Conformer-selective Photodissociation of Nanohydrated Biomolecules : Structure – Photophysics Relationship

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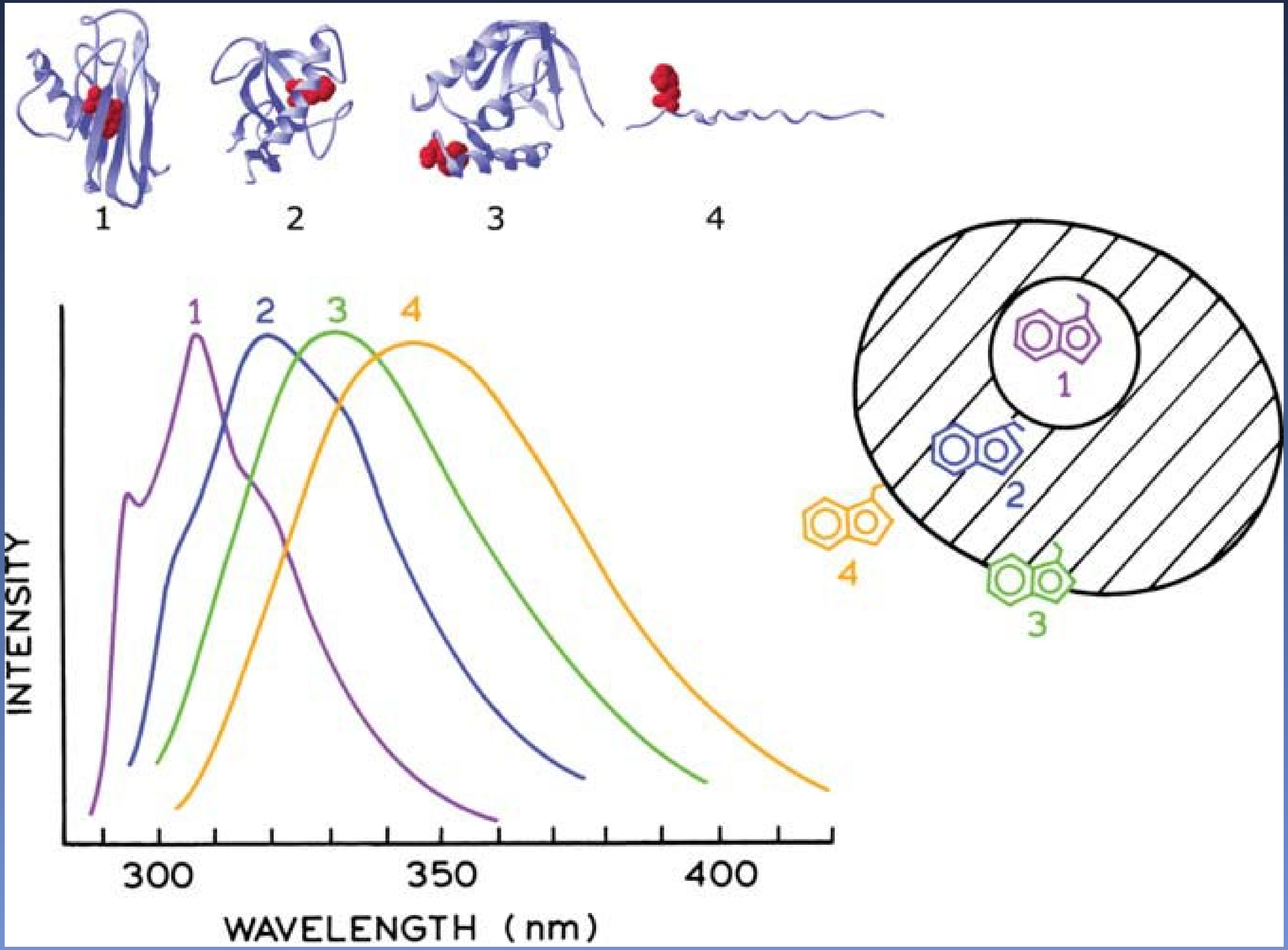
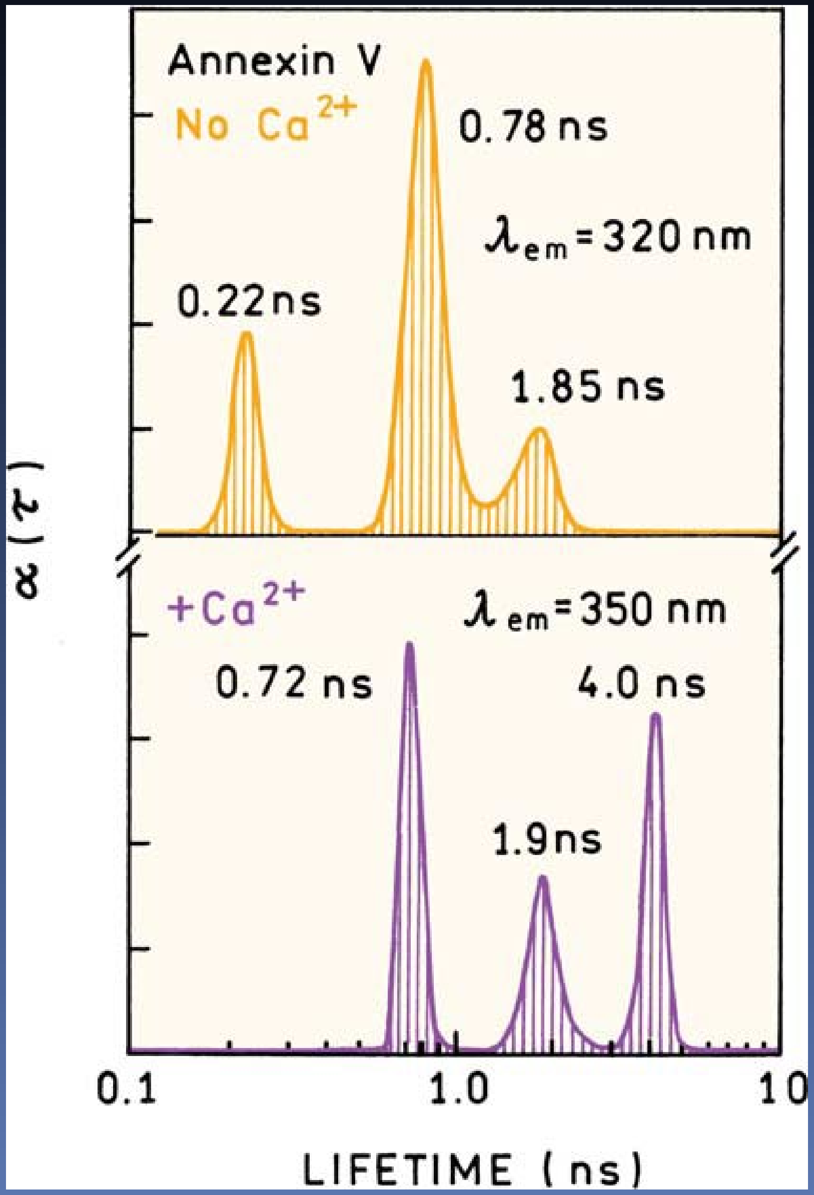
Tryptophan Fluorescent Properties

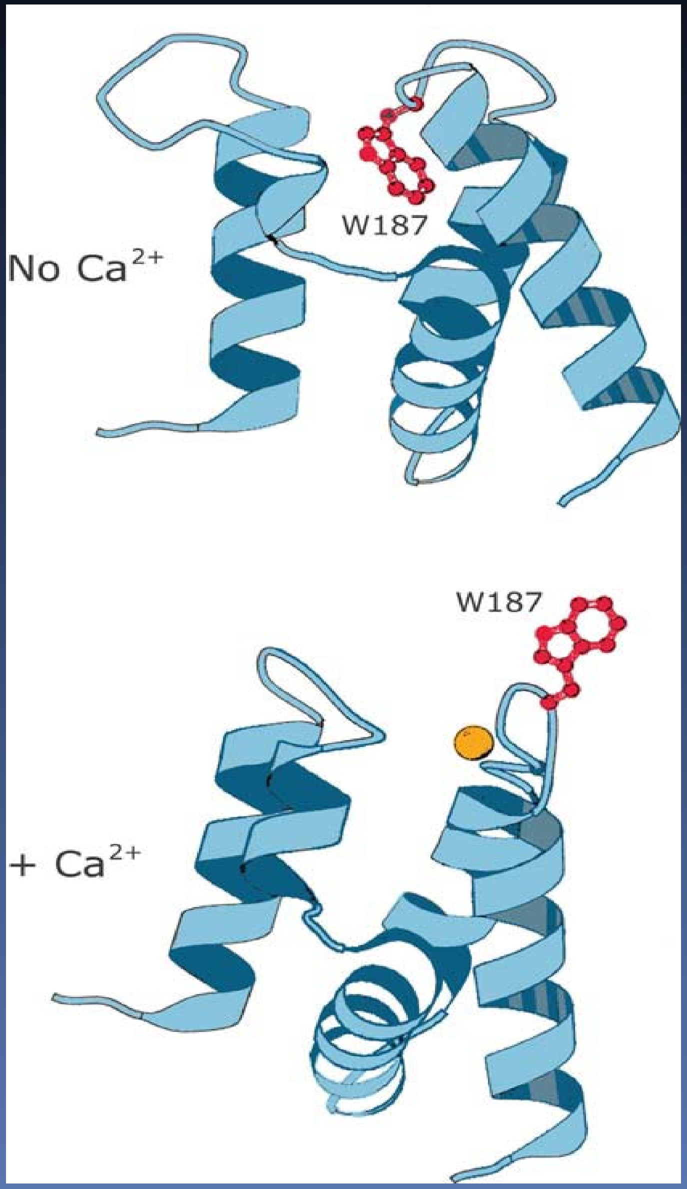
Effect of tryptophan environment on the emission spectra :
Folding / Unfolding of proteins



Principes of Fluorescence Spectroscopy, J. Lakowicz, Springer

Hydration effect on excited state lifetime

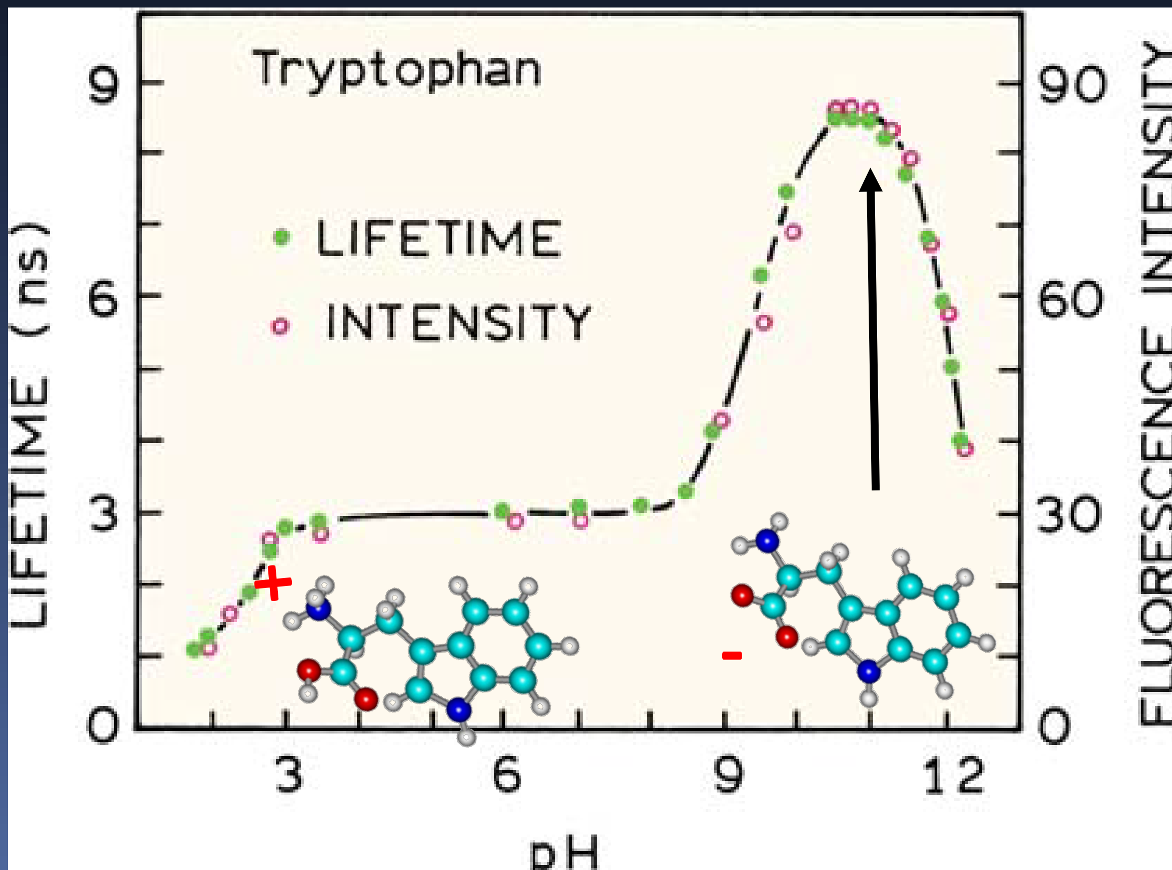




Structure of Annexin V in the absence (top) and presence (bottom) of Ca2+.

- ✓ Buried tryptophan residues seem to display shorter lifetimes !
- ✓ The longer lifetimes of exposed tryptophan residues have been puzzling because exposure to water is expected to result in shorter lifetimes.
- ✓ It is now known that peptide bonds and charged residues can quench tryptophan emission.

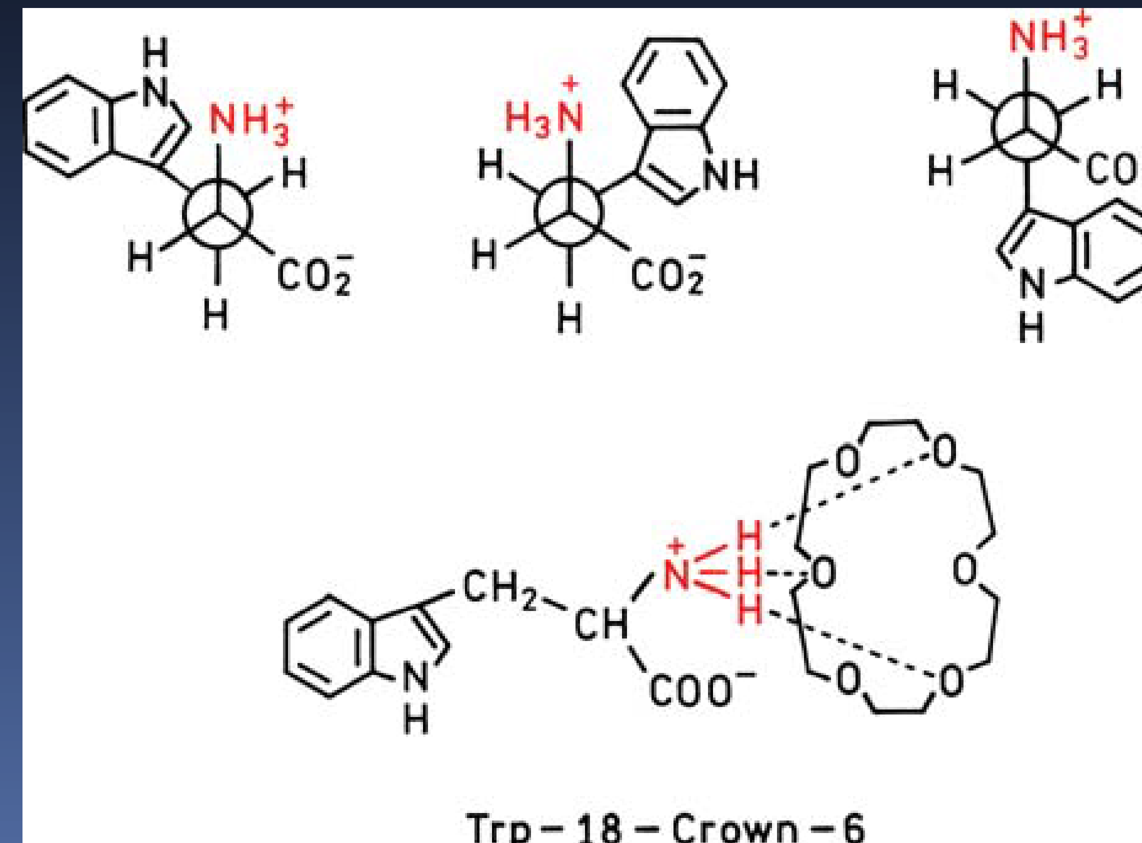
Trp Fluorescent Properties



Quenching by the ammonium group

- At neutral pH : 2 time constants (0.5 and 3 ns)
- Shortening of excited state lifetime at low PH
- At high PH : the quantum yield and mean lifetime increase approximately threefold.

The rotamer model to explain the multi exponential time decay

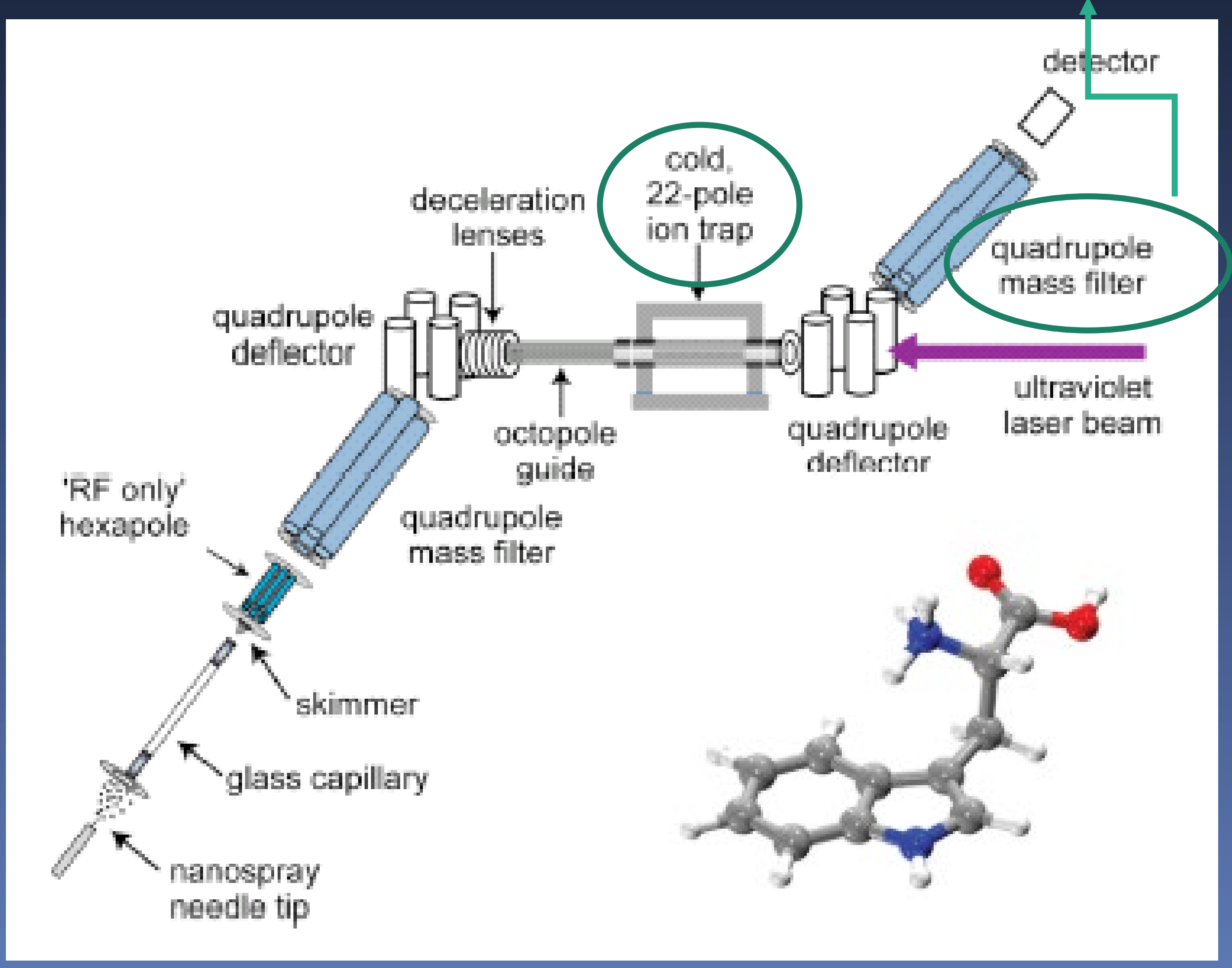


Rotational isomers of tryptophan.

