Electronic Supplementary Information

N-Methylamide-Structured SB366791 Derivatives with High TRPV1 Antagonistic Activity: Toward PET Radiotracers to Visualize TRPV1

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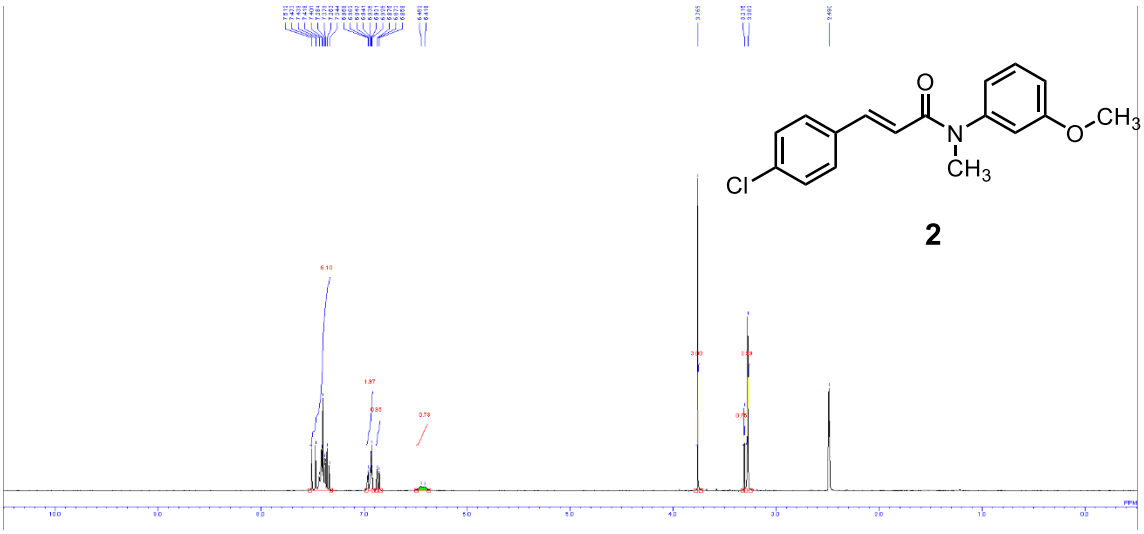
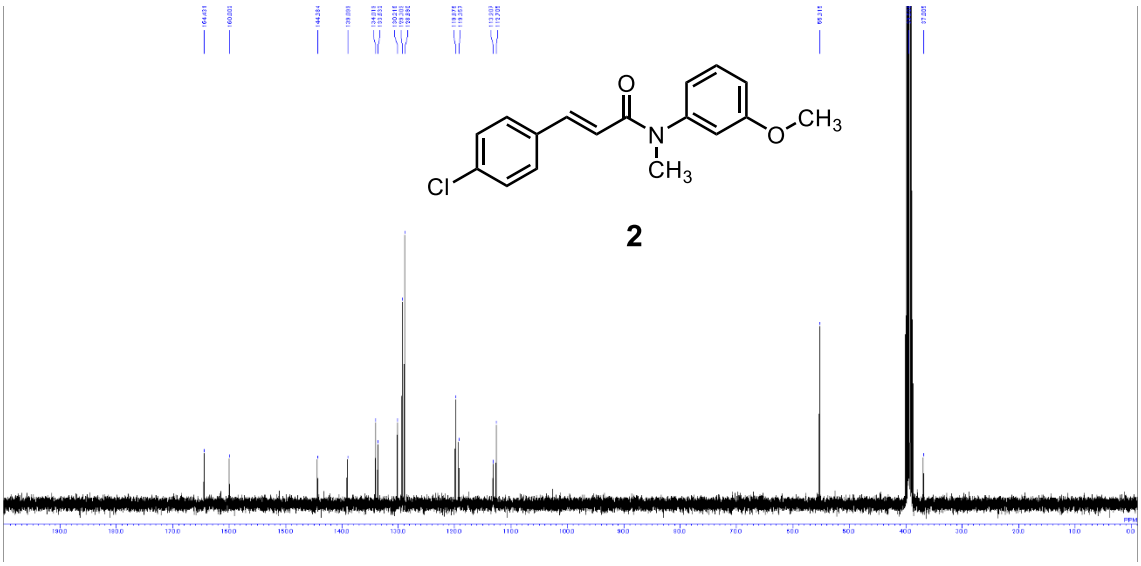
Chapter 1: Compound characterization data

