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A BODIPY-based fluorescent sensor for amino acids bearing thiol

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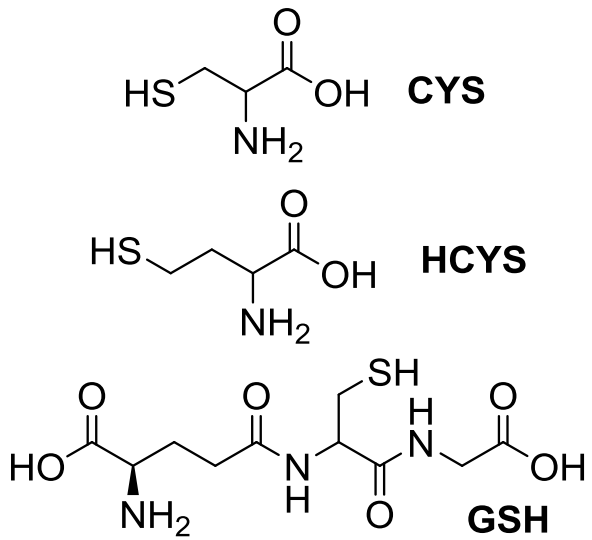
A BODIPY-based fluorescent sensor for amino acids bearing thiol

1. Introduction
2. Objective
3. Molecular Design
4. Synthesis
5. Photophysics
6. Sensing
7. Conclusions

Sensing of biomolecules

- Visualization of biochemical events
- Early detection of diseases
- Unravel mechanism of physiological anomalies

One of the most seeking targets are amino acids (AA) like those bearing thiols:
Cysteine (CYS), Homocysteine (HCYS) and Glutathione (GSH)



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Fluorescent chemosensors

Ligth emission as a tool to transform chemical information (AA presence or concentration) into analitical information (fluorescence signal)

Design:

- High afinity for the target analyte
- Induce spectroscopic changes into the dye photophyscs
- Upon target binding, reverse the aforementioned change into the spectroscopic signatures

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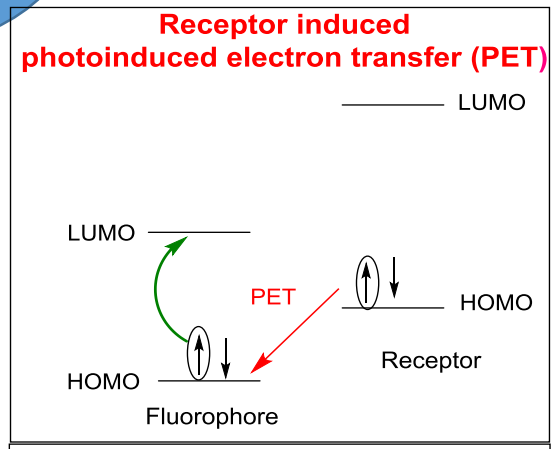
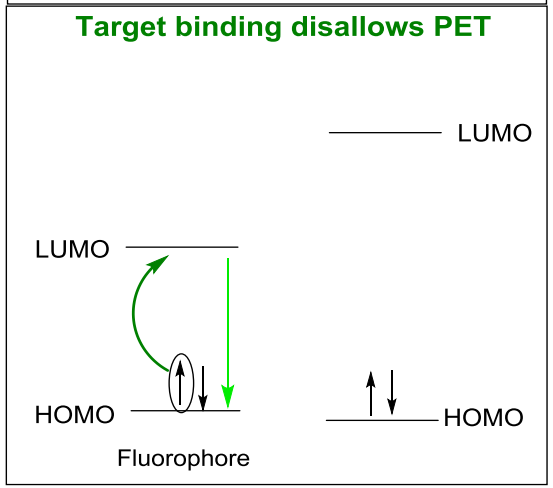
Main strategies to develop fluorescent chemosensors:

