectroscopy. Finally, QD-SH were mixed with DNA-maleimide and allowed to react overnight (Schemes 1 and SI1). The conjugated QDs were then purified by SEC in PBS 1× pH 7.4, 25 °C.

2.3. Characterization of QD-DNA Conjugates. *2.3.1. Qualitative Detection of DNA on QDs.* The conjugation of DNA on QD was verified by several approaches (Figure 2). In the first approach, purified QD-DNA conjugates are mixed with a biotinylated complementary DNA sequence (DNA_b-Bt) and then incubated with streptavidin agarose beads for 5 min. The agarose beads are washed three times to remove nonspecifically associated QDs and observed under fluorescent lamp or microscope. The beads appear brightly fluorescent (Figure 2B (iii)). In contrast, the same experiment performed with noncomplementary DNA sequence (as a negative control) does not give fluorescent beads (Figure 2B (iv)). This also proved that the ss-DNA conjugated on the QD is accessible for hybridization, in contrast to that observed in several DNA functionalized nanoparticles.[40,41] To assess the nonspecificity of the above assay, streptavidin agarose beads were mixed with QD and QD-DNA without biotin-labeled complement. Beads from both these samples showed negligible fluorescence (Figure 2B (i,ii)). To assess whether addition of biotinylated DNA enhances any nonspecific adsorption of QD-DNA on the beads, biotinylated DNA of random (noncomplementary) sequence was mixed with QD-DNA followed by incubation with beads. These beads remained non fluorescent, hereby asserting the specificity of this assay (Figure 2B (iv)).

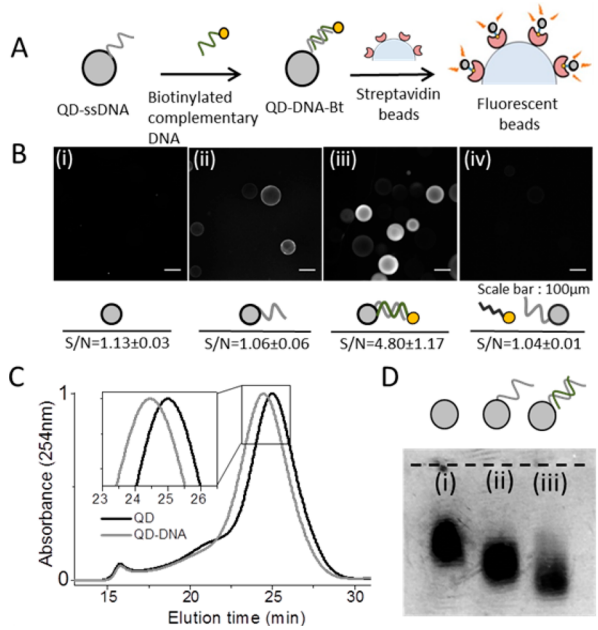


Figure 2. Validation of DNA coupling on QD. (A) Schematic of QD-DNA conjugate hybridized to complementary DNA labeled with biotin mixed with streptavidin agarose beads. (B) Fluorescence microscopy image of streptavidin agarose beads mixed with (i) QD, (ii) QD-DNA, (iii) QD-DNA with complementary biotin labeled DNA, and (iv) QD-DNA with noncomplementary DNA labeled with biotin. Signal to Noise ratio (S/N) corresponding to each image is indicated. (C) Chromatograms of QD (Black) and QD-DNA (gray). Inset: magnified chromatogram from 23 to 26.5 min. (D) Gel electrophoresis showing migration difference between (i) QD, (ii) QD-ssDNA, and (iii) QD-dsDNA.

QD-DNA conjugates were further characterized by size exclusion chromatography (SEC). QD samples elute with peak maxima at 25.0 ± 0.1 min, whereas QD-DNA conjugates elute at 24.4 ± 0.2 min (Figure 2C). To confirm that change in retention time of QD-DNA conjugates in comparison to QD was due to only covalently conjugated DNA (and not nonspecifically adsorbed DNA), a random sequence of Cy5 labeled DNA was incubated with QD and QD-DNA samples for 48 h and injected into the SEC. The elution chromatograms of both these mixtures were obtained for absorbance at 350 and 640 nm for QD and Cy5, respectively. No signal from Cy5 was observed in the region of QD/QD-DNA elution (Figure SI2), thereby proving the absence of nonspecific absorption.

