

# **Biotransformation of eugenol by an endophytic fungus *Daldinia sp.* IIMF4010 isolated from *Rosmarinus officinalis***

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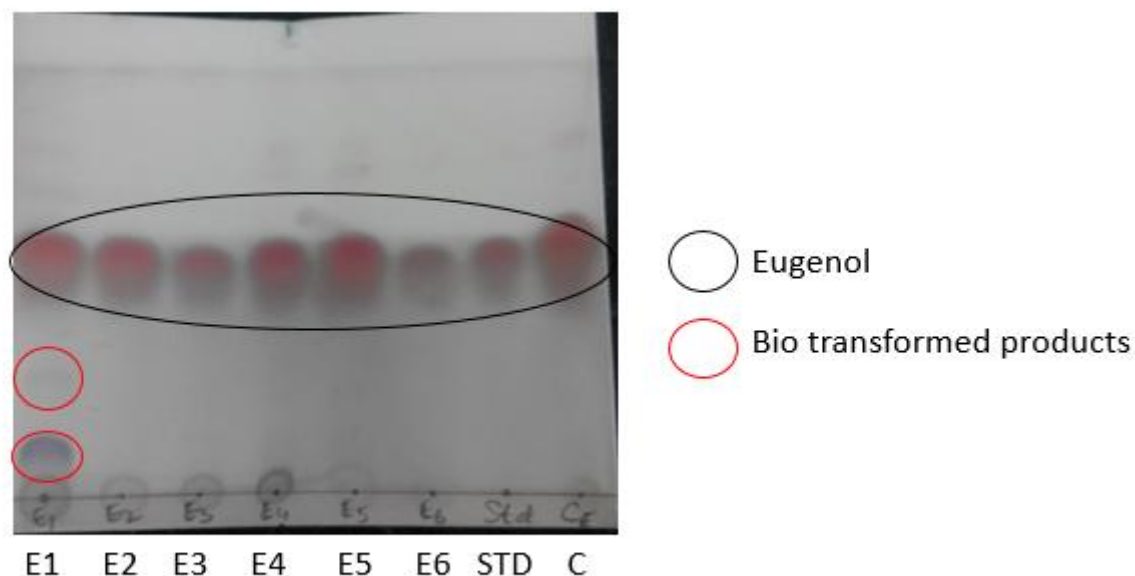
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## **Abstract**

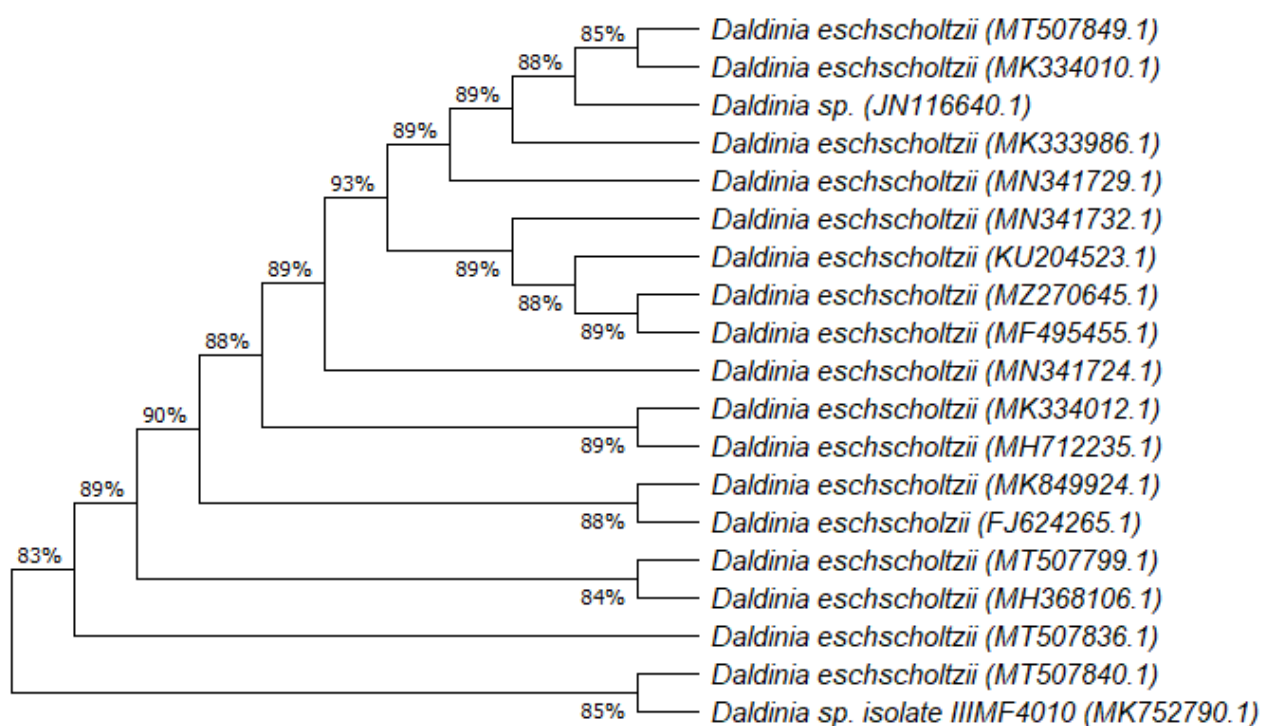
Natural value-added compounds produced from biological sources have attained immense significance in medicinal, food, flavourings, and agrochemical industries. Further, biotransformation is a powerful tool used to produce value-added compounds cost-effectively and selectively. In the present study, biotransformation of eugenol using an endophytic fungus *Daldinia sp.* IIMF4010 isolated from the fresh leaves of the plant *Rosmarinus officinalis* leads to the production of two known value-added compounds. The biotransformation reaction of eugenol (50 mM) resulted in the production of eugenol- $\beta$ -D-glucopyranoside (6.2%) and vanillin (21.8%). These biotransformed products were further characterized by liquid chromatography-mass spectroscopy (LC-MS) and nuclear magnetic resonance (NMR).

