Supplementary Information

Evaluation of pH and concentration effects on antioxidant and pro-oxidant activities of lysine- and lysine methyl ester-based antioxidant tetramers, each containing four units of either syringaldehyde or vanillin

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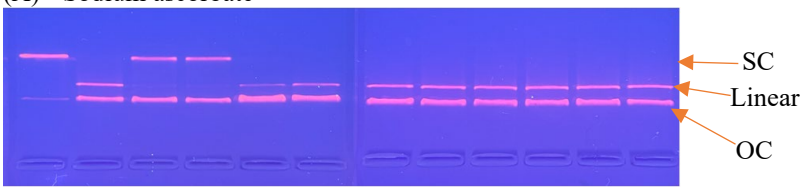
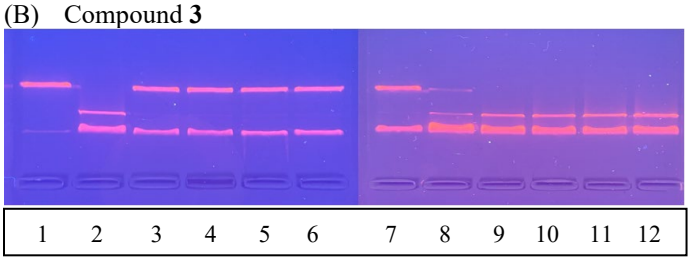
Note: In all gels, SC stands for supercoiled native DNA, OC refers to open circular damaged DNA, and Linear denotes the cleaved linear form of DNA.

Hydrophilic Antioxidants

(A) Sodium ascorbate

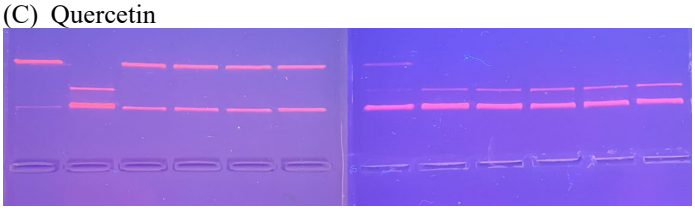


(B) Compound **3**

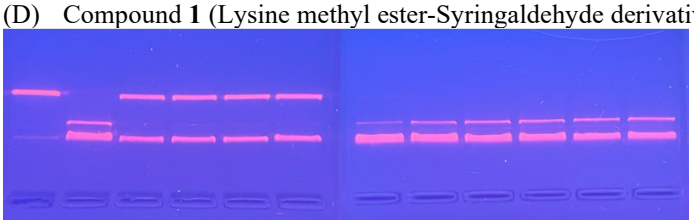


Hydrophobic Antioxidants

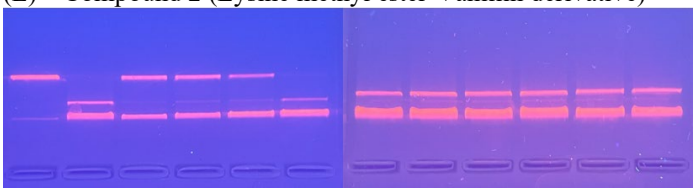
(C) Quercetin



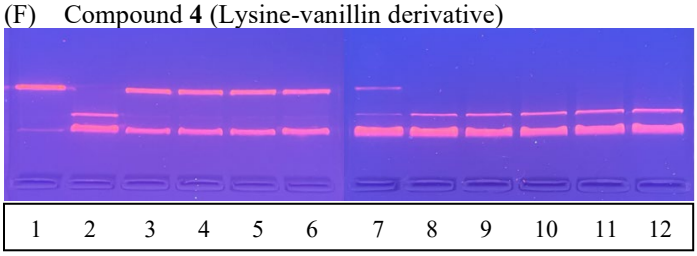
(D) Compound **1** (Lysine methyl ester-Syringaldehyde derivative)



(E) Compound **2** (Lysine methyl ester-Vanillin derivative)



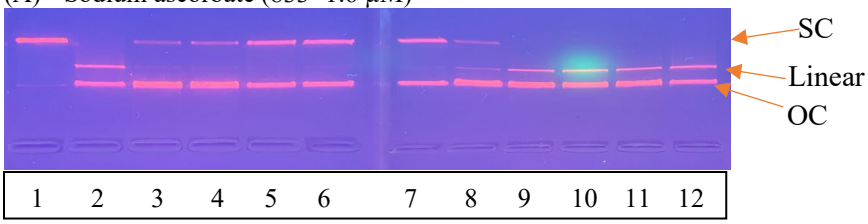
(F) Compound **4** (Lysine-vanillin derivative)



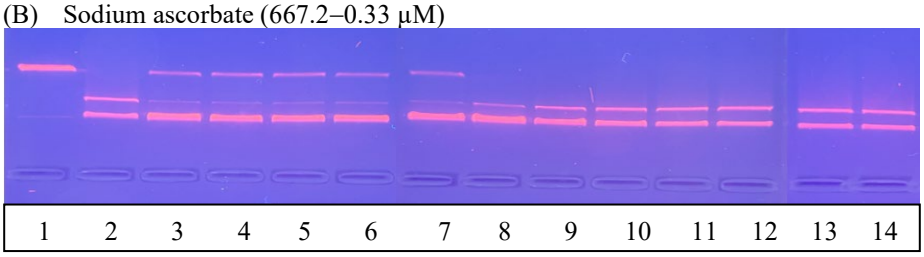
S1. Protection against AAPH-induced DNA oxidation by antioxidants (in PBS). Lane 1 (native DNA), lane 2 (AAPH + DNA, without antioxidant), and lanes 3–12 (AAPH + DNA with antioxidant at various concentrations: 83.3 µM, 41.7 µM, 20.8 µM, 10.4 µM, 5.2 µM, 2.6 µM, 1.3 µM, 0.65 µM, 0.33 µM, and 0.16 µM, respectively. (A) Sodium ascorbate; (B) Compound **3**; (C) Quercetin; (D) Compound **1**; (E) Compound **2**; (F) Compound **4**.

Hydrophilic Antioxidants

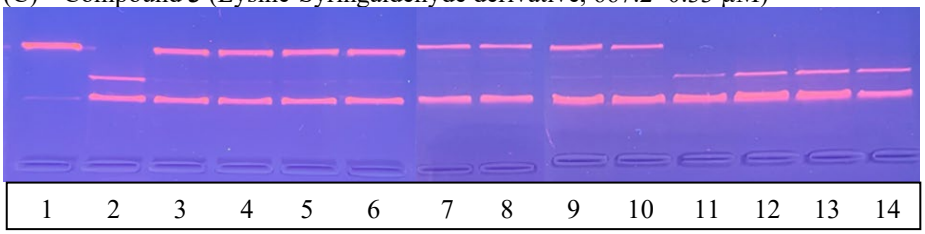
(A) Sodium ascorbate (833–1.6 µM)



(B) Sodium ascorbate (667.2–0.33 µM)