QD-DNA conjugates were also characterized by agarose gel electrophoresis. QD-DNA conjugates (QD-ssDNA and QD-dsDNA) migrated faster than QD (Figure 2D). The trend of faster migration in electrophoresis is consistent with that reported in the literature.[42]

2.3.2. Quantification of DNA per QD. To quantify the number of DNA molecules conjugated per QD, two approaches were taken. The primary approach involves the estimation of concentration of DNA by absorbance at 260 nm. First, absorbance spectra of QD and purified QD-DNA were normalized at 220 nm. Then from the spectrum of QD-DNA conjugate, QD was subtracted to obtain the spectra of conjugated DNA. The concentration of DNA was then estimated by the Beer-Lambert law.

To evaluate the precision of the quantification of DNA by absorbance in the deep-UV region, several reactions were quantified by a more conventional approach, using fluorophore-

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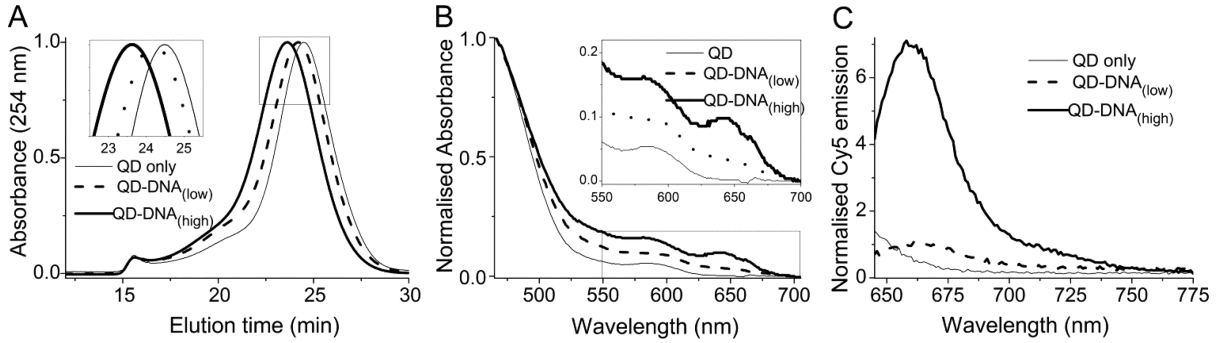

Figure 3. Effect of salt on conjugation of DNA to QD: Chromatogram of QDs (short dash), QDs conjugated to 15mer ssDNA in the presence of 140 mM NaCl (low salt, long dash) and 1400 mM NaCl (high salt, solid line). Inset: Magnification of chromatogram between 22.5 and 25.5 min. (B) Absorption spectra of QD, QD-DNA-$Cy5_{low\ salt}$, and QD-DNA-$Cy5_{high\ salt}$ normalized at 460 nm. Inset: magnification of absorption spectra between 560 and 700 nm. (C) Normalized fluorescence emission spectra of QD, QD-DNA-$Cy5_{low\ salt}$, and QD-DNA-$Cy5_{high\ salt}$ with λ_{exc} = 630 nm, $\lambda_{em\ s}$ = 645−800 nm (for Cy5).

labeled DNA. For these reactions, we used a double-stranded DNA with a NH_2 at the 5′ end on one stand and Cy5.5 at the 5′ end on complementary strand. After coupling of this ds-DNA on the QDs and its SEC purification, the quantification of DNA is performed using either the absorbance of DNA at 260 nm or the absorbance of Cy5.5 at 680 nm. Both approaches gave similar results (Figure SI3). In this work, most quantification was carried out by the first approach, unless specified otherwise.

2.4. Optimization of Coupling Conditions. *2.4.1. Effect of Reducing Agent.* The copolymer used to make the QDs water-soluble has a statistical distribution of zwitterionic moieties and dithiol groups that can bind on the QDs. Not all the dithiol groups from the polymer are bound to the QD surface, and free thiols can be used to conjugate DNA−maleimide, but the number of DNA conjugated on a QD with this approach is low (Table SI2). We explored the influence of TCEP as a reducing agent used to increase the number of thiol groups available for conjugation. We observe that addition of TCEP (up to 400/QD) improves the DNA conjugation yield. Higher molar excess of TCEP produces QD instability and rapid QD degradation under UV excitation. When 400 molar excess of TCEP is used, we could conjugate up to ∼3 ss-DNA per QDs, whereas when no TCEP is used, a negligible fraction of the QDs was conjugated with DNA (TableSI2).