Characterization data for compound **2**

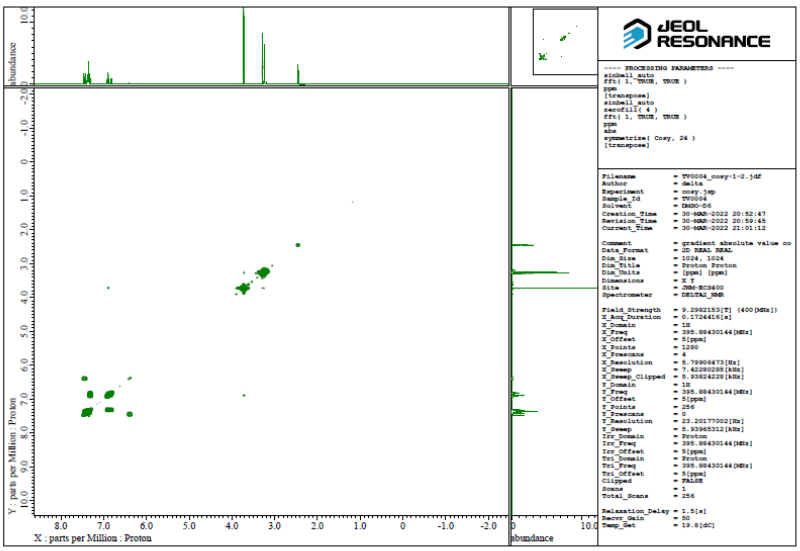
^{1}H NMR (400 MHz, DMSO-d_6) spectrum

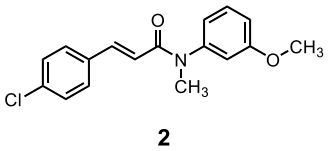


^{13}C{^{1}H} NMR (100 MHz, DMSO-d_6) spectrum



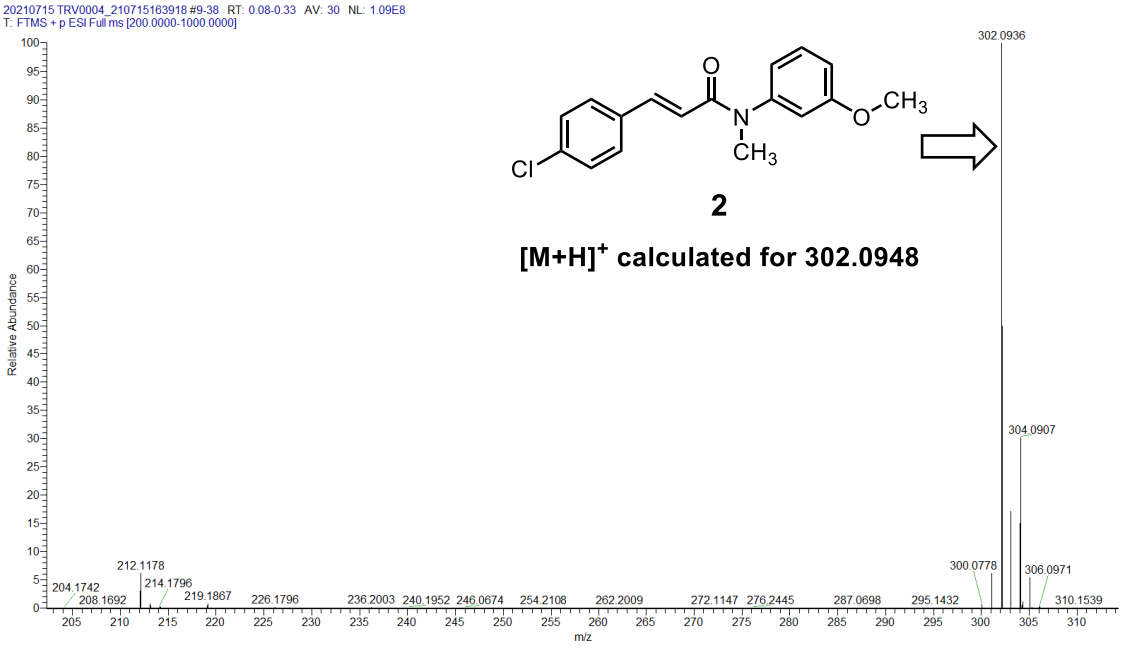
^{1}H–^{1}H COSY (DMSO-d_6) spectrum





2

High-resolution mass spectrum

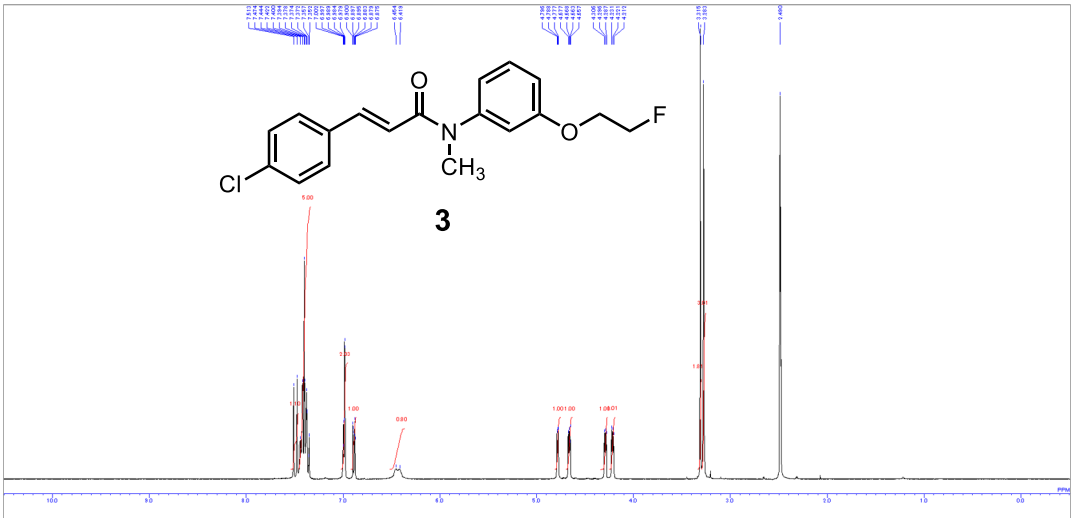


2

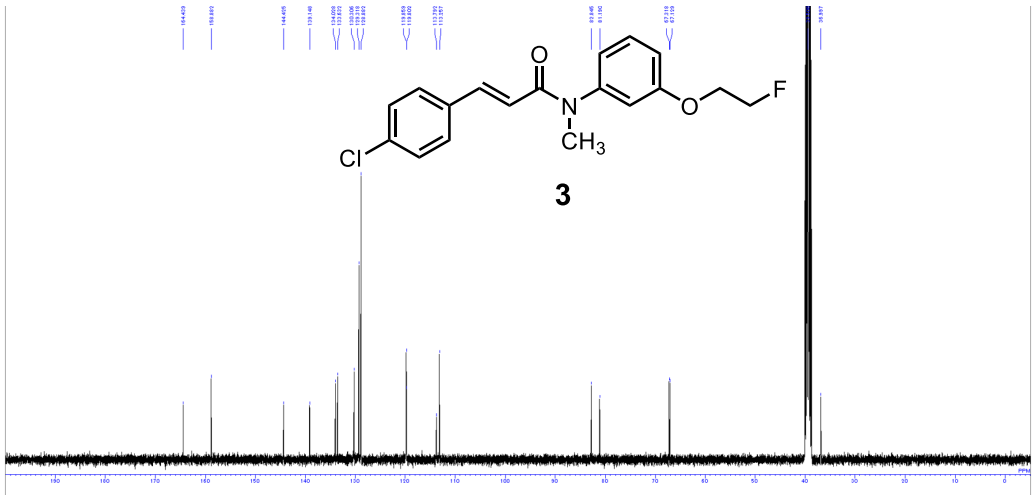
$[M+H]^+$ calculated for 302.0948

Characterization data for compound **3**

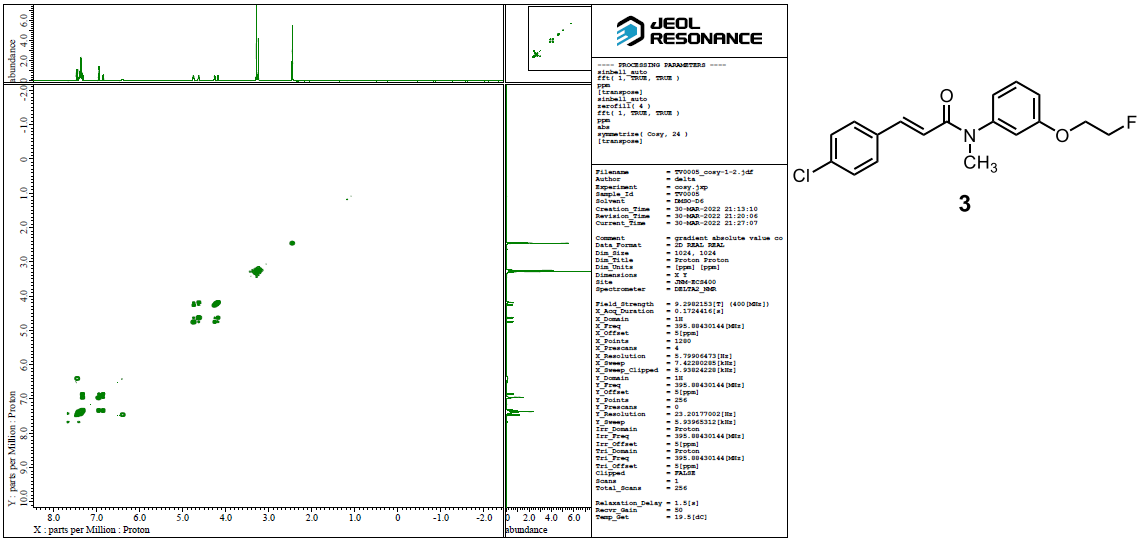
^{1}H NMR (400 MHz, DMSO-d_6) spectrum



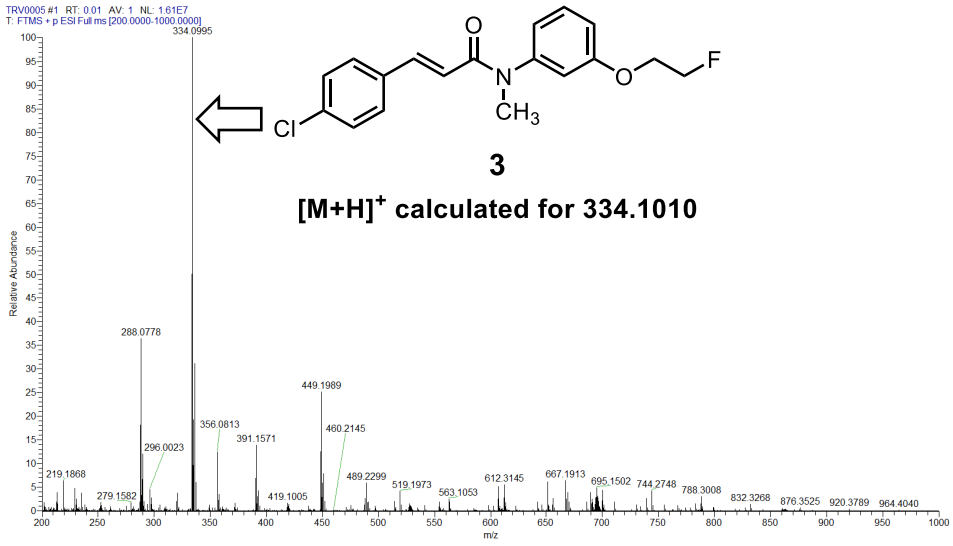
^{13}C{^{1}H} NMR (100 MHz, DMSO-d_6) spectrum



