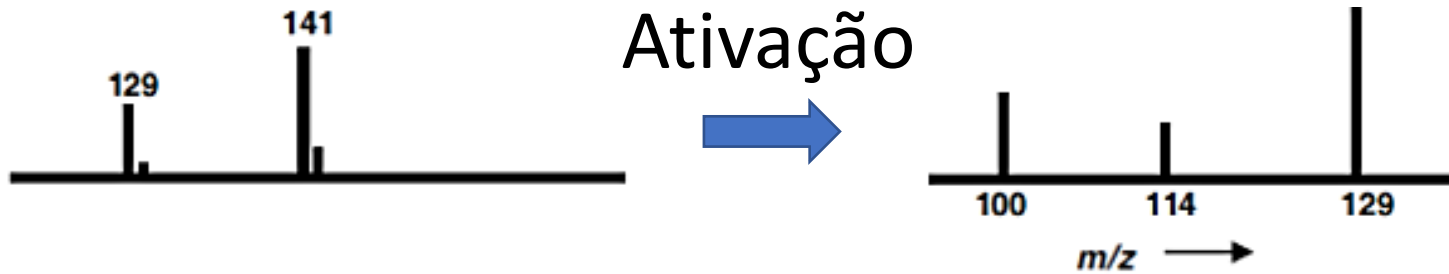


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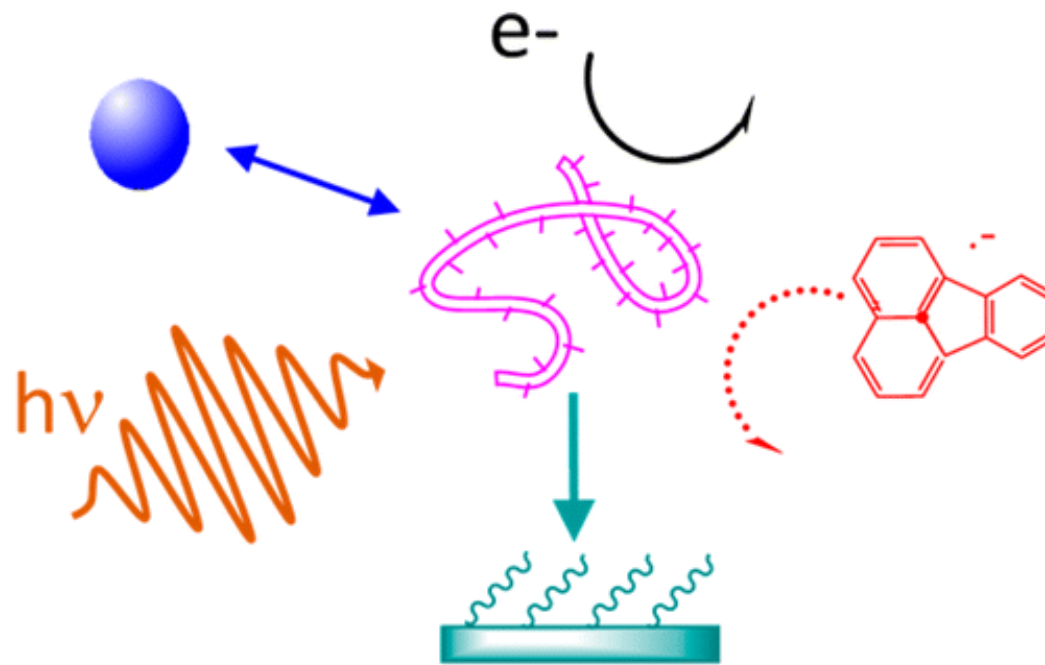
Prof. Dr. Thiago C. Correra

Métodos espectroscópicos acoplados à MS – Espectroscopia IRMPD

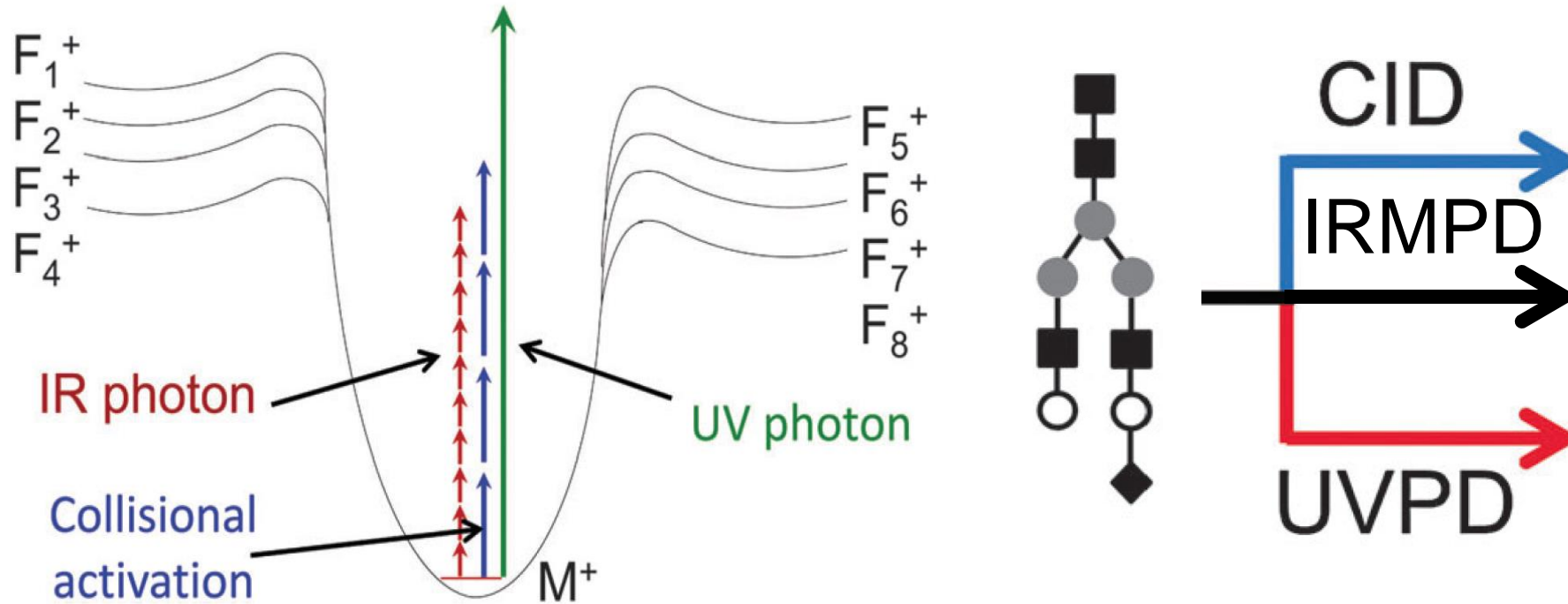
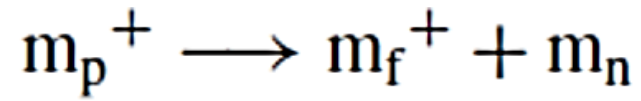
Métodos de ativação de isômeros



- Induzida por colisão
- Induzida por superfície
- Induzida por elétrons
- ...
- Fotoinduzida

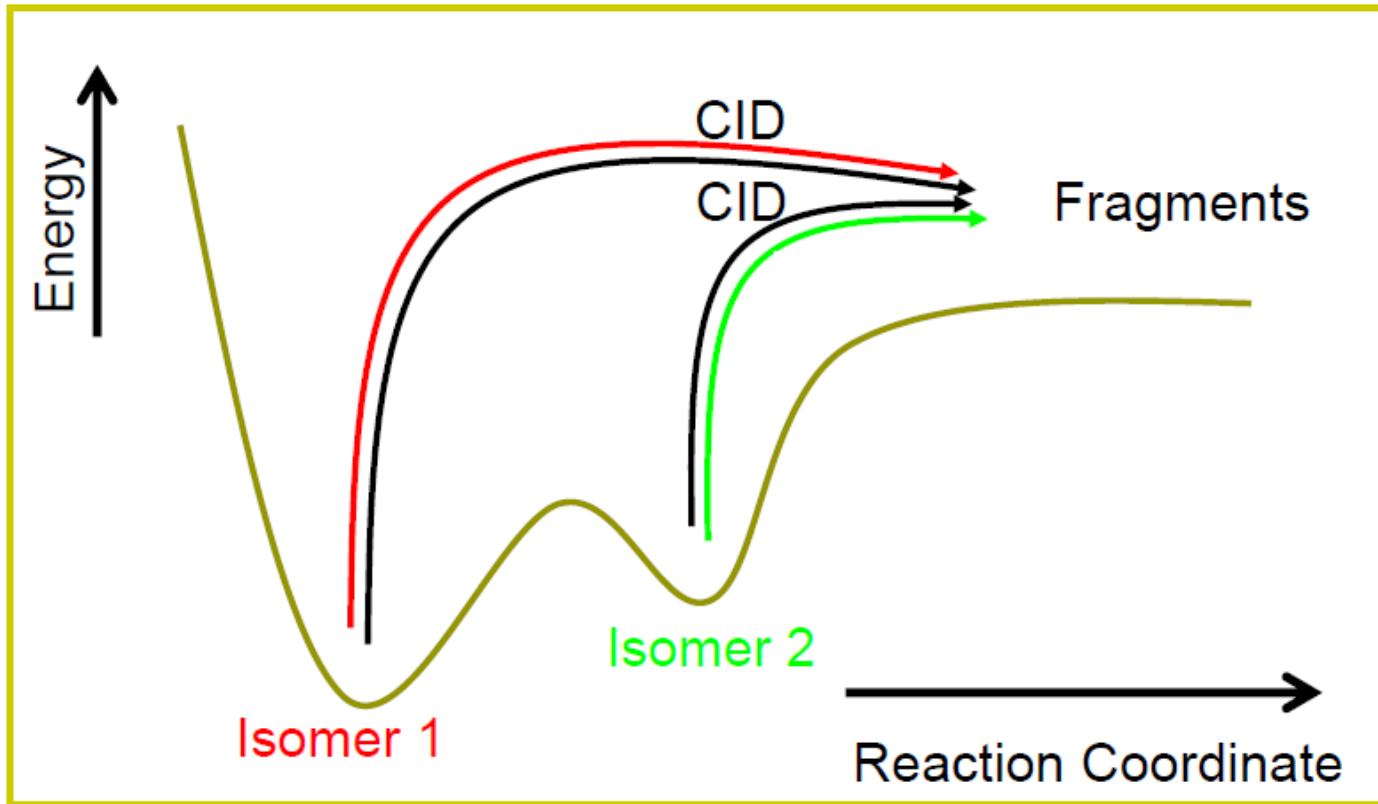


CID, UVPD and IRMPD



Riveros, J. M.; Cap. 4 - Infrared Photodissociation, Encyclopedia of Mass Spectrometry, Vol. 1, Amsterdam, **2003**.
Brodbelt, J. S.; *Chem. Soc. Rev.* **2014**, 43 (8), 27575–2783.

