

**Analytical characterization and pharmacological evaluation of the new psychoactive substance 4-fluoromethylphenidate (4F-MPH) and differentiation between (±)-*threo*- and (±)-*erythro*- diastereomers.**

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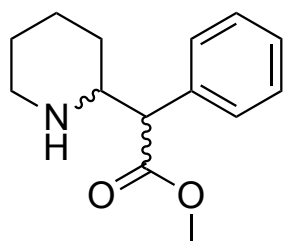
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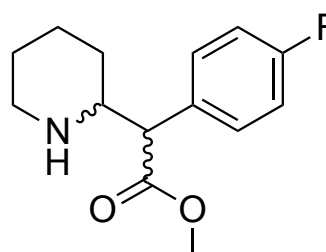
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Methylphenidate

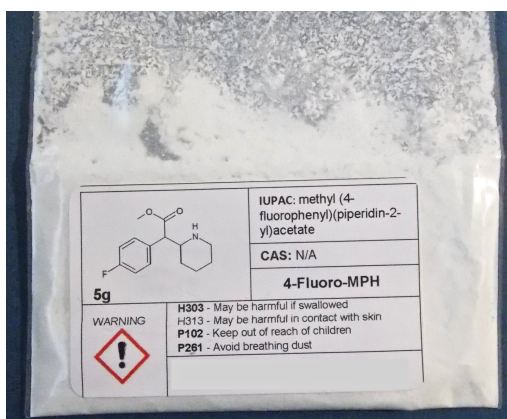
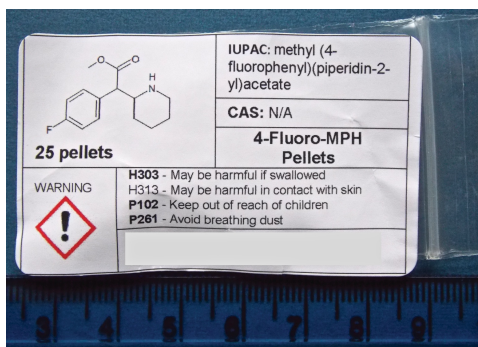


4-Fluoromethylphenidate

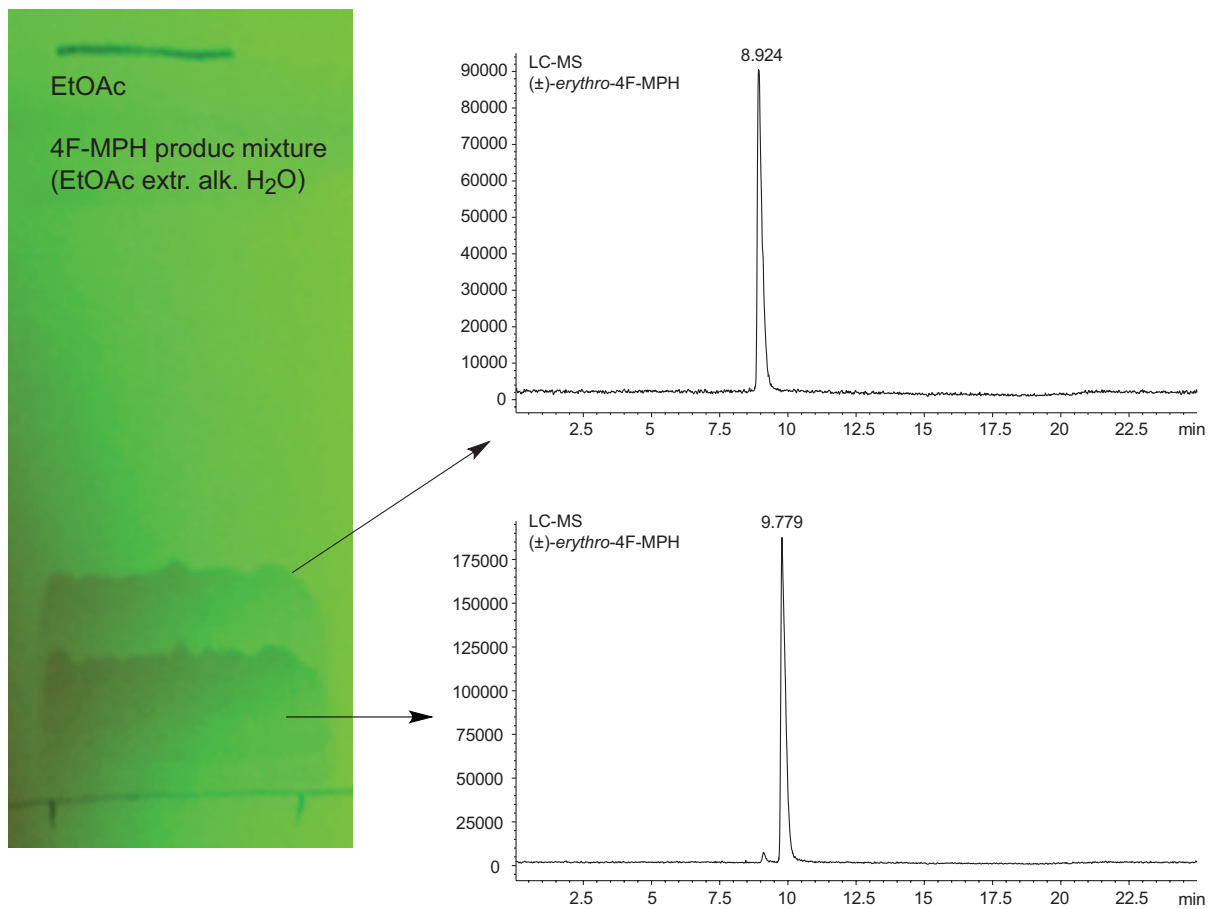
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1. Examples of powdered 4F-MPH and tablet products



2. TLC analysis of vendor sample containing both ( $\pm$ )-*threo*- and ( $\pm$ )-*erythro*-racemates of 4F-MPH, followed by LC-MS analysis of the isolated bands.

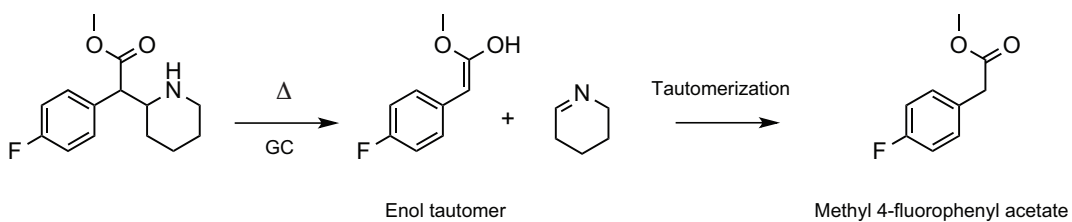
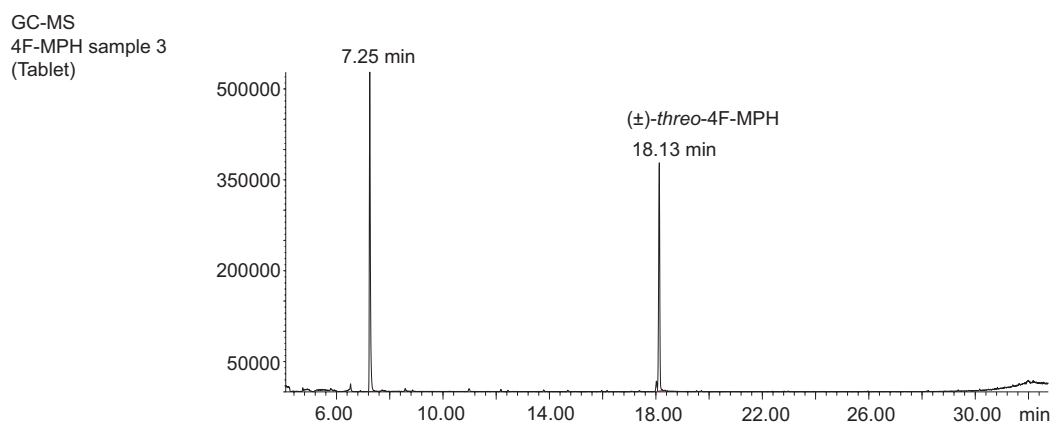
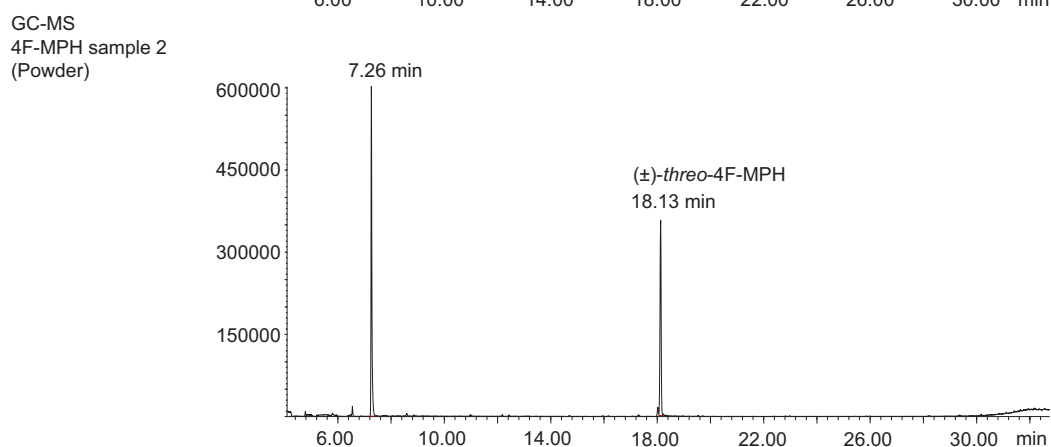
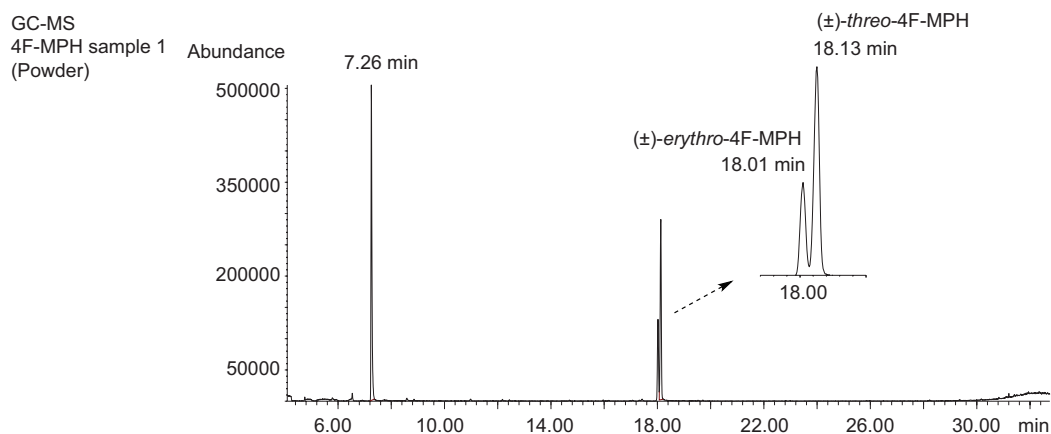


### 3. Preparation of 4F-MPH tablets for GC-MS/LC-MS analysis

For analysis of the 4F-MPH tablets by gas chromatography mass spectrometry (GC-MS), the tablet was crushed using a mortar and pestle and 10 mg was added to 1 mL of methanol. This solution was added to a Corning® Costar® Spin-X® centrifuge tube filter (cellulose acetate membrane, 0.45 µm) (Corning Inc, United States) and centrifuged at 2500 rpm for 3 minutes. Furthermore, 100 µL of this filtered solution was added to 900 µL methanol in a GC vial.