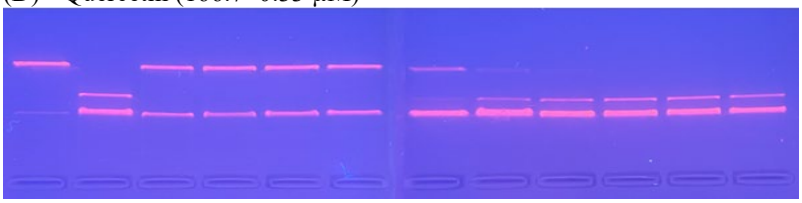


(C) Compound **3** (Lysine-Syringaldehyde derivative, 667.2–0.33 µM)

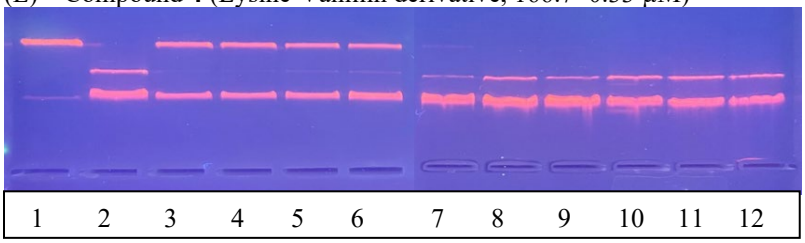


Hydrophobic Antioxidants

(D) Quercetin (166.7–0.33 µM)



(E) Compound **4** (Lysine-Vanillin derivative, 166.7–0.33 µM)



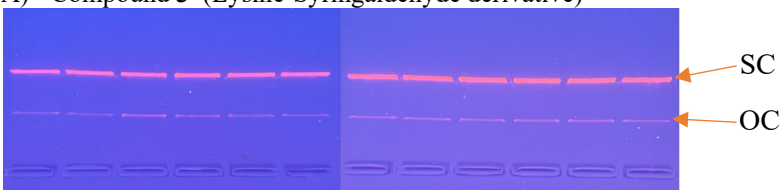
S2. Protection against AAPH-induced DNA oxidation in PBS at high concentrations. Lane 1 (native DNA), lane 2 (AAPH + DNA, without antioxidant), and lanes 3–12 (AAPH + DNA + antioxidant at various concentrations). (A) Sodium ascorbate; (B) Compound **3**; (C) Quercetin; (D) Compound **4**.

S3. The summary of pH effects on AO activities of hydrophilic and hydrophobic antioxidants, obtained from gel electrophoresis.

	Antioxidant	pH 4	pH 7	pH 9
Hydrophilic AO	Sodium ascorbate	Very slight protection at 41.7 μM	Good protection at 41.7 μM	Good protection at 41.7 μM Some protection at 20.8 μM.
	Compound **3**	Very slight protection at 41.7 μM	Slight protection in 41.7–10.4 μM	Good protection in 41.7–2.6 μM
Hydrophobic AO	Quercetin	No protection	Good protection in 41.7–10.4 μM Slight protection at 5.2 μM	Good protection in 41.7–5.2 μM
	Compound **1**	No protection	Good protection in 41.7–20.8 μM	Good protection in 41.7–10.4μM Slight protection at 5.2 μM
	Compound **2**	No protection	Good protection at 41.7μM Slight protection at 20.8 μM	Good protection in 41.7–20.8 μM Slight protection in 10.4–5.2 μM
	Compound **4**	No protection	Good protection in 41.7 – 10.4 μM	Good protection in 41.7–5.2 μM Some protection in 2.6–1.3 μM.

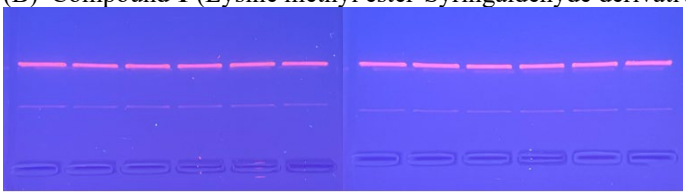
Hydrophilic Antioxidants

A) Compound **3** (Lysine-Syringaldehyde derivative)

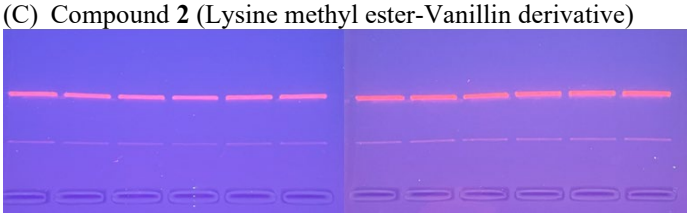


Hydrophobic Antioxidants

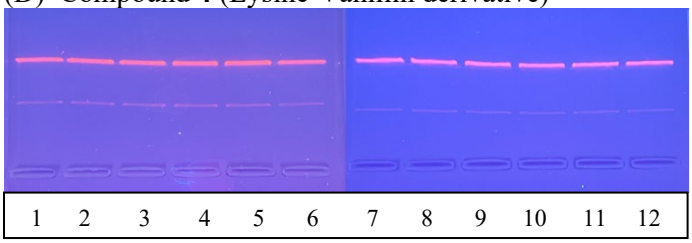
(B) Compound **1** (Lysine methyl ester-Syringaldehyde derivative)



(C) Compound **2** (Lysine methyl ester-Vanillin derivative)

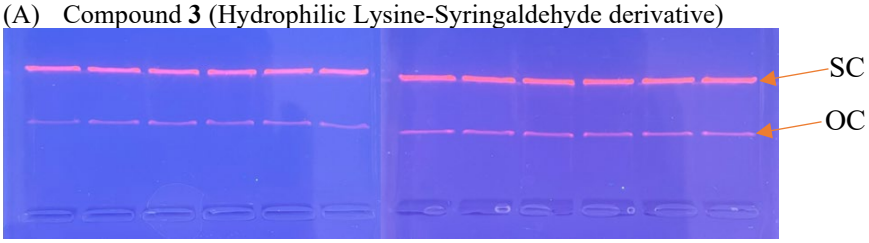


(D) Compound **4** (Lysine-Vanillin derivative)

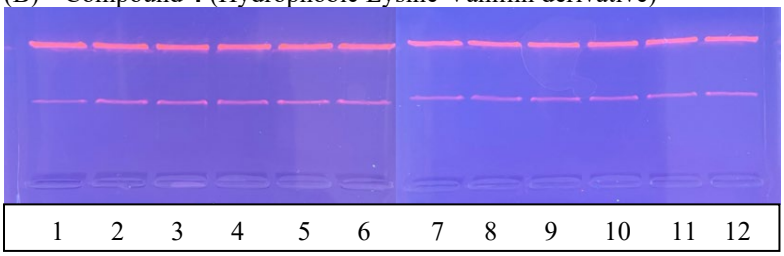


S4. Pro-oxidant effects of tetrameric antioxidants (in PBS).
(A) Lane 1 (native DNA), lane 2 (native DNA + Cu(II) ions, without antioxidant), and lanes 3–12 contain native DNA + Cu(II) ions + antioxidant at various concentrations: 83.3 µM, 41.7 µM, 20.8 µM, 10.4 µM, 5.2 µM, 2.6 µM, 1.3 µM, 0.65 µM, 0.33 µM, and 0.16 µM, respectively. (A) Compound **3**; (B) Compound **1**; (C) Compound **2**; (D) Compound **4**.

