MS SERVICE OF SMALL MOLECULES

phone lines:
Office: 117
Spectrometers: ESI, APCI: 282, 461
EI, CI: 505
MALDI: 508

e-mail: msrequest@uochb.cas.cz

THE MS SERVICE WHICH WE PROVIDE

- a confirmation of a molecular structure by determination of exact mass and assigning a suitable formula
- MALDI of high molecular compounds such as proteins, oligonucleotides and some polymers
- a help with an identification of a structure according to fragmentation by EI or CI and measurement MS/MS analysis
- GC/LC-MS analysis
- GC-MS self-service - after training and making a schedule
- +/- ESI self -service with low resolution, using direct injection or LC - after training and making a schedule

THE MASS SPECTROMETRY SERVICE

WHAT SHOULD YOU DO, IF YOU LIKED TO GET YOUR SAMPLES MEASURED?

- go to IOCB internal pages ➡ forms (formuláře) ➡ MS sample submission
- fill the registration “not registered?” and insert a new application “create a new request”

- the more information you give us, the quicker and easier the measurement will be for us (please fill the “storage and the special handling”, the “solubility”, the “request of the sample returning” and the most important is “submit the structure”)
- each sample has to be written under its own request separately
- PRINT THE FORM AND BRING IT WITH THE SAMPLE TO US

WHERE YOU FIND THE MS LABORATORY/OFFICE AND YOUR RESULTS?

- lab + office in the basement, number of the door 24
- results and MS spectra on the request system 

- more information and software downloads “available services” 

- exact mass calculator
- MSreVIEW software (mMass)

MSreVIEW SOFTWARE - mMass (*.raw*)

mMass *belongs to the wide family of open source software. It is written mostly in Python language, uses wxPython libraries for graphic user interface and is released under GNU General Public License. Therefore it is portable to different computer platforms and has a good potential to be easily modified or extended by modules of specific needs.*

-using this software you can open your data, which are in a . raw file type and simply work with them

! The advantages over *.pdf!*:

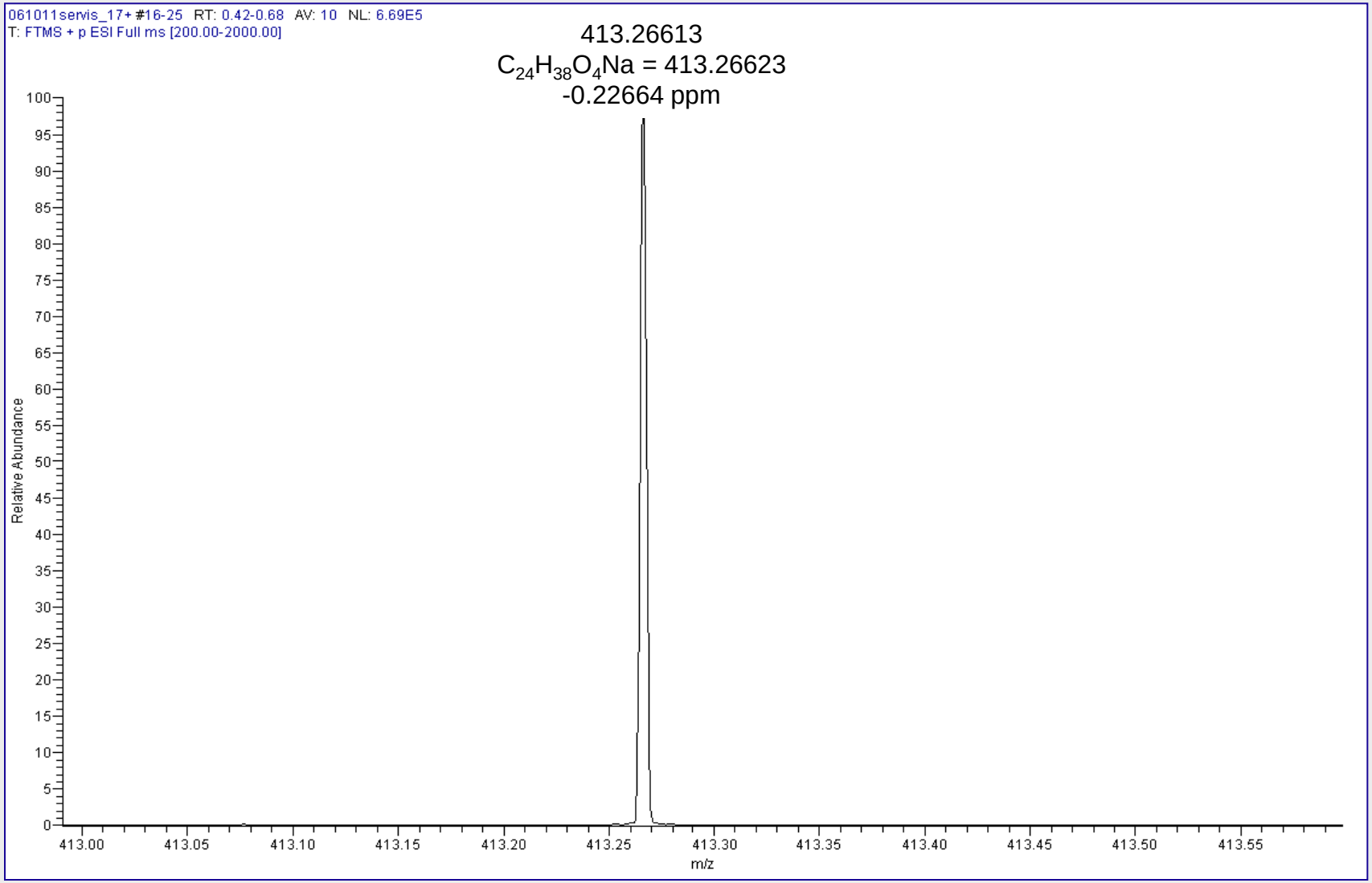
- zoom and normalize (to eliminate different intensity ranges provided by different measurement or instruments) spectra
- manually or automatically label peaks and annotate them
- generate molecular formula for measured mass (only for HR spectra)
- *Mass calculator* - generate a list of ion series and model isotopic pattern