**Keywords:** Eugenol, Biotransformation, Eugenol- $\beta$ -D-glucopyranoside, Vanillin, *Daldinia*

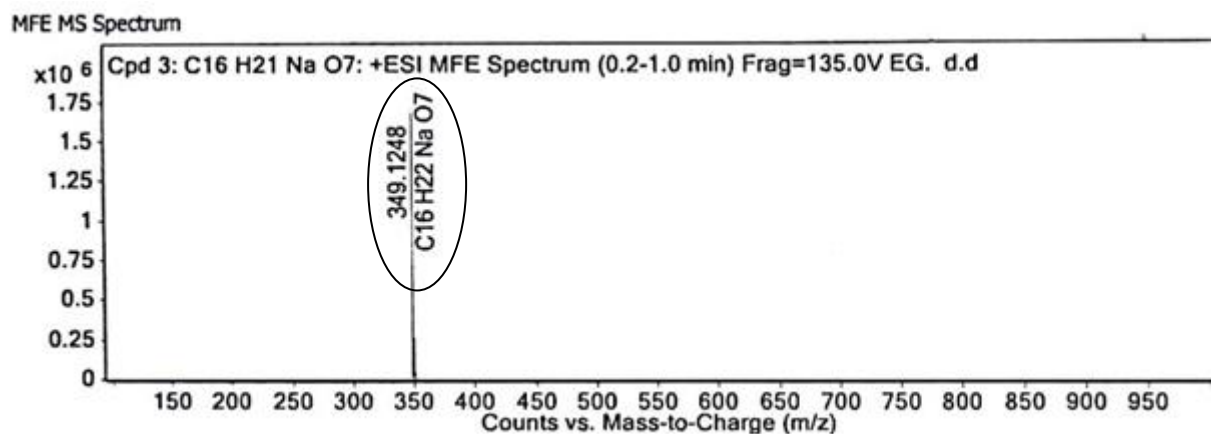


E1-6: Reaction mixtures of screened cultures; STD : Eugenol standard; C: Control

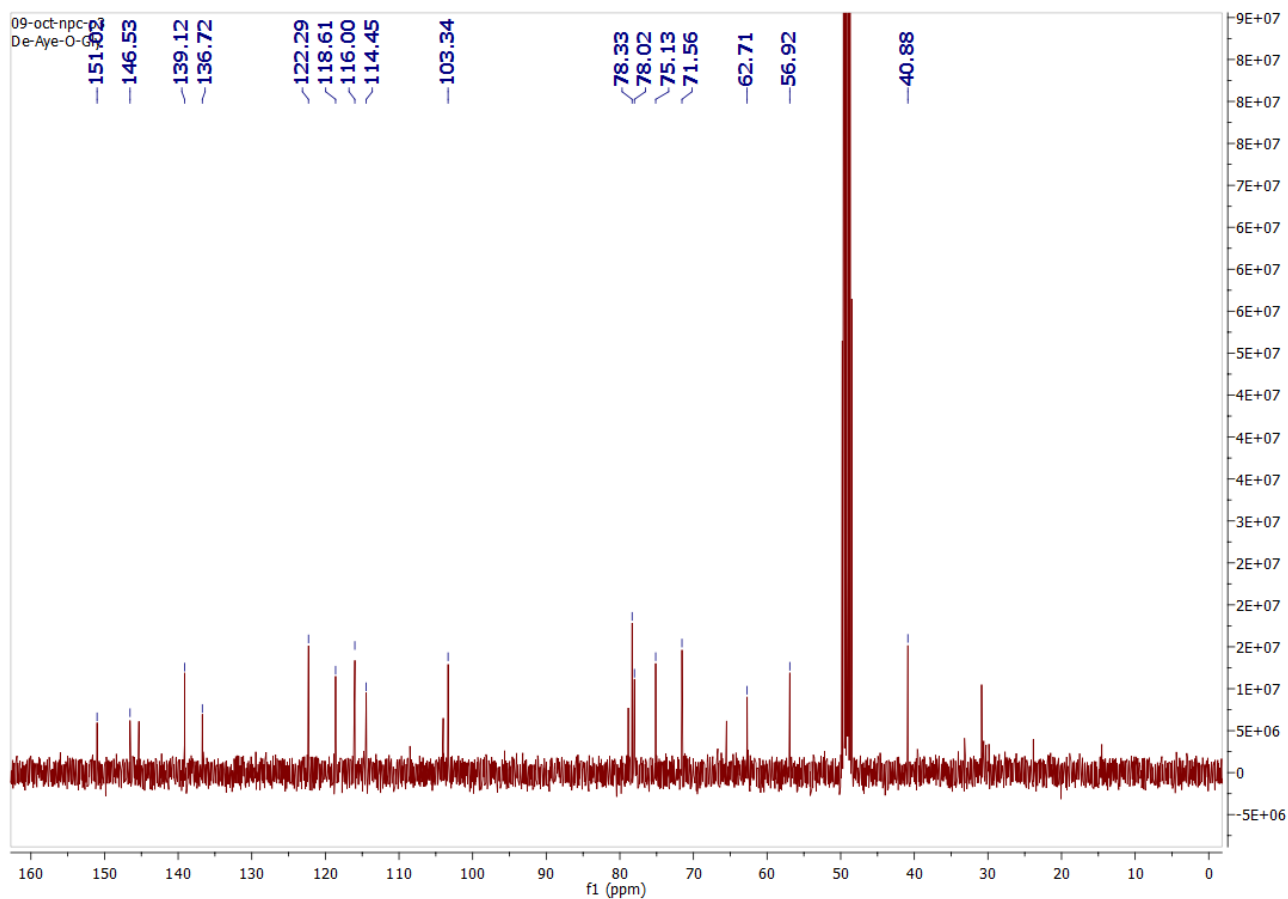
**Figure S1:** TLC (developed in anisaldehyde) of screened cultures (E1-6) used in the biotransformation reaction.