2.4.2. Effect of Salt Concentration. To test whether addition of excess salt affects the reaction yield, several reactions were set at 140 mM NaCl and 1400 mM NaCl (Figure 3). For these experiments, a 15mer ss-DNA_a with a NH_2 on the 5′ end was used for conjugation and the quantification of the conjugation was done based on absorbance at 260 nm. On increasing the salt concentration from 140 to 1400 mM in the final reaction, the number of DNA conjugated per QD increased by a factor of 4 (Table 1). The influence of the salt on the DNA conjugation efficiency is first visible on the SEC data: the QD-DNA conjugates produced with 1400 mM NaCl elute faster from the column than both conjugates with 140 mM salt and unconjugated QDs, confirming higher conjugation yields (Figure 3A). To further confirm these results, a similar ss-DNA with a Cy5 (DNA_a-Cy5) at the 3′ end was conjugated to QD at 140 mM and 1400 mM NaCl concentration and the purified product was quantified with both absorbance and fluorescence of Cy5. Similar higher yields in the presence of 1400 mM NaCl were observed for these reactions as well (Figure 3B and C). Hence, we conclude that in our case, higher salt concentration indeed improves the conjugation efficiency of DNA to QD.

Table 1. Coupling Efficiency of DNA on QD at Different Salt Concentration and DNA Length

[NaCl][a] mM	length (bases)[b]	excess DNA[c]	#DNA/QD[d]	% DNA conjugated[e]
140	15	40	3.0 ± 1.0	7.5 ± 2.5
1400	15	25	11.5 ± 4.0	46.0 ± 16.0
1400	45	25	4.3 ± 1.0	17.3 ± 3.8

[a]NaCl concentration in the final reaction buffer. [b]Number of bases of DNA used for reaction. [c]Initial molar excess of DNA added in reaction compared to QD. [d]Number of DNA per QD determined by absorbance of Cy5 labeled DNA on purified reaction. [e]Percentage of total DNA finally conjugated w.r.t initial excess of DNA.

2.4.3. Effect of DNA Length. To test whether conjugation in the presence of salt is independent of molecular weight of DNA, two lengths of ssDNA (15mer and 45mer) were conjugated on QD in the presence of 1400 mM NaCl. We observed that the coupling efficiency depended on the DNA length. The average number of DNA/QD for ssDNA was 11.5 ± 4 for 15mer (DNA_a) and fell to 4.3 ± 1.0 for 45mer (DNA_c) DNA. We also observed that the conjugation of ds-DNA was not as efficient as ss-DNA of same length (4.7 ± 1.2 ds DNA/QD for 15 bp).

It is thus possible to conjugate both ss DNA and ds DNA from 15 to 45 bases long. The elution time of QD conjugated with 45-bases-long DNA is marginally faster than that of 15-bases-long, but substantially faster than unconjugated QDs (Figure 4A).

2.5. Stability of QD-DNA Conjugates. *2.5.1. Photostability of QD-DNA Conjugates.* Several articles report a loss of fluorescence quantum yield of QDs up to 50% or more, upon ligand exchange and conjugation with biomolecules including DNA.[32,43] However, in the method of conjugation presented in this work, the quantum yield (QY) of the QDs remained constant within 10% after ligand exchange and DNA conjugation (using Rhodamine 6G as a reference)[44] up to 1 month of storage in 0.2 M $NaHCO_3$ at 4 °C in the dark. Similar results were found with different types of QDs coated with the same polymer, subsequently conjugated with DNA. The QD-DNA conjugates could be lyophilized and readily redissolved in $NaHCO_3$ without loss of QY.