MSreVIEW SOFTWARE – mMass (*.raw*)

- *Deisotoping* - determine peaks charge and remove unwanted isotopes from the final peak list
- *Deconvolution* - recalculate all the multiply-charged species into its singly-charged form
- *Peak differences* - tool simply generates a table of differences between all the peaks in a peak list
- *Compare Peak Lists* - tool can be utilized to compare peak lists between open documents

Do not afraid of MSreVIEW software it is easy to work with, furthermore you can find a link for really well arranged manual on our Request site.

LR and HR ESI SPECTRA (*.pdf*)



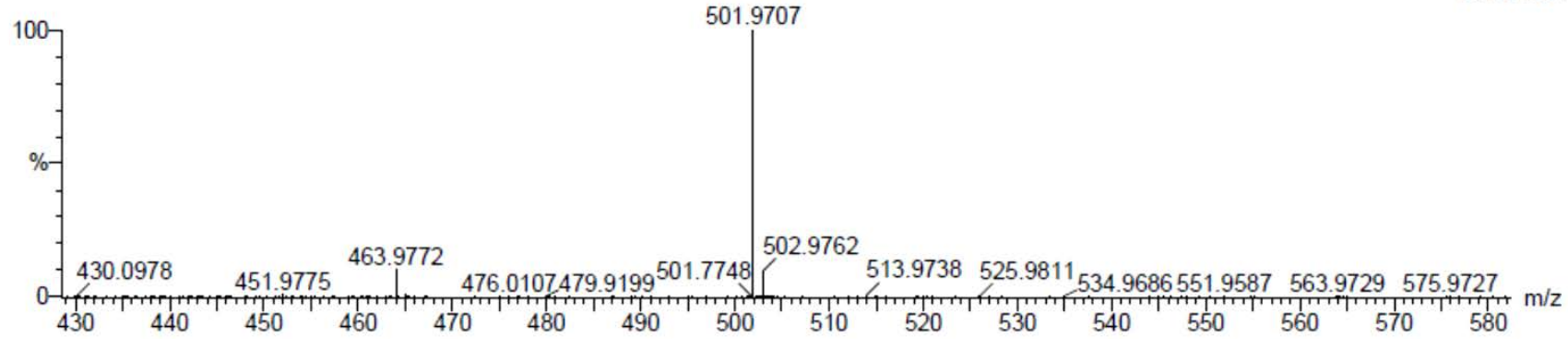
THE LR and HR EI/CI SPECTRA (.pdf)

Elemental Composition Report **Page 1**

Single Mass Analysis (displaying only valid results)
Tolerance = 10.0 PPM / DBE: min = -100.0, max = 1000.0
Element prediction: Off

Monoisotopic Mass, Odd and Even Electron Ions
17 formula(e) evaluated with 1 results within limits (up to 50 closest results for each mass)
Elements Used:
C: 3-15 H: 0-1 N: 1-1 F: 0-30
heptacosa
heptacosa 162 (2.701) Cm (160:191)

TOF MS EI+
8.82e+004



Minimum:				-100.0		
Maximum:		5.0	10.0	1000.0		
Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	Formula
501.9707	501.9711	-0.4	-0.8	0.5	3495.1	C9 N F20

SAMPLES

Solid samples:
It is better to submit a solid sample (100 µg - 1.0 mg).

Dissolved samples:
Please use only solvents for LC/MS or distilled solvents.

- samples should be fully dissolved.

All instruments are usually capable of detecting at least 1 µg/mL.

However, it is good habit to submit around 1 mg/mL.

RECOMMENDED SOLVENTS

Which solvent can I use? - it depends on the type of ionization and whether you want to use GC chromatography before MS or not.

- **ESI:** the most preferred solvents are **MeOH**, **H_2O** and **ACN** - a solution should be free from nonvolatile buffers and and another additives.

  $CHCl_3$ and **acetone** are also acceptable in mixture with MeOH or ACN.
- **APCI: MeOH, ACN, IPA, $CHCl_3$, EtAc, EtOH** and **acetone**
- **EI/CI with direct probe:** solid samples or dissolved in volatile solvents.
- **EI/CI coupled with GC:** hexan, ether, $CHCl_3$, CH_2Cl_2, MeOH, EtOH. The samples for GC/MS must be free from strong acid, bases and oxidizing compounds.
- **MALDI:** in a solution free from nonvolatile buffers, solvents and surfactants

! Please, avoid solvents with high boiling points (DMSO, DMF) !