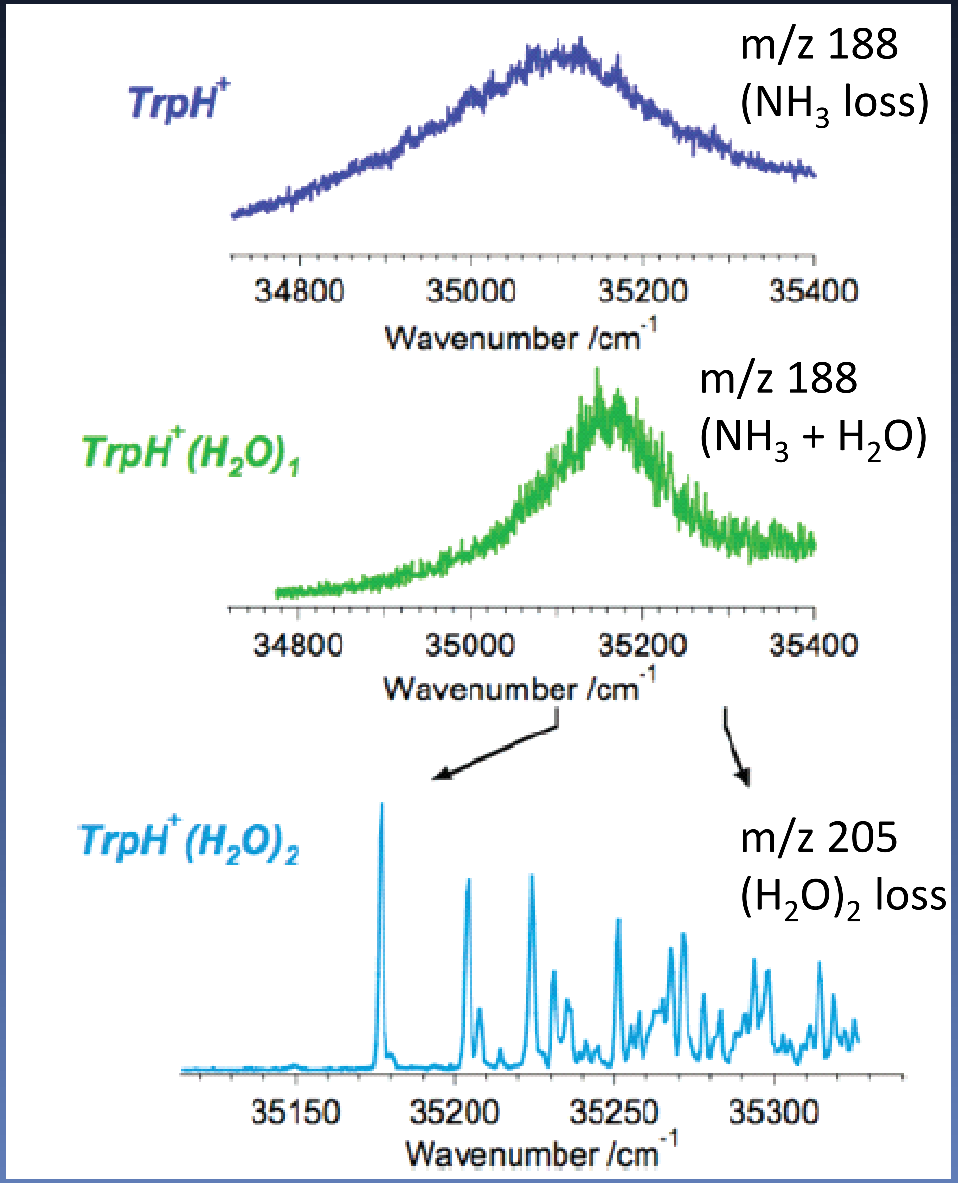
- The rotamers on the left is thought to be responsible for the 0.5 ns decay time.
- 18-Crown-ether prevents quenching by the ammonium group.

The seminal study of Boyarkin and Rizzo (2006)
Cryogenic ion spectroscopy of (hydrated) $TrpH^+$ at the band origin

$$m/z\ 205 + h\nu \longrightarrow m/z\ 188 + NH_3$$

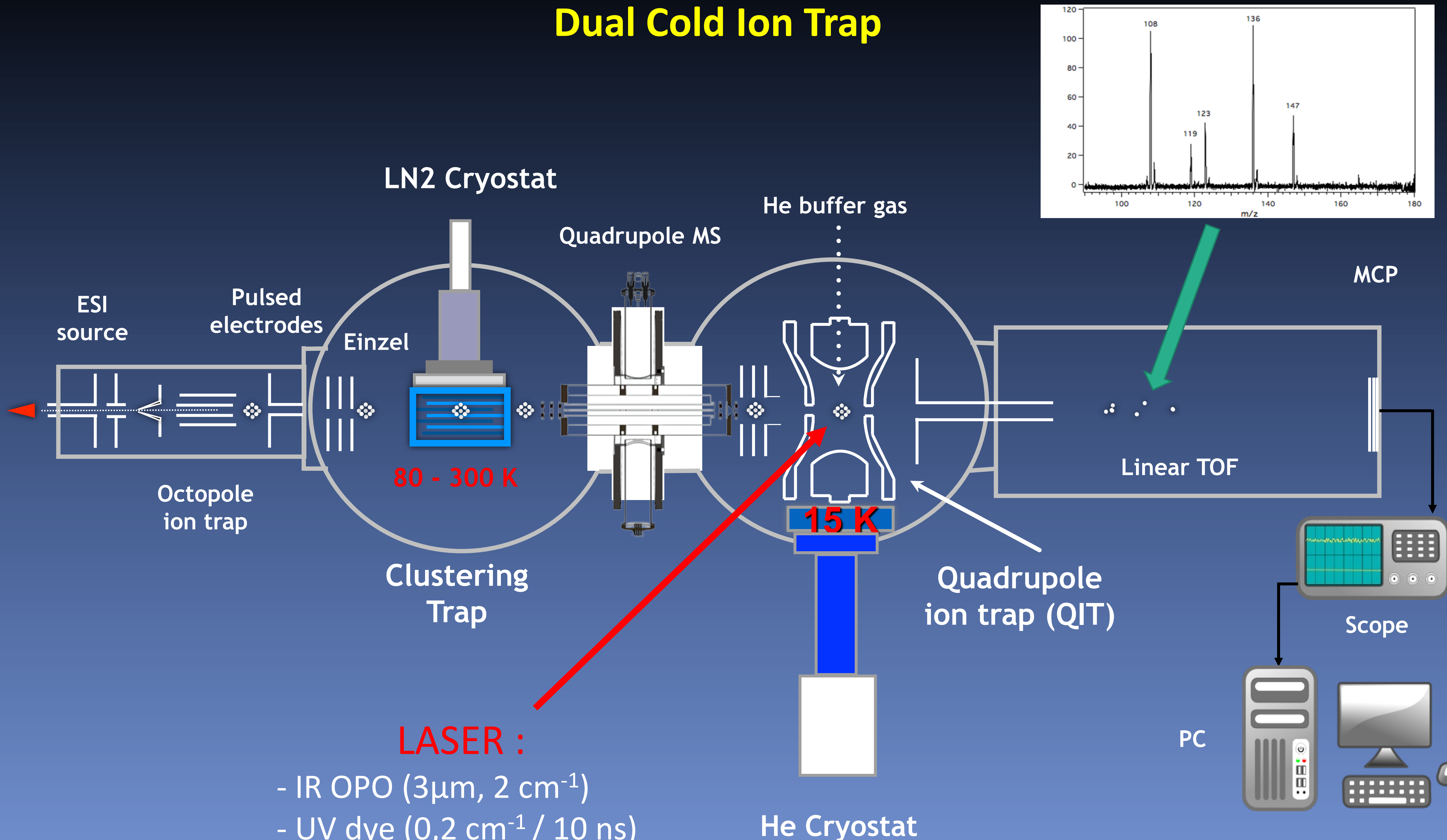


JACS, ***128***, *2816 (2006)*



- **Broadened excitation spectrum of $TrpH^+$ at band origin : short excited lifetime (fs)**
- **Sharp vibronic transitions for $TrpH^+$-$(H_2O)_2$: Unexpected solvation effect !!**

Orsay Experimental Setup
Dual Cold Ion Trap



LASER :

- IR OPO (3µm, 2 cm^{-1})
- UV dye (0,2 cm^{-1} / 10 ns)

- UV OPO (10 cm^{-1} / **16 ps**)

Reactivity and Dynamics of Photo-Induced Processes in Protonated Molecules

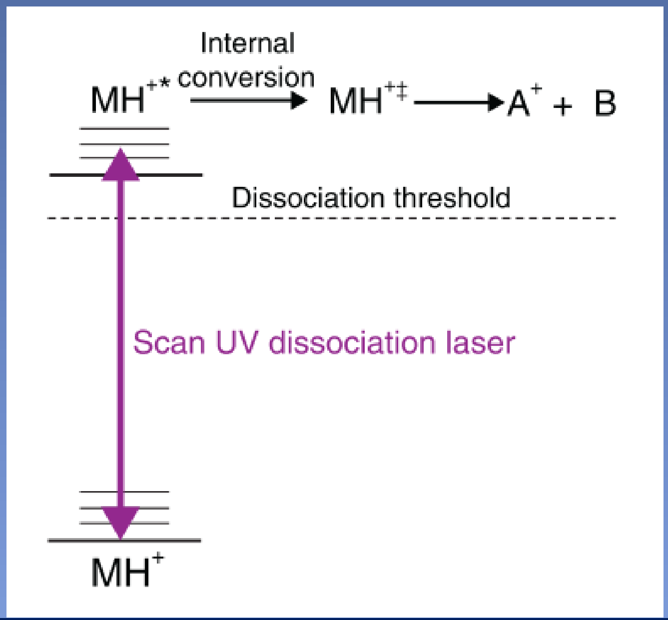
Combine UV laser spectroscopy with mass spectrometry

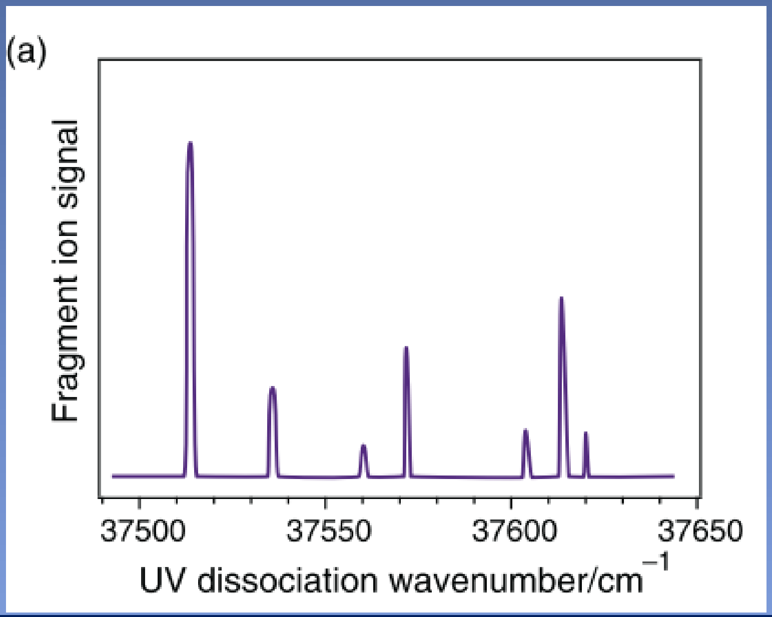
- Control of the entrance channel : mass-selection m/z
- Control of the excess energy : photon ; tunable laser
- Control of the exit channel : detection of all fragmentation channels

Cold Ions (10-20 K)

- Well resolved spectroscopy
- Conformer selectivity
- Direct comparison with QC calculations

UV Photo Dissociation Spectroscopy (UV-PD)



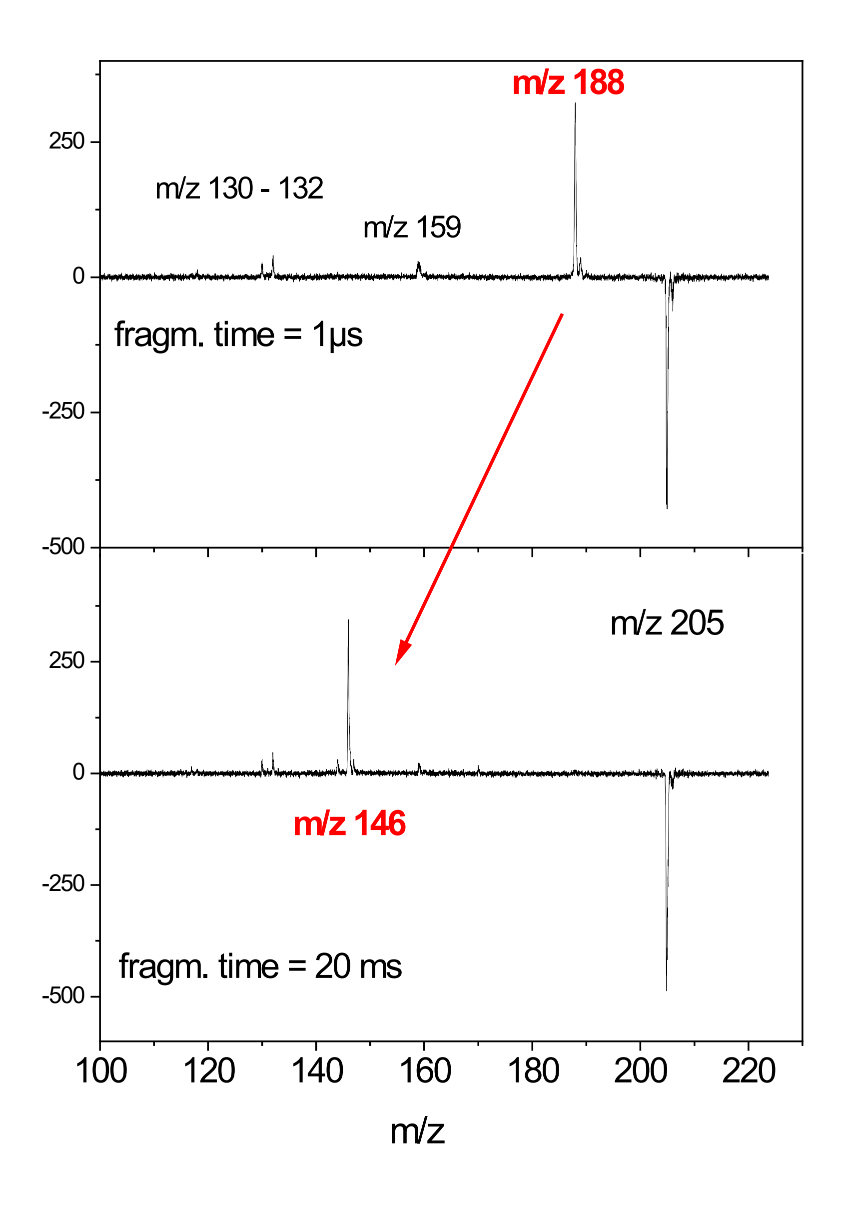


UV Photodissociation spectroscopy of cold $TrpH^+$

Main fragmentation channel NH_3 loss : m/z 188

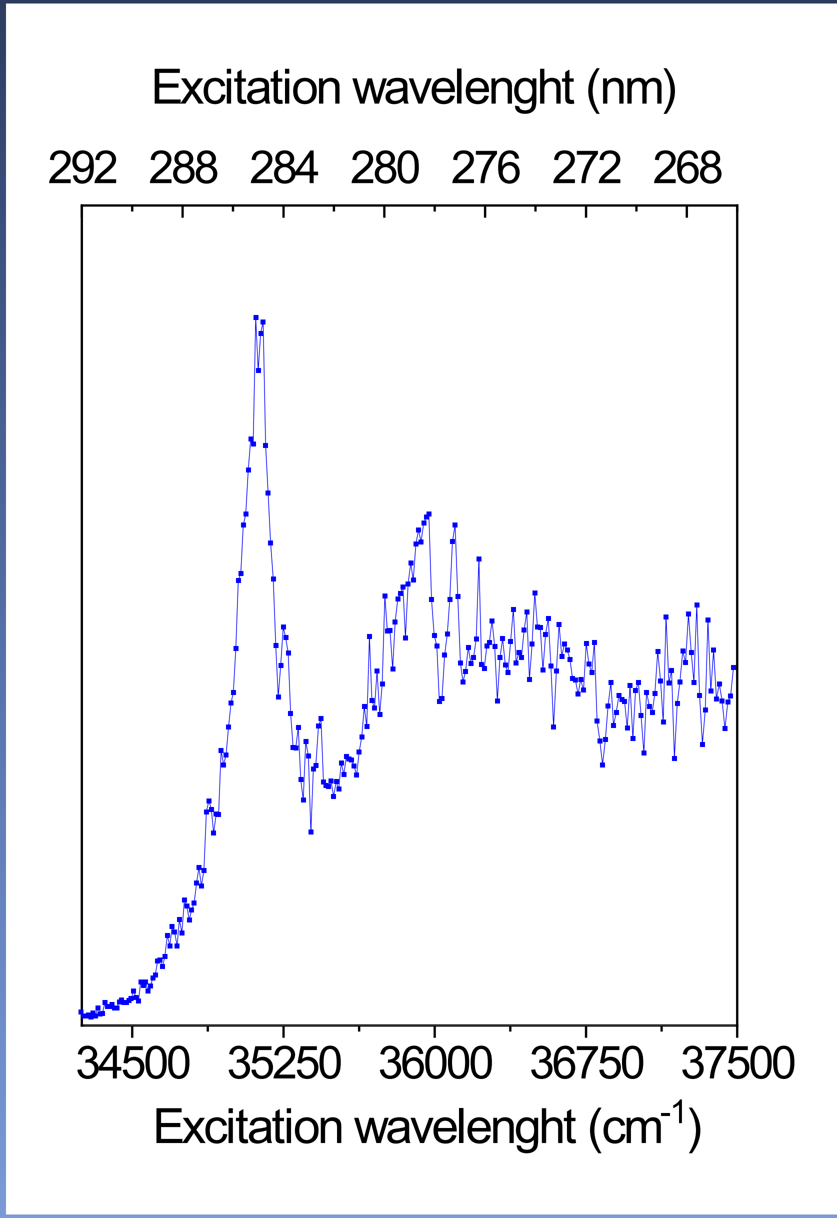
Secondary fragmentation m/z 188 → m/z 146
NH_3 and CH_2CO loss : m/z 146

CID like fragmentation : Internal conversion



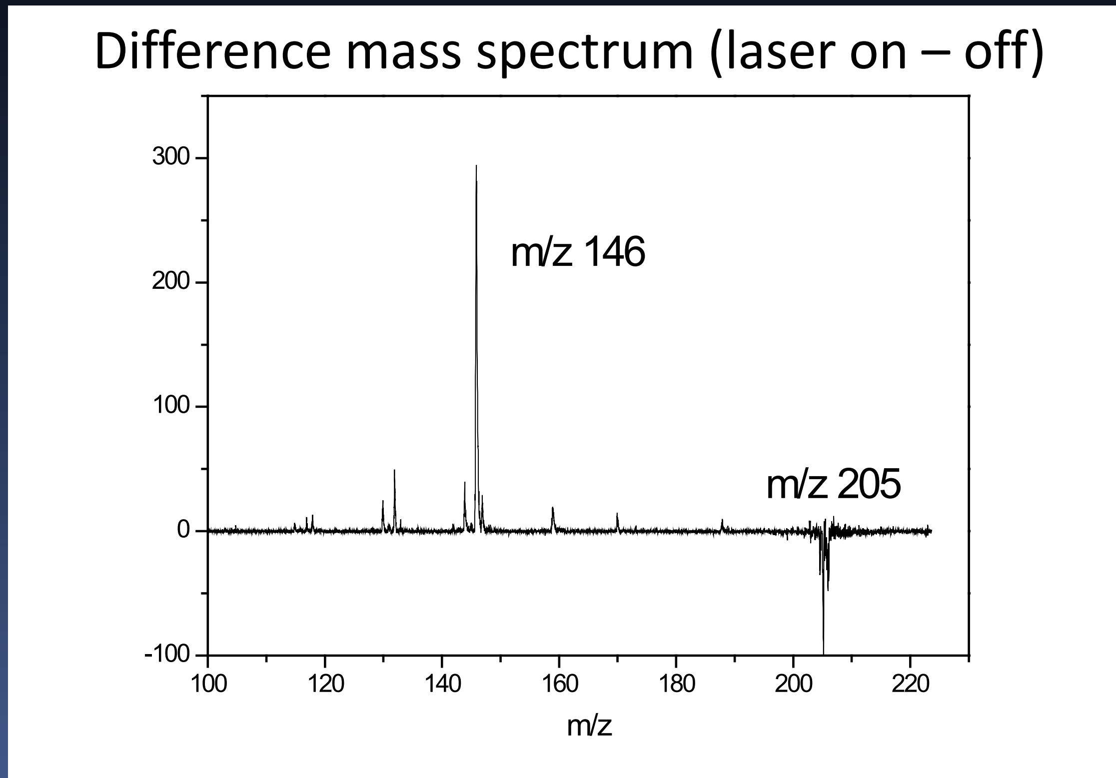
Broadened electronic spectroscopy
Sub picosecond $\pi\pi^*$ excited state lifetime

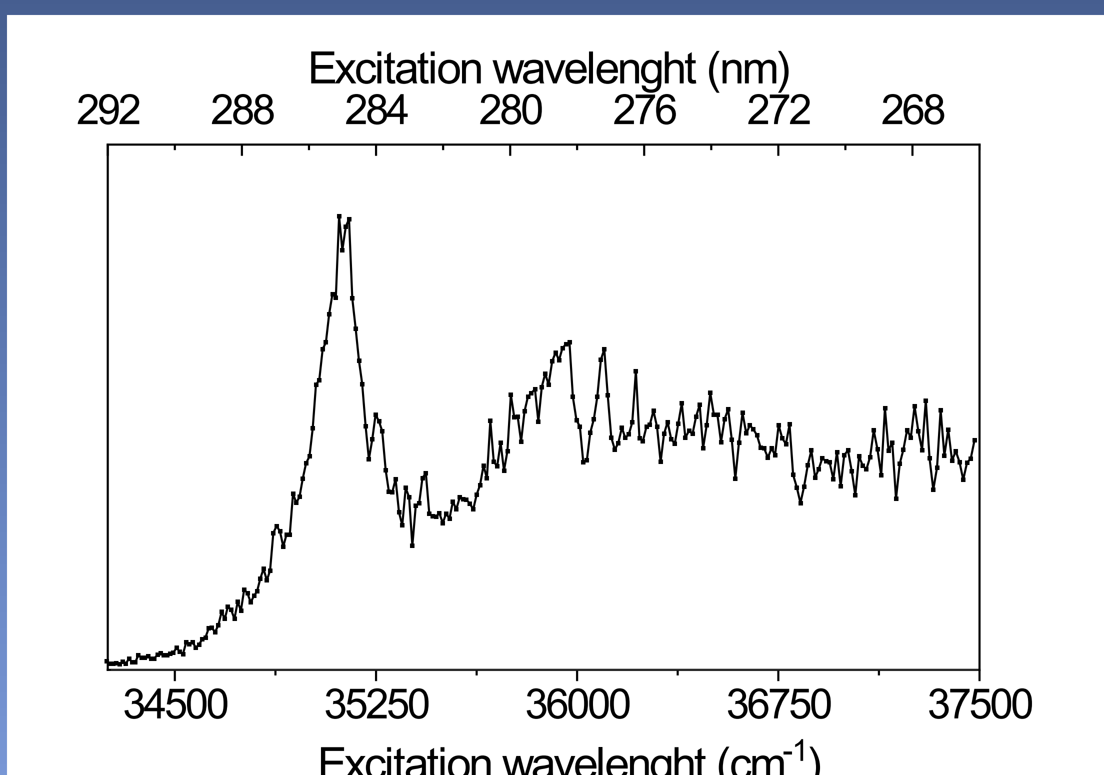
Boyarkin et al. JACS, **128**, 2815 (2006)