- Need for structural information in MS/MS
 - Fragmentation mass Spectra, especially EI
 - Infrared Spectroscopy could be useful
- IR spectroscopy integrated to MS
 - It relies on SPECIFIC fragmentation of an **ion isomer** based on SPECIFIC activation



CID : non specific activation

Isomer 1 $\xrightarrow{\text{CID}}$ Fragments

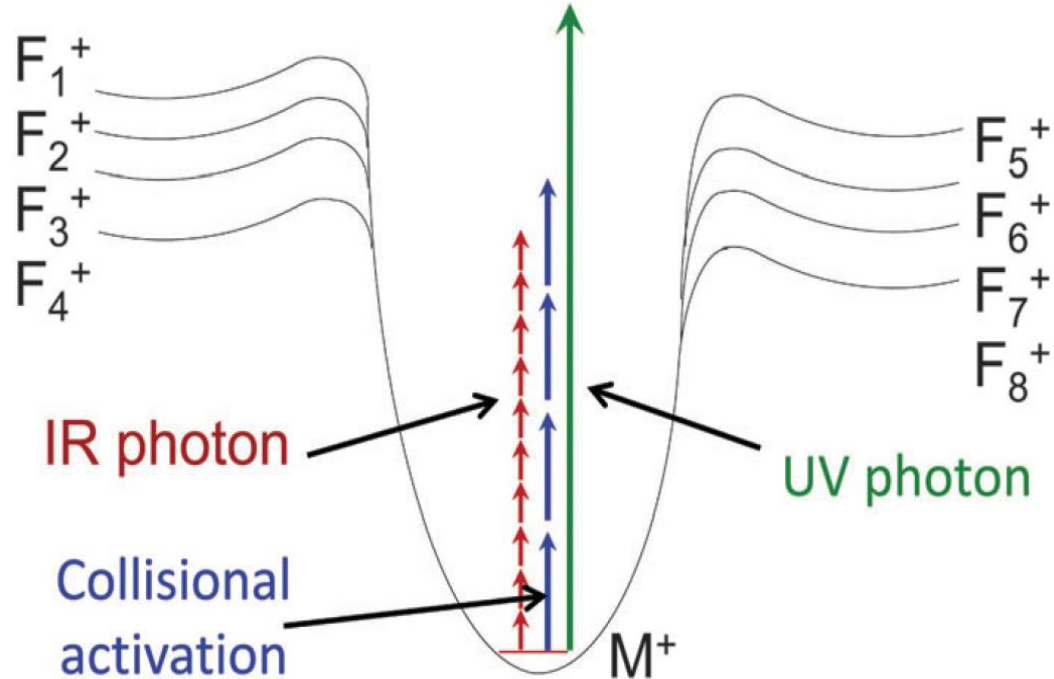
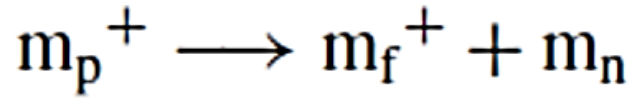
Isomer 2 $\xrightarrow{\text{CID}}$ Fragments

IR : specific

Isomer 1 $\xrightarrow{\text{IR } (\lambda_1)}$ Fragments

Isomer 2 $\not\xrightarrow{\text{IR } (\lambda_1)}$

Espectroscopia de íons



Avaliação da taxa de dissociação em função do comprimento de onda do fóton gera uma medida indireta da absorção dos fótons

Faixas espectrais (e fontes de radiação) diferentes podem ser utilizadas, com mecanismos diferentes:

- IR, UV, UV de vácuo

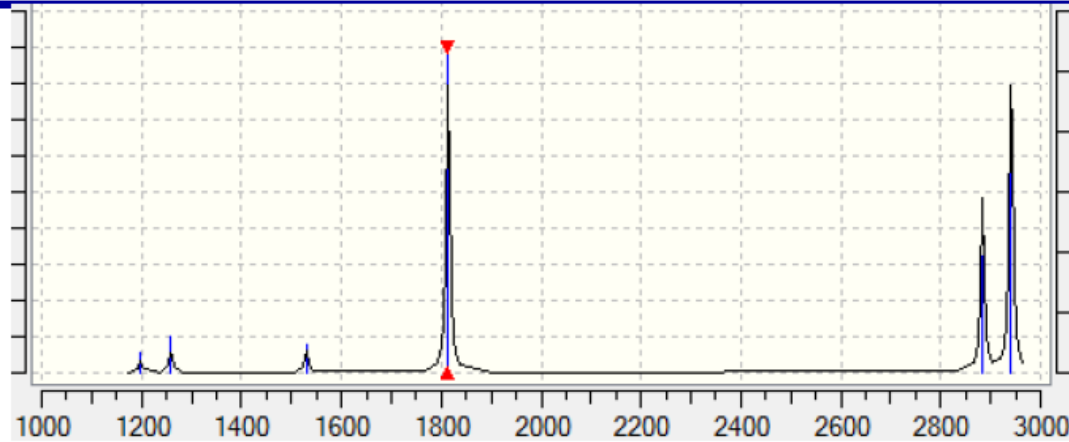
Em técnicas diversas:

- Dupla ressonância (IR-IR), FRET, fótons em faixas espectrais diferentes (UV-IR, UV-IR²)

Fontes diversas:

- OPO/OPA, FEL, Sincrotron (sobretudo para PEPICO)

Absorção no IR



Units used:

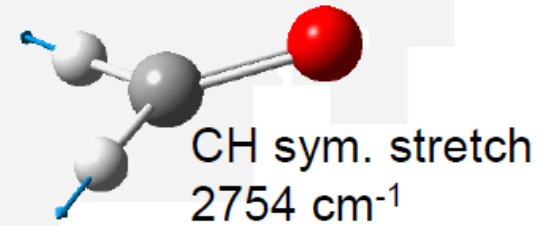
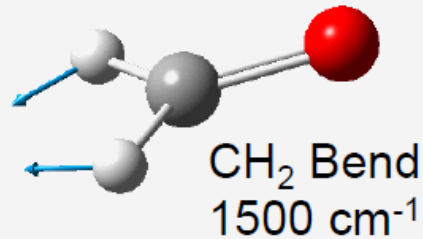
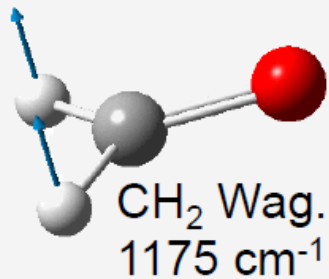
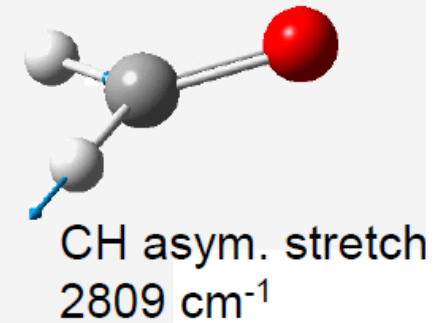
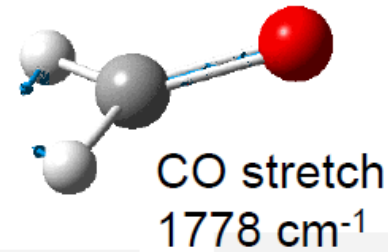
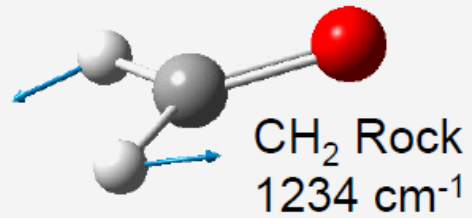
$$E = h \nu$$

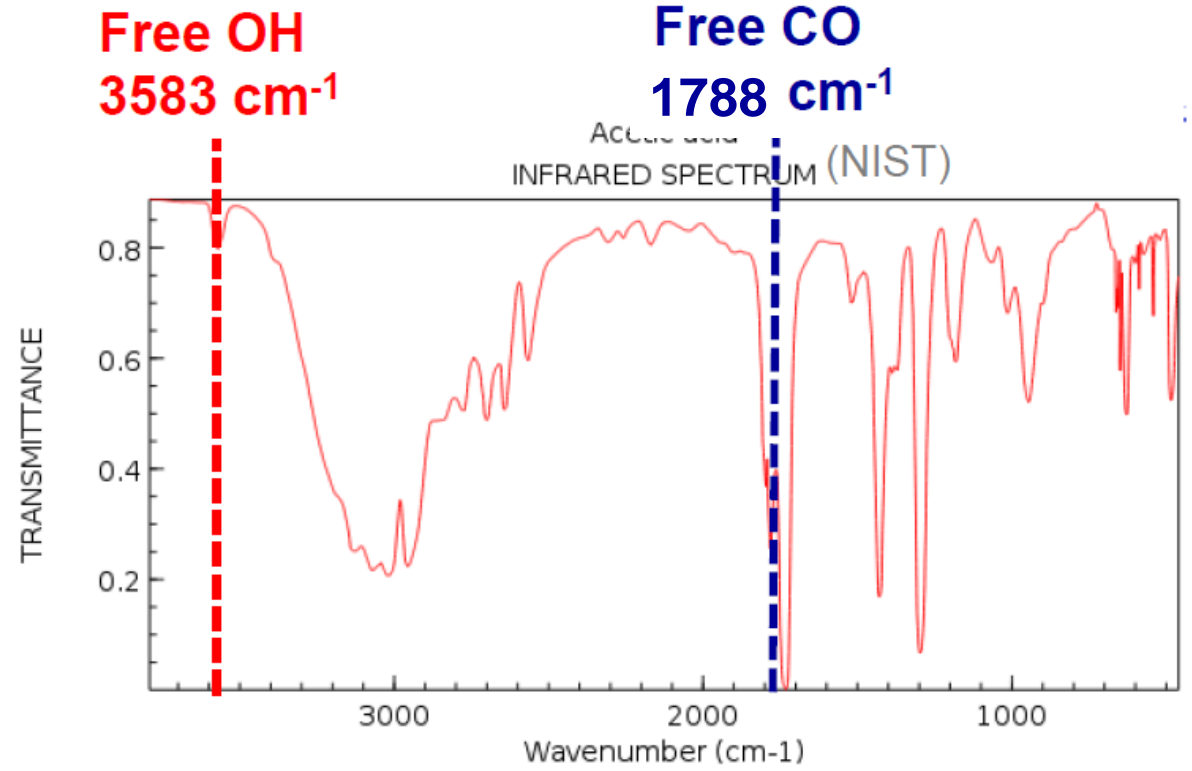
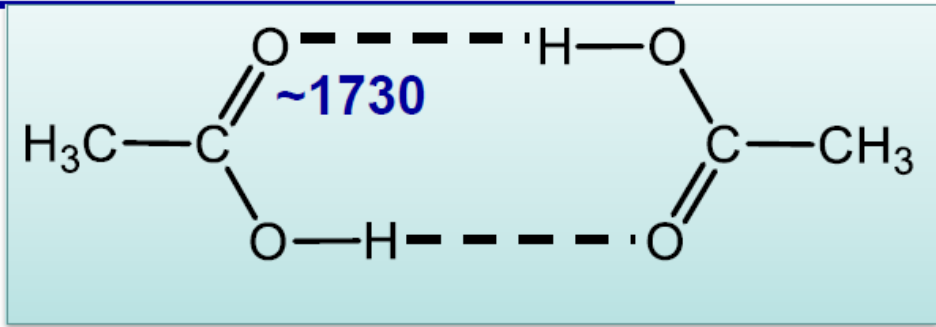
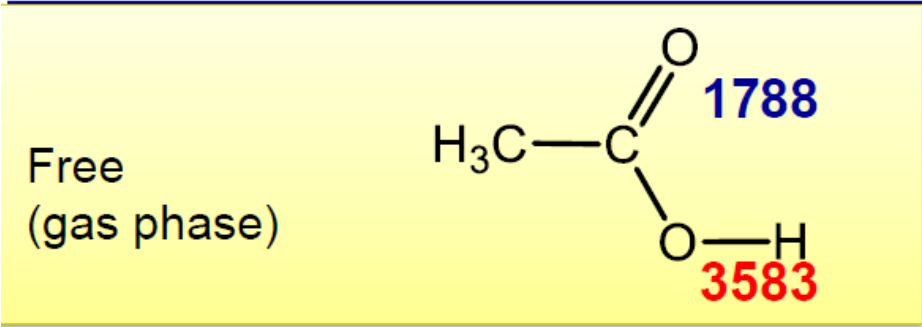
$$= h * C * 1/\lambda$$

$$= h * C * \bar{\nu}$$

where λ is the wavelength (μm)