USUAL CONTAMINANTS

- **Phthalates** - from plastics, contaminated solvents

  diisobutylphthalate (masses EI: 149, 205, 223, 278, ESI: 279, 301)

  diisooctylphthalate (masses EI: 149, 167, 279, 390, ESI: 391, 413)

- **Antioxidants** - from plastics : irganox, irgafos

  (the most common masses EI: 316 ,591, 647, 662, ESI: 663, 685)

- **Polysiloxans** - from silicone rubber, teflon lined caps from vials

  (the most common masses EI: 73, 147, 221, 295, 355, ESI: 297, 371, 445, 519)

- **PEGs** - extracted polymer from teflon/silicon septum (+44 series)

- **Amides** - from plastics - oleamide (ESI: 282), stearamide (ESI: 284), erucamide (ESI: 338)

- **Detergents** - Triton X-100

- **Fatty acids** - palmitic and oleic acid from skin (masses negESI : 255, 283)

SALTS, BUFFERS and ANOTHER ADDITIVES

- **volatile** - *TFA, FA, acetic acid, ammonium acetate, ammonium formate etc.*

  - can be used in lower concentrations (up to 10mM)

- **nonvolatile** - *phosphate, sulfate buffers, SDS, CHAPS, TRIS, HEPES etc.*

  - should not be used because of decreasing signal, salt clusters creation and unbearable contamination of mass spectrometers

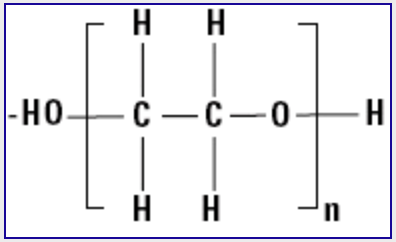
	MW	MALDI	MALDI	ESI	ESI
Surfactant/ buffer/ salt	(g/mol)	(mM)	(wt.%)	(mM)	(wt.%)
TRIS	121	100	1	n.a.	n.a.
HEPES	238	100	2,4	n.a.	n.a.
Urea	60	500	3	n.a.	n.a.
Dithiotreitol	154	500	7,7	n.a.	n.a.
Guanidine	96	250	2,4	n.a.	n.a.
Glycerol	92	130	1,2	n.a.	n.a.
Triton X-100	628	1,6	0,1	1,6	0,1
Tween20	1228	n.a.	n.a.	0,81	0,1
SDS	288	0,35	0,01	0,34	0,01
CHAPS	615	0,16	0,01	1,6	0,1
Sodium Azide	65	15	0,1	3,1	0,02
NaCl	58	50	0,29	n.a.	n.a.
Sodium Acetate	82	50	0,41	n.a.	n.a.
TFA	114	n.a.	n.a.	4,4	0,05
$NaHPO_4$	120	10	0,12	10	0,12

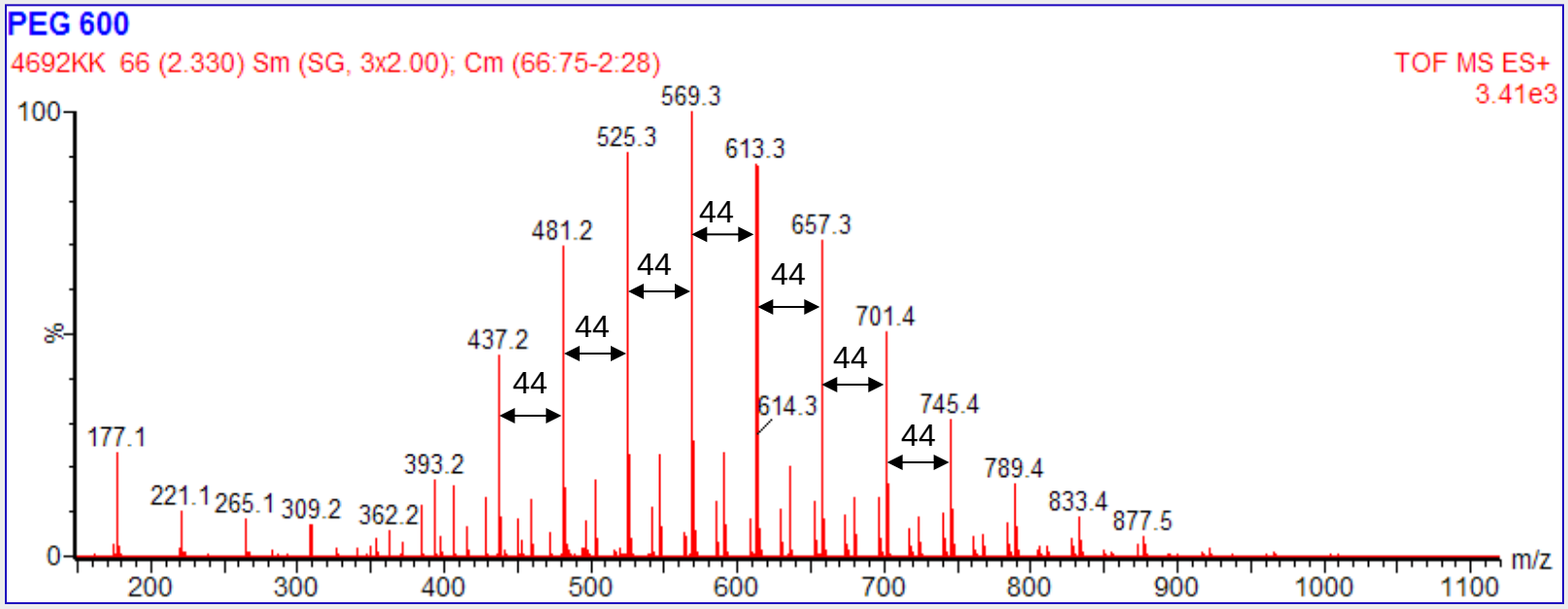
http://masspec.scripps.edu/services/proteomics/saltol.php