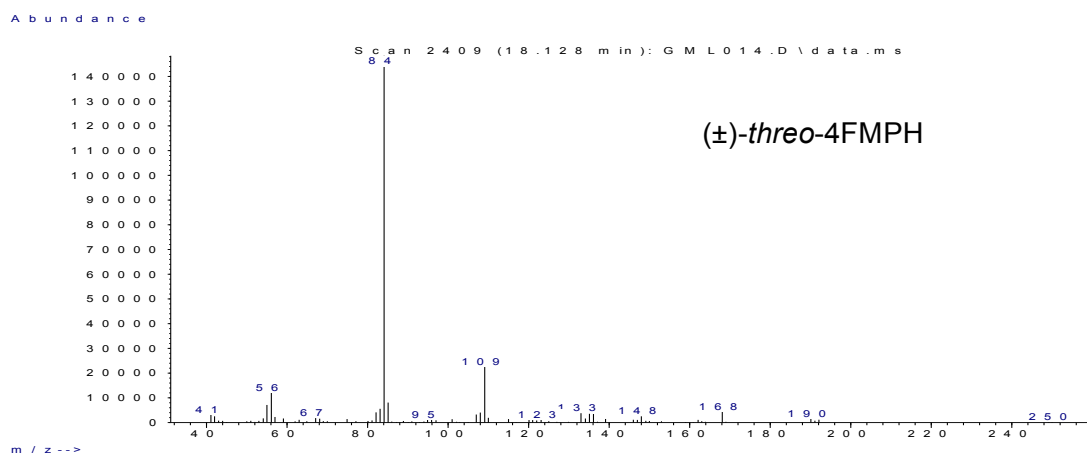
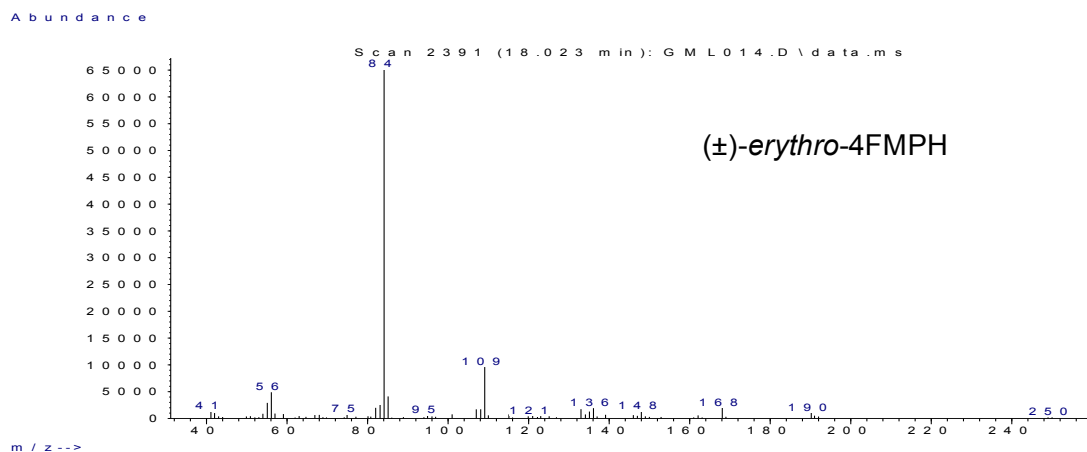
For analysis of the 4F-MPH tablets by liquid chromatography mass spectrometry (LC-MS), the tablet was crushed using a mortar and pestle and 10 mg was added to 1 mL of acetonitrile/water (1:1) with 0.1% formic acid. This solution was added to a Corning® Costar® Spin-X® centrifuge tube filter (cellulose acetate membrane, 0.45 µm) (Corning Inc, United States) and centrifuged at 2500 rpm for 3 minutes. Furthermore, 20 µL of this filtered solution was added to 980 µL acetonitrile/water (1:1) with 0.1% formic acid in a LC vial.

4. GC-induced thermal degradation of 4-fluoromethylphenidate isomers (4F-MPH) to methyl 4-fluorophenyl acetate (7.26 min)

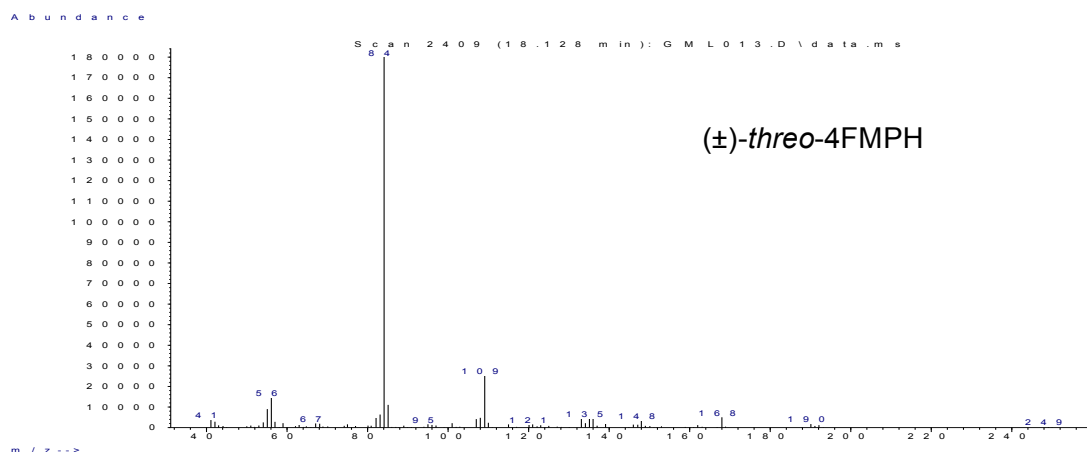


## 5. EI-MS data obtained for vendor samples 1-3

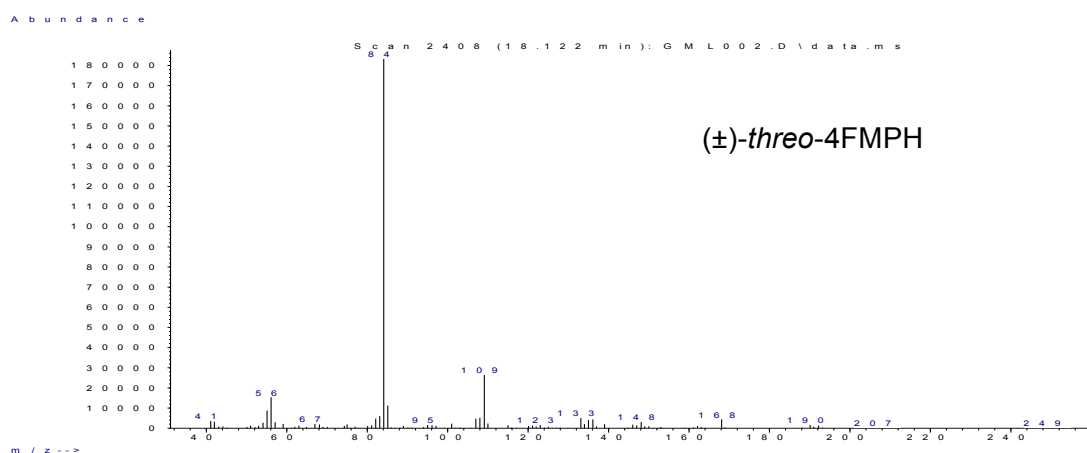
Vendor 1: both ( $\pm$ )-*threo*- and ( $\pm$ )-*erythro* racemates of 4F-MPH (Powder)



Vendor 2: ( $\pm$ )-*threo*-4F-MPH only (Powder)

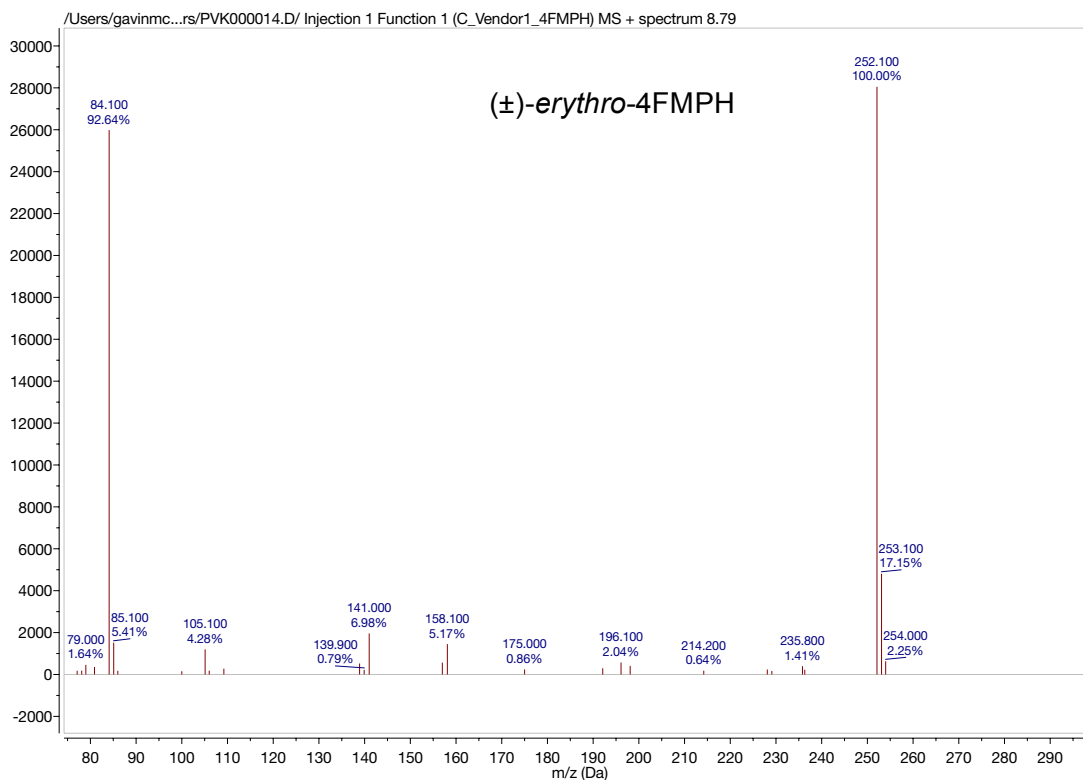


Vendor 3: ( $\pm$ )-*threo*-4F-MPH only (Tablet)

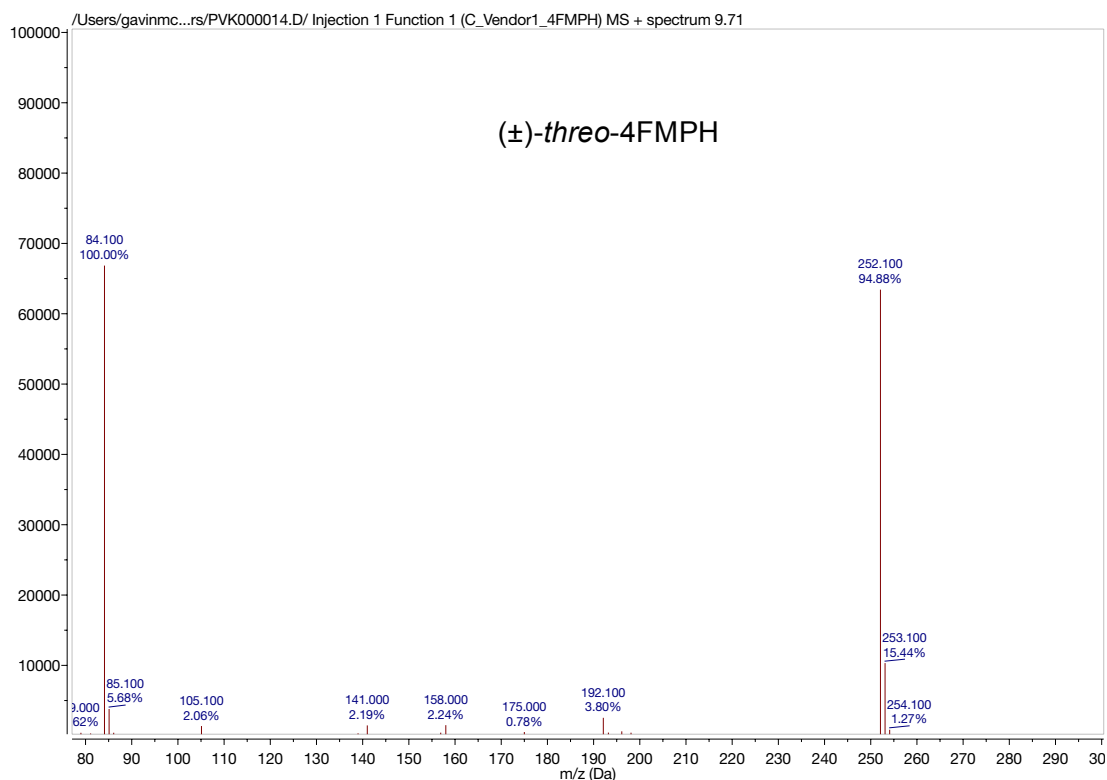


6. LC-ESI-MS of vendor samples 1-3

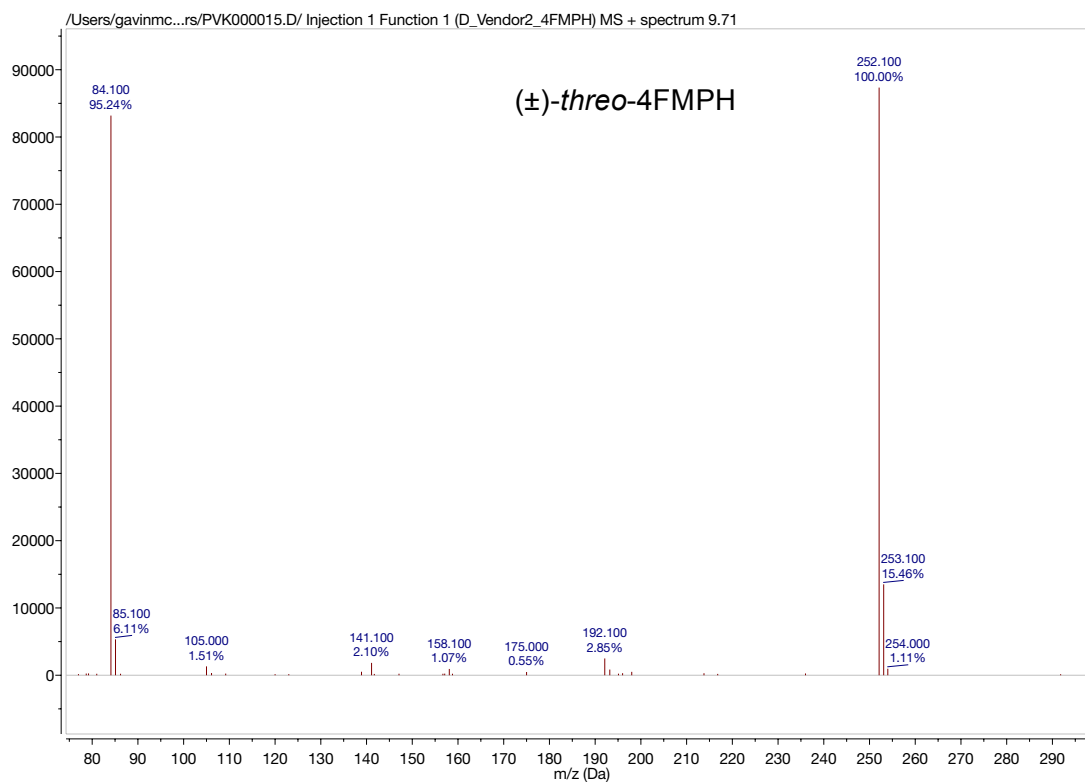
Vendor 1: both ( $\pm$ )-*threo*- and ( $\pm$ )-*erythro* racemates of 4F-MPH (Powder)