2.5.2. Solution Stability of QD-DNA Conjugates. Many reports in the past have discussed the loss of stability of QD

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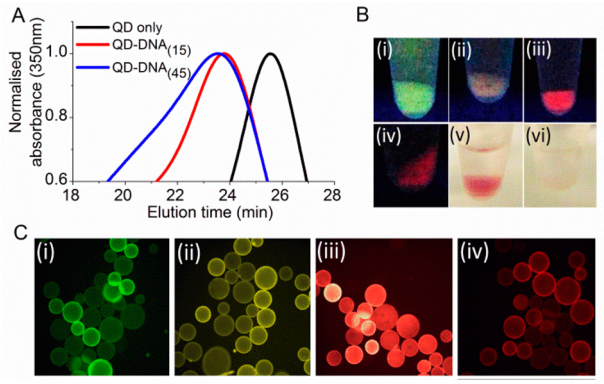

Figure 4. Coupling of 15mer and 45mer DNA to QDs of different colors and to GNP. (A) Chromatogram showing different retention time of QD (black), QD-DNA_{15mer} (red), and QD-DNA_{45mer} (blue). (B) Streptavidin agarose beads under UV lamp mixed with QD-DNA hybridized to complementary DNA labeled with biotin (i) QD550, (ii) QD580, (iii) QD610, and (iv) QD650. Streptavidin agarose beads under white light mixed with (v) GNP-DNA hybridized to complementary DNA labeled with biotin and (vi) GNP-DNA without biotin complement. (C) Fluorescent microscopy images of beads. (B) (i–iv) using Excitation filter: 450 ± 25 nm, Emission filter: 485 nm long pass. The images have been colored using *ImageJ*.

due to ligand desorption in dilute solutions over time.[45] To assess the stability of the DNA conjugated QDs a simple assay was designed. The QD-DNA-Bt sample was incubated with streptavidin agarose beads for 5 min followed by washing and imaging as before. These beads were stored at 4 °C, rewashed (3×), and reimaged. This procedure was repeated for 2 days. The beads remained fluorescent over time, hereby suggesting that the specific polymer strands that were coupled to DNA were stable on the QD in highly dilute solutions over days (Figure SI4). In addition, solutions of QD-DNA conjugates did not show any visible precipitates for >3 months upon storage at 4 °C in the dark.

2.6. Applicability of the Coupling Strategy to Other Nanoparticles. *2.6.1. Conjugation of DNA to QDs Emitting at Different Wavelengths.* To assess the scope of this strategy to different nanoparticles, 15mer ss-DNA_a was conjugated to QDs emitting at 550, 580, or 650 nm and purified by SEC. The presence of DNA on QD was tested with streptavidin agarose assay as described before. All four types of QD-DNA conjugates could be observed on the agarose beads after hybridization with biotin-labeled complementary DNA (Figure 4B(i–iv),C). In the absence of complementary biotinylated DNA, QD-DNA conjugates did not associate with the streptavidin agarose beads. This confirmed both the presence and the availability for hybridization of the conjugated DNA.

2.6.2. Conjugation of DNA to Gold Nanoparticles. The conjugation strategy was also tested on gold nanoparticles (GNP; see Experimental Section in Supporting Information). The protocol is similar to the conjugation of DNA onto QDs, but the molar excess of TCEP had to be increased to 50 000 with an excess of DNA/GNP of 50 in 1400 mM NaCl to obtain efficient conjugation of DNA on the GNP. Under these conditions, DNA could be conjugated on GNP and subsequently hybridized to a complementary sequence with biotin and attached to streptavidin agarose beads (Figure 4B (v,vi)).

DISCUSSION

QD-DNA conjugates have been synthesized by several methods in the past. For application of these conjugates, optimization of parameters such as reaction time, yield of conjugation, QY, and long-term stability is highly desirable.[46−48] Additionally, methods of purification can dramatically affect the yield and colloidal stability of the final product.[49] Here, we present a fast and efficient method to conjugate DNA to QDs and systematically study the role of different variables that influence the reaction. The QD-DNA conjugates obtained can have up to 12 DNA strands per QDs; they retain photophysical and structural properties of QD and DNA, respectively, and display long-term stability in solution.

