

# Structural elucidation of new hallucinogenic phenethylamines and tryptamines identified on the drug market



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# Hallucinogens

## Psychedelic substances

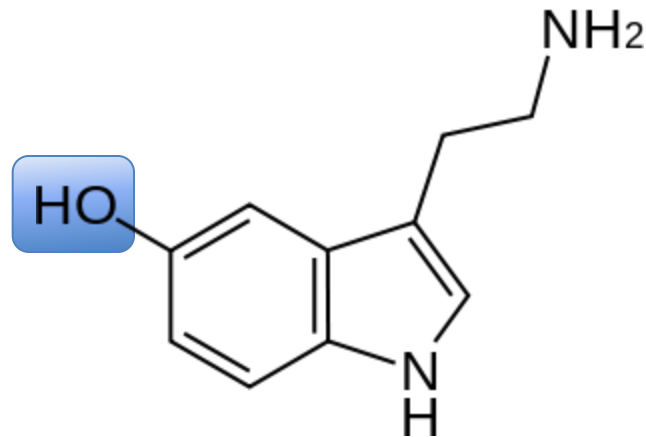
- **Psychedelic** (*def.*) – relating to or denoting new or altered perceptions or sensory experiences
- A **psychedelic substance** is a psychoactive drug whose primary action is to alter cognition and perception
- **Psychedelic** – the Greek *psukhe* (mind) + *deloun* (reveal, make visible)

# Serotonin receptors

- Many hallucinogens are known to show affinities for various serotonin (5-HT) receptors in different ways and levels, and may be classified by their activity at different 5-HT sub-sites, such as 5-HT<sub>1A</sub>, 5-HT<sub>1B</sub>, 5-HT<sub>2A</sub>, etc.
- It is almost unanimously agreed that serotonergic psychedelics produce their effect by acting as strong partial agonists at the **5-HT<sub>2A</sub>** receptors

# Serotonin

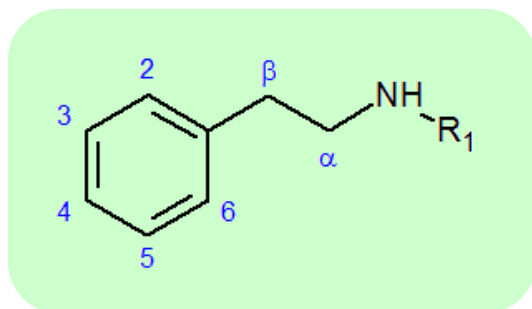
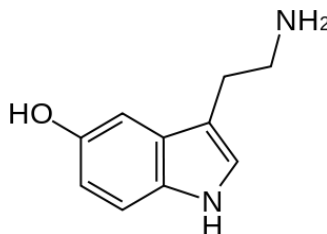
Serotonin (often referred to as **5-HT**, short for its full chemical name **5-hydroxytryptamine**) is a naturally occurring neurotransmitter which is tied to positive mood, certain involuntary muscle control, and countless other functions



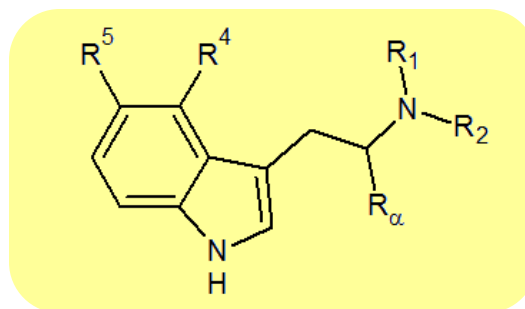
# Serotonergic psychedelics

Many serotonergic psychedelics have very strong structural similarities to serotonin itself, which partially explains the affinity for certain 5-HT sites

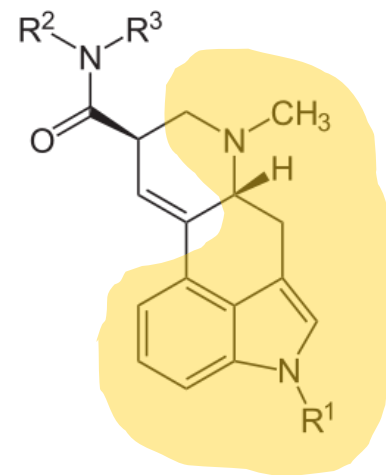
**Serotonin**



**Phenethylamines**

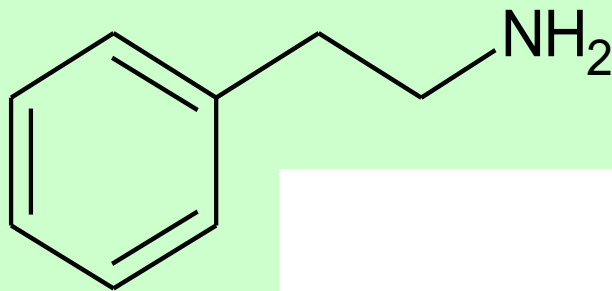
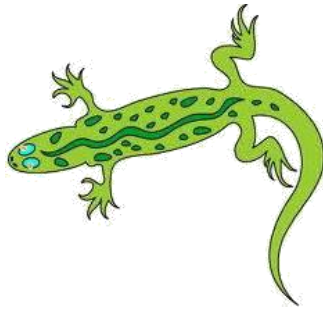


**Tryptamines**

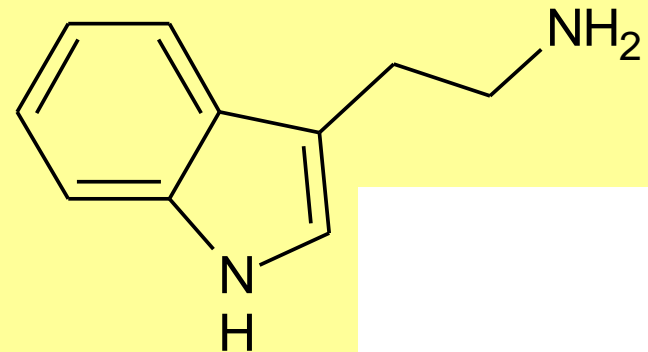
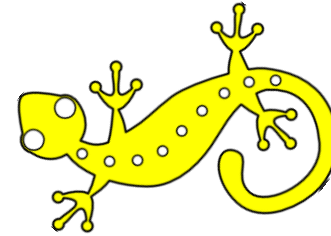


**Ergolines**

# Chemical structures

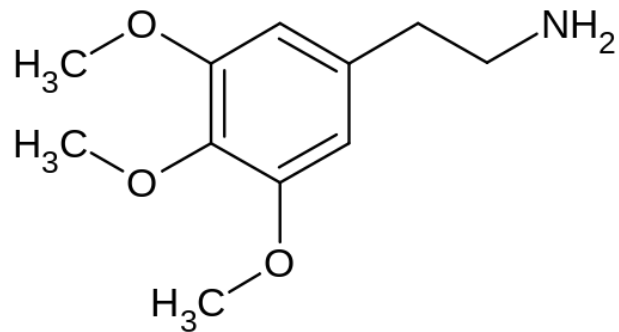


**Phenethylamine**  
**PEA**



**Tryptamine**  
**TRYP**

# Natural PEA



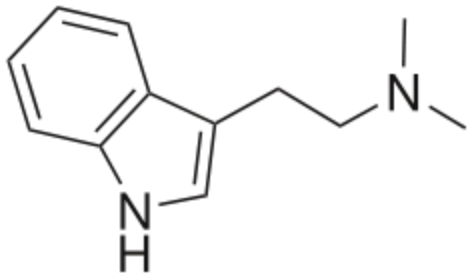
## Mescaline

2-(3,4,5-trimethoxyphenyl)ethanamine



- Peyote cactus (*Lophophora williamsii*)
- San Pedro cactus (*Echinopsis pachanoi*)
- Peruvian torch (*Echinopsis peruviana*)

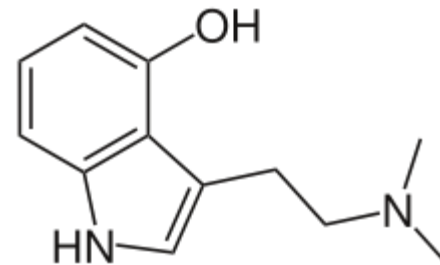
## Natural TRYP



N,N-dimethyltryptamine  
(DMT)



*Ayahuasca*

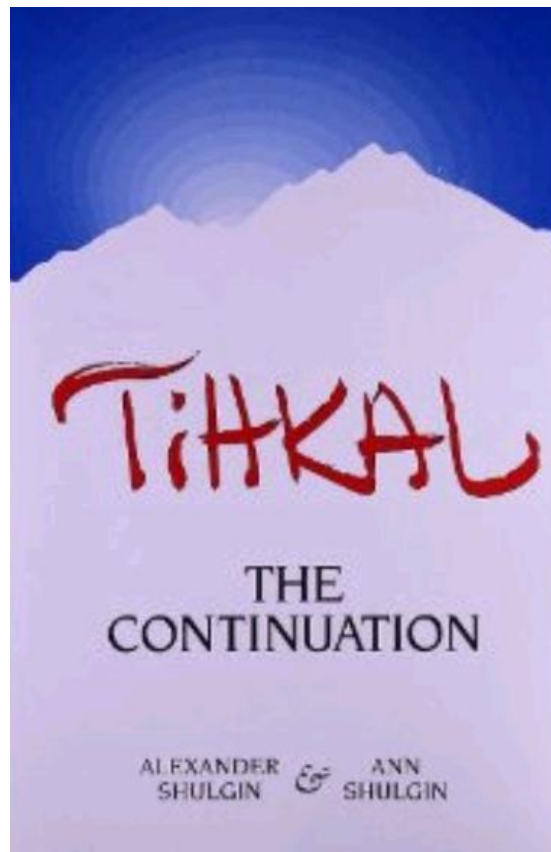
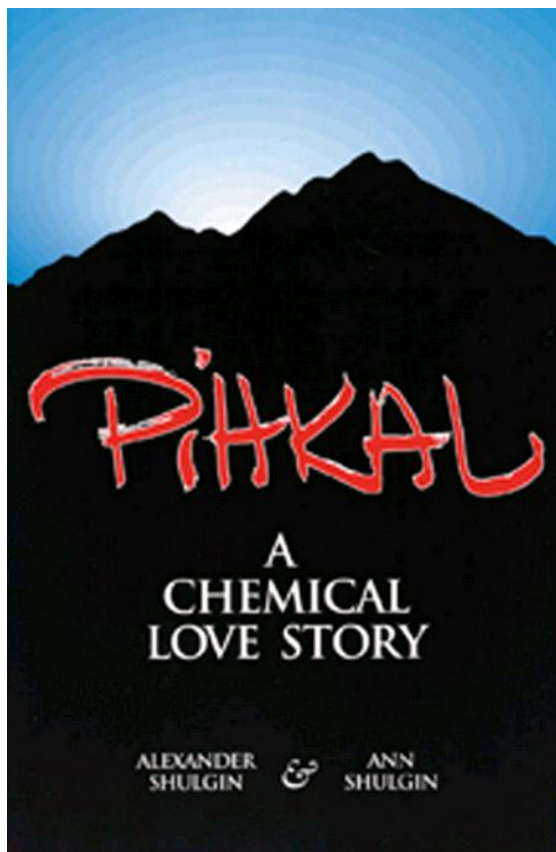