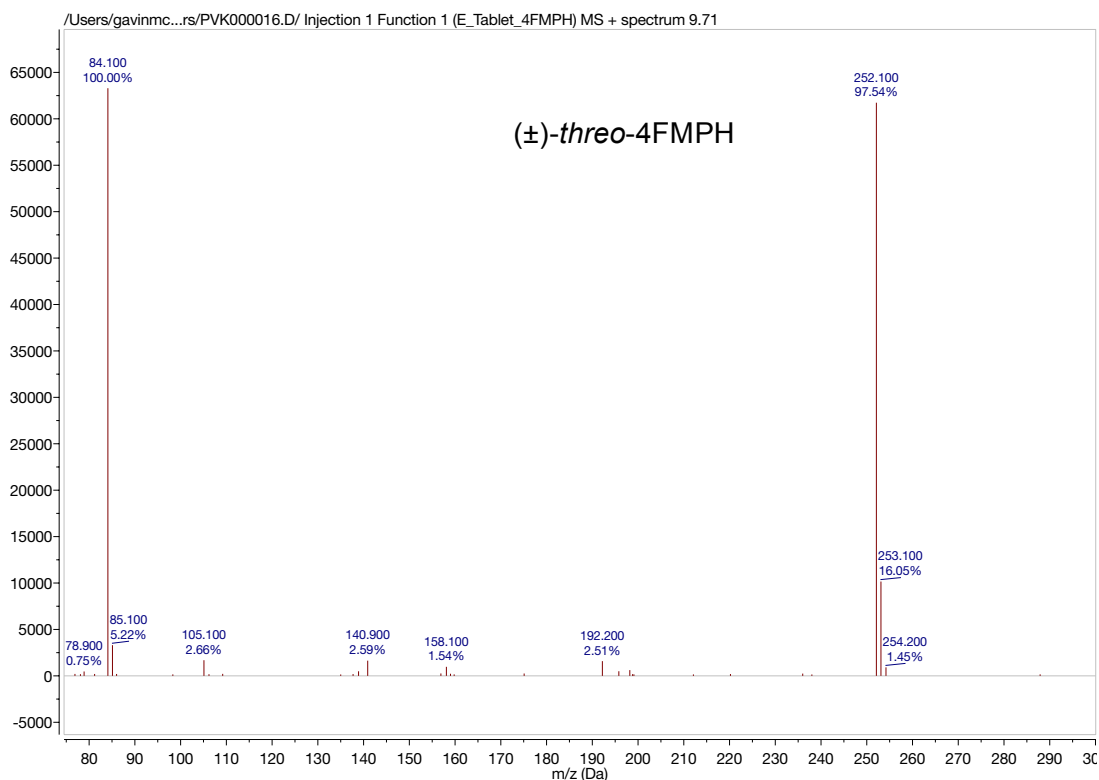




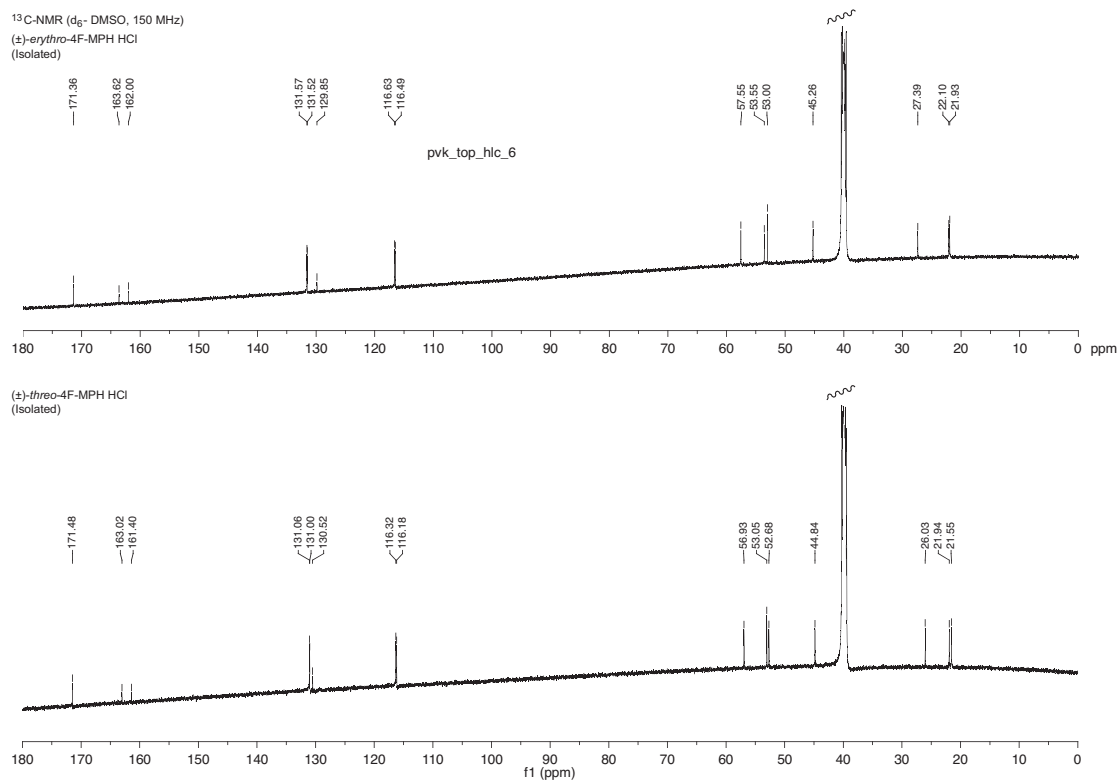
Vendor 2: (±)-*threo*-racemate of 4F-MPH (Powder)



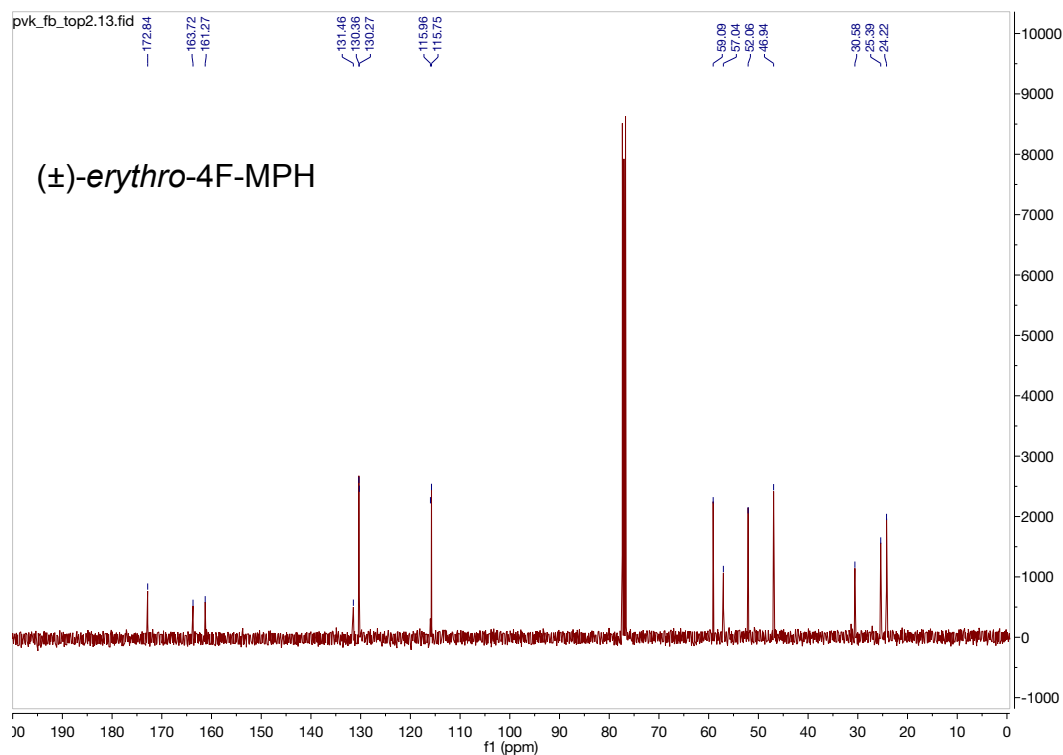
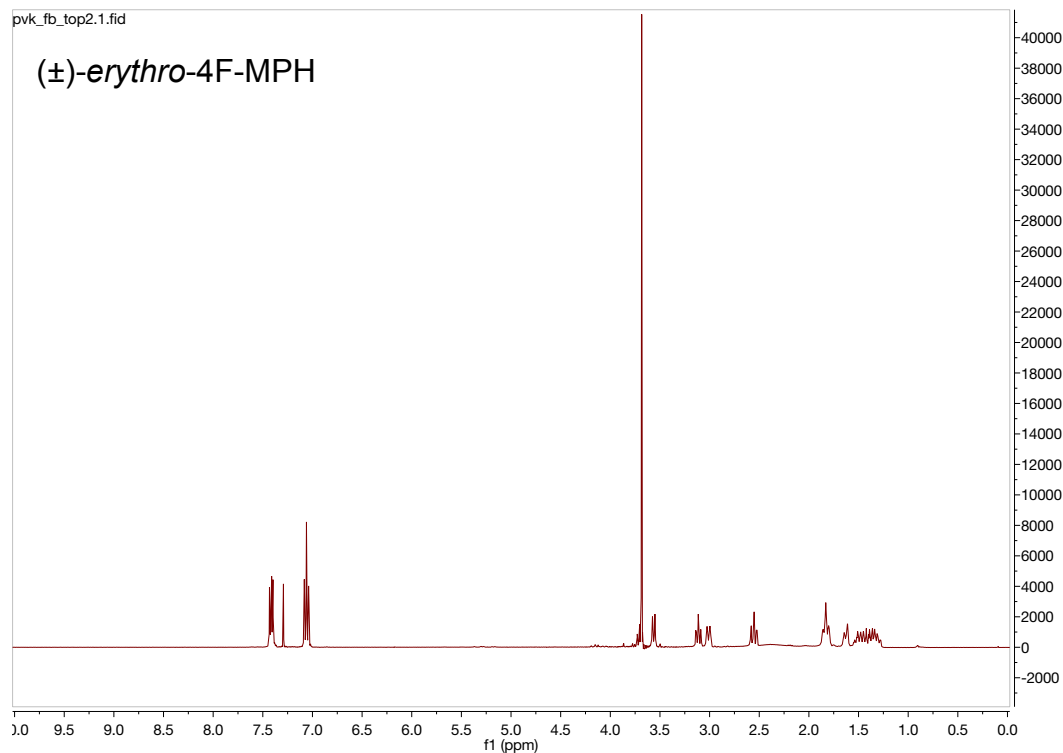
Vendor 3: ( $\pm$ )-*threo*-racemate of 4F-MPH (Tablet)