ADDUCTS AND CLUSTER PEAKS IN +/-ESI

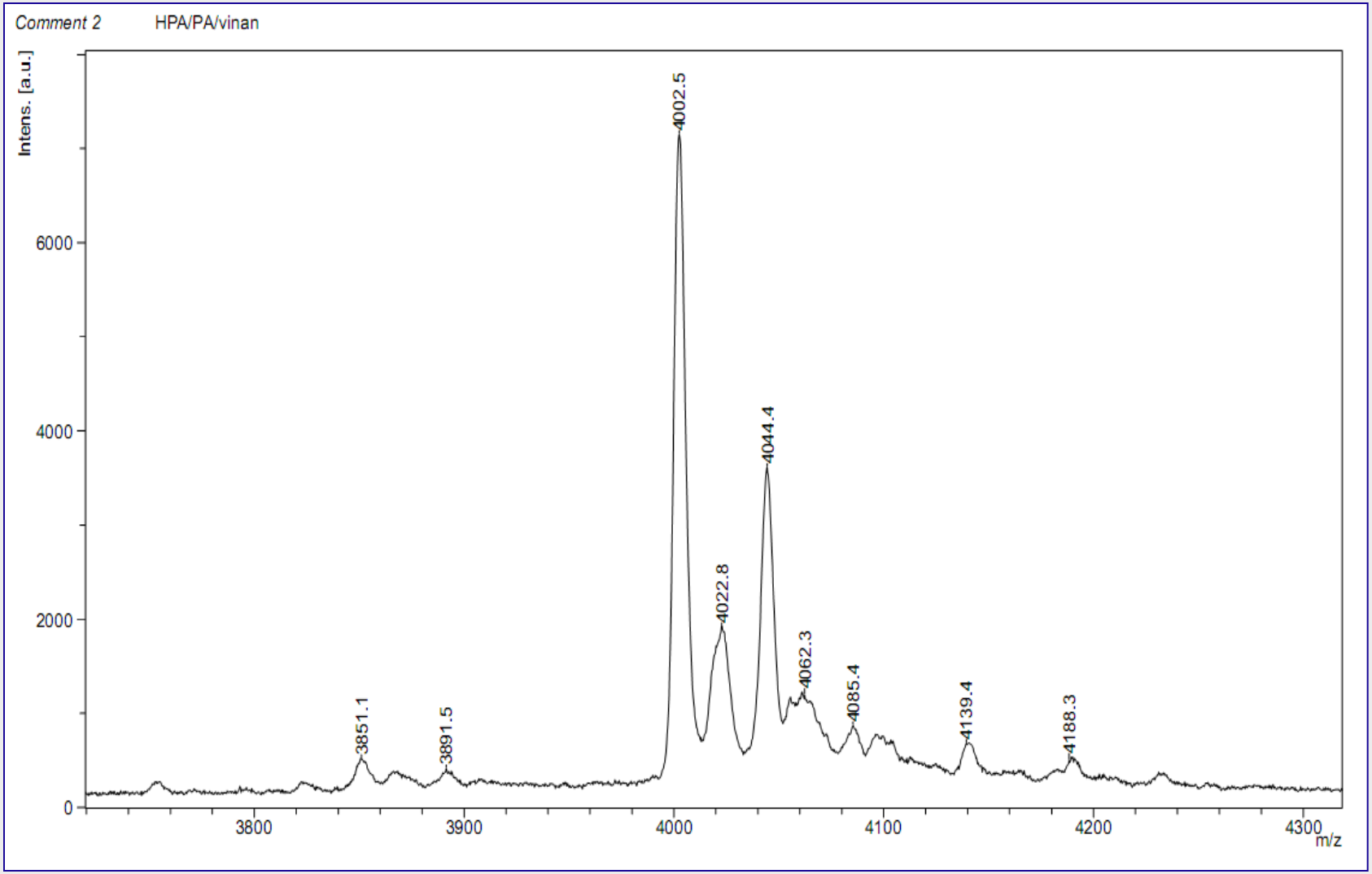
- +23n….sodium adducts….+ESI
- +32n….methanol adducts….+ESI
- +39n….potassium adducts….+ESI
- +41n….acetonitrile adducts….+ESI
- +44n…..polyethylene glycol related (-CH_2CH_2O-)n....+ESI
- +53n…..ammonium chloride adducts (NH_4Cl)….+ESI
- +63n…..ammonium formate adducts ($HCOONH_4$)….+ESI
- +68n…. sodium formate adducts (HCOONa)….+/-ESI
- +74n…..polysiloxanes ($Si(CH_3)_2O$)….+/- ESI
- +77n…..ammonium acetate salts (CH_3COONH_4)….+ESI
- +82n.….sodium acetate adducts (CH_3COONa)....+/-ESI
- +114n ...TFA (trifluoroacetic acid) adducts (CF_3COOH)….-ESI
- +136n….sodium TFA (trifluoroacetic acid) adducts (CF_3COONa)….+/-ESI
- +288n …SDS (sodium dodecylsulfate) adducts….-ESI