We demonstrate that the thiols present on the polymer coating the QDs can be used for conjugation with DNA by addition of reducing agent TCEP. TCEP may have two modes of action for improving conjugation efficiency: first, by reducing the polymer dithiol bonds unlinked to the QD surface, making them reactive to DNA-maleimide; second, since phosphine-based molecules have affinity toward the surface of the QDs.[50,51] TCEP can replace the original thiolated polymer. At high excess of TCEP (10 000 times molar excess), sufficient numbers of thiols are replaced causing the polymers to desorb from the QD surface, thereby making the QDs unstable in solution. This is also supported by our results on conjugation of DNA to GNP. We find that the minimum concentration of TCEP required for conjugation of DNA on GNP is 125 times higher than for QD. Since thiols have higher affinity for GNP than QD, a stronger reducing environment is necessary to enhance coupling with the polymer.[52] Even at 50 000 excess of TCEP, yield of conjugation is lower in the case of GNP. Some reports of conjugation of DNA to GNP include long incubation steps of increasing salt concentrations;[53,54] which is unnecessary in this case. The conjugated DNA does not undergo conformational distortion as seen by its ability to hybridize with complementary sequence.[6,41] Further optimization to improve yields of DNA/GNP is being carried out.

We also find that salt dramatically improves the yield of this reaction from 3.0 ± 1.0 to 11.5 ± 4.0 DNA/QD. This improvement by a factor of 4 in conjugation yield in the presence of 1400 mM NaCl can be explained by electrostatics. High salt improves the solvation of zwitterions present on the polymer of QDs[55] along with reducing the electrostatic repulsions between DNA molecules in solution[30,56] and between the negatively charged thiolates on the QD and DNA, thereby favoring the reaction.

Generally, QD-DNA conjugates are purified by gel electrophoresis or gradient ultracentrifugation.[38,57] Both these approaches are time and labor intensive and multistep recovery of the final product is often difficult and with poor yields.[49] Most of these issues are overcome by SEC that provides a rapid (30 min) and automated method to purify QD-DNA conjugates from excess DNA. We also observed that retention time of QD-DNA conjugates were reproducibly sensitive to the yield of reactions, and could be used as a qualitative marker for successful conjugation.

For quantification of the number of DNA/QD, most approaches reported previously have measured either absorbance and fluorescence of a fluorophore on DNA, or FRET.[58,59] Owing to high yields of this reaction, we have been able to quantify DNA on QD by directly measuring absorbance of conjugated DNA at 260 nm. To verify the quantification, we

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also tested similar reactions with DNA labeled with different types of fluorophores (Cy5 and Cy5.5), and obtained similar results by both absorption and fluorescence spectroscopy. This approach is reliable, label free, and economically favorable.

Robust polymer design from our previous work[27] and systematic experiments in this work allowed us to find a balance between fast and optimum conjugation and long-term stability of conjugates. Although several thiolated ligands have been reported to quench the photoluminescence of QDs,[60,61] we do not observe change in QY upon ligand exchange and subsequent conjugation (<10%). Additionally, even post conjugation, a sufficient number of thiols continue to anchor the QD maintaining good colloidal stability. The conjugates can be stored in lyophilized form and readily resolubilized without loss of quantum yield, thereby facilitating long-term storage of conjugates.

CONCLUSIONS

This work presents a new approach to conjugate DNA to polymer coated QDs, and systematically studies parameters that influence the coupling reaction. We show that when optimized, this approach is fast, efficient, and generalizable to different nanoparticles.

QD-DNA conjugates presented in this work have several desirable properties such as (1) higher number of DNA strand per QD, up to twelve 15mer ss-DNA per QDs; (2) accessibility of the QD-DNA for hybridization; (3) high QY; and (4) long-term stability in solution. The QDs upon conjugation with DNA retain >90% of the QY, which is so far the best reported to our knowledge. QD-DNA conjugates have been obtained for QDs of four different colors and DNA has also been coupled to gold nanoparticles using the same surface chemistry and solubilization.

The conjugation strategy presented in this work has been also applied to peptides and proteins[39] and should help extend the use of functional nanoparticles in biomedical applications.

MATERIALS AND METHODS

All experimental details, formulas, and additional figures can be found in the Supporting Information.

ASSOCIATED CONTENT

Supporting Information

Materials and methods, sequences of DNA, effect of TCEP on conjugation of DNA to QD, characterization of QDs, detailed purification procedure and stepwise characterization of conjugation reaction, assessment of nonspecific interaction of DNA with QDs, quantification of DNA/QD and stability in dilute conditions. The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.bioconjchem.5b00221.

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Notes

The authors declare no competing financial interest.

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