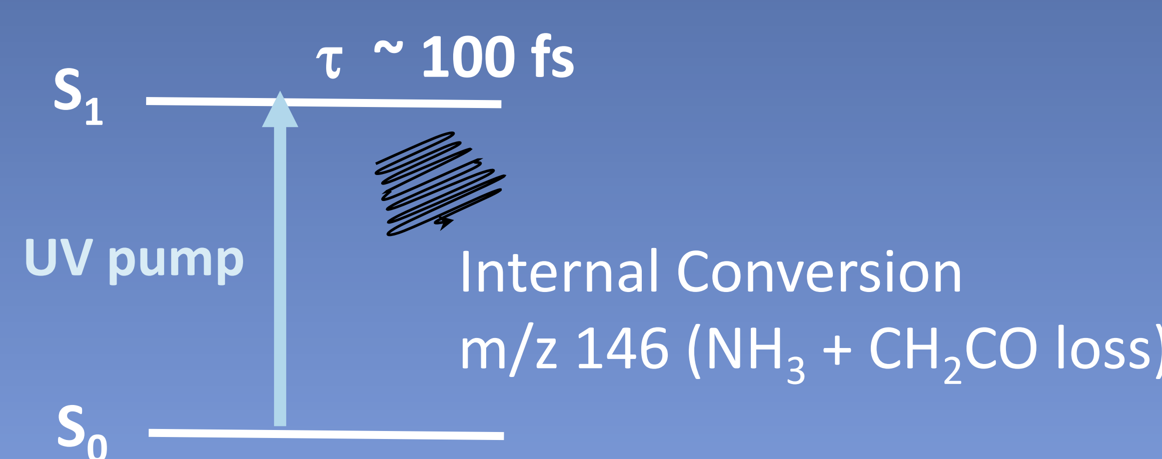


pump-probe photodissociation spectroscopy at the band origin

Pump only







pump-probe photodissociation spectroscopy at the band origin

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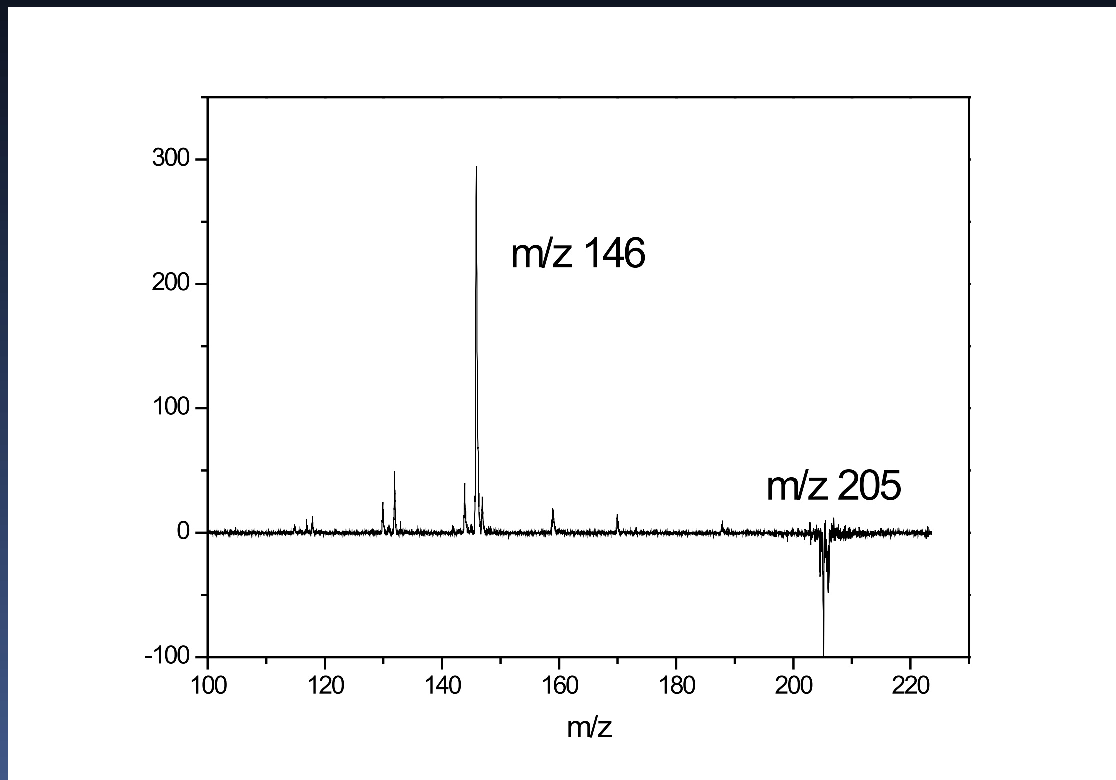
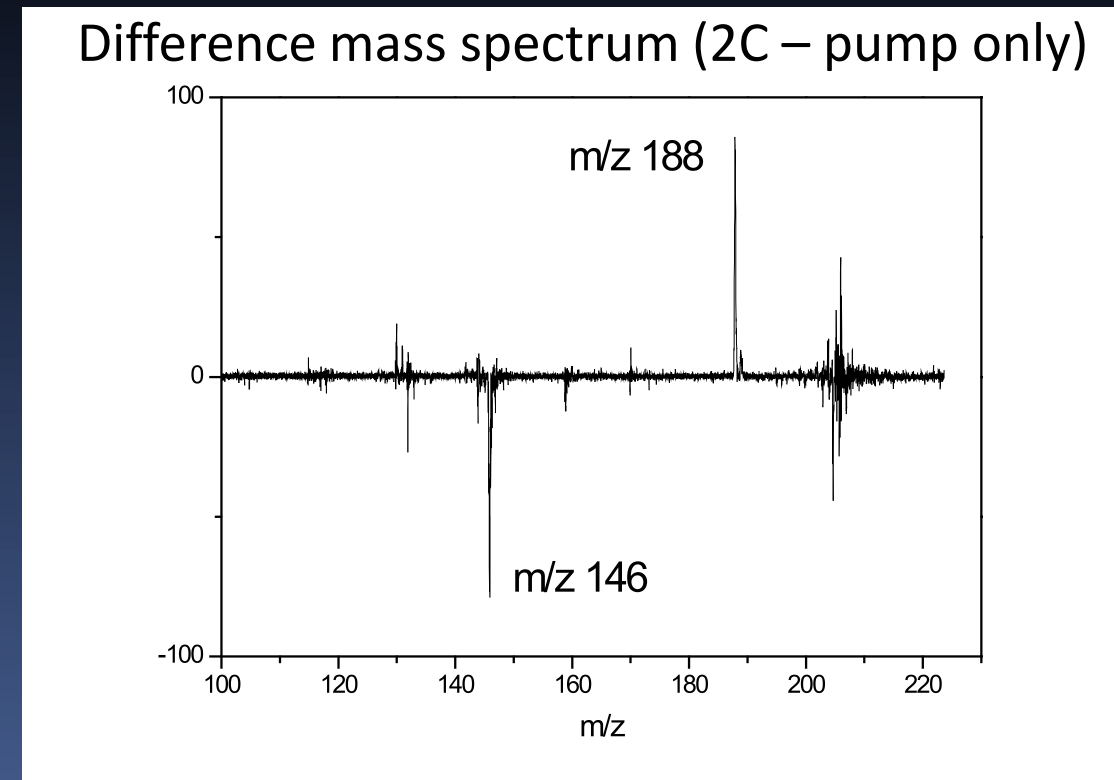
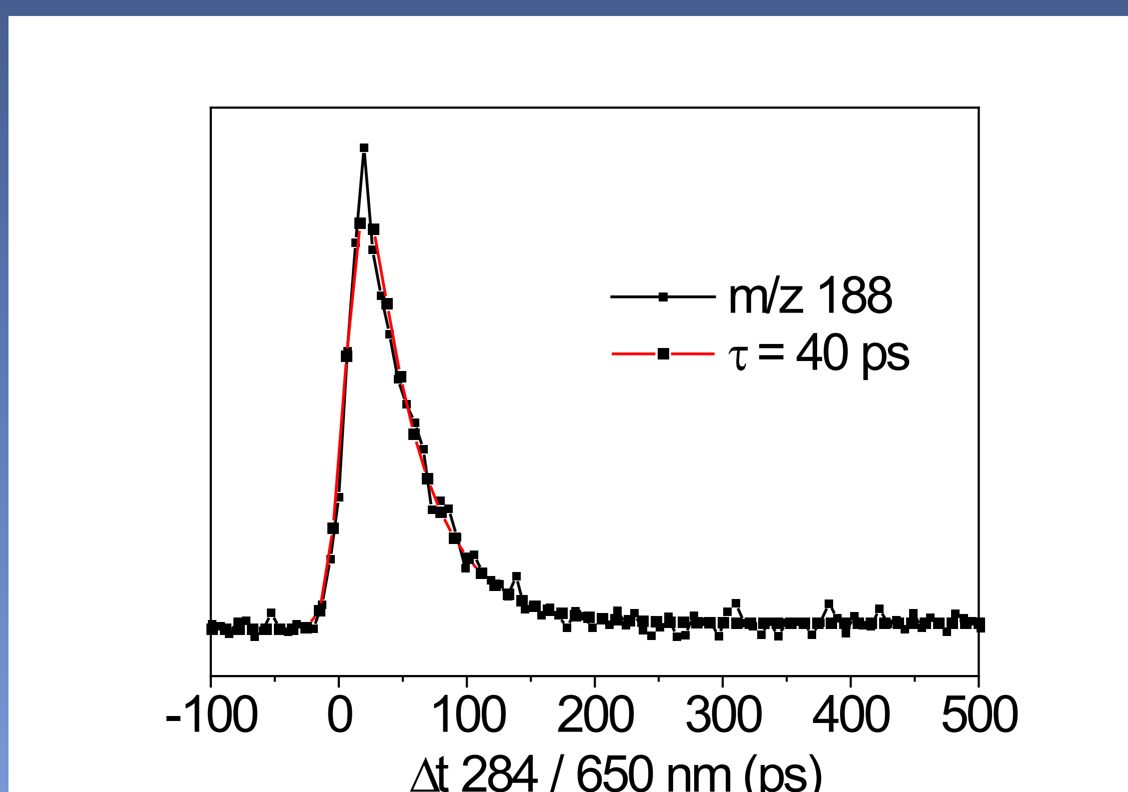
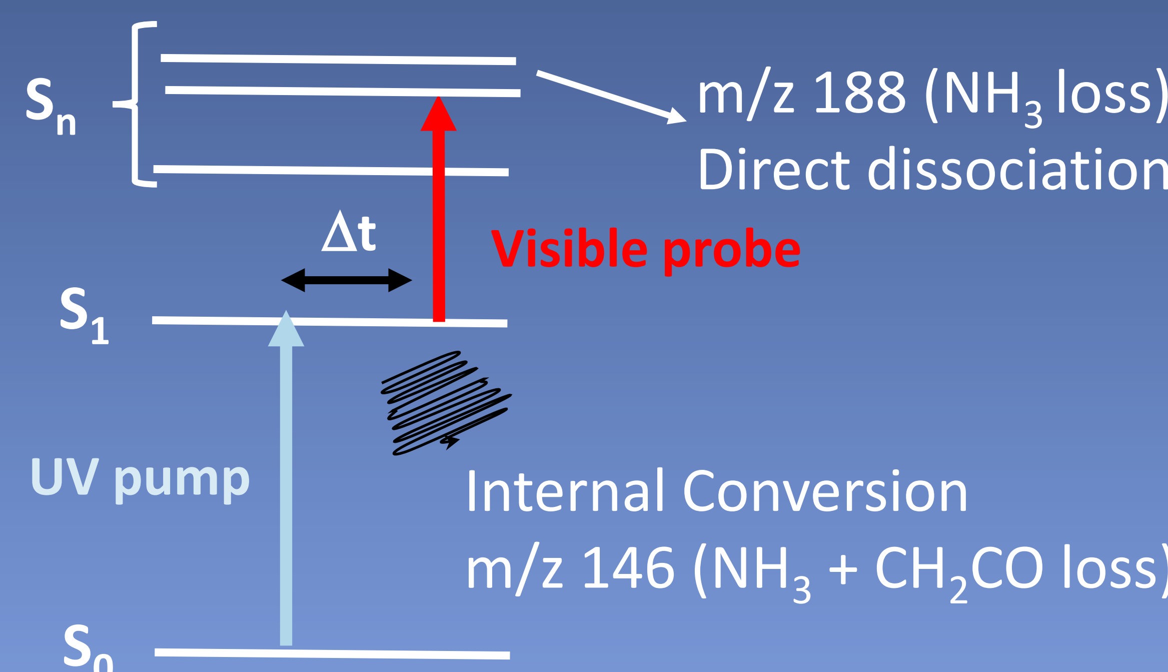


Pump + 650 nm



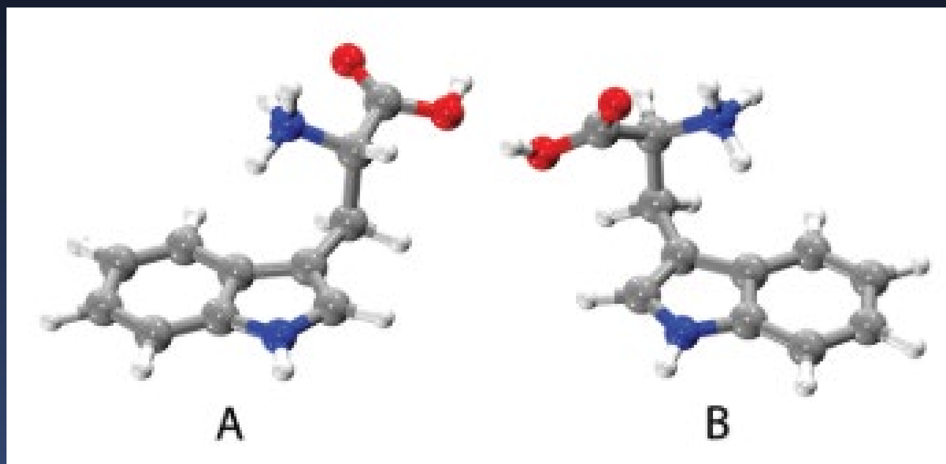


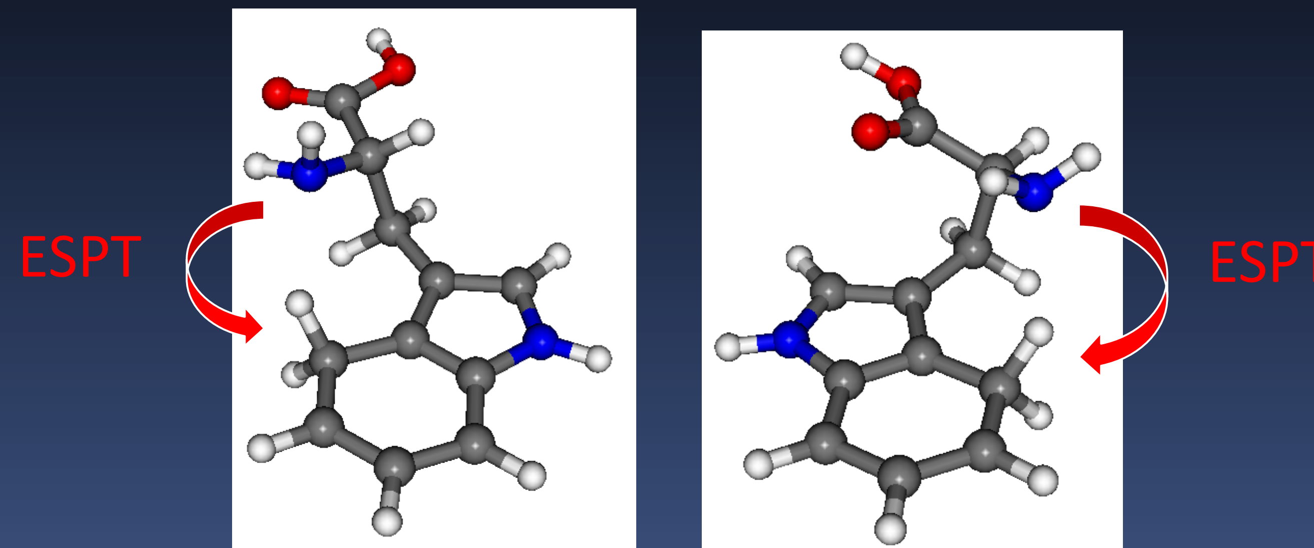
NH_3 loss (from an excited state)



Barrierless Excited-State Proton Transfer (ESPT) at the band origin (284 nm)

Gregoire et al. JACS, **129**, 6223 (2007)

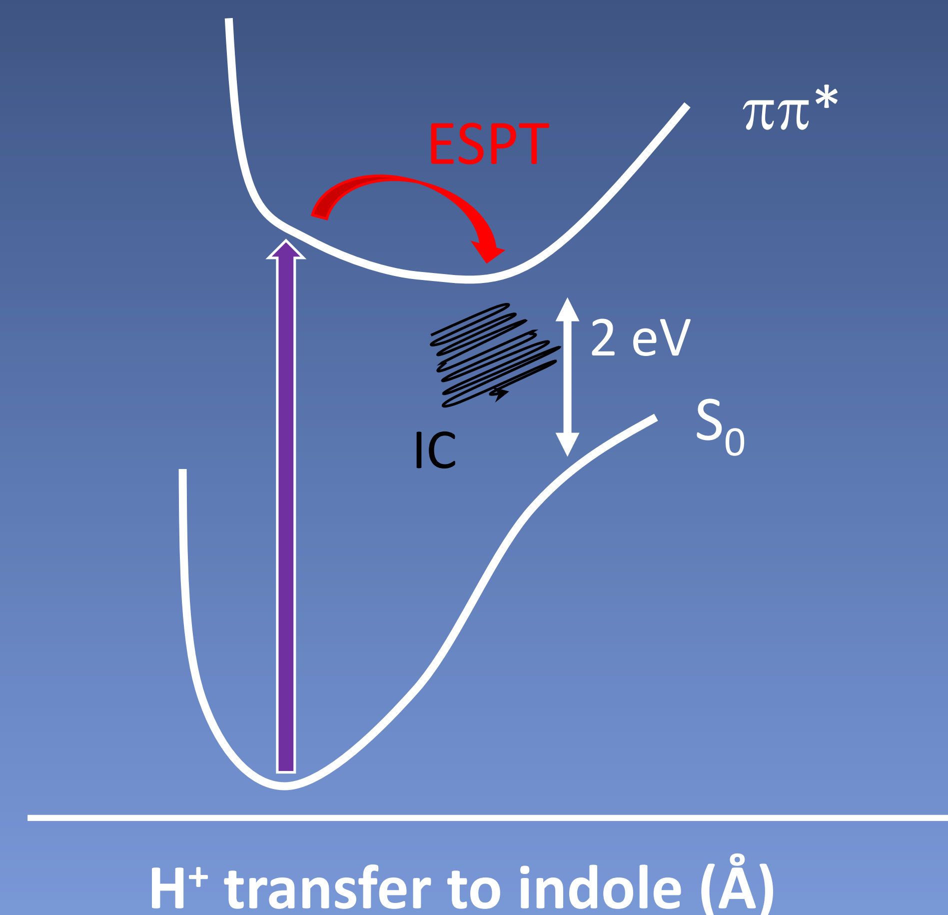




- S_0 and S_n geometry optimizations performed at CC2-SCS / aug-cc-pVDZ level.