ESI SPECTRUM OF POLYETHYLENE GLYCOL 600

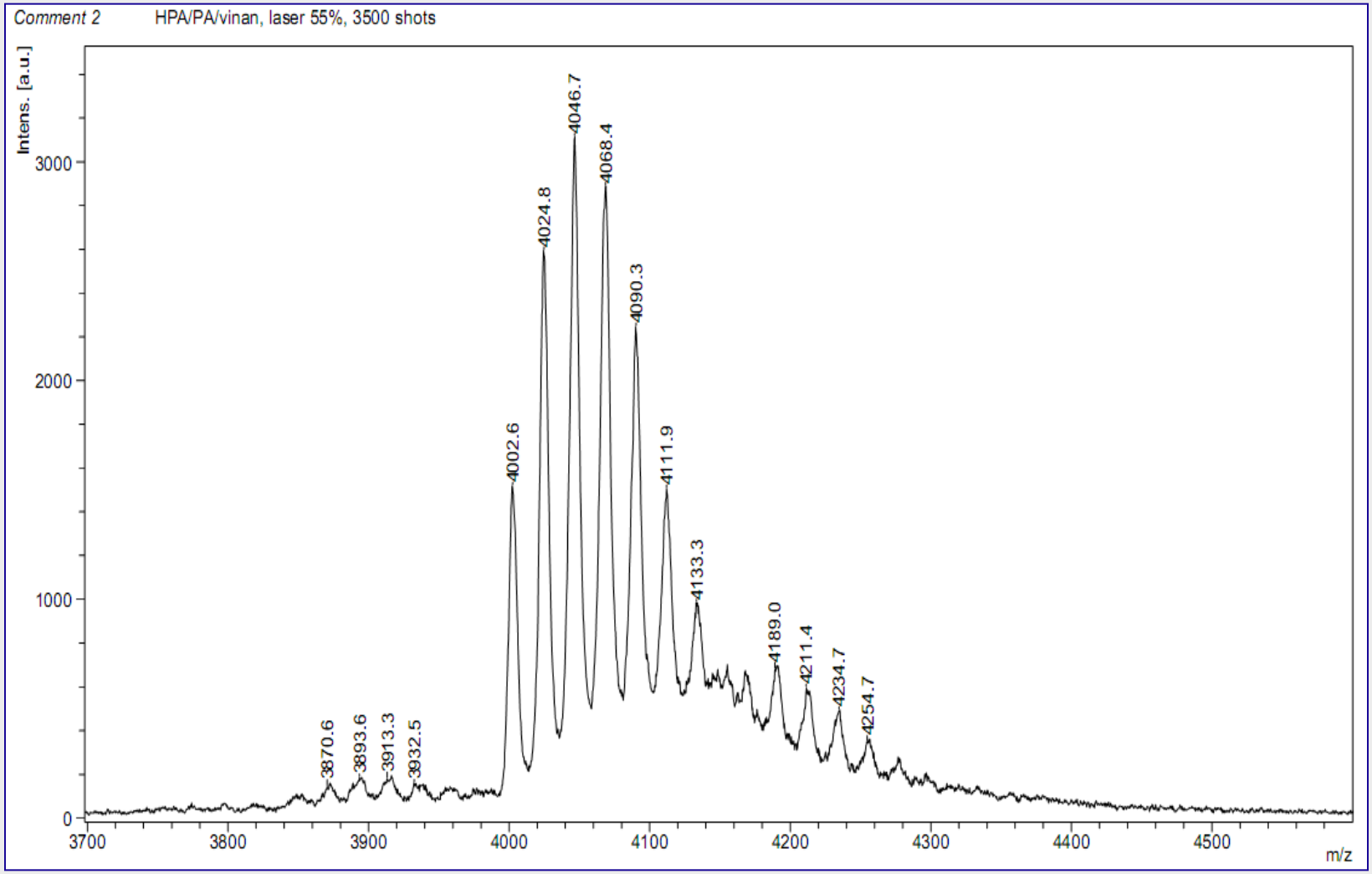




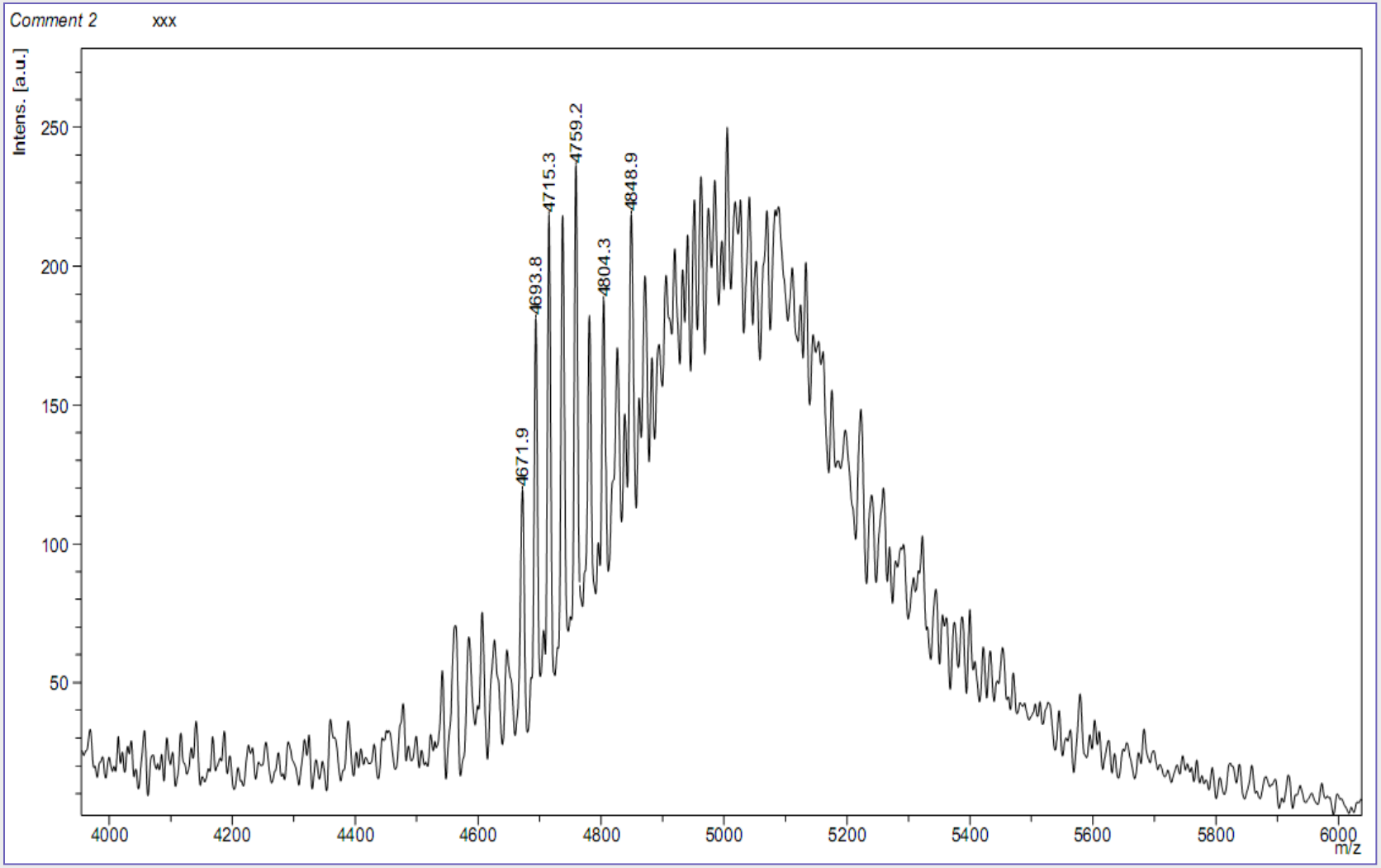
