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Micropatterning of quantum dots monolayer for quantitative FRET-based imaging

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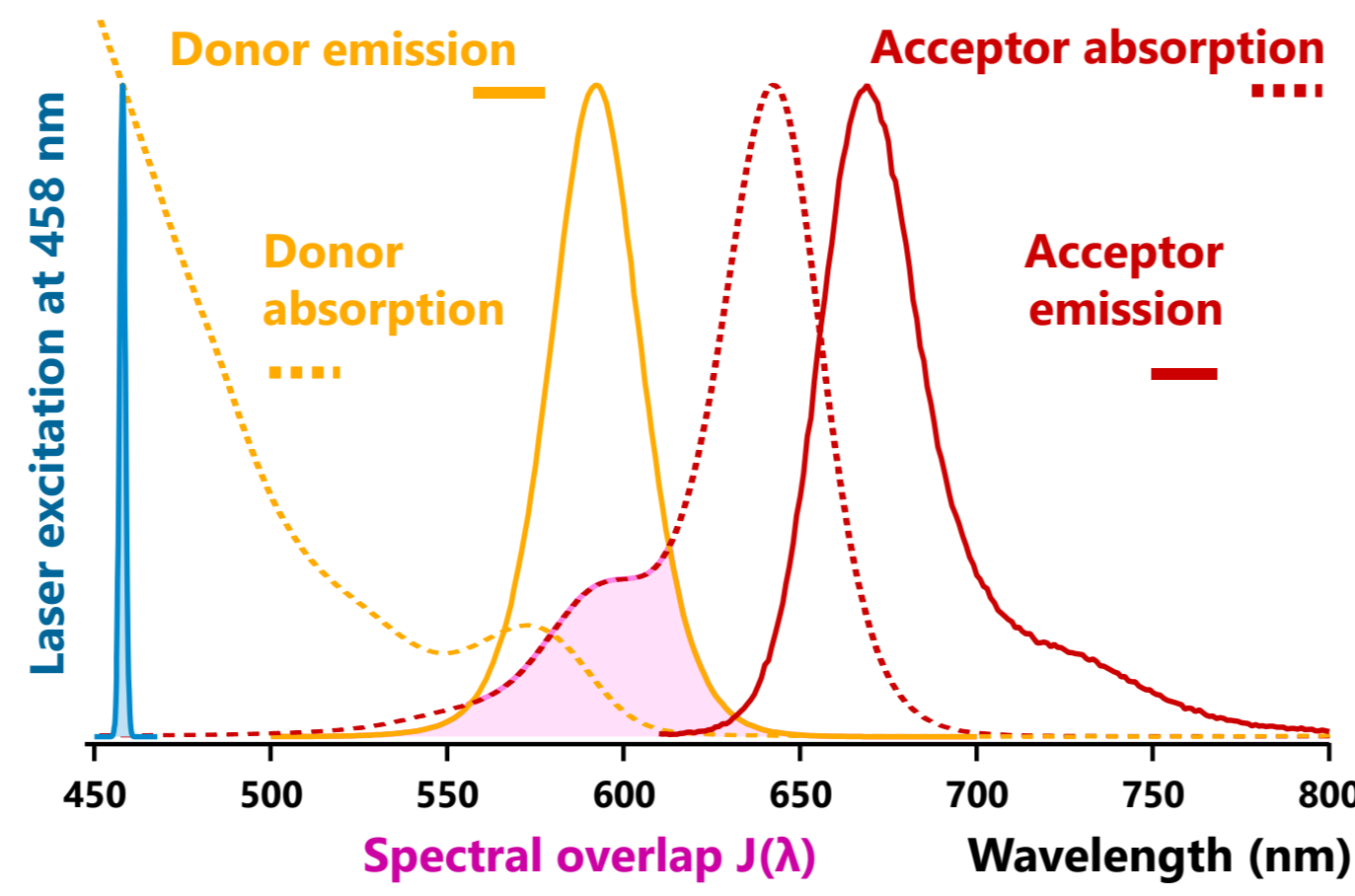
Theoretical & Experimental Basis

Förster Resonance Energy Transfer (FRET) Efficiency:

$$E = \frac{R_0^6}{r^6 + R_0^6} = 1 - \frac{F_{DA}}{F_D}$$

$$R_0^6 = 8,79 \cdot 10^{-5} \cdot \frac{\kappa^2}{n^4} \cdot QY_D \cdot J(\lambda)$$

F_{DA} : Donor emission with Acceptor presence
 F_D : Donor emission without Acceptor presence
 R_0^6 : Distance of 50% transfer efficiency
 κ^2 : Orientation factor
 n : Refractive index
 QY_D : Quantum Yield of the Donor