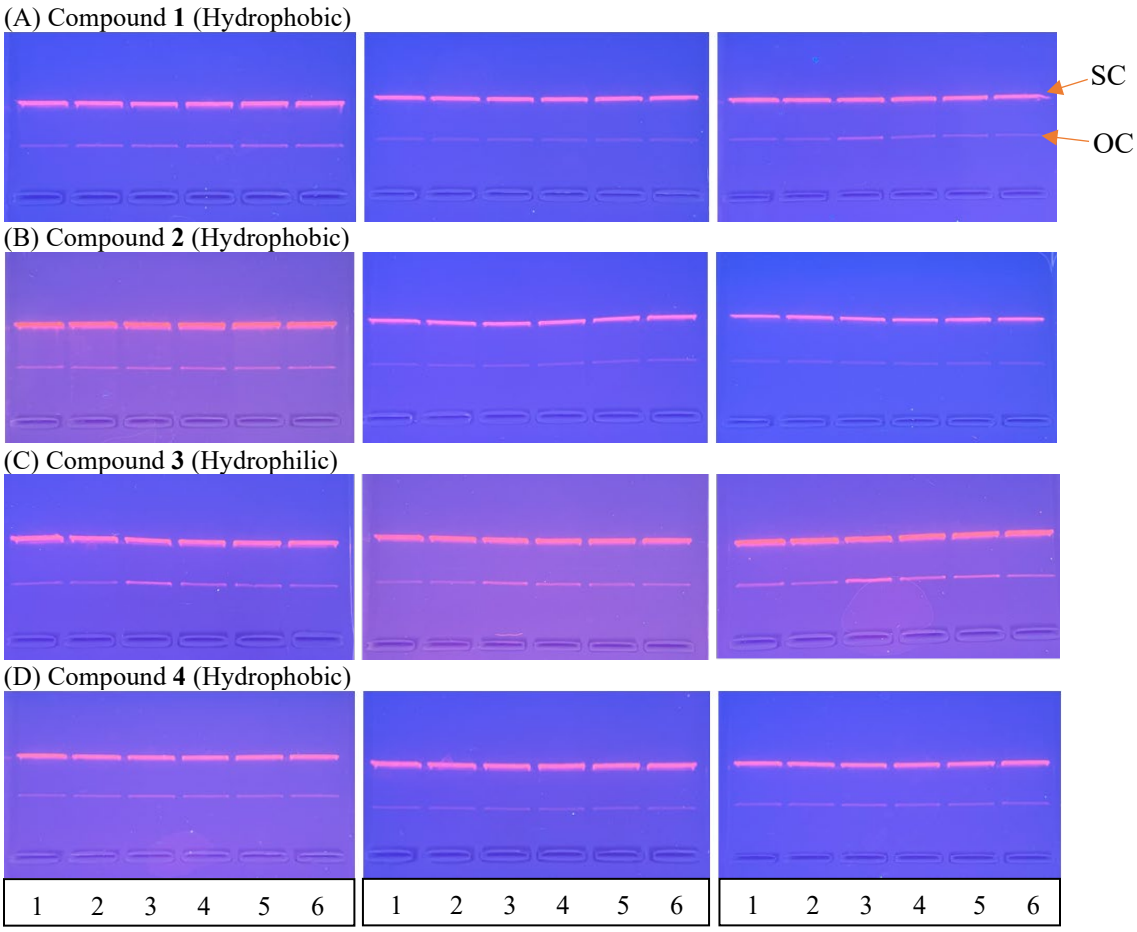
(A) Compound **3** (Hydrophilic Lysine-Syringaldehyde derivative)



(B) Compound **4** (Hydrophobic Lysine-Vanillin derivative)



S5. Pro-oxidant effects at high concentrations (166.7 µM–0.33 µM) in PBS. Lane 1 (native DNA), lane 2 (native DNA + Cu(II) ions, without antioxidant), and lanes 3–12 contain native DNA + Cu(II) ions + antioxidant at various concentrations: 166.7 µM, 83.3 µM, 41.7 µM, 20.8 µM, 10.4 µM, 5.2 µM, 2.6 µM, 1.3 µM, 0.65 µM, and 0.33 µM, respectively. (A) Compound **3**; (B) Compound **4**.

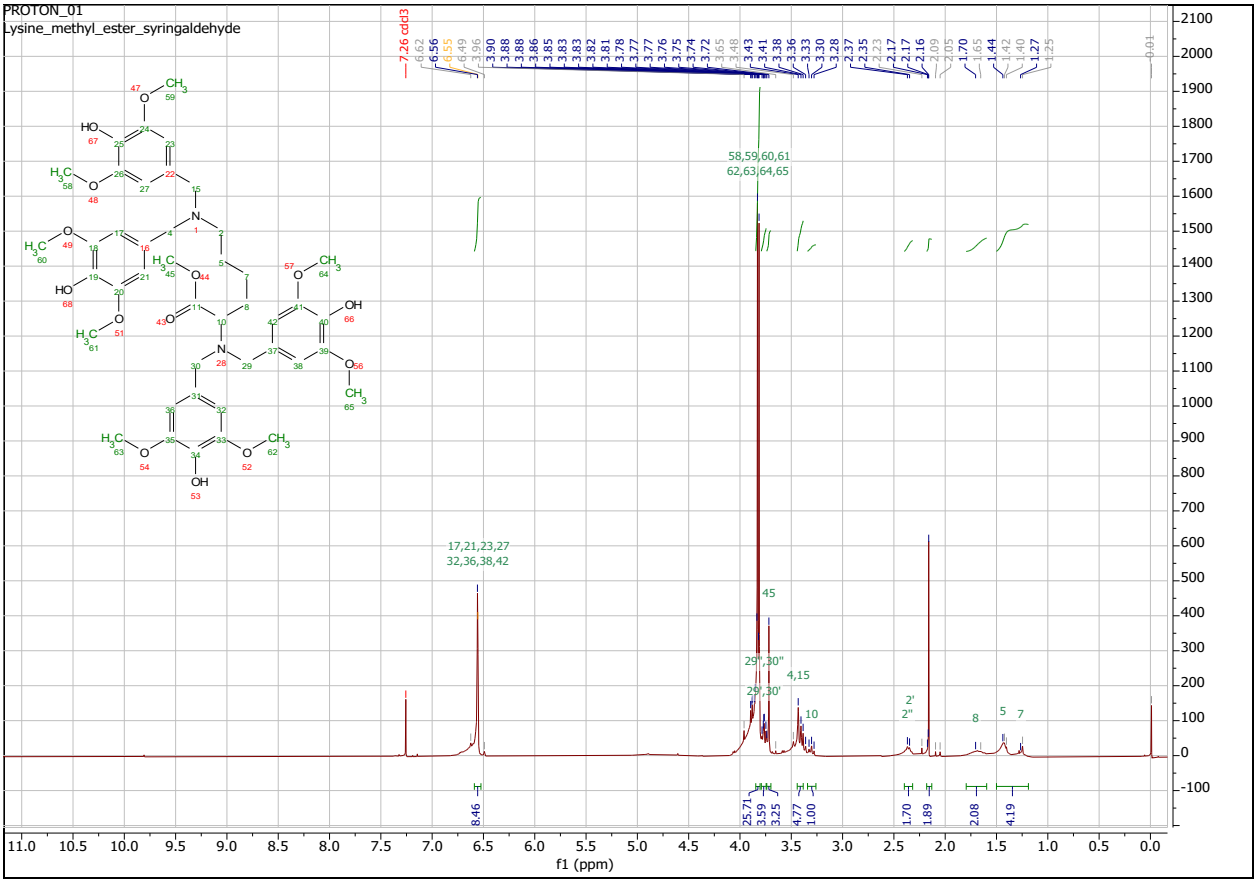


S6. Effects of pH on the pro-oxidant activities of tetrameric antioxidants. All controls and antioxidants contain 1% DMSO. The left panels show samples at pH 4, the middle panels at pH 7, and the right panels at pH 9. In each panel, lane 1 contains native DNA and lane 2 contains DNA + Cu(II) ions. Lanes 3-6 contain DNA + Cu(II) ions + antioxidants at various concentrations: 41.7 µM, 20.8 µM, 10.4 µM, and 5.2 µM, respectively. (A) Compound **1**; (B) Compound **2**; (C) Compound **3**; (D) Compound **4**.