Psilocin  
(4OH-DMT)





# PIHKAL and TIHKAL



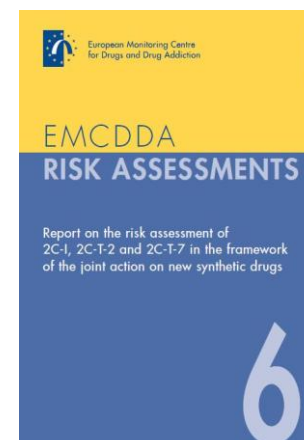
- Synthesis
- Dosage
- Duration
- Qualitative comments
- Extensions and commentaries



European Monitoring Centre  
for Drugs and Drug Addiction

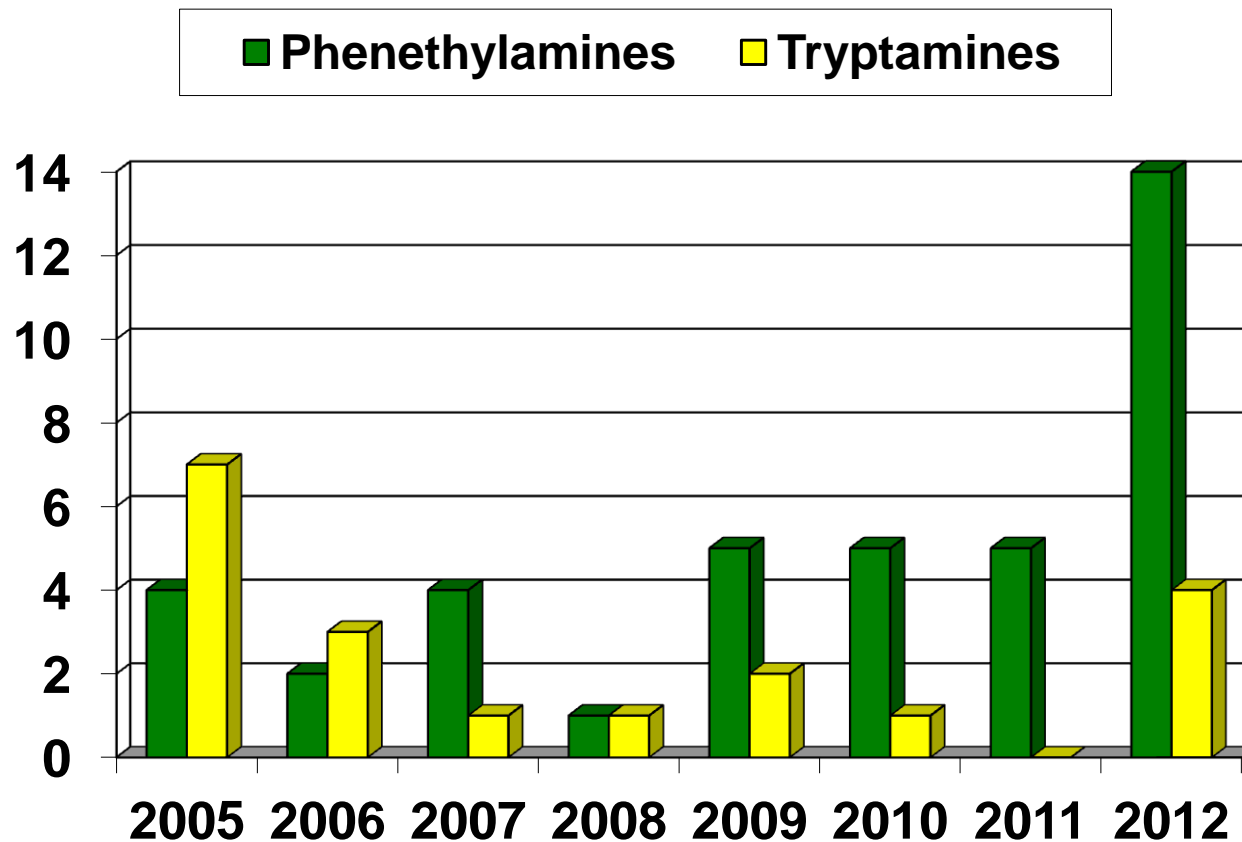
# NEW DRUGS 2014

## Risk assessment

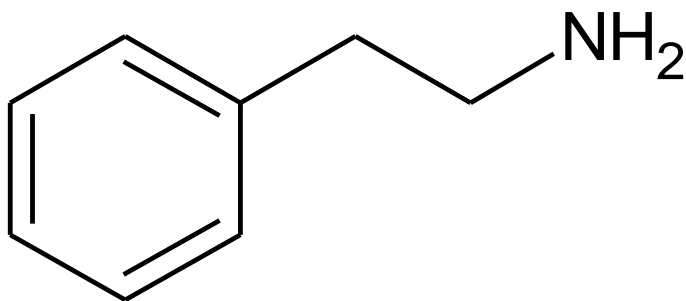


Year	Substance(s)
1999	<b>MBDB; 4-MTA</b>
2002	GHB
2003	<b>PMMA</b>
2004	<b>TMA-2; 2C-I; 2C-T-2; 2C-T-7</b>

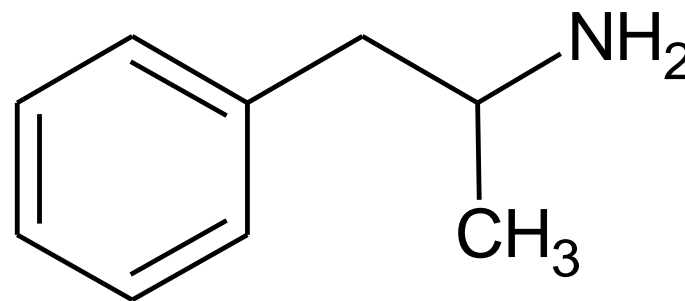
# Number of notifications through the EWS



# PEA and Amphetamine



Phenethylamine  
(2-phenylethanamine)

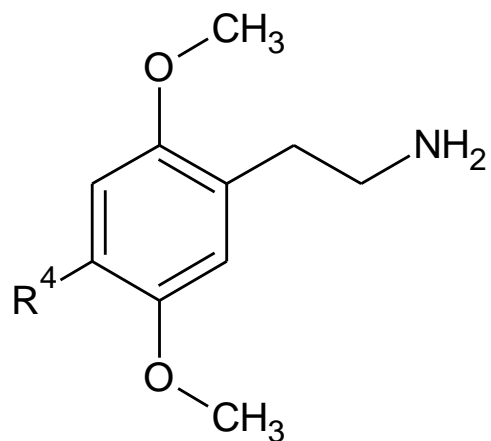


Phenylisopropylamine  
(1-phenylpropan-2-amine)  
AMPHETAMINE

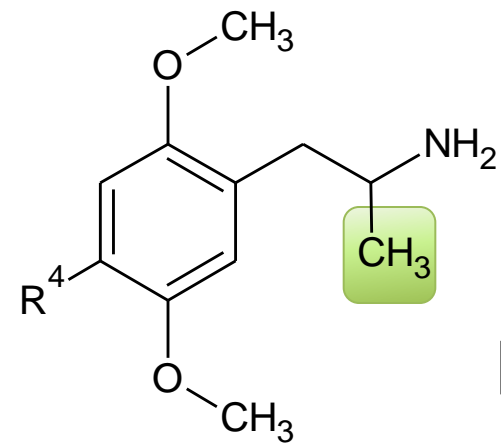
# Reported Phenethylamines

- 71 substances
  - 2C group: 12 representatives (+ two 3C) + ,multimethoxy' derivatives (trimethoxy, Fly's)
  - NBOMe's: 11 representatives + 25NBMD
  - DOx group: ~ 10
  - Many amphetamine-type stimulants (ATS)

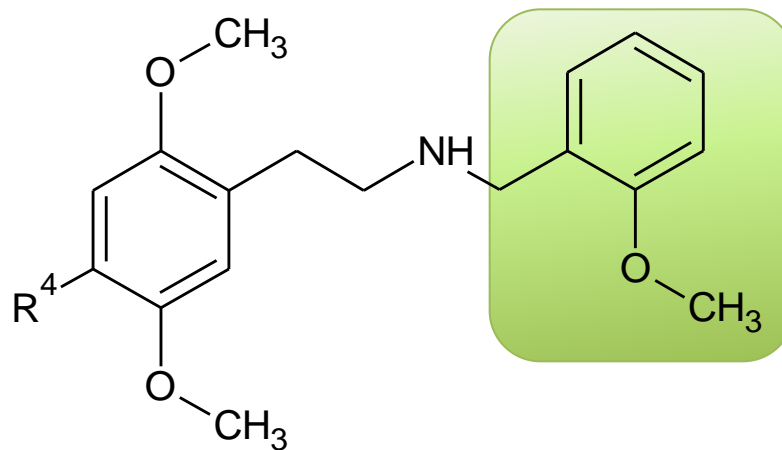
## PEA



2C



DOx



NBOMe

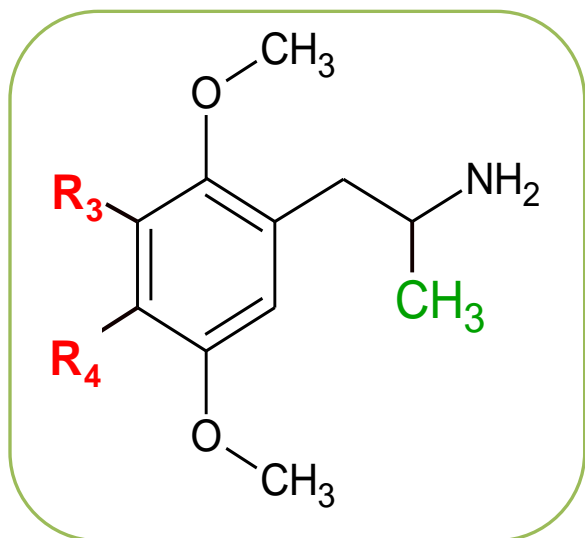
# Reported 2C's

Substance	Substituent @ C4
2C-B	Br
2C-C	Cl
2C-D	CH <sub>3</sub>
2C-E	C <sub>2</sub> H <sub>5</sub>
2C-G	CH <sub>3</sub> (+CH <sub>3</sub> @C3)
2C-H	H
2C-I	I
2C-N	NO <sub>2</sub>
2C-P	C <sub>3</sub> H <sub>7</sub>
2C-T-4	SC <sub>3</sub> H <sub>7</sub> (iso)
2C-T-7	SC <sub>3</sub> H <sub>7</sub>