ON-OFF switches

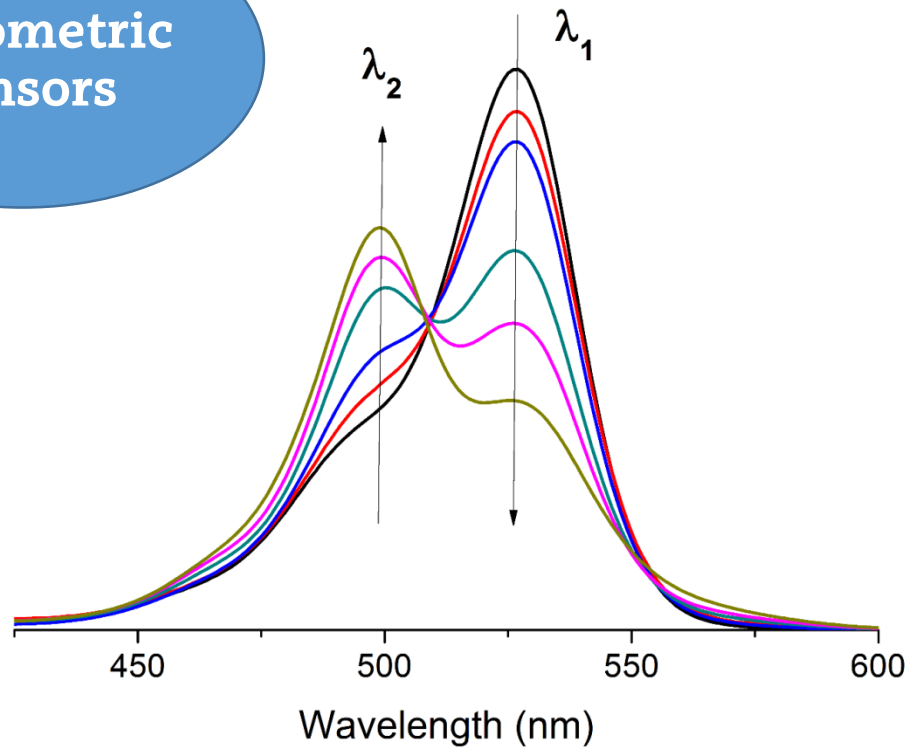
OFF state



ON state



Ratiometric sensors



Binding of the target promotes spectral splitting
Different channels for detection (signal loss at λ_1 or signal growing at λ_2)

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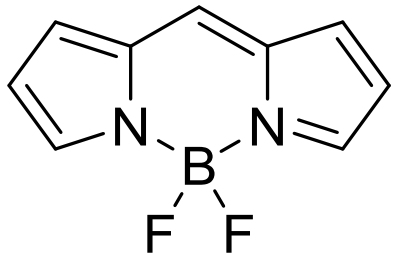
BODIPY chromophore suitable molecular scaffold as fluorophore:



4,4-difluoroboron-3a,4a-diaza-s-indacene

BODIPY

- Chemically stable
- High photo and termal stability
- Excellent photophysical signatures (strong absorption and fluorescent bands)
- **Outstanding chemical versatility (multitude of synthetic routes applied)**
- Fine modulation of the spectroscopic properties by tailoring the molecular structure

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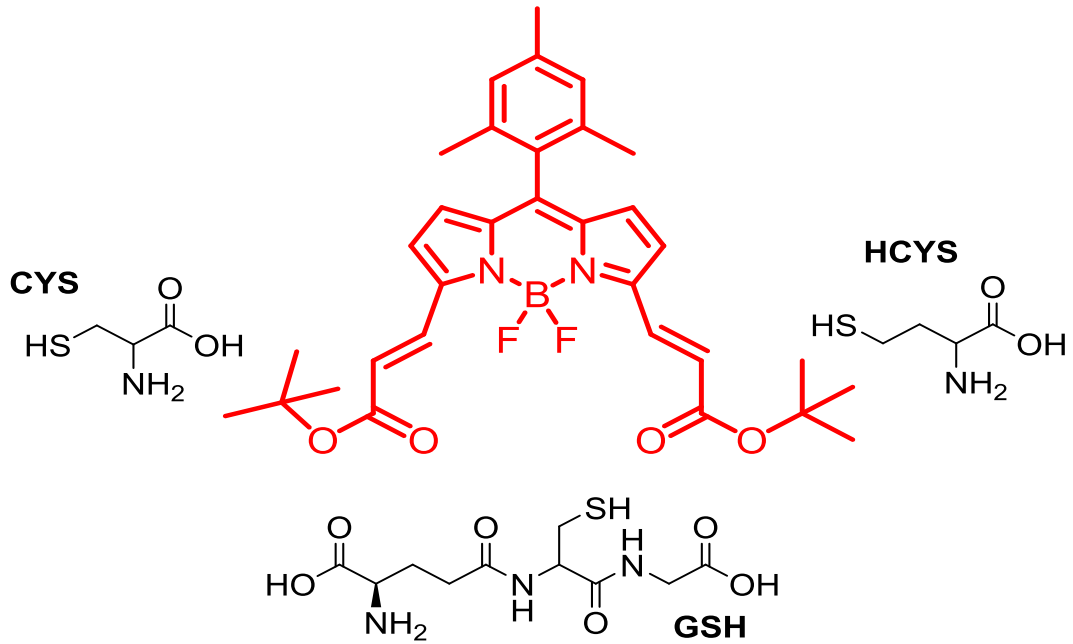
Test the viability of the designed and synthesized BODIPY based ratiometric chemosensor for the detection of CYS, HCYS and GSH