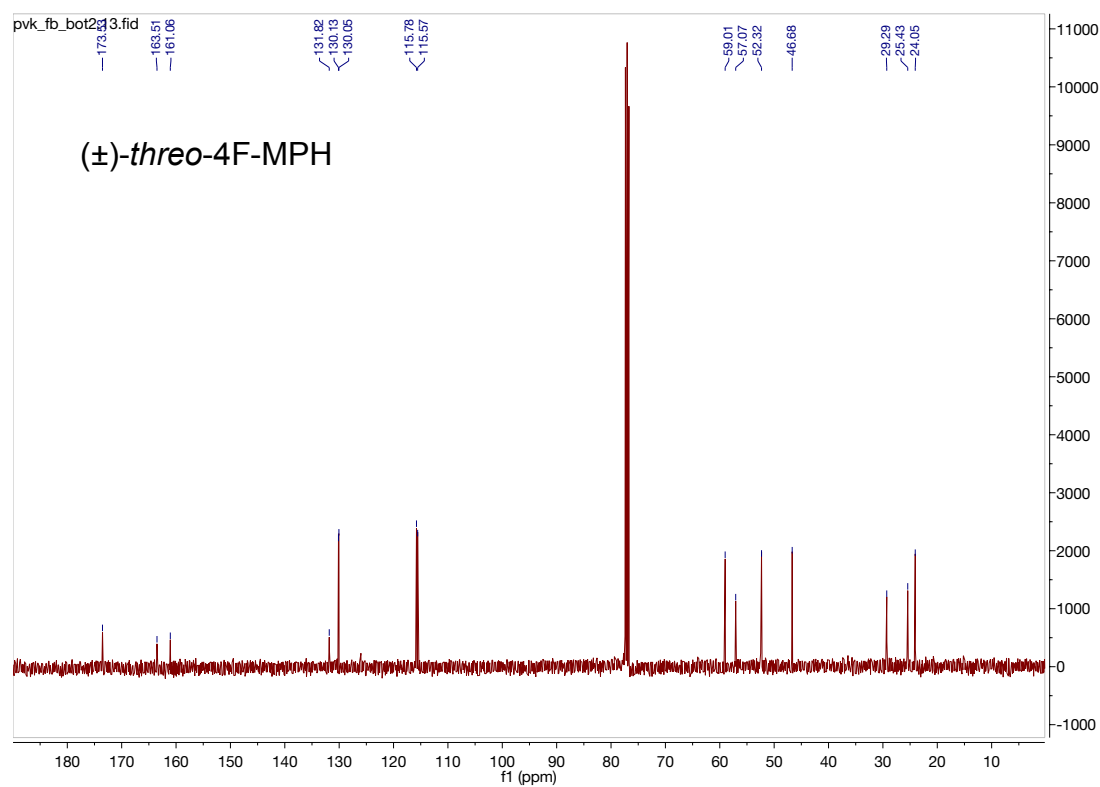
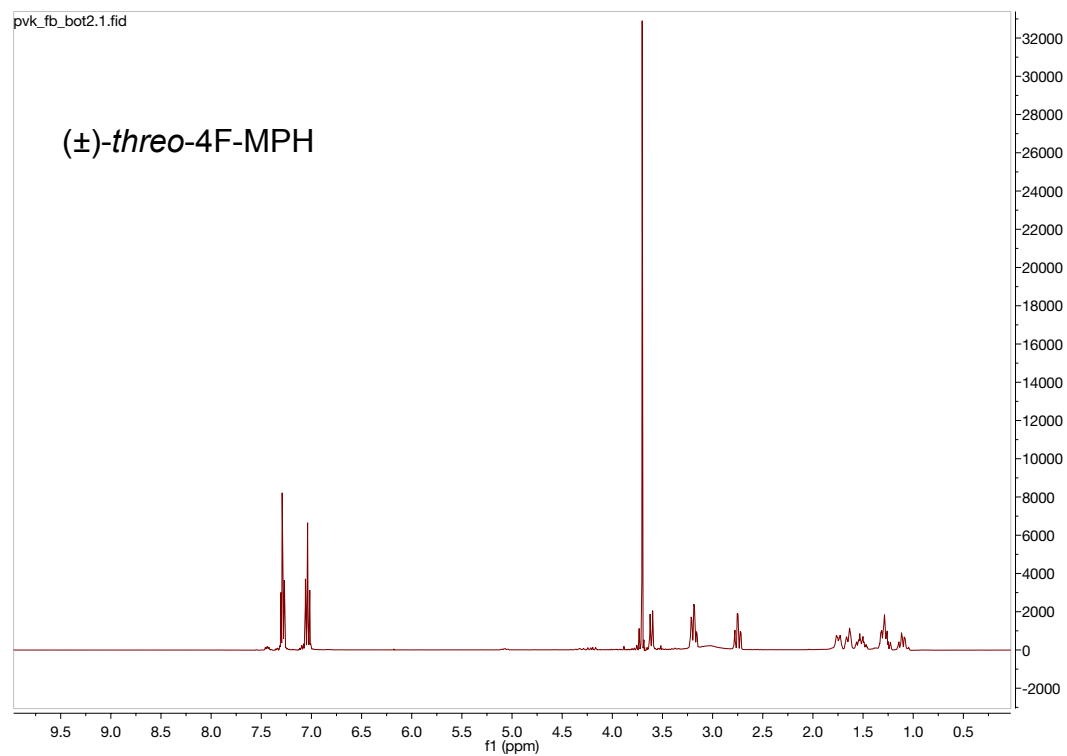


7.  $^{13}\text{C}$  NMR spectra obtained for the isolated ( $\pm$ )-*erythro*- and ( $\pm$ )-*threo*-racemates of 4F-MPH (HCl salt in DMSO- $\text{d}_6$ ).

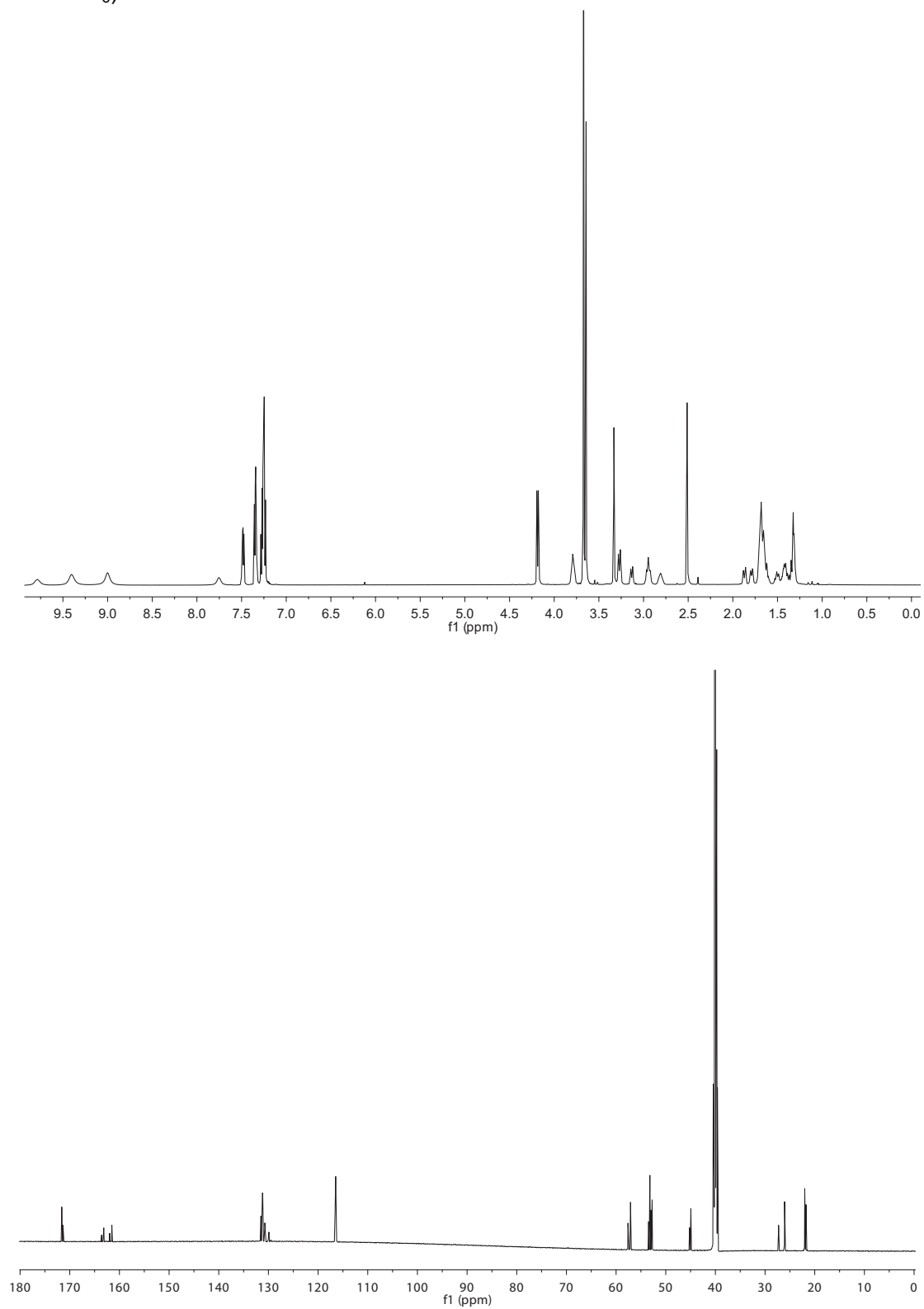


8.  $^1\text{H}$  and  $^{13}\text{C}$  NMR spectra obtained for the isolated ( $\pm$ )-*erythro*- and ( $\pm$ )-*threo* racemates of 4F-MPH (Free Base in  $\text{CDCl}_3$ )

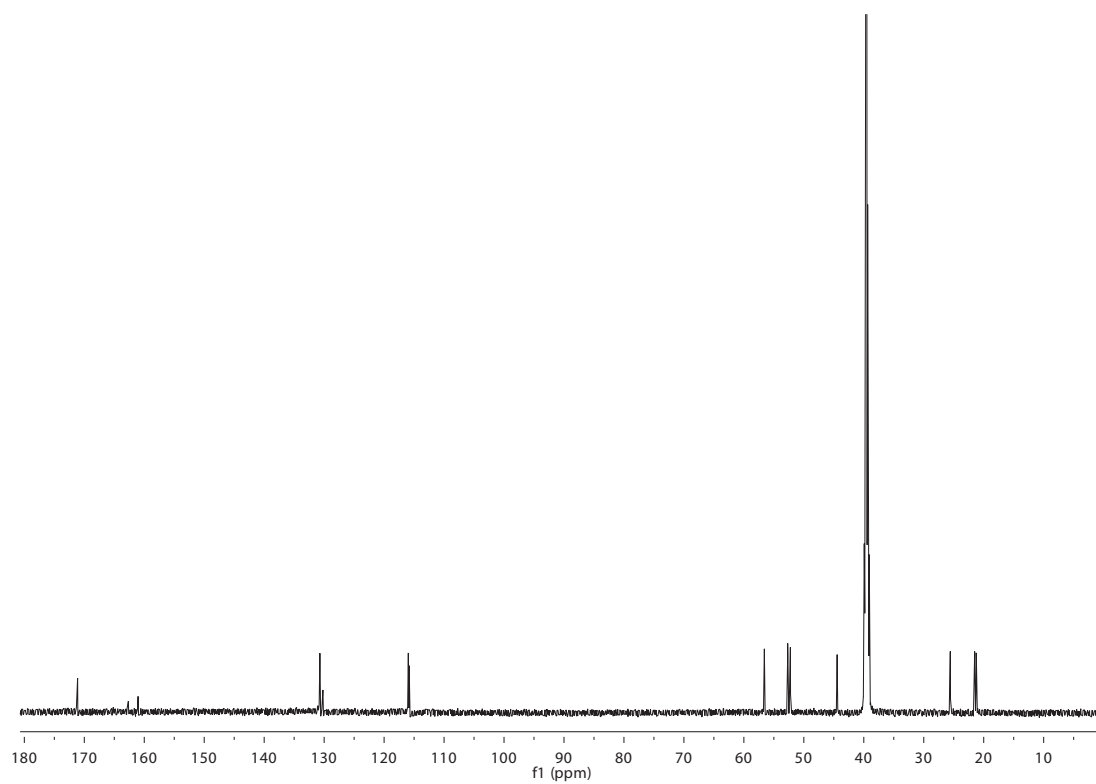
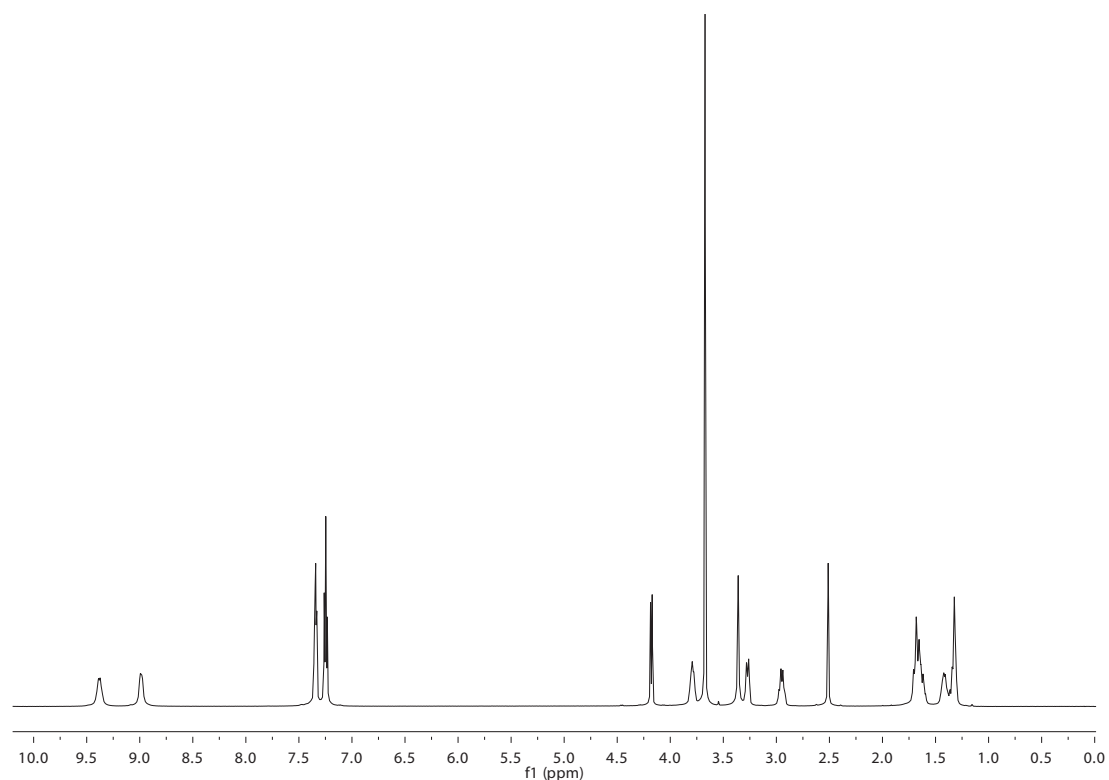




