

Targeting fatty-acid amide hydrolase with prodrugs for CNS-selective therapy

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Supporting Information

Table S1. COS-7 cell homogenate not transfected with the FAAH1 plasmid retains little substrate hydrolysis activity compared to homogenate overexpressing FAAH. Substrates were tested at 100 μ M.

| Substrate | FAAH (nmol * mg ⁻¹ * min ⁻¹) | Mock (nmol * mg ⁻¹ * min ⁻¹) |
|-------------------|---|---|
| AEA | 69.2 \pm 19.74 | 0 ^a |
| Previous lead (3) | 1.36 \pm 0.02 | 0.067 \pm 0.002 |
| Sob-AM1 (6) | 7.2 \pm 1.2 | 0.03 \pm 0.005 |
| Sob-AM2 (7) | 5.6 \pm 0.5 | 0.102 \pm 0.007 |

^aBelow limit of detection

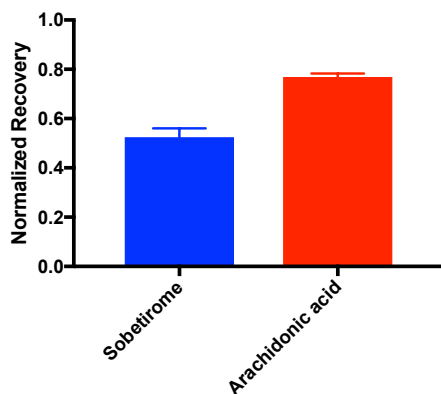


Figure S1. Extraction efficiencies for both sobetirome and arachidonic acid were determined for the extraction from the cell homogenate reaction. For both products, known amounts of the deuterated sample were spiked in prior to extraction while unlabelled sample was spiked in at the end of the extraction. The relative ratio of labelled/unlabelled products was determined by comparing ion ratios for matched fragments in the typical LC/MS-MS assay. Extraction efficiencies for both sobetirome (0.48) and arachidonic acid (0.76) were used to correct for differences in Figure 1B and Table 1.

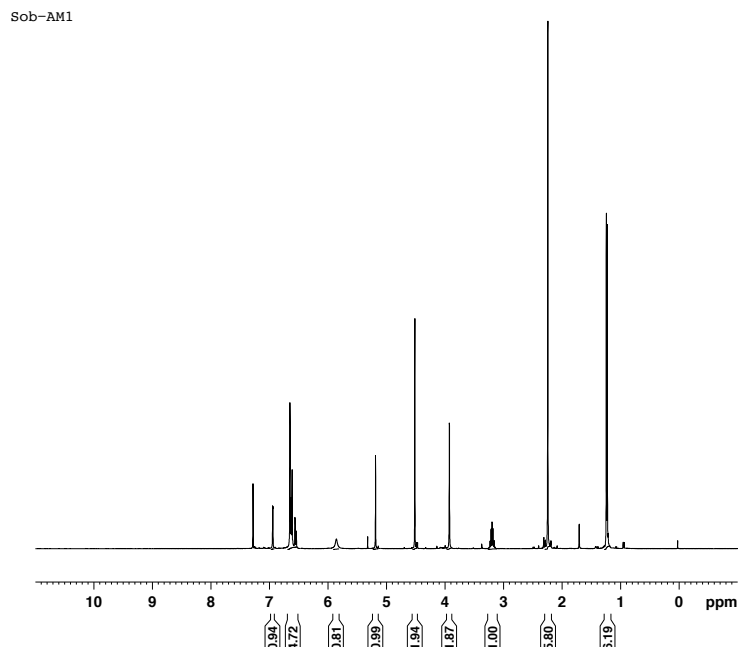


Figure S2. ¹H NMR spectra of Sob-AM1 (6)

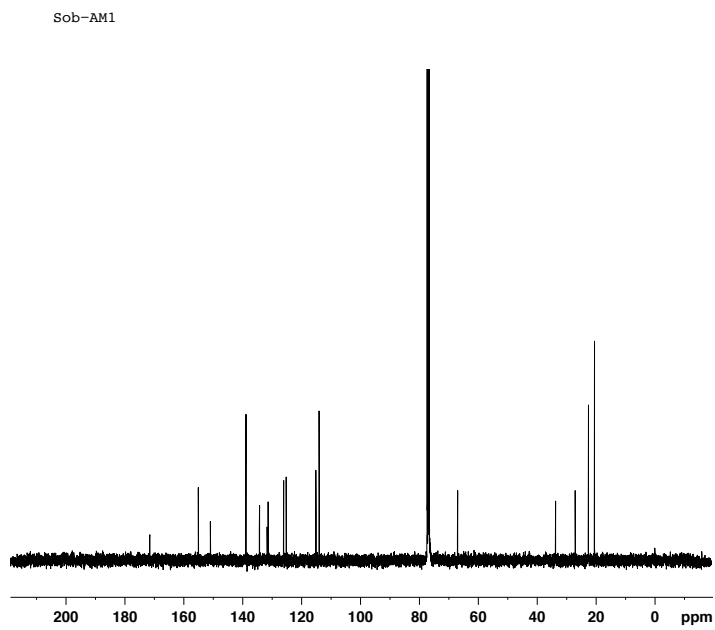


Figure S3. ¹³C NMR spectra of Sob-AM1 (6)