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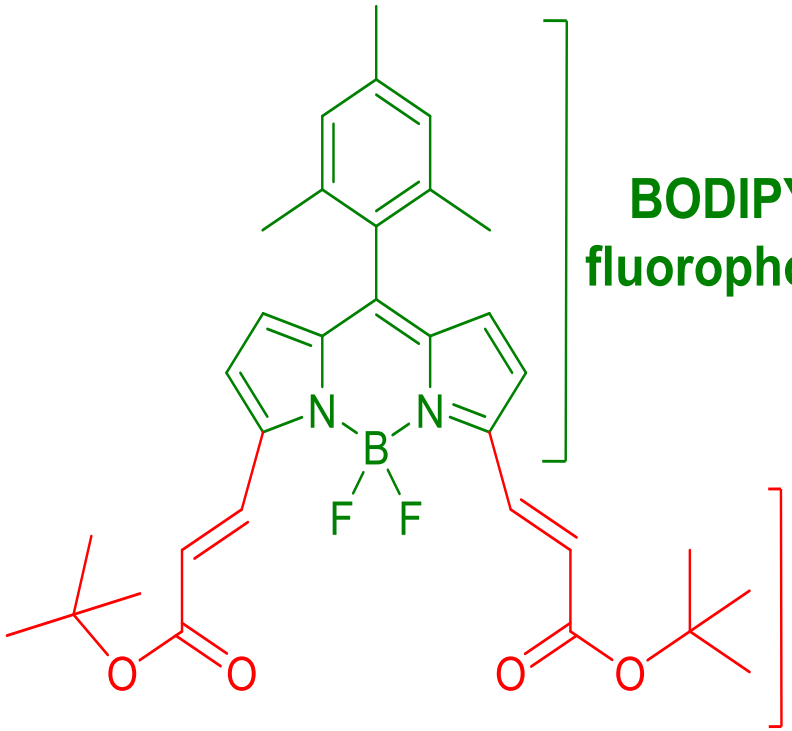

BODIPY fluorophore

- Rigidized molecular structure to ensure high fluorescent response
- Modulable spectroscopic signatures tailoring the molecular structure by the subtitution pattern at the dipyrrin core

Amino acid receptor

- Reaction sites (vinyl and carbonyl) for binding of the AA
- Aromatic moiety at the α-pyrrolic position to extent the chromophoric π-system
- Shift of the emission band to the red edge to increase the ligth penetration into tissues
- Binding of the AA should alter the extention of the conjugated π-system and the photophysics

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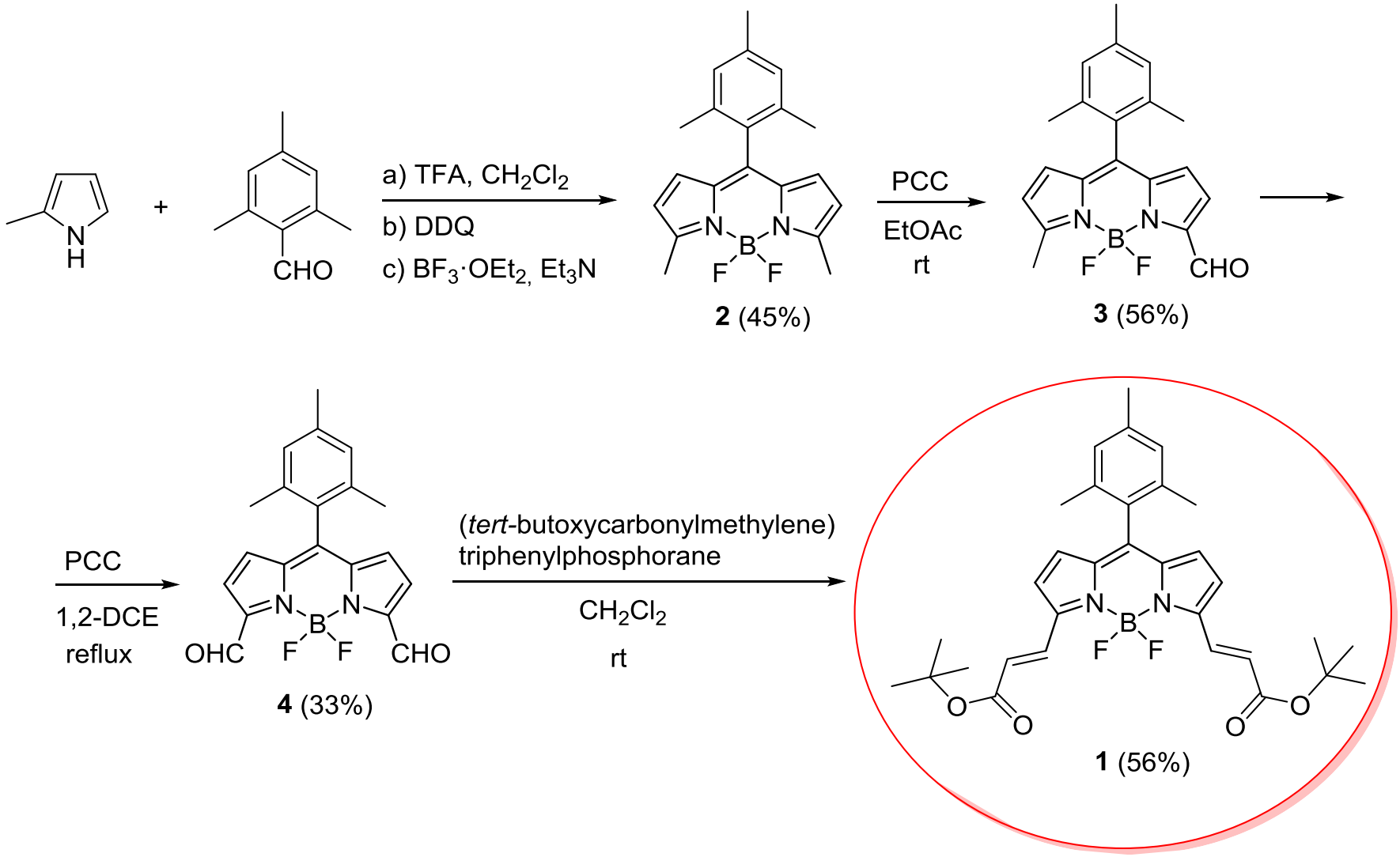