## Others:

- Escaline
- Methallylescaline
- Allylescaline
- Proscaline
- 3C-E
- 3C-P

# DOx (examples)



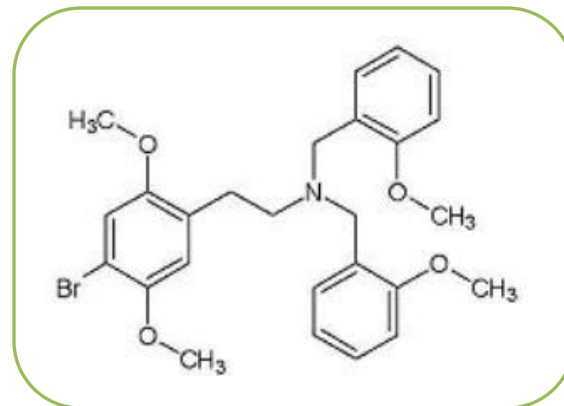
R <sub>4</sub>	R <sub>3</sub>	Name
-H	-H	DMA
-CH <sub>3</sub>	-H	DOM
-C <sub>2</sub> H <sub>5</sub>	-H	DOET
-OCH <sub>3</sub>	-H	TMA-2
-OCH <sub>3</sub>	-OCH <sub>3</sub>	TMA*
-Br	-H	DOB
-I	-H	DOI
-Cl	-H	DOC

\* - does not contain -OCH<sub>3</sub> group in position 2

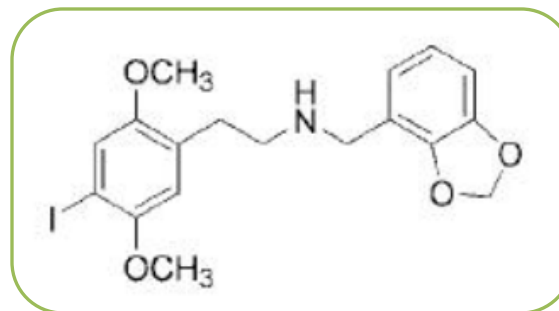


# List of reported NBOMe's

- 25B-N(BOMe)<sub>2</sub>
- 25B-NBOMe
- 25C-NBOMe
- 25D-NBOMe
- 25E-NBOMe
- 25G-NBOMe
- 25H-NBOMe
- 25I-NBOMe
- 25iP-NBOMe
- 25N-NBOMe
- C30-NBOMe
- 25I-NBMD

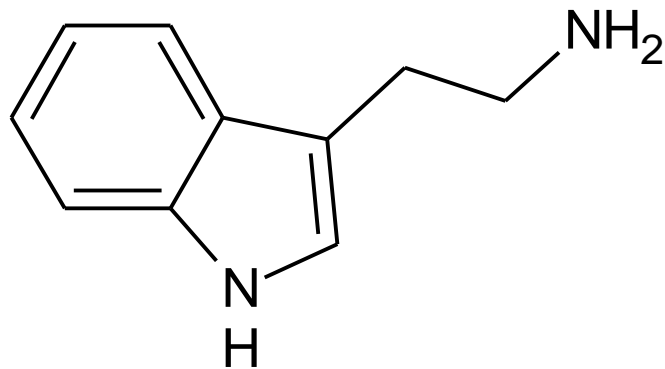


25B-N(BOMe)<sub>2</sub>

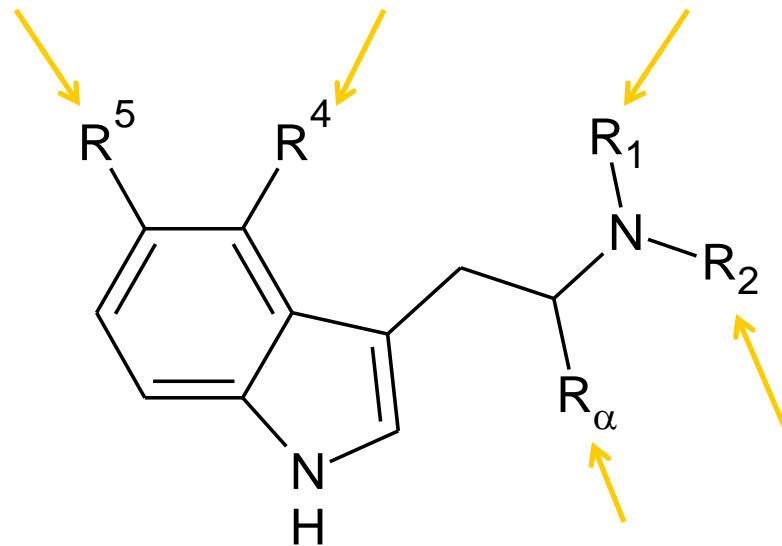


25I-NBMD

# General structure of TRYP

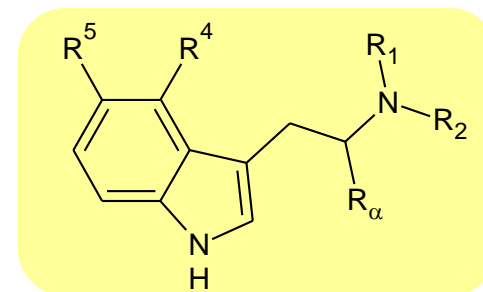


Tryptamine



Tryptamines

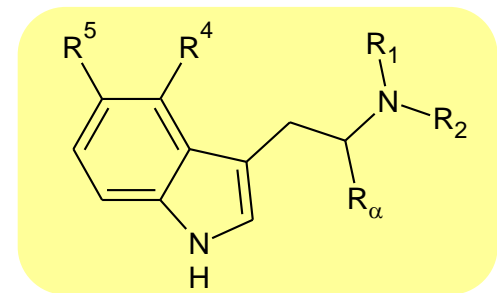
# Reported tryptamines



## • 28 substances

- $R_\alpha$ : No substituent or methyl group
- $R_1, R_2$ :
  - No substituent (only hydrogens) – Tryptamine (T)
  - dialkyl
    - dimethyl - DMT,
    - diethyl – DET,
    - dipropyl – DPT,
    - diisopropyl - DiPT,
  - mixed alkyl groups (MET, MiPT),
  - allyl (DALT)

# Reported tryptamines



## • 28 substances

- Indole (pyrrole ring fused to a benzene ring) – Core 'ring'
- $R_4$ : Hydroxyl or Acetoxy
- $R_5$ : Methoxyl
- $R_2$ : Methyl (one substance - 2-Me-DMT)

# **HOW TO IDENTIFY AND DIFFERENTIATE?**

## Analytical methods

- GCMS (EI)
- LCQTOFMS (ESI)
- Other
  - NMR
  - FTIR
  - (U)HPLC-PDA



# IDENTIFICATION PROTOCOL



# Determination of a molecular mass

- *Difficult to obtain for both PEA and TRYP using 'classical' EI/GCMS*
- Soft ionisation is a better choice
  - Chemical ionisation (CI) or Field desorption (FD) in GCMS,
  - Electrospray ionisation (ESI) in LCMS



# Mass accuracy in QTOFMS

$$ppm = 10^6 \times \frac{m_{measured} - m_{teoretical}}{m_{teoretical}}$$



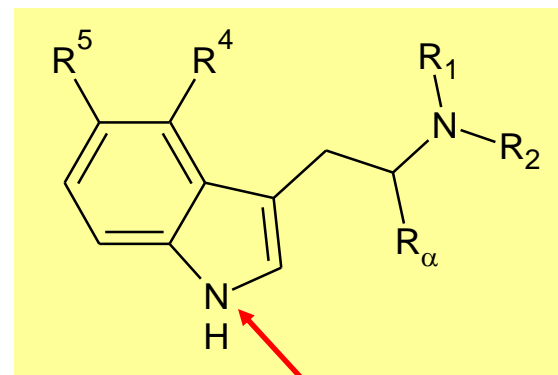
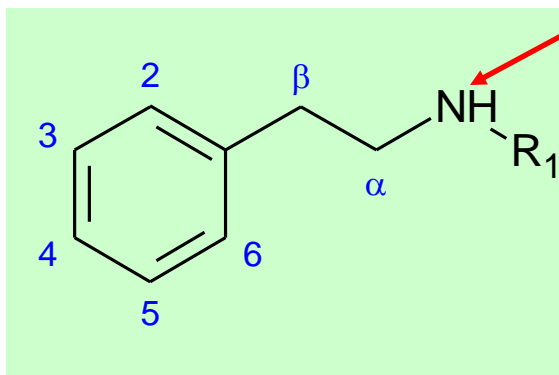
Mass [amu]	Accuracy [amu]	$\Delta m$ [ppm]
100	$\pm 0.0005$	5.0
250	$\pm 0.0005$	2.0
500	$\pm 0.0005$	1.0
750	$\pm 0.0005$	0.65
1000	$\pm 0.0005$	0.5

Mass [amu]	Accuracy [amu]	$\Delta m$ [ppm]
200	$\pm 0,0200$	100
200	$\pm 0,0020$	10
200	$\pm 0,0010$	5
200	$\pm 0,0004$	2
200	$\pm 0,0002$	1

Mass of electron =  $0.000548579903$  amu

# Determination of molecular mass

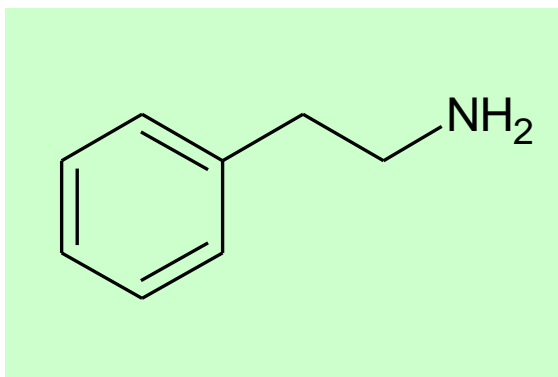
- Derivatisation, e.g., with TFAA
  - PEA: Nitrogen in the side chain
  - TRYP: Nitrogen in the indole ring