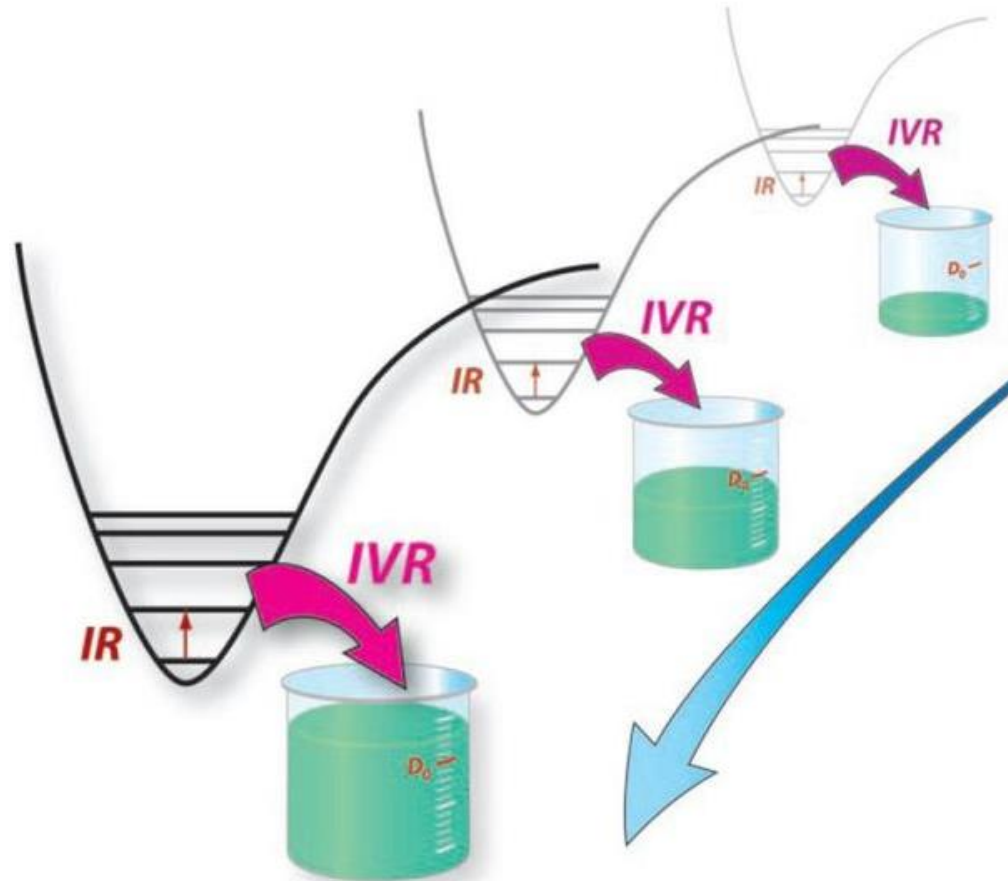
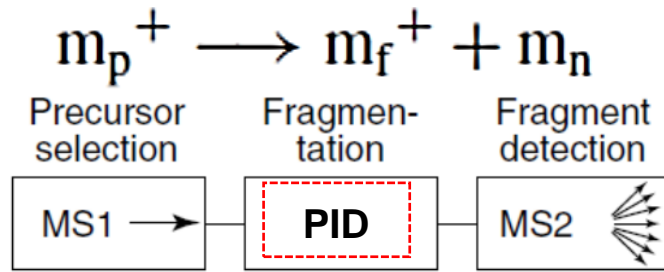
and $\bar{\nu}$ is the wavenumber per cm



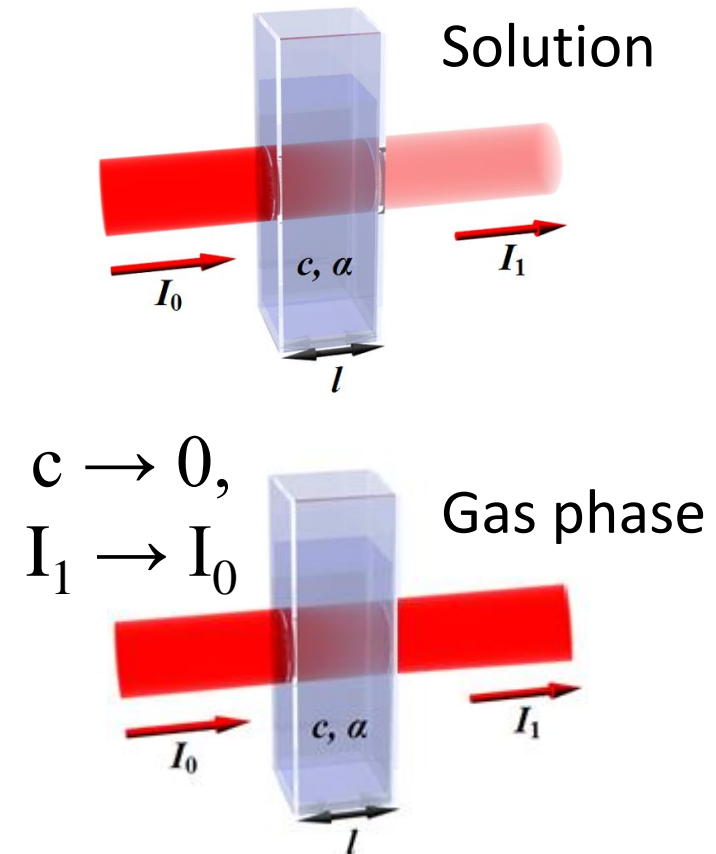


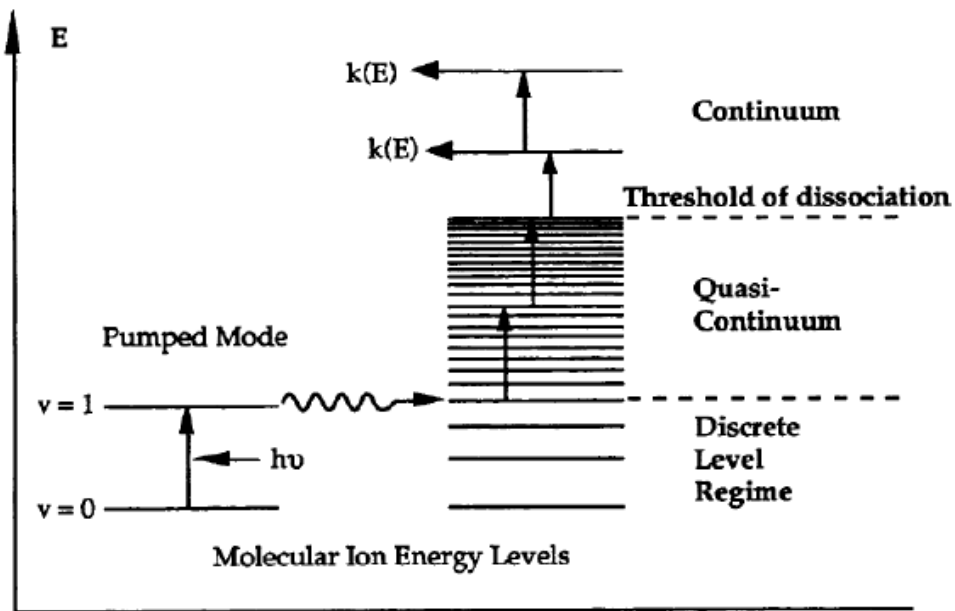
Upon formation of hydrogen bonds, red-shifts (i.e. bond weakening) are observed.

Infrared Multiple photon Dissociation (IRMPD)



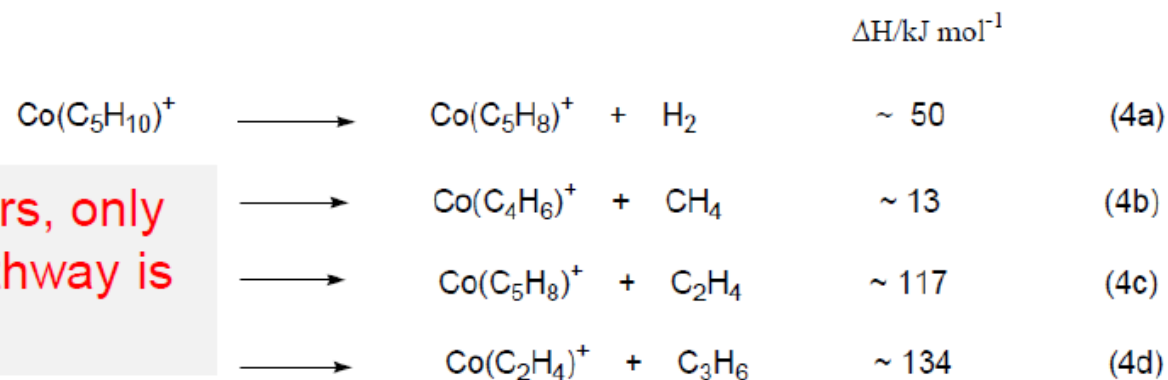
No measurable absorption in the gas phase!





Riveros, J.M. Infrared photodissociation. Encyclopedia of mass spectrometry, Vol. 1. Amsterdam: Elsevier. **2003** pp. 262–271.

Using (relatively) low-intense IR lasers, only the lowest-energy fragmentation pathway is observed



Hanratty, M. A.; Paulsen, C. M.; Beauchamp, J. L. Multiphoton infrared laser activation of organometallic species: A novel probe of the potential energy surfaces for reactions of cobalt ions with C_5H_{10} isomers. J. Am. Chem. Soc. **1985**, **107**, 5074-5080.



<https://www.youtube.com/watch?v=IFWXjzhH8a0>

Early IRMPD spectroscopy studies

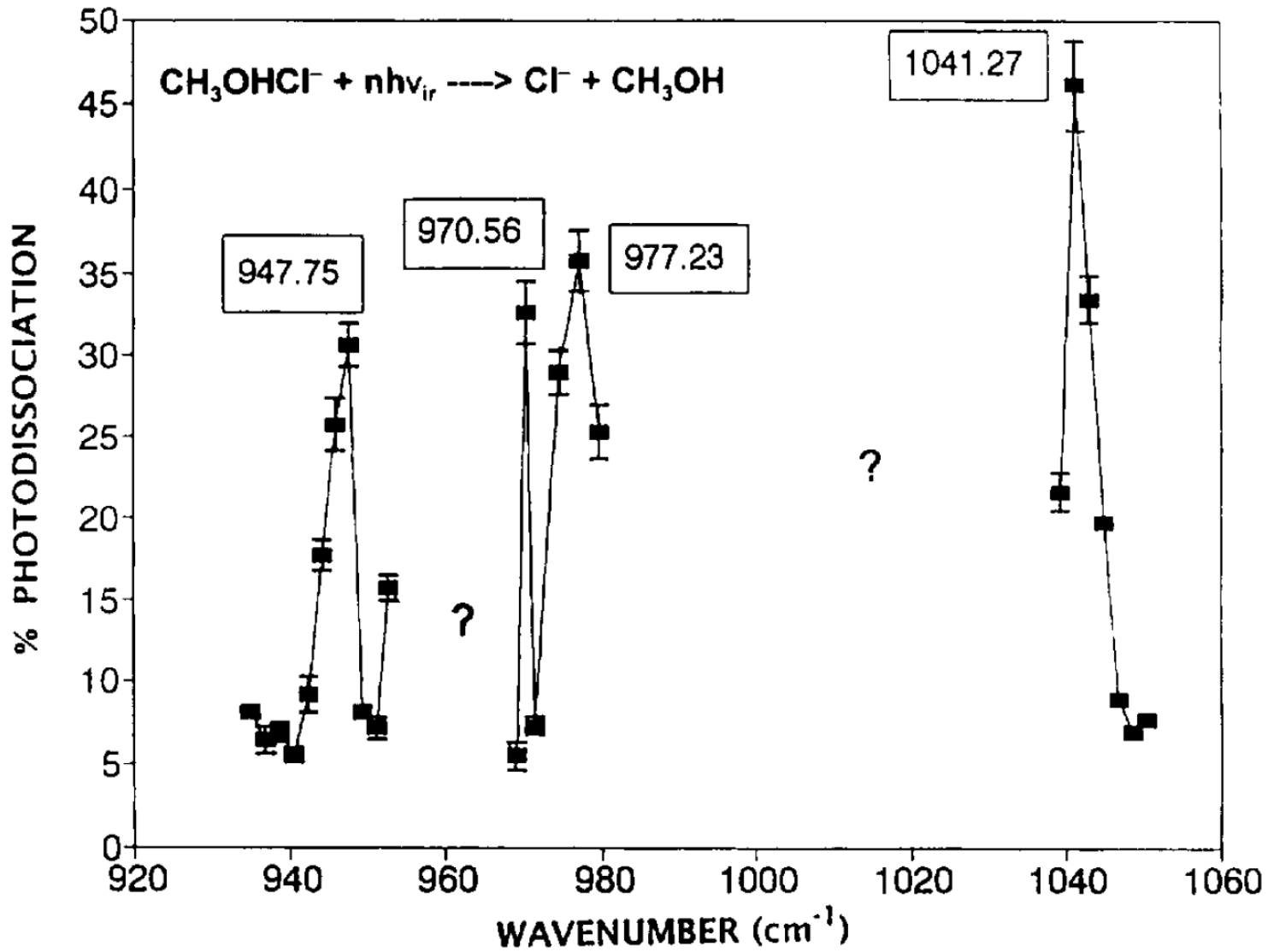
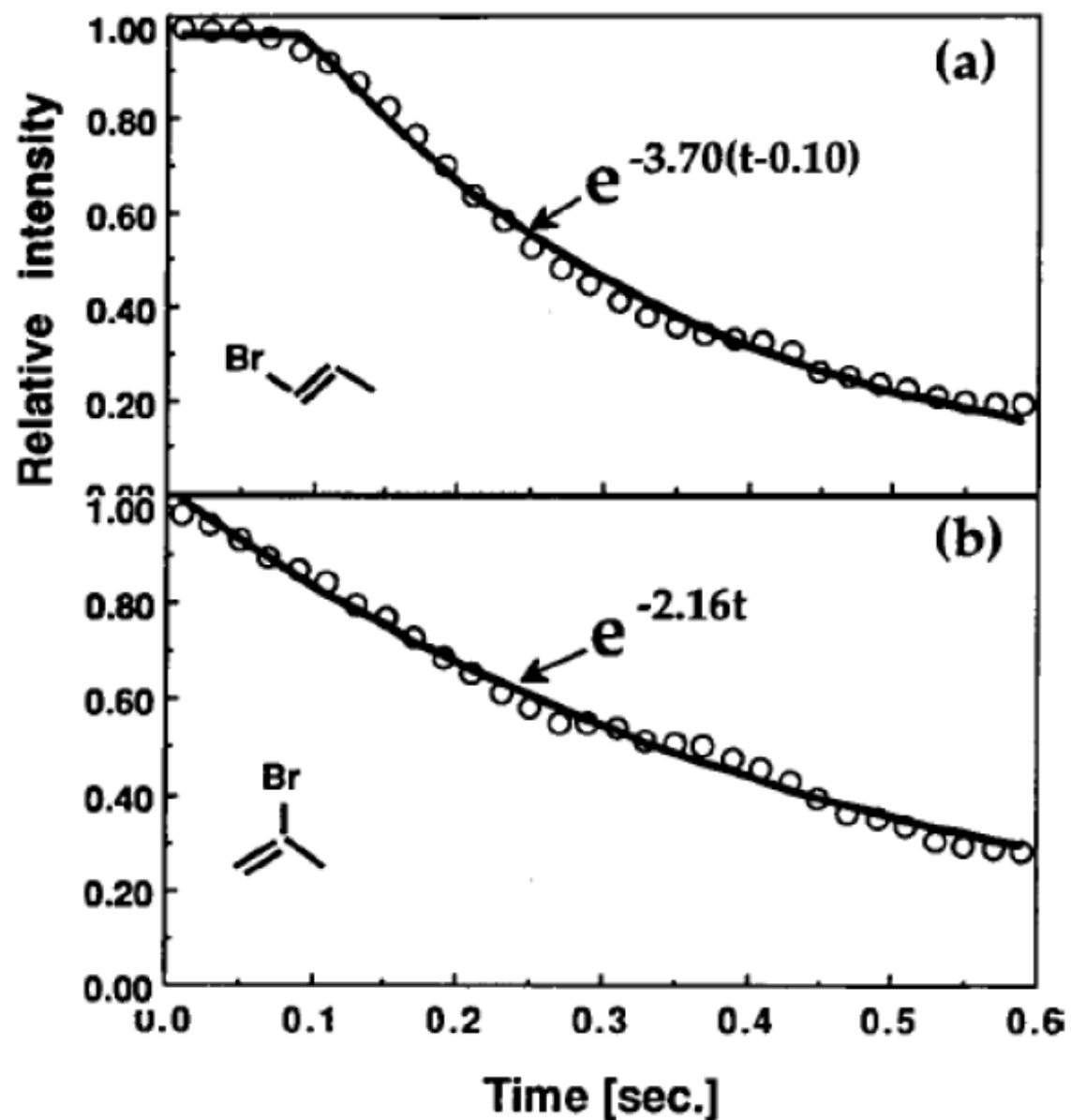