9.  $^{13}\text{C}$  and  $^1\text{H}$  NMR spectra obtained for one of the powdered 4F-MPH products identified as mixed ( $\pm$ )-*erythro*- and ( $\pm$ )-*threo*- racemates (HCl salt in DMSO- $\text{d}_6$ )



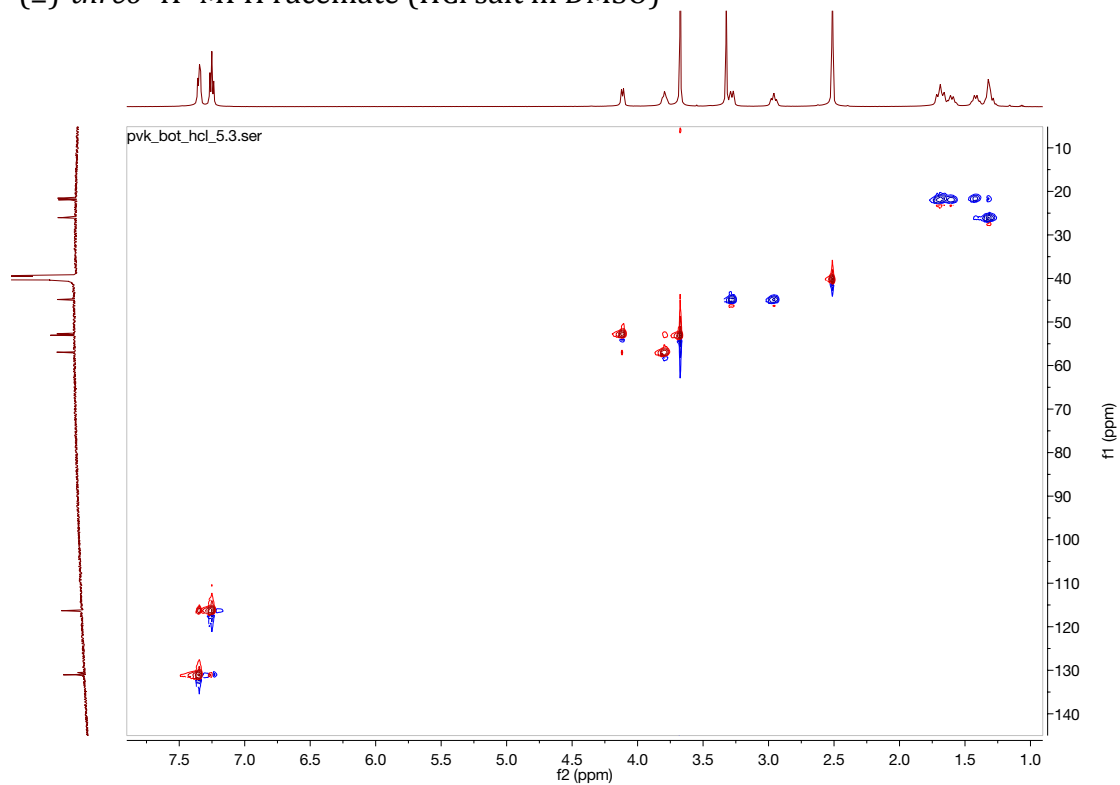
10.  $^{13}\text{C}$  and  $^1\text{H}$  NMR spectra obtained for one of the powdered 4F-MPH products identified as the ( $\pm$ )-*threo*-racemate (HCl salt in  $\text{DMSO-d}_6$ )



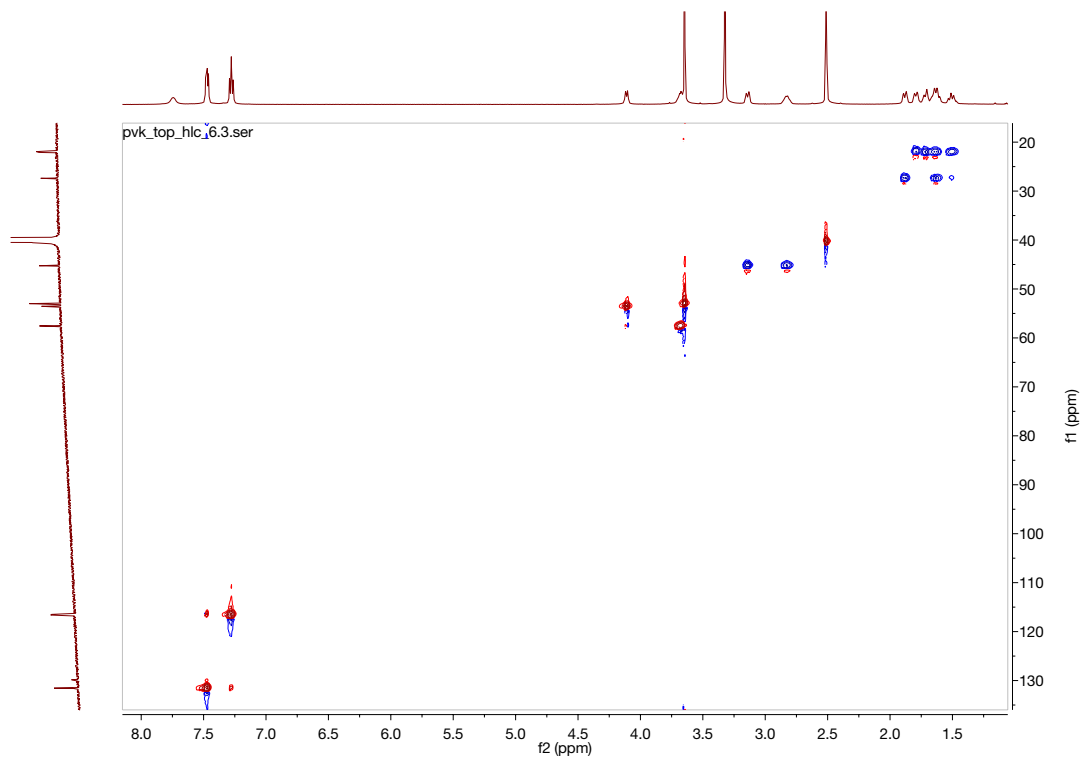
## 11. Two-dimensional NMR experiments

- Heteronuclear single quantum coherence spectroscopy (HSQC)

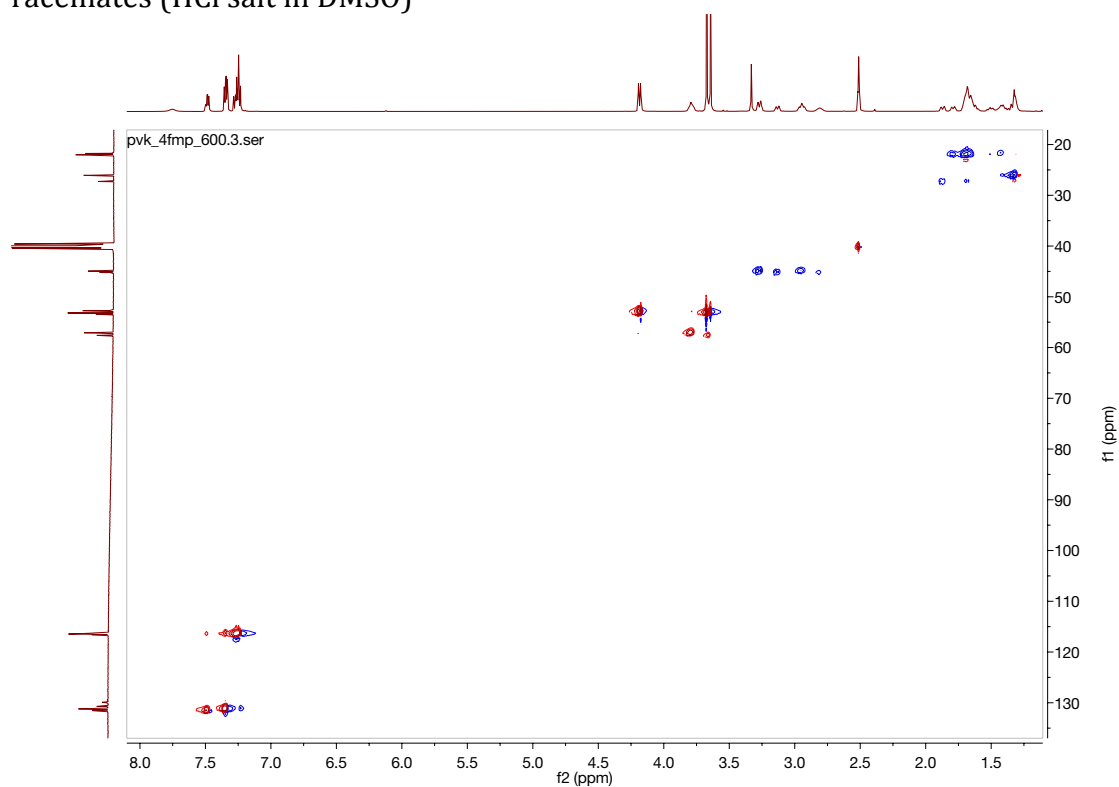
(±)-*threo*-4F-MPH racemate (HCl salt in DMSO)



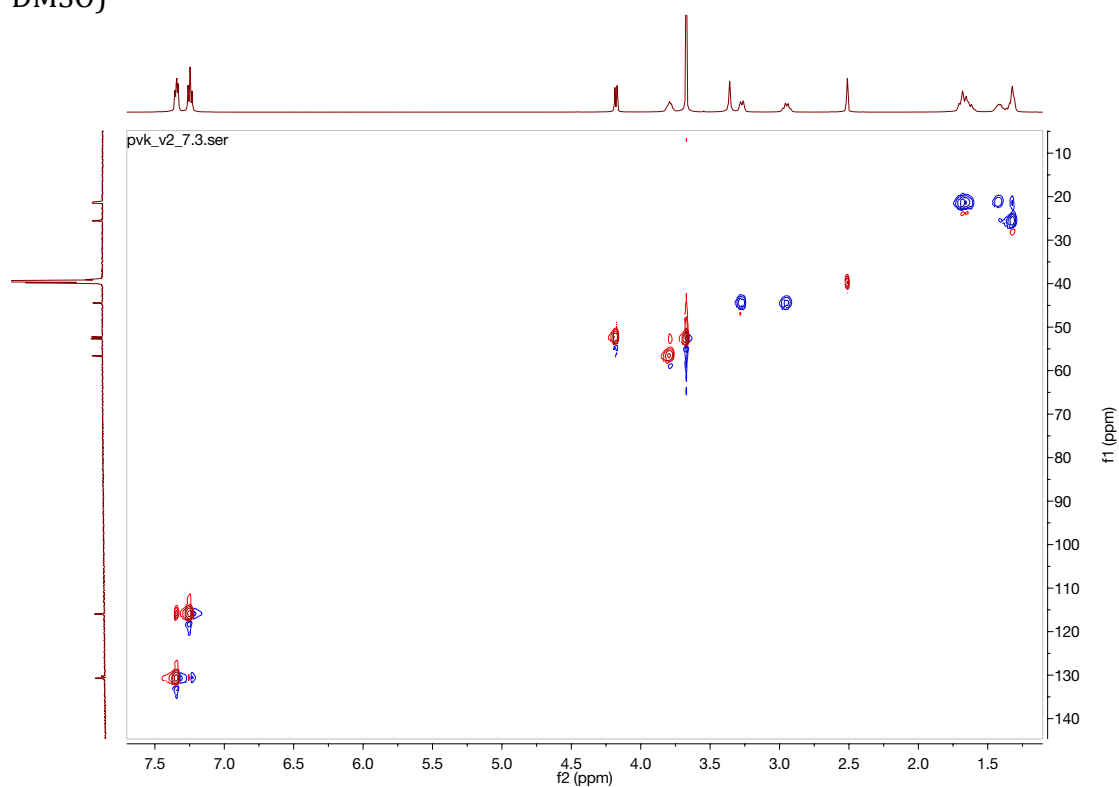
(±)-*erythro*-4F-MPH racemate (HCl salt in DMSO)



