SUPPORTING MATERIAL INFORMATION

Conjugation of 2-(1'-Hexyloxyethyl)-2-Devinylpyropheophorbide-a (HPPH) to Carbohydrates Changes its Subcellular Distribution and Enhances Photodynamic Activity *in Vivo*†

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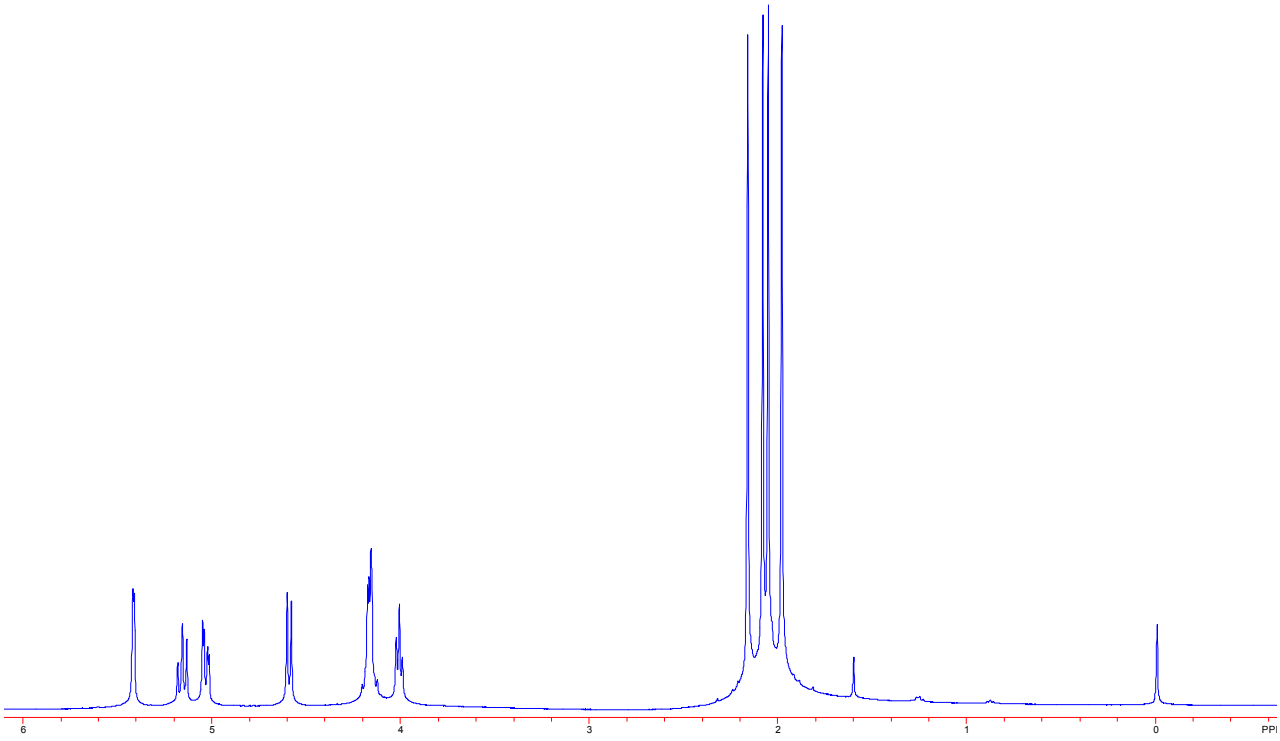
Contents:

Compound 1c



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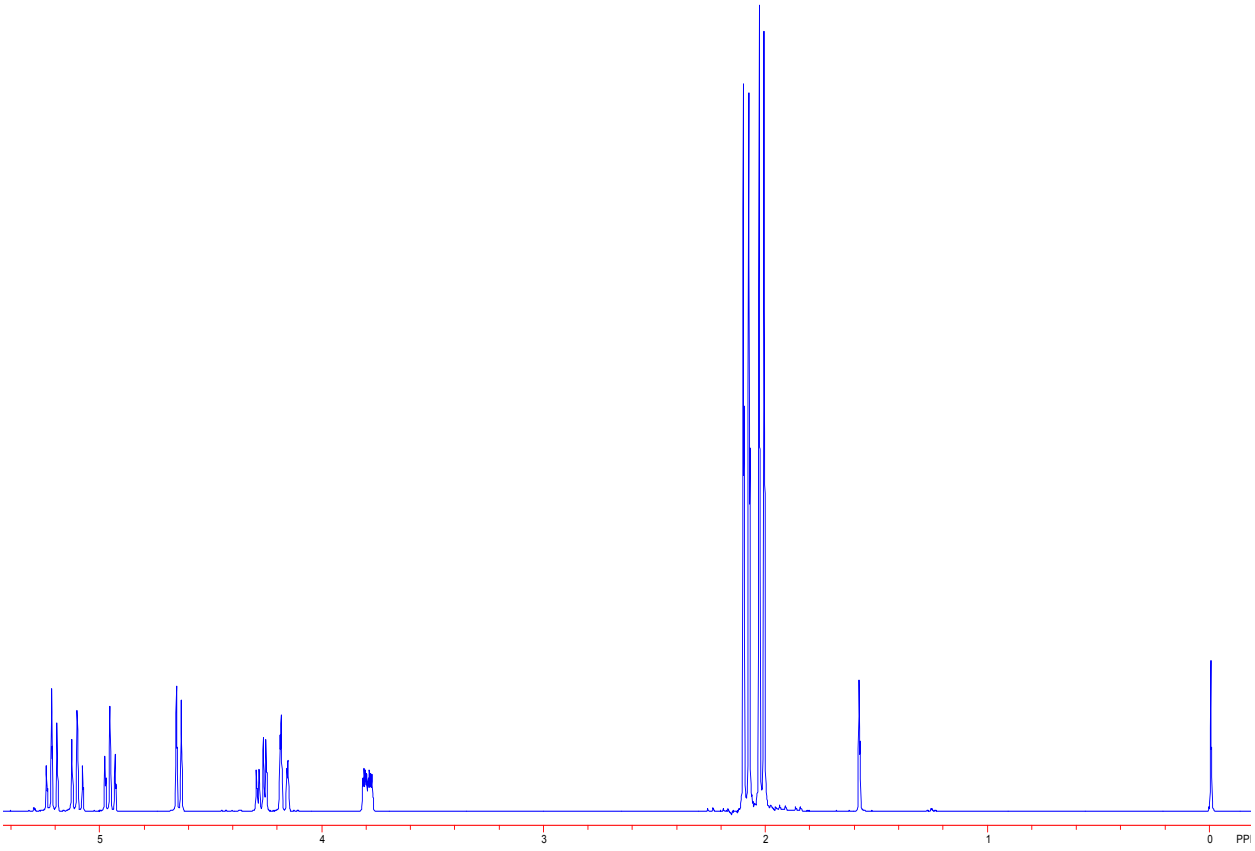

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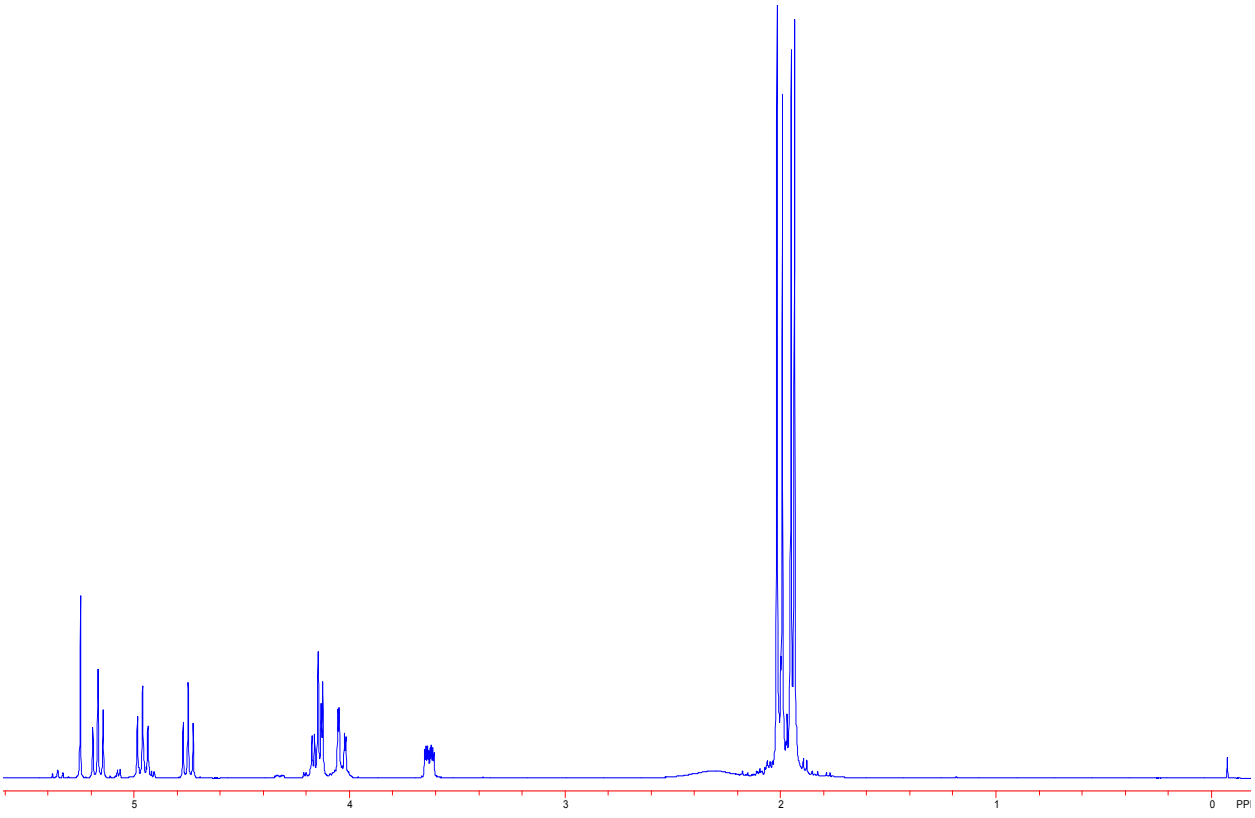