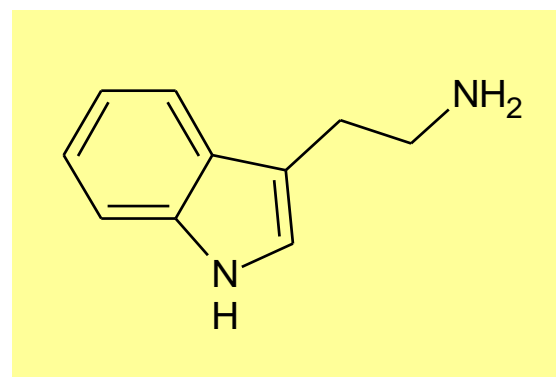
# Parity of molecular ion (mass)

## Nitrogen rule

- **odd** nominal mass indicates an odd number of nitrogen atoms in the molecular ion
- **even** nominal mass indicates an even number of nitrogen atoms



1



2

# Side chain investigation

Immonium ion(s)



Group	R <sub>α</sub>	R <sub>1</sub>	R <sub>2</sub>	Ion
2C	H	H	H	30
DOx	CH <sub>3</sub>	H	H	44
NBOMe	H	H	BOMe	150



TIP

The *m/z* range should begin with 30 (or lower)

# Side chain investigation

## Immonium ion(s)

- Straight chain:

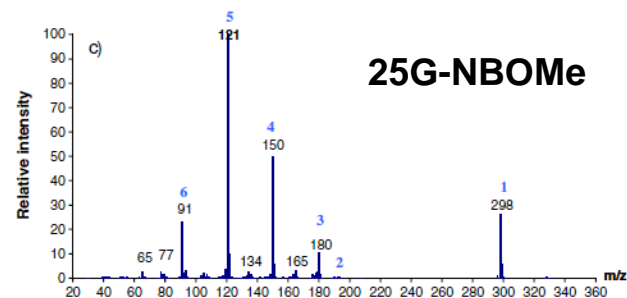
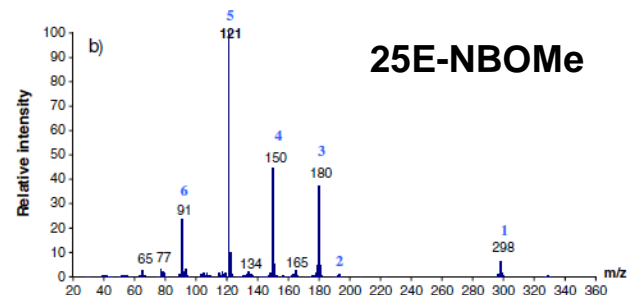
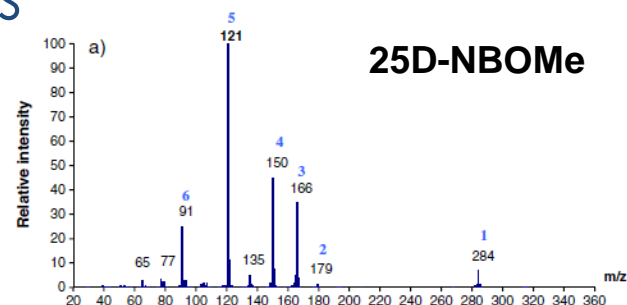
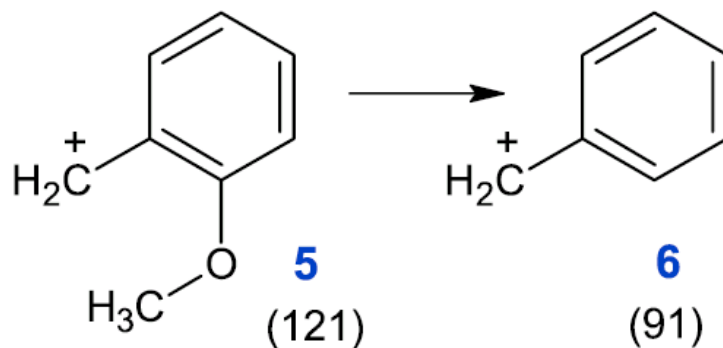
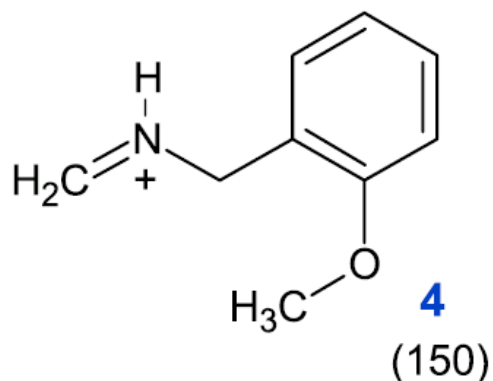


The ions are observed at  $m/z$  30, 44, 58, 72, 86, ... (i.e.  $16 + 14n$ )

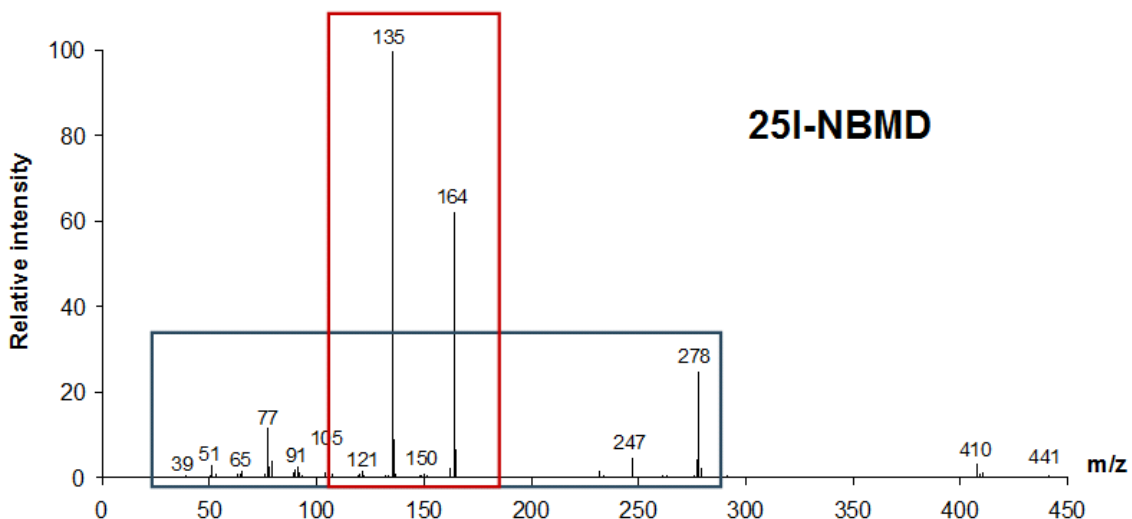
These ions are also observed in cathinones

# Side chain investigation

Characteristic peaks of NBOMe's

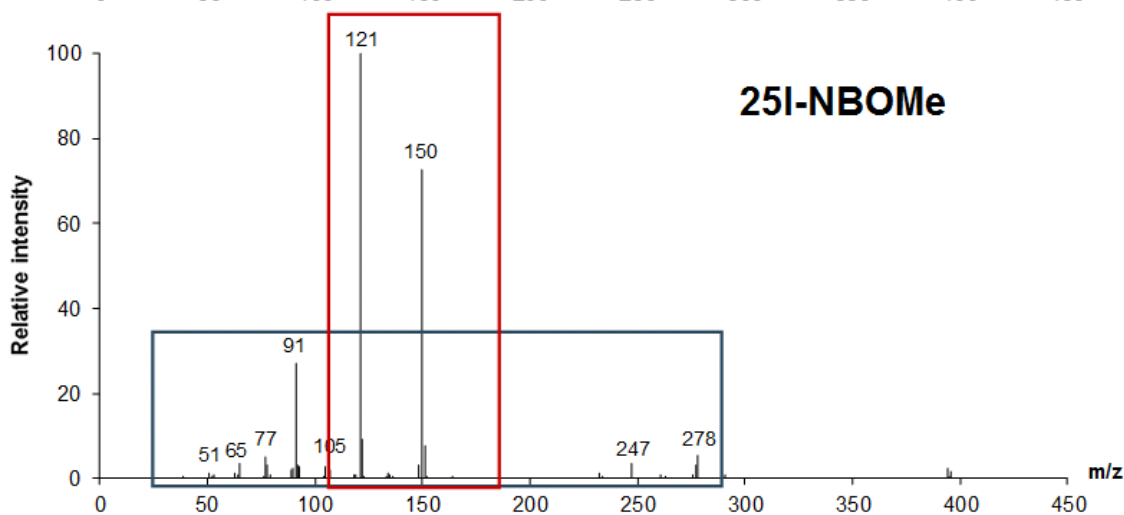
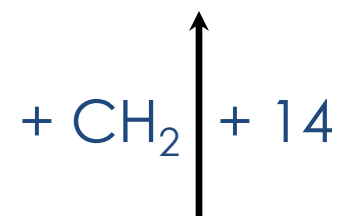


# Side chain investigation



• 164

• 135

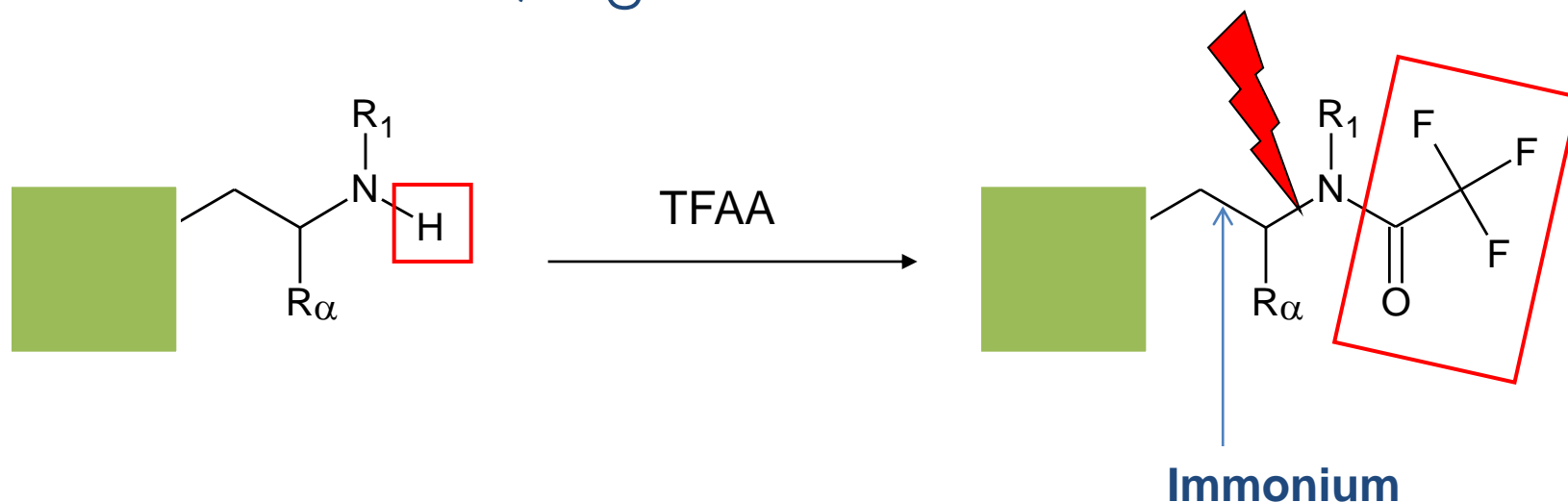


• 150

• 121

# Side chain investigation

- How to check where methyl (alkyl) group is located?
  - Derivatisation, e.g. with TFAA