MALDI OF CLEAN SAMPLE



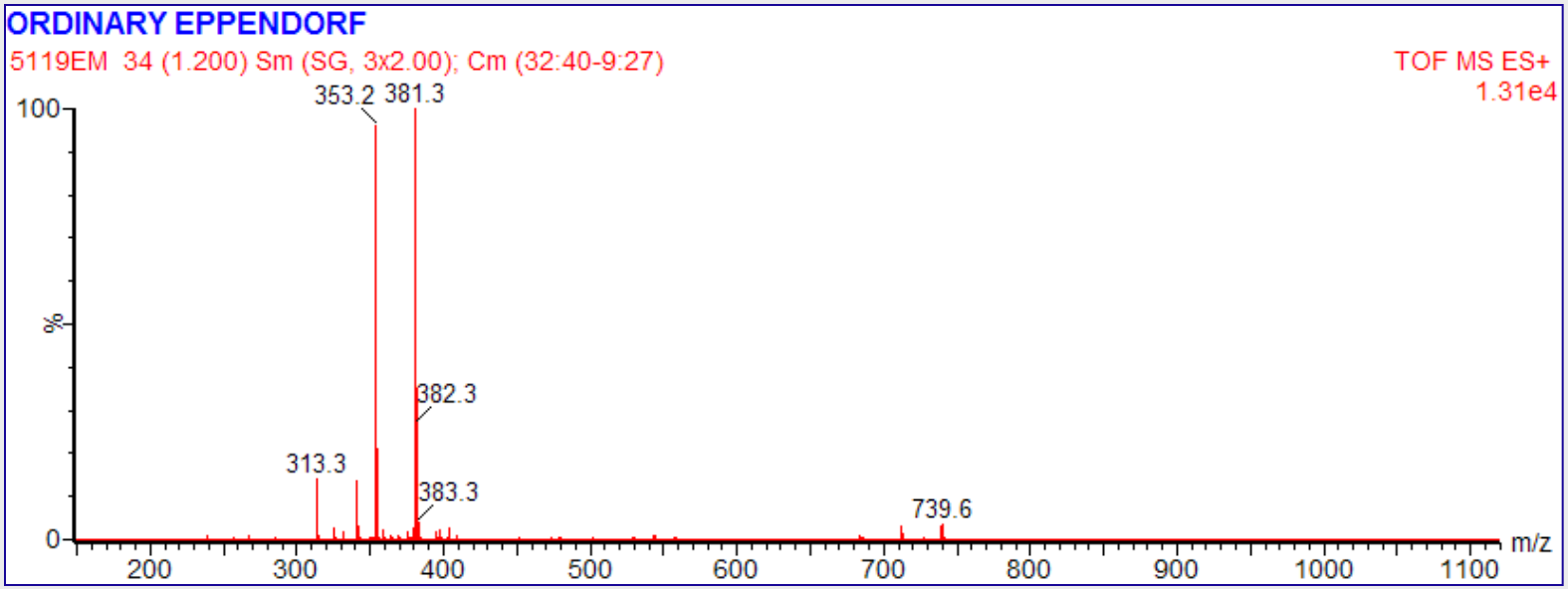
MALDI OF SAMPLE WHICH IS SLIGHTLY CONTAMINATED WITH SALTS