4F-MPH vendor product identified as ( $\pm$ )-*threo*- and ( $\pm$ )-*erythro*-4F-MPH racemates (HCl salt in DMSO)



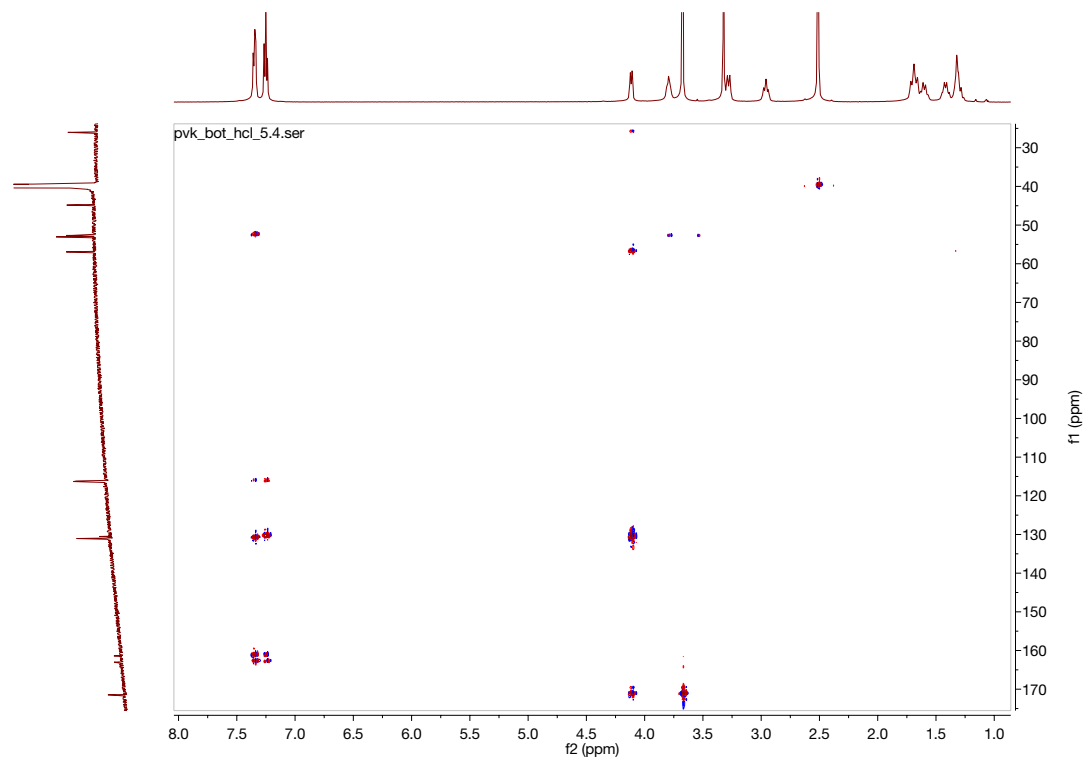
4F-MPH vendor product identified as ( $\pm$ )-*threo*-4F-MPH racemate (HCl salt in DMSO)



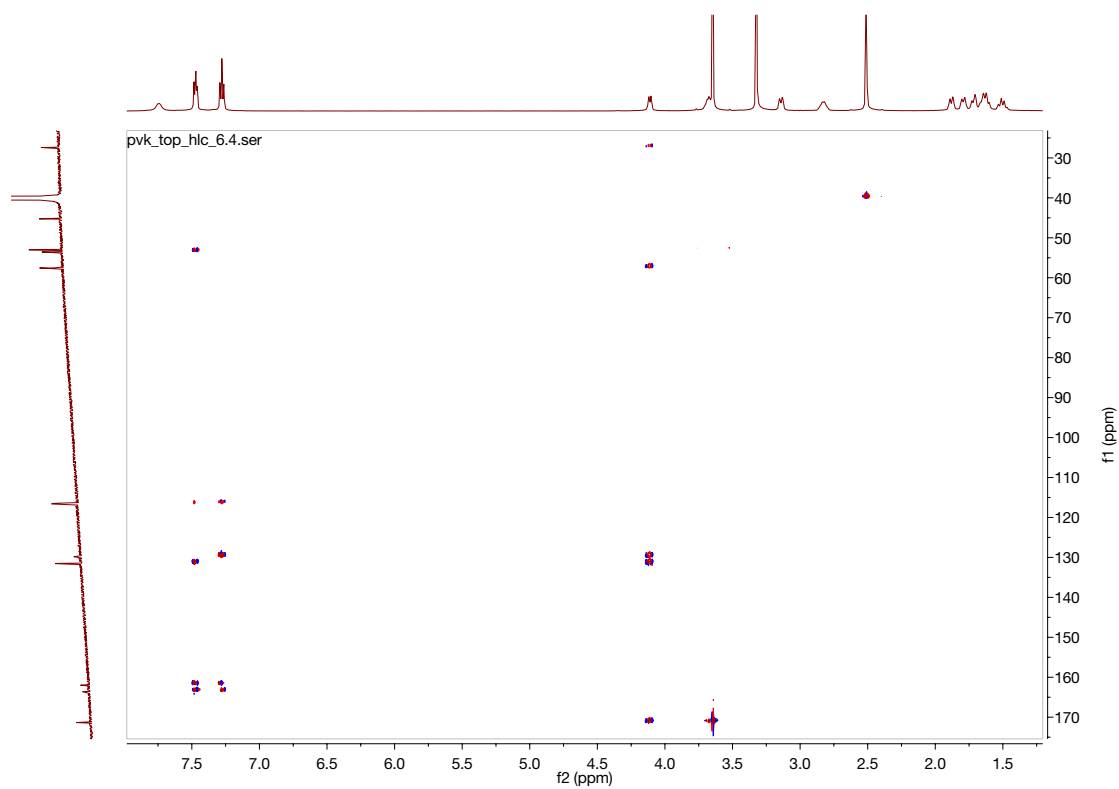


## Heteronuclear Multiple Bond Correlation (HMBC)

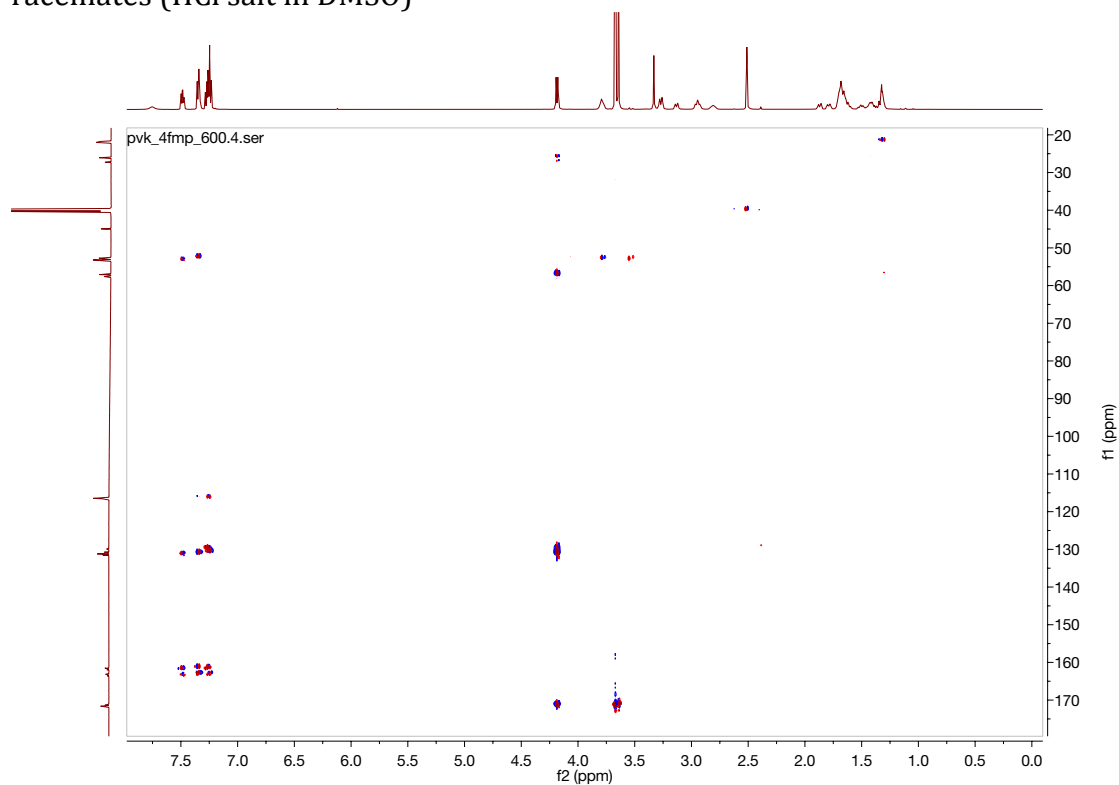
(±)-*threo*-4F-MPH racemate (HCl salt in DMSO)



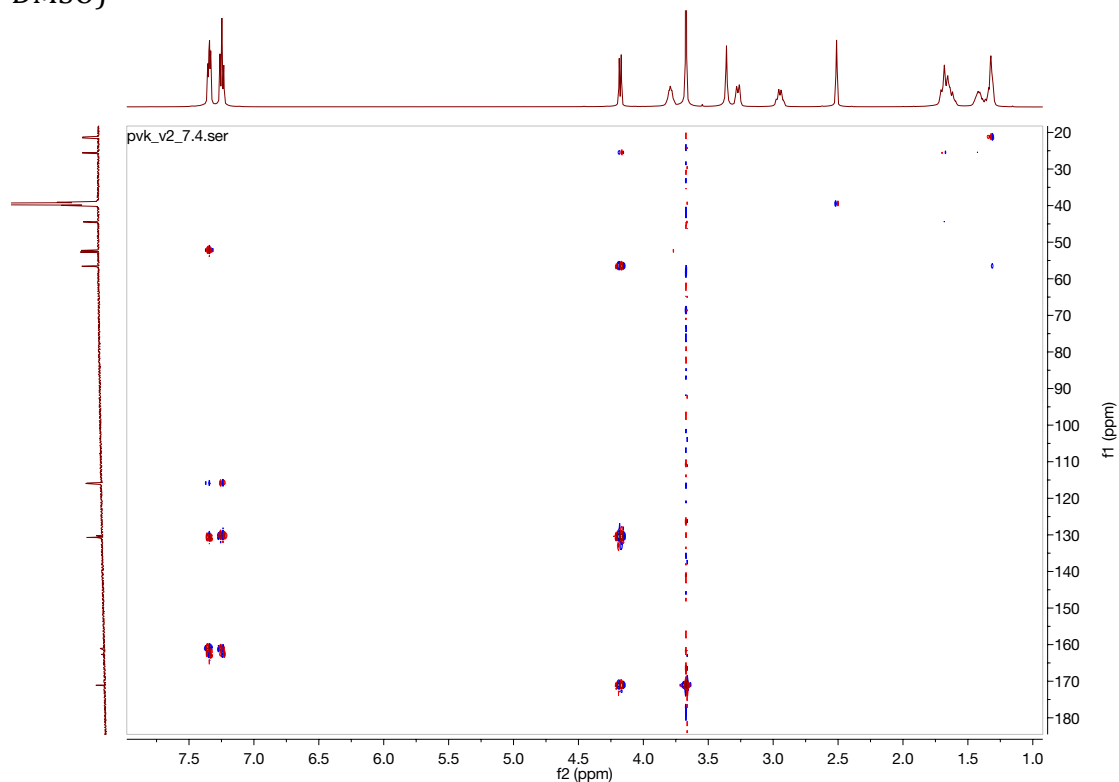
(±)-*erythro*-4F-MPH racemate (HCl salt in DMSO)