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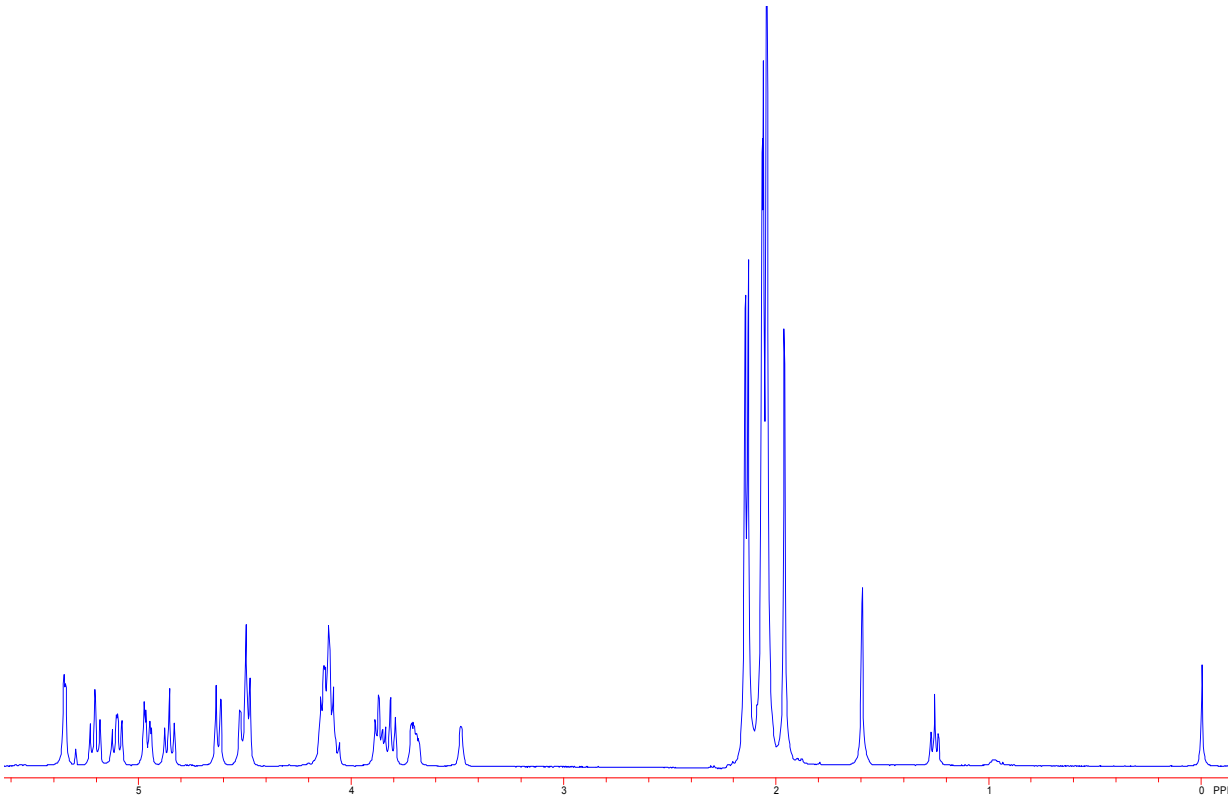

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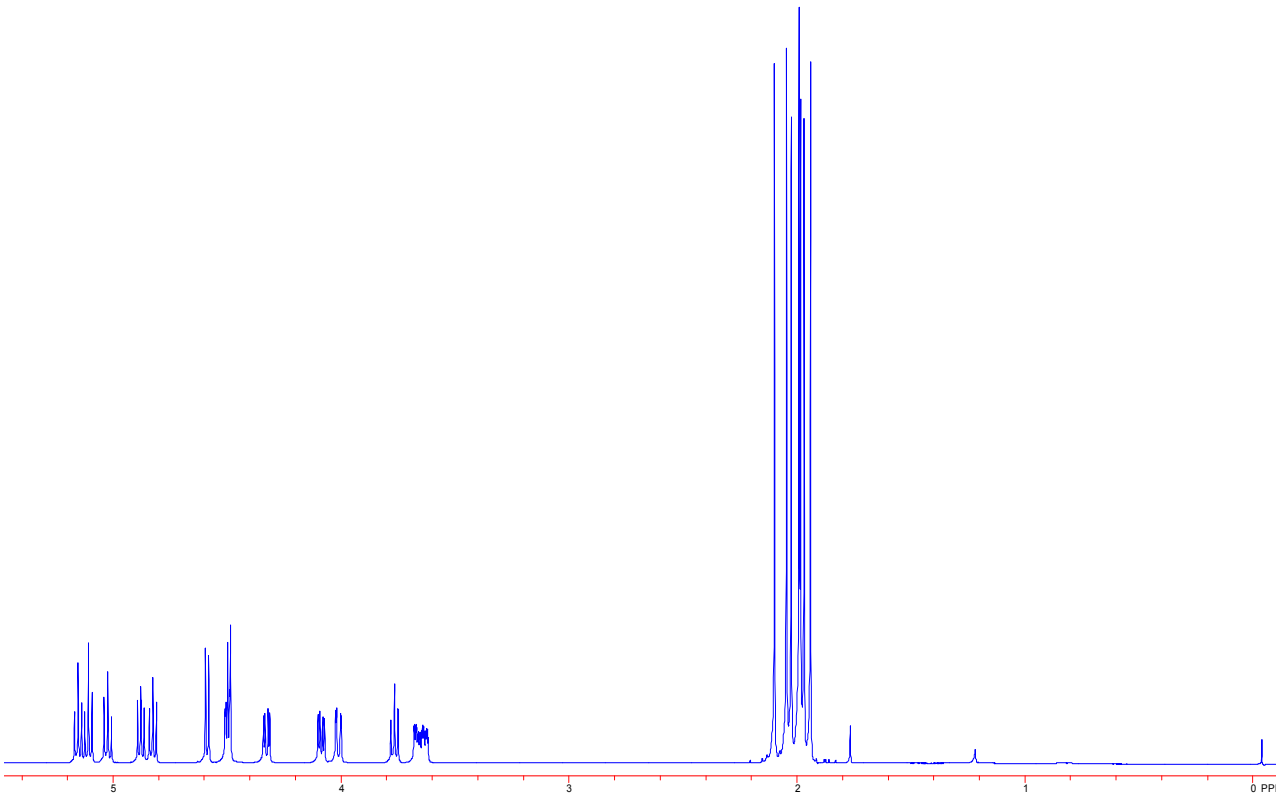

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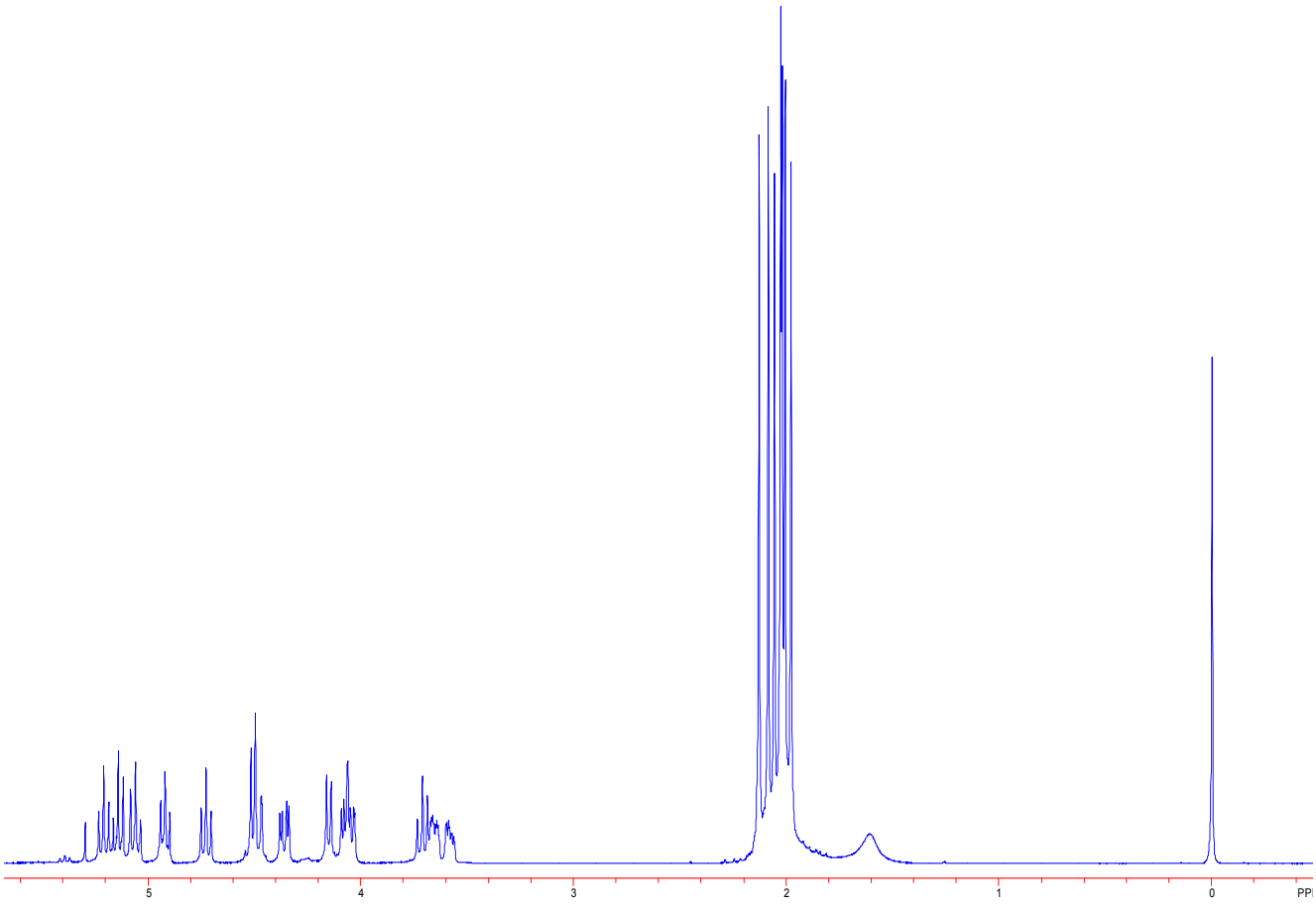