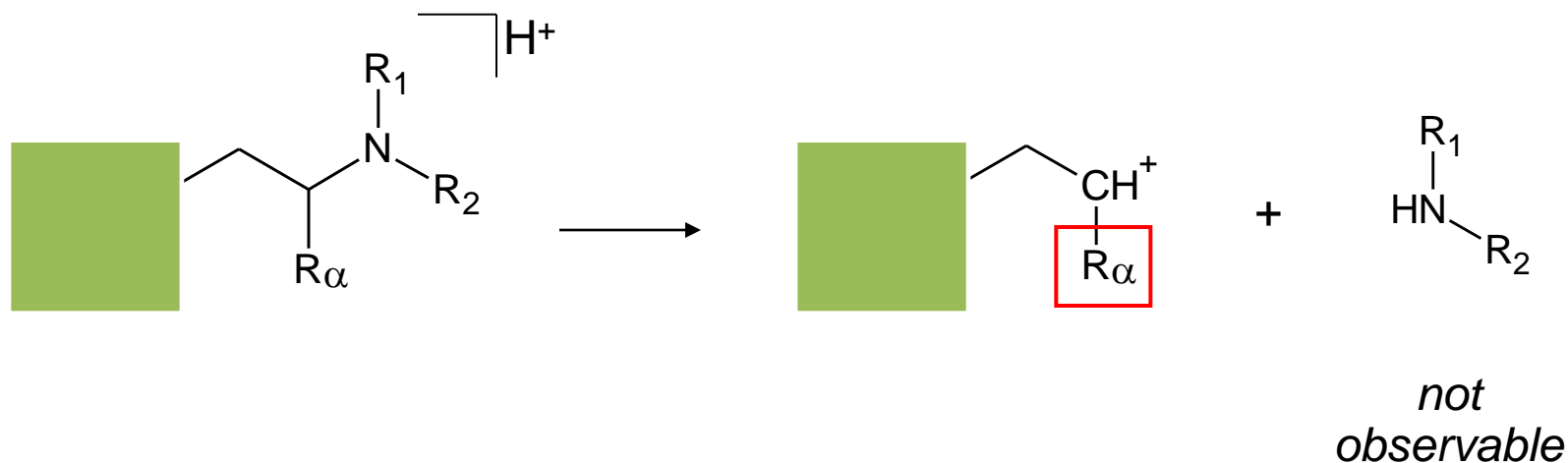


- Use of other ionization technique (e.g. ESI)



# Side chain investigation

## • ESI/QTOFMS

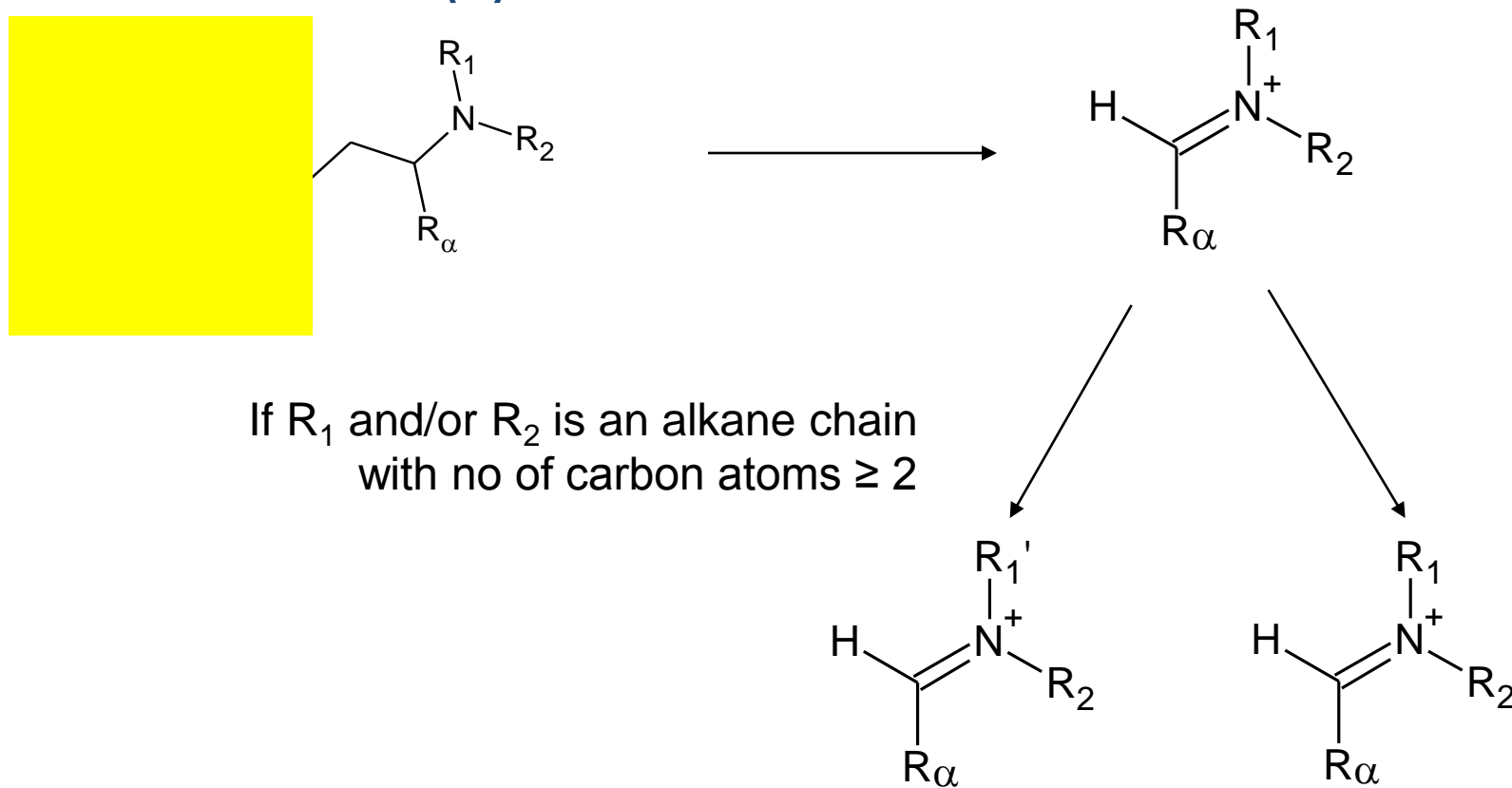


2C and DOx:	$[M+H^+] - 17.0265$ , $[M+H^+] - 31.0422$ , $[M+H^+] - 45.0578$ , $[M+H^+] - 59.0735$ , ...
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NBOMe:	$[M+H^+] - 122.0732$ ( $- 137.0841$ ; $- 152.1075$ )
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# Side chain investigation (TRYP)

Immonium ion(s)



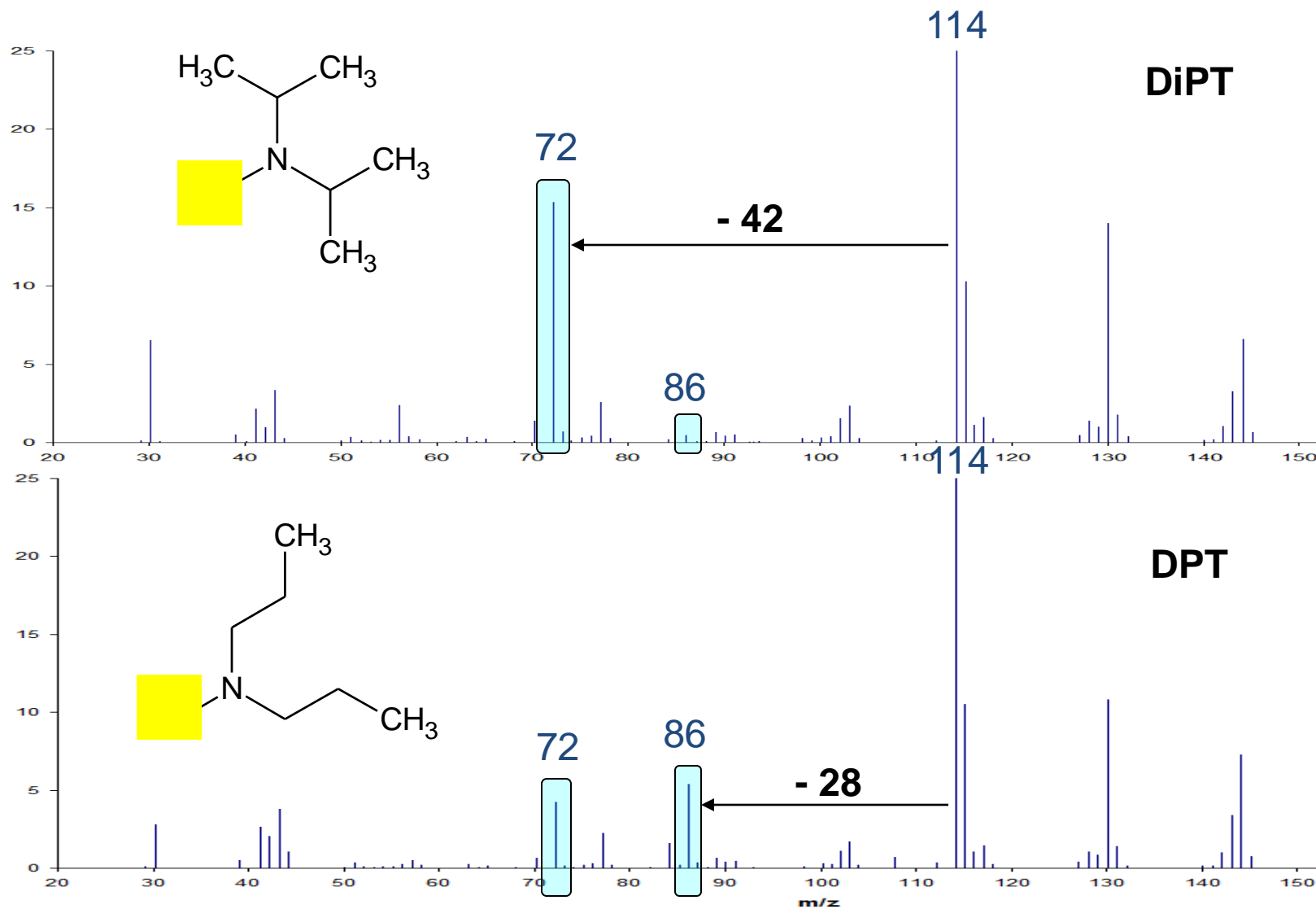
# Side chain investigation

Immonium ion(s)