- Excited state optimization of the $^1\pi\pi^*$ states (L_b and L_a) leads to barrierless ESPT (fs time scale)

➢ **40 ps : Lifetime of the ESPT structure**



Conclusions $TrpH^+$

$TrpH^+$ photodynamics not as simple as supposed :

- Internal conversion but not from the locally excited $\pi\pi^*$ state
- **Barrierless Excited State Proton Transfer** : fs lifetime of $\pi\pi^*$ state
- Lifetime of ESPT form : tens of ps (with about 0.8 of excess energy)

- Specific photofragments as a function of the probe wavelength:

Direct dissociation in the excited states

The deactivation processes in the excited state are not straightforward to understand

CHEMICAL REVIEWS
Review
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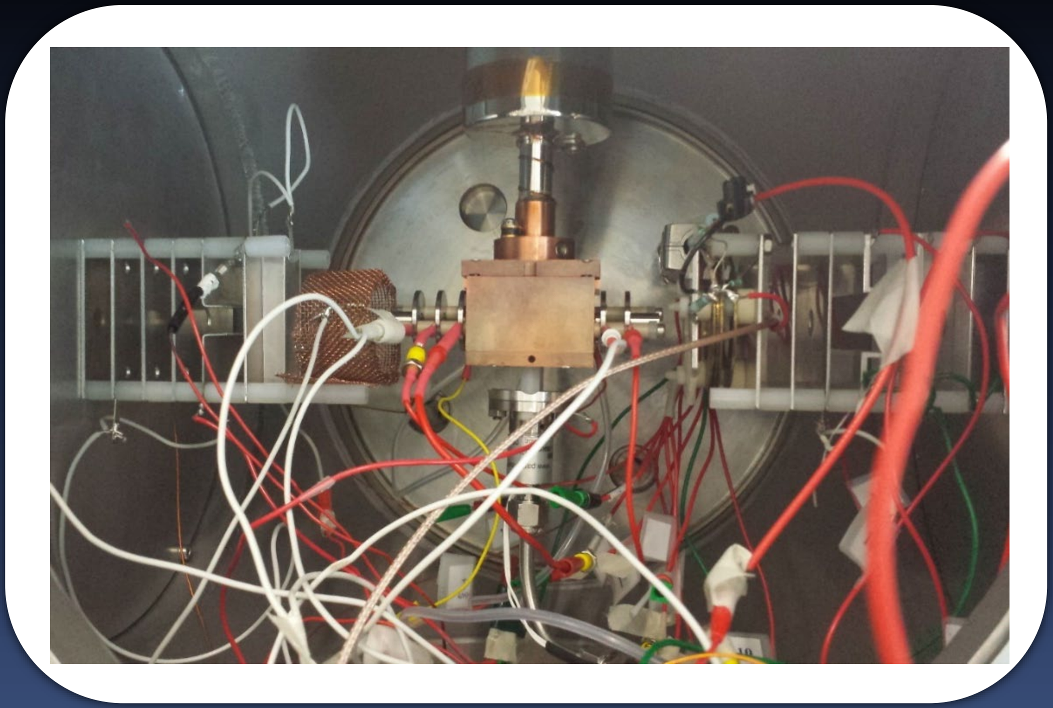
pubs.acs.org/CR

UV Photoinduced Dynamics of Conformer-Resolved Aromatic Peptides

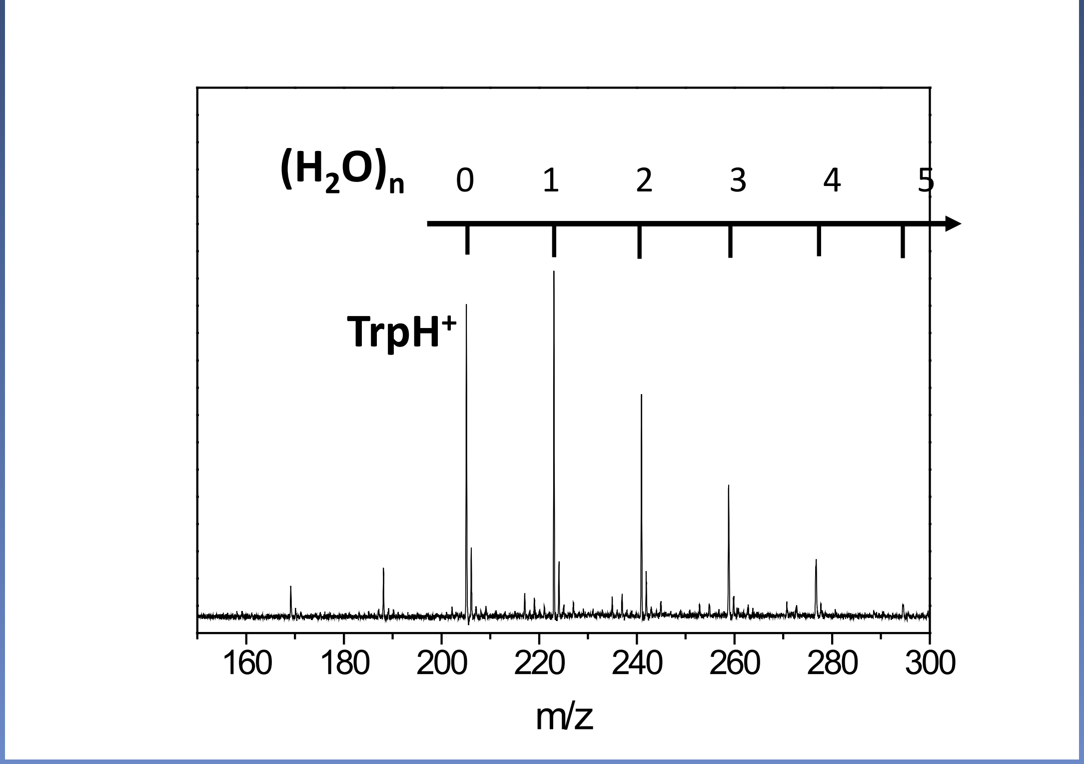
Satchin Soorkia,[†] Christophe Jouvet,[‡] and Gilles Grégoire*[,†]

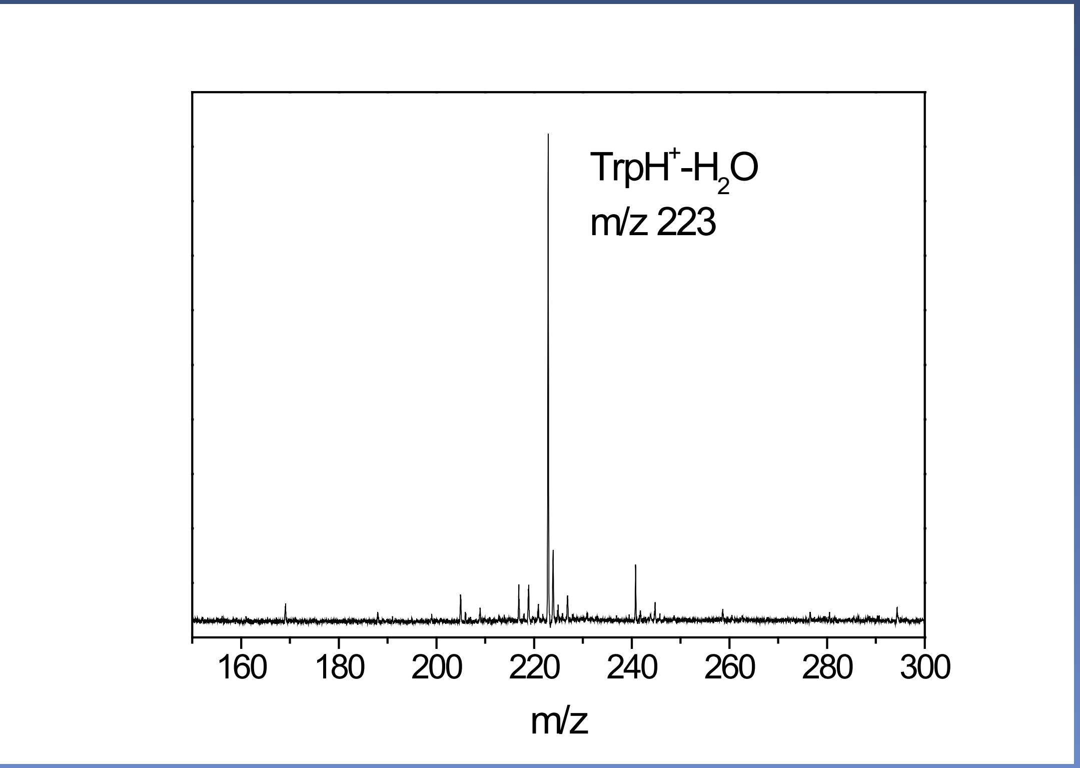
$TrpH^+$-$(H_2O)_n$



Home-made octopole trap :
Liquid Nitrogen cooling

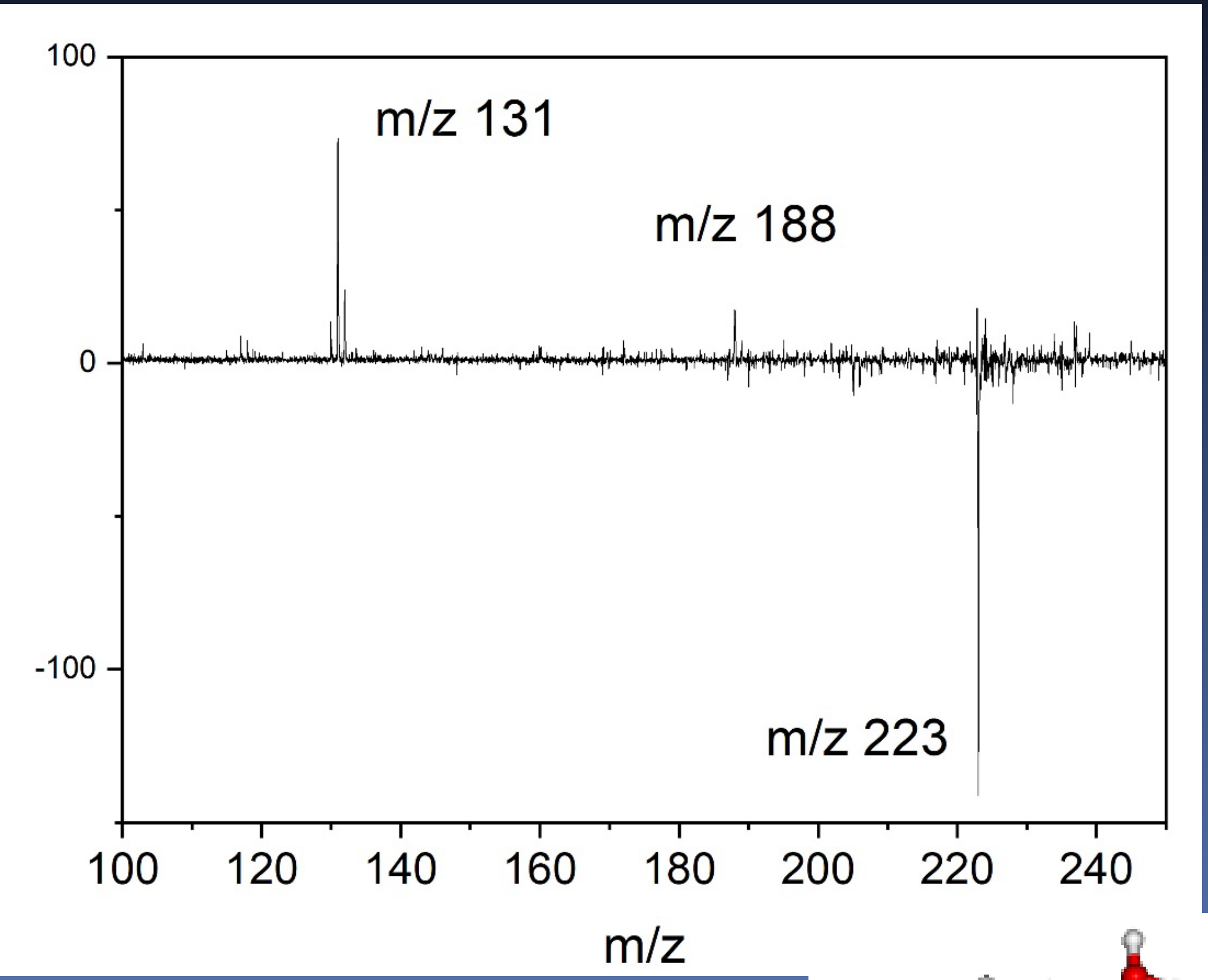
Mass Selection : Quadrupole mass filter

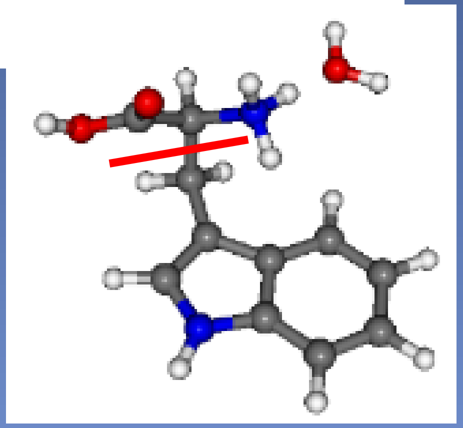




- Temperature below 200 K
- Trapping time (1-10 ms)

UV Photodissociation of $TrpH^+$-H_2O

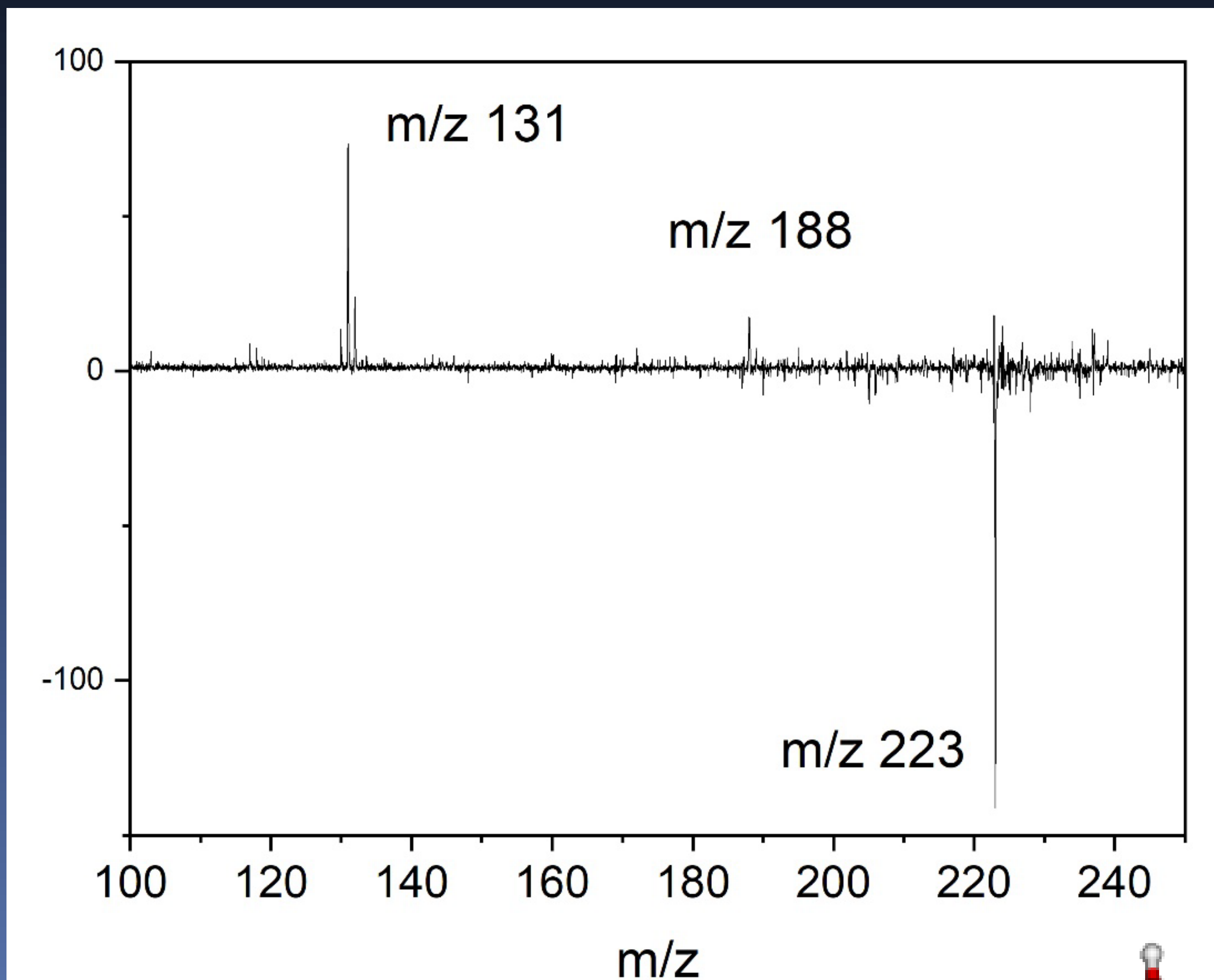


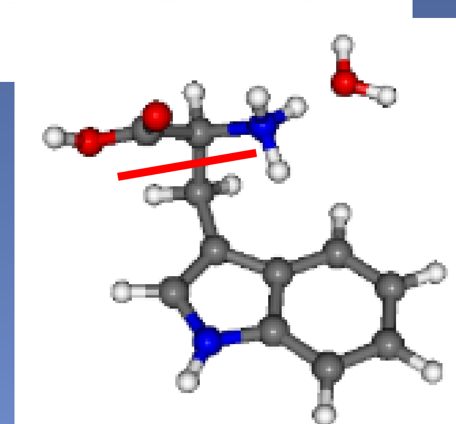


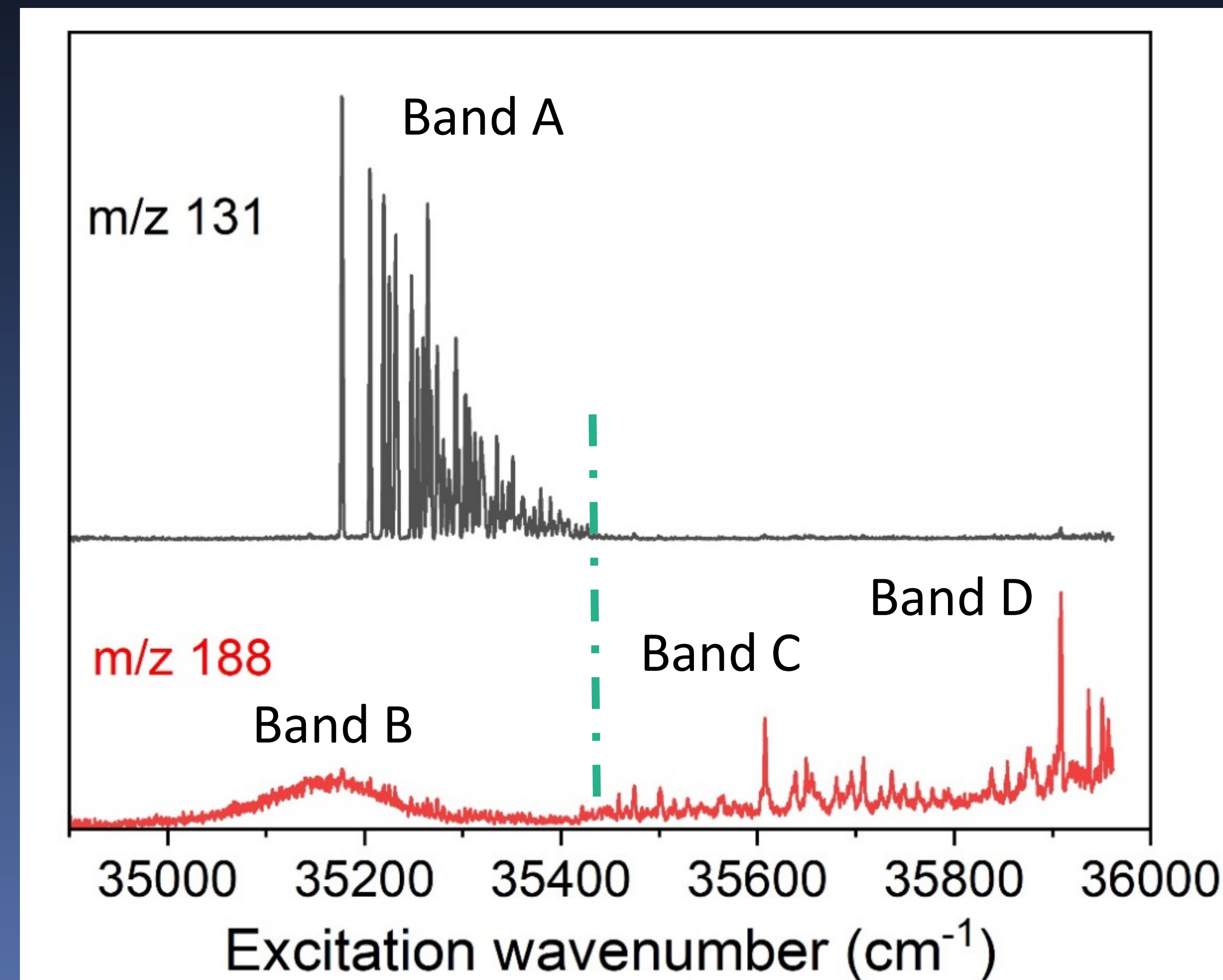
Main fragmentation channels :

- m/z 131 : C_α-C_β bond break
- m/z 188 : H_2O and NH_3 loss (IC)

UV Photodissociation of $TrpH^+$-H_2O







Main fragmentation channels :