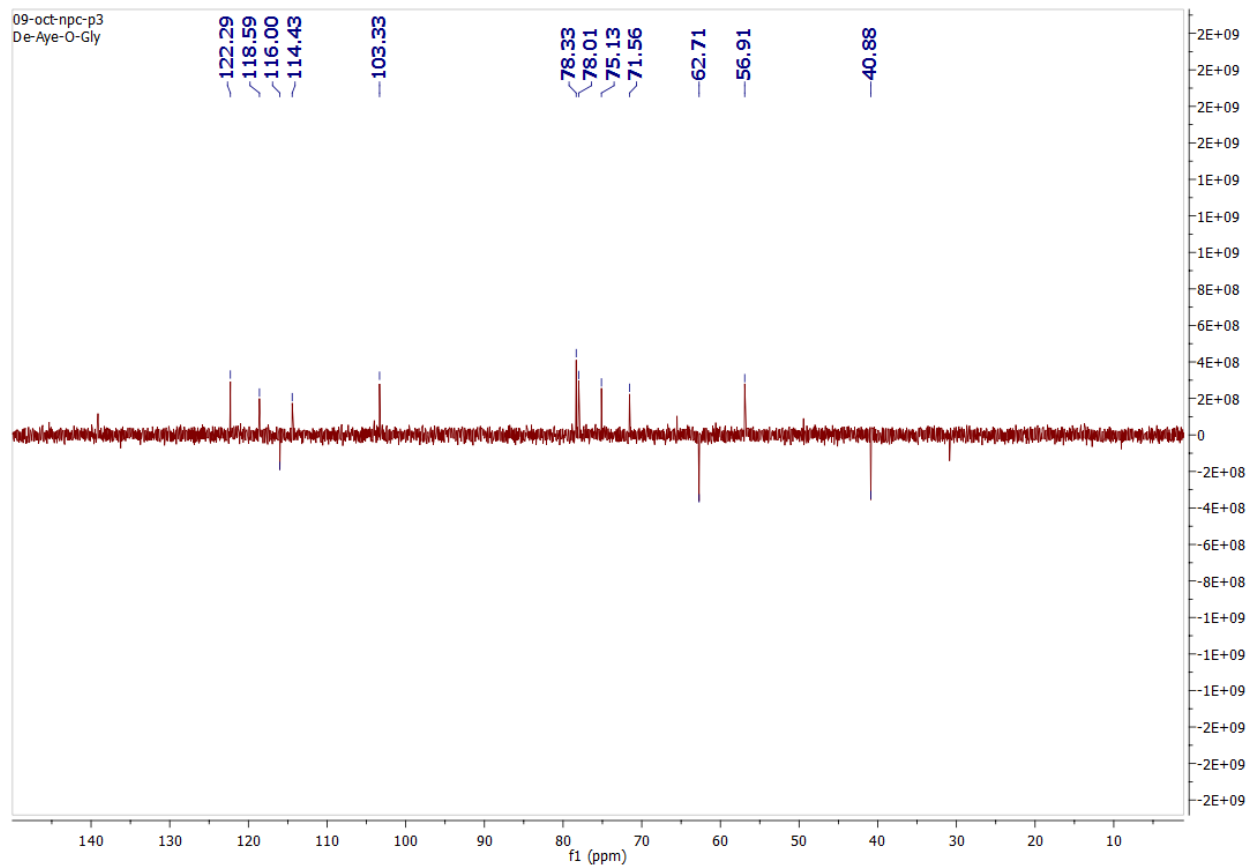
**Figure S2:** Phylogenetic tree showing evolutionary relationship of *Daldinia sp.* IIMF4010 with closely related strains based on the ITS based rDNA sequences. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (500 replicates) are shown next to the branches. The evolutionary