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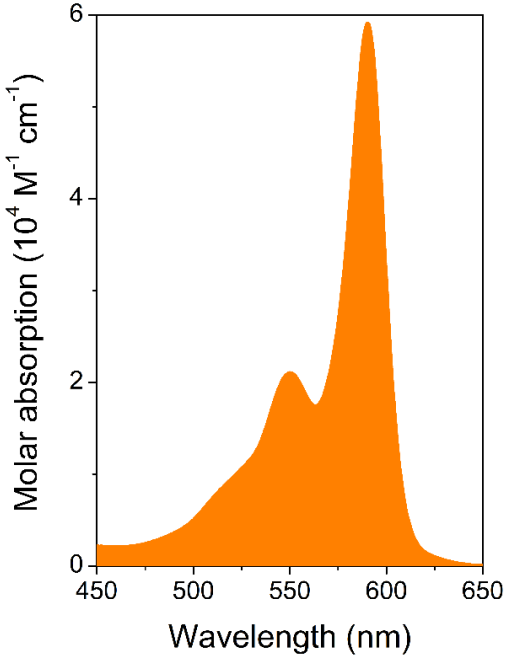
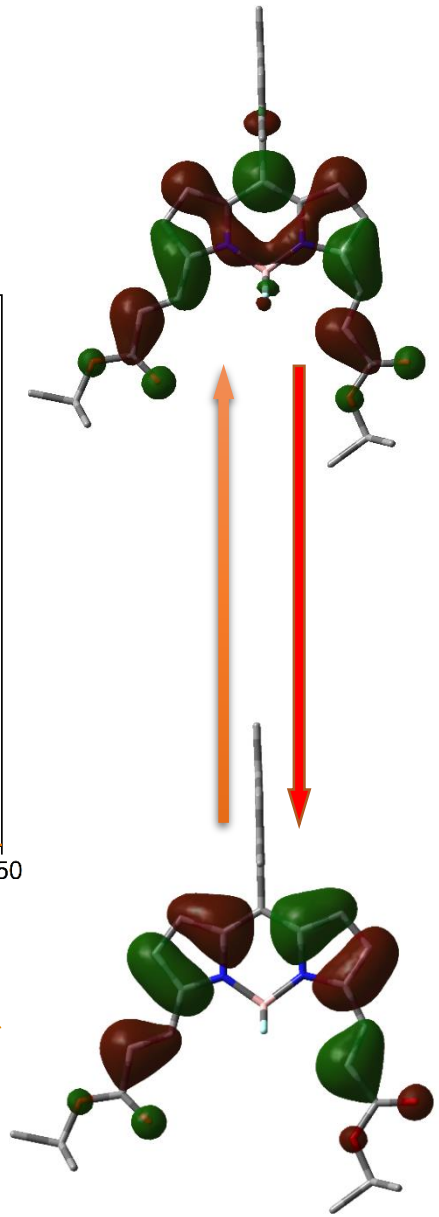
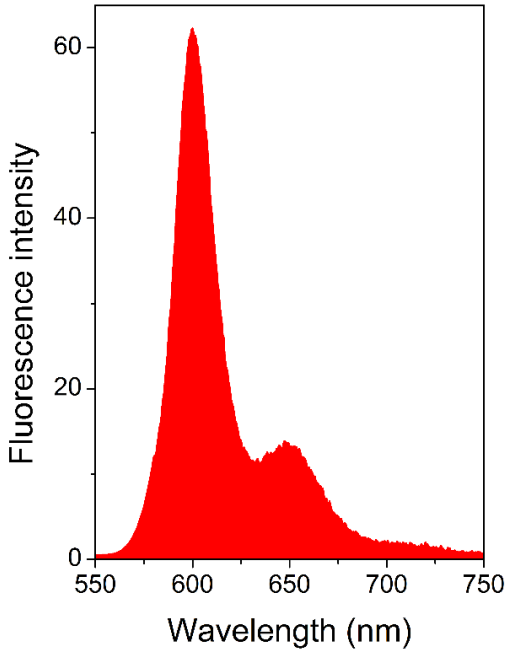