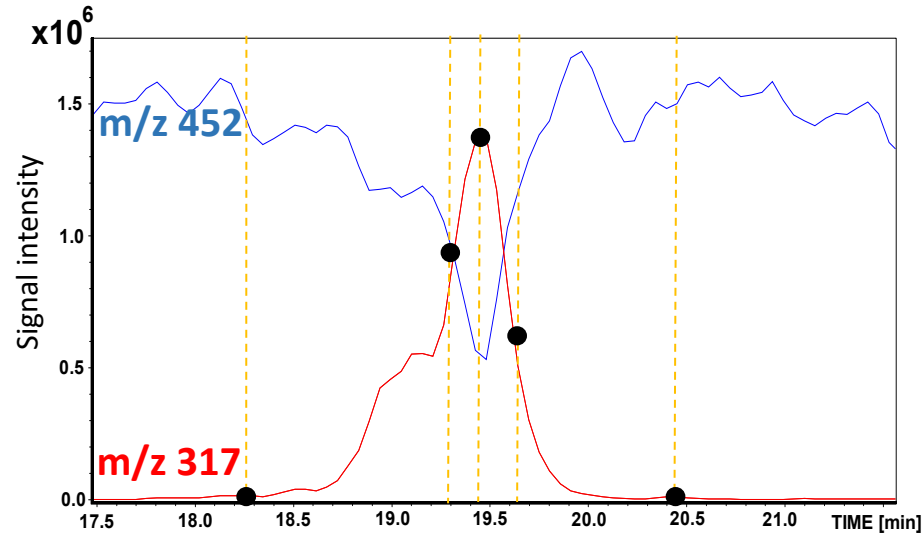
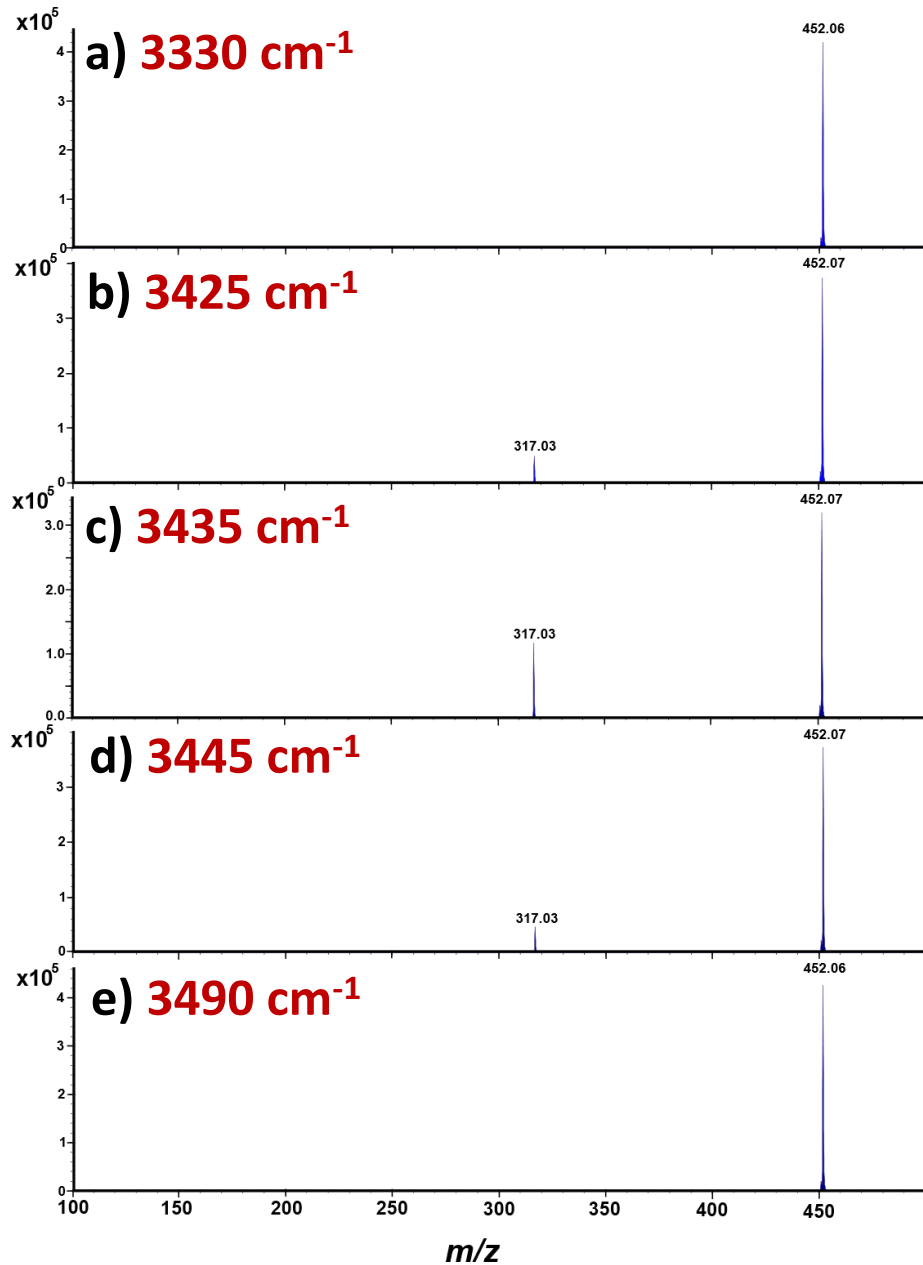
Fig. 4. IRMPD spectrum of methanol solvated chloride ion (CH_3OHCl^-).



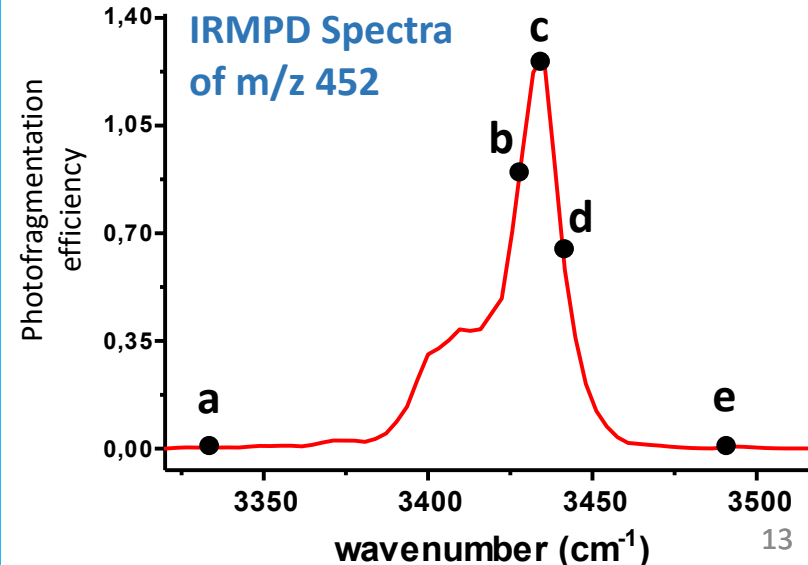
Gaumann, T.; Zhu, Z.; Kida, M. C.;
Riveros, J. M. Kinetic and Spectroscopic
Characterization of the Allyl Bromide
Molecular Ion. *J. Am. Soc. Mass Spectrom.*
1991, 2, 372-378.