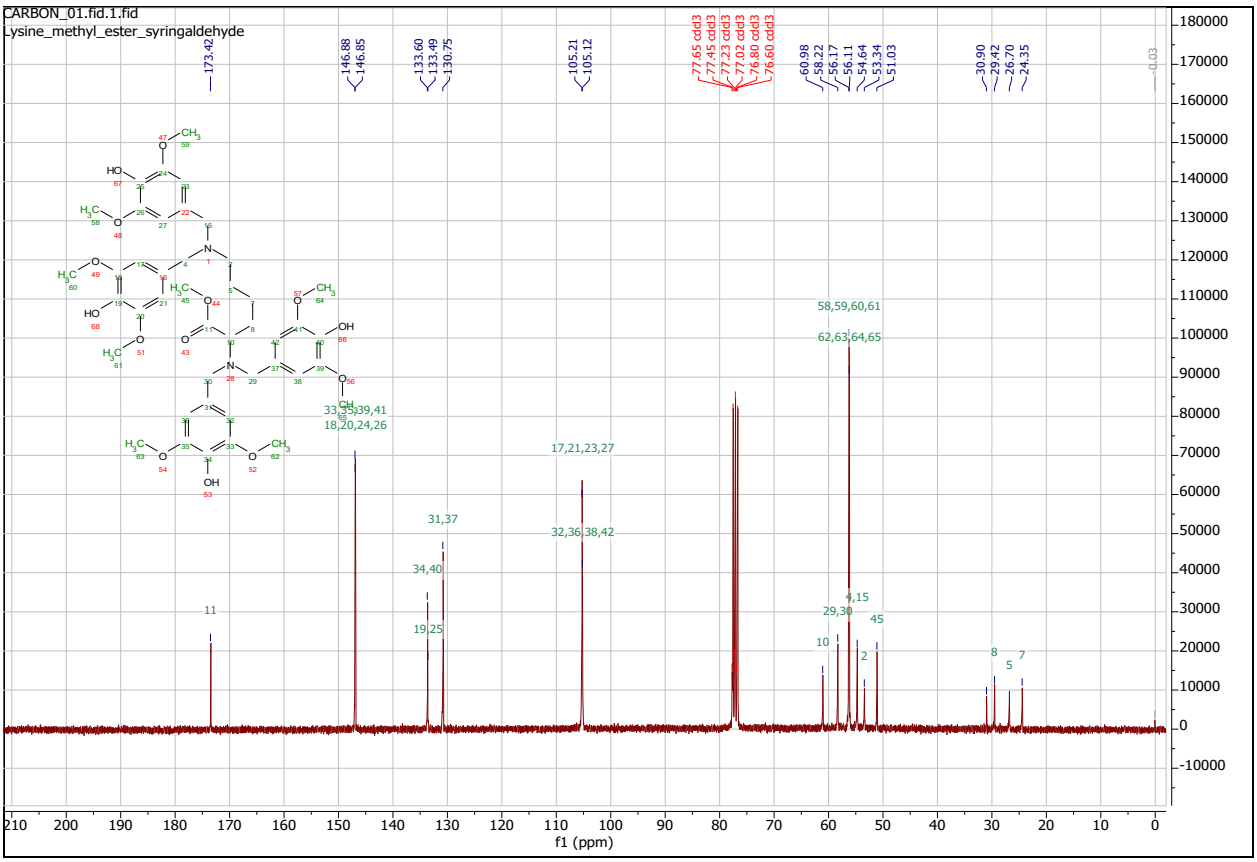
S7. ^{1}H NMR, ^{13}C NMR, and HR-MS of compounds **1**-**4**