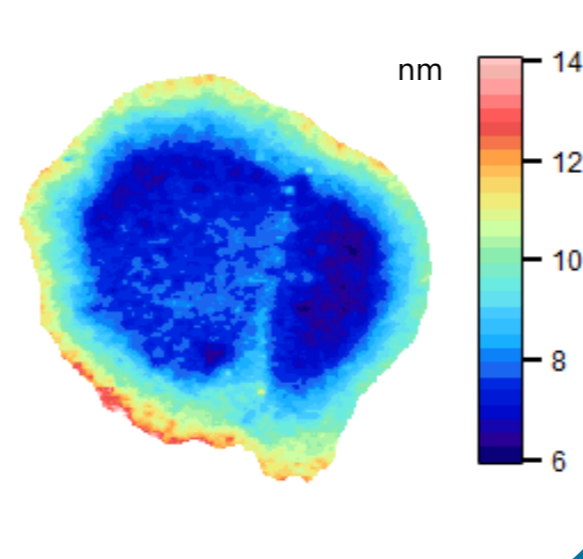
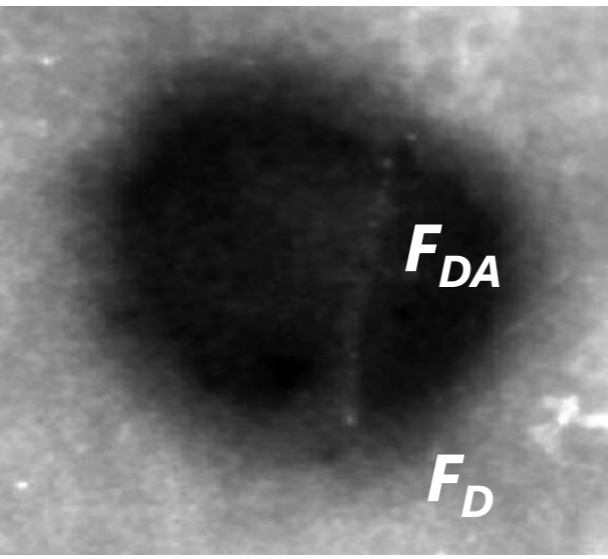
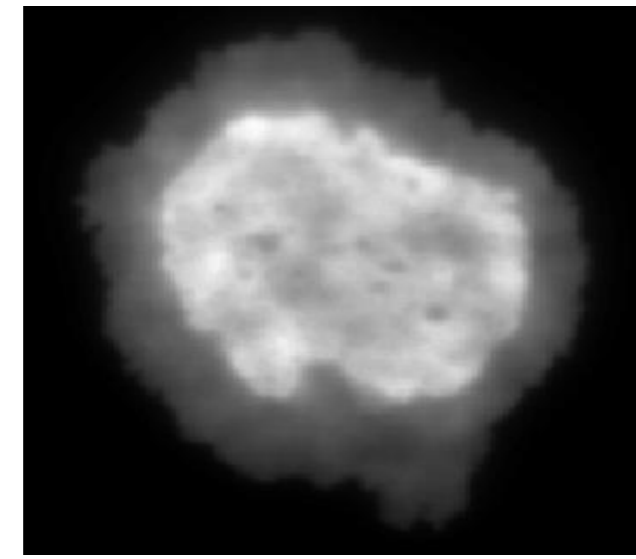
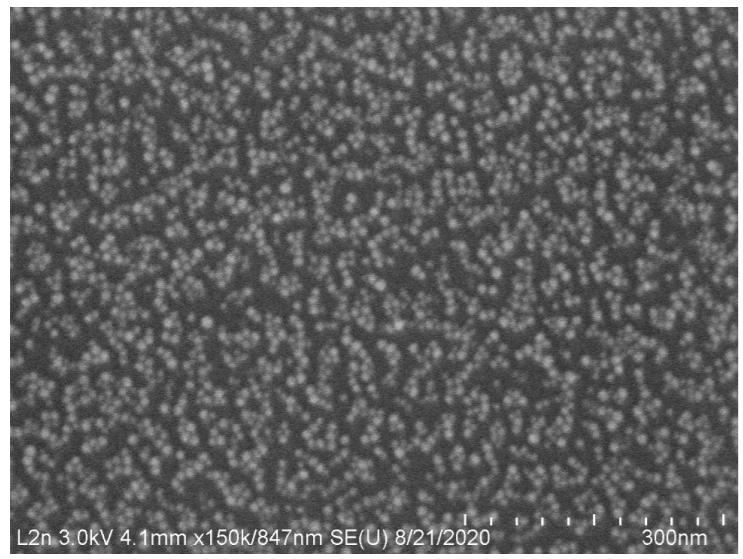