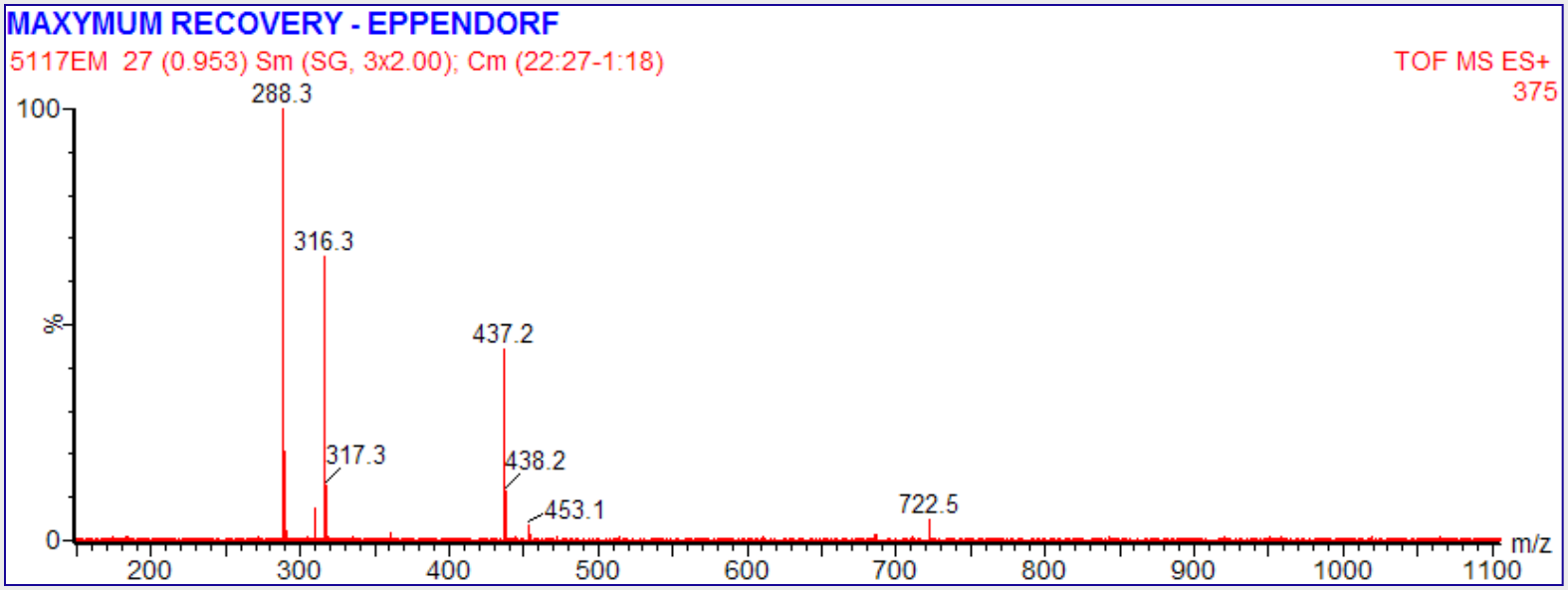


MALDI OF SAMPLE WHICH IS HIGHLY CONTAMINATED WITH SALTS

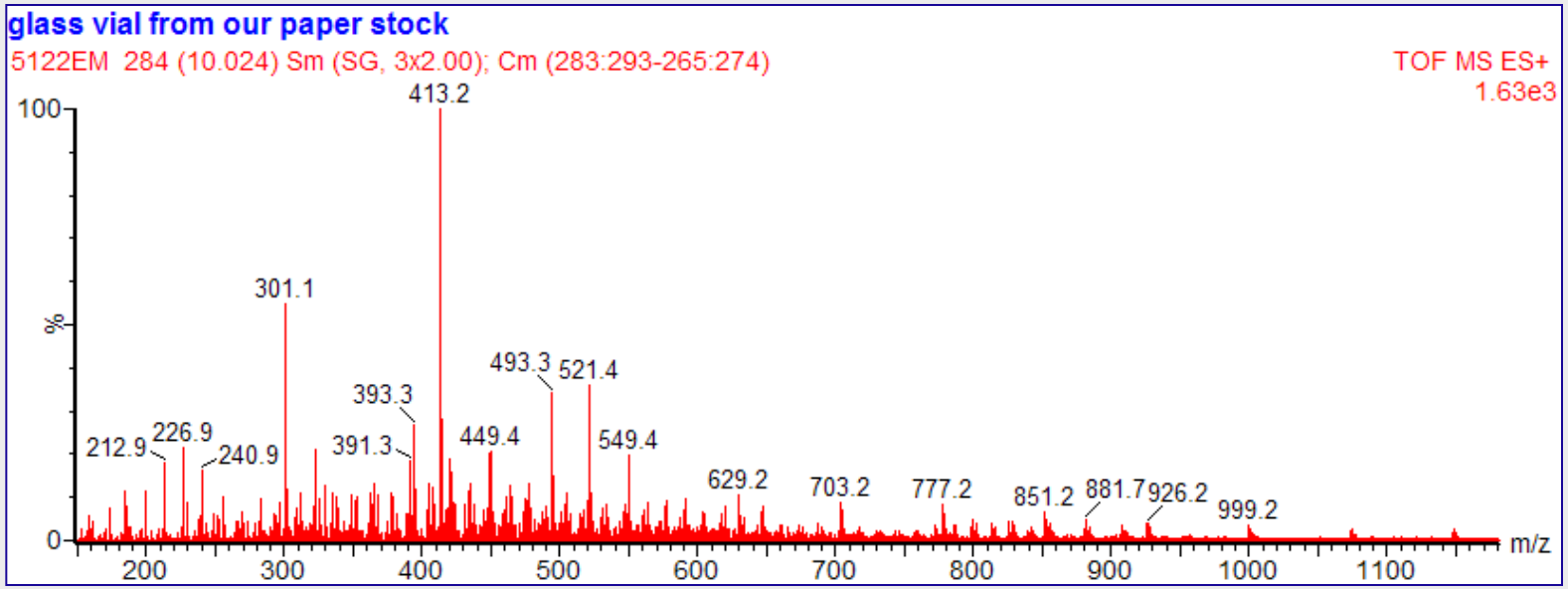