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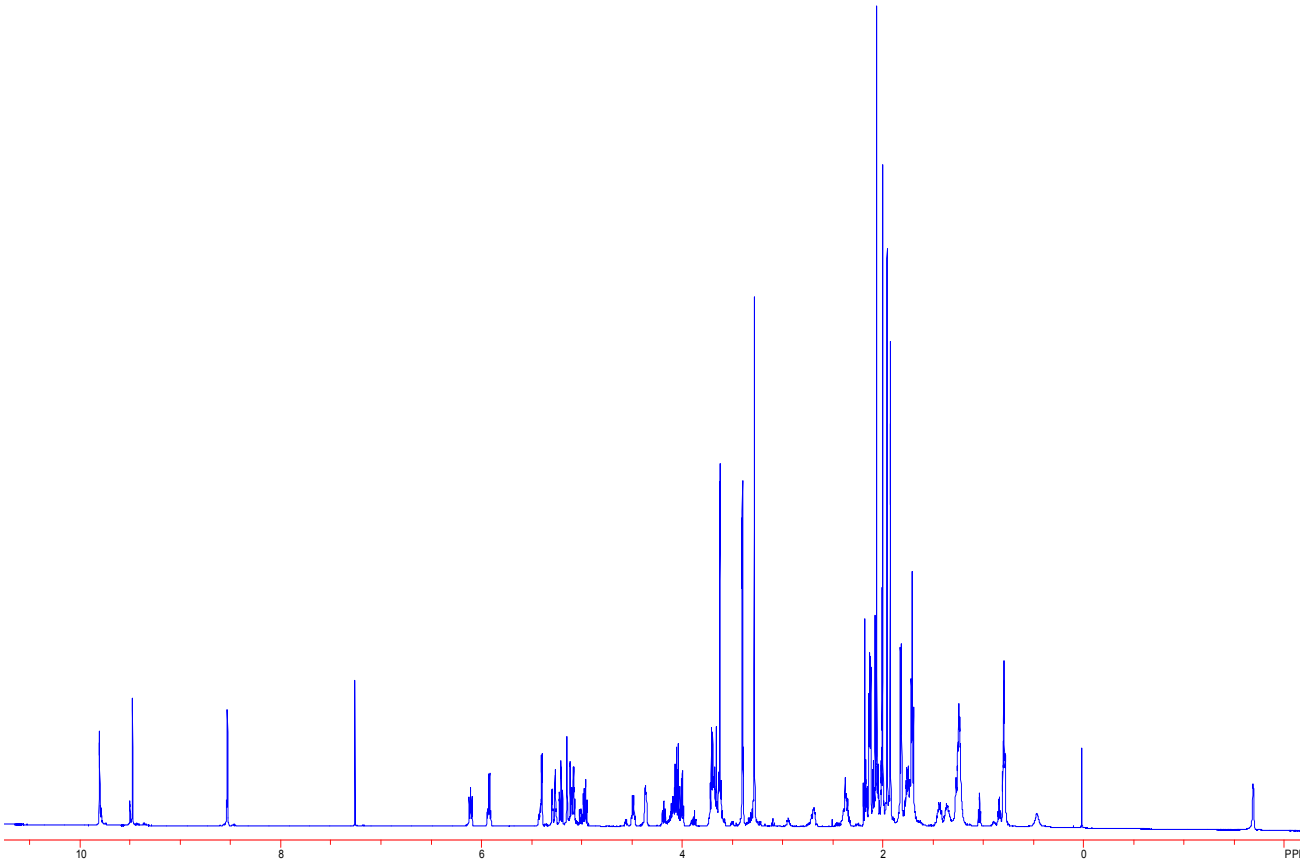

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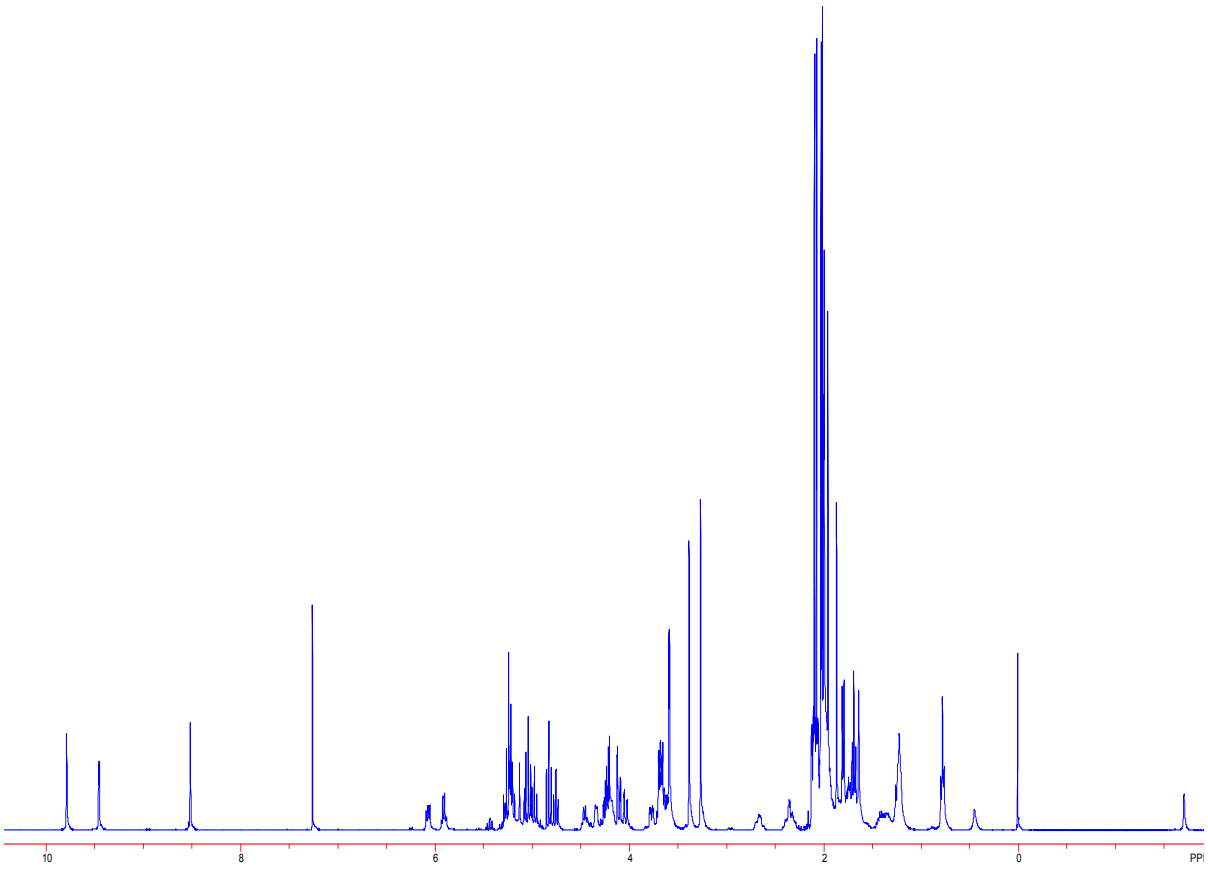

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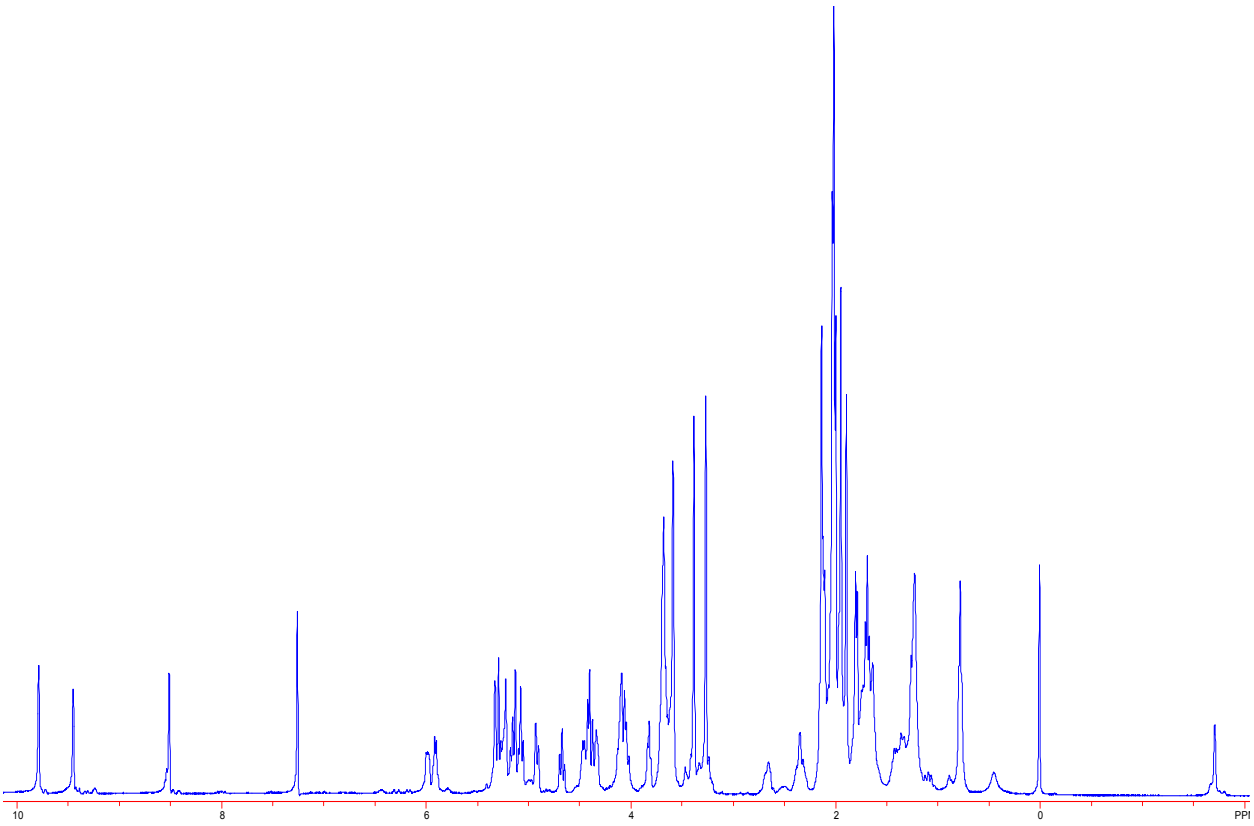

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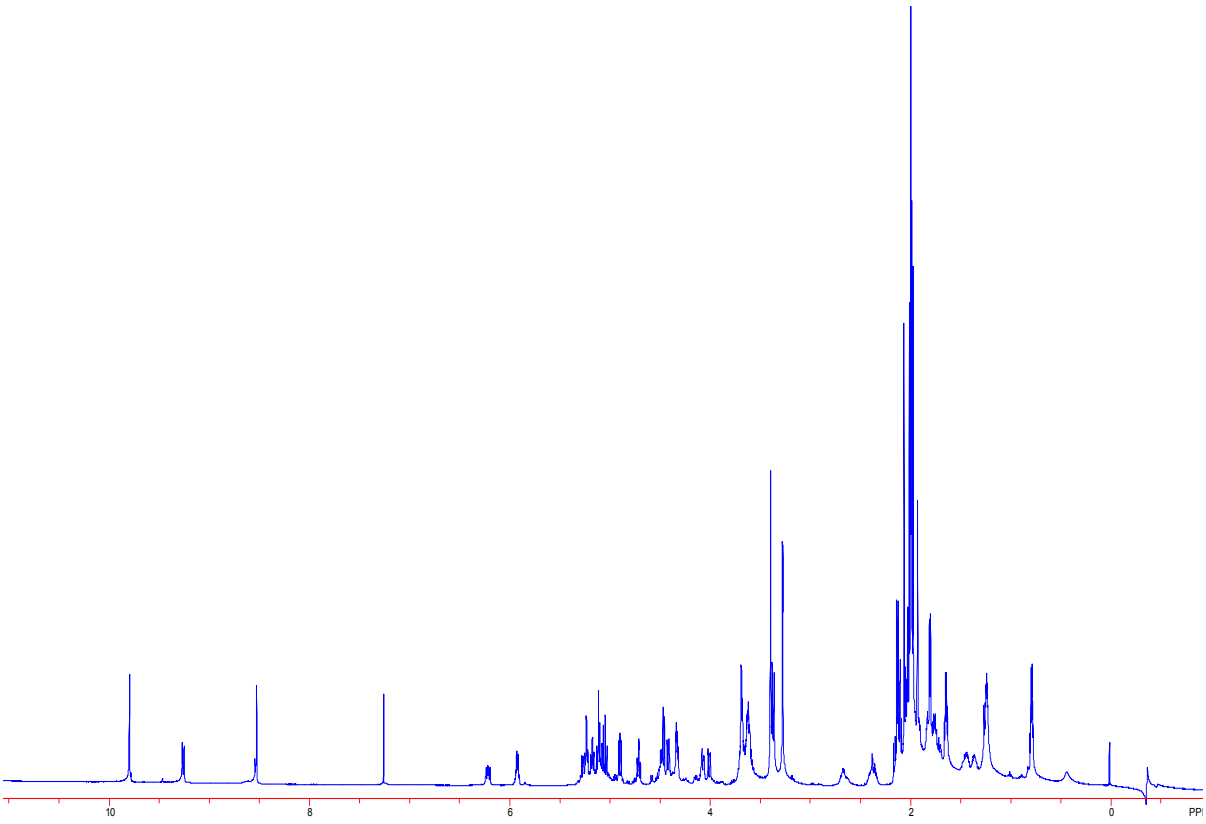