Donors monolayer:
up to 4000 QDs/μm²

Acceptor
FRET signal

Donor
Quenching

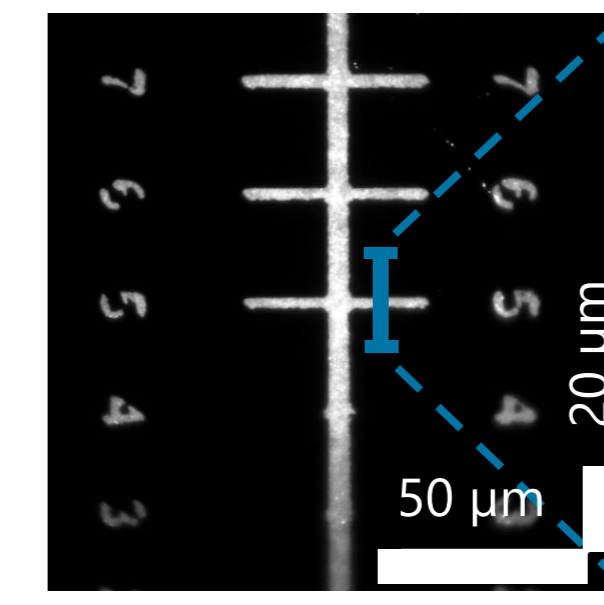
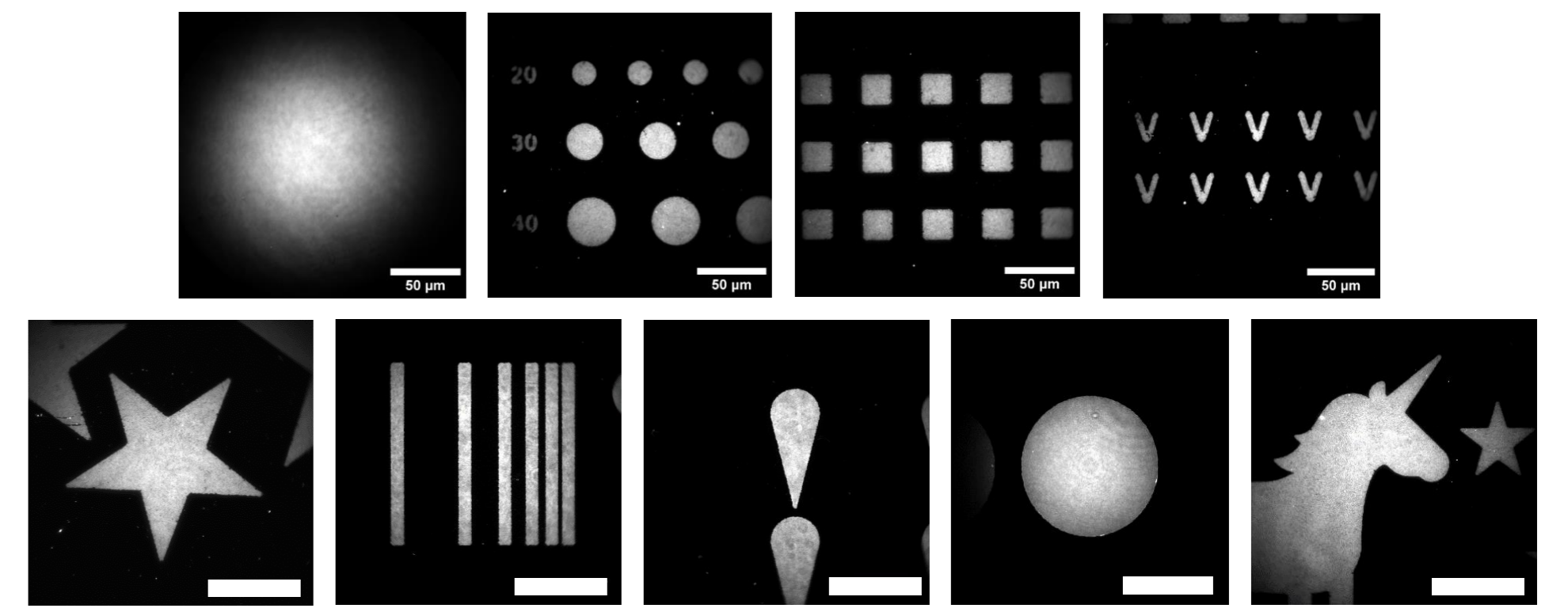
Computed
Height r