Compound **1**: Lysine methyl ester-Syringaldehyde derivative

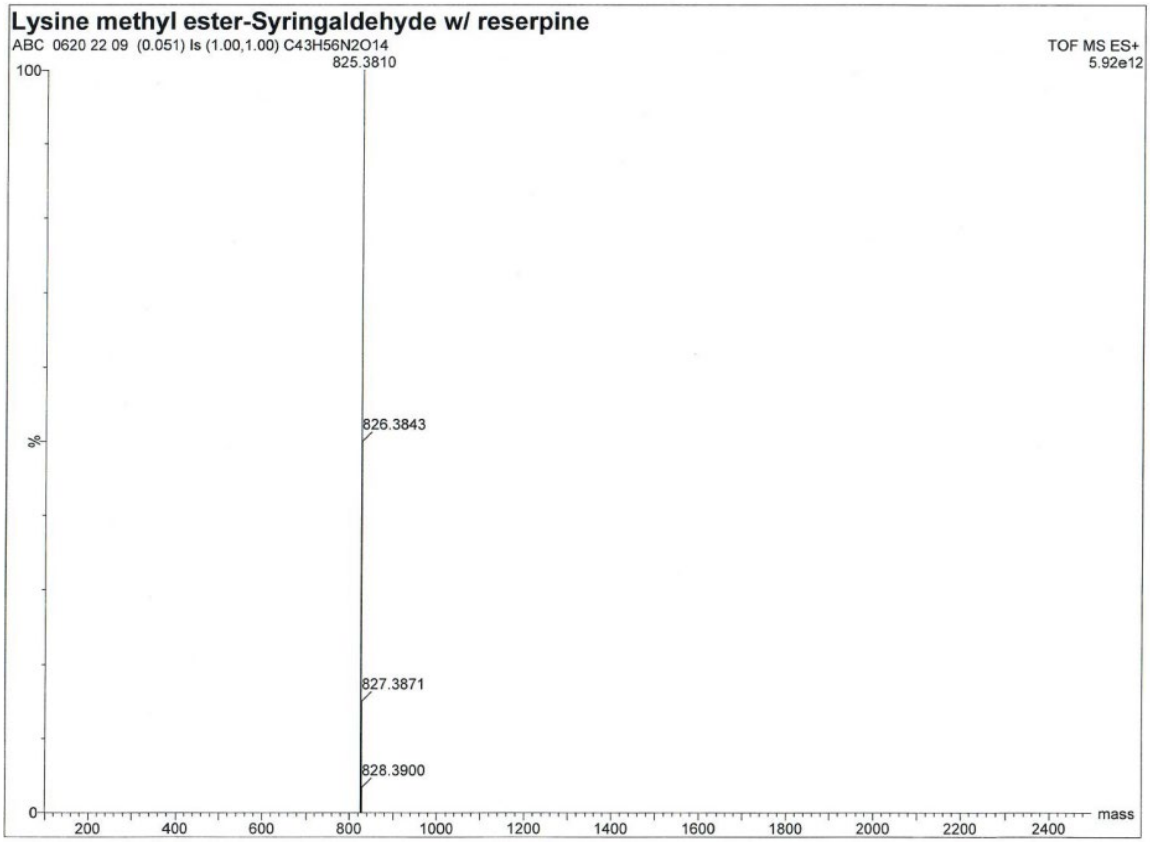
^{1}H NMR spectrum of Compound **1**



^{13}C NMR spectrum of Compound **1**

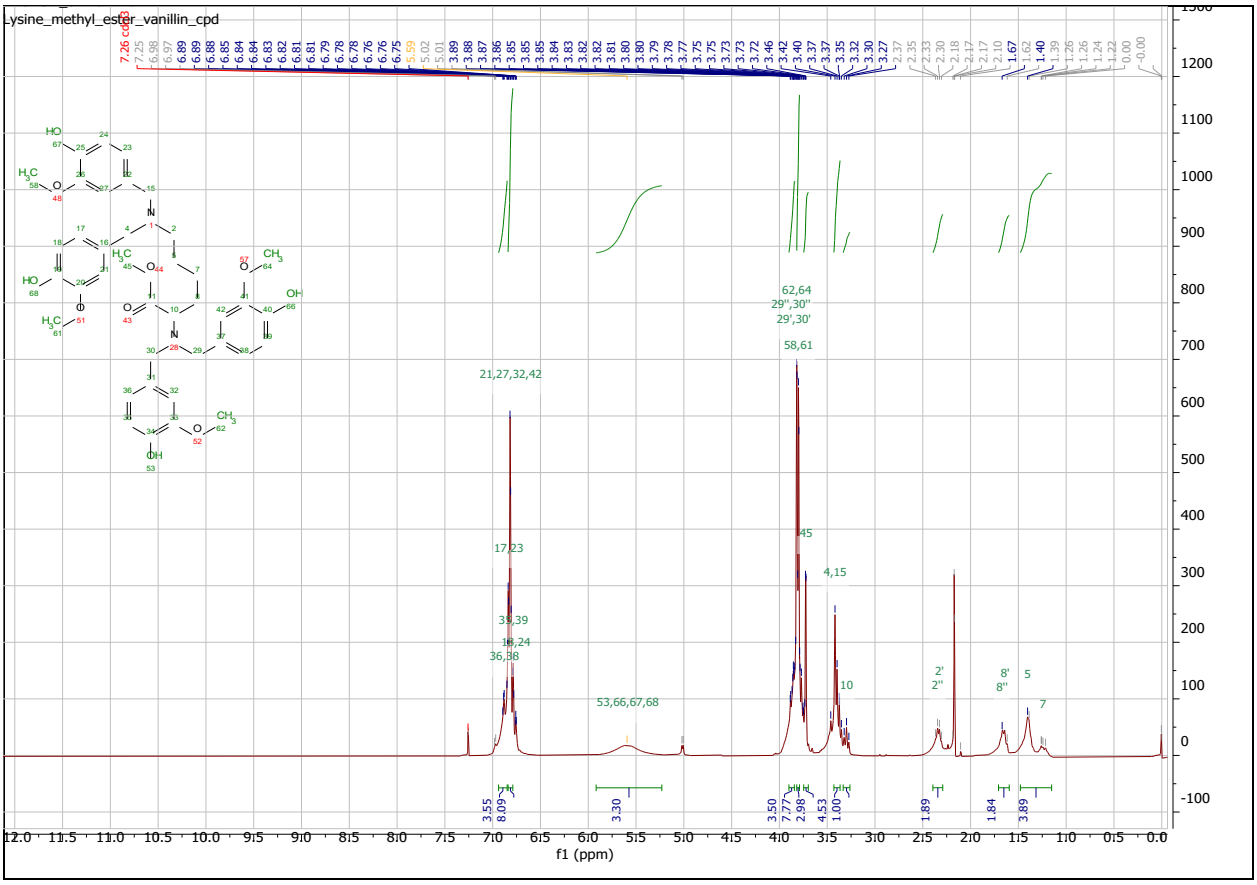


HR-MS of Compound **1**