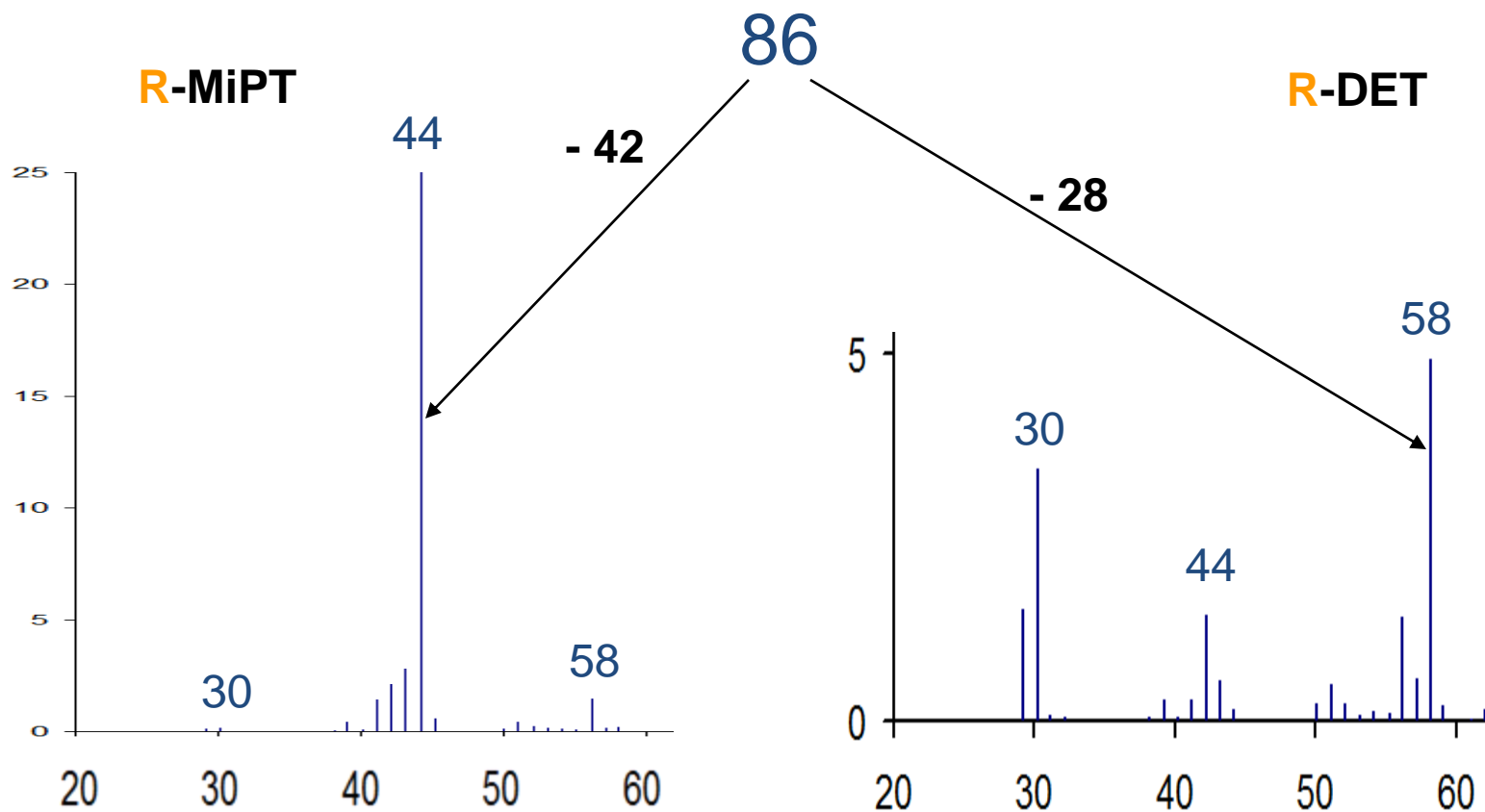
Group	R <sub>a</sub>	R <sub>1</sub>	R <sub>2</sub>	Ion(s)
<b>T</b>	H	H	H	30
<b>AMT</b>	CH <sub>3</sub>	H	H	44
<b>DMT</b>	H	CH <sub>3</sub>	CH <sub>3</sub>	58
<b>MET</b>	H	CH <sub>3</sub>	C <sub>2</sub> H <sub>5</sub>	72 (+ 44)
<b>DET</b>	H	C <sub>2</sub> H <sub>5</sub>	C <sub>2</sub> H <sub>5</sub>	86 (+ 58)
<b>MiPT</b>	H	CH <sub>3</sub>	C <sub>3</sub> H <sub>7</sub>	86 (+ 44)
<b>DALT</b>	H	C <sub>3</sub> H <sub>5</sub>	C <sub>3</sub> H <sub>5</sub>	110
<b>DiPT</b>	H	C <sub>3</sub> H <sub>7</sub>	C <sub>3</sub> H <sub>7</sub>	114 (+ 72)
<b>DPT</b>	H	C <sub>3</sub> H <sub>7</sub>	C <sub>3</sub> H <sub>7</sub>	114 (+ 86 + 72)

Step 1 Step 2 Step 3 Step 4

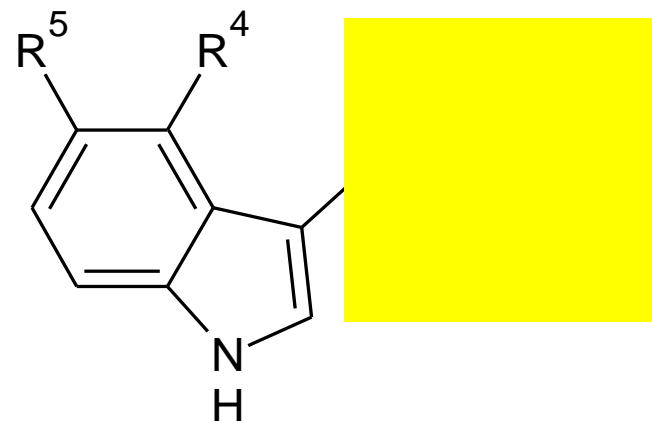
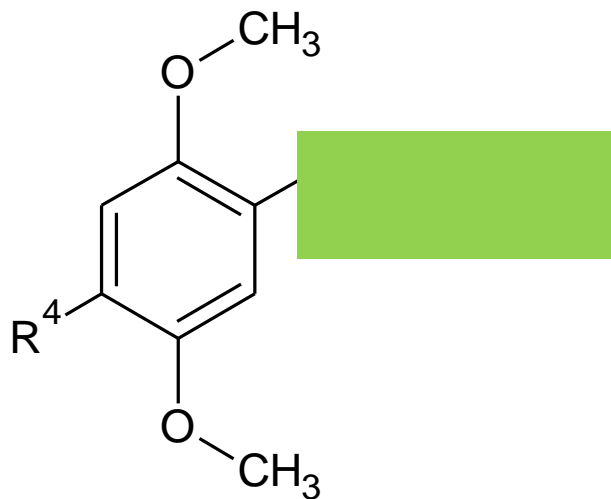
# DiPT vs DPT