distances were computed using the neighbour-joining method and are in the units of the number of base substitutions per site. Evolutionary analyses were conducted in MEGA 10.1.8.



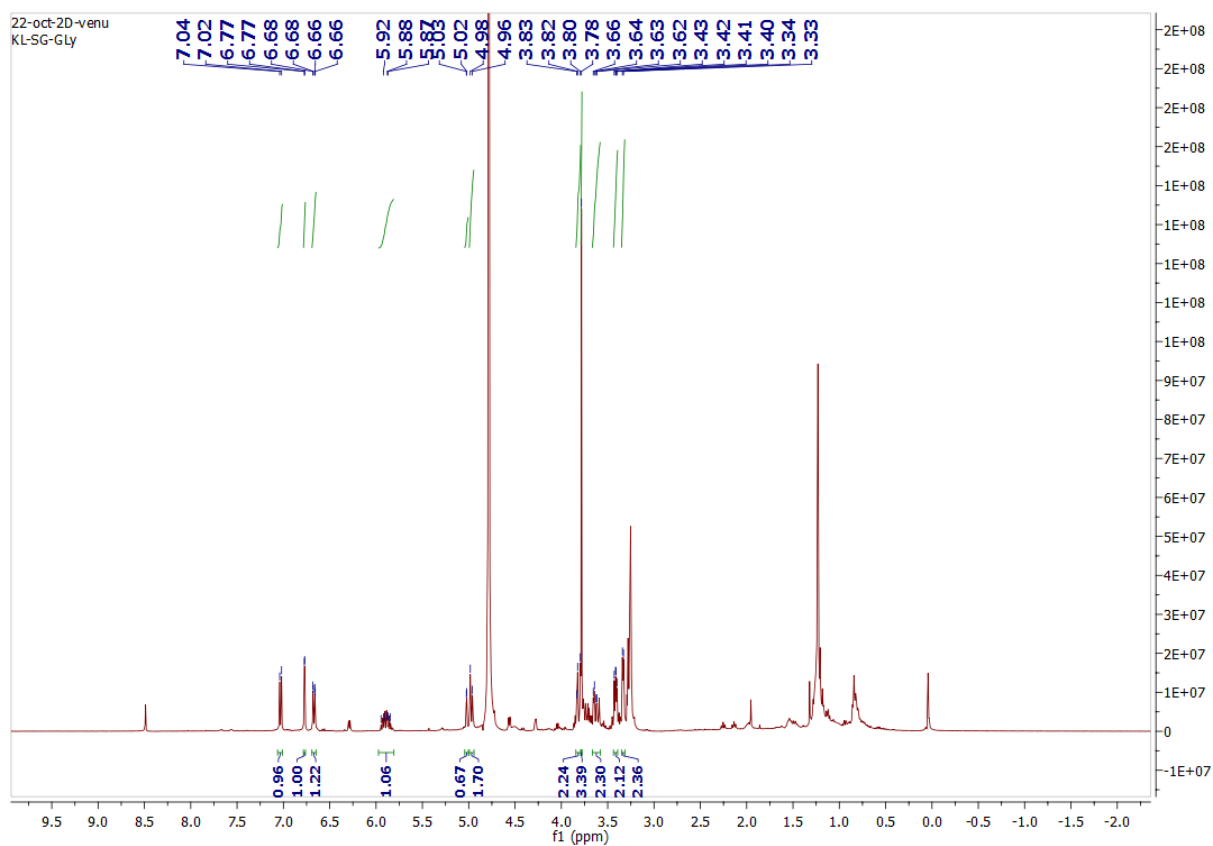
**Figure S3:** ESI-MS with a mass peak at  $m/z$  349  $[M+Na]^+$  of Compound **1**