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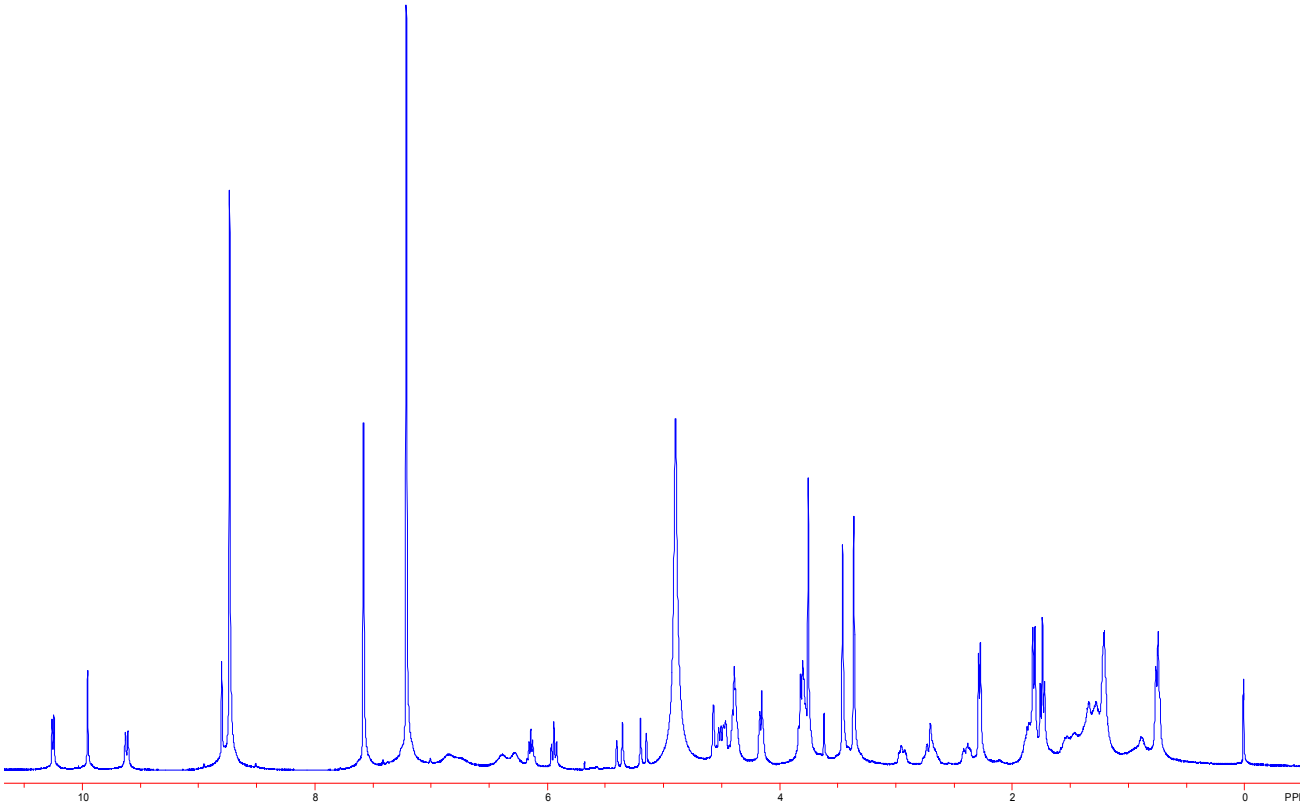

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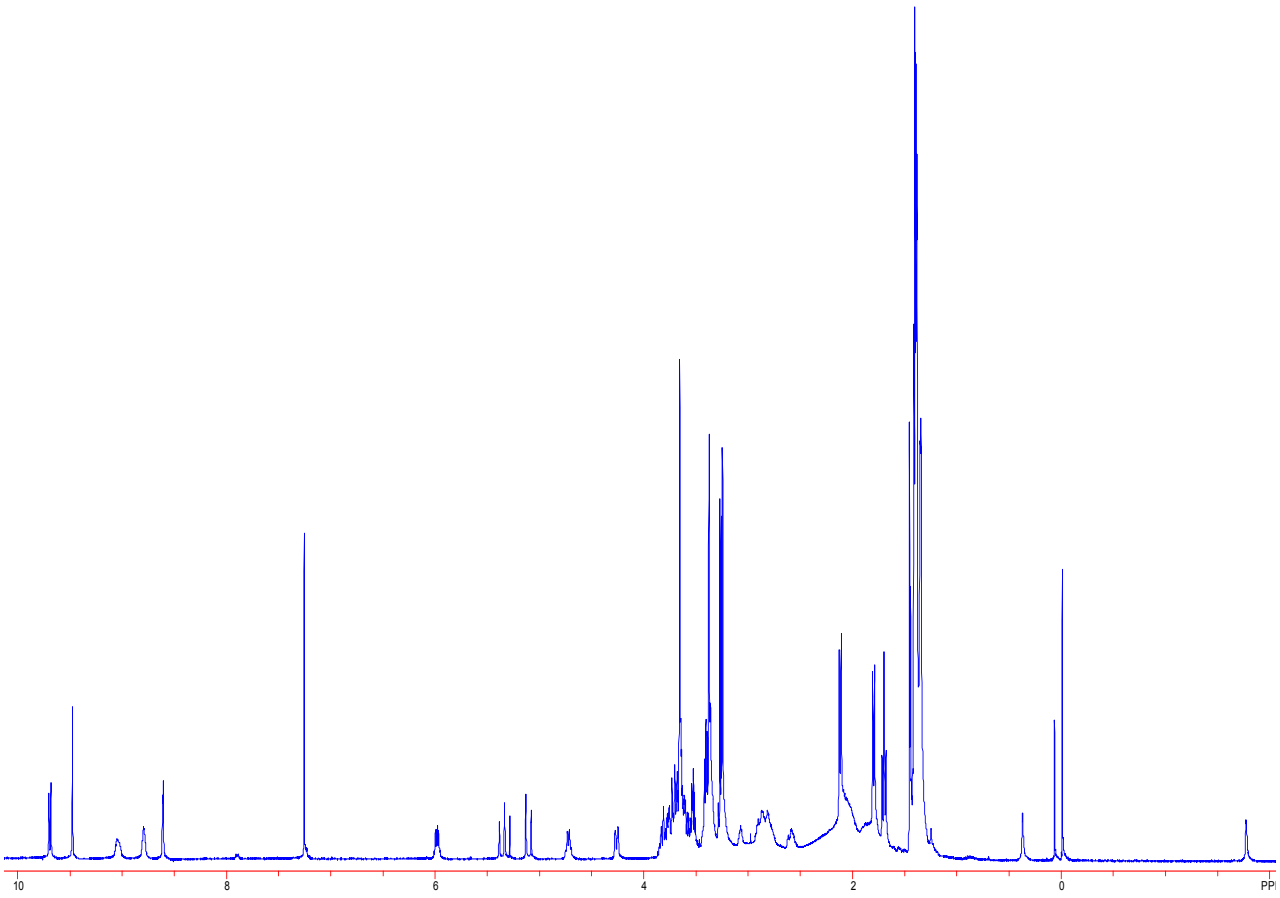

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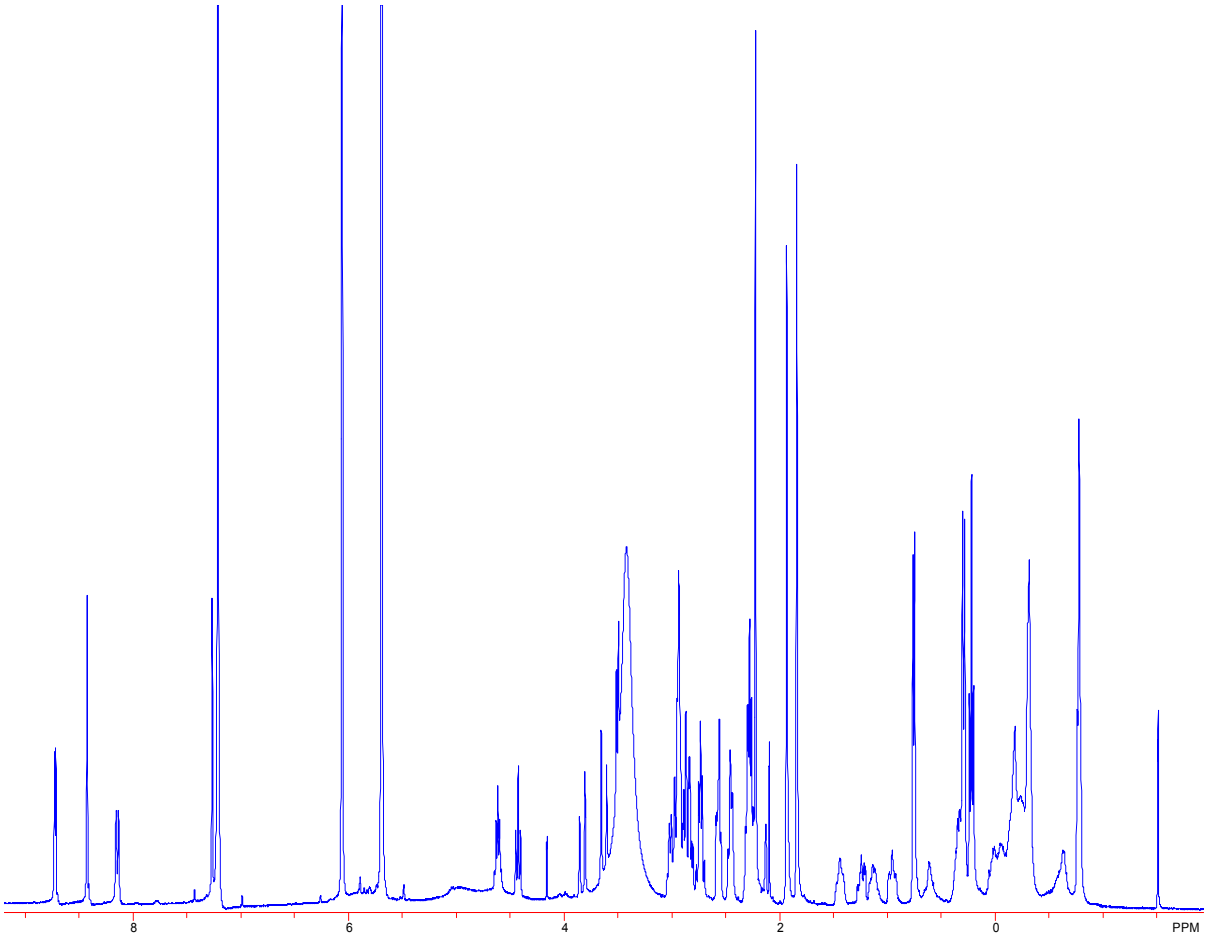