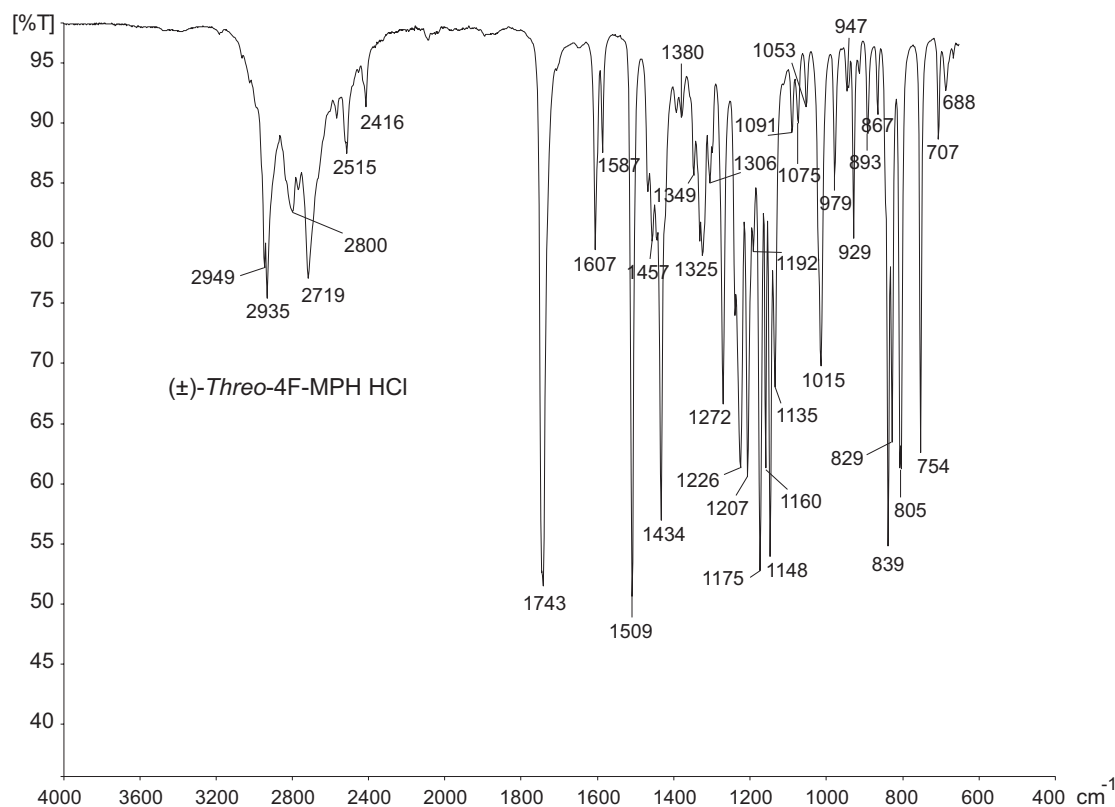
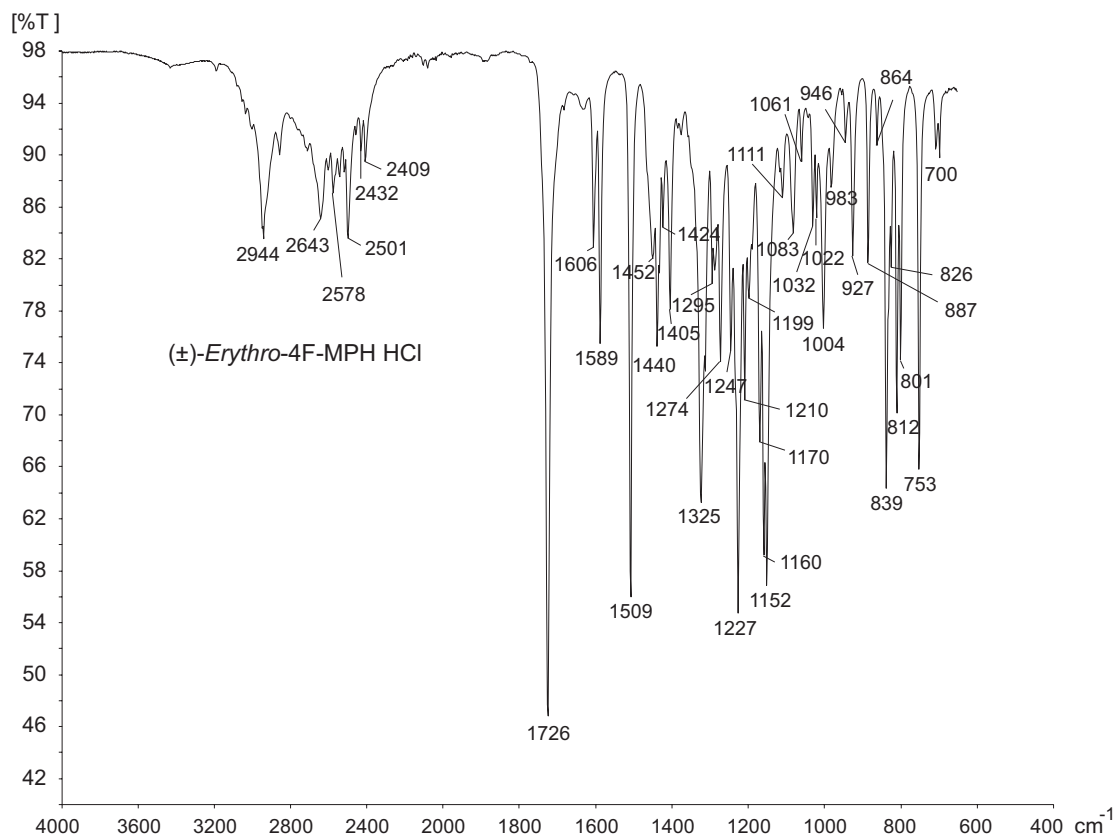
4F-MPH vendor product identified as ( $\pm$ )-*threo*- and ( $\pm$ )-*erythro*-4F-MPH racemates (HCl salt in DMSO)

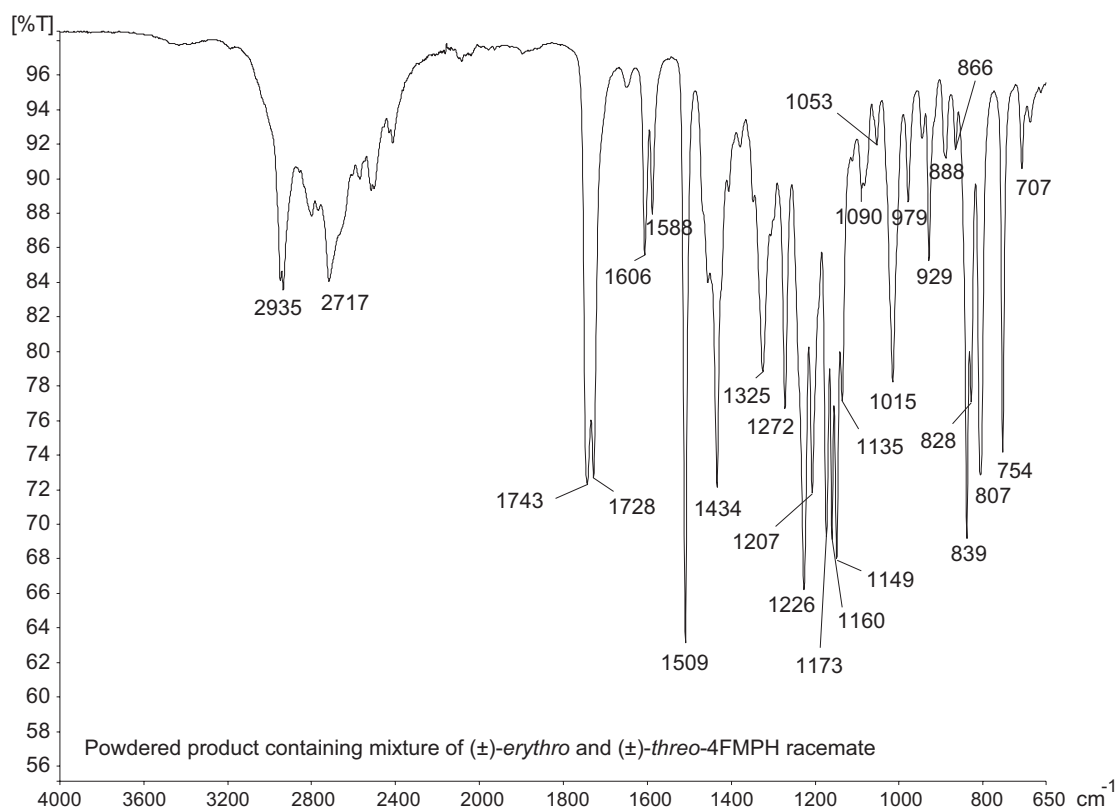
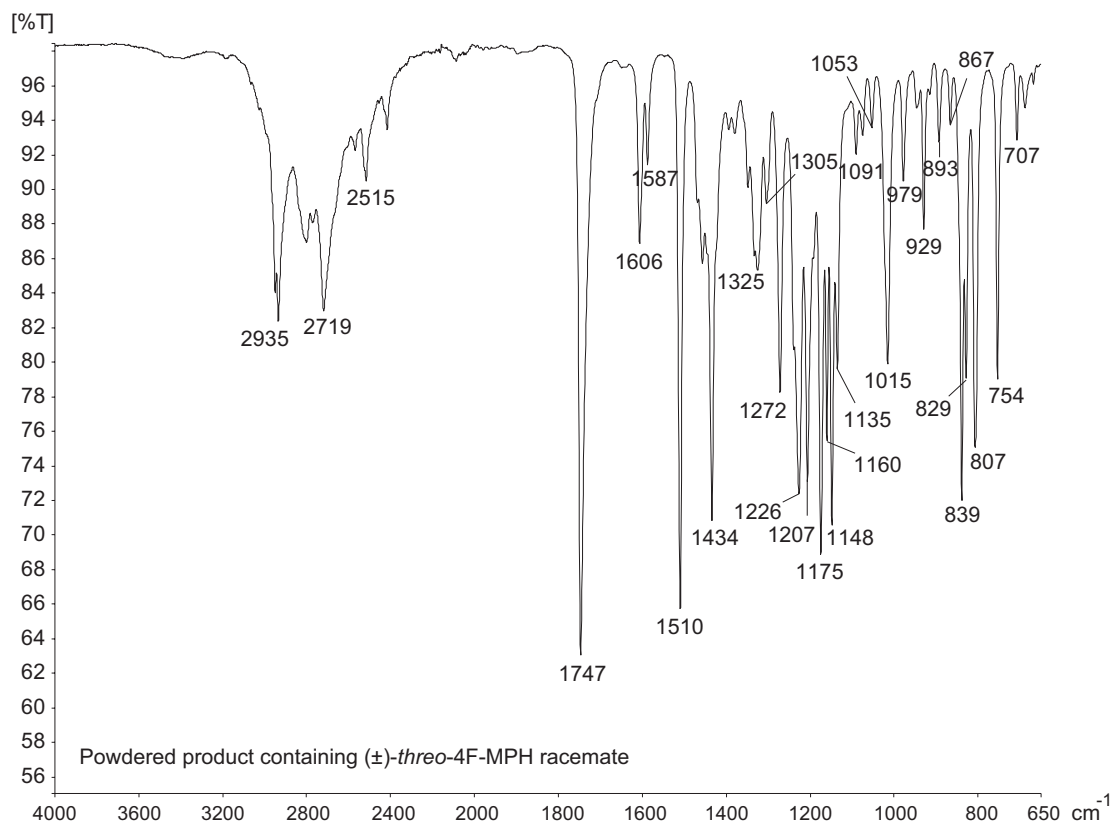