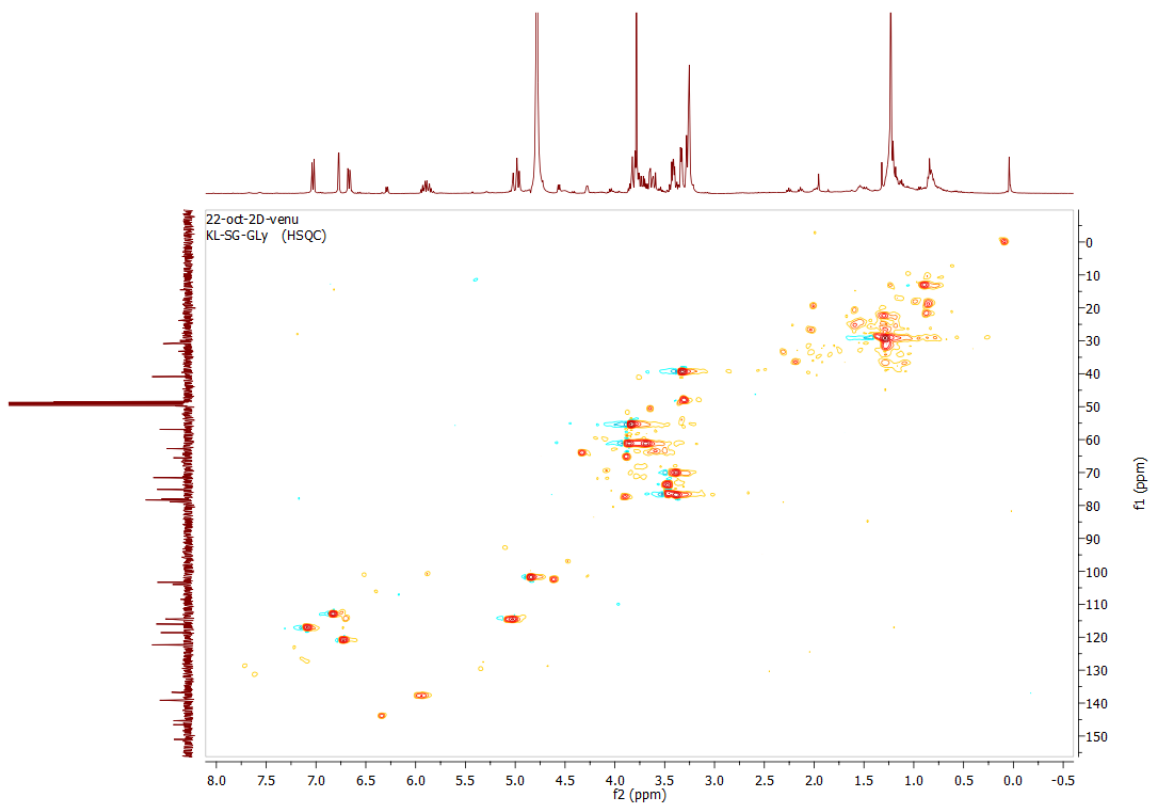
**Figure S4:**  $^{13}C$  of compound **1**