^{1}H–^{1}H COSY (DMSO-d_6) spectrum

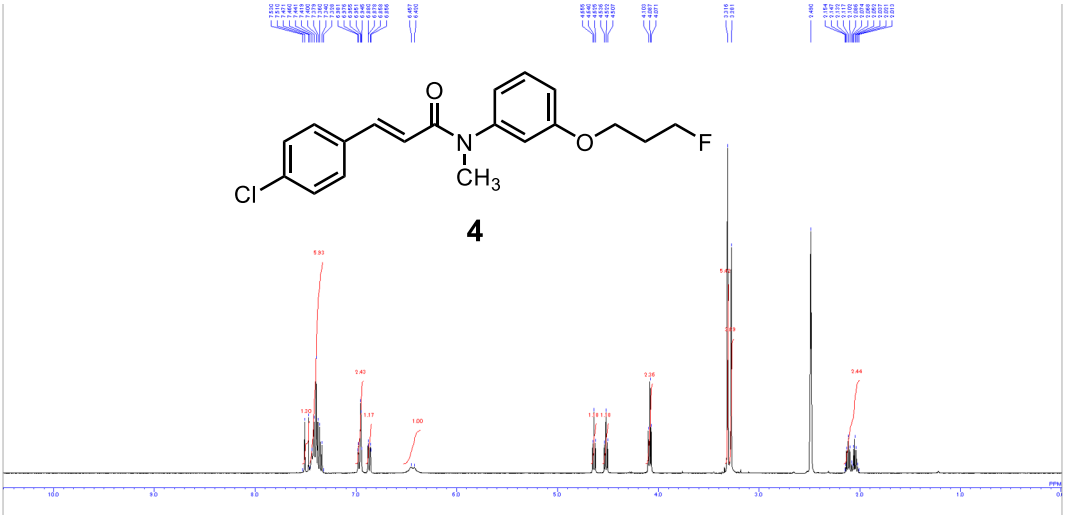


High-resolution mass spectrum

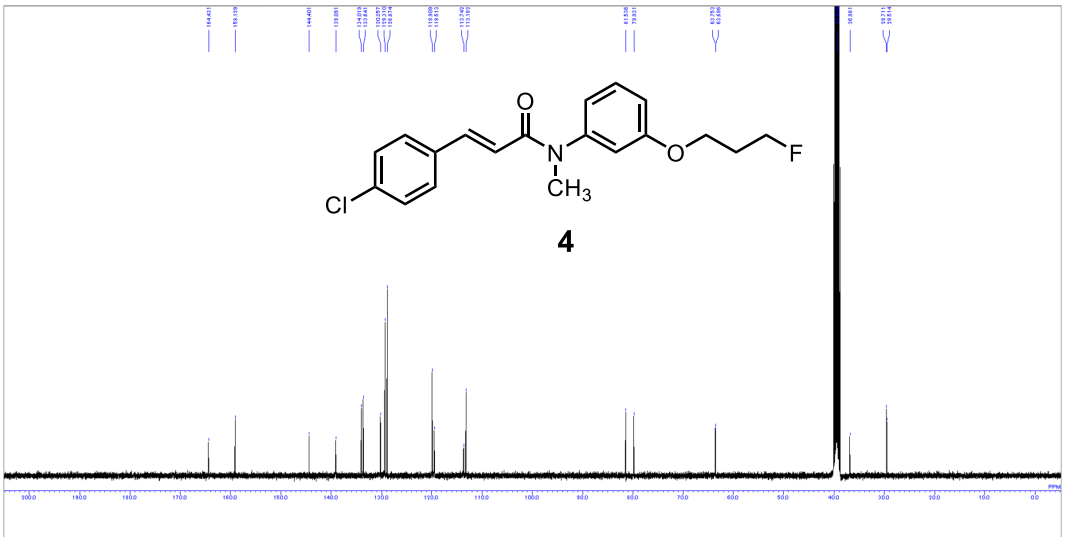


Characterization data for compound **4**

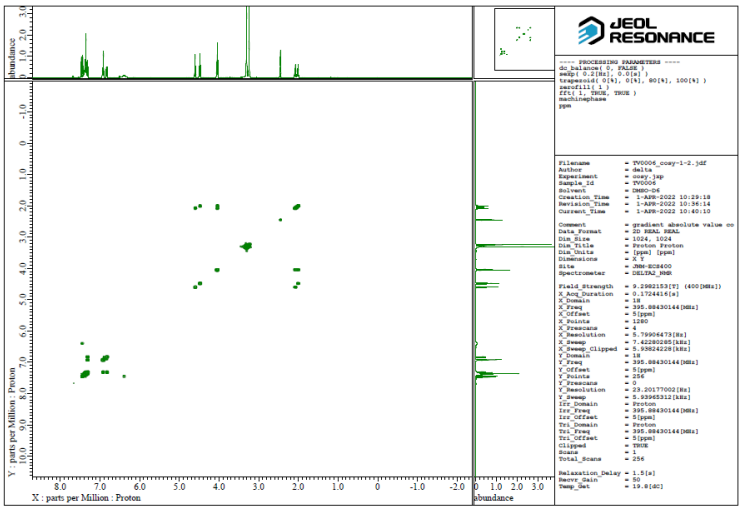
^{1}H NMR (400 MHz, DMSO-d_6) spectrum

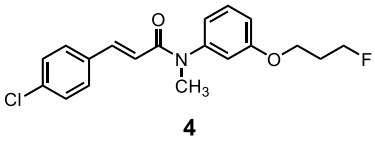


^{13}C{^{1}H} NMR (100 MHz, DMSO-d_6) spectrum



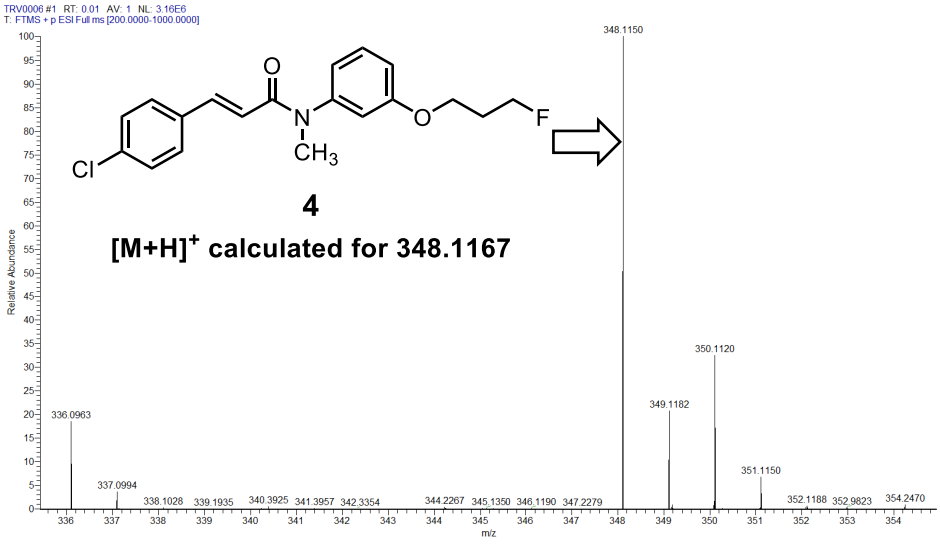
^{1}H–^{1}H COSY (DMSO-d_6) spectrum





4