λ_{ab} = 589.5 nm
ε_{max} = 59000 $M^{-1}cm^{-1}$
(ethanol)





λ_{fl} = 598.5 nm
ϕ_{fl} = 62%
τ_{fl} = 5.42 ns
(ethanol)

The constrained and ortogonal 8-mesityl ensures rigid structure and avoids non-radiative relaxations.

The α-unsaturated diesters induce a pronounced bathochromic shift

High fluorescence at the red edge of the visible

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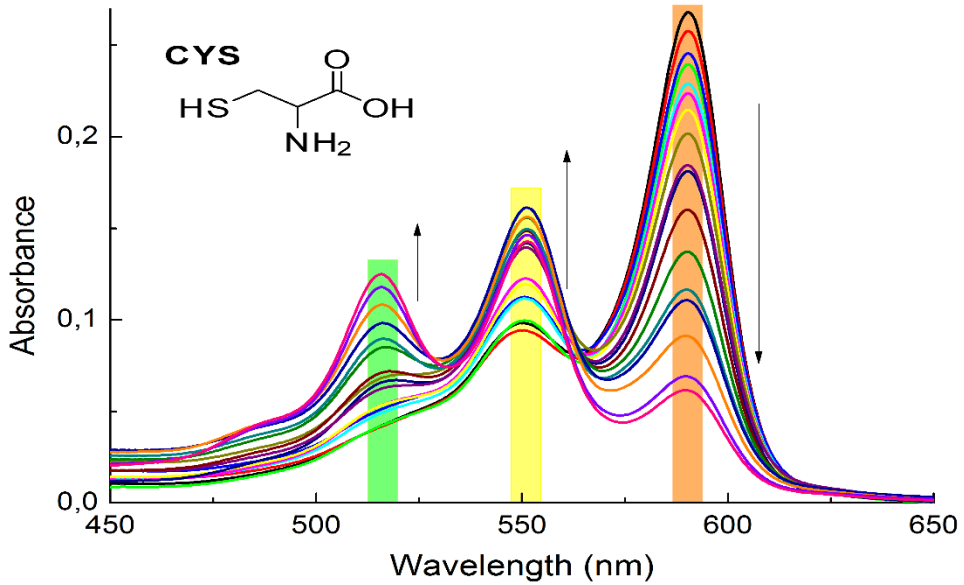
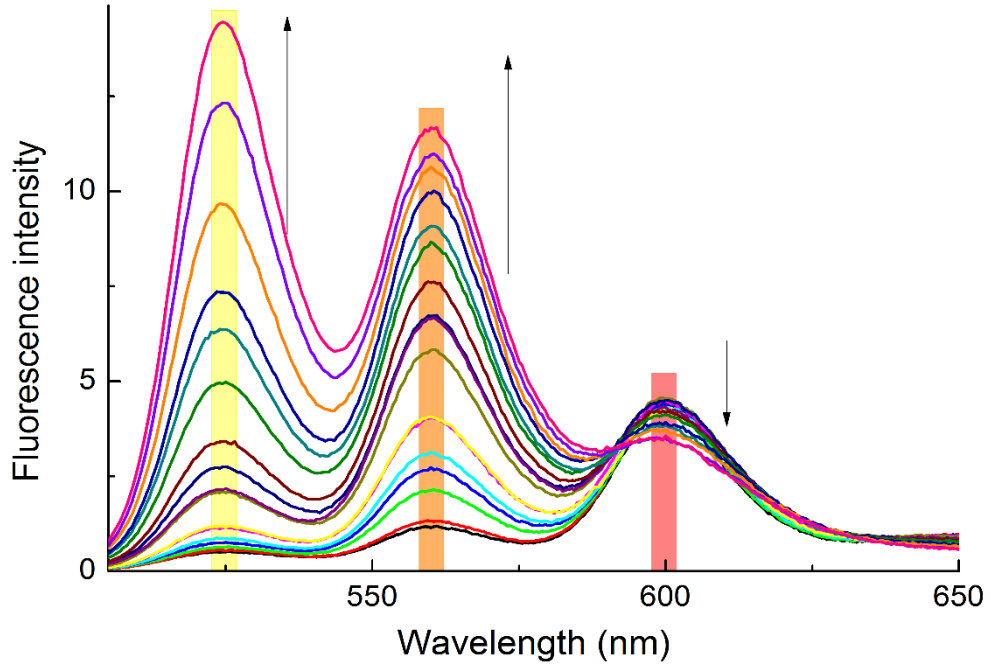
CYSTEINE