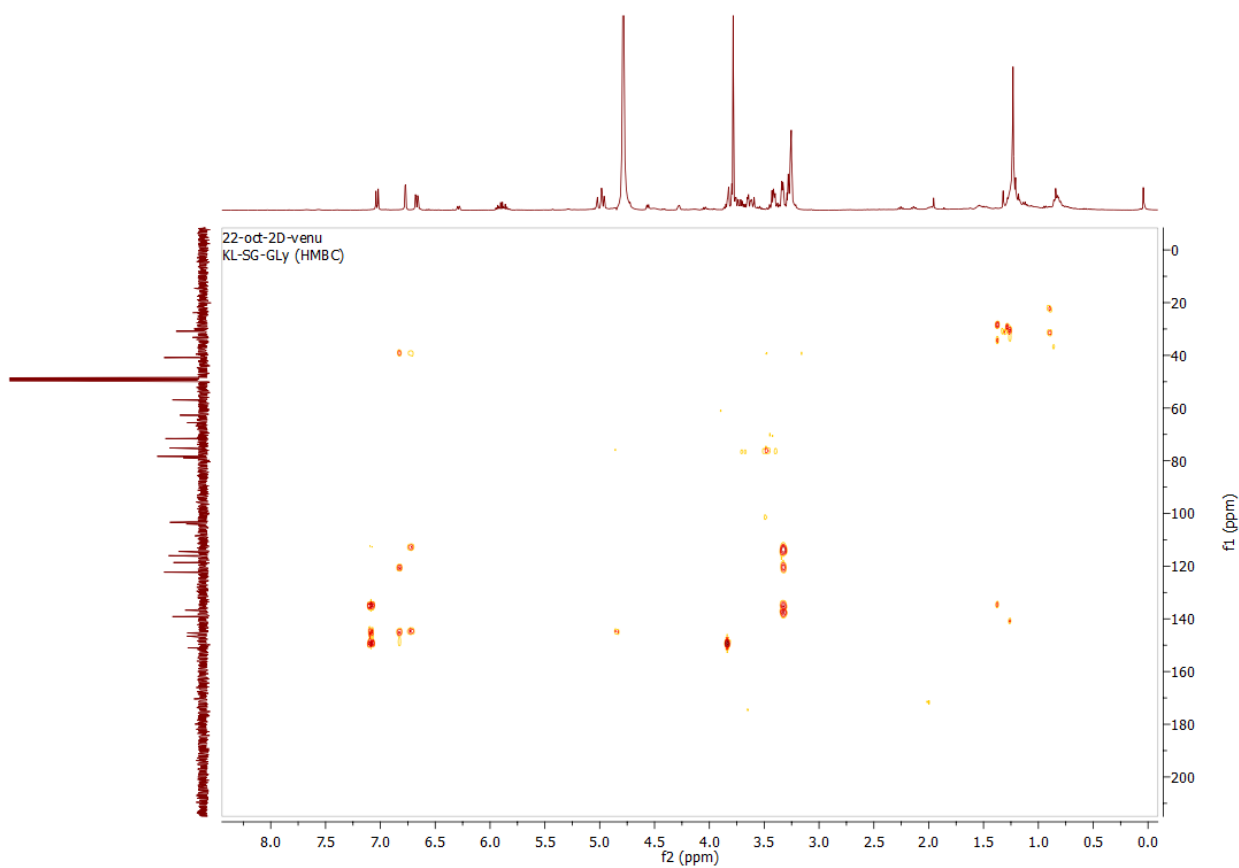
**Figure S5: DEPT of compound 1**