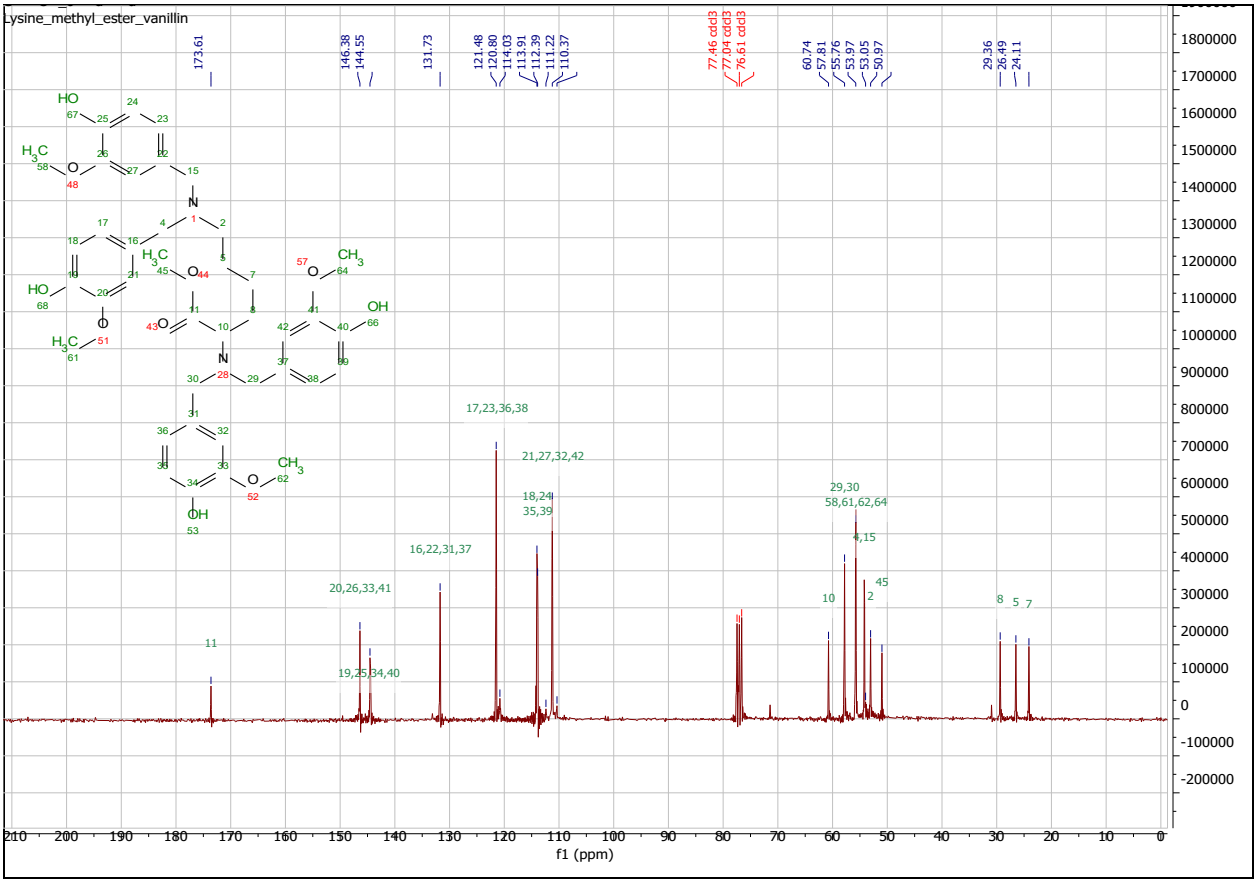


Compound **2**: Lysine methyl ester-Vanillin derivative

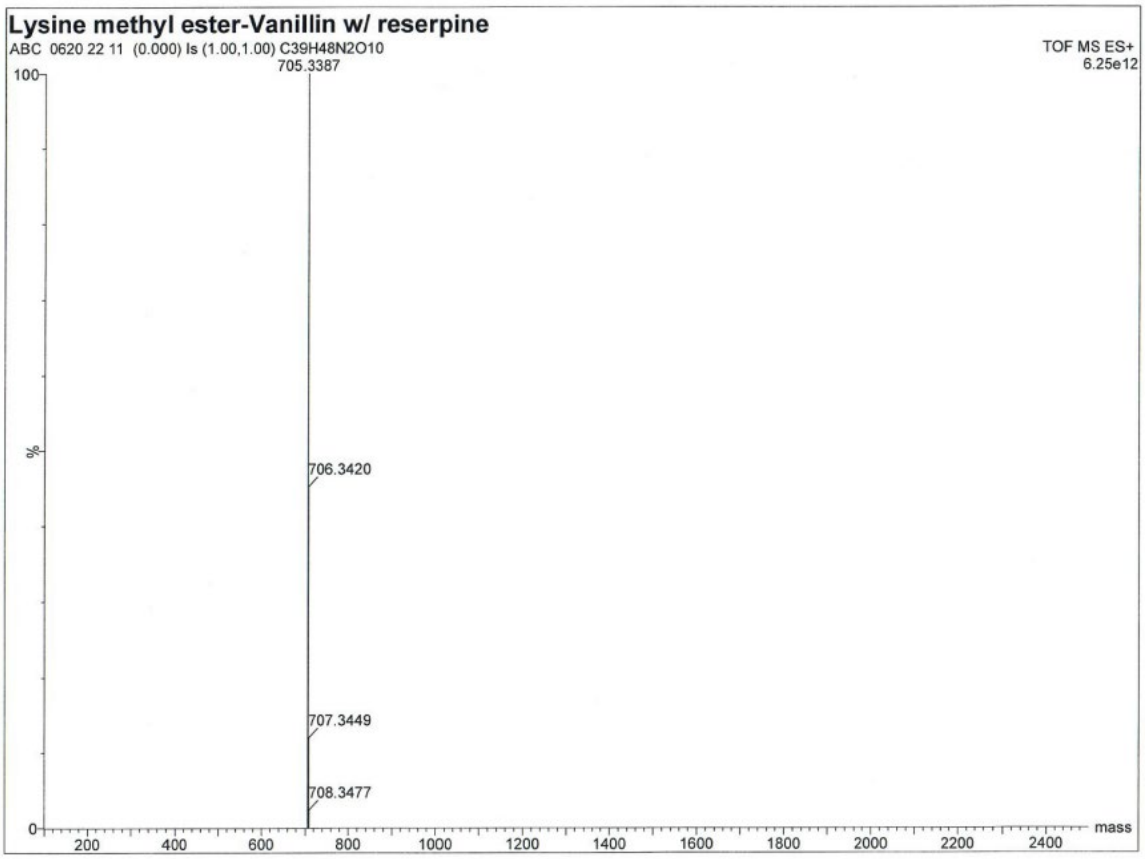
^{1}H NMR spectrum of Compound **2**



^{13}C NMR spectrum of Compound **2**

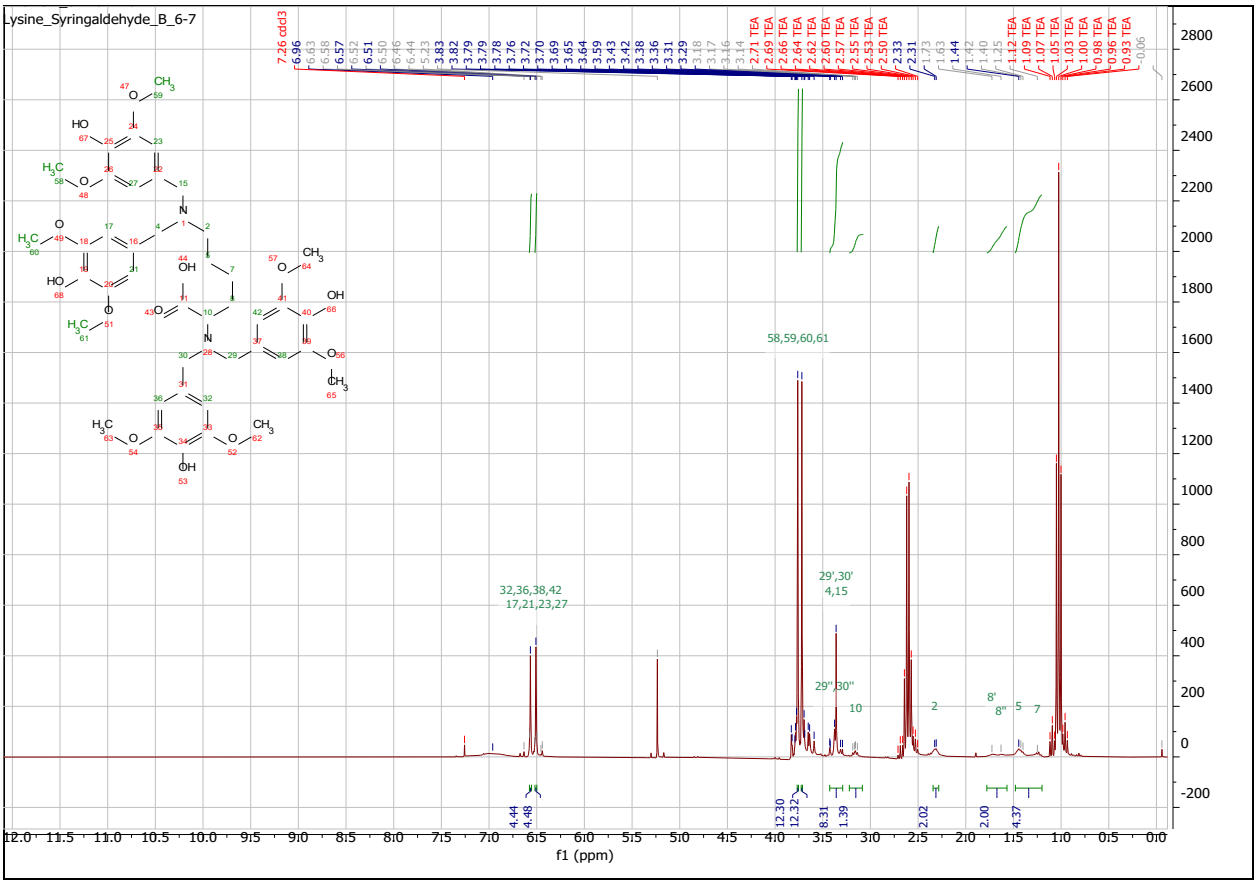