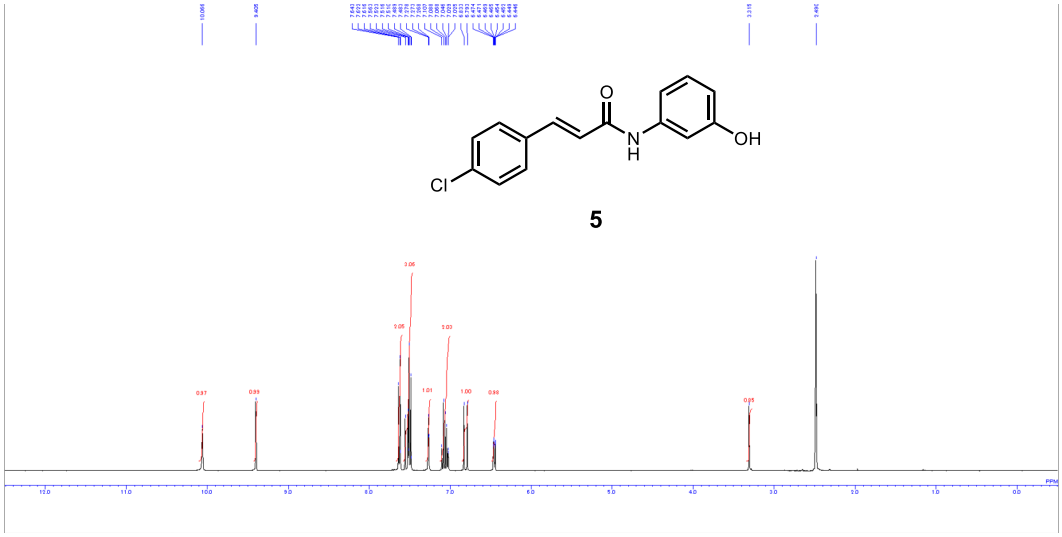
High-resolution mass spectrum



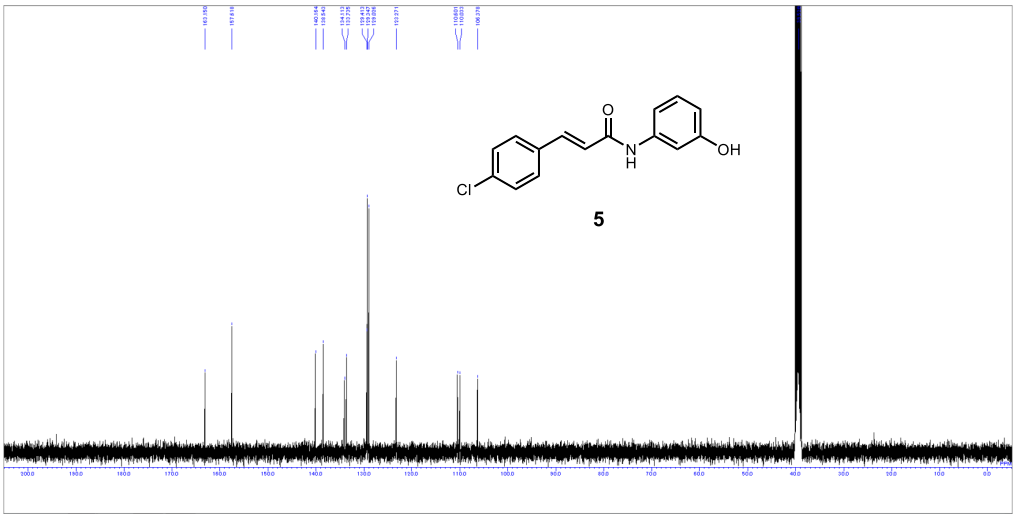
Characterization of compound **5**

^{1}H NMR (400 MHz, DMSO-d_6) spectrum

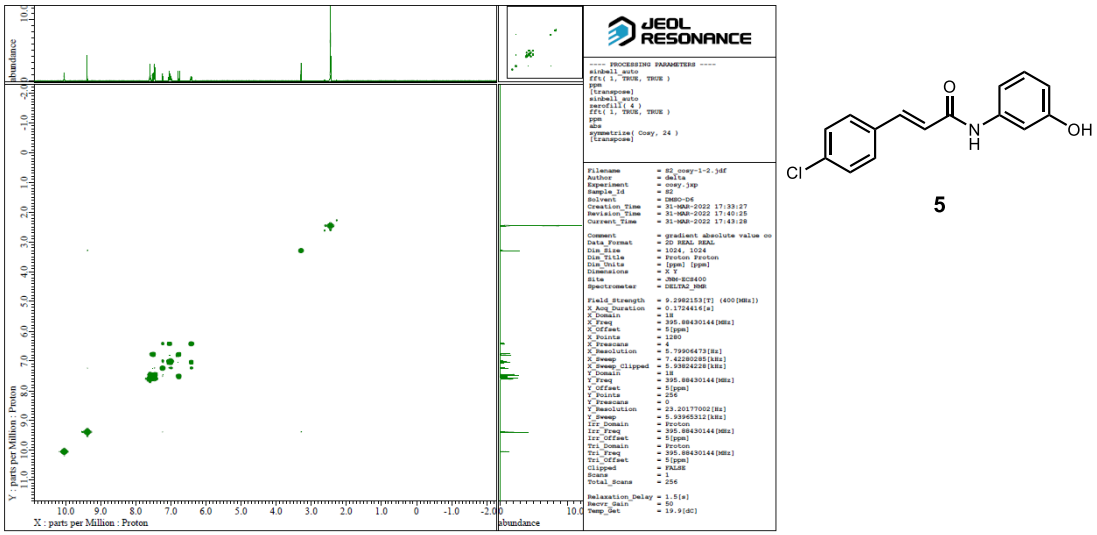
This ^{1}H NMR spectrum is consistent with that of a previous report (see D. V. Veghel *et al.*, *Nucl. Med. Biol.*, 2013, **40**, 141–147).



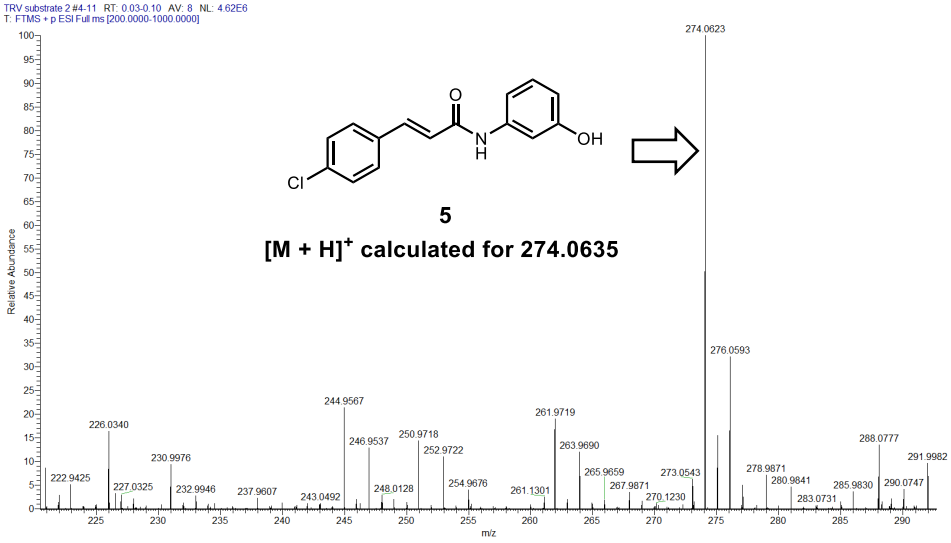
^{13}C{^{1}H} NMR (100 MHz, DMSO-d_6) spectrum