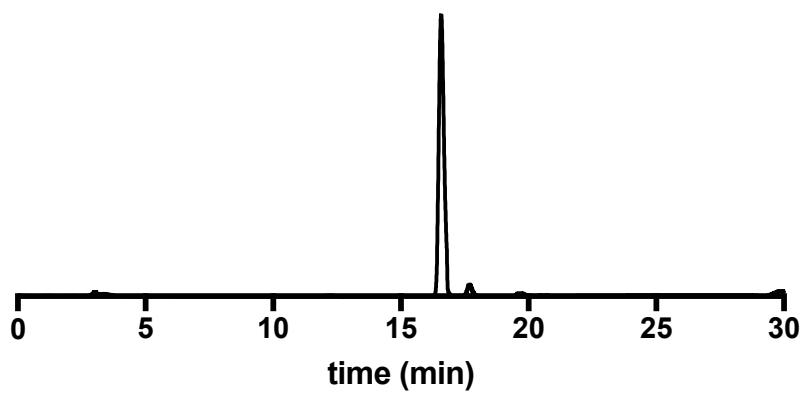


Figure S4. HPLC trace of Sob-AM1 (6)

Sob-AM2

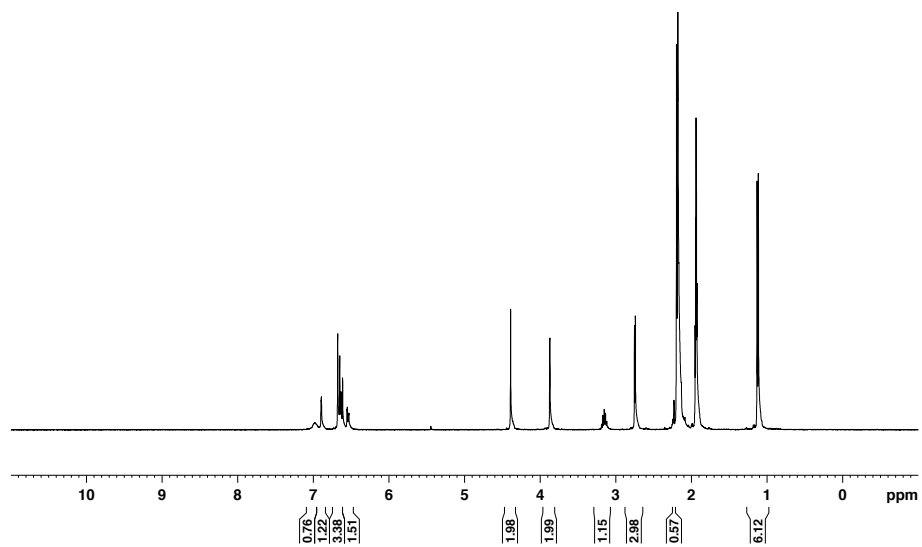


Figure S5. ¹H NMR spectra of Sob-AM2 (7)

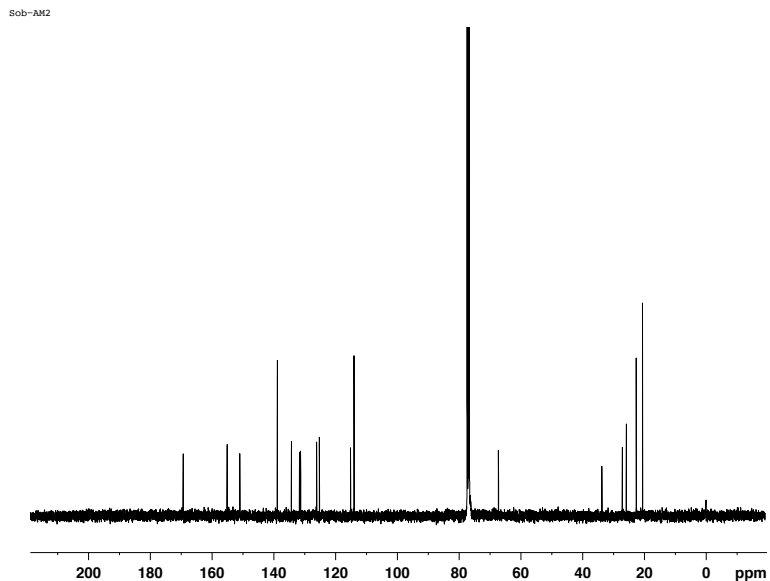


Figure S6. ¹³C NMR spectra of Sob-AM2 (7)