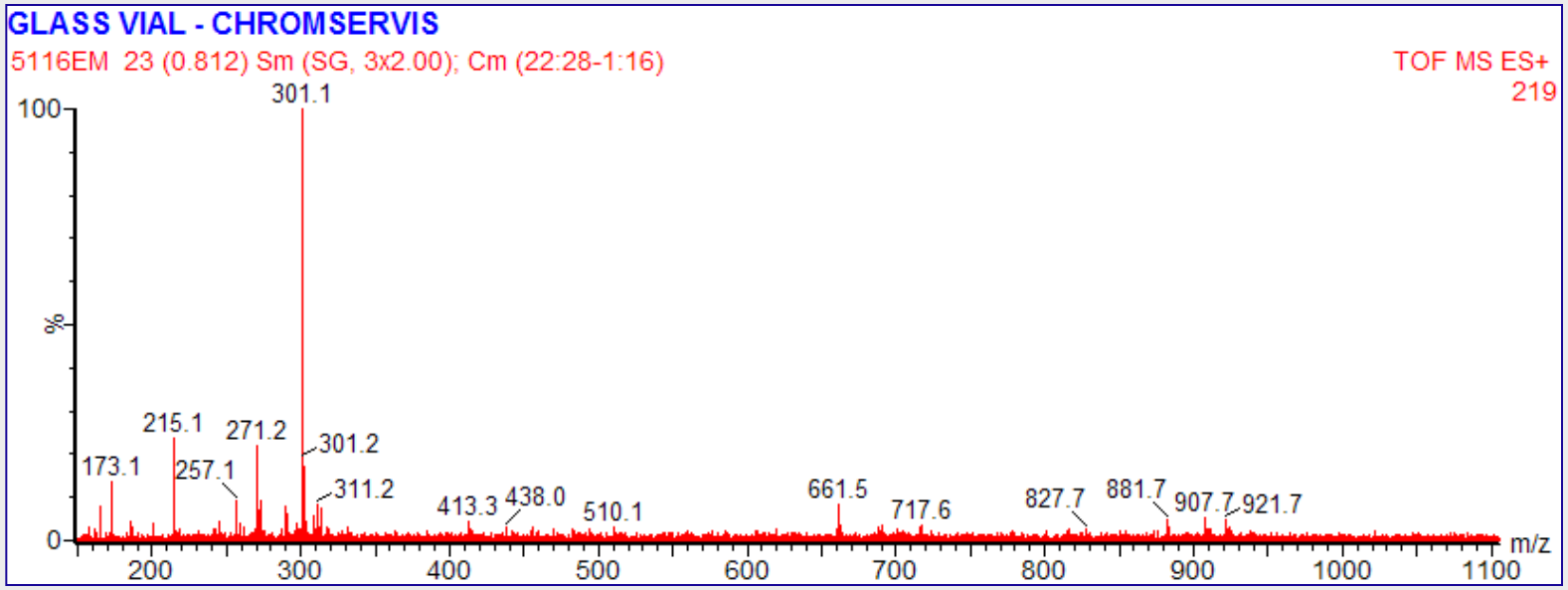
MeOH FROM EPPENDORFS





MeOH FROM GLASS VIALS





Thank you for your attention