Using using an 8 Watt CO₂ laser the
IRMPD kinetics of the molecular ion
obtained from 1- and 2-bromopropene are
clearly different

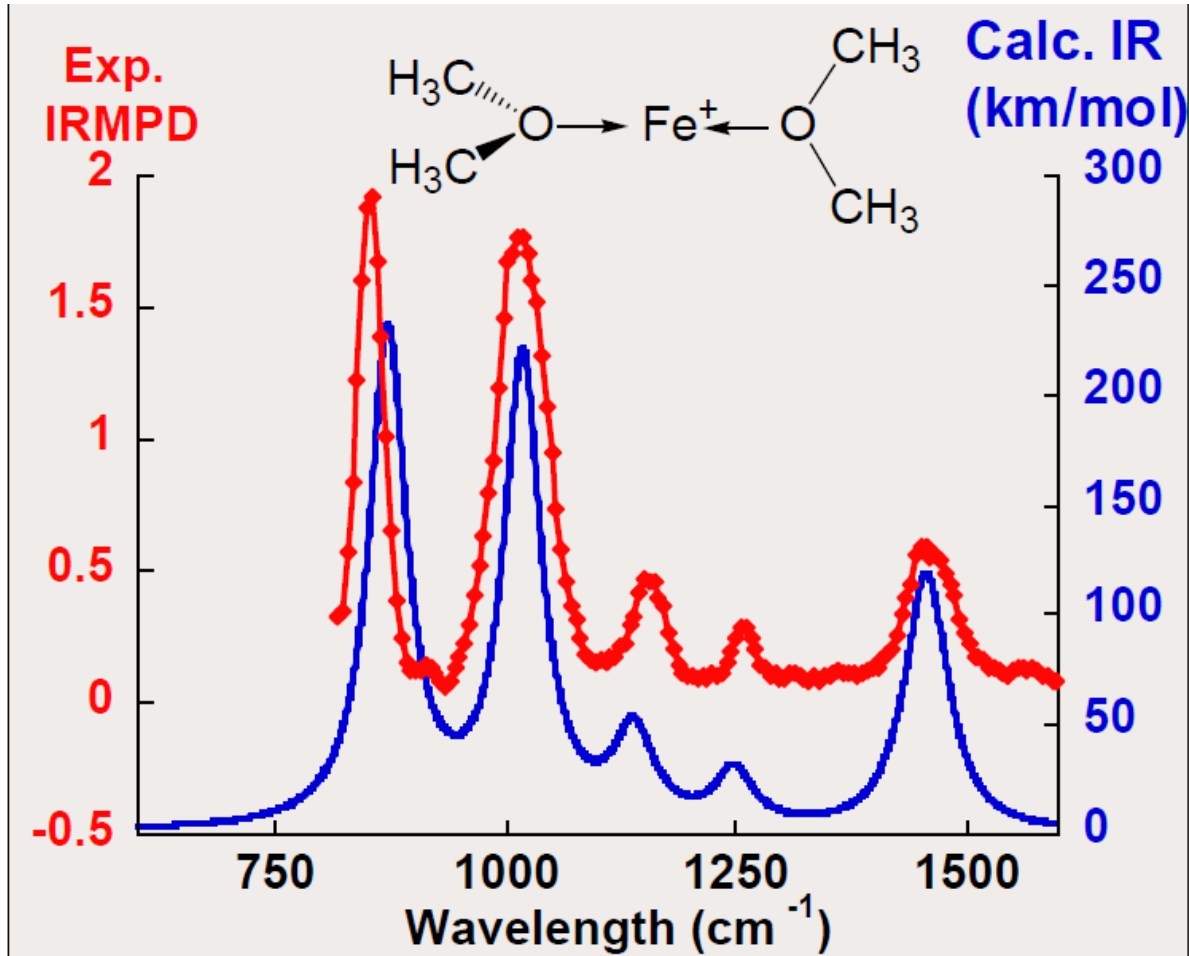
Espectroscopia de íons - IRMPD



$$\text{eff} = -\ln\left(\frac{I_P}{I_P + \sum I_{F_n}}\right) = -k_{\text{obs}}t$$



Espectroscopia de íons - IRMPD

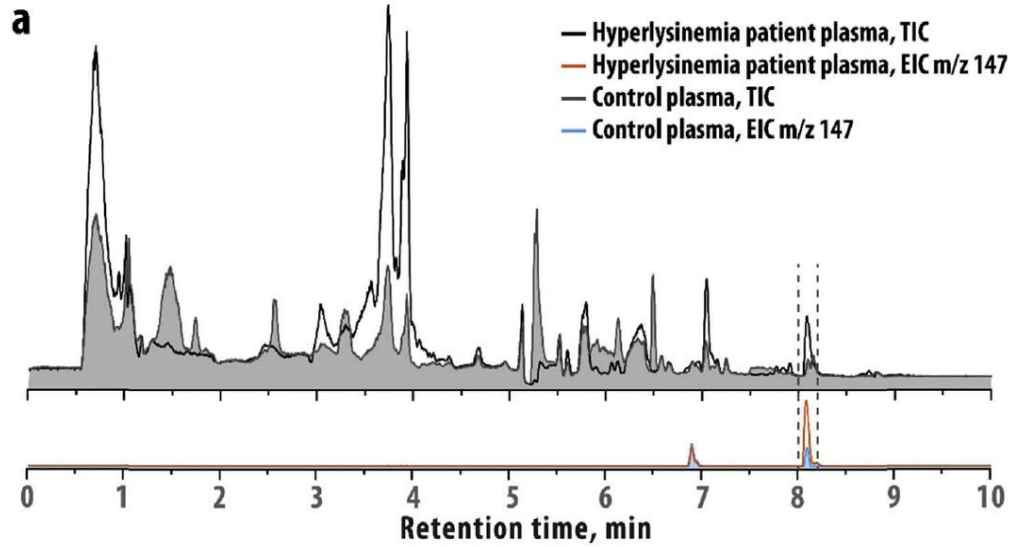


IRMPD permite comparação direta com cálculos de estrutura eletrônica

Free Electron Laser Workshop, Chicago, 09/2002
-> Nuc. Instr. And Method A, **2003**, 507, 541
Chem. Phys. Lett., **2004**, 385, 273

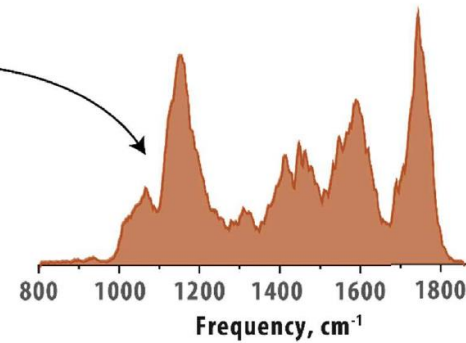
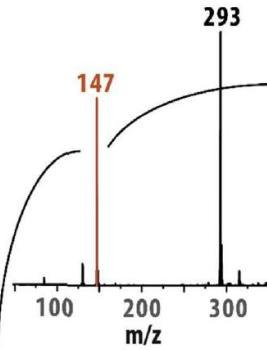
Reference-standard metabolite identification

Lysine metabolism disorder



— MS - Hyperlysine patient plasma, fraction 8.02-8.13 minutes

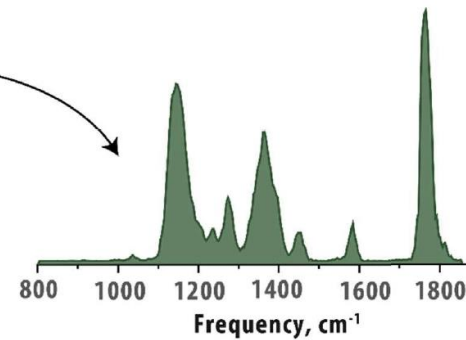
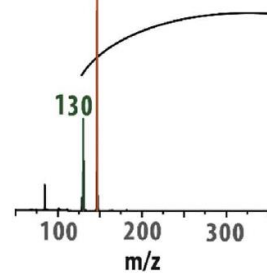
c — IR - Hyperlysine patient plasma, fraction 8.02-8.13 minutes - m/z 147



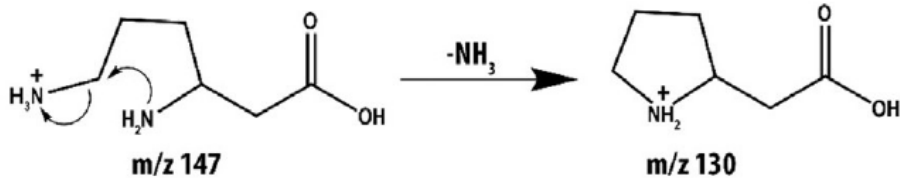
e — CID MS/MS - Hyperlysine patient plasma, fraction 8.02-8.13 minutes - m/z 147

— IR - Hyperlysine patient plasma, fraction 8.02-8.13 minutes - m/z 130 fragment of m/z 147