AA
0
25 μM
50 μM
75 μM
0,1 mM
0,15 mM
0,2 mM
0,25 mM
0,3 mM
0,4 mM
0,5 mM
0,7 mM
0,9 mM
1 mM
1,25 mM
1,5 mM
1,75 mM

EtOH/HEPES (1:1)
2 μM





Remarkable spectral band **splitting** of the BODIPY **induced by** the presence of **CYS** in the media.

Especially clear in fluorescence, with **new and strong hypsochromically shifted emissions.**

Similar trends in the rest of AAs.

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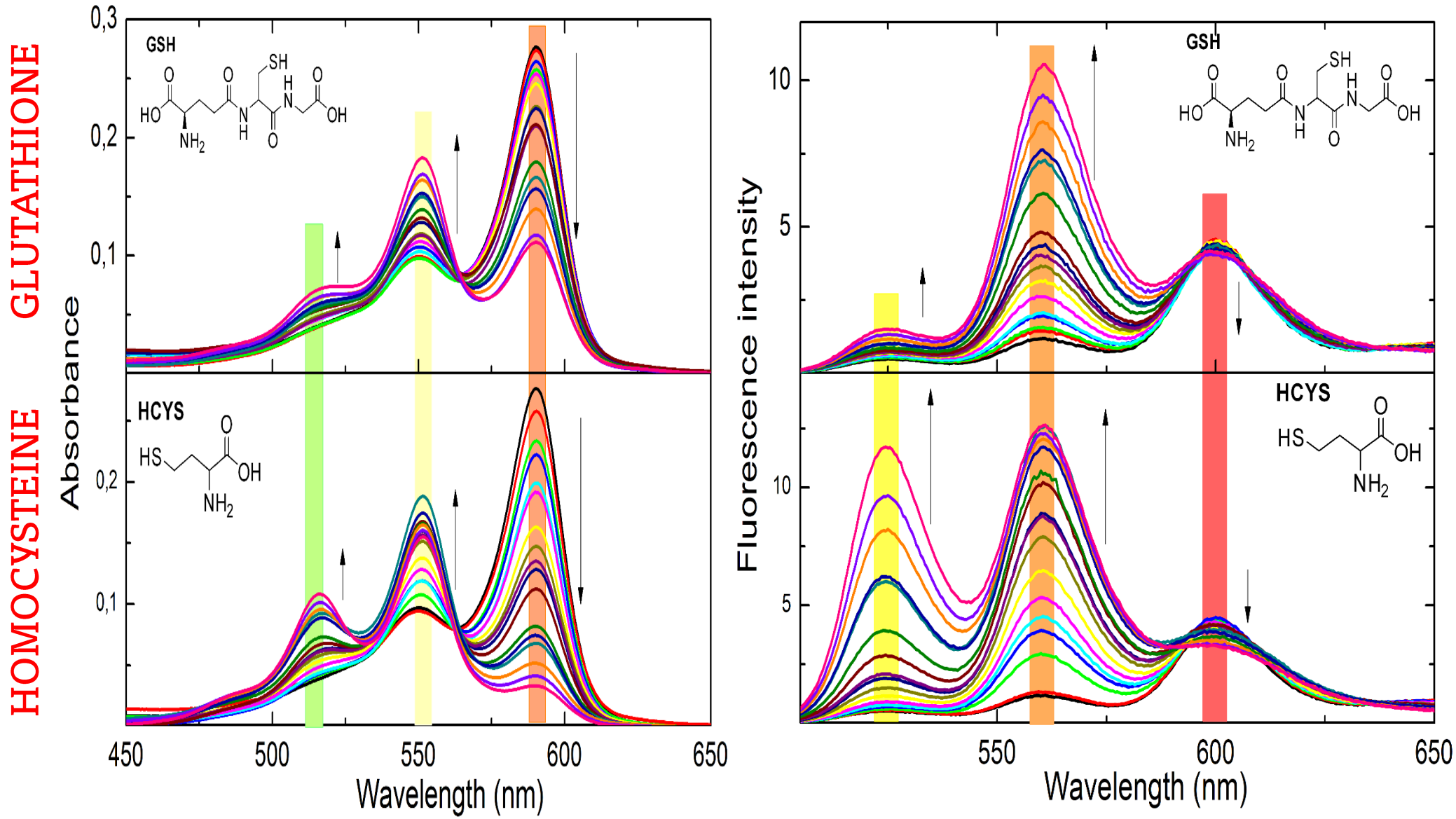

