CHARACTERIZATION OF QUANTUM DOTS CONJUGATES WITH ANTIBODIES BY CAPILLARY ELECTROPHORESIS

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Preparation

1st step: preparation of hydrogen telluride

$4\ NaBH_4 + 2\ Te + 7\ H_20 \rightarrow 2\ NaHTe + Na_2B_4O_7 + 14\ H_2$

2nd step: quantum dots formation

$CdCl_2$ + NaHTe + MPA or MA + heating

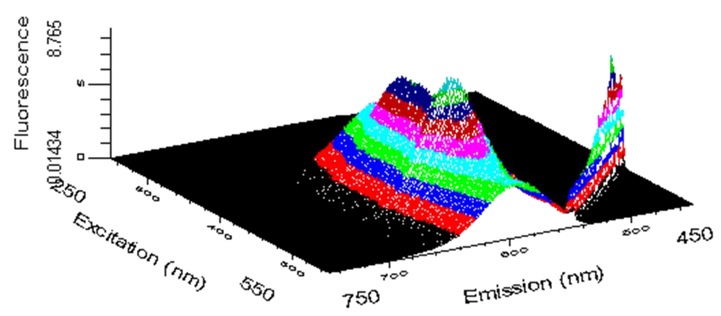
MPA: 3-mercaptopropionic acid: $HS\text{-}CH_2\text{-}CH_2\text{-}COOH$
MA: 2-mercaptoethylamin : $HS\text{-}CH_2\text{-}CH_2\text{-}NH_2$

3rd step: coating

CdS: CdTe + $CdCl_2$ + Na_2S + MPA or MA + heating

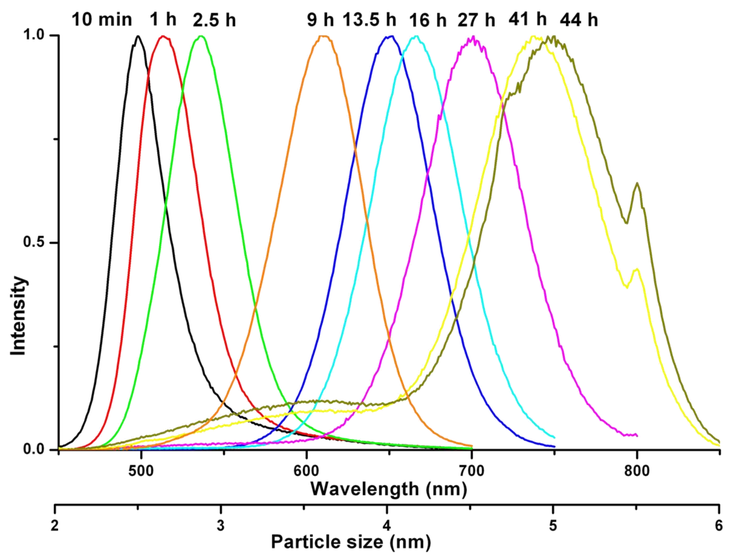
ZnS: CdTe/CdS + $ZnCl_2$ + Na_2S + MPA or MA + heating

Luminescence spectra



- wide excitation spectra with maximum at 469 nm
- narrow emission spectra with maximum at 600 nm
- bandwidth 58 nm at half height

Emission spectra



Quantum yield

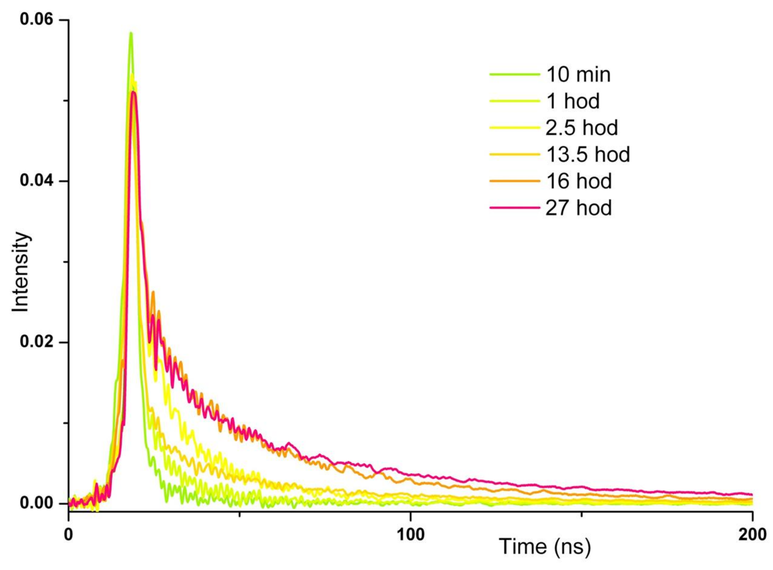
Coating	Ligand	
	MPA	MA
-	9.66	21.08
CdS	18.18	25.54
CdS/ZnS	9.08	27.96