# MiPT vs DET

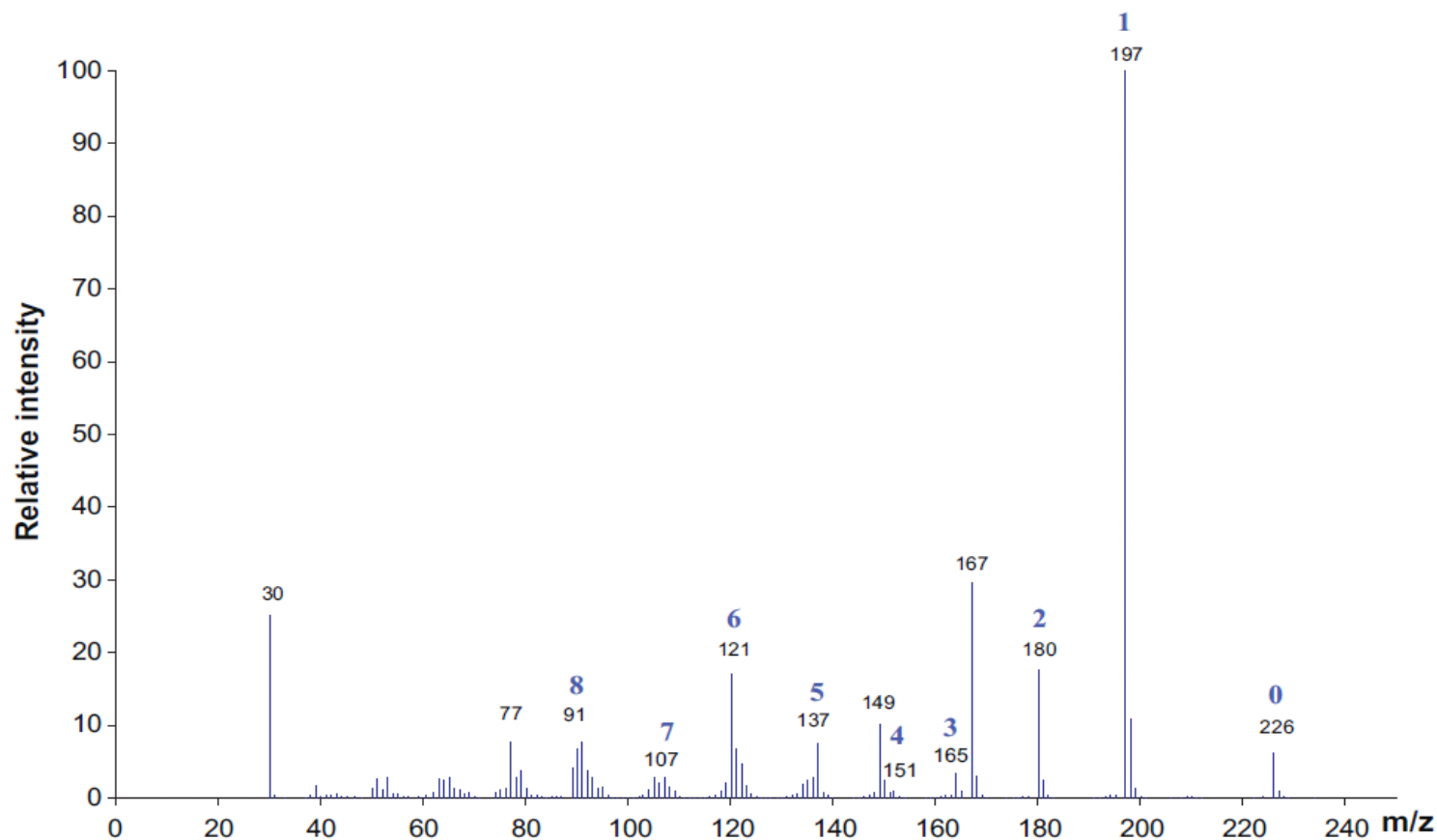


# Core (body) investigation



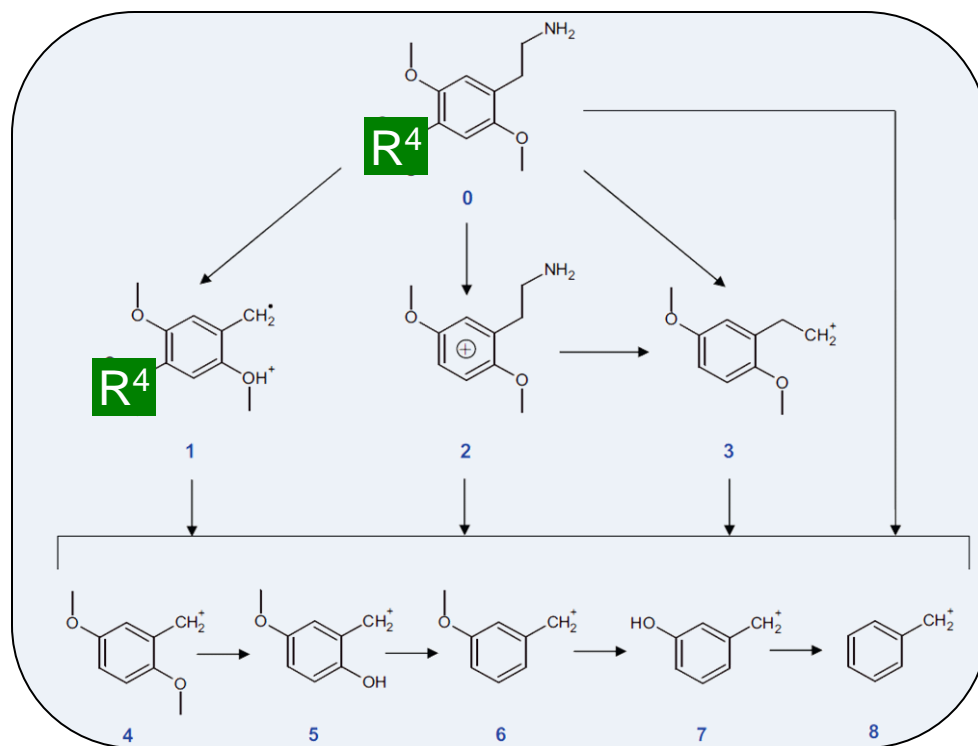
# Core investigation

EI/GCMS spectrum of 2C-N



# Core investigation

EI/GCMS spectra of 2C-series compounds

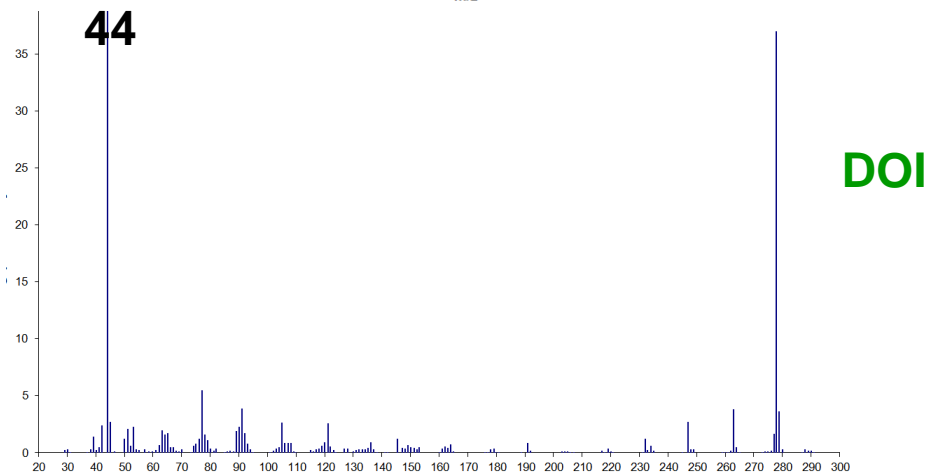
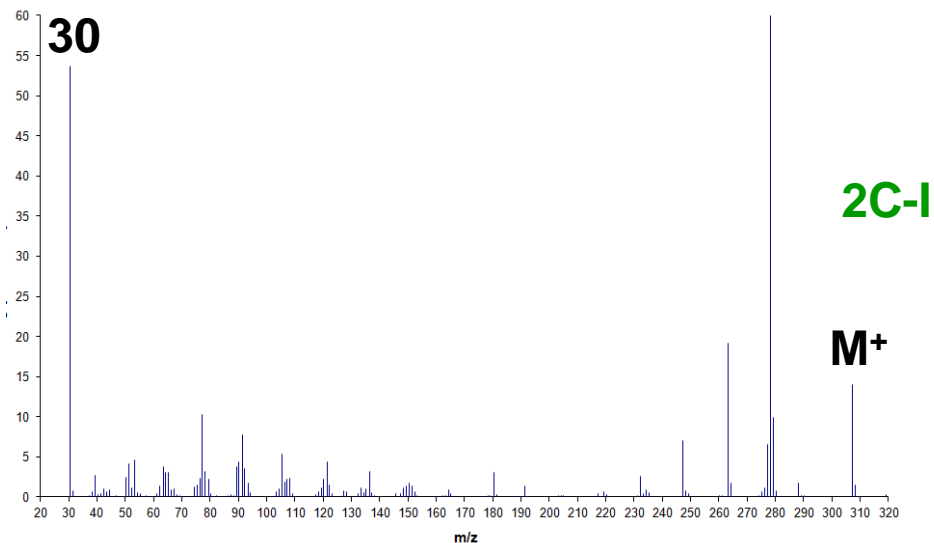


Number	Ion
2	180
3	165
4	151
5	137
6	121
7	107
8	91
	77
	30



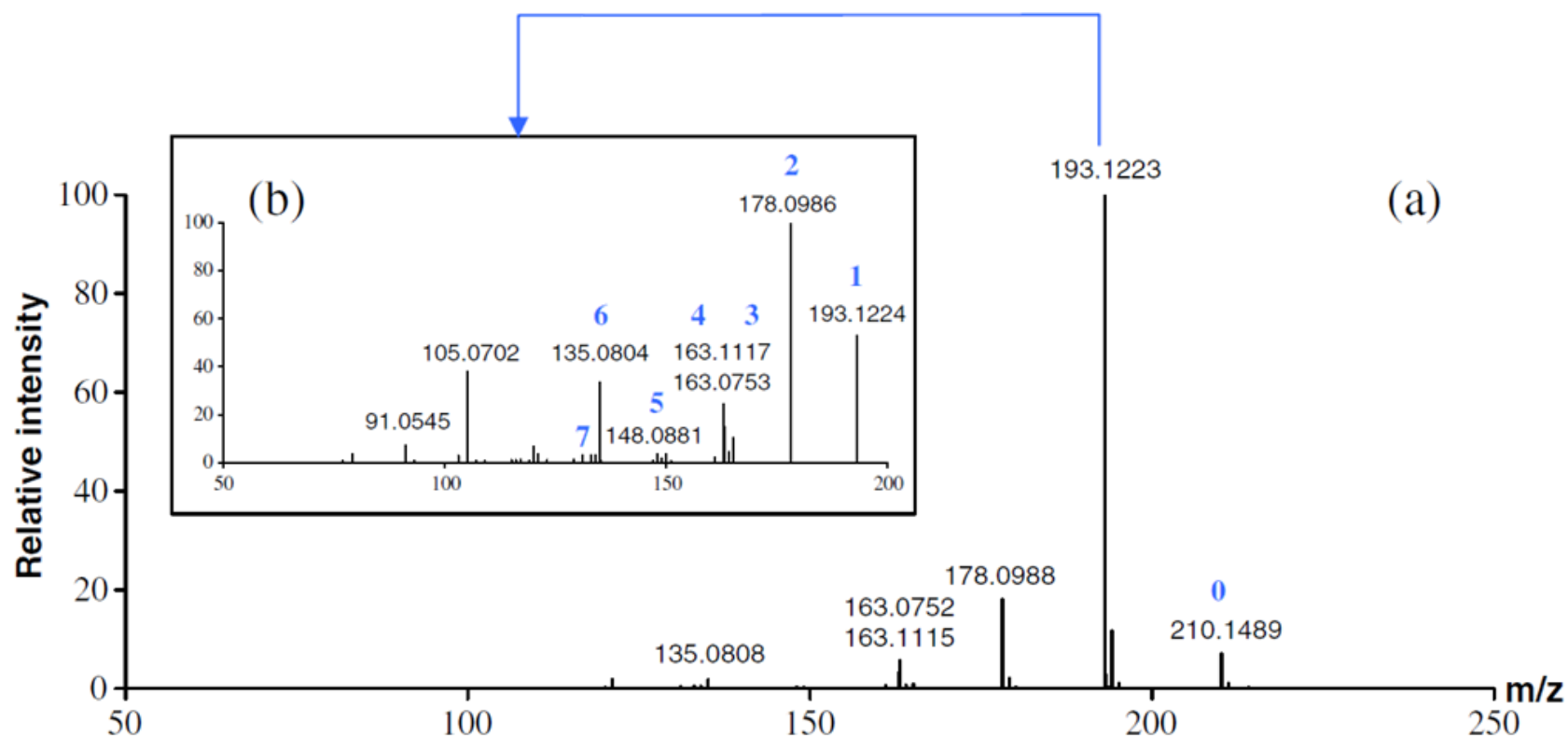
# 2Cs vs DOx vs NBOMe

As the core part of the compounds is identical, many ions are observed at the same  $m/z$  values



# Core investigation

ESI/QTOFMS (2C-E)