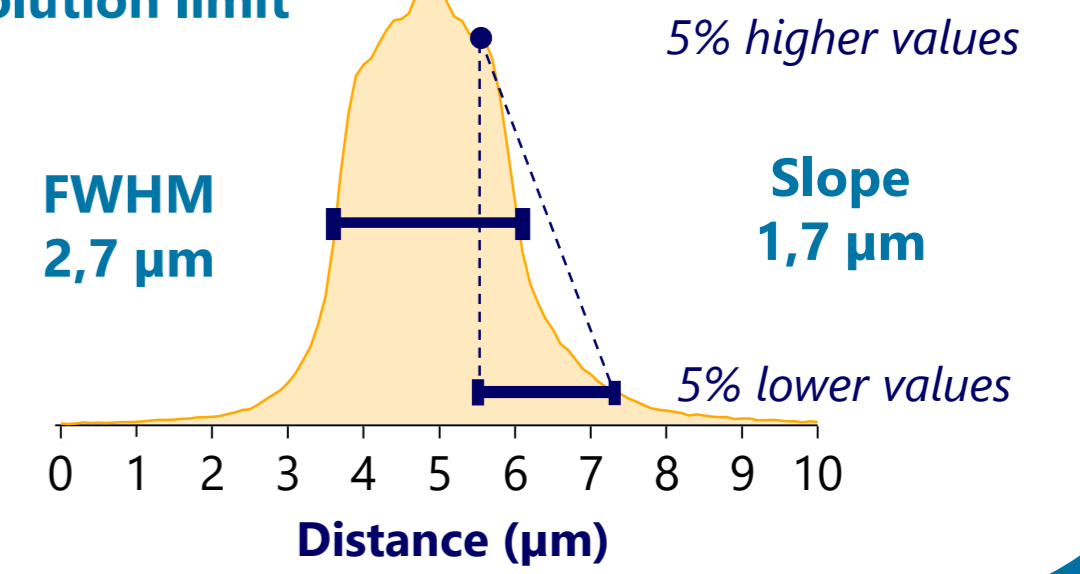
Micropatterning TIRF characterization

Easy design using Alveole's PRIMO setup:
Any digital image (1824×1140 px) can be
projected onto the glass substrate

