CHEMBIOCHEM

Supporting Information

© Copyright Wiley-VCH Verlag GmbH & Co. KGaA, 69451 Weinheim, 2009

Supporting Information

for

DNA Monofunctionalization of Quantum Dots

Helen M. J. Carstairs, Kostas Lymperopoulos, Achillefs N. Kapanidis, Jonathan Bath, and Andrew J. Turberfield*

Experimental Section

Yield: QD655 (Invitrogen) were conjugated to 52 bp duplexes, purified and run on a 1% agarose TAE gel as described in the paper. A Pharos FX Plus Molecular Imager (Bio-Rad) was used to image the gel with 488 nm laser excitation and 640 nm band pass. Care was taken to ensure that the excitation strength did not result in the detector becoming saturated at any point. Quantity One analysis software (Bio-Rad) was used to measure the intensities of bands. The total intensity of the band was calculated by integrating the signal over the area of the band.

The intensity of the monofunctionalized band in the unpurified sample and the recovered monofunctionalized band were compared. Taking into account the relative volumes of the sample which had been loaded onto the gel and purified on the column the yield of monofunctionalized QDs was calculated to be 86%. This is a higher yield than the comparable step in gel purification, which is extraction from the gel. The overall yield from the starting materials could be improved by optimizing the conjugation ratios of QDs to DNA to ensure that monofunctionalized QDs were the majority product.

For presentational purposes the levels of gel images were rescaled linearly after analysis.

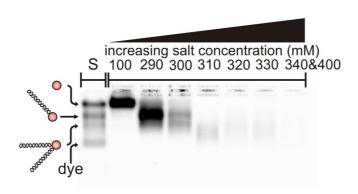


Figure S1. To assess the yield, a sample of QD655 and 52bp duplexes were conjugated, purified and run on a 1% agarose TAE gel. Comparison of the bands shows that the recovery of purified, monofunctionalized QDs in the 290 mm NaCl fraction is 86%.

Purification of QDs functionalized with ssDNA: The method described in the paper can also be used to purify QDs decorated with single-stranded DNA. Figure S2 shows the fractions obtained from a sample of QD655 conjugated to a 74 base oligomer. A longer single strand was used in order to improve the resolution of bands in the agarose gel. The monofunctionalized QDs are recovered in the 270 mm NaCl fraction.

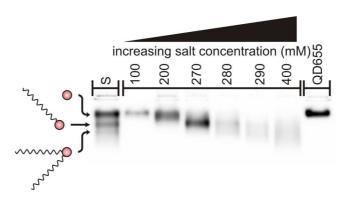


Figure S2. Purification of ssDNA-QD conjugates by ion exchange. The 1% agarose, TAE gel shows the heterogeneous sample of DNA-QD conjugates (S) applied to a DEAE Sepharose spin column and the concentrated eluted fractions. The ionic concentration of the elution buffer is shown above each lane. The gel was run at room temperature at 3.5 V/cm for 1.5 h. QDs monofunctionalized with ssDNA are recovered at 270 mM NaCl. A 74 base strand was used to improve the resolution in the agarose gel of bands containing functionalized QDs.

DNA Sequences

For the experiment shown in Figures 1, 2 & S1 the following duplex was used.

- 52 base pair duplex:
- 5'-CTAGCCGCCCGATCTACGCCCGATCTACGCCCGATCTACGCCCGATCCTTCC-3'

For the experiment shown in Figure 1b&c, the additional following duplexes were used.

- 19 base pair duplex:
- 5'-biotin GGAAGGATCGGGCGGCTAG-3'
- 5'-CTAGCCGCCCGATCCTTCC-3'
- 30 base pair duplex:
- 5'-biotin GGAAGGATCGGGCGTAGATCGGGCGGCTAG-3'
- 5'-CTAGCCGCCCGATCTACGCCCGATCCTTCC-3'
- 41 base pair duplex:
- 5'-biotin GGAAGGATCGGGCGTAGATCGGGCGTAGATCGGGCGCTAG-3'
- 5'-CTAGCCGCCCGATCTACGCCCGATCCTTCC-3'
- 63 base pair duplex:
- 5'-biotin GGAAGGATCGGGCGTAGATCGATAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGATAGATCGGGCGTAGATCGATAGATCGGGCGTAGATCGATAGATCGGGCGTAGATCGATAGATCGATAGATCGGGCGTAGATCGATAGATCGGGCGGTAGATCGATAGATCGGGCGGATAGATCGGGCGTAGATCGATAGATCGGGCGGATAGATCGGGCGGTAGATCGATAGATCGGGCGGATAGATCGGGCGTAGATCAGATCGGGCGATAGATCGGGCGGATAGATCGGATCGATAGATCGGGCGGATAGATCGA
- 74 base pair duplex:
- 5'-biotin GGAAGGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAG-3'

For the experiment shown in Figure 3, the following duplexes with 20-nt sticky ends were used.

QD655 linker:

(29 bases):

5'-biotin ACGTCCTAGCTCGGCCTTCATTAATCGAG-3'

(49 bases):

5'-TTCCGTCATGCAGAGATCTCCTCGATTAATGAAGGCCGAGCTAGGA CGT-3'

QD565 linker:

(52 bases):

5'-biotin TTATCTATATAGCGAATTCCTGCCTTTACTGACTTACGGCATCGCGTACTCT-3'

(72 bases):

5'-GAGATCTCTGCATGACGGAAAGAGTACGCGATGCCGTAAGTCAGTAAAGGCAGG-AATTCG-3'

For the experiment shown in supporting information Figure S2, the following strand was used.

(74 bases):

5'-biotin GGAAGGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAGATCGGGCGTAG-3'