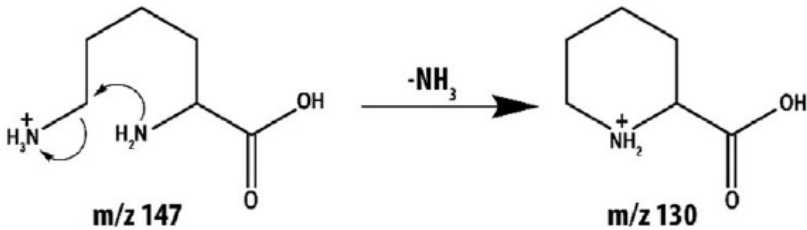
d



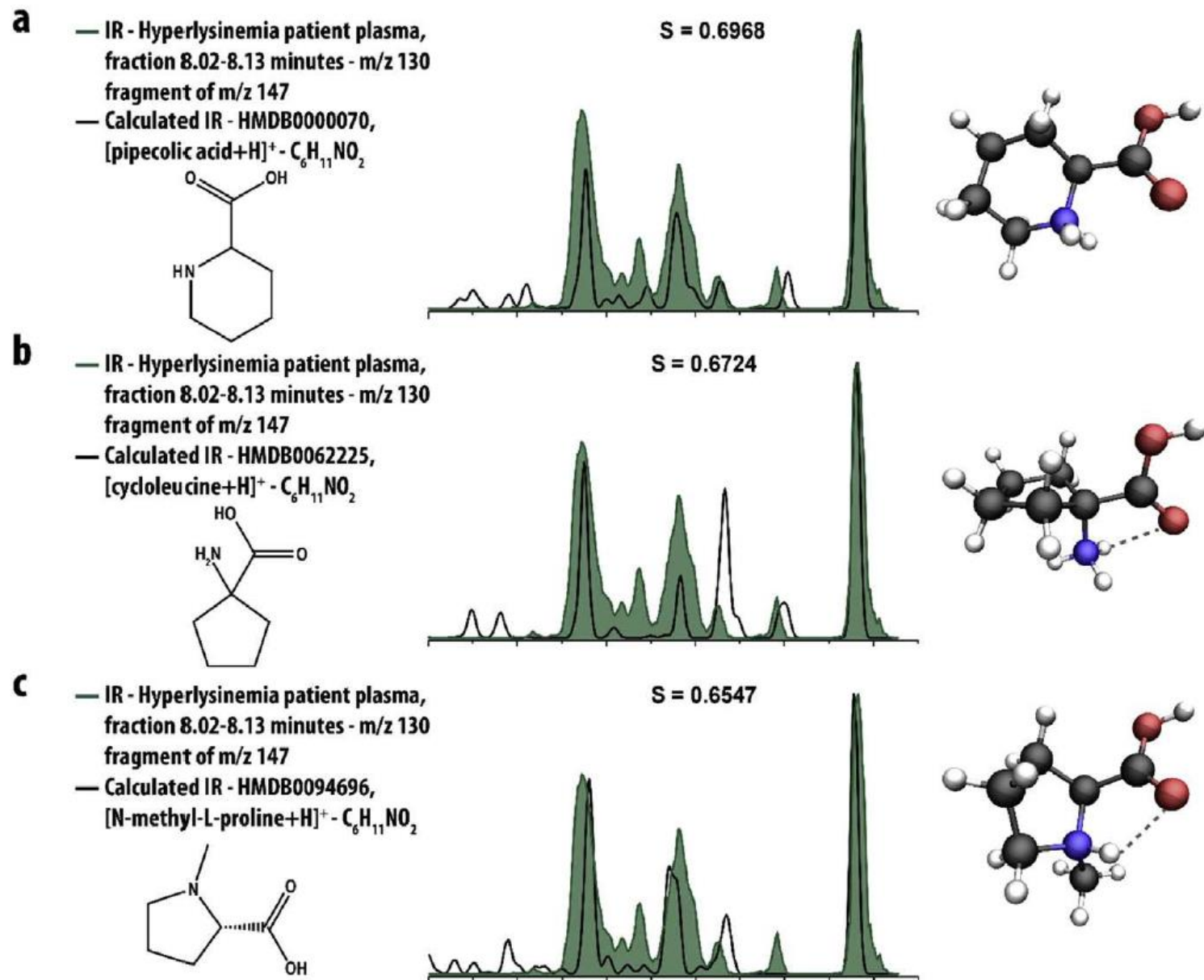
a



b



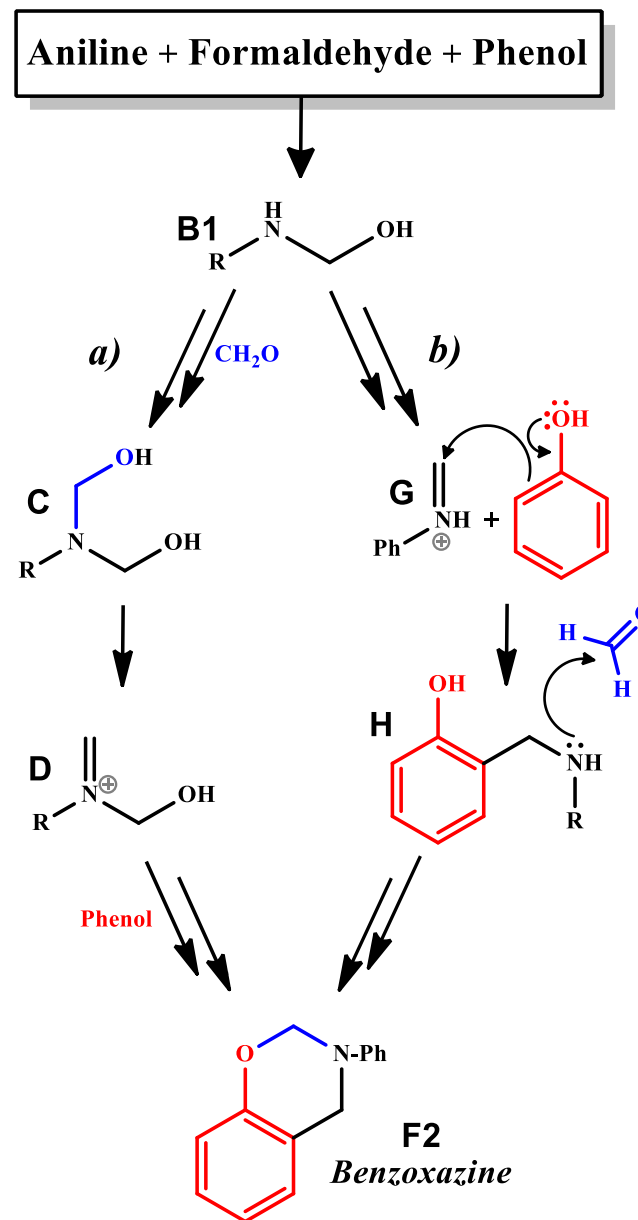
Reference-standard metabolite identification



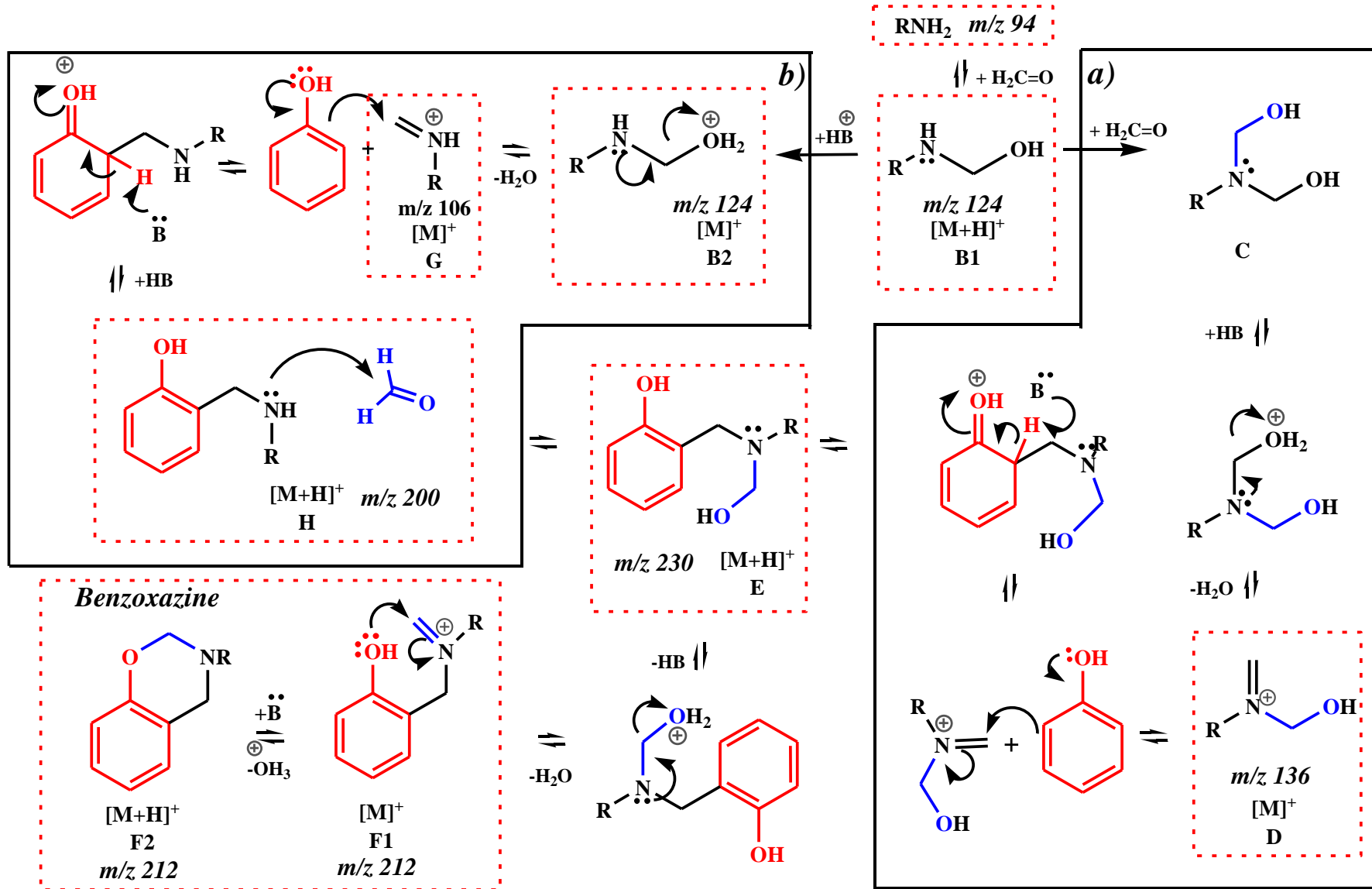
Estudo de mecanismos de reação



Podemos avaliar mecanismos de reação por ESI-IRMPD?

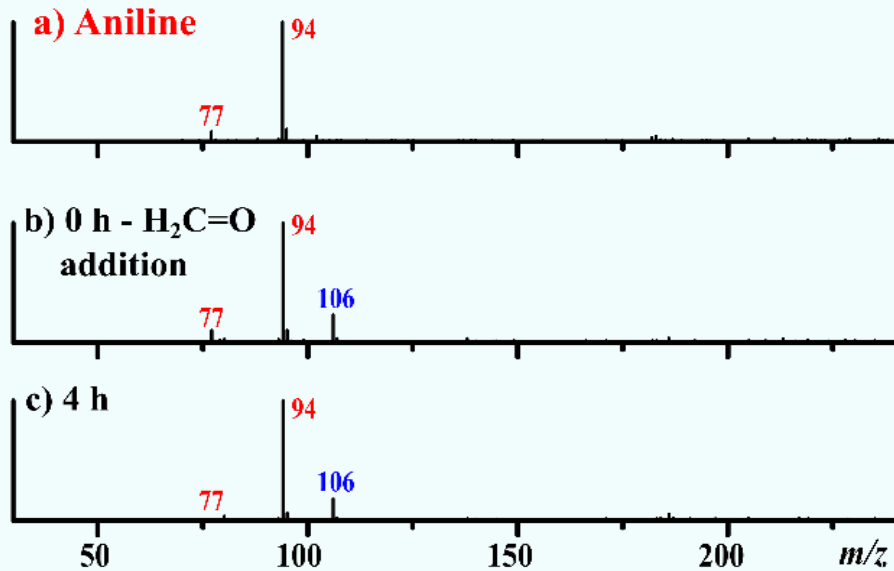


Estudo de mecanismos de reação

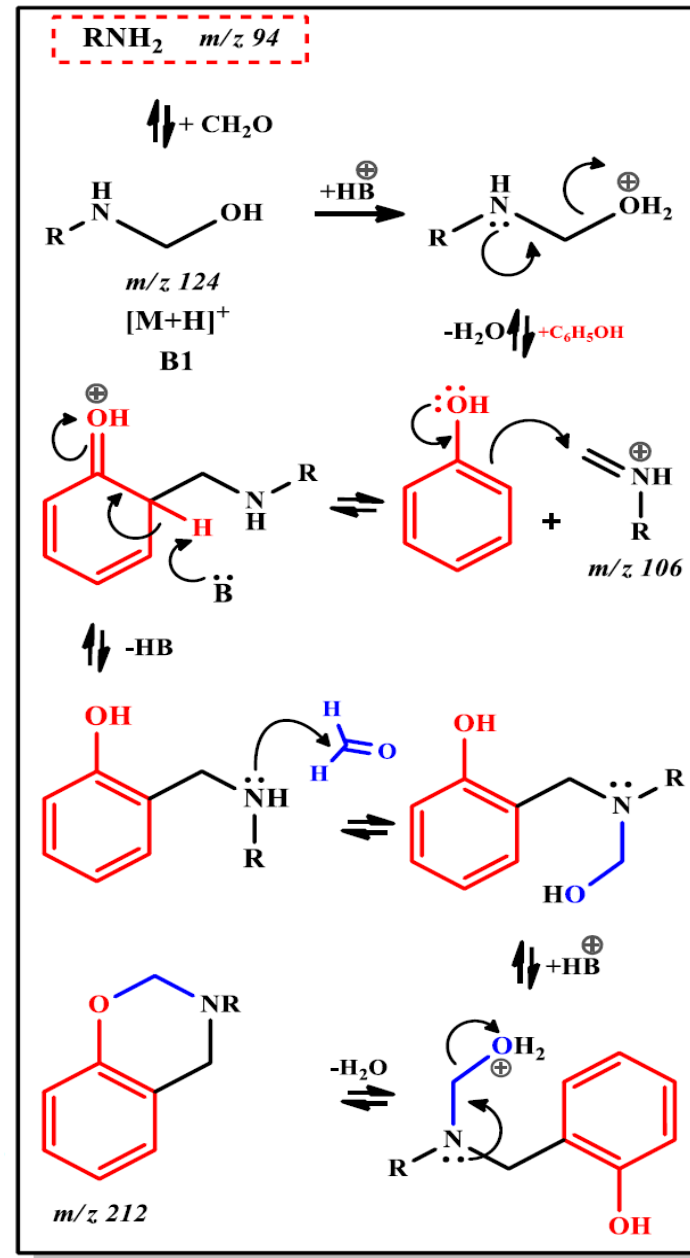
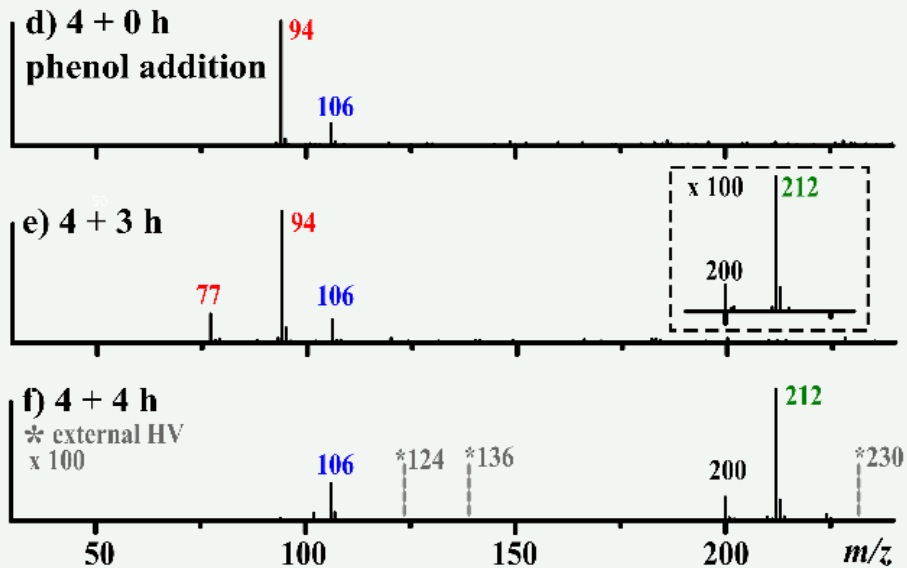