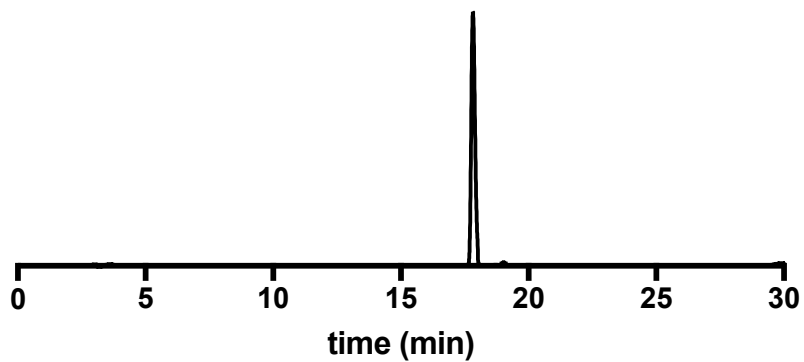


Figure S7. HPLC trace of Sob-AM2 (7)

d6-Sob-AM2

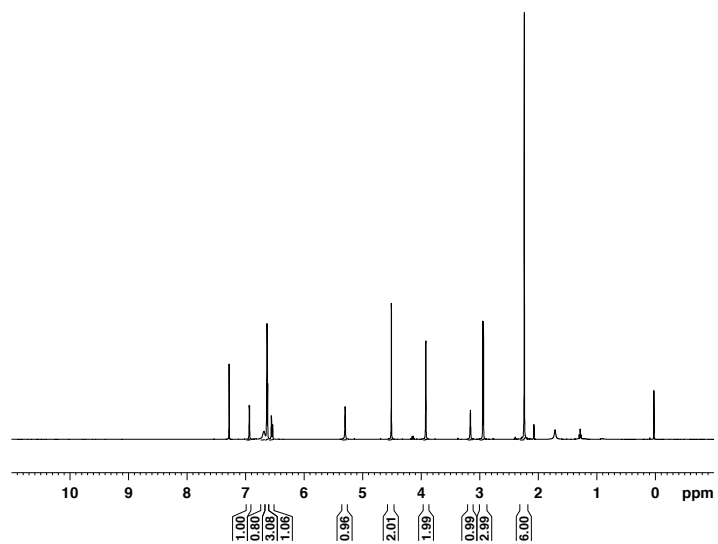


Figure S8. ¹H NMR spectra of d₆-Sob-AM2 (8)

d6-Sob-AM2

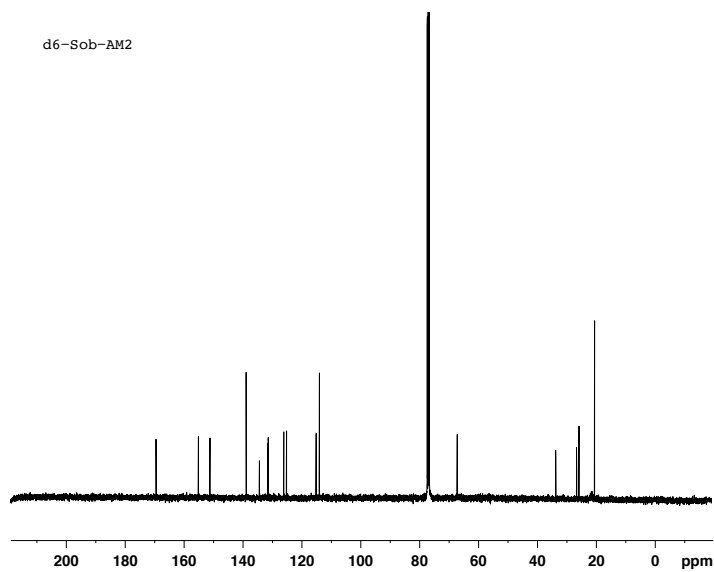


Figure S9. ¹³C NMR spectra of d₆-Sob-AM2 (8)

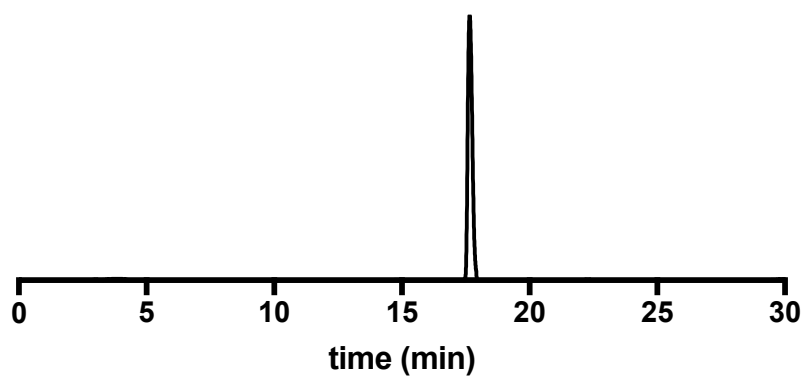


Figure S10. HPLC trace of d_6 -Sob-AM2 (8)