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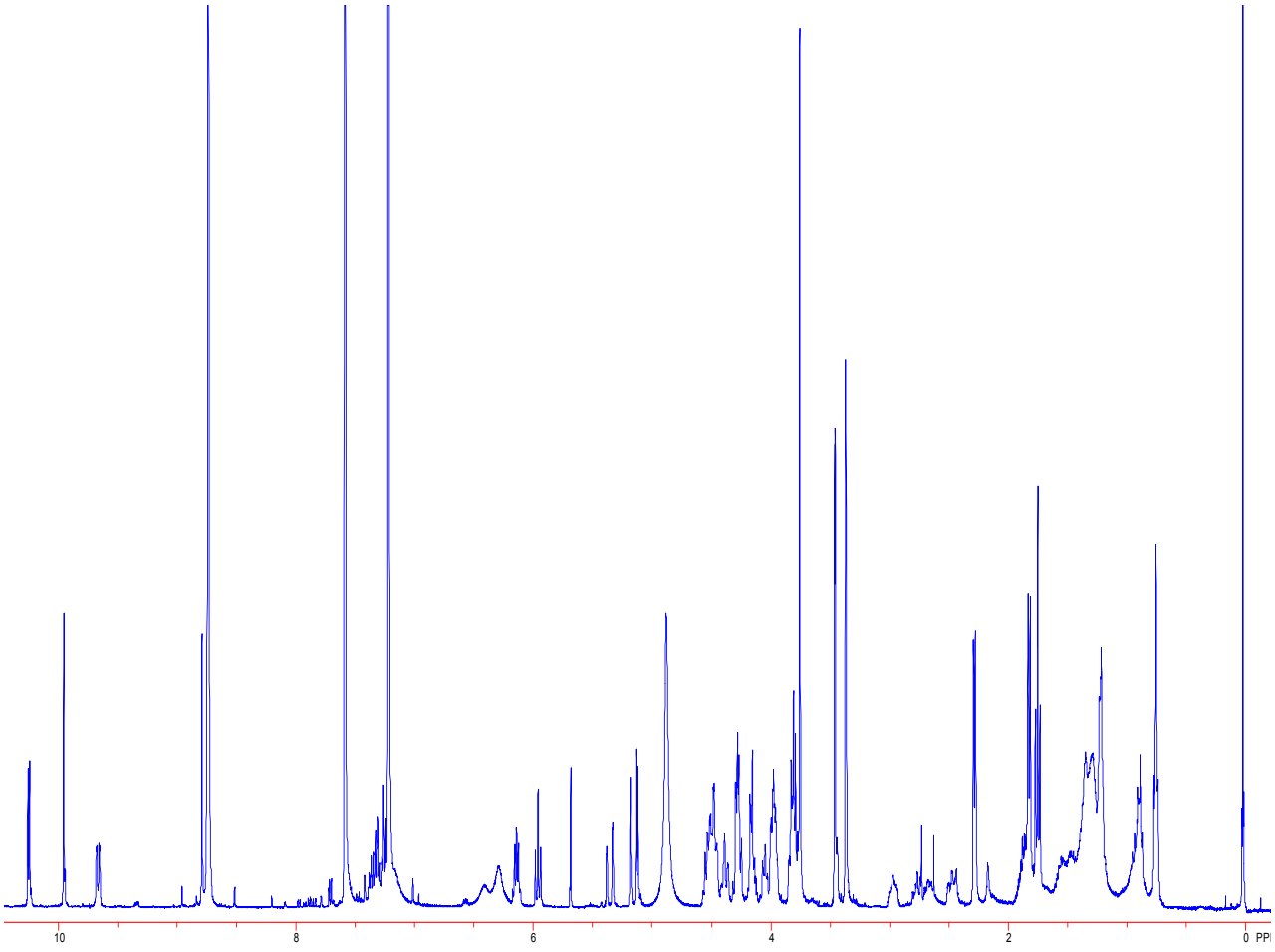

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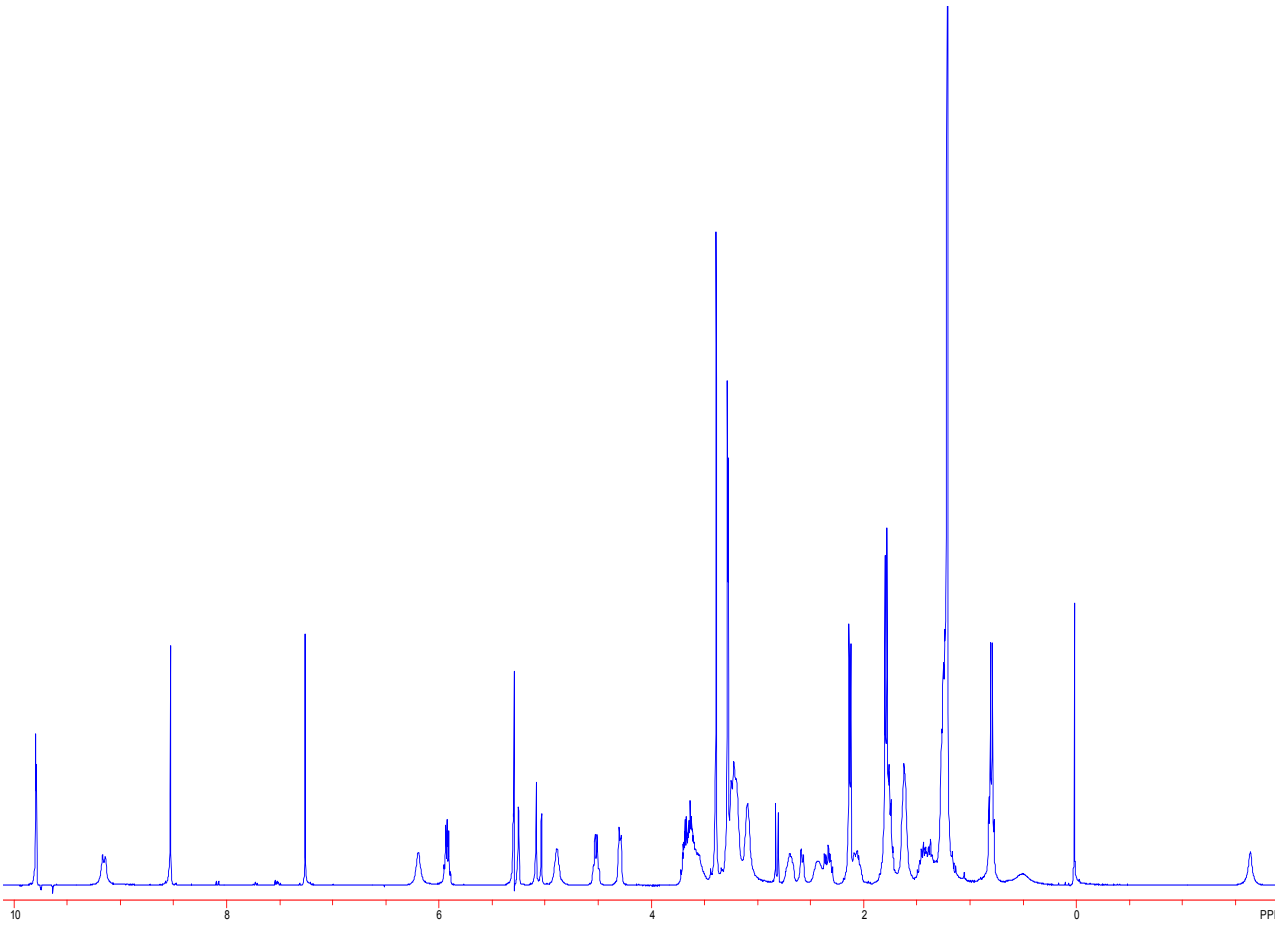

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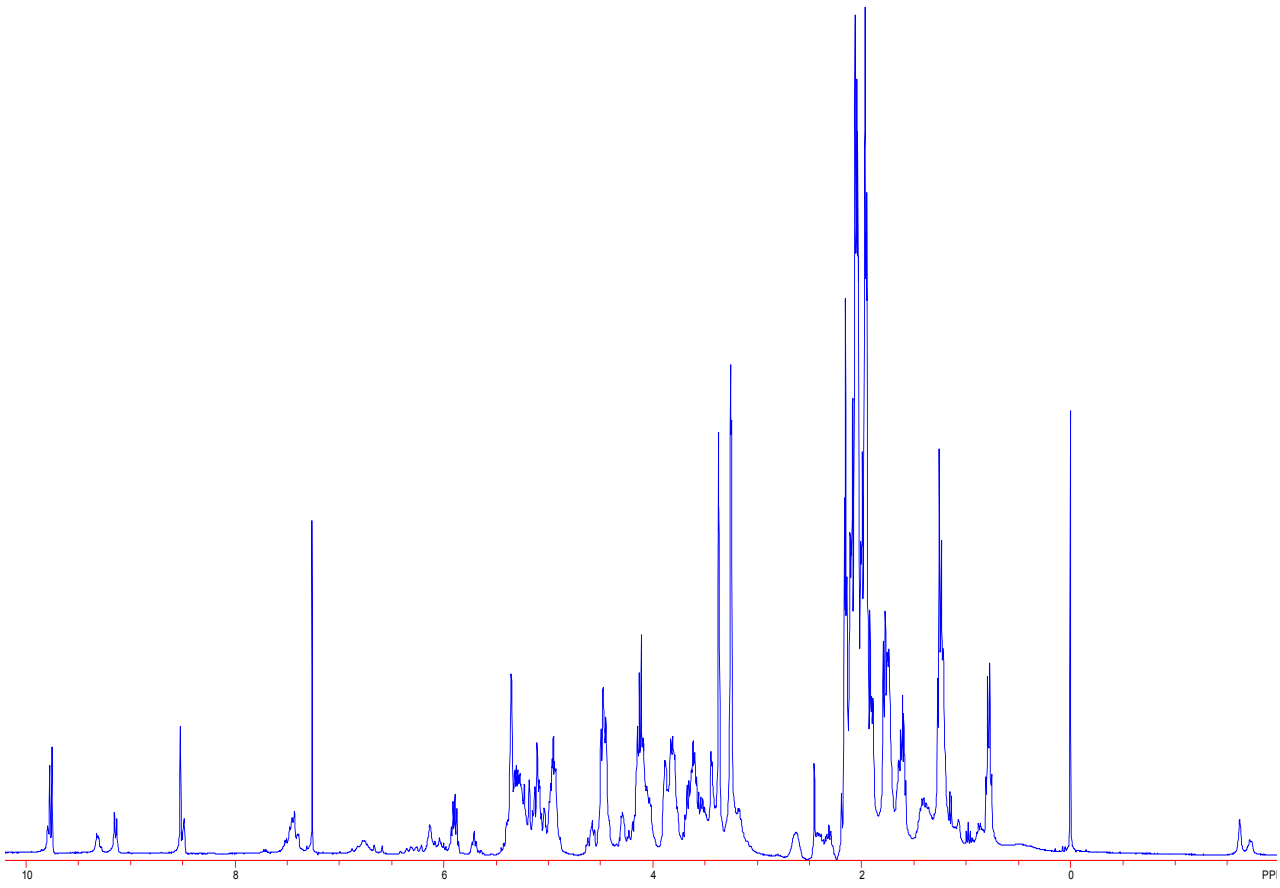