HR-MS of Compound **2**

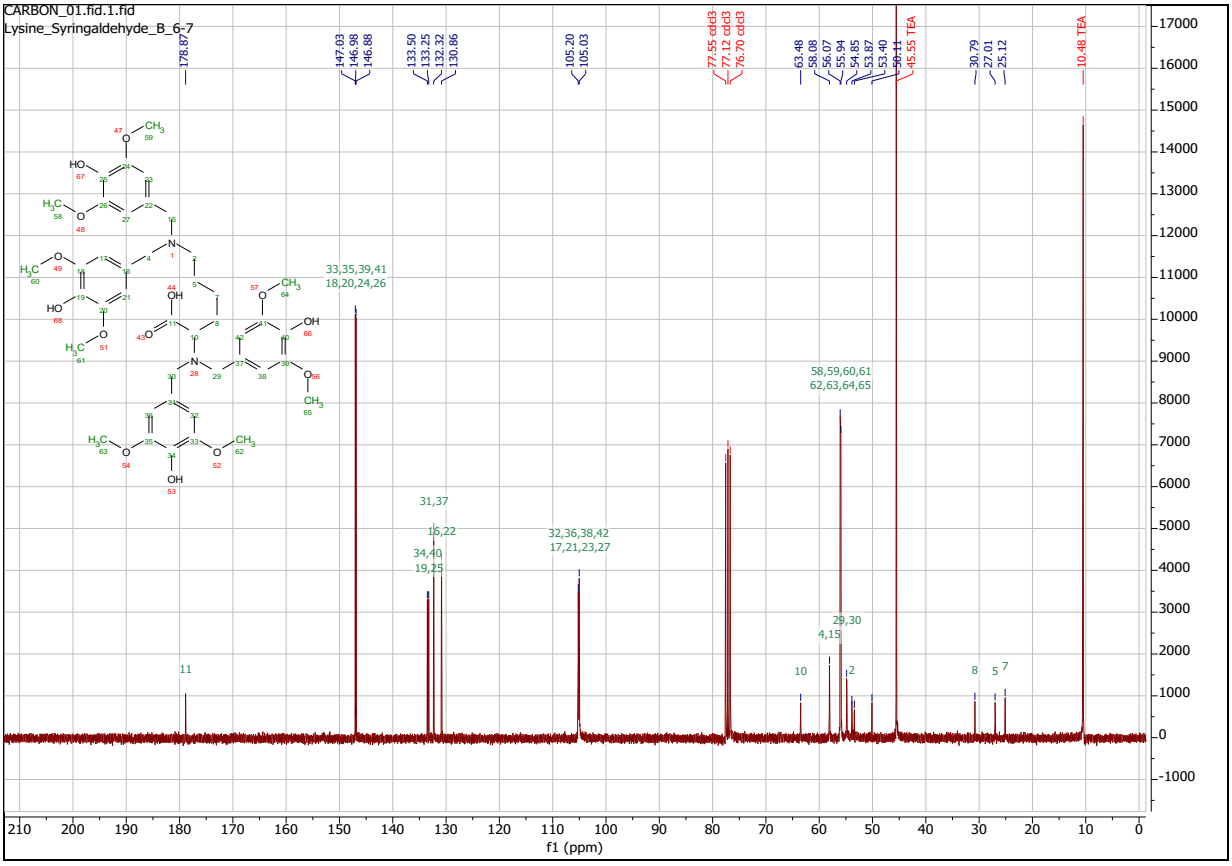


Compound **3**: Lysine-Syringaldehyde derivative

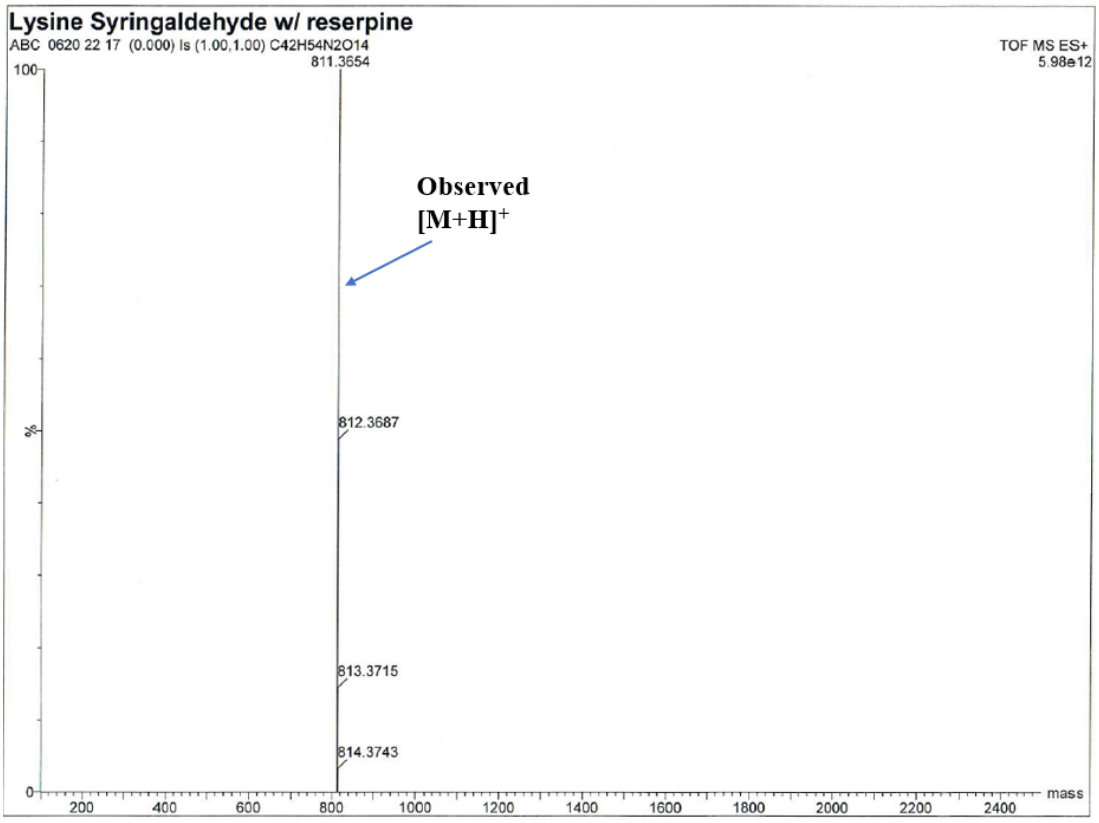
^{1}H NMR spectrum of Compound **3**



^{13}C NMR spectrum of Compound **3**