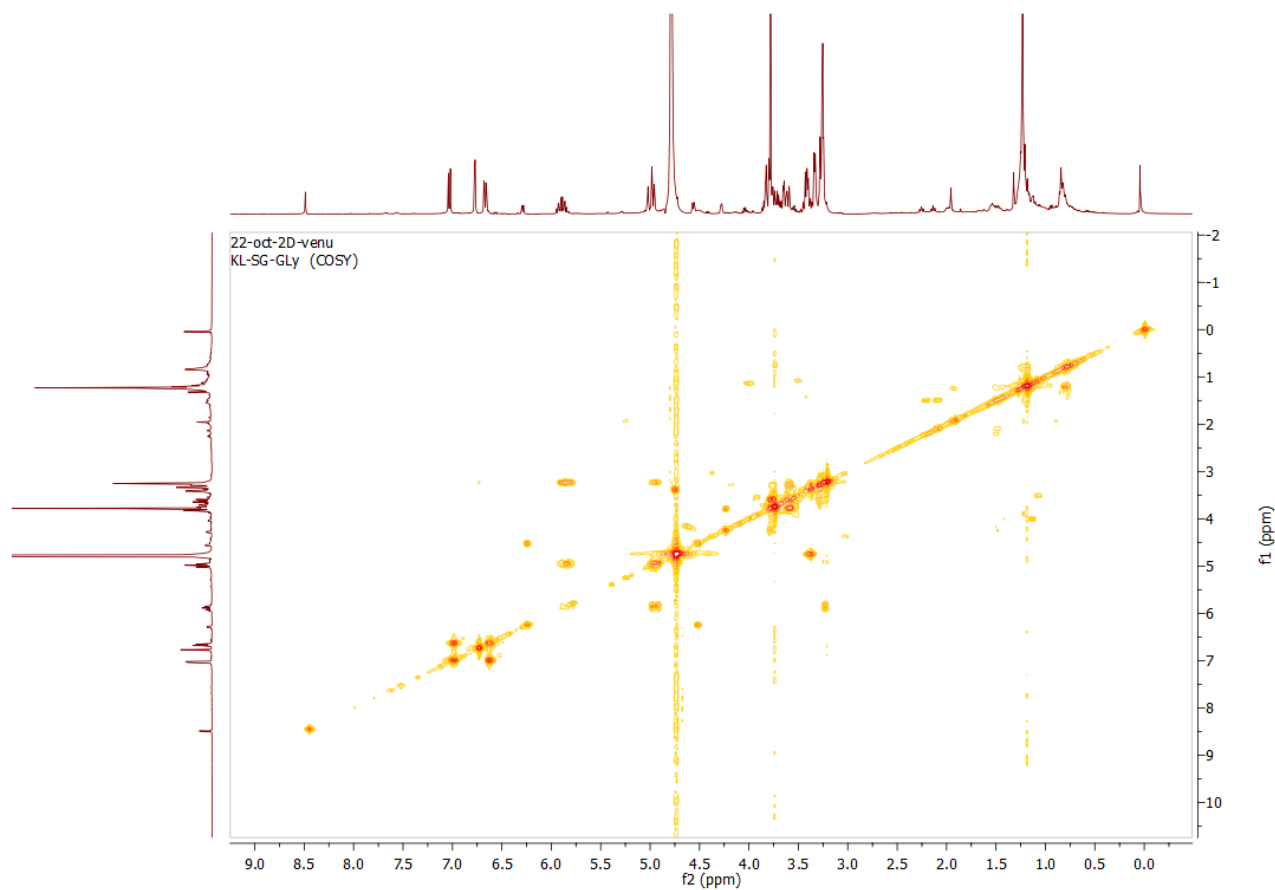
**Figure S6:  $^1\text{H-NMR}$  of compound 1**