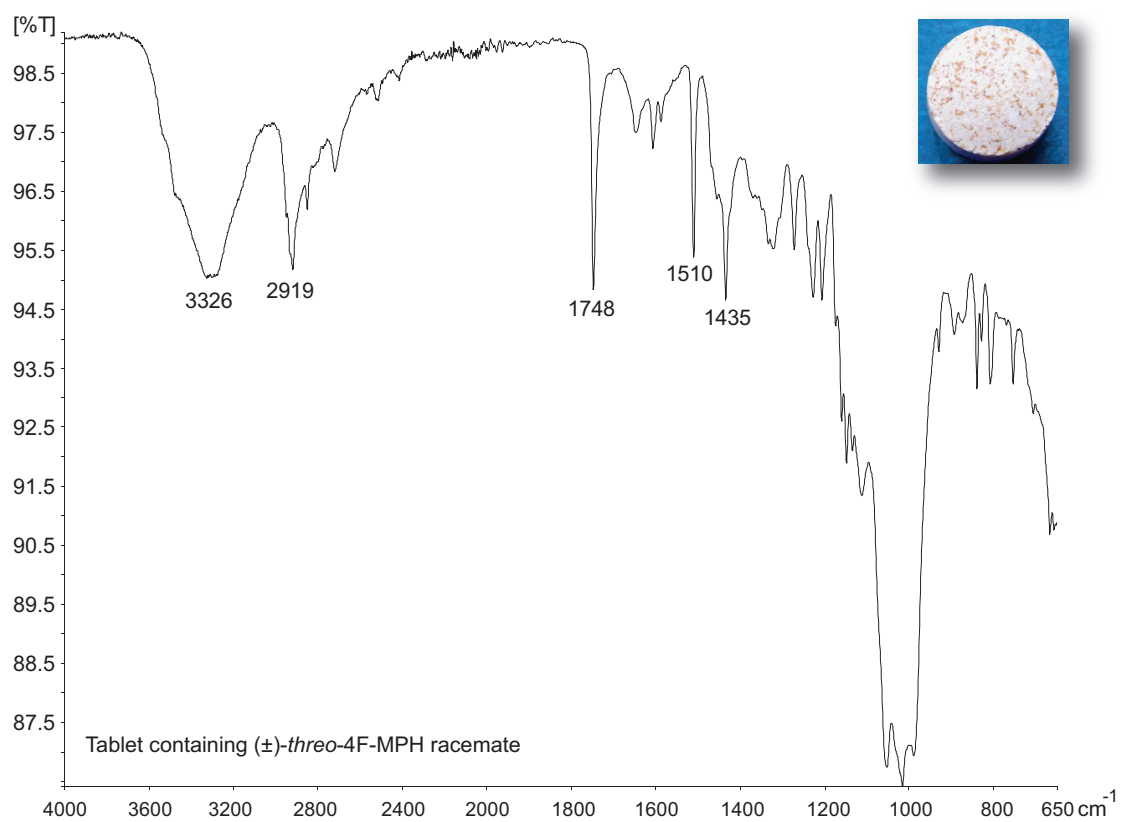
4F-MPH vendor product identified as ( $\pm$ )-*threo*-4F-MPH racemate (HCl salt in DMSO)



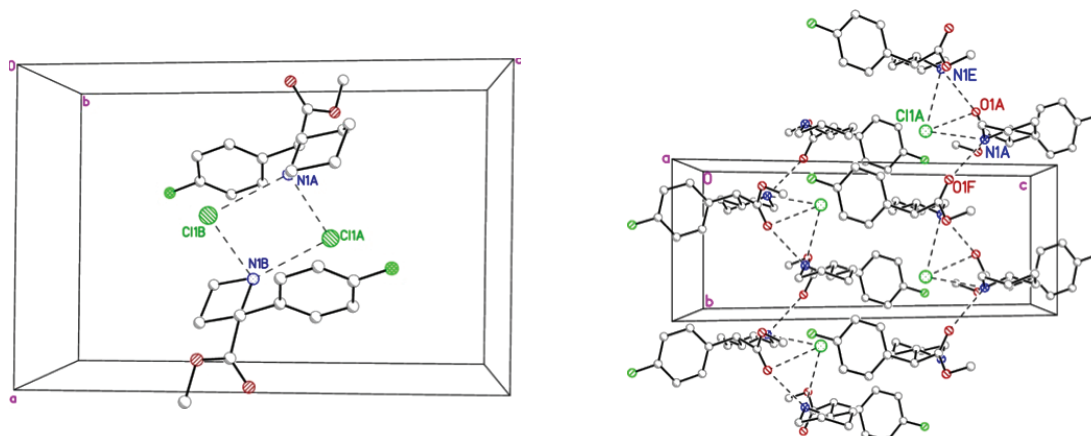
12. Infrared Spectroscopy - IR spectra for isolated ( $\pm$ )-*threo*-racemate, isolated ( $\pm$ )-*erythro*-racemate, and vendor samples 1-3







### 13. X-ray crystallography supporting information



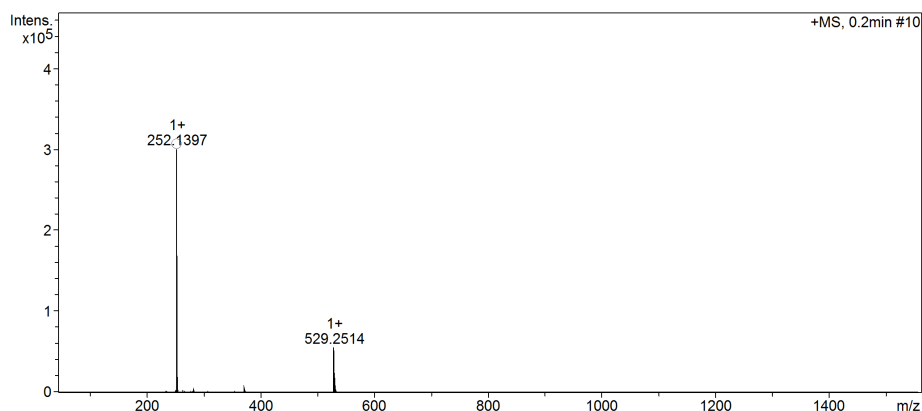
Left: *eythro* salt hydrogen bonded dimer with hydrogen atoms omitted for clarity. The cell is shown. Only atoms involved in H-bonding interactions are labelled. Right: *threo* salt hydrogen bonding network extending parallel to the *b*-axis

14. High resolution mass spectrometry data obtained for isolated ( $\pm$ )-*threo*-racemate, isolated ( $\pm$ )-*erythro*-racemate, and vendor samples

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## Bruker Open Access LC-MS - Formula Identification Report

Sample-ID		Station	
Submitter		Supervisor	
Analysis Name	GML_Erythro_4FMPH_RA6_01_4048.d	Acquisition Date	06/07/2016 15:04:11
Sample Description			



Meas. m/z	#	Ion Formula	m/z	err [mDa]	err [ppm]	rdb	N-Rule	e <sup>-</sup>	Conf	mSigma
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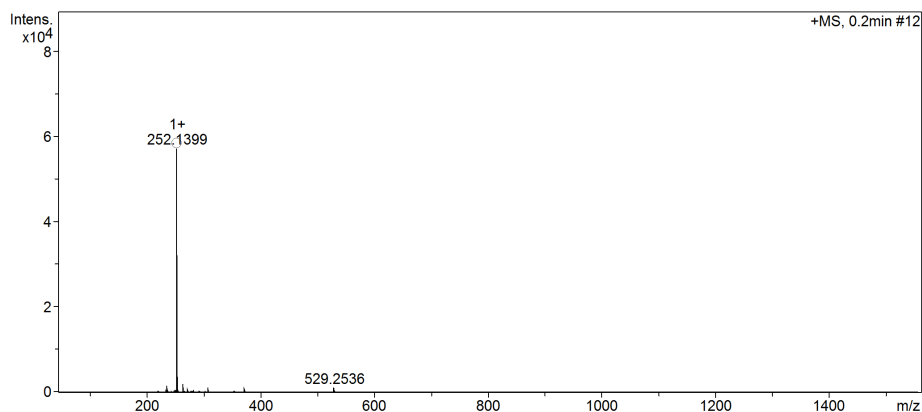
## SmartFormula Settings

Low value of mSigma indicates good isotopic pattern match



Trinity College Dublin  
Bruker Open Access LC-MS - Formula Identification Report

Sample-ID		Station	
Submitter		Supervisor	
Analysis Name	GML_Threo_4FMPH_RA7_01_4049.d	Acquisition Date	06/07/2016 15:07:29
Sample Description			



Meas. m/z	#	Ion Formula	m/z	err [mDa]	err [ppm]	rdb	N-Rule	e <sup>-</sup> Conf	mSigma
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SmartFormula Settings

Low value of mSigma indicates good isotopic pattern match

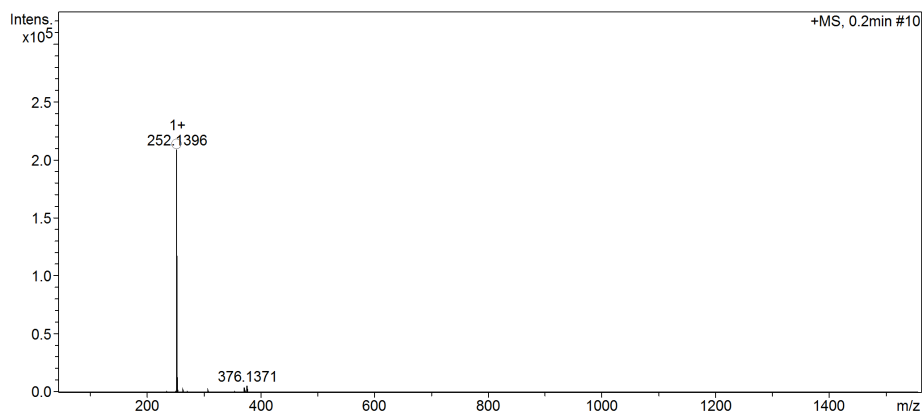




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Bruker Open Access LC-MS - Formula Identification Report

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Submitter		Supervisor	
Analysis Name	GML_Vendor_1_RA8_01_4050.d	Acquisition Date	06/07/2016 15:10:46
Sample Description			



Meas. m/z	#	Ion Formula	m/z	err [mDa]	err [ppm]	rdb	N-Rule	e <sup>-</sup> Conf	mSigma
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SmartFormula Settings

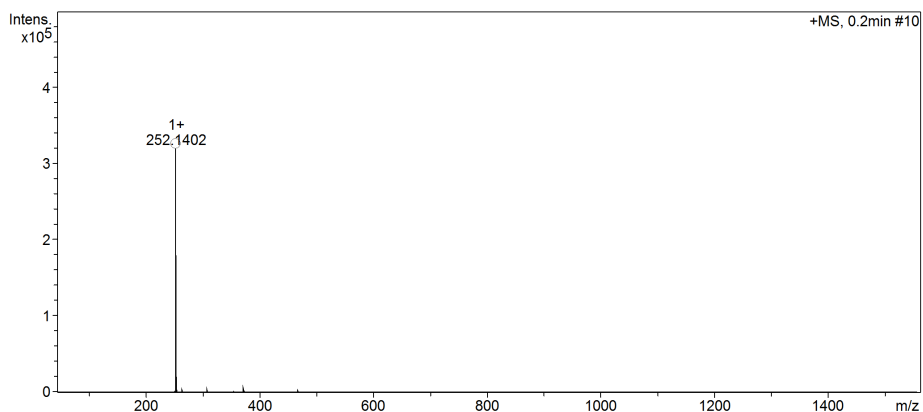
Low value of mSigma indicates good isotopic pattern match



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Bruker Open Access LC-MS - Formula Identification Report

Sample-ID		Station	
Submitter		Supervisor	
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Sample Description			



Meas. m/z	#	Ion Formula	m/z	err [mDa]	err [ppm]	rdb	N-Rule	e <sup>-</sup> Conf	mSigma
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SmartFormula Settings

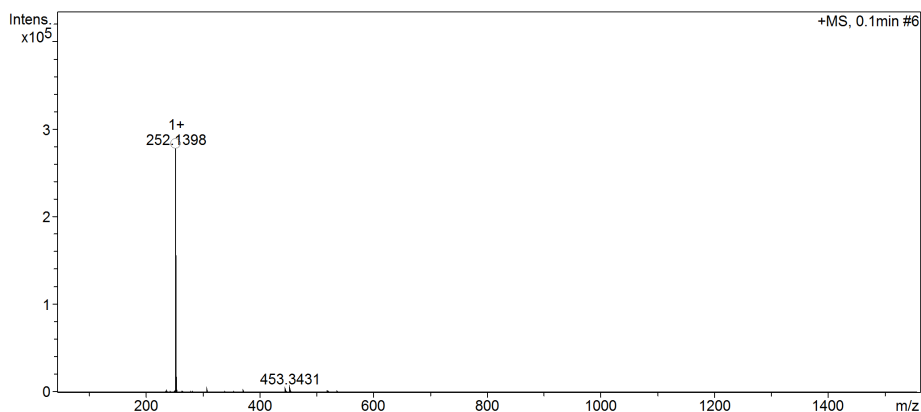
Low value of mSigma indicates good isotopic pattern match



Trinity College Dublin

Bruker Open Access LC-MS - Formula Identification Report

Sample-ID		Station	
Submitter		Supervisor	
Analysis Name	GML_Vendor_3_Tablet_RB2_01_4052.d	Acquisition Date	06/07/2016 15:17:21
Sample Description			



Meas. m/z	#	Ion Formula	m/z	err [mDa]	err [ppm]	rdb	N-Rule	e <sup>-</sup> Conf	mSigma
252.139850	1	C <sub>14</sub> H <sub>19</sub> FNO <sub>2</sub>	252.139433	-0.4	-1.7	5.5	ok	even	0.7

SmartFormula Settings

Low value of mSigma indicates good isotopic pattern match