^{1}H–^{1}H COSY (DMSO-d_6) spectrum

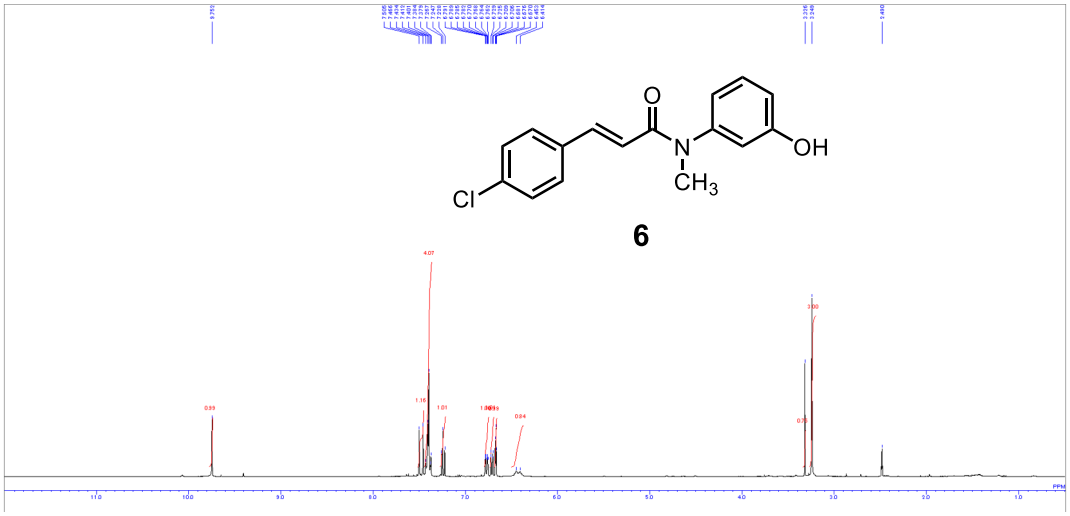


High-resolution mass spectrum

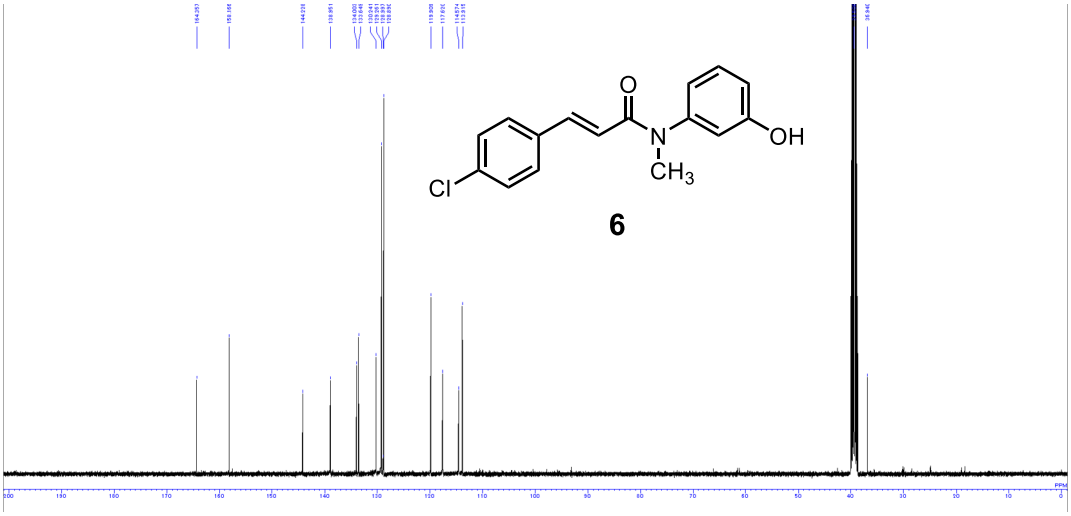


Characterization data for compound **6**

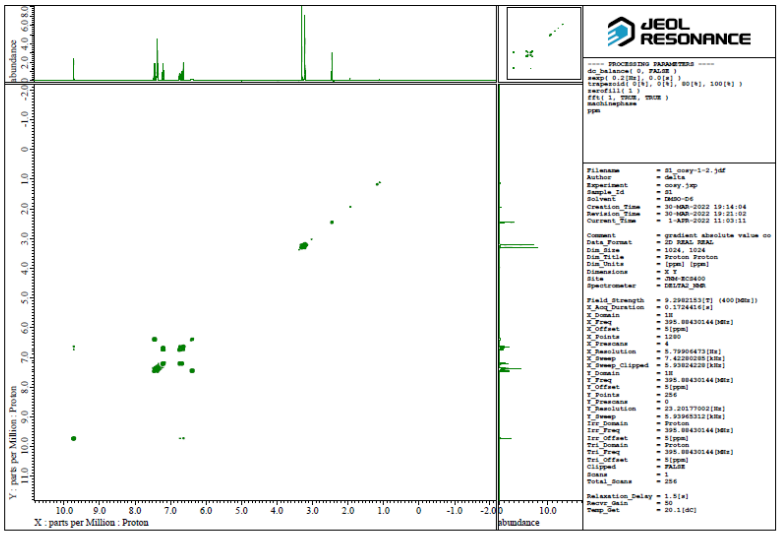
^{1}H NMR (400 MHz, DMSO-d_6) spectrum

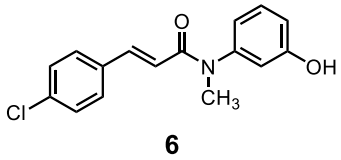


^{13}C{^{1}H} NMR (100 MHz, DMSO-d_6) spectrum



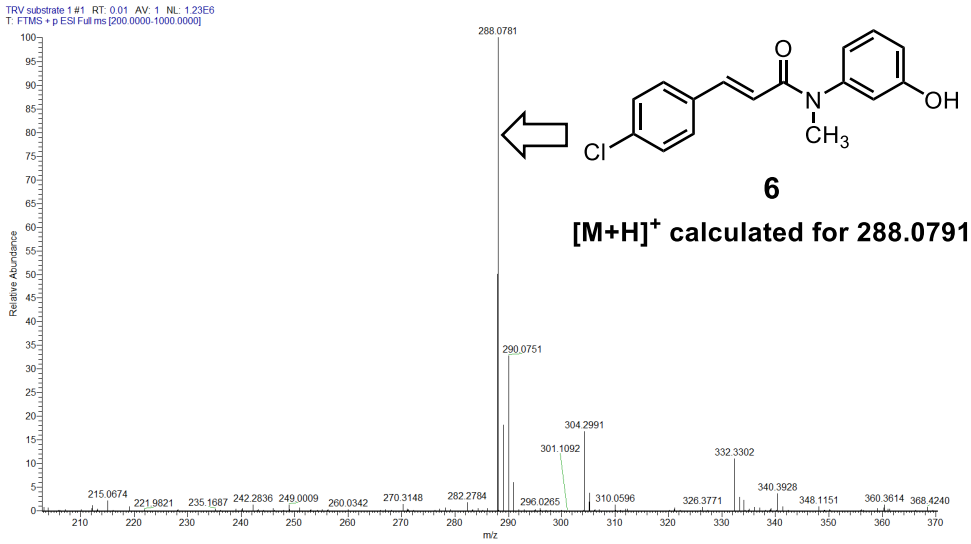