- m/z 131 : C_α-C_β bond break
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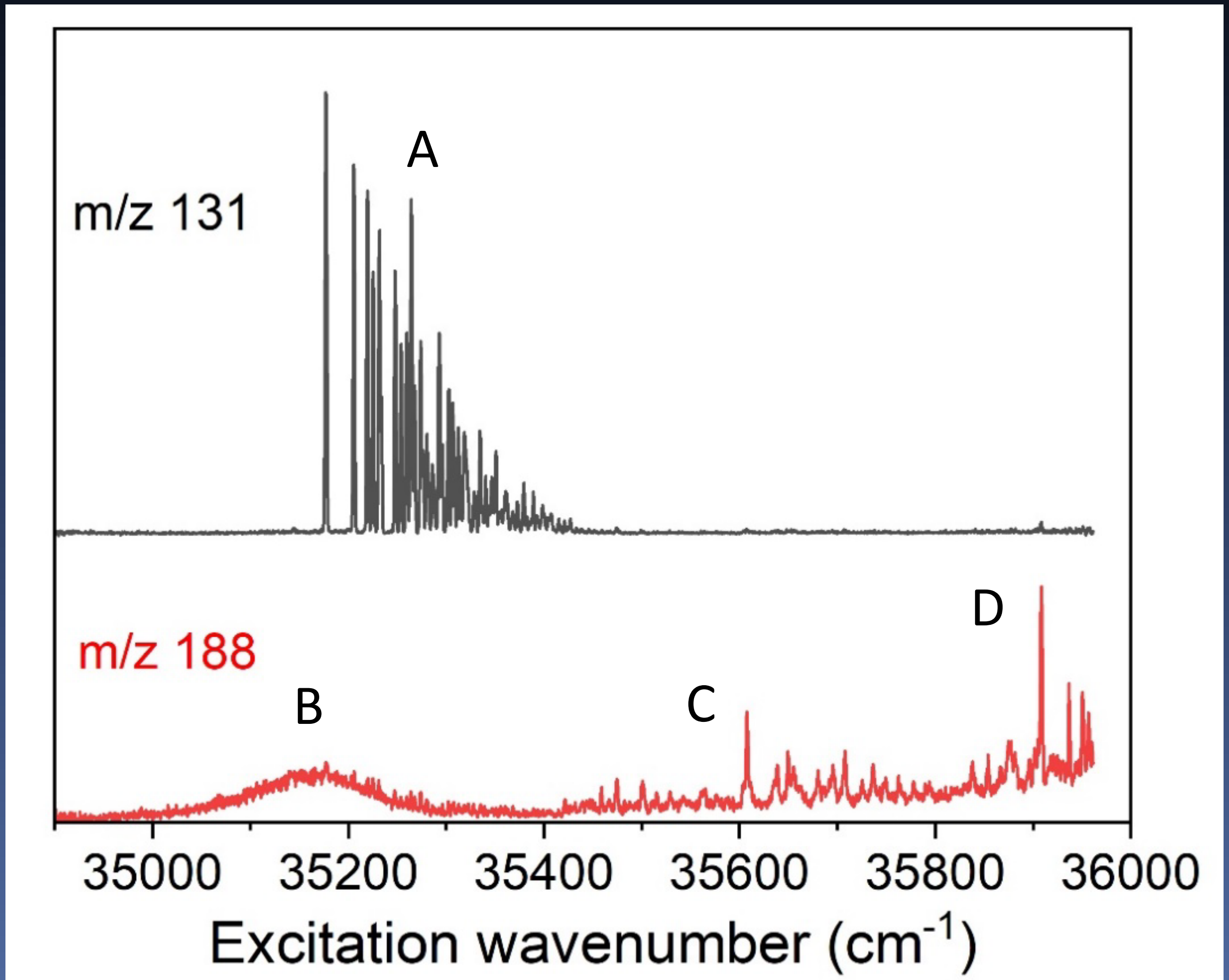
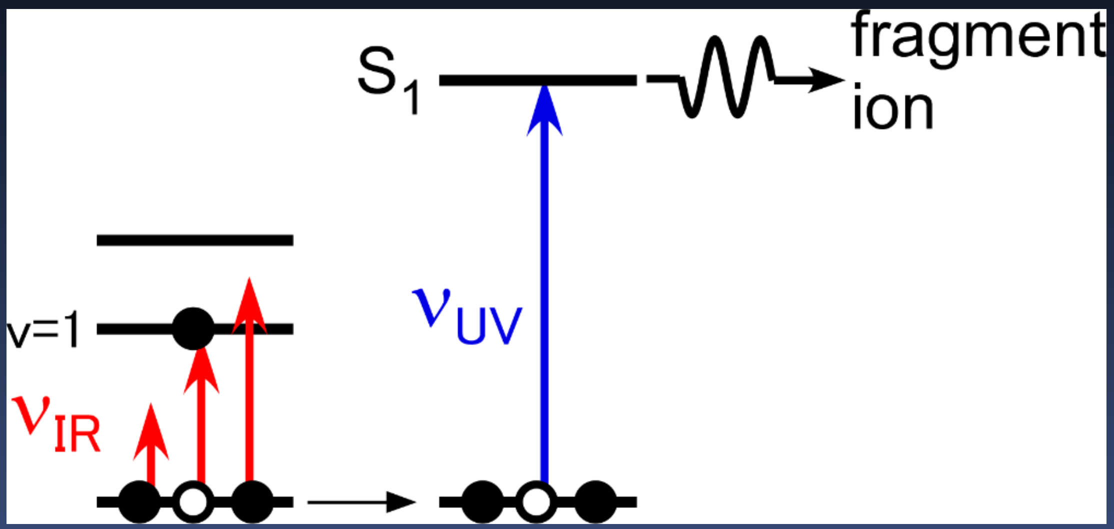
Two different vibronic spectra :

- Sharp transitions observed on m/z 131
- Broadened absorption band on m/z 188
- Sharp transitions further to the blue on m/z 188

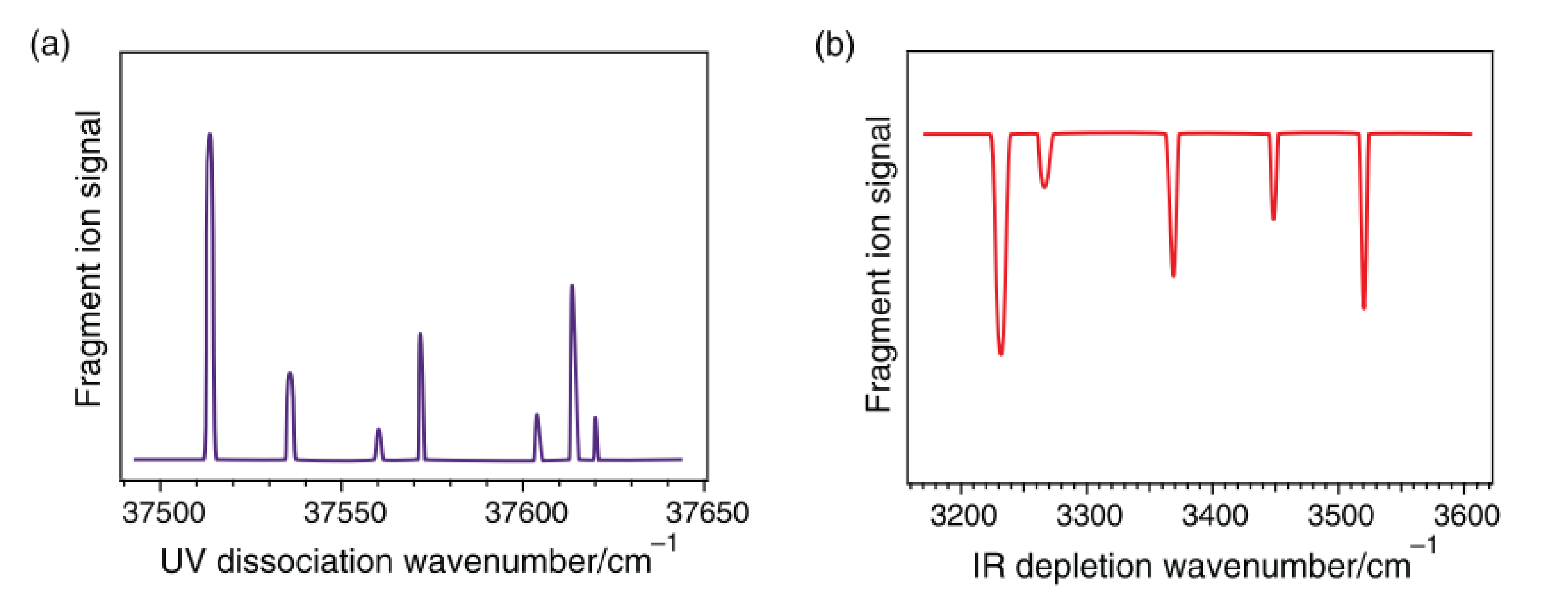
➢ **How many conformers ?**

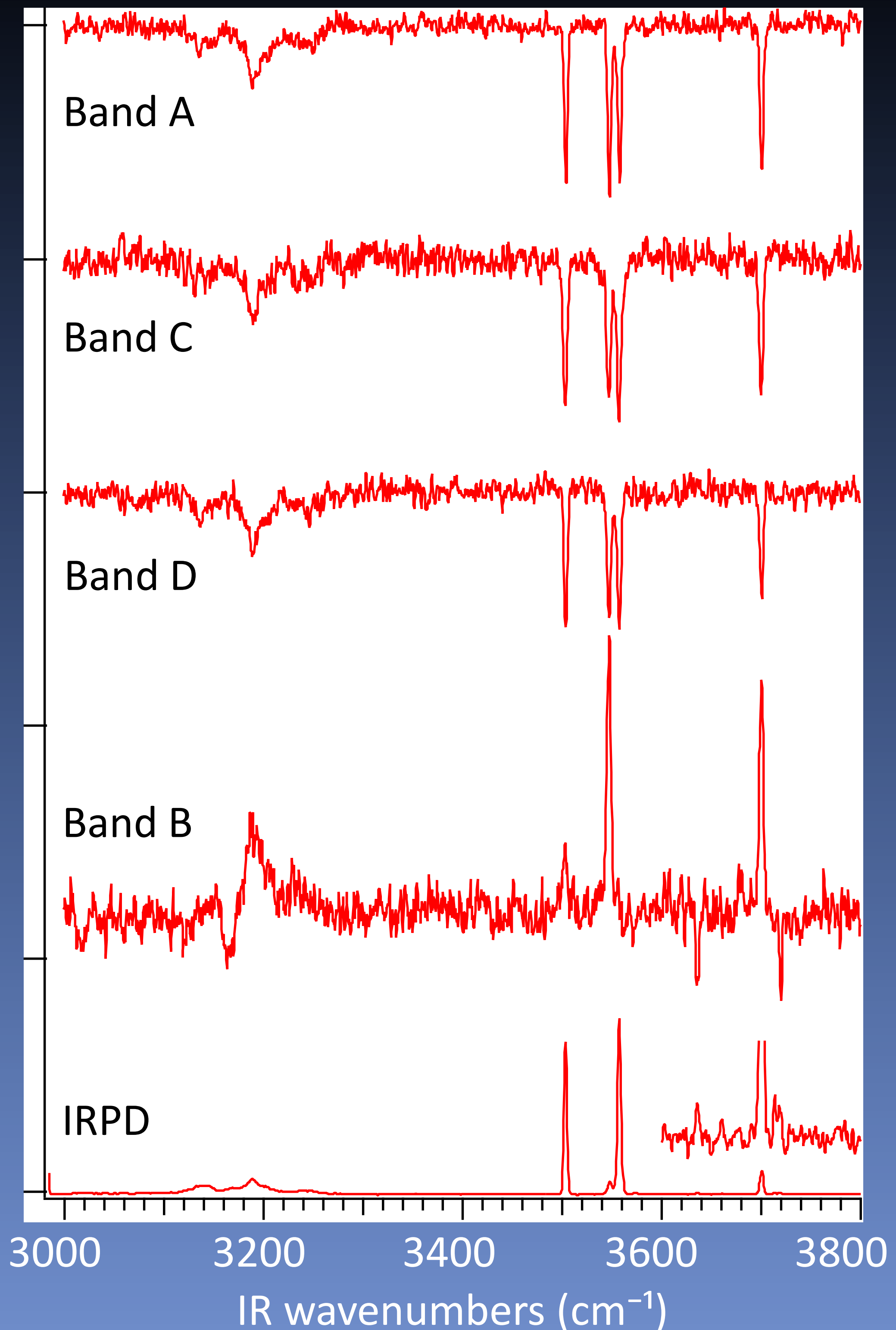
IR-UV dip spectroscopy

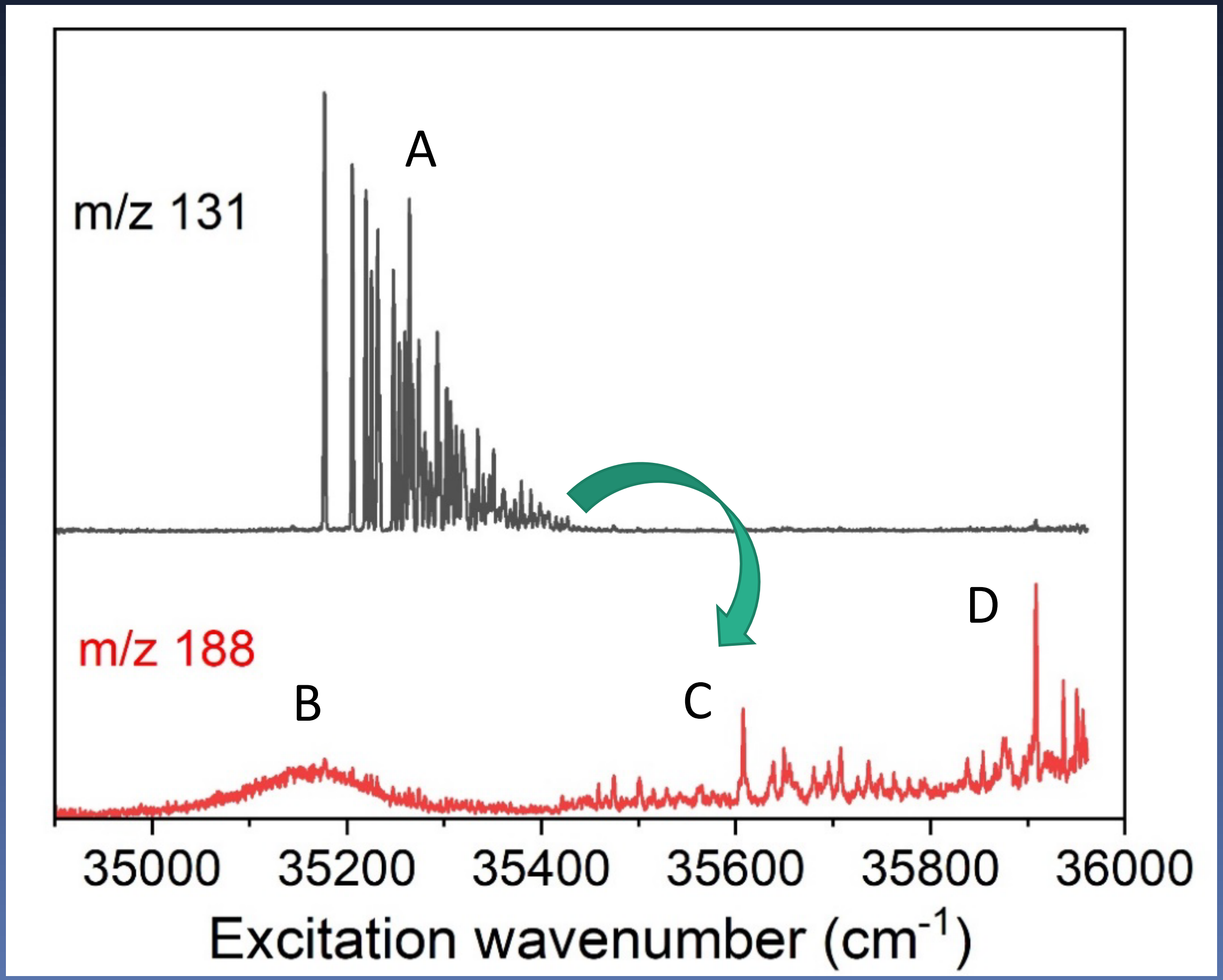




1. Set the UV on specific vibronic transition
2. Scan the IR laser before the UV
3. Record the IR dip spectrum



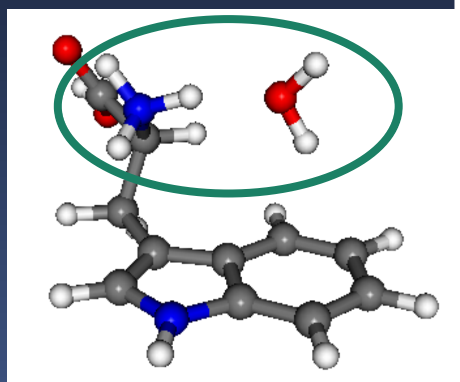




- Bands A/C/D belong to the same conformer
- Band B: IR different ➡ Two conformers
- Fragmentation branching ratio change within 300 cm^{-1}

Conformer assignment

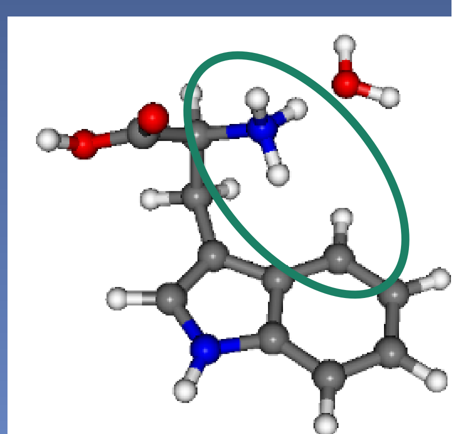
DFT : B3LYP-D3 / cc-pVTZ

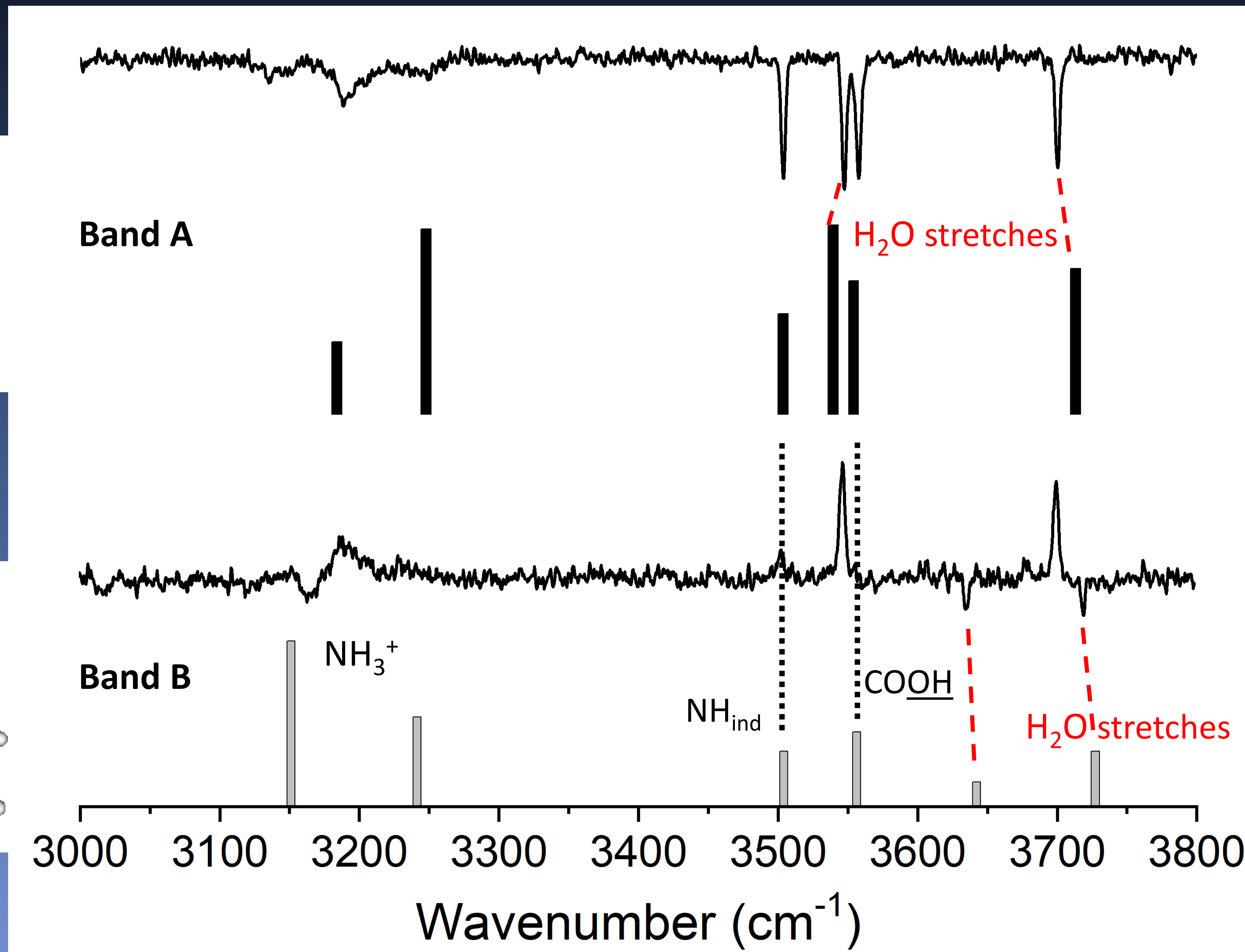


0

ΔG_{180K}

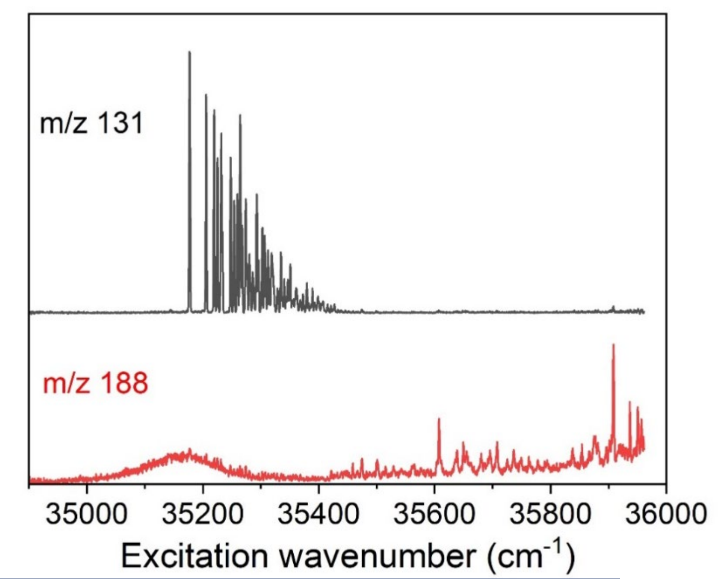
1,4 kcal/mol

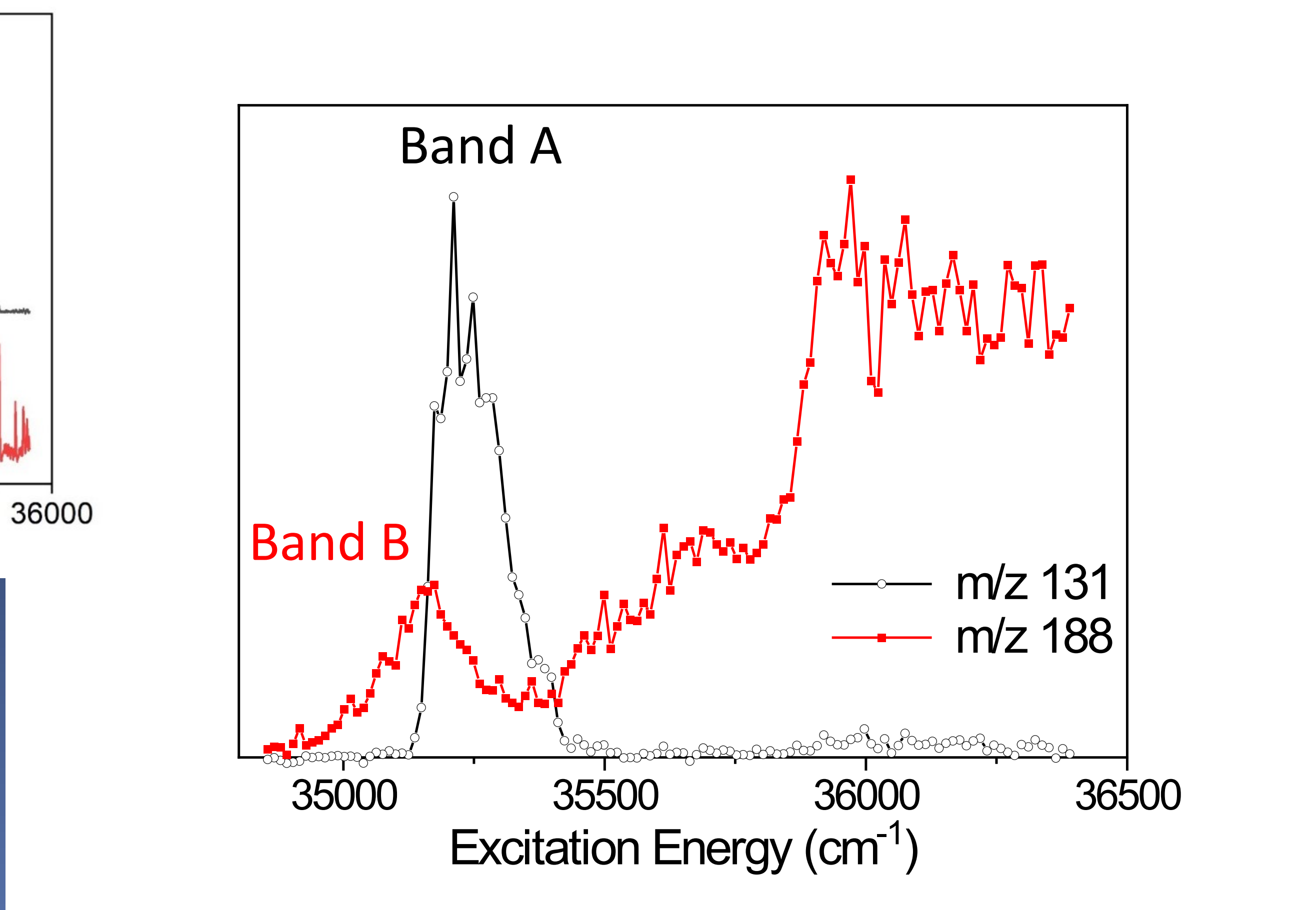




Picosecond pump-probe photodissociation spectroscopy

ps laser : Spectral resolution too low (10 cm^{-1}) to observe vibronic transitions but still able to discriminate band A (black, m/z 131) and band B (red, m/z 188)