$$QY_{QD} = QY_S \frac{A_S I_{QD}}{I_S A_{QD}}$$

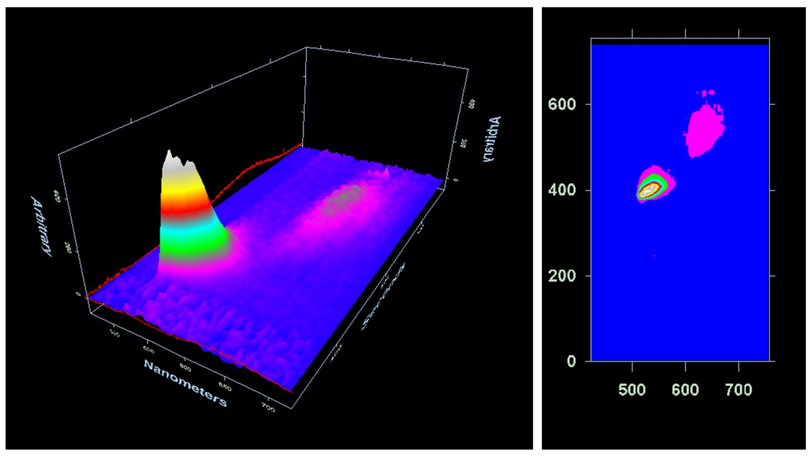
S – standard
(fluorescein QY = 0.90*)

* Demas J.N, Crosby G.A., J Phys. Chem., 75, 991 (1971)

Luminescence lifetimes

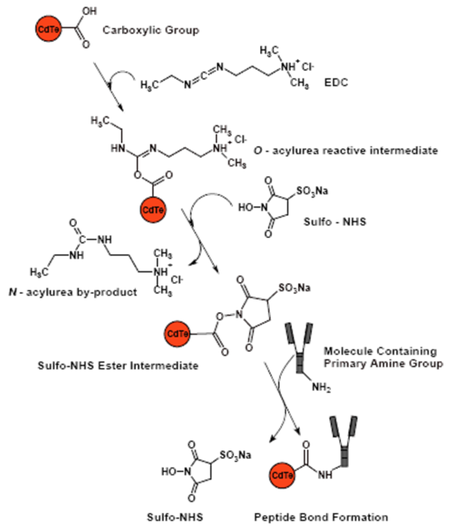


Electrophoresis in replaceable sieving media



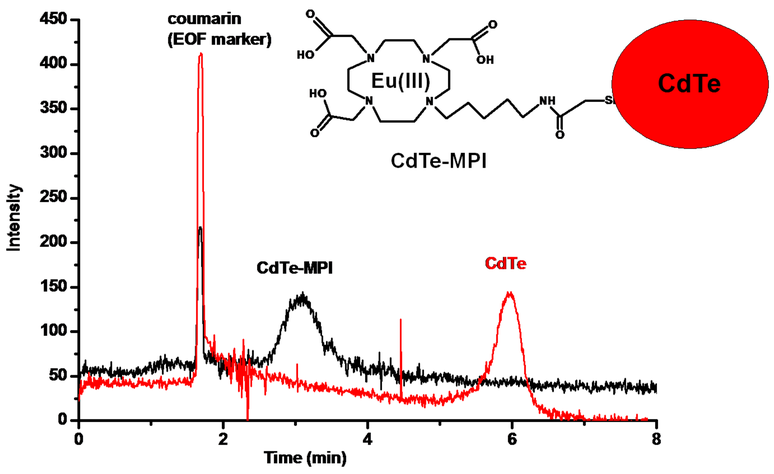
PVA coated capillary 20/30 cm, i.d. 75mm, separation buffer 3% LPA 10 MDa in 50 mM TRIS/TAPS buffer, pH = 9, QD 2.8 and 3.7 nm (525 and 610 nm 1:1), injection time 10 s, separation voltage 3 kV

Conjugation using zero-cross linkers



- formation of peptide bond
- carboxylic group of MPA on the QD surface
- amino group from antibody
- catalyzers: EDC (carbodiimide) HNS (succinimide)