^{1}H–^{1}H COSY (DMSO-d_6) spectrum





6

High-resolution mass spectrum

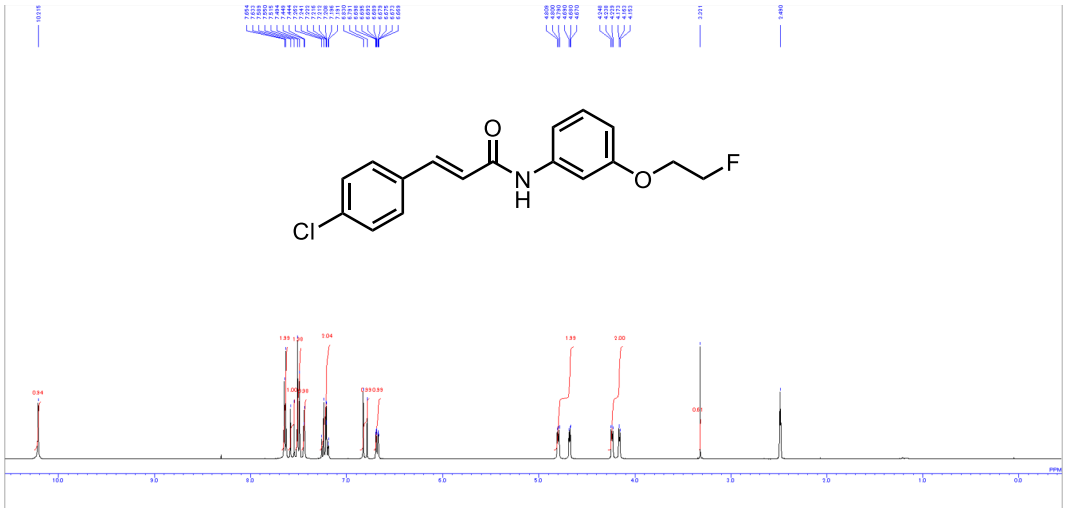


6

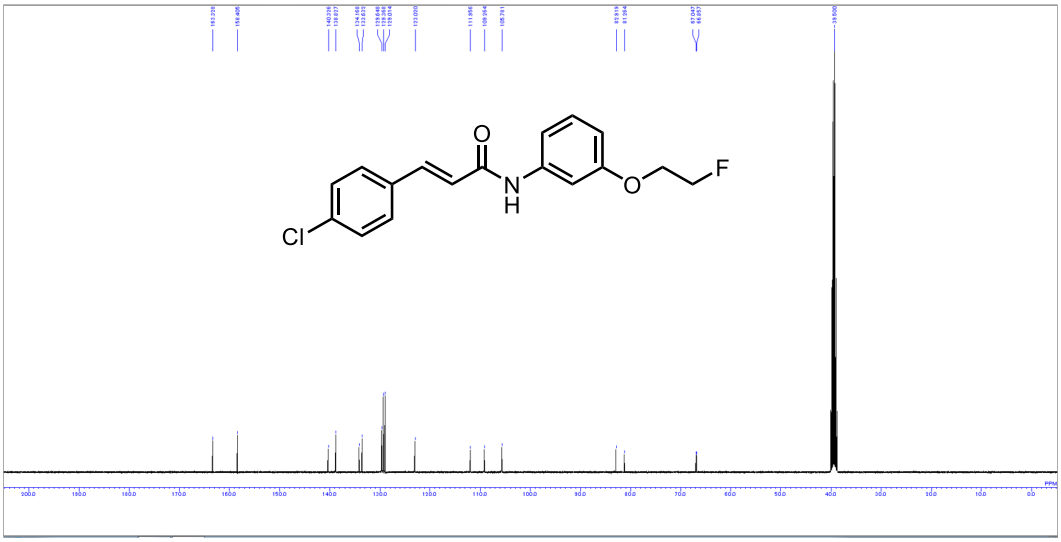
[M+H]$^{+}$ calculated for 288.0791

Characterization of *N*-{3-(2-fluoroethoxy)phenyl}-4-chlorocinnamamide

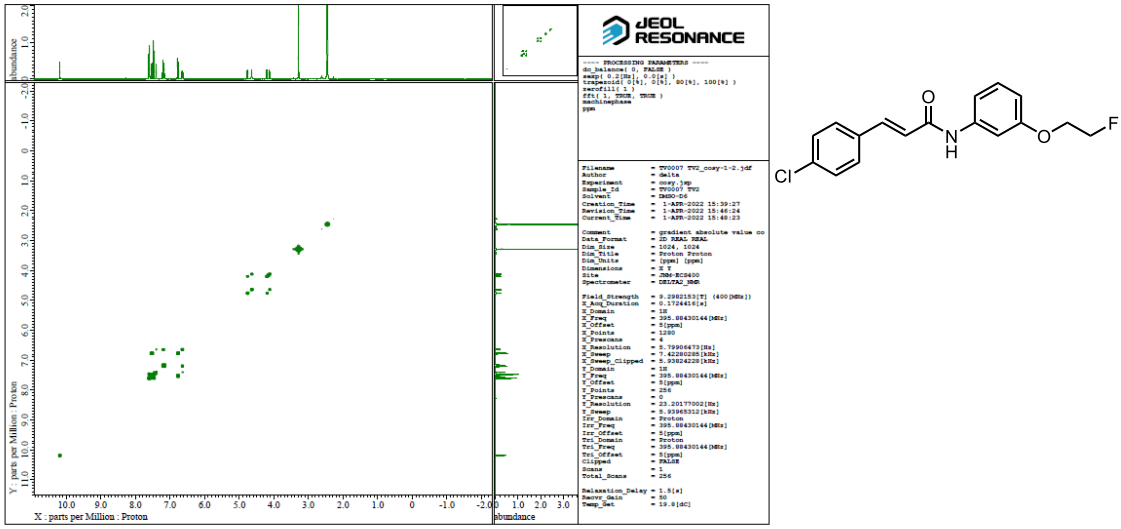
^{1}H NMR (400 MHz, DMSO-d_6) spectrum



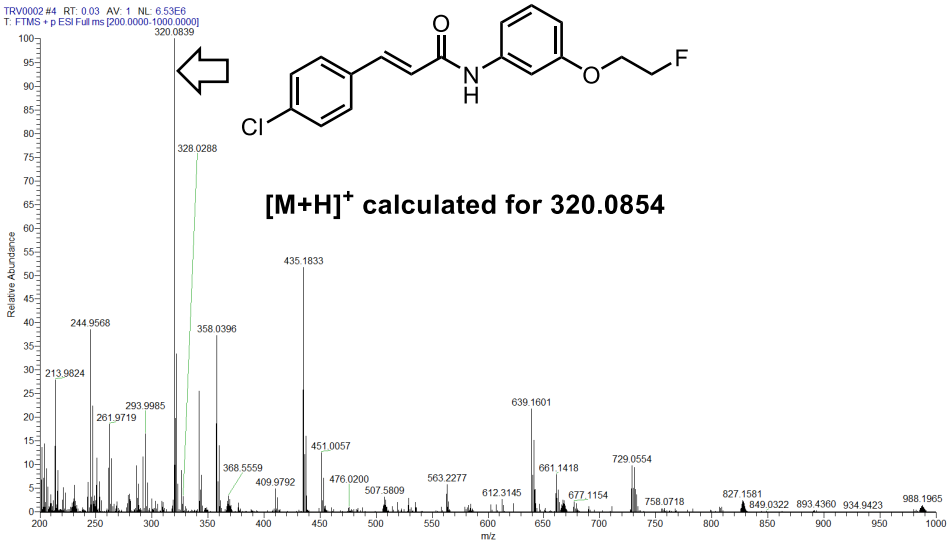
^{13}C{^{1}H} NMR (100 MHz, DMSO-d_6) spectrum