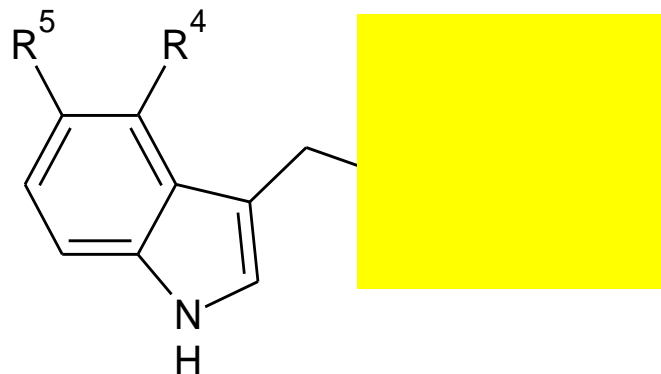


(a) – MS mode; (b) – MS/MS mode

# Core investigation

## Characteristic ions

$R_4, R_5$	Ions
-(H,H)	130
-OH	146
-OCH <sub>3</sub>	160
-OOCH <sub>3</sub>	(176)

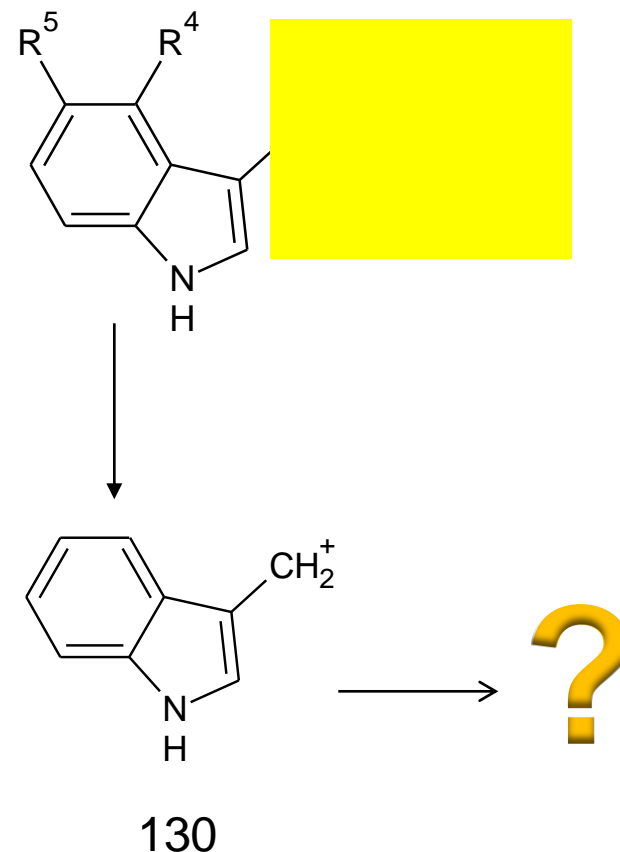


# Core investigation

## ESI/QTOFMS

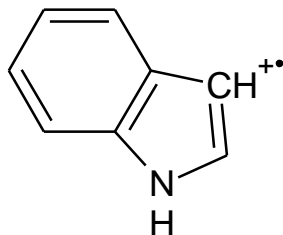
- Ions observed in most TRYP:

- 130
- 117
- 115
- 103
- 91
- 77
- 65 ...

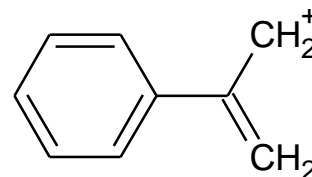


# Fragmentation under ESI

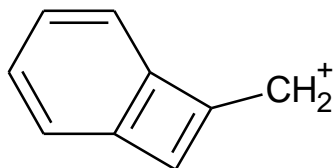
## ESI/QTOFMS



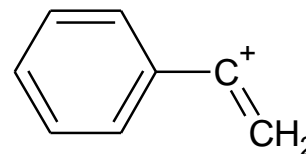
Molecular Formula =  $C_8H_7N$   
 Monoisotopic Mass = 117.057301 Da



Molecular Formula =  $C_9H_9$   
 Monoisotopic Mass = 117.069877 Da



Molecular Formula =  $C_9H_7$   
 Monoisotopic Mass = 115.054227 Da

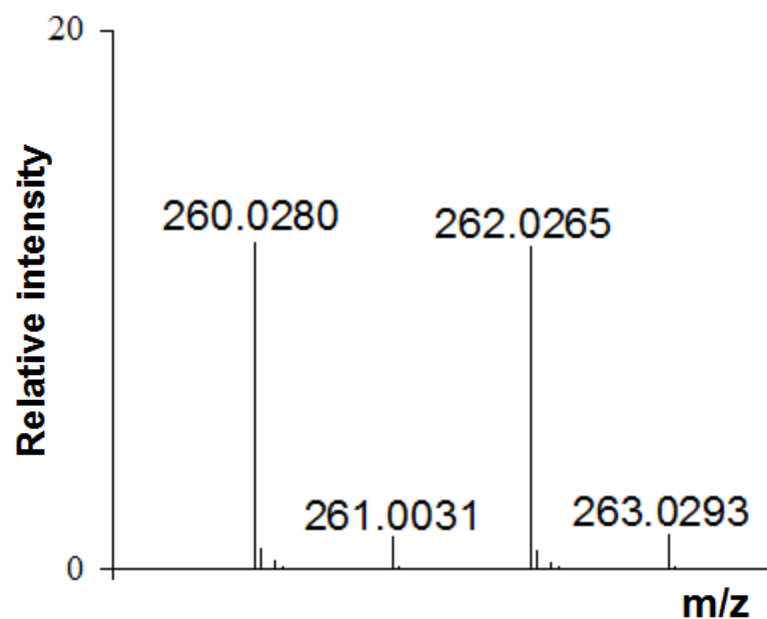


Molecular Formula =  $C_8H_7$   
 Monoisotopic Mass = 103.054227 Da

## **OTHER USEFUL FEATURES**

# Isotope pattern

Compounds (and ions formed during fragmentation) containing bromine and/or chlorine atoms have characteristic isotope pattern



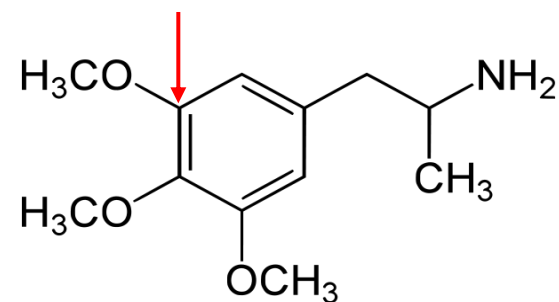
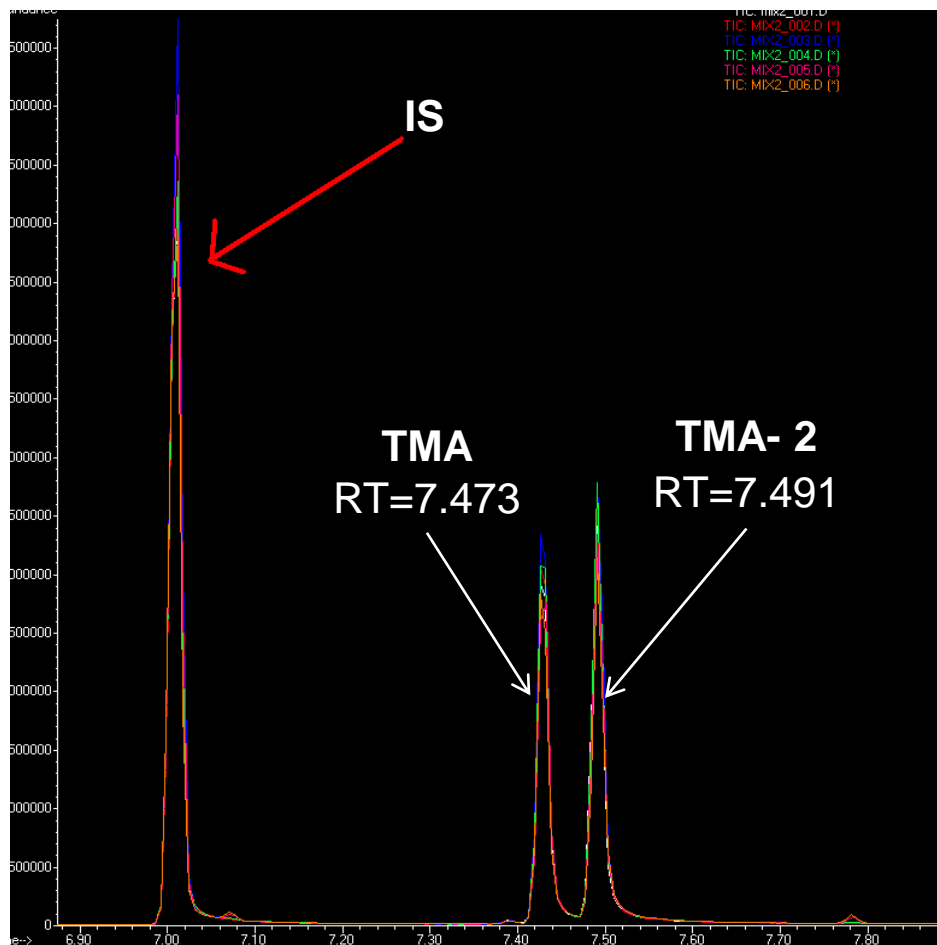
$m/z$	Relative intensity [%]	
	theoretical	observed
260.0281	100.00	100.00
261.0313	11.43	10.14
262.0261	98.28	98.65
263.0293	11.19	10.46
264.0316	0.98	0.84

# Retention time locking (RTL)

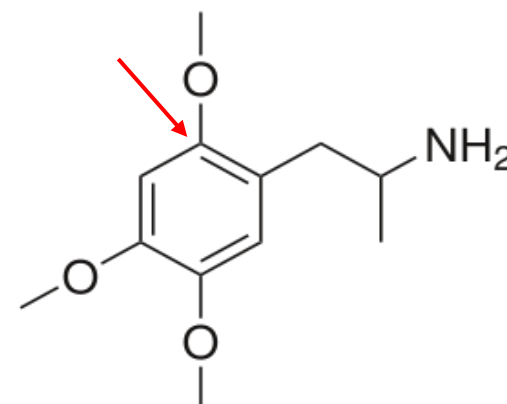
- 'Locking' of retention time of the internal standard (IS) significantly improves repeatability of retention times of analytes
- Based on the analysis of 34 substances from different chemical classes, it was estimated that:
  - intraday precision was 0.0046 min (0.27 s), which corresponded to  $RSD = 0.06\%$ ,
  - interday precision was 0.0078 min (0.47 s), which corresponded to  $0.10\%$ .
- It indicates HIGH IDENTIFICATION POWER



# RTL – Example



**TMA**



**TMA-2**

# SUMMARY

- Identification of new psychoactive substances is a complex process, due to similarities in their chemical structures and physicochemical properties
- Mass spectra of PEA and TRYP contain many characteristic peaks
- Detailed analysis of fragmentation processes allows us to predict mass spectra of new substances

# References

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