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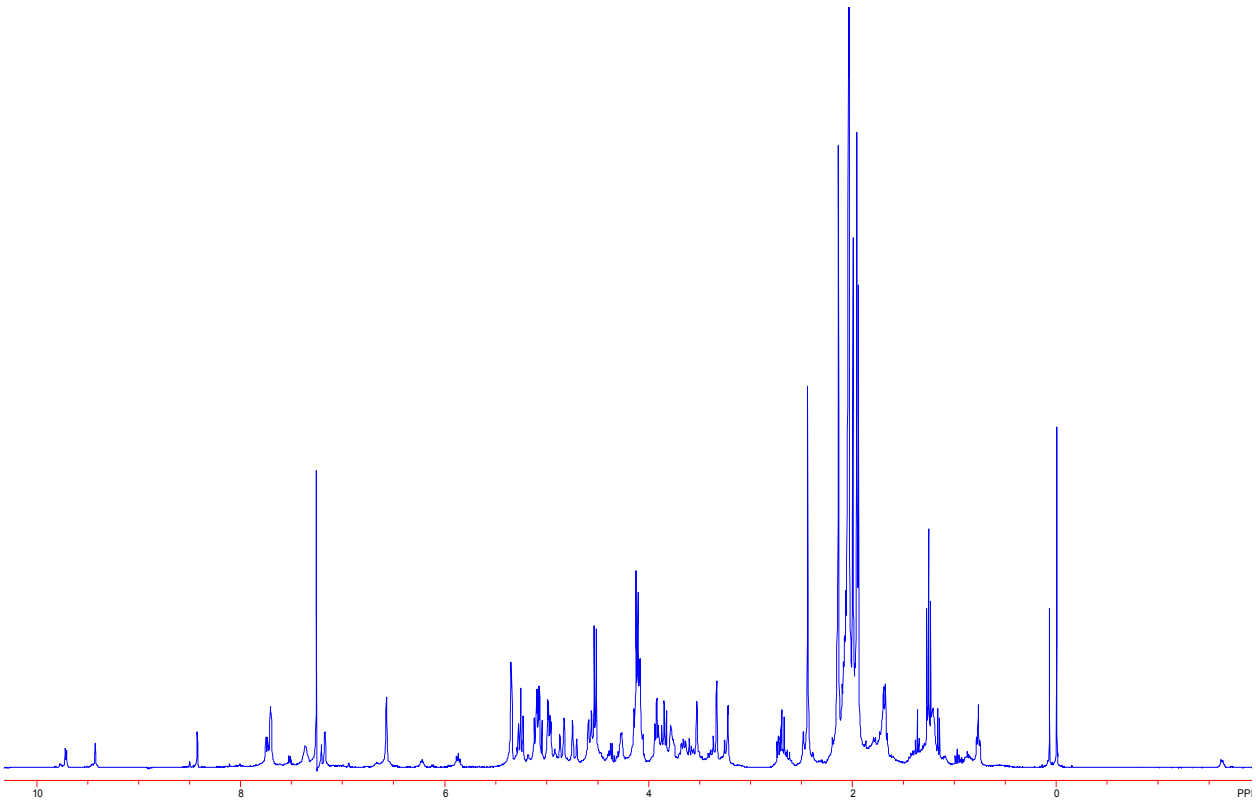

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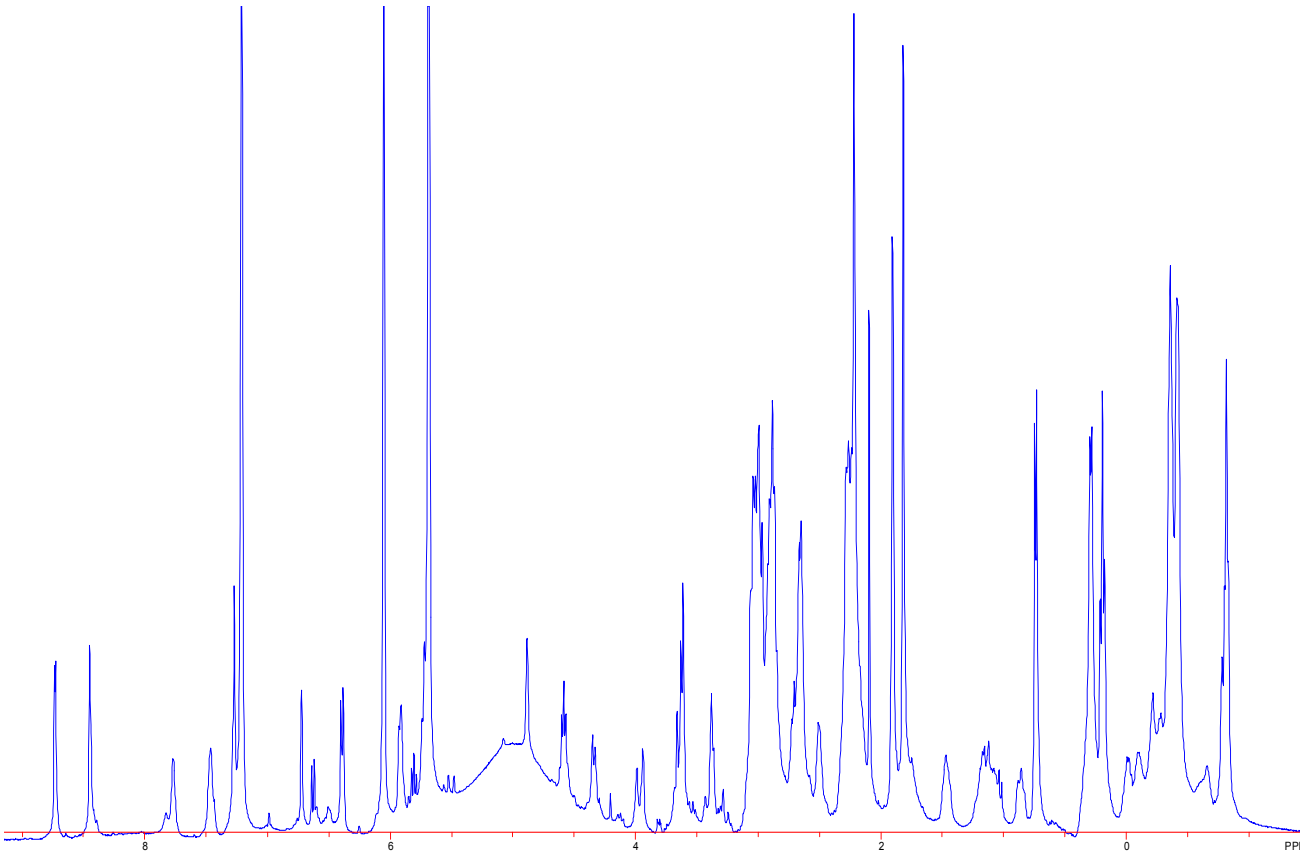

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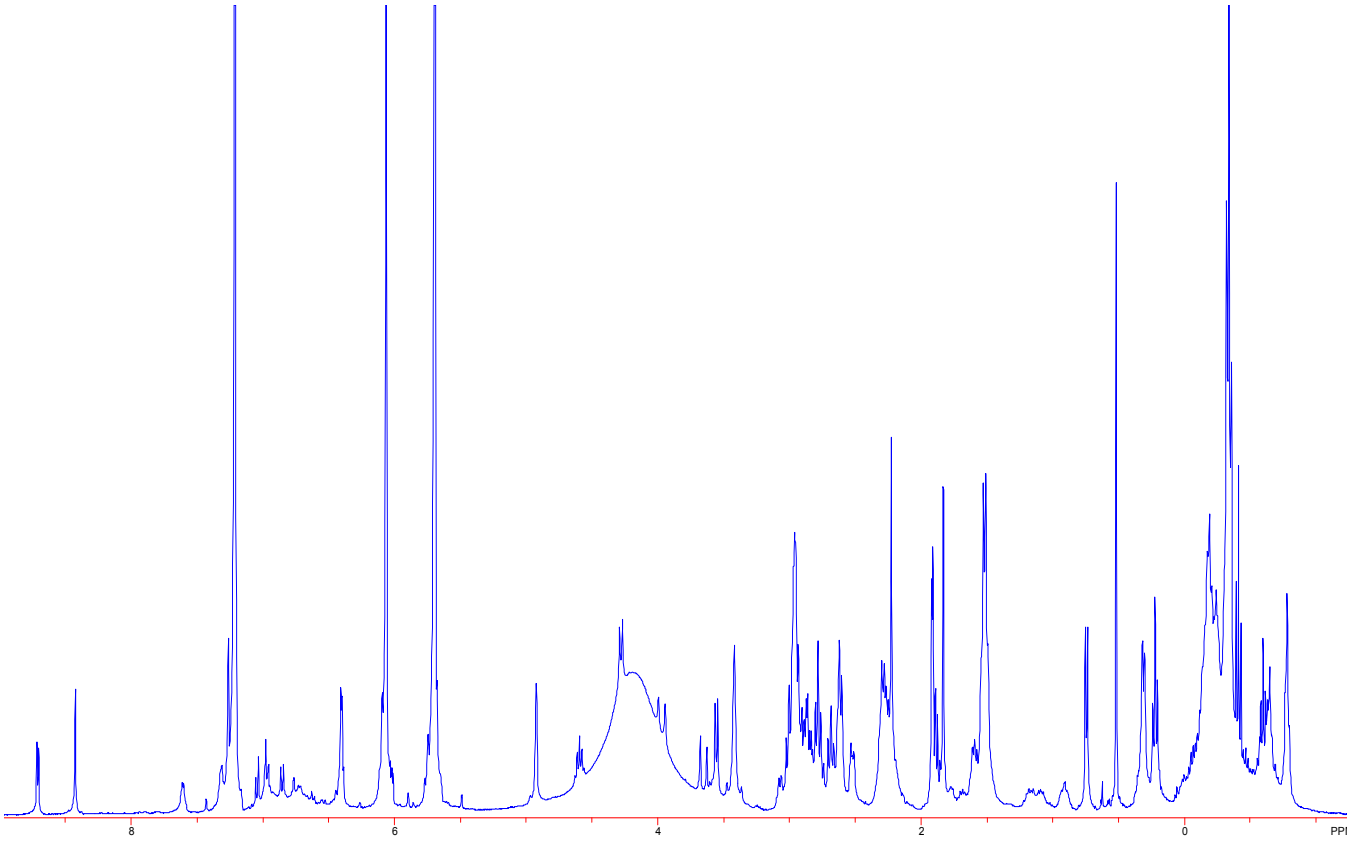