×10 Objective ≈ 0,5 μm/px
S1813 G2 photoresist
min UV dose = 1,6 mJ/mm²



Resolution limit



Micropatterning Protocol

Piranha cleaning for surface
activation

Spin coating of S1813 G2
4000 RPM ; 3000 ACC ; 30 s

Softbake on hotplate
115°C ; 1 min



UV illumination
With Alveole's PRIMO setup

Bake on hotplate
110°C ; 10 min

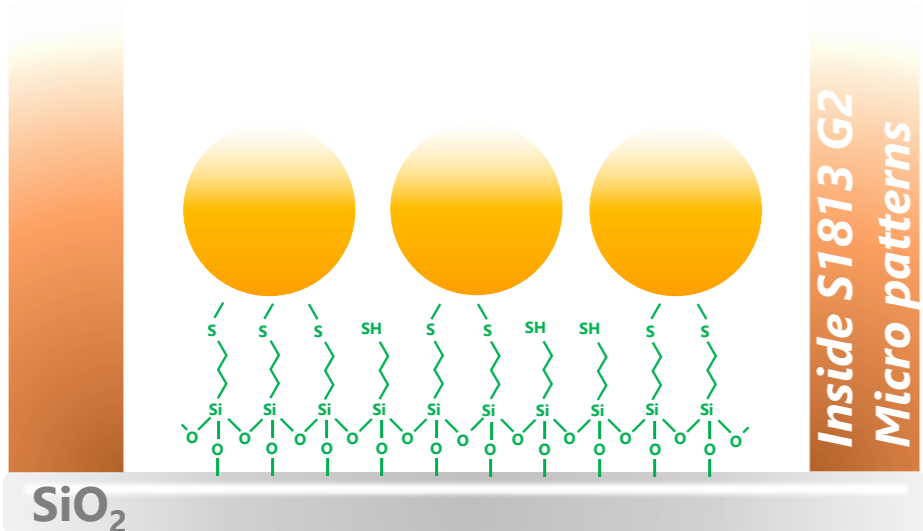


Development
MF26-A ≈ 1 min + Plasma-Air
cleaning 2 min

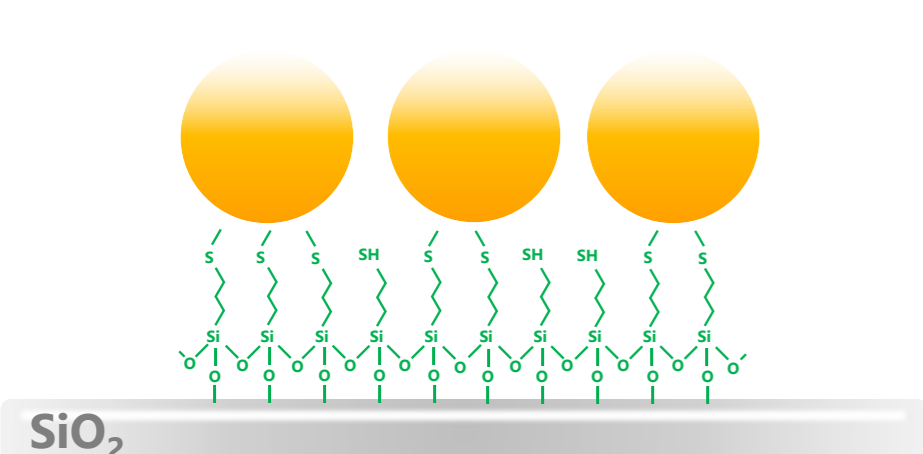


Silanzation with MPTMS