^{1}H–^{1}H COSY (DMSO-d_6) spectrum

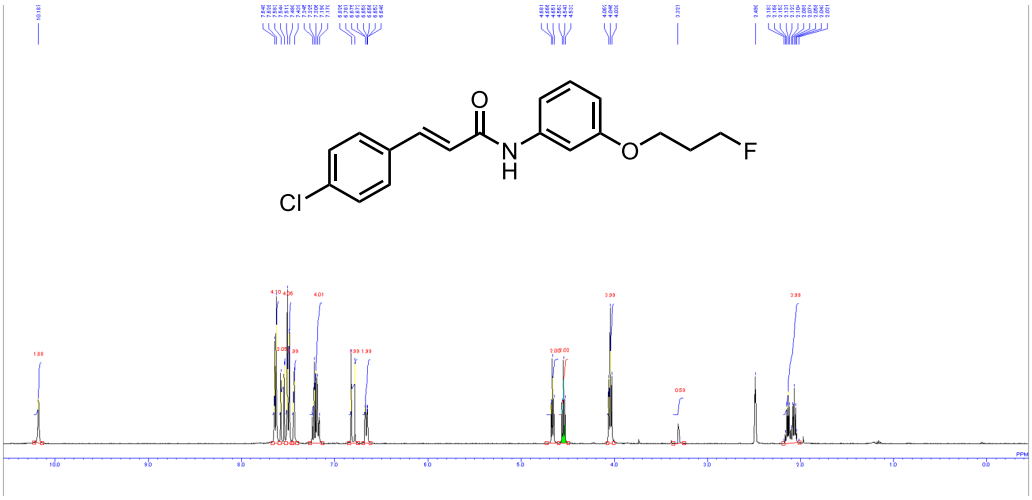


High-resolution mass spectrum

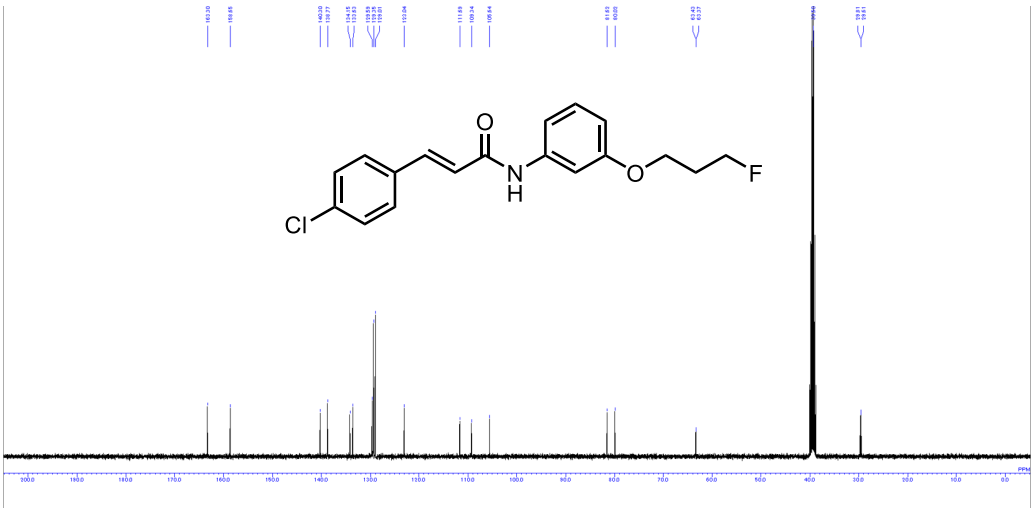


Characterization of *N*-{3-(3-fluoropropoxy)phenyl}-4-chlorocinnamamide

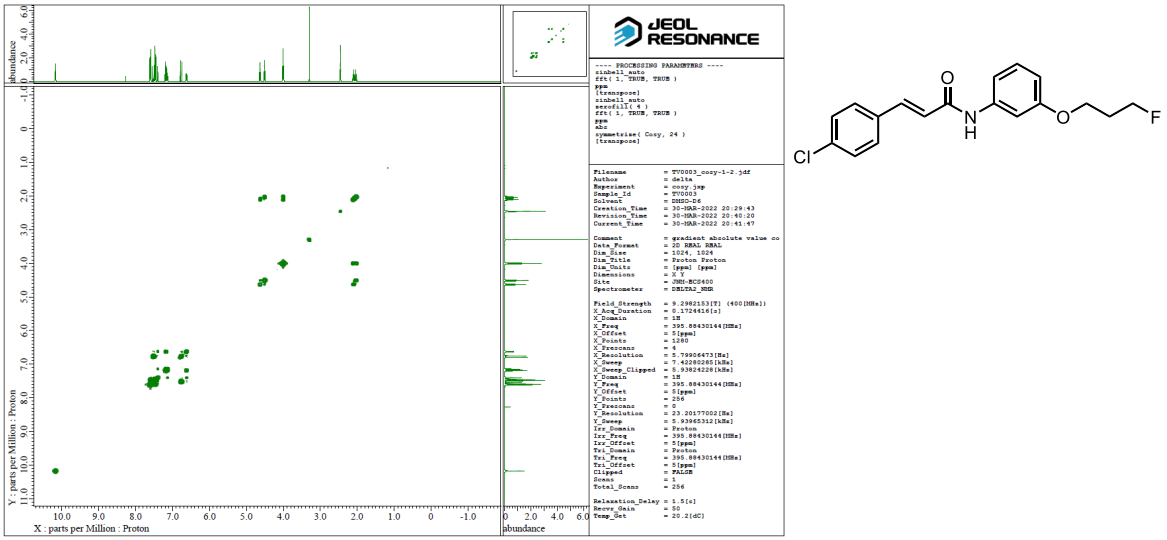
^{1}H NMR (400 MHz, DMSO-d_6) spectrum



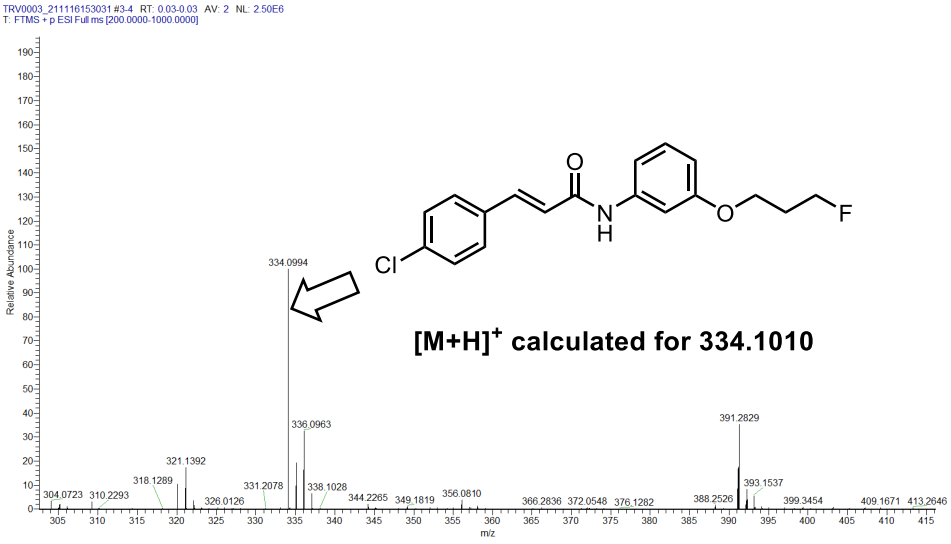
^{13}C{^{1}H} NMR (100 MHz, DMSO-d_6) spectrum



^{1}H–^{1}H COSY (DMSO-d_6) spectrum



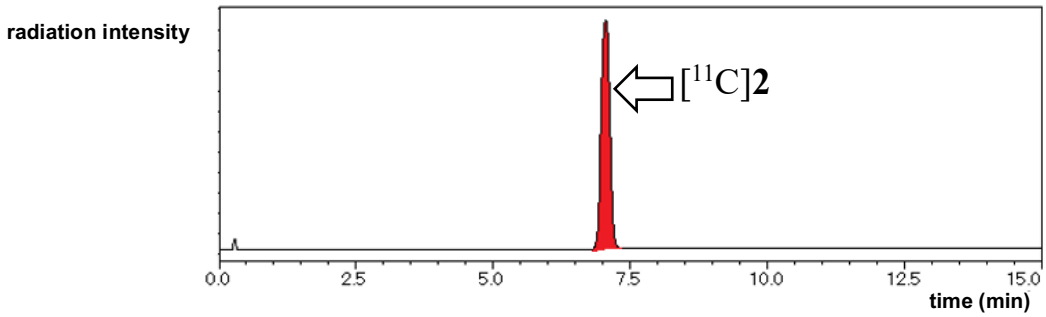
High-resolution mass spectrum



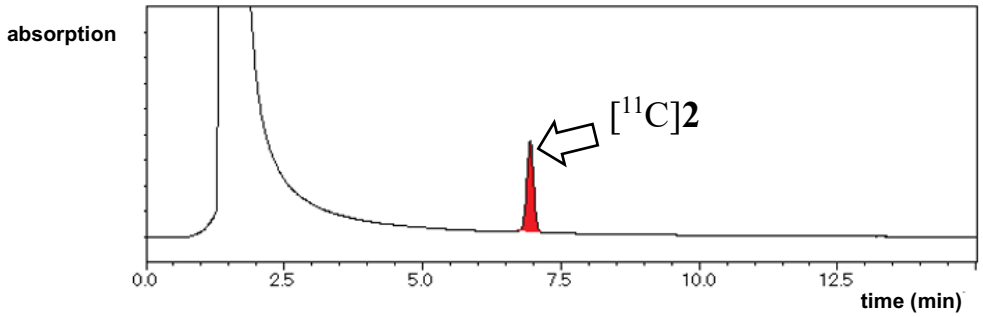
Chapter 2: Analytical data for radiolabeled tracers [^{11}C]**2** and [^{18}F]**4**

Purity analysis of [^{11}C]**2**: (a) radio-HPLC analysis, and (b) UV-HPLC analysis; the peak at 1.5–4.0 min is largely attributed to the use of ascorbic acid as an additive.

(a) Radio-HPLC analysis



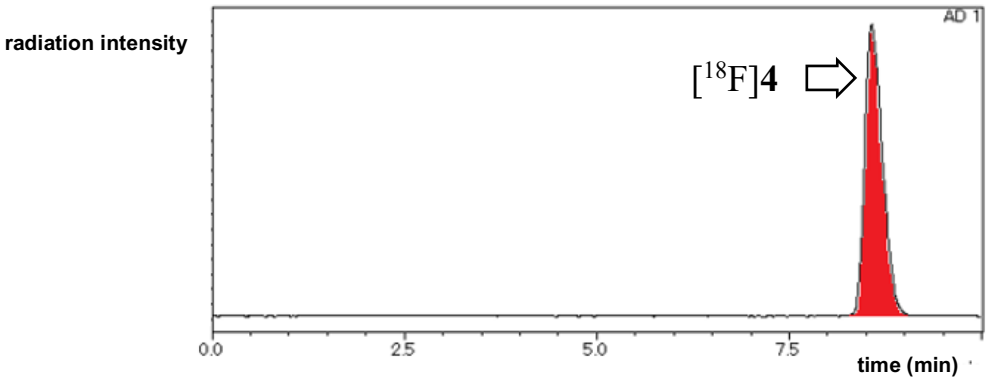
(b) UV-HPLC analysis



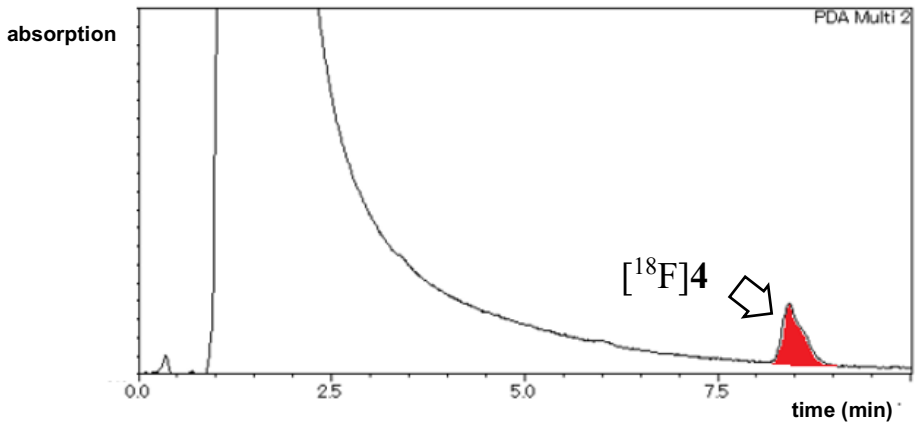
HPLC conditions: column: COSMOSIL, 5C_{18}-MS-II, 4.6 × 150 mm; mobile phase: acetonitrile/H_2O = 60:40; flow rate: 1.0 mL/min; column temperature: 25 °C; UV detection: 254 nm; retention time 7.0 min.