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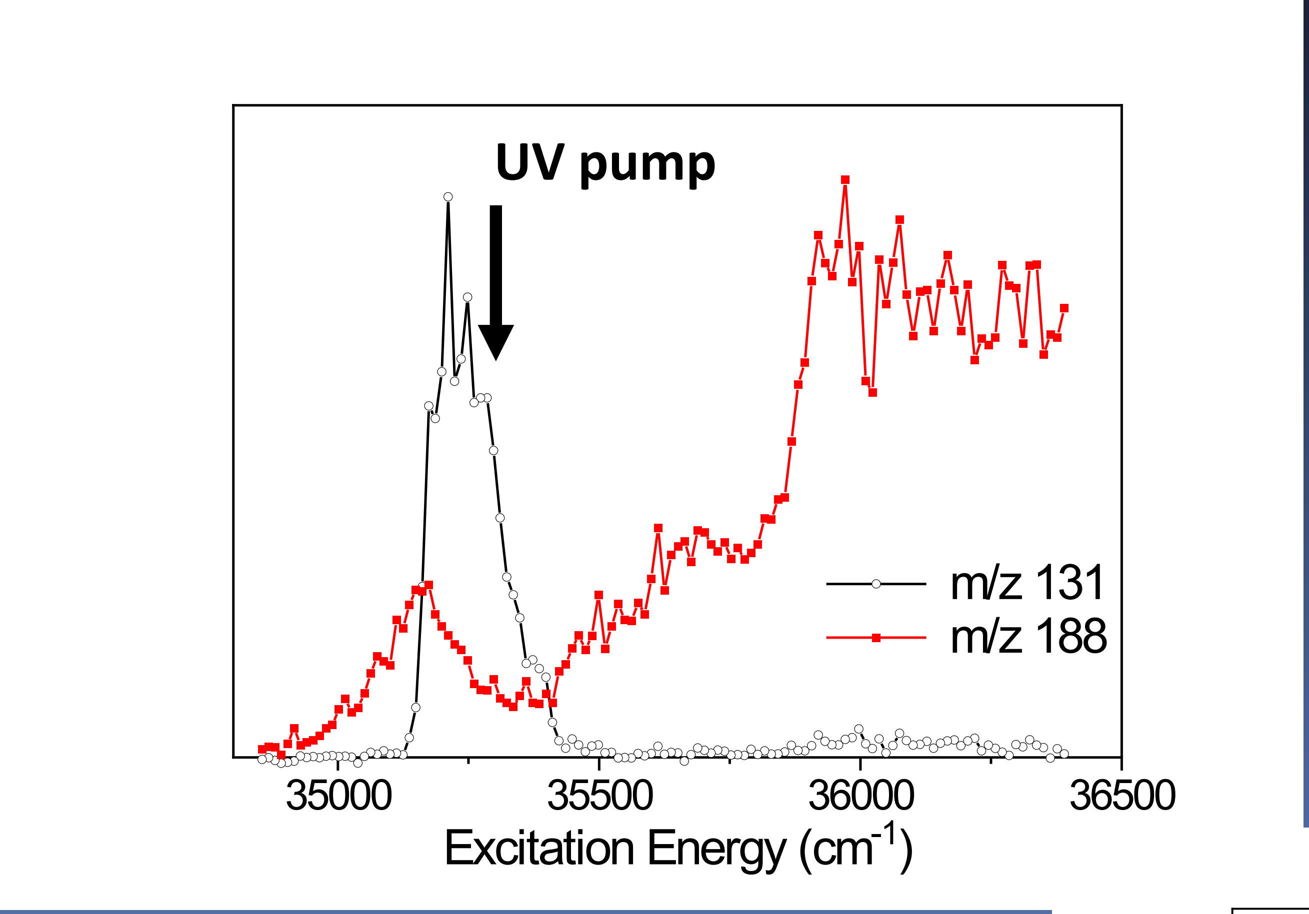
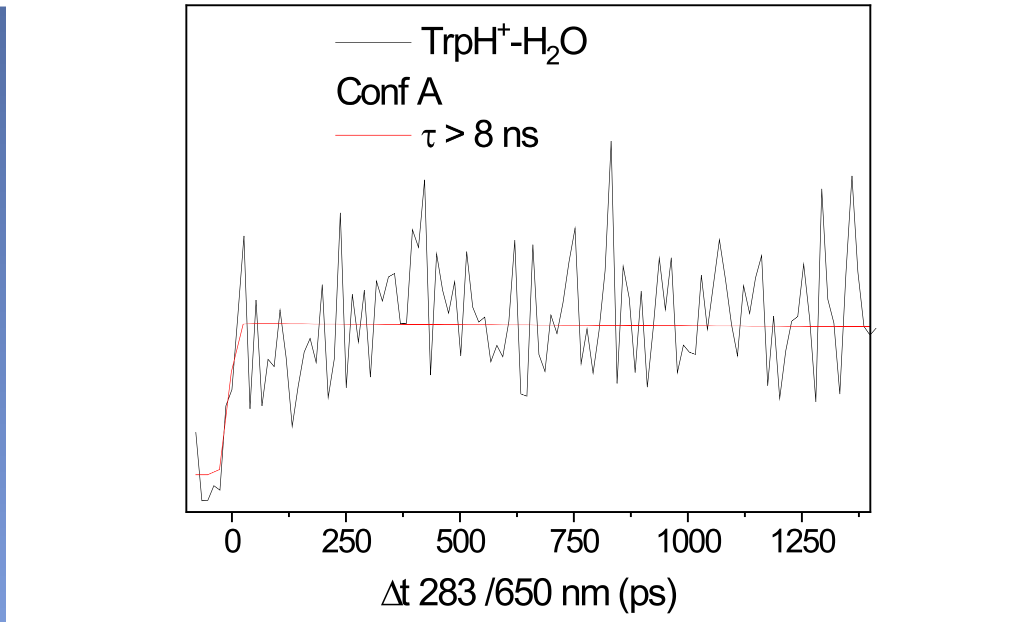


ns lifetime :
Consistent with sharp vibronic spectrum



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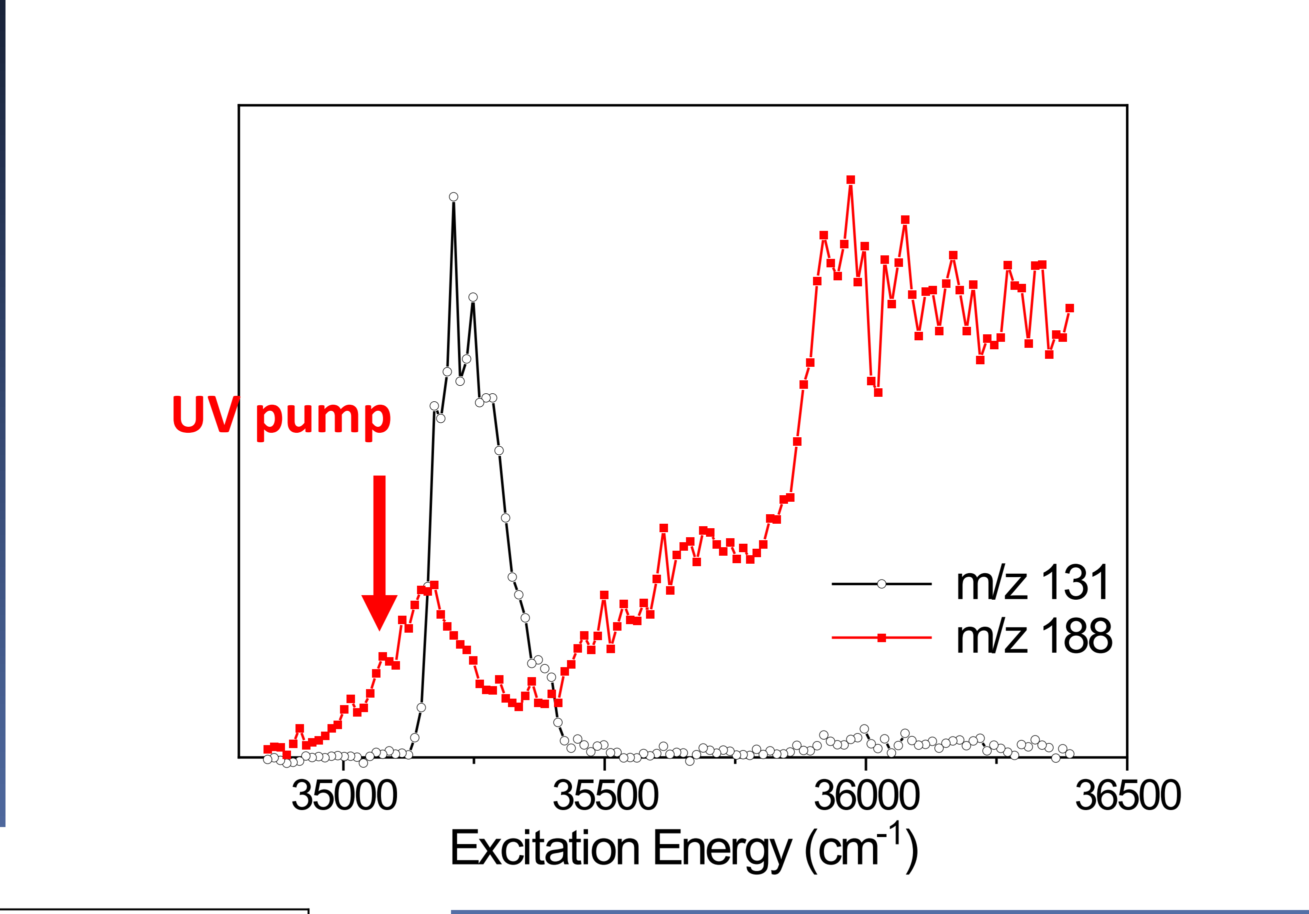
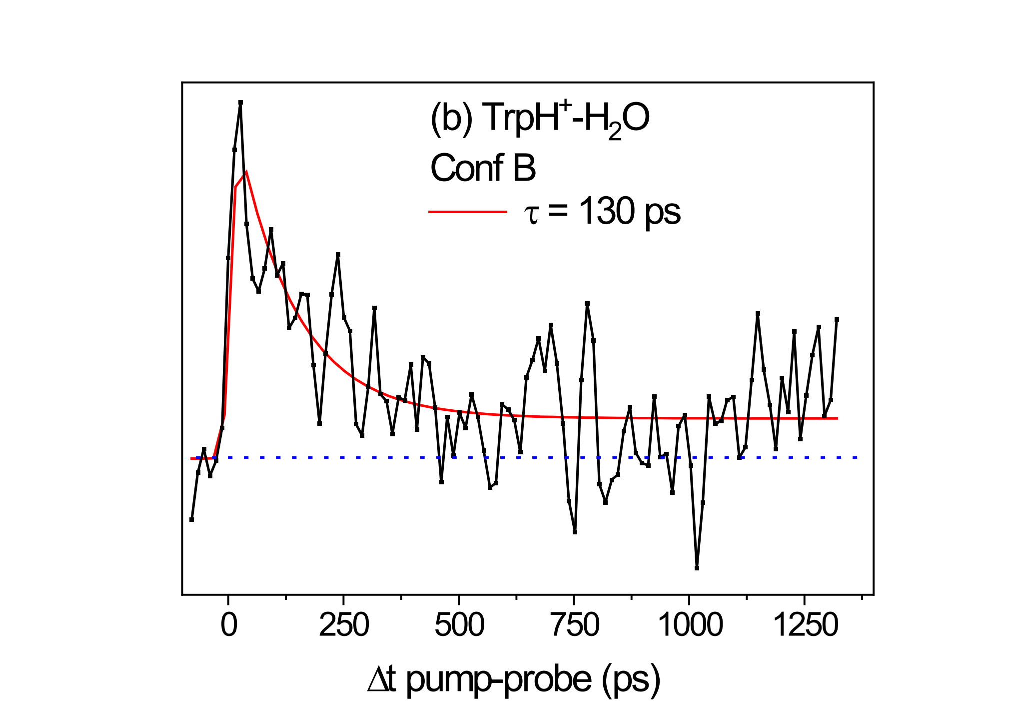


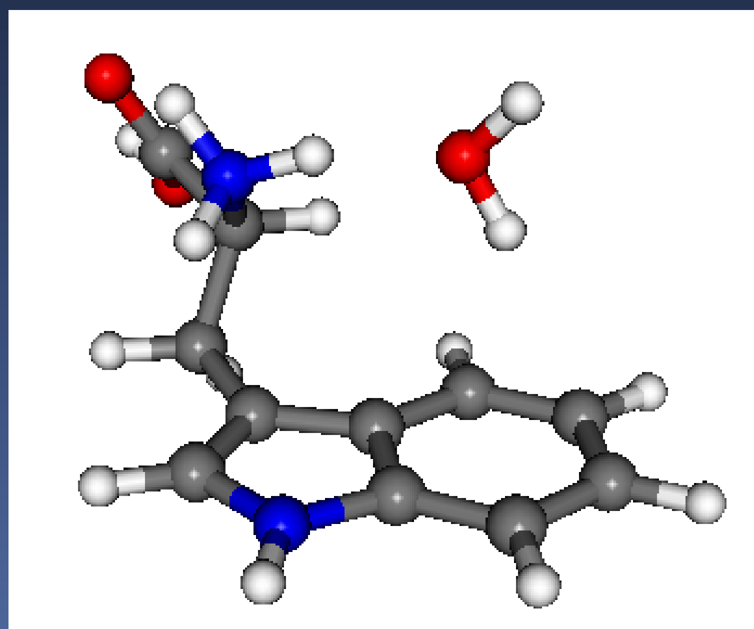
100 ps lifetime
as for the ESPT form



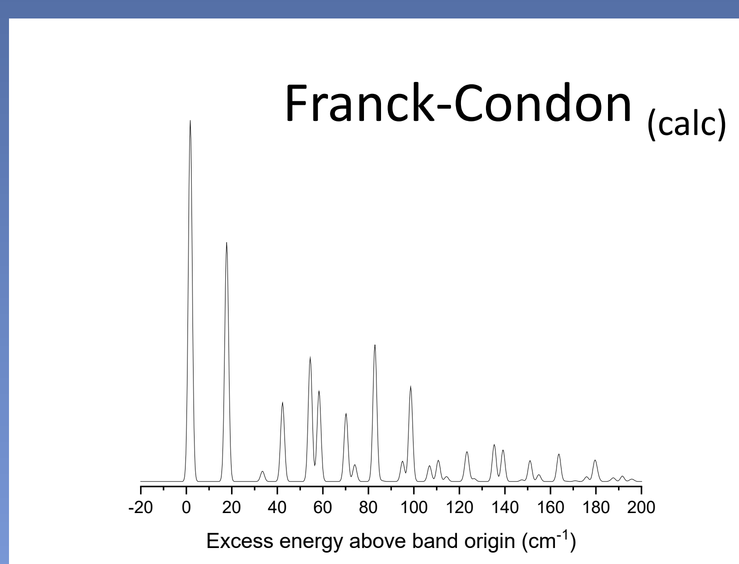
$TrpH^+$-H_2O : conformer-specific photodynamics

CONF A : sharp vibronic transitions
Water inserted between the indole and NH_3^+

ESPT blocked, ns lifetime of the $\pi\pi$ * state



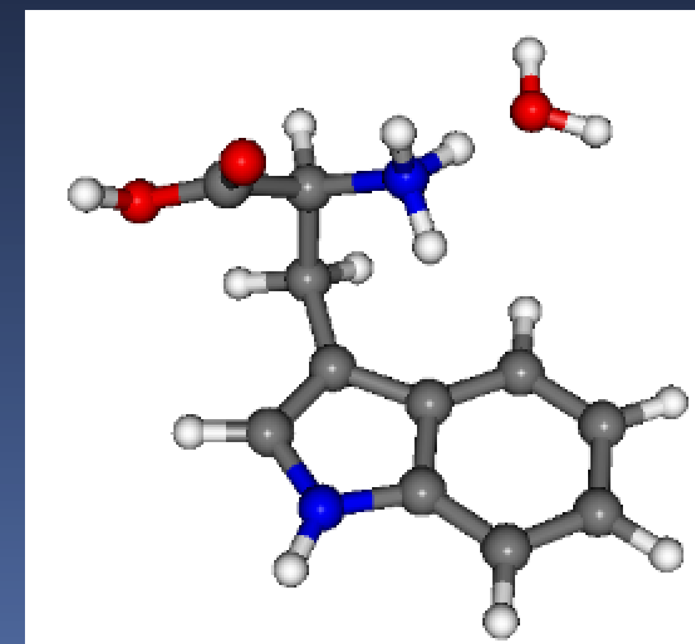
0_0^0 $_{(calc)}$ = 4,36 eV vs 4,31 ev (exp)



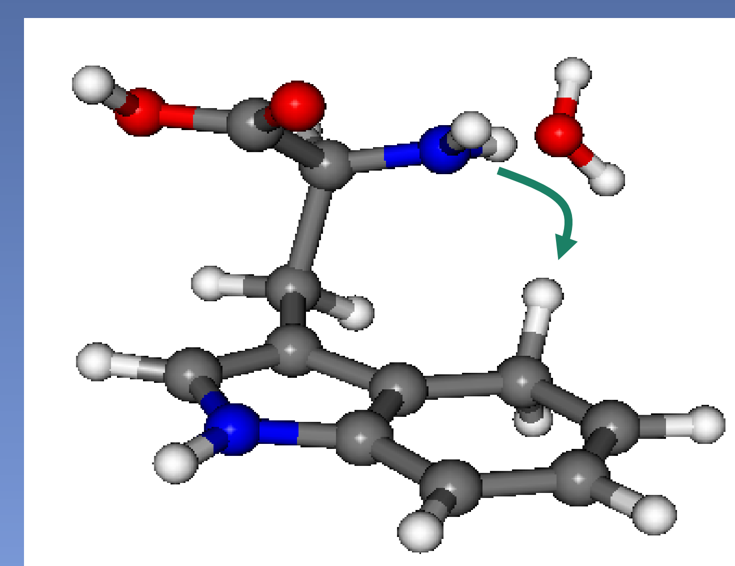
CC2-SCS / aug-cc-pVDZ level
Opt + freq (S_0 and $\pi\pi$* states)