Estudo de mecanismos de reação

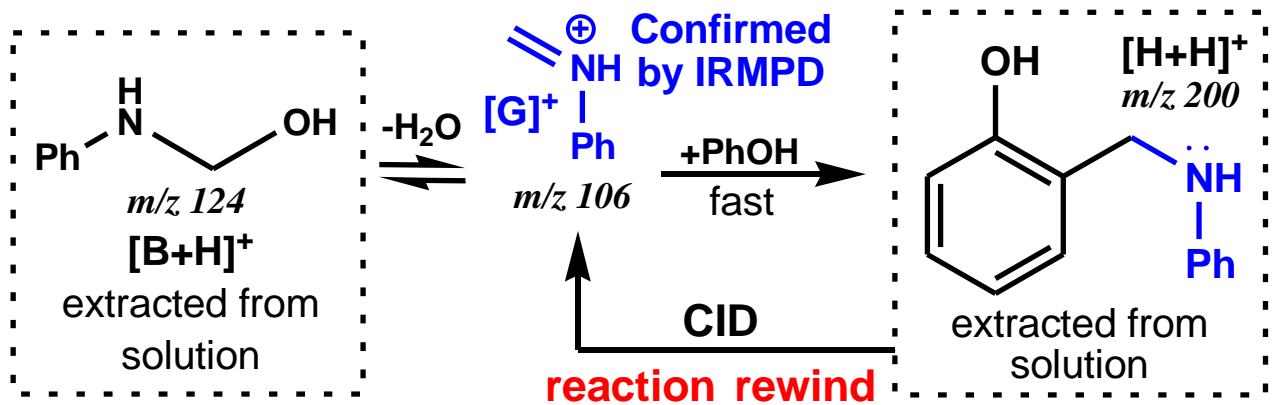
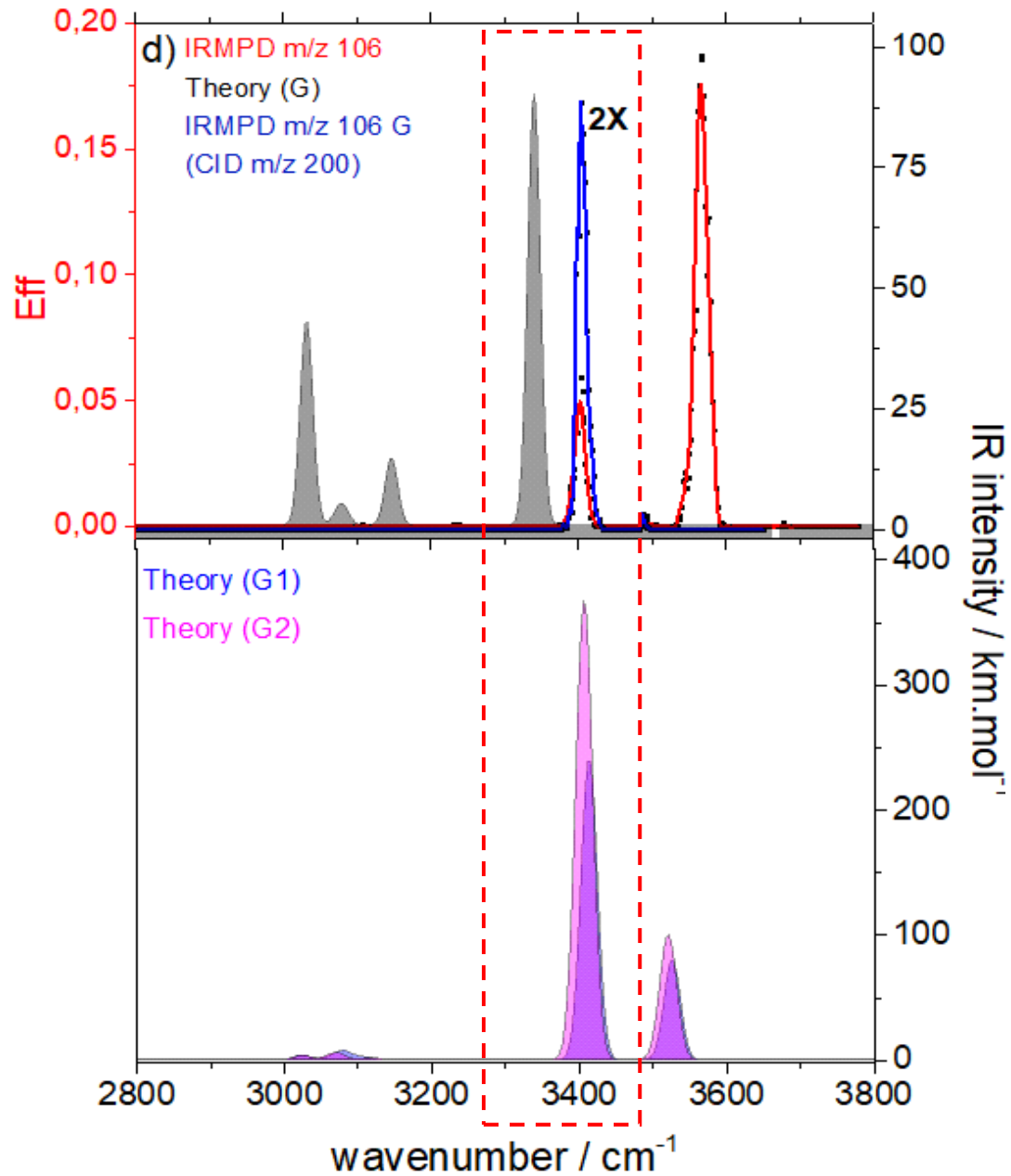
"Imine" formation



"Benzoxazine" formation

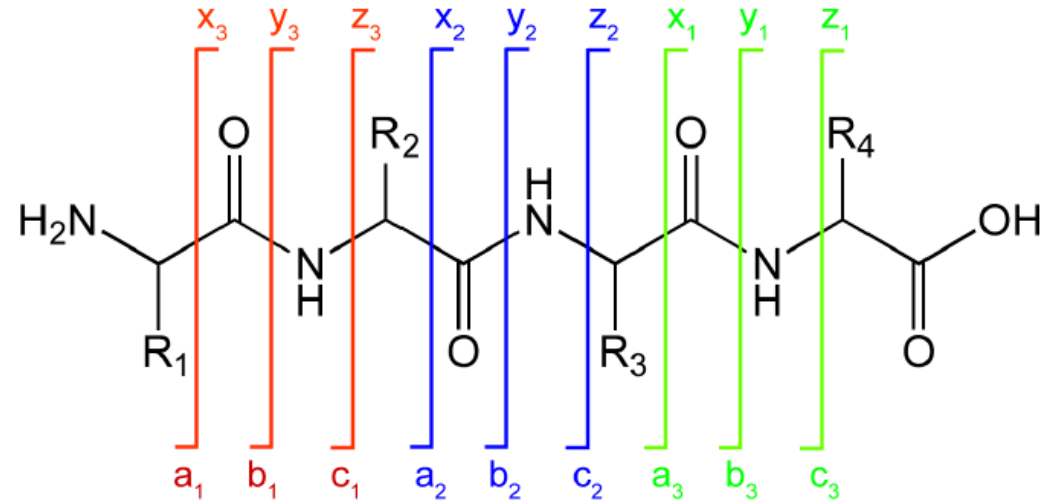
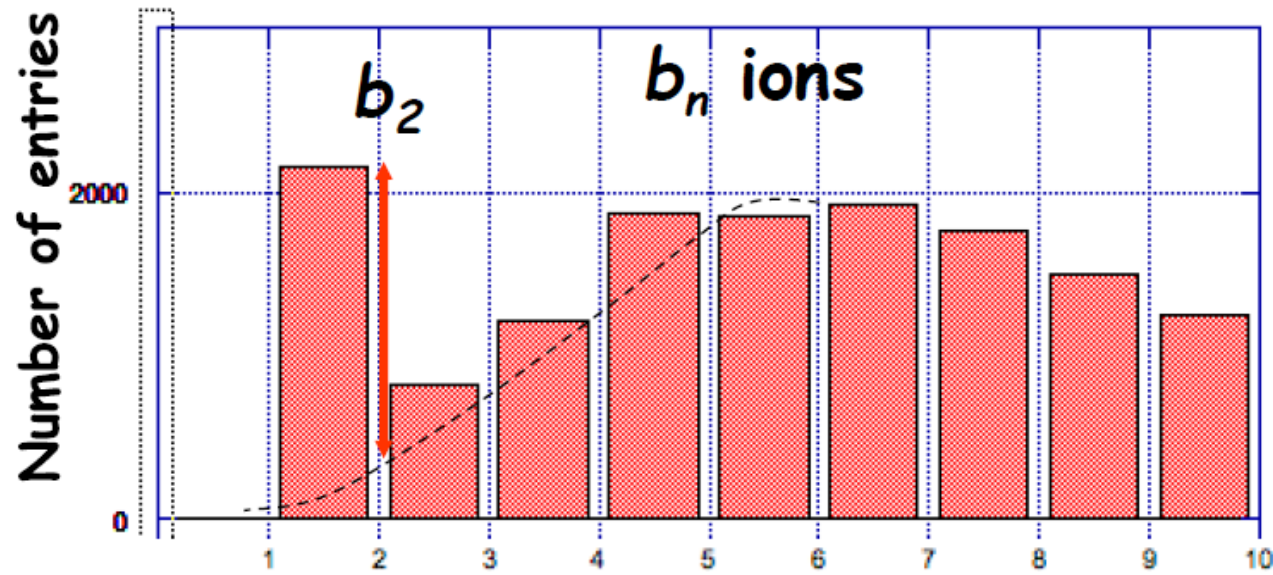


Síntese de intermediários em fase gasosa



Fragmentação preferencial de peptídeos

Statistical Analysis of the SwedCAD Spectral Database

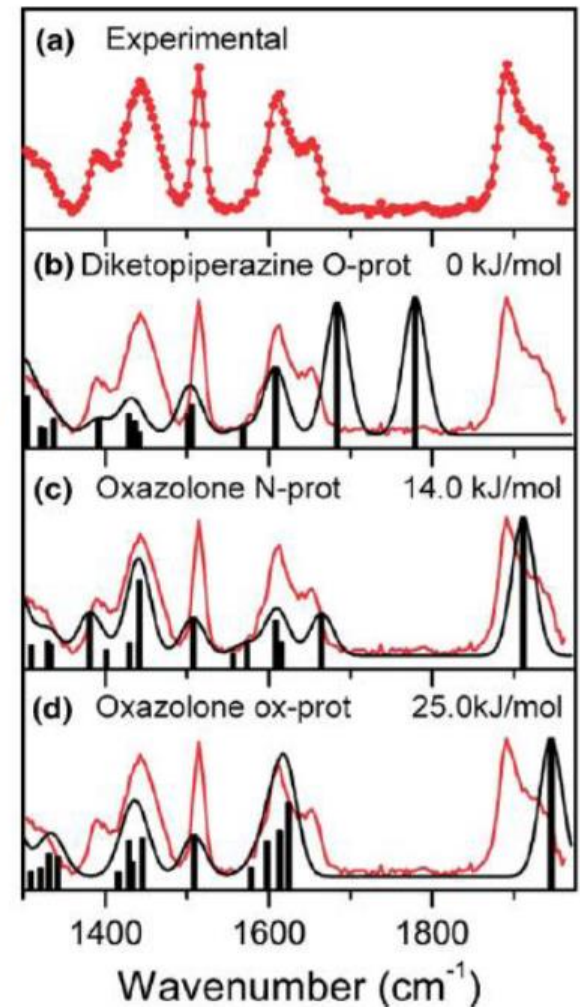
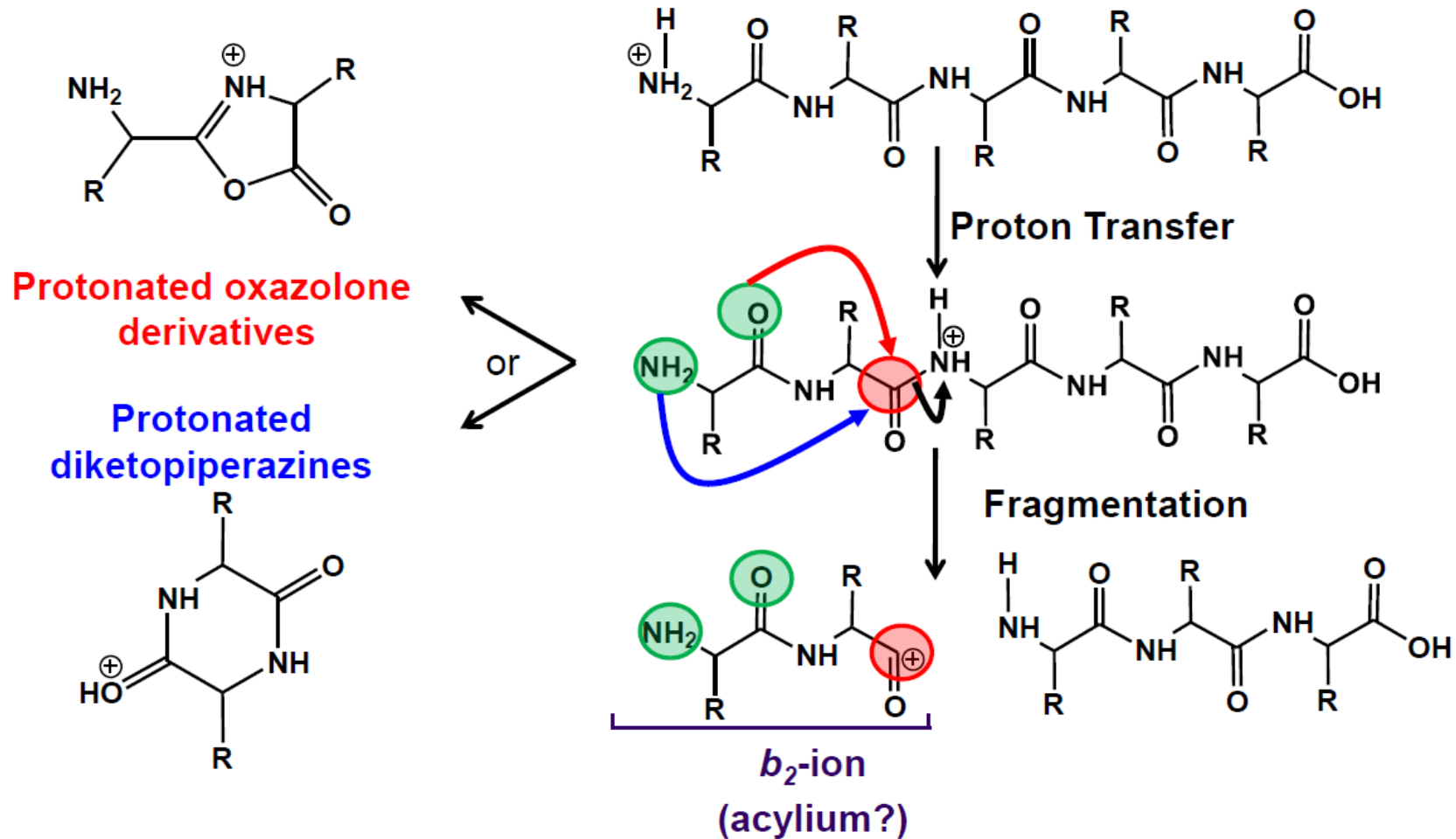