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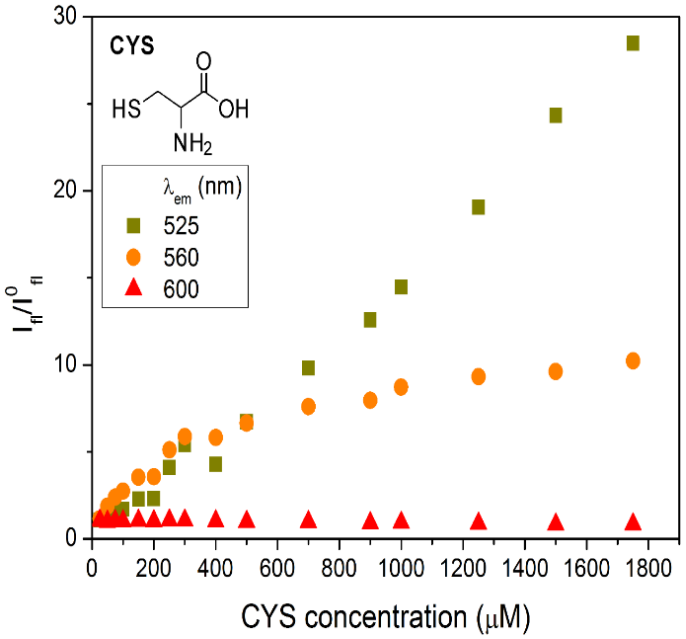
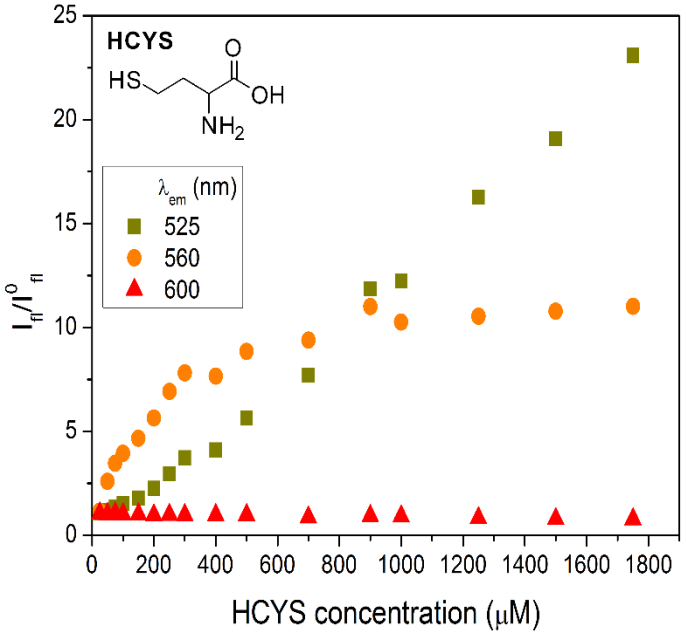
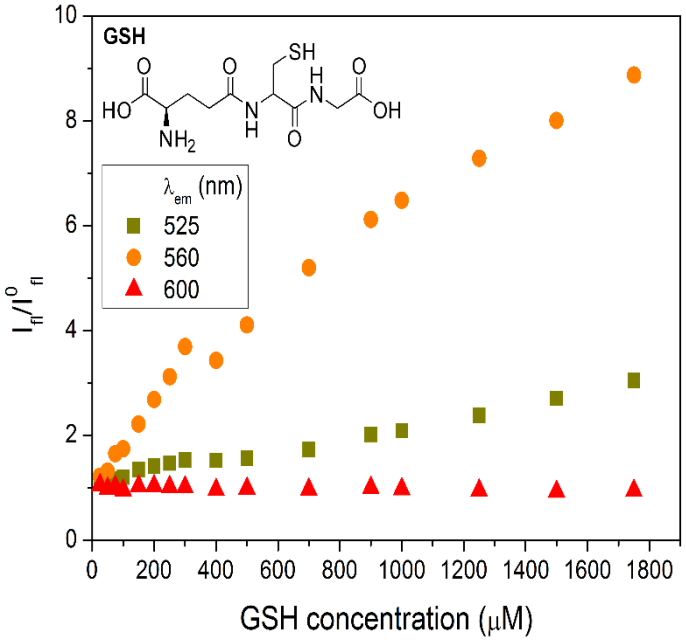
DETECTION CHANNELS