[2-5] % in anhydrous toluene
Overnight

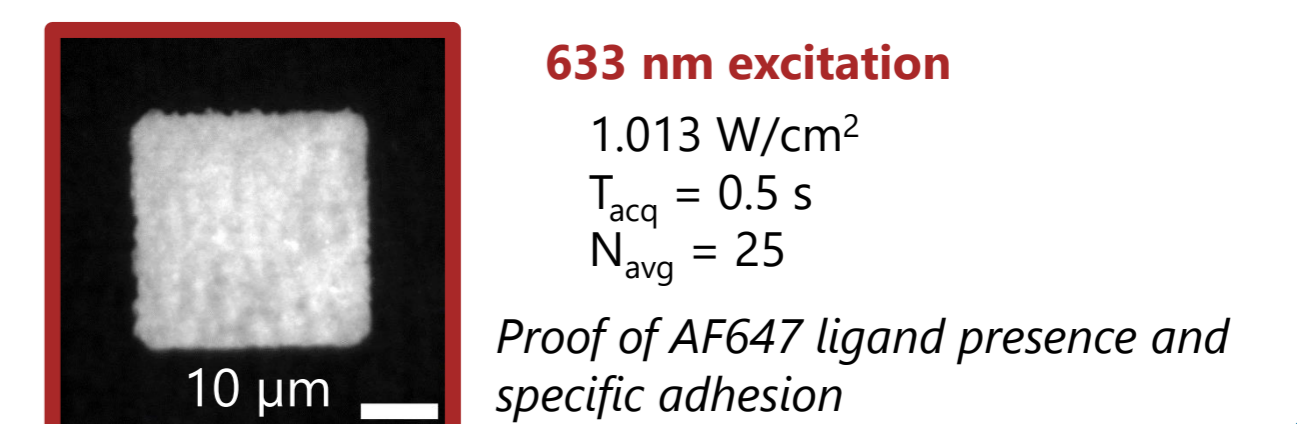
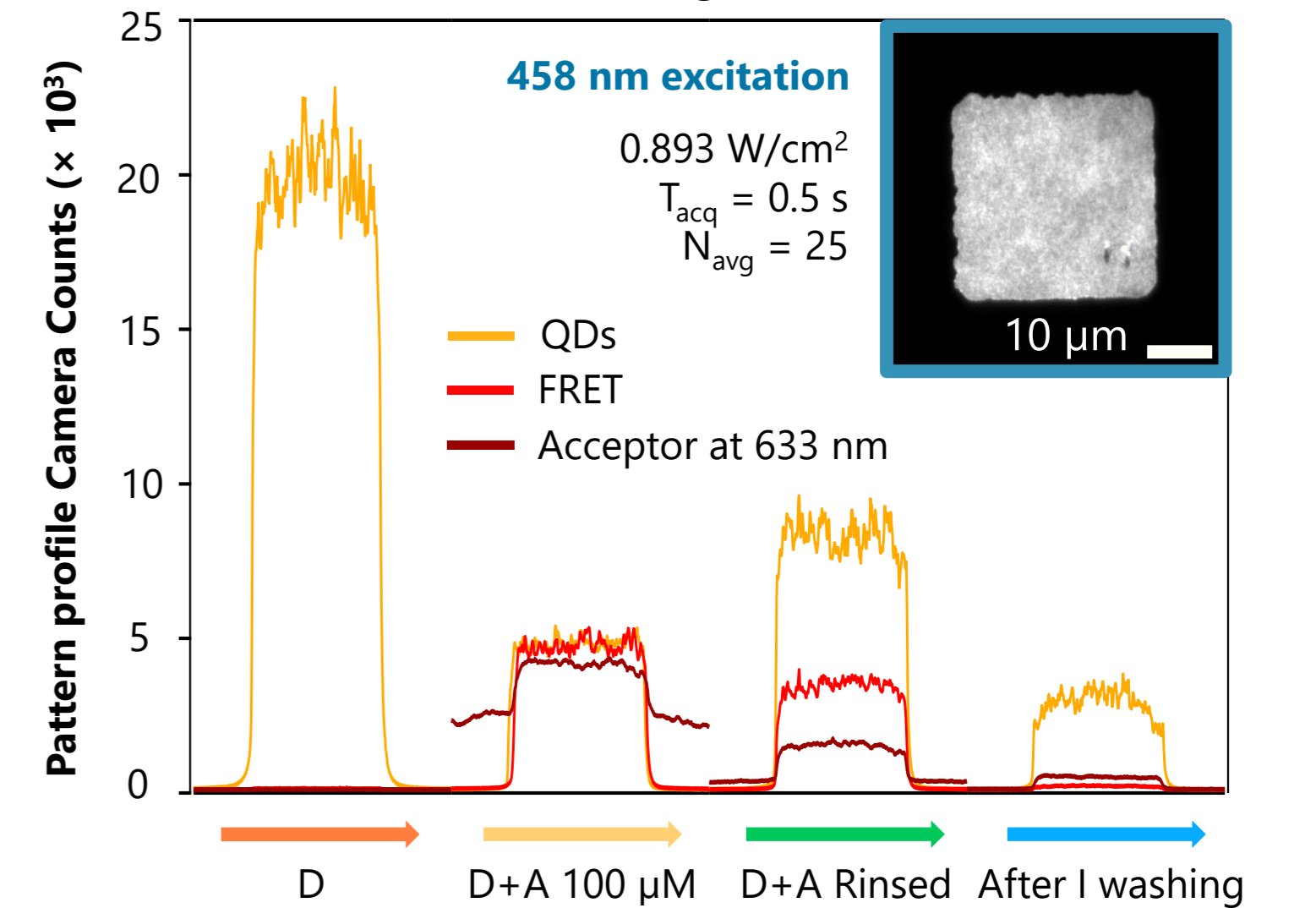
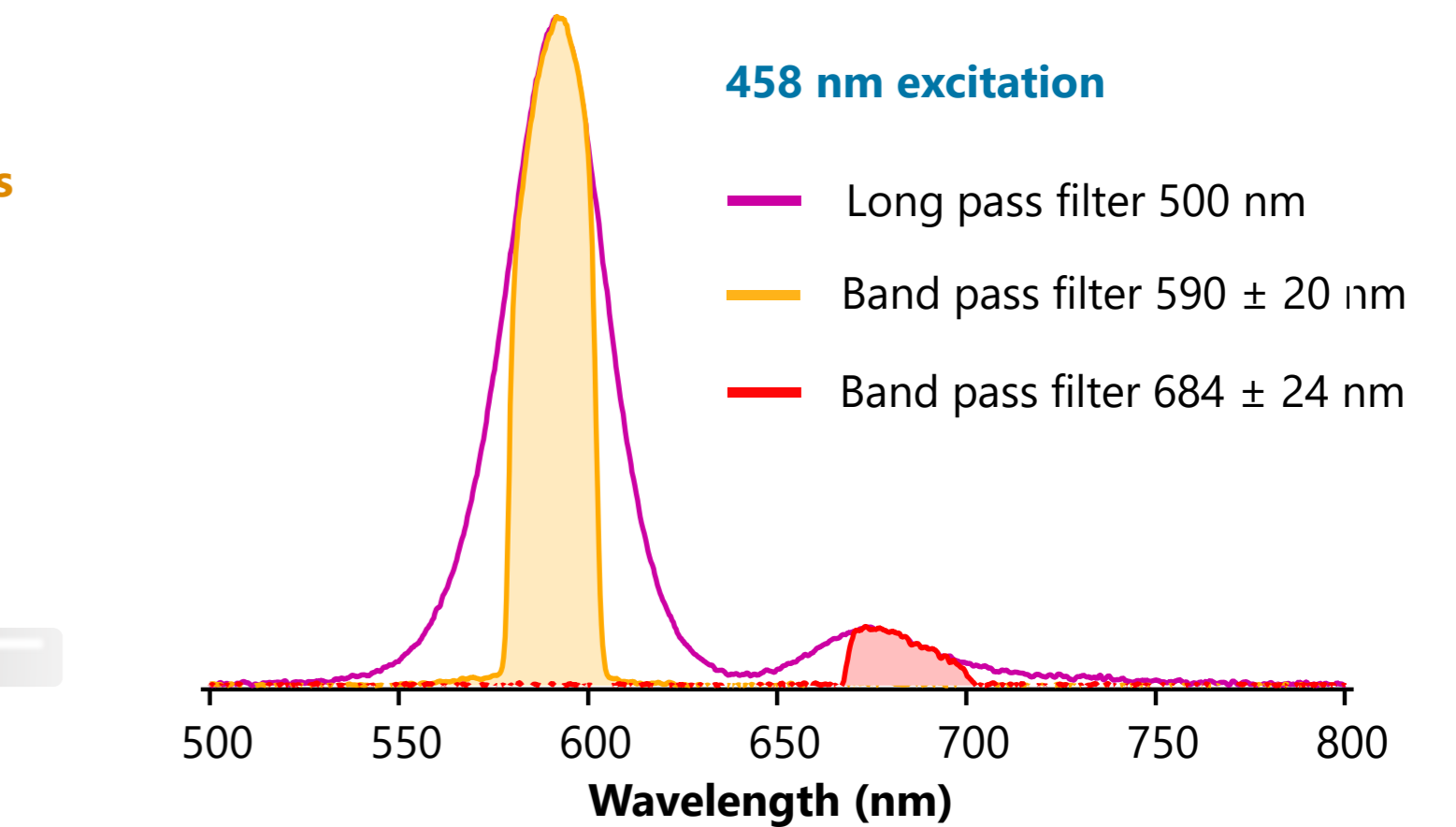
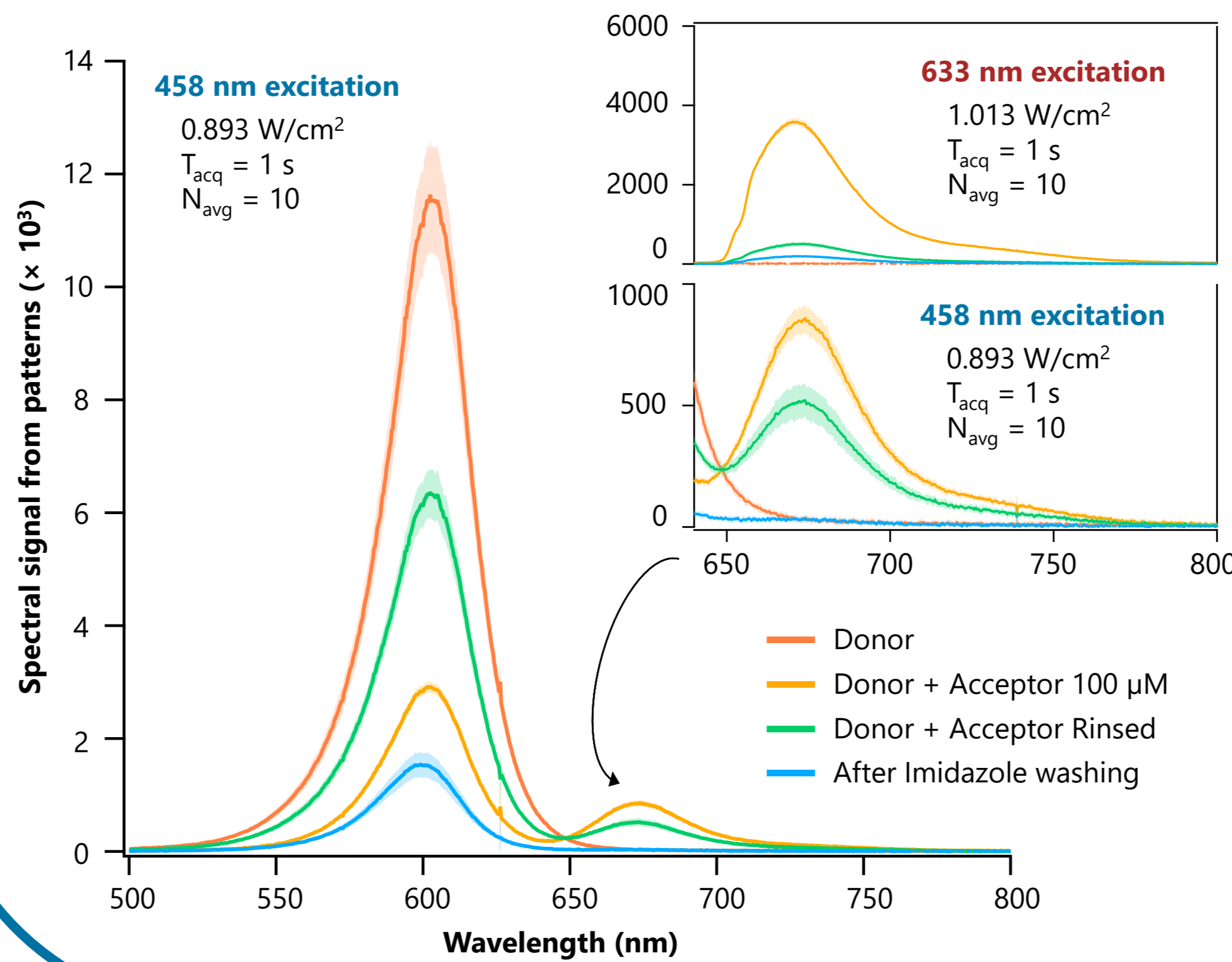
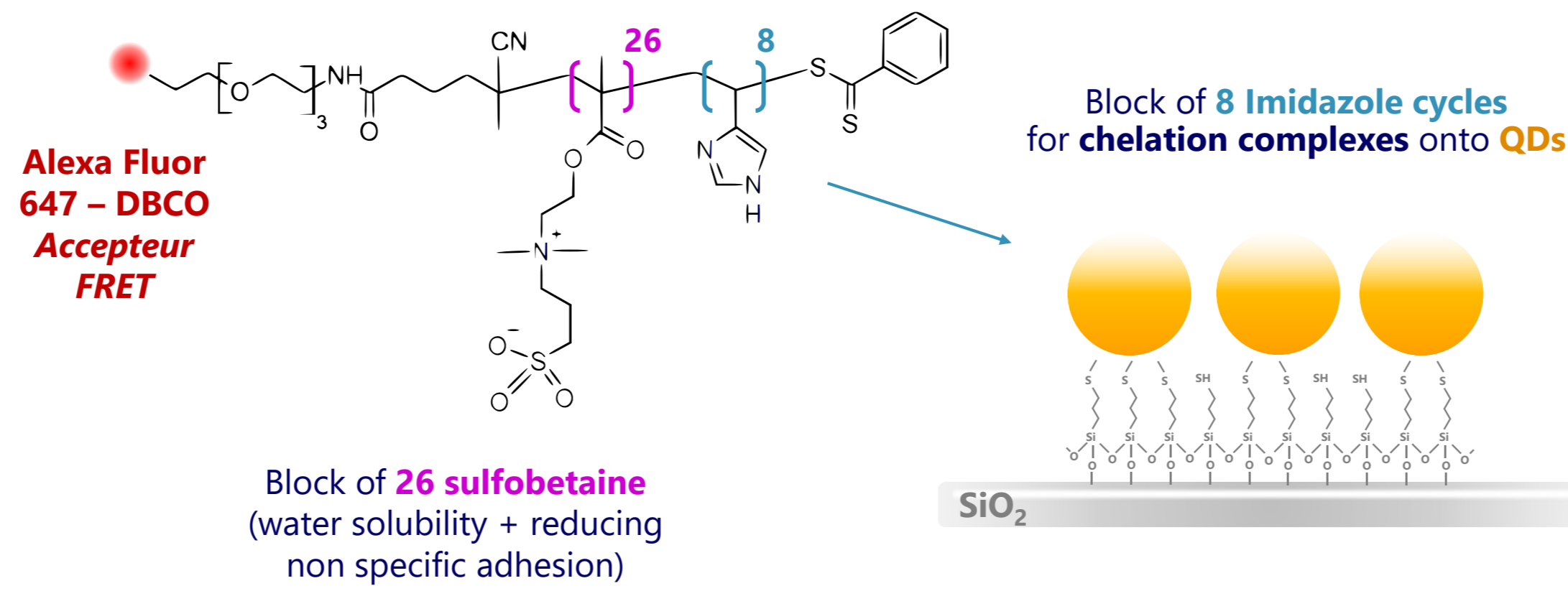
Quantum dots grafting
through disulphuric bridge
[0.01-24] h in μM solution



Lift-off in acetone
with 5 min of sonication



Proof of concept for proteins of adhesion scaffold



Cell micropatterning demonstration

RGD Ligand: peptidic sequence GRGDSPKGAGAKG-HHHHHHHH