b_2 ions are more abundant than other b_n fragments

Are b_2 ions of tryptic peptides really oxazolones?
Or diketopiperazines?

www.bmms.uu.se/CAD

Íons b_2



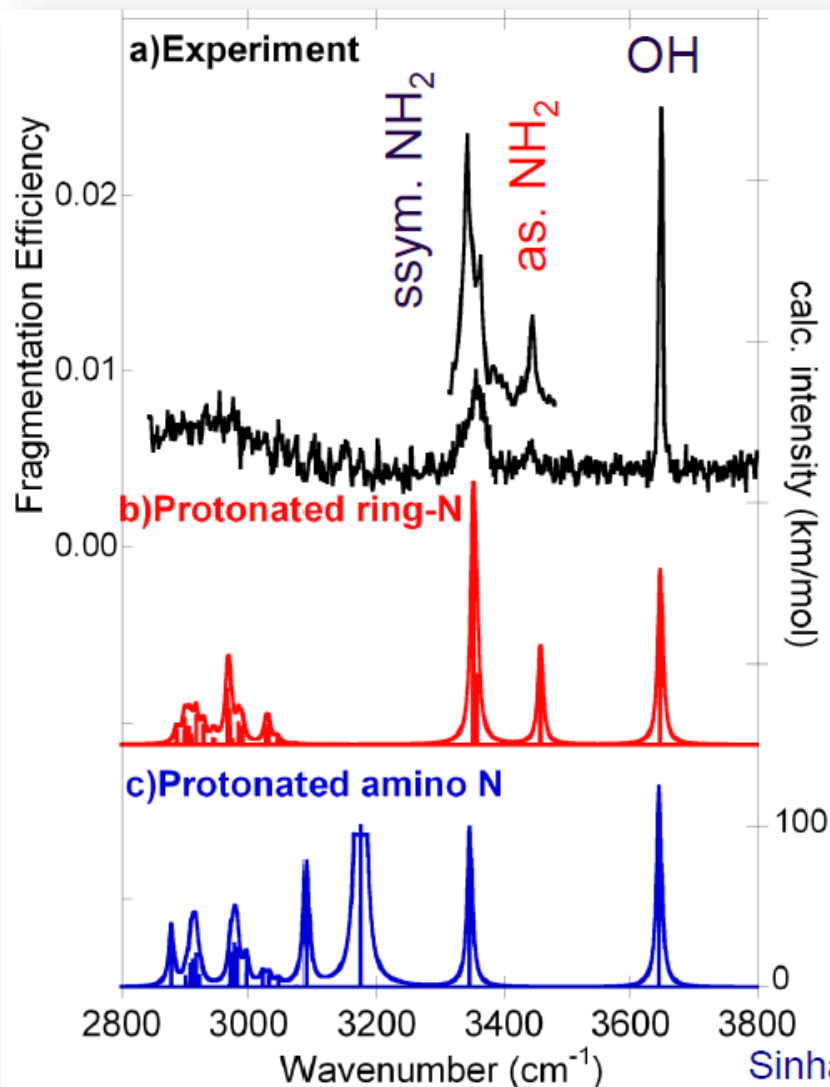
Yalcin, Csizmadia, Peterson, Harrison, *J. Am. Soc. Mass Spectrom.* 1996, 7, 233

Cordero, Houser, Wesdemiotis, *Anal. Chem.* 1993, 65, 1594.

Polfer and coworkers: *J Am Soc Mass Spectrom* 2010, 21, 1313.

Diferenciando múltiplas populações com “apenas” um fóton

Fragment b_2 of doubly protonated YIGSR

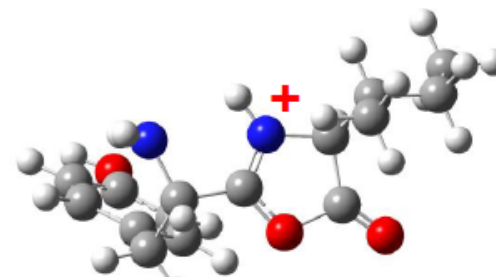


Diagnostic band at 3445 cm^{-1}

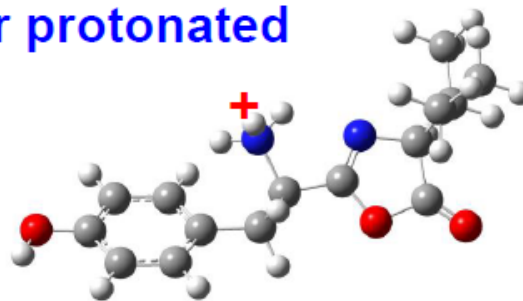
Characteristic of the asymmetric NH_2 N-H stretching mode

Could only be observed using an auxiliary CO_2 laser combined with the OPO/OPA

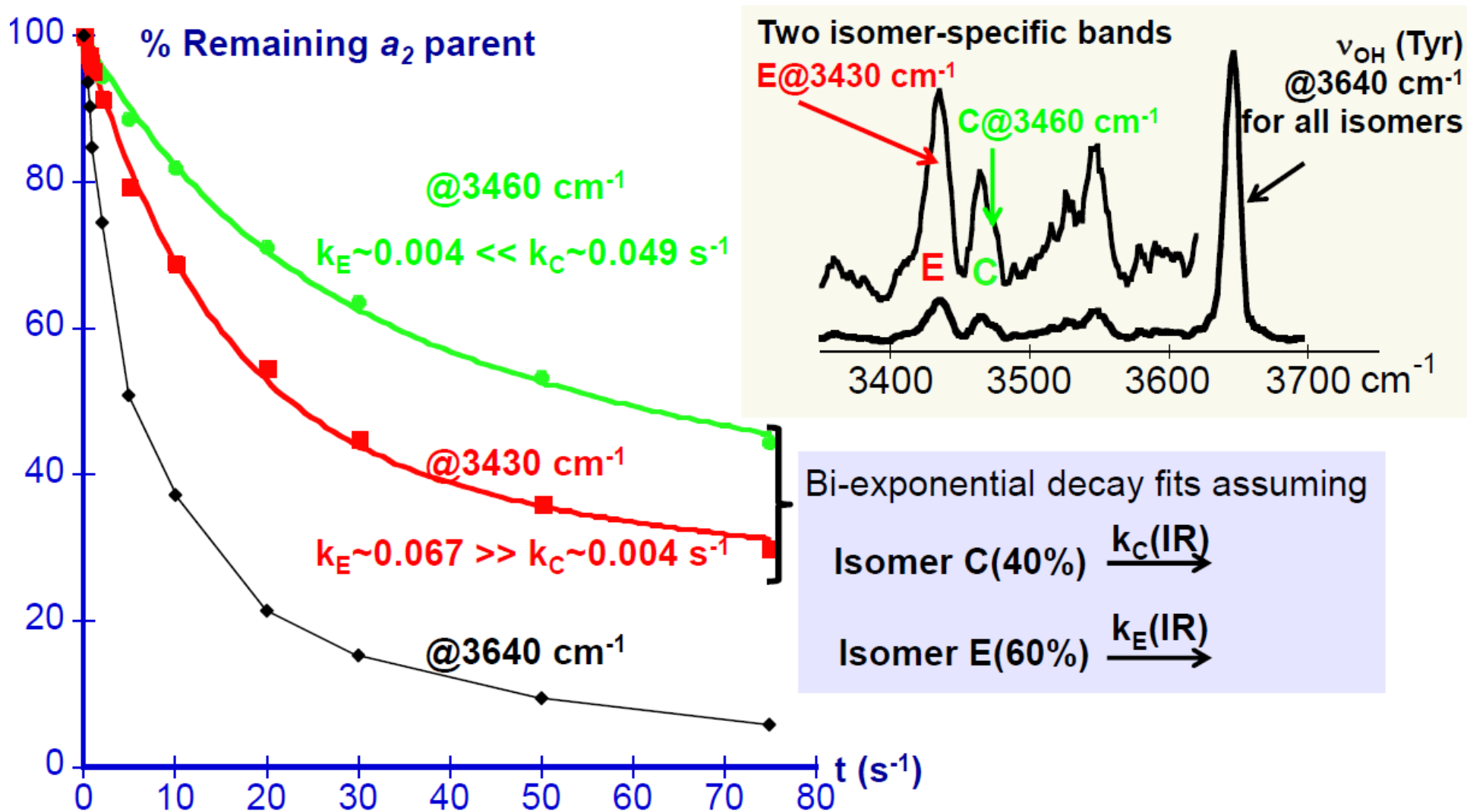
N-ring protonated



N-ter protonated

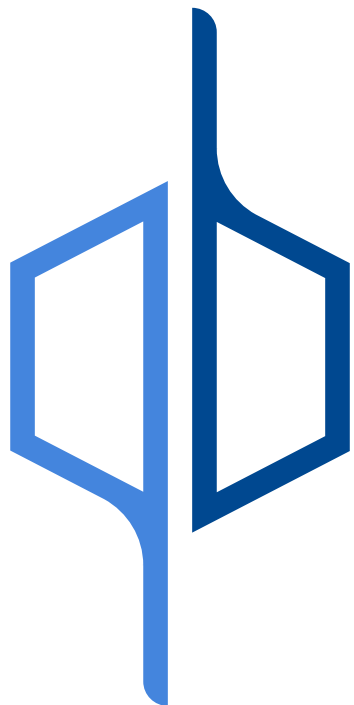


Taxas de dissociação diferentes para íons diferentes



Hydration Isomers of Protonated Phenylalanine and Derivatives: Relative Stabilities from Infrared Photodissociation.

Prell, J.S., Chang, T.M., O'Brien, J.T., and Williams, E.R., *J. Am. Chem. Soc.* 2010, 132, 7811-7819.



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