Up to 3 **detection channels** to monitor and quantify the presence of AA:

- The **fluorescence loss at 600 nm**
- The **fluorescence growing at 525 nm and 560 nm**

These last two channels are the most recommended ones

The sensor is **less sensitive to GSH** rather than to CYS and HCYS

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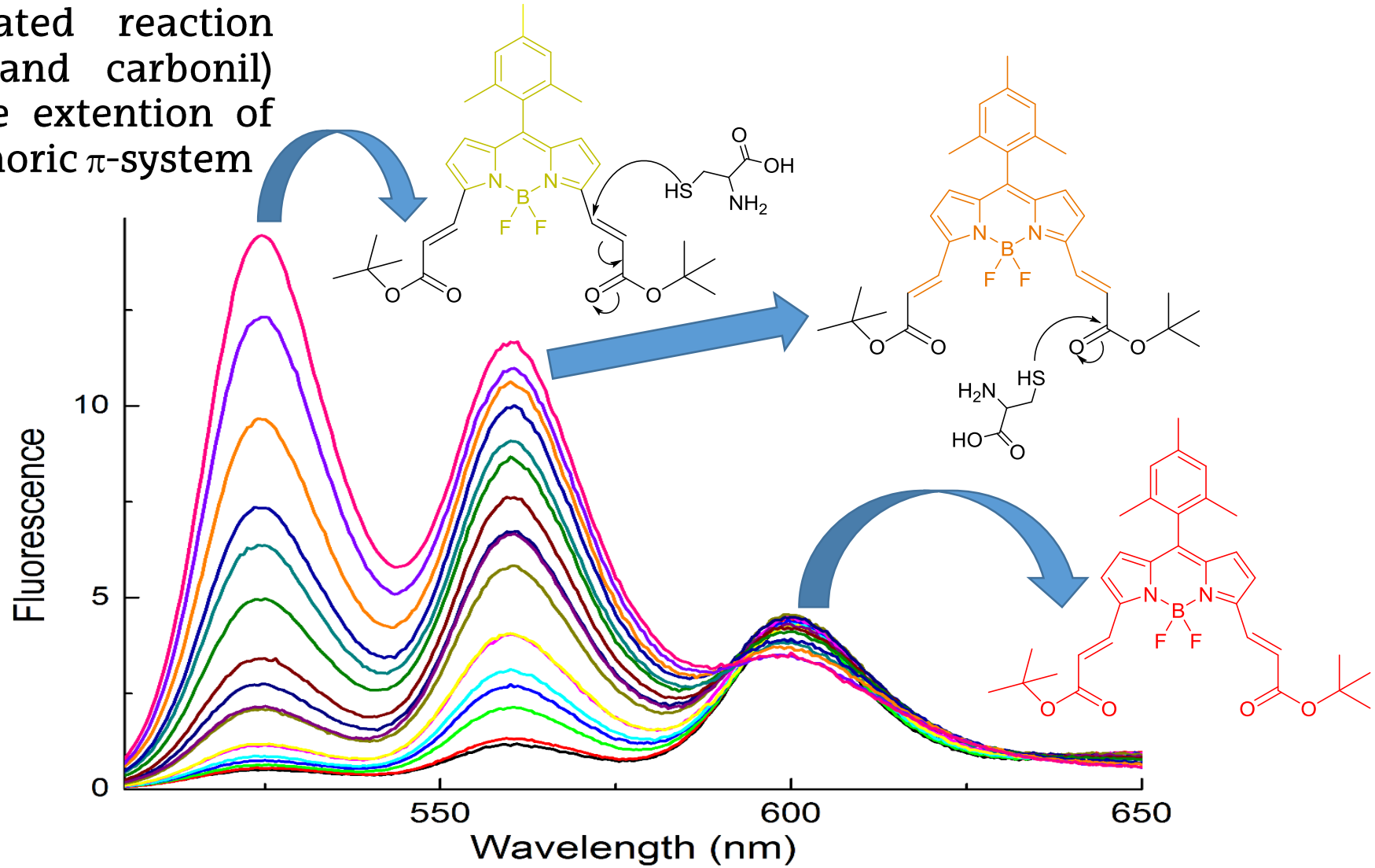
PROPOSED SENSING MECHANISM

The binding of the AA with the unsaturated reaction sites (vinyl and carbonil) decreases the extention of the chromophoric π-system



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The designed BODIPY-based chemosensor for **thiolated AAs** allows:

- **Easy and versatile detection**, just by the emission color.
- **Quantification** of the AA concentration by up to three detection channels.
- Sensitive detection down to **micromolar**.