Purity analysis of [^{18}F]**4**: (a) radio-HPLC analysis and (b) UV-HPLC analysis; the peak at 1.5–4.0 min is largely attributed to the use of ascorbic acid as an additive.

(a) Radio-HPLC analysis



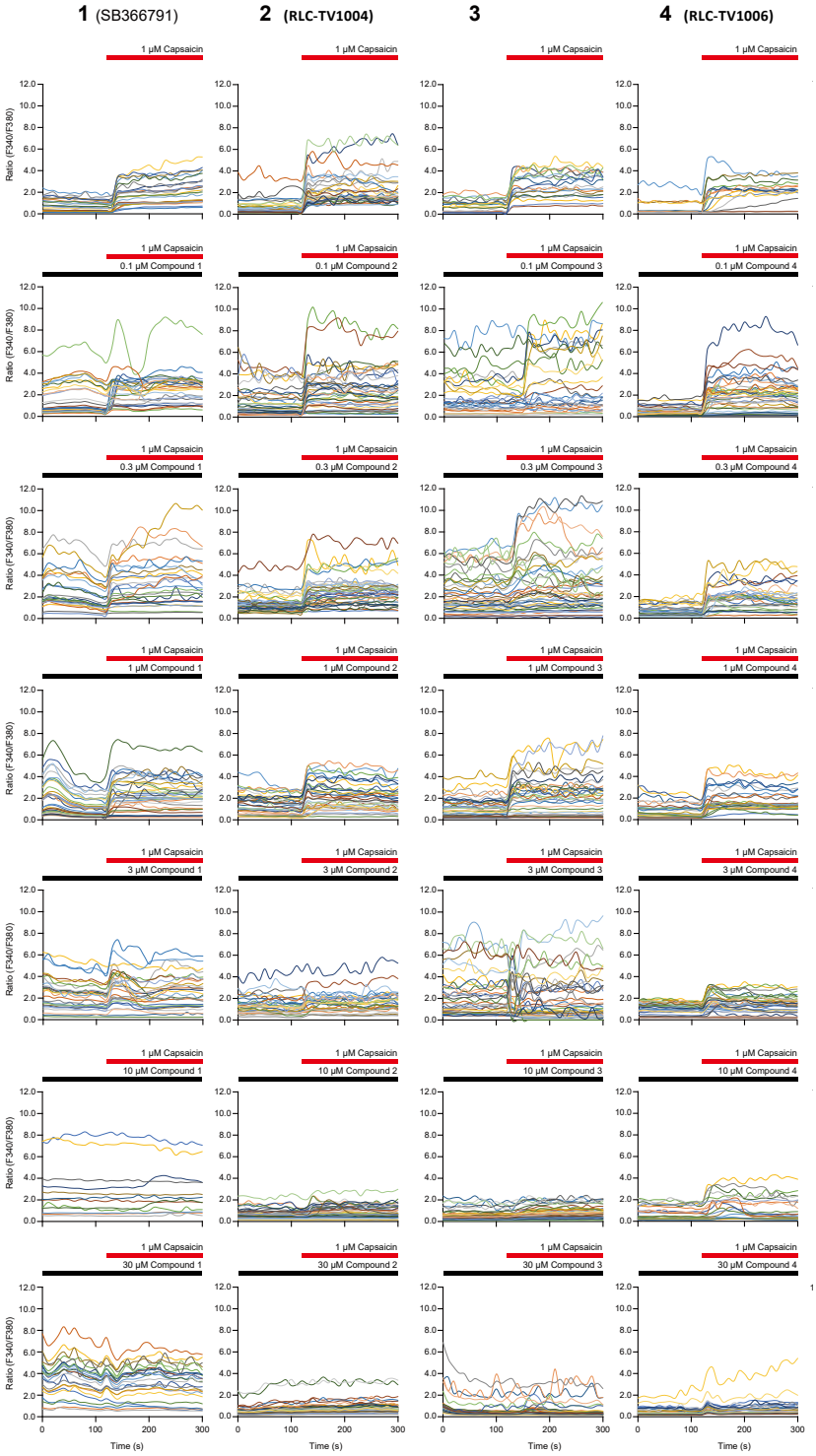
(b) UV-HPLC analysis



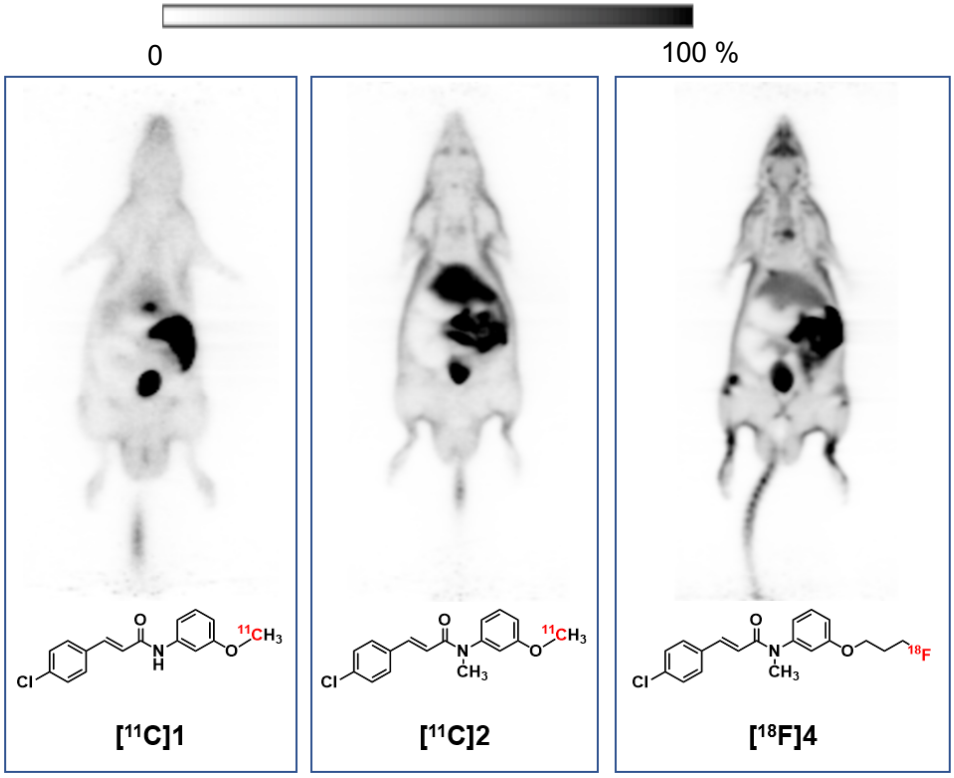
HPLC conditions: column: COSMOSIL, 5C_{18}-MS-II, 4.6 × 150 mm; mobile phase: acetonitrile/H_2O = 60/40; flow rate: 1.0 mL/min; column temperature: 25 °C; UV detection: 280 nm; retention time 8.6 min.

Chapter 3: Analytical data for calcium(II) influx assays using **1**–**4**

Ca^{2+} influx assay using rat TRPV1-expressing HEK293 cells and a 1.0 μM solution of capsaicin as an agonist; regarding the test compounds **1**–**4** (specified concentration), fura-2 was excited at 340 and 380 nm and the ratio of the emissions was measured.



Chapter 4: Whole-body PET images of three radiotracers [^{11}C]**1**, [^{11}C]**2**, and [^{18}F]**4**



The PET radiotracers [^{11}C]**1**, [^{11}C]**2**, and [^{18}F]**4** were intravenously injected into the tail vein of rats, and the summated PET images (5–60 min after tracer injection) were reconstructed. A clear whole-body image of [^{11}C]**2** with a high S/N ratio has been presented in the text. For the experimental procedure, please refer to the Experimental section.