CONF B: broadened excitation spectrum
NH_3^+ pointing toward indole

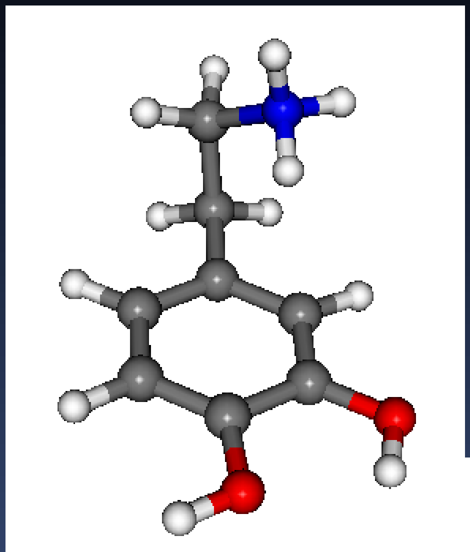
Prone for ESPT



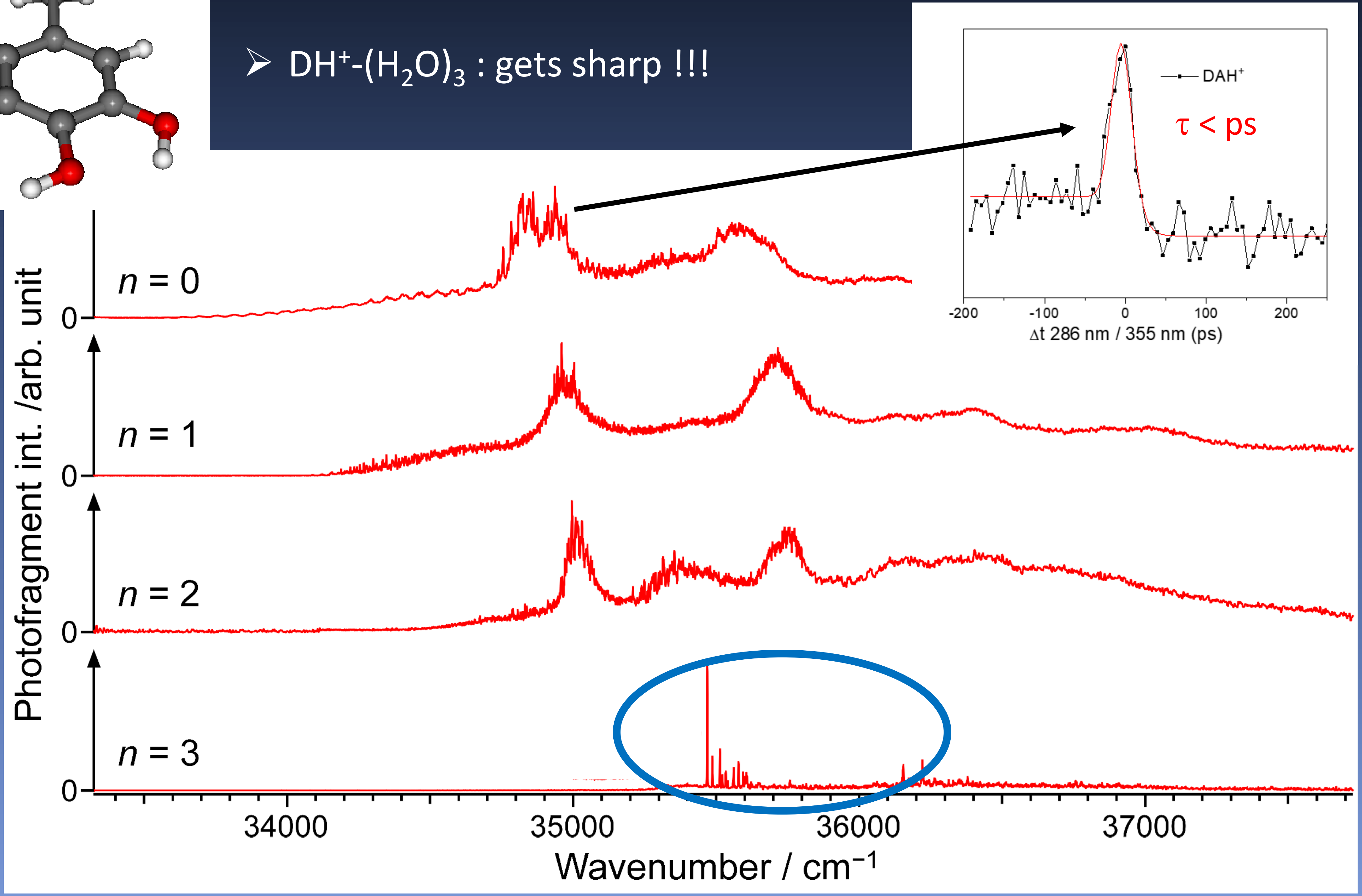
Barrierless ESPT



Dopamine Water Clusters
$DH^+\text{-}(H_2O)_{n=0\text{-}3}$

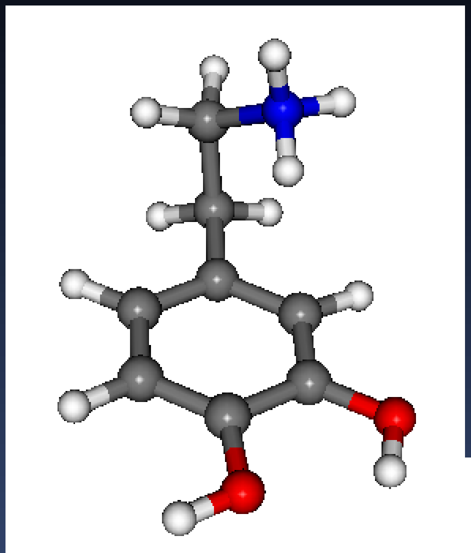


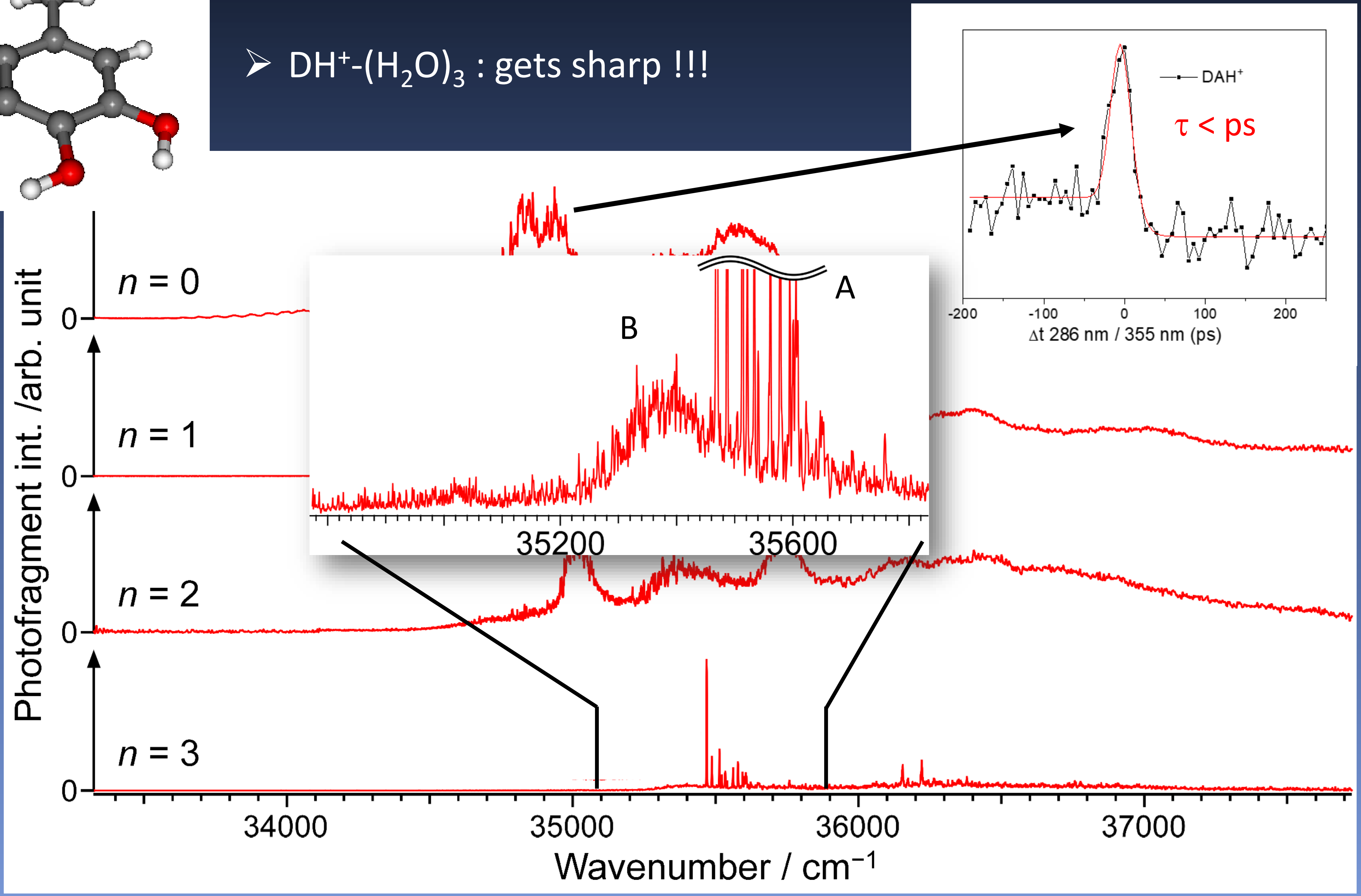
- $DH^+\text{-}(H_2O)_{n=0\text{-}2}$: Broadened excitation spectrum
- $DH^+\text{-}(H_2O)_3$: gets sharp !!!



Dopamine Water Clusters
$DH^+-(H_2O)_{n=0-3}$

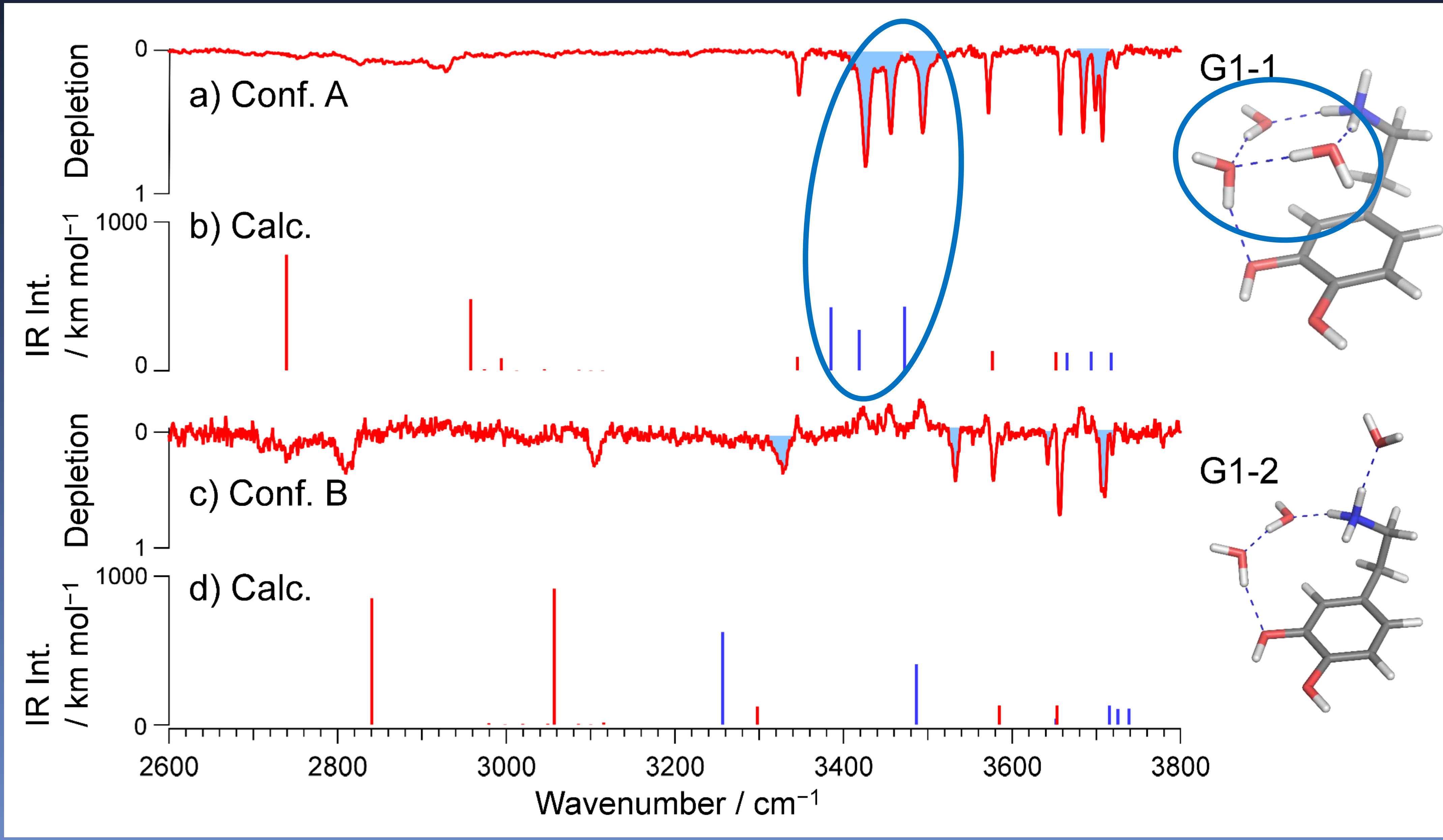
- $DH^+-(H_2O)_{n=0-2}$: Broadened excitation spectrum
- $DH^+-(H_2O)_3$: gets sharp !!!





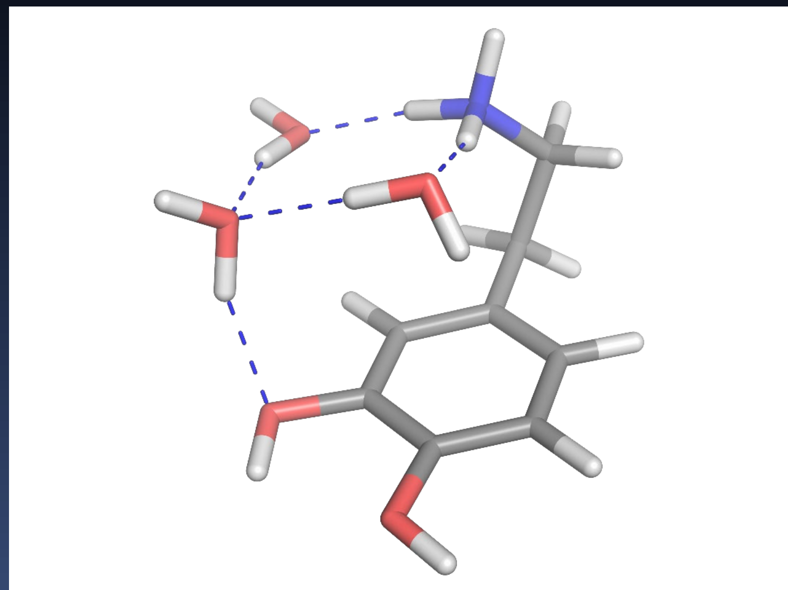
DH^+-$(H_2O)_3$: 2 conformers

IR-UV dip spectroscopy + B3LYP-D3/cc-pVTZ calculations

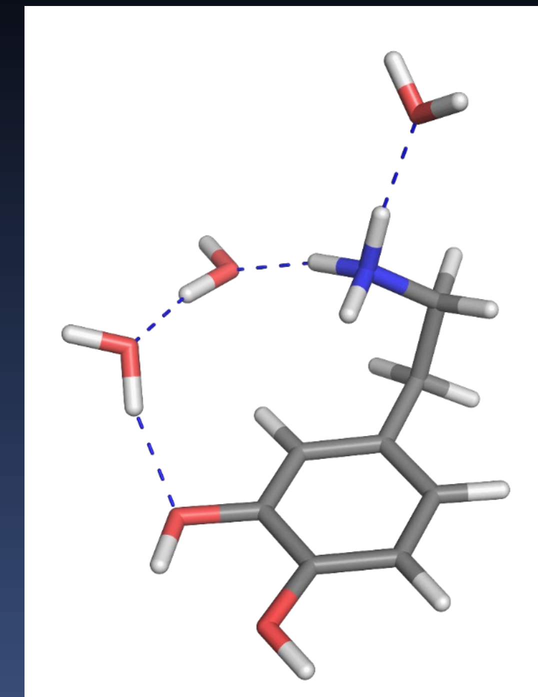


- ✓ Conf A : G1-1 : cyclic water cluster, prevent NH_3^+ - catechol ring interaction
- ✓ Conf B : G1-2 : still one NH (NH_3^+) pointing towards catechol ring

DH^+-$(H_2O)_3$: conformer-specific photodynamics

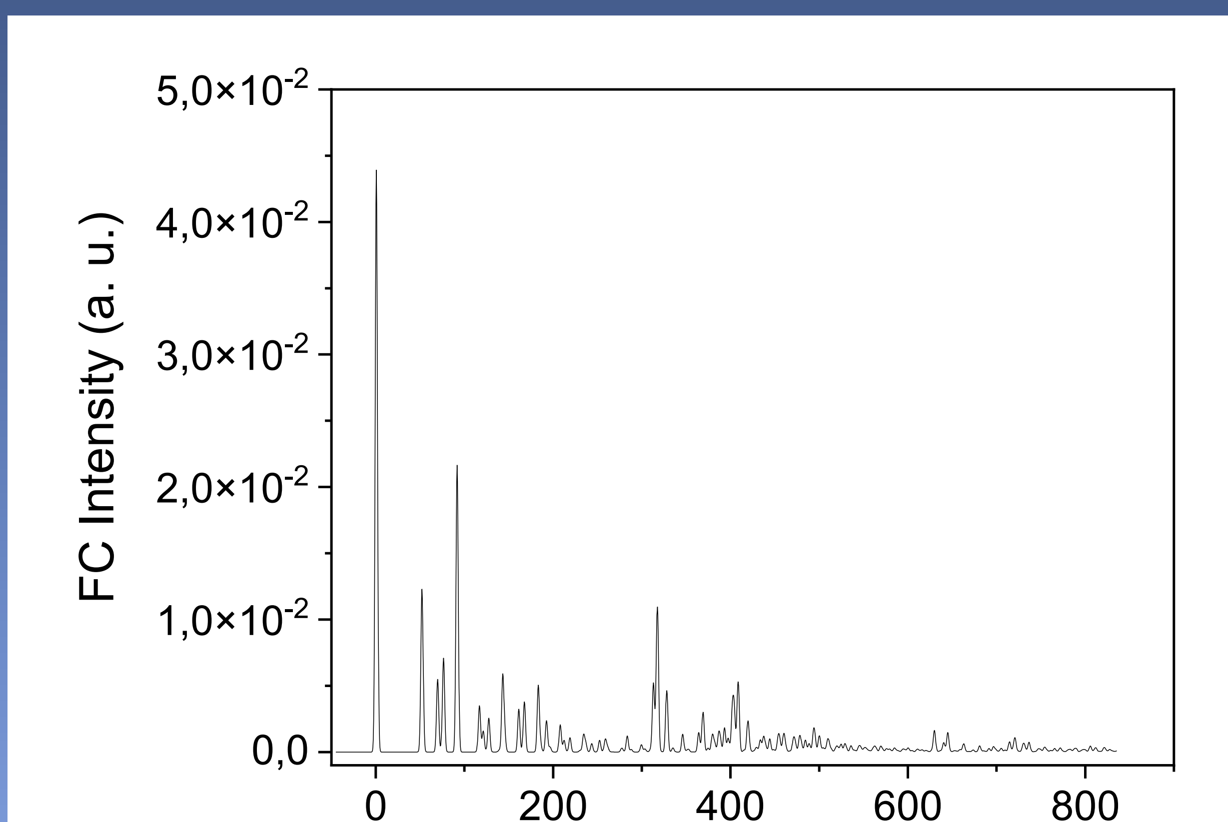


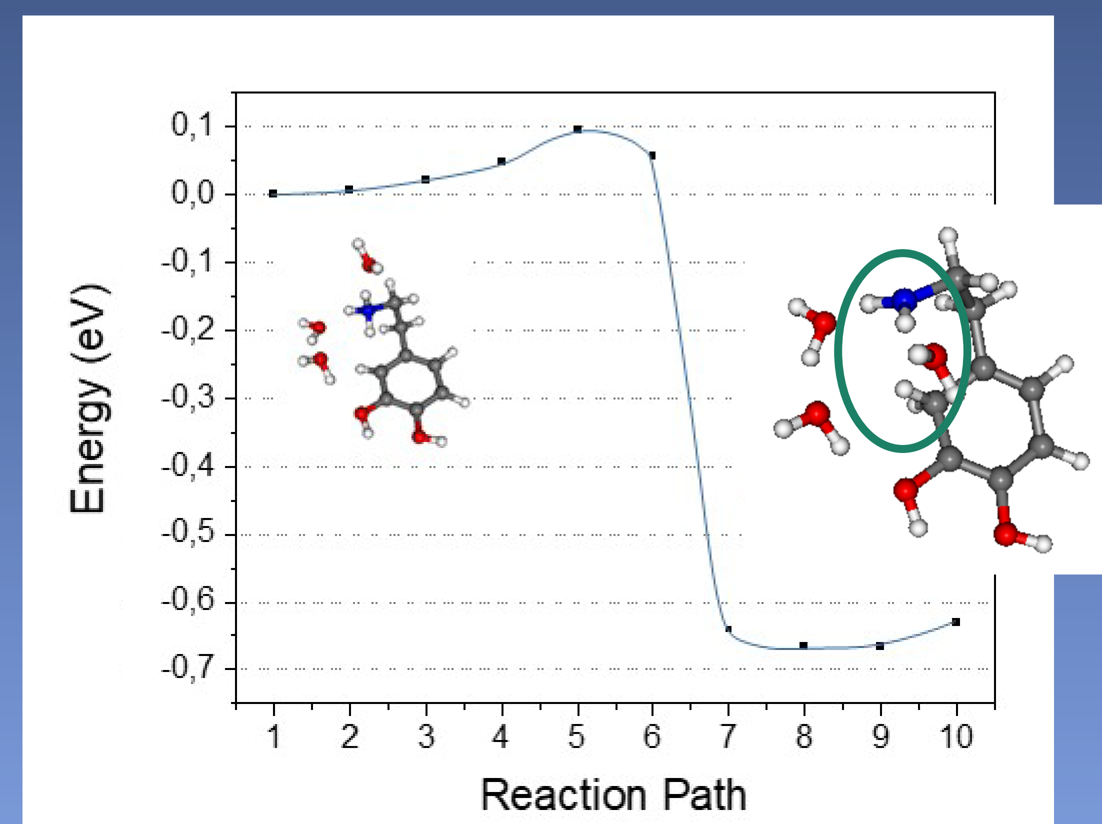
CC2-SCS / aug-cc-pVDZ level
Opt + freq (S_0 and $\pi\pi^*$ states)