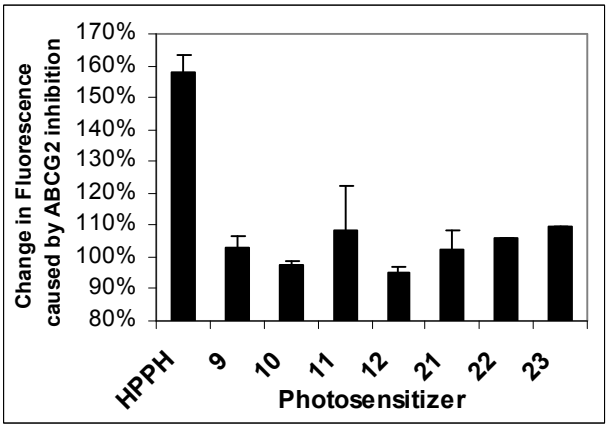


Figure 1. Effect of an inhibitor of ABCG2 activity (imatinib mesylate) on retention of HPPH and HPPH-carbohydrate conjugates in RIF cells. Inhibitor 10 μM or PBS was added to cells in medium containing 2% FCS at for 30 minutes at 37^{o}C followed by HPPH or the conjugates at 1 μM for 1 h at 37^{o}C. Cells were placed on ice and fluorescence measured by flow cytometry. The size of the increase in fluorescence in the presence of inhibitor indicates relative substrate specificity forABCG2.

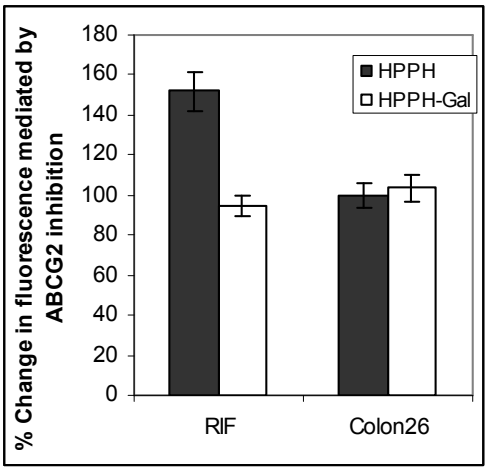
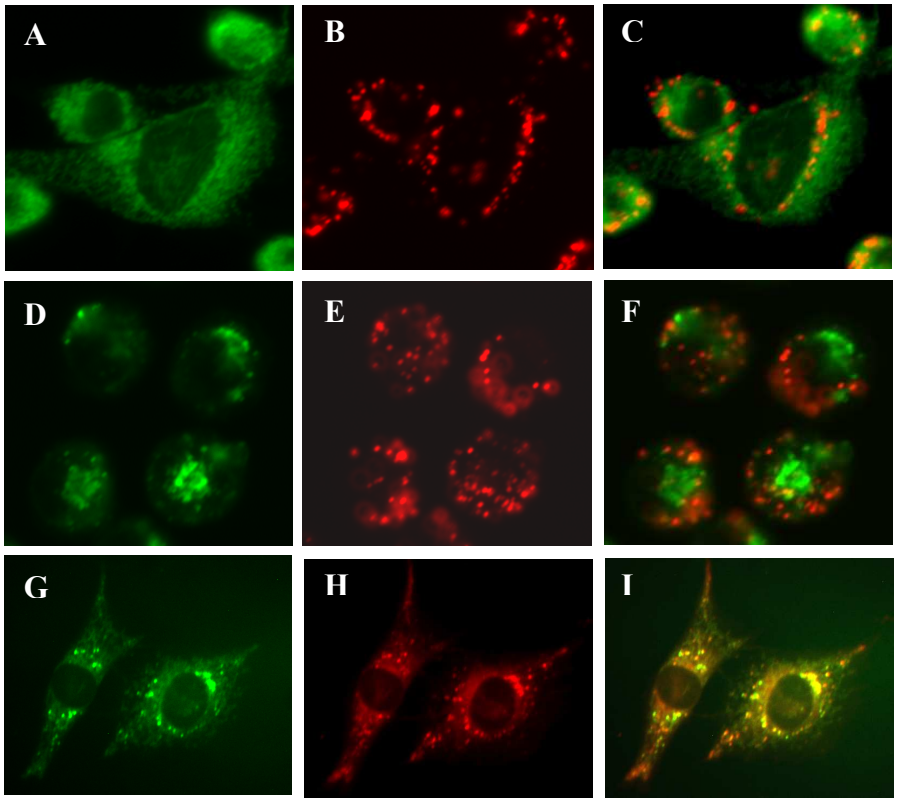


Figure 2. Comparative effects of ABCG2 inhibitor (imatinib mesylate) on retention of HPPH or HPPH-Gal in RIF and Colon26 cells. No change observed for either HPPH or HPPH Gal in the ABCG2 negative Colon26 cells.

Figure 3: Comparative intracellular localization of HPPH-carbohydrate conjugates with MitoTracker Green (a mitochondrial probe), Bodipy C5 ceramide (a golgi apparatus probe) and Lyso Tracker Green (a lysosomal probe) respectively in RIF cells: **A**) MitoTracker Green; **B**) PS conjugate; **C**) A and B merged; **D**) Bodipy C5 ceramide; **E**) PS conjugate; **F**) D and E merged; **G**) Lyso Tracker; **H**) PS conjugate; **I**) G and H merged

(i) Figure 3a:

9 (HPPH-Galactose) – 24 h incubation



(ii) Figure 3b:

10 (HPPH-Glucose) – 24 h incubation

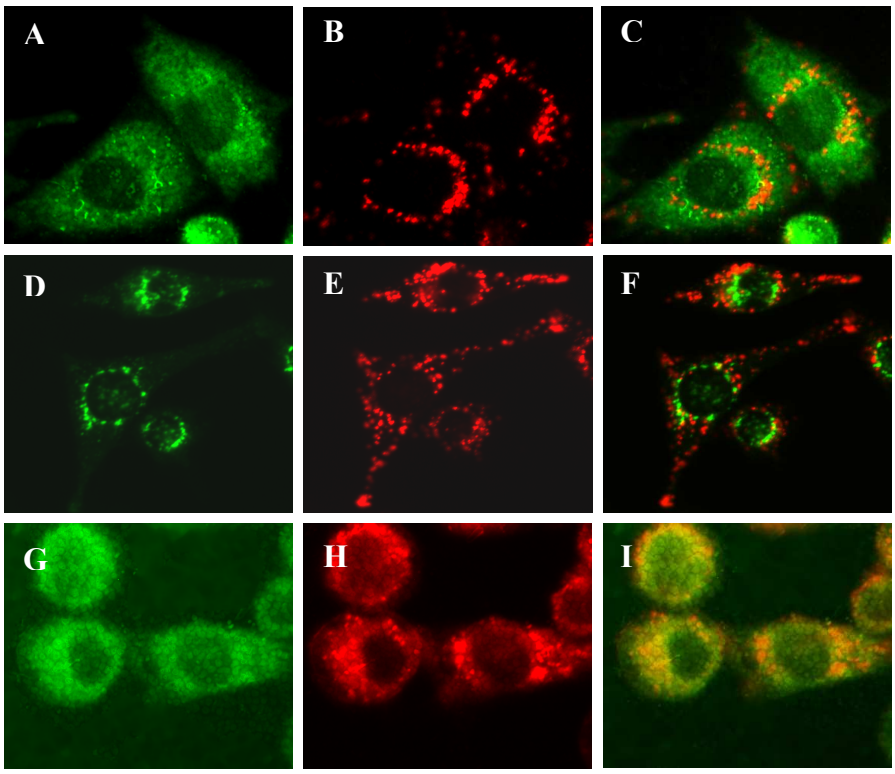


Figure 3c:

11 (HPPH-Lactose) – 24 h incubation

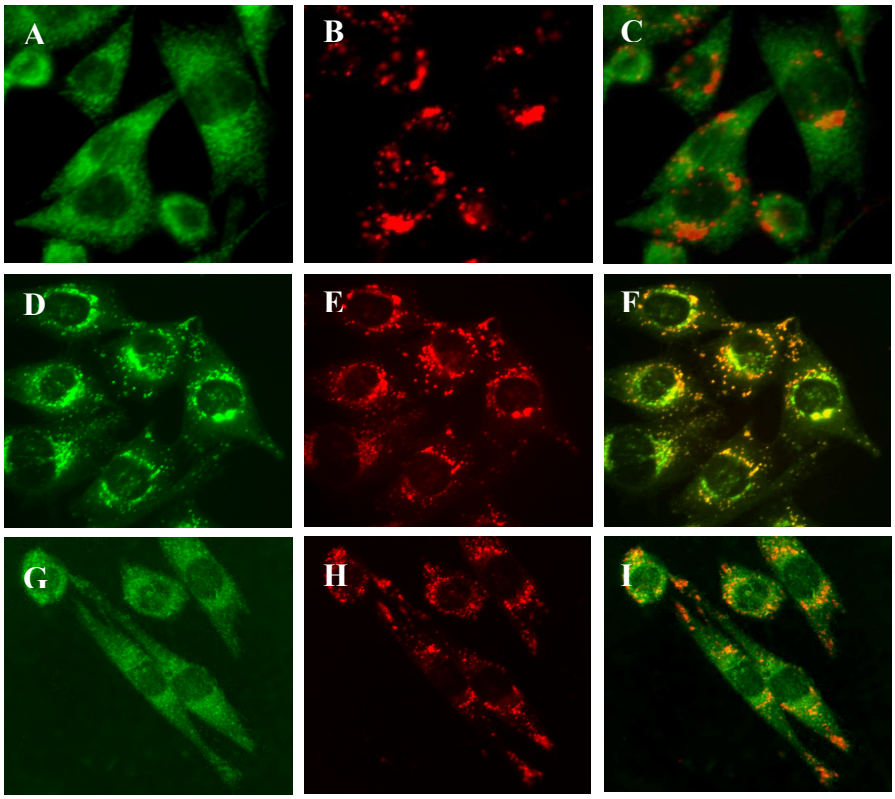


Figure 3d:

12 (HPPH-Cellobiose) – 24 h incubation

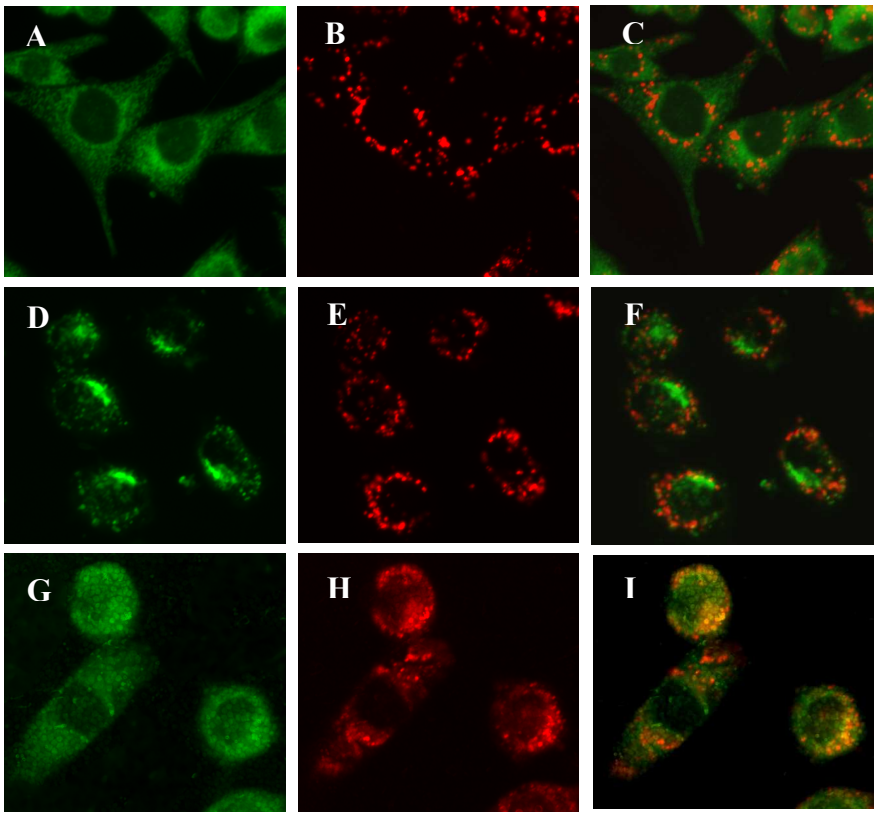


Figure 3e:

21 (HPPH-Monolactose) – 24 h incubation

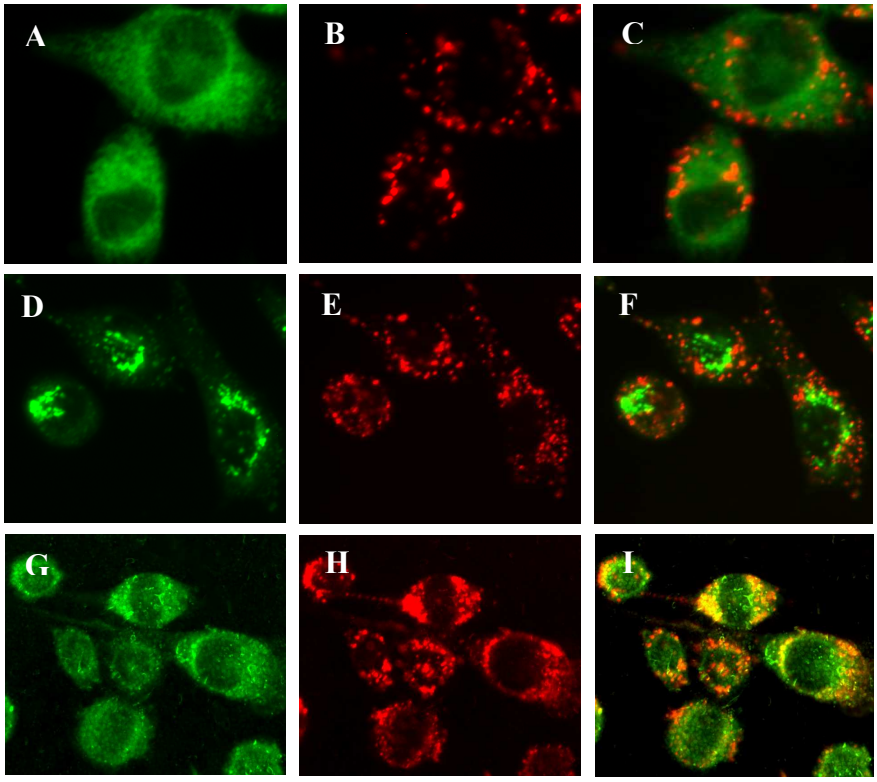


Figure 3f:

22 (HPPH-Dilactose) – 24 h incubation

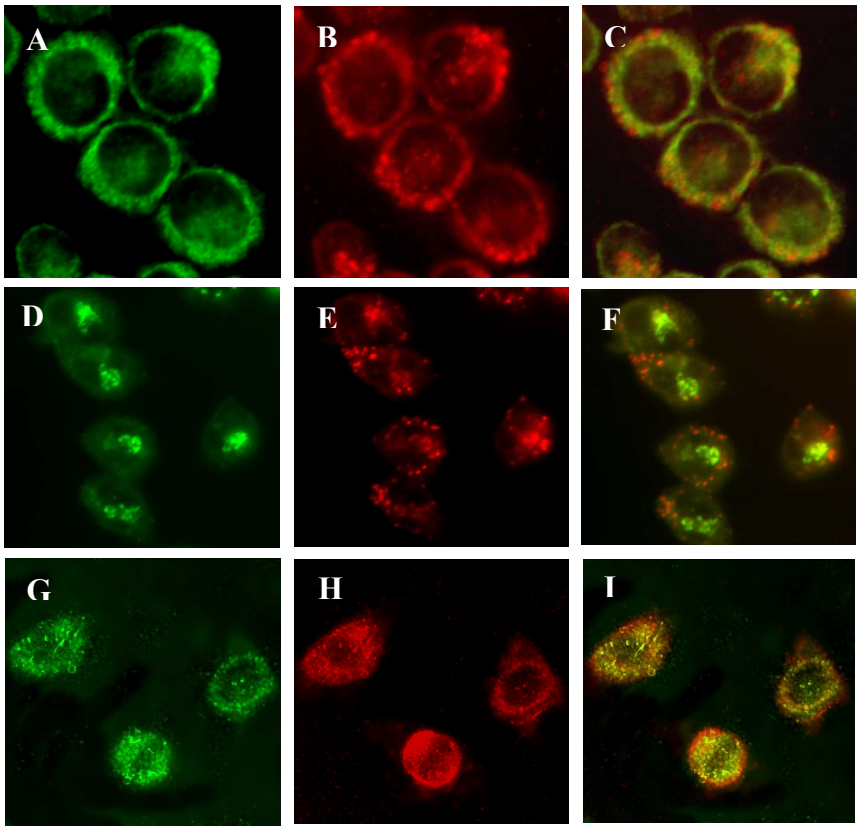


Figure 3g;

23 (HPPH-Tetralactose) – 24 h incubation

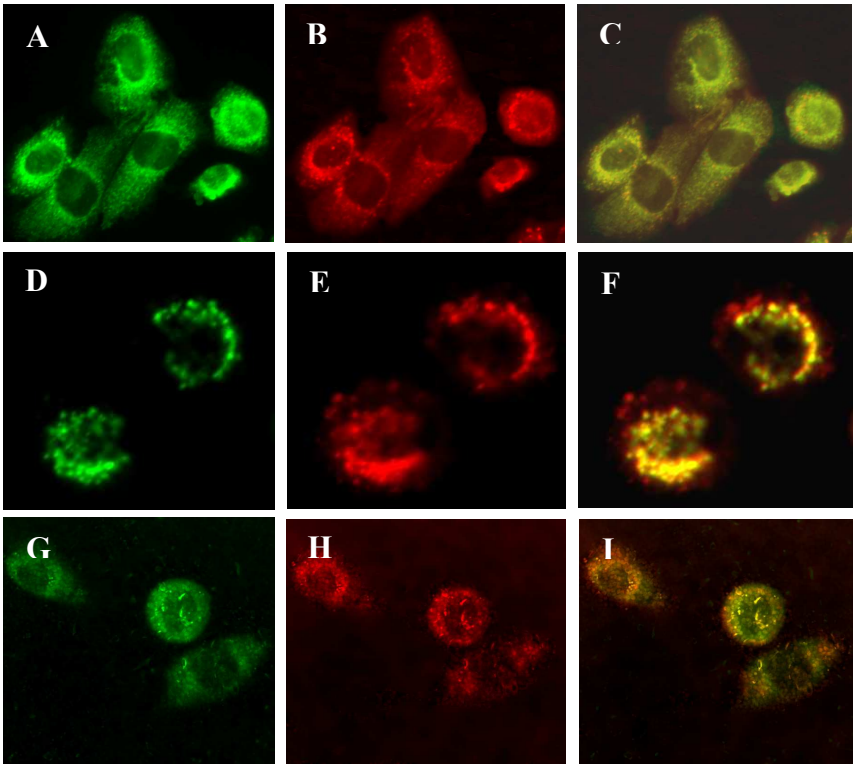


Table 1 Intracellular localization of **HPPH** and its carbohydrate conjugates

Compound	HPPH	9 (Gal)	9 (Gal)	10 (Glc)	11 (Lac)	12 (Cel)	21 (Monolac)	22 (Dilac)	23 (Tetralac)
Incubation time (h)	**24**	**3**	**24**	**24**	**24**	**24**	**24**	**24**	**24**
Mitochondria	+	-	-	-	-	-	-	+	+
Golgi apparatus	-	-	-	-	+	-	-	+	+
Lysosome	-	+	+	+	-	+	+	+	+

Table 2: HPLC analysis of the final products*

Compound	Retention time (min)	Purity (%)
HPPH-Galactose **9**	11.15	97.5
HPPH-Glucose **10**	10.61	97.9
HPPH-Lactose **11**	9.84	97.6
HPPH-Cellobiose **12**	10.26	96.3
HPPH-Regid Monolactose **21**	7.75	97

* Under the HPLC conditions (see the experimental section), compounds **22** and **23** retained in the column for a long time. However, as the corresponding acetoxy-analogs these compounds were >95% pure.