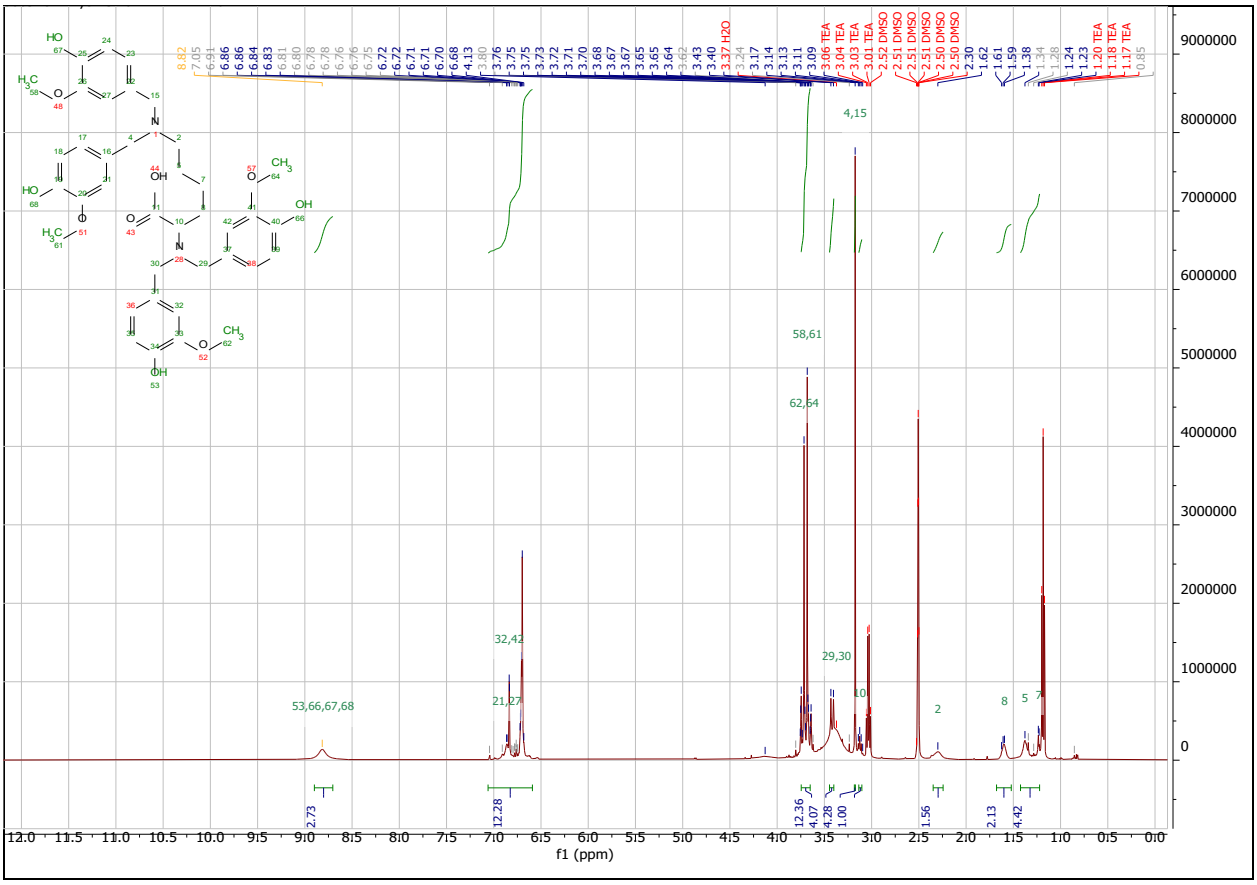


HR-MS of Compound **3**

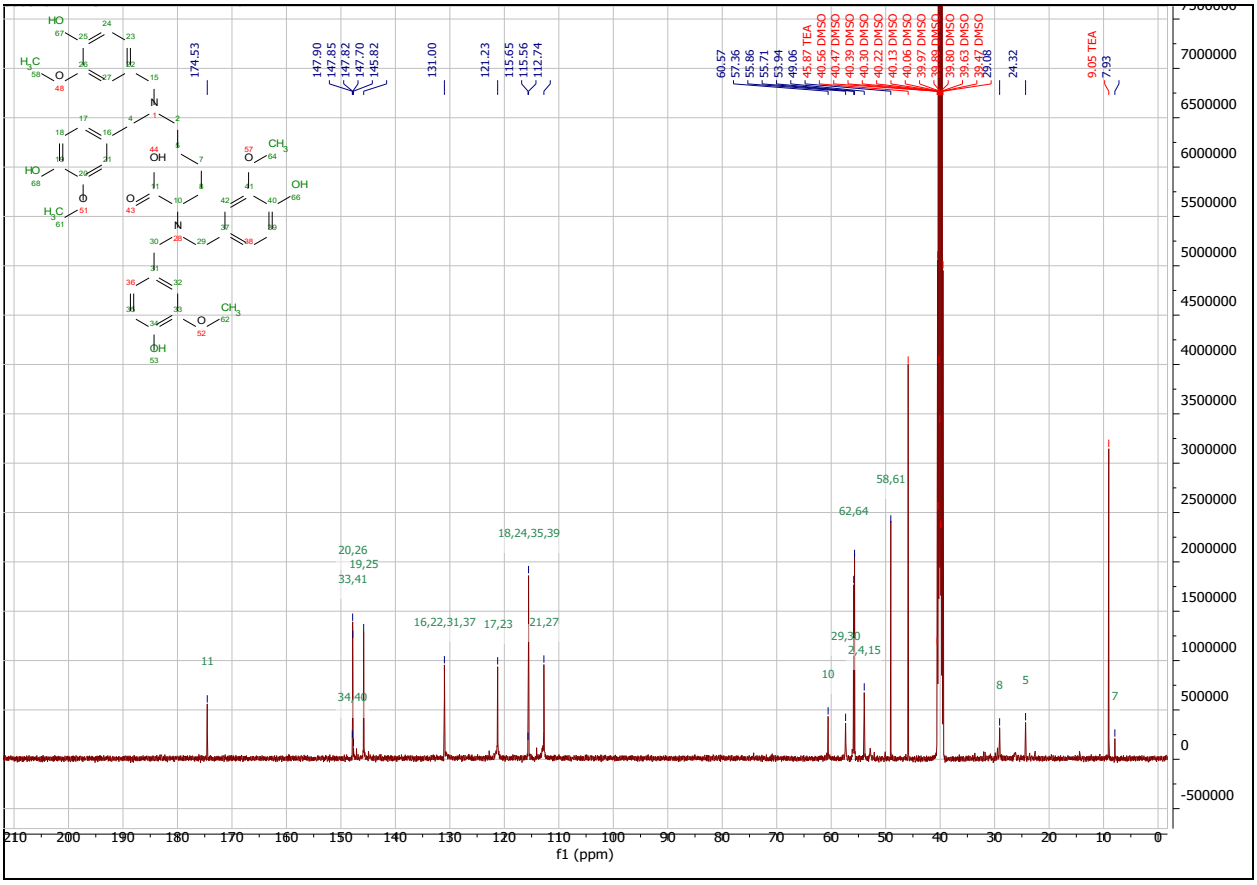


Compound **4**: Lysine-Vanillin derivative

^{1}H NMR spectrum of Compound **4**



^{13}C NMR spectrum of Compound **4**



HR-MS of Compound **4**