Conformer A
Well-resolved FC spectrum

Conformer B
Small energy barrier for ESPT





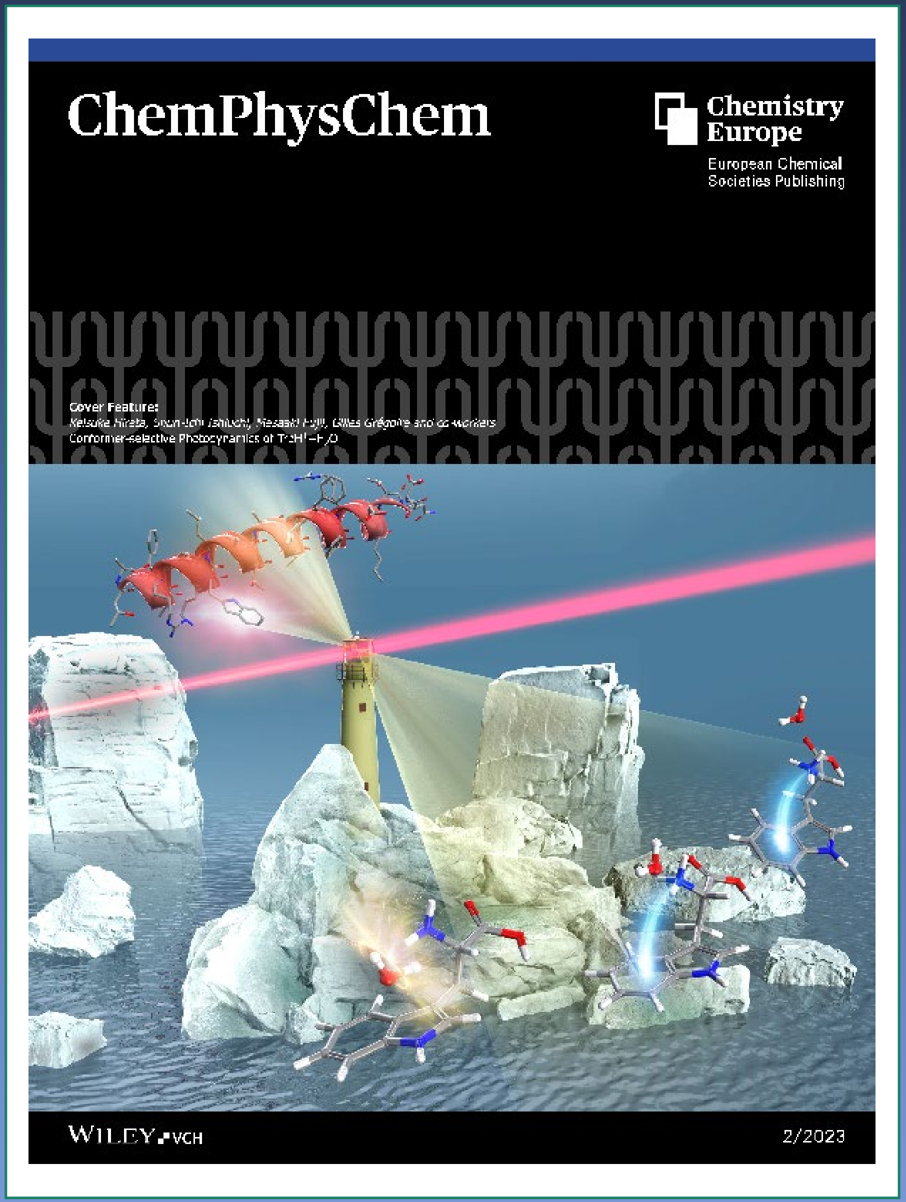
Conclusions

conformer-selective photodynamics of hydrated biomolecules

$TrpH^+$-$(H_2O)_n$

ESPT blocked by a single water molecule

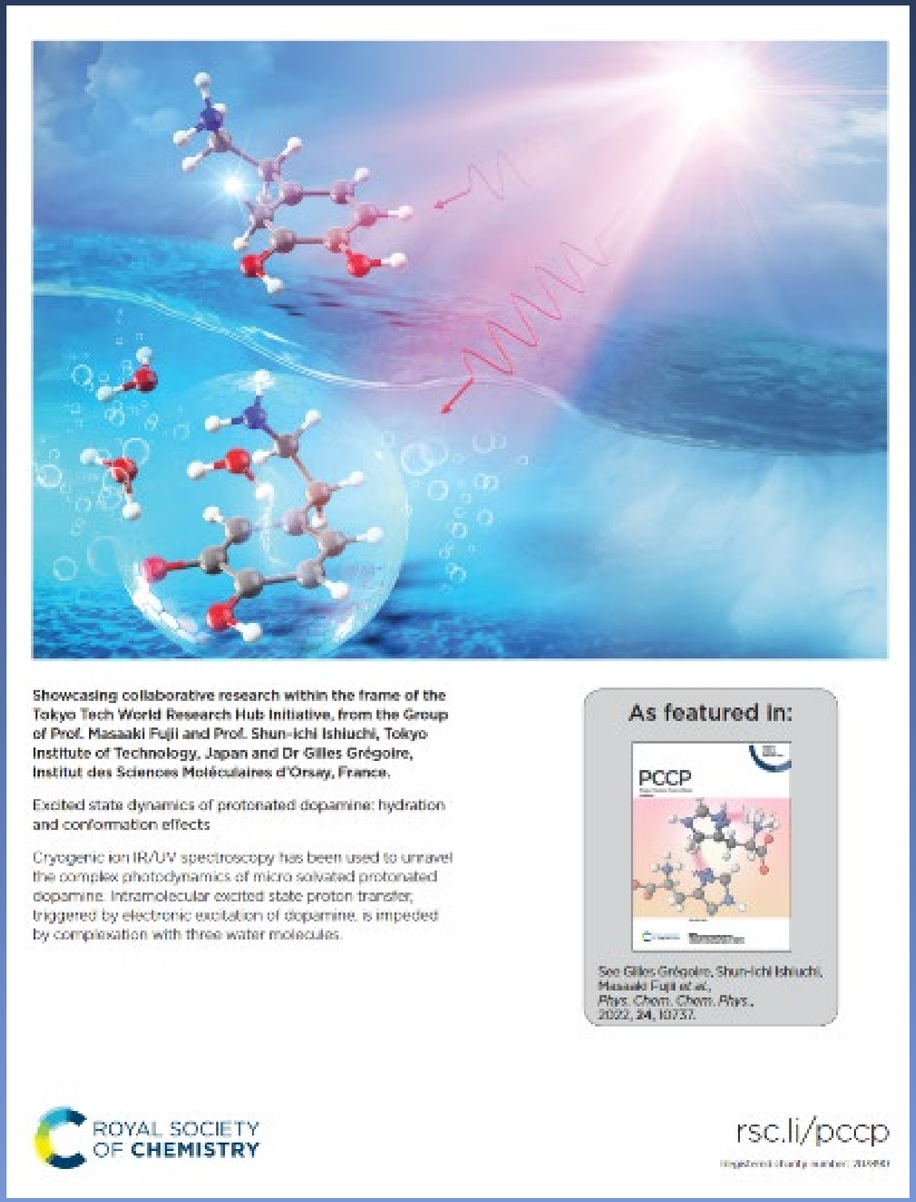
ChemPhysChem 2023



DH^+-$(H_2O)_n$

ESPT blocked in DH^+-$(H_2O)_3$ clusters

PCCP 2022 & JCP 2021 Editor Choice



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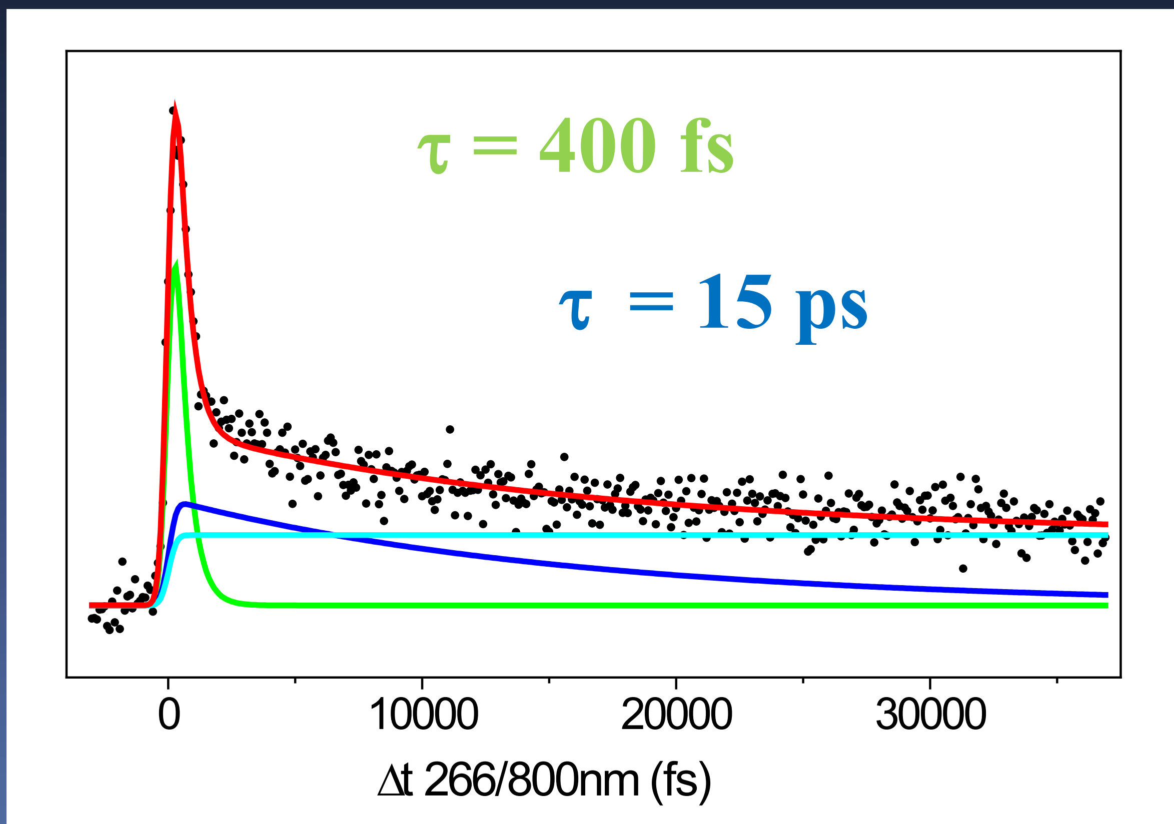
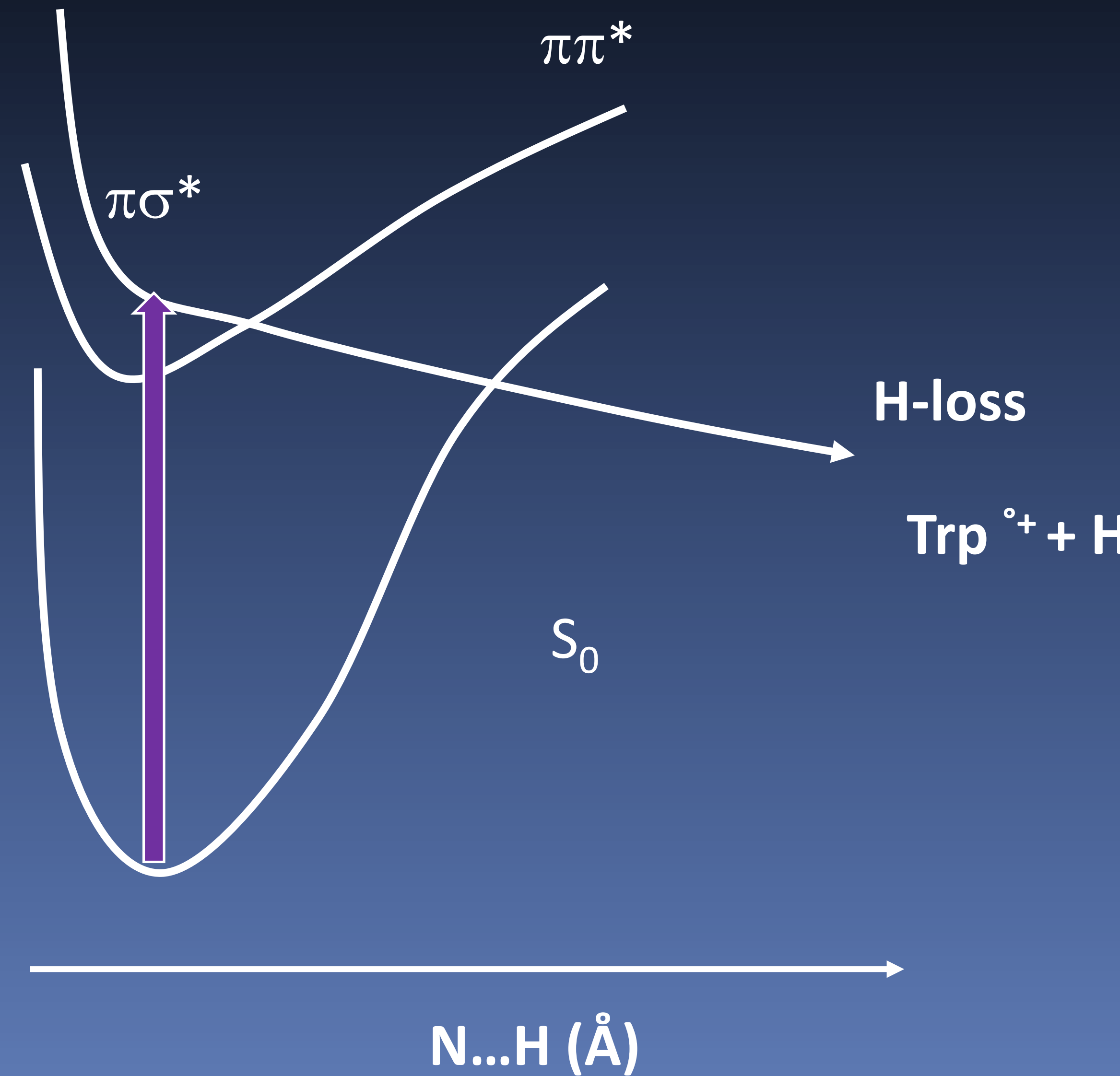
Thank you for your attention !!

In the Gas Phase : Excited state lifetime of « hot » $TrpH^+$ @ 266 nm fs pump-probe photodissociation (2005)



JPC A, ***109***, *2417 (2005)*
PCCP, ***7***, *394 (2005)*

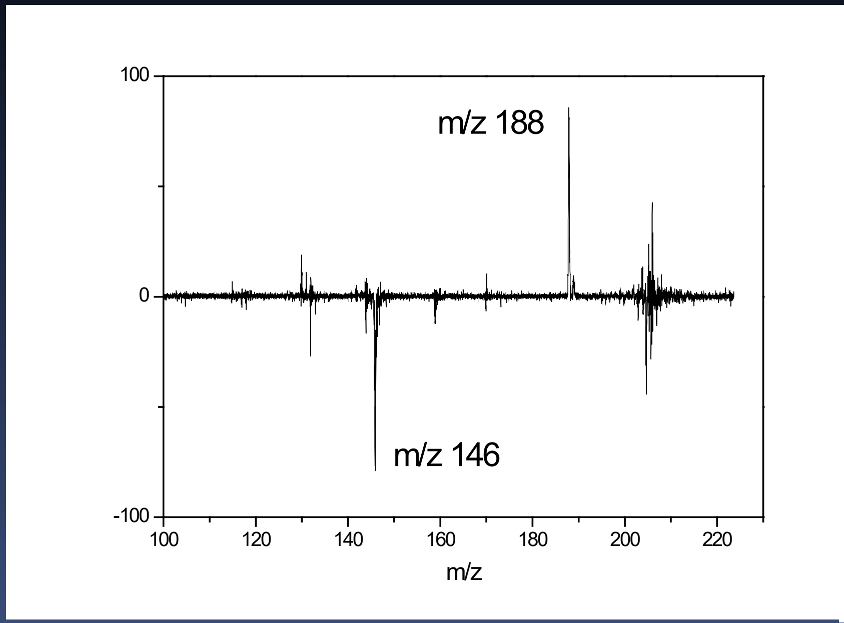


- **400 fs** : dynamics on the πσ* state and formation of the radical Trp °+ (plateau)
- **15 ps** ?

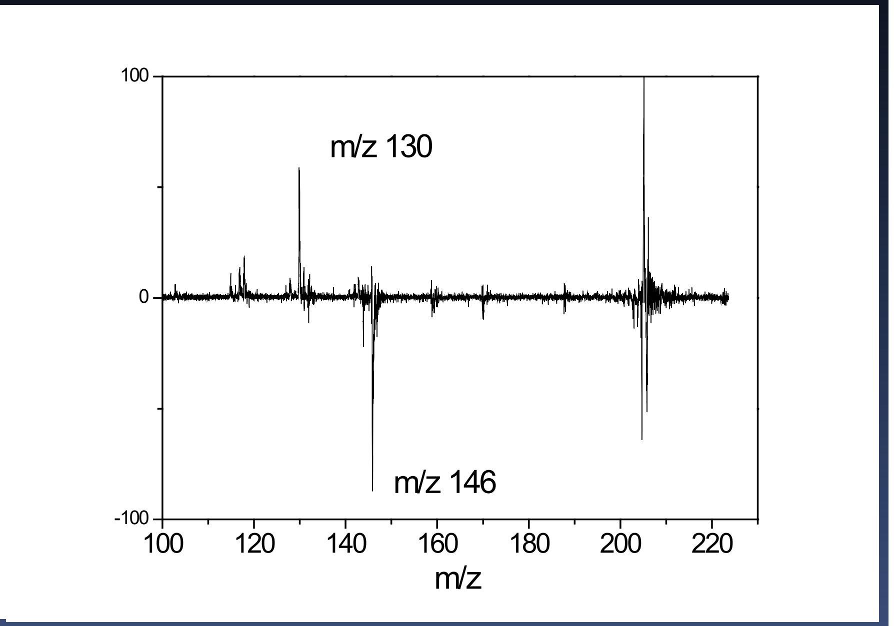
No conformer selection – Fixed pump wavelength

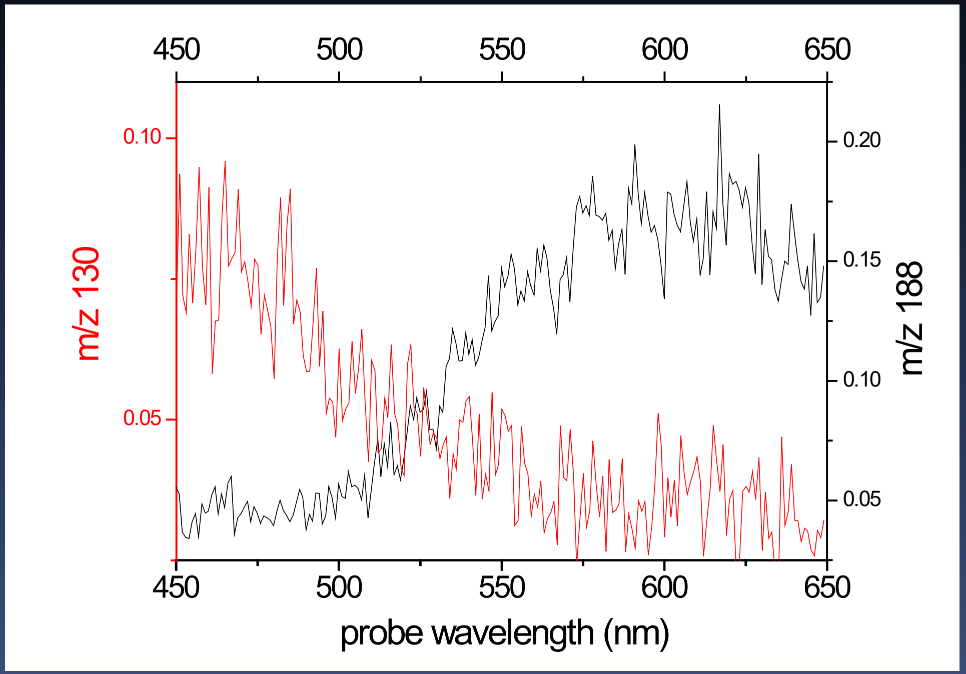
Effect of the probe wavelength

650 nm probe



450 nm probe

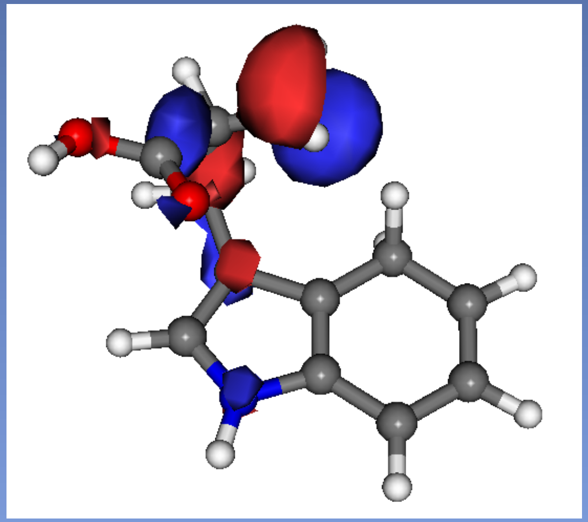


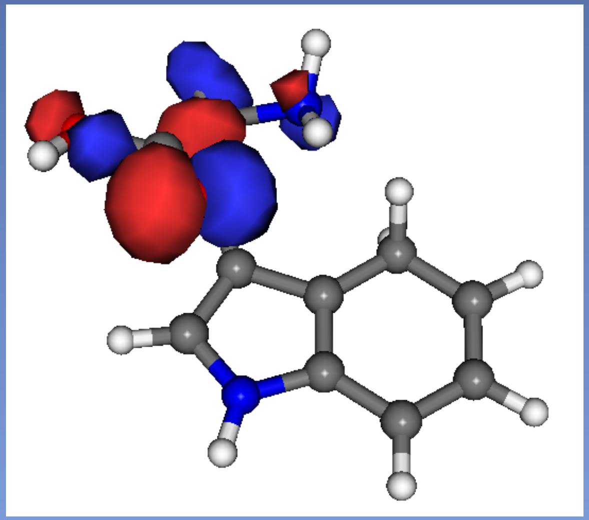


- Fragm. branching ratio changes with the probe wavelength:
  - → @ 650 nm : direct NH_3 loss (from an excited state)
  - → @ 450 nm : C_α-C_β bond cleavage

- Access to excited states of the ESPT structure

S_3 : + 1.7 eV
(740 nm)
$n_{NH2} - \pi^*$





S_4 : + 2.7 eV
(460 nm)
$n(O) - \pi^*$