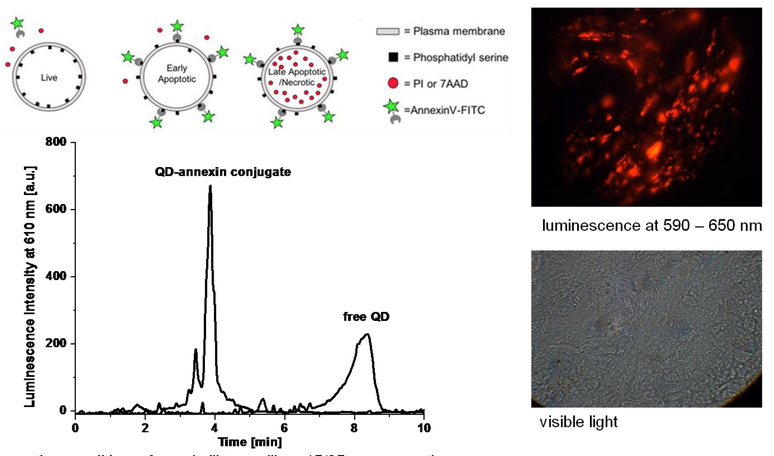
Capillary electrophoresis of conjugates



Separation conditions: fussed silica capillary 15/25cm, separation voltage 6 kV, injection time 15 s, separation buffer 100 mM TRIS/TAPS, pH = 8.3

Capillary electrophoresis of conjugates

Apoptotic cells of mouse duodenum labeled by QD-annexin conjugate



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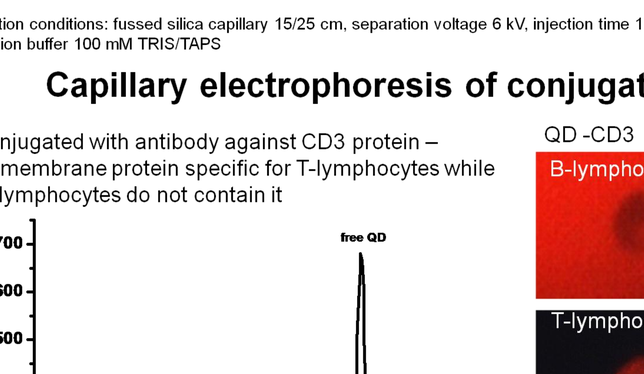
Capillary electrophoresis of conjugates



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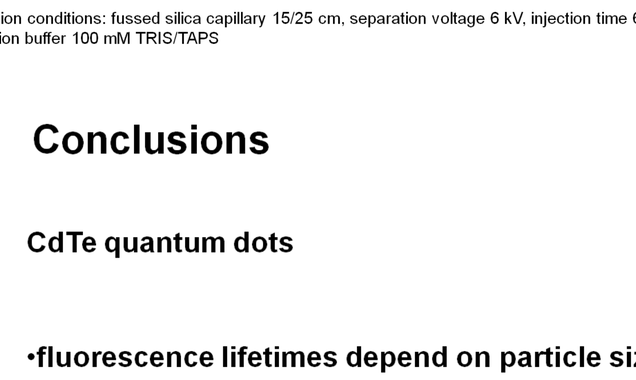
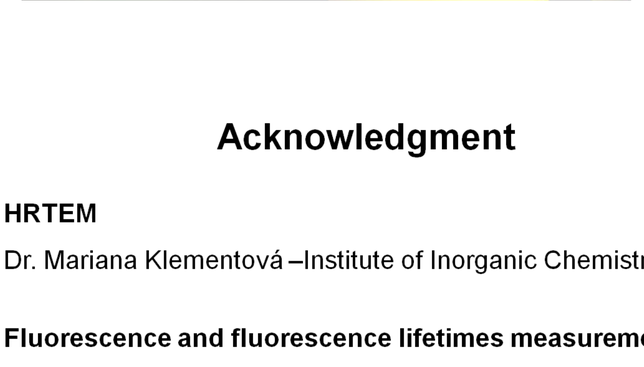
Capillary electrophoresis of conjugates



Separation conditions: fussed silica capillary 15/25 cm, separation voltage 6 kV, injection time 6s, separation buffer 100 mM TRIS/TAPS

Conjugation of magnetic beads with Anti-ovalbumin



Capillary electrophoresis of conjugates

- conjugated with antibody against CD3 protein – a membrane protein specific for T-lymphocytes while B-lymphocytes do not contain it

Separation conditions: fussed silica capillary 12/20 cm, separation voltage 6 kV, injection time 15 s, separation buffer 50 mM CAPS, pH = 11.2

Conclusions

CdTe quantum dots

- **fluorescence lifetimes depend on particle size**
- **small selectivity of size separation using sieving media**
- **conjugation with antibody and macrocyclic ligand**
- **capillary electrophoresis luminescence immunoassay**
- **selective cell labeling**

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Fluorescence and fluorescence lifetimes measurement

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