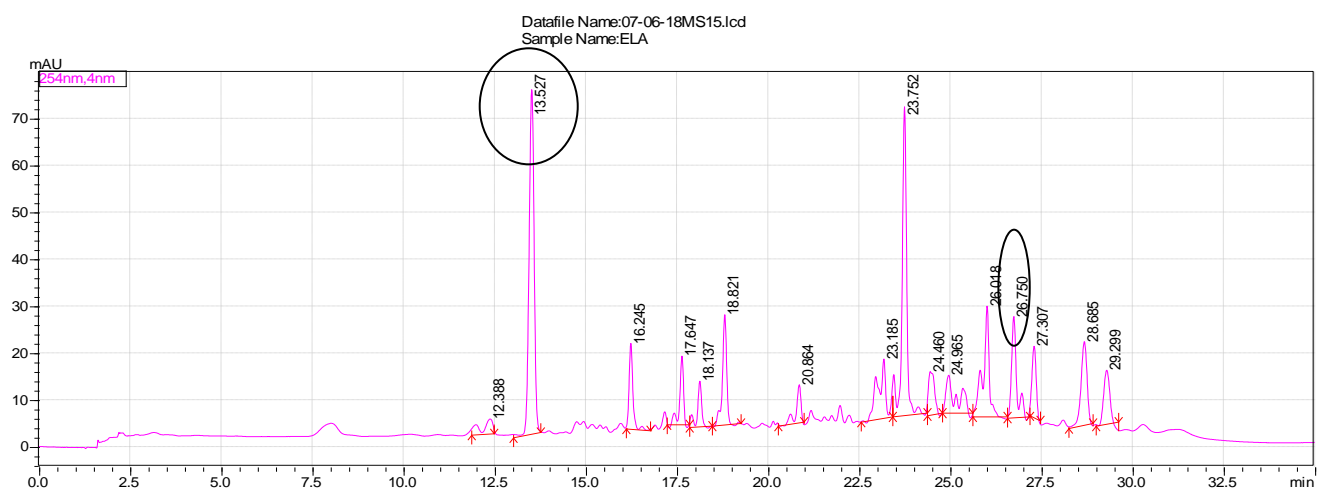
**Figure S7:** HSQC of compound **1**



**Figure S8:** HMBC of compound **1**

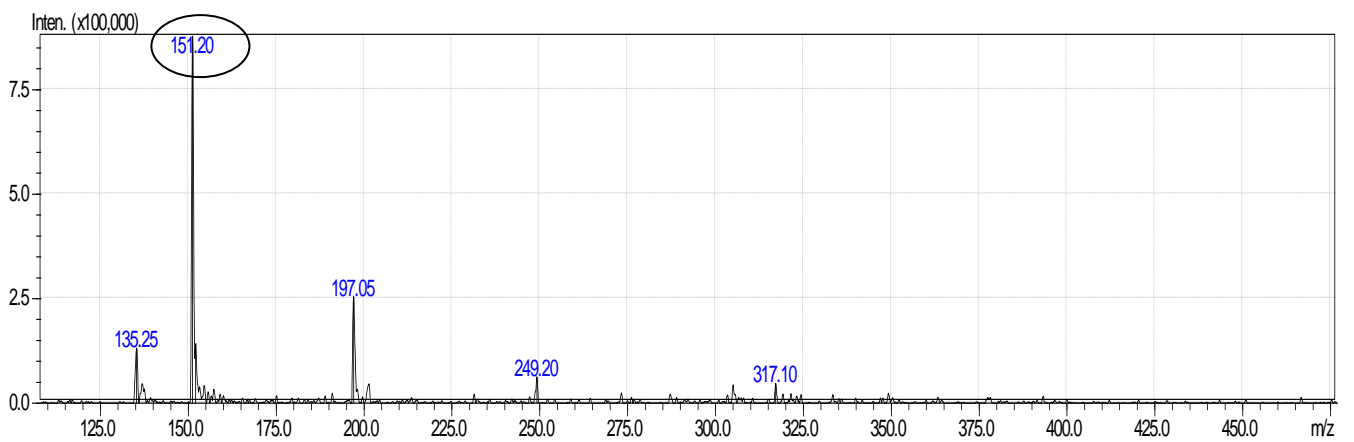
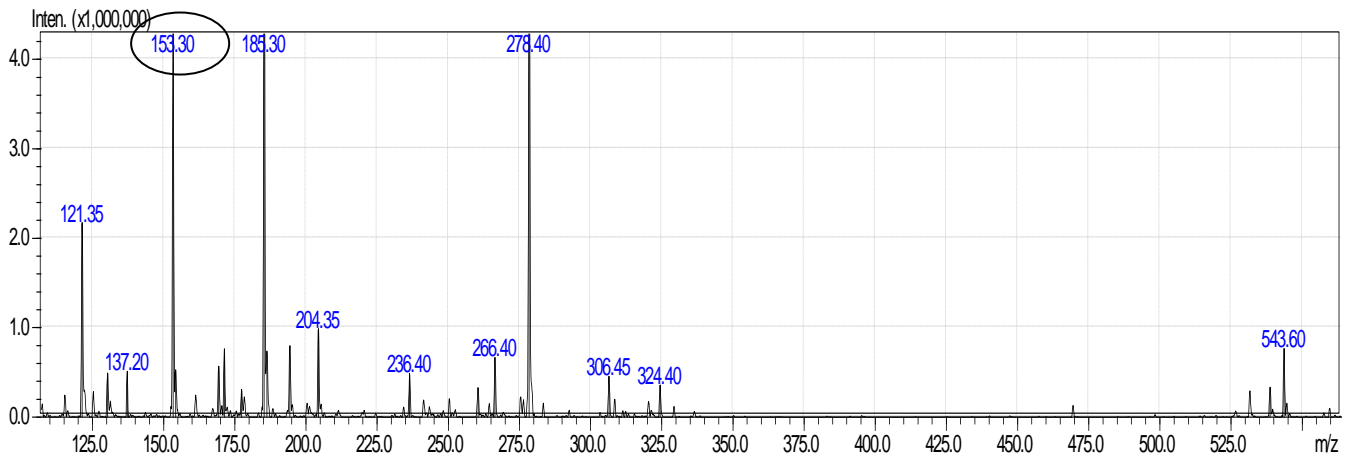


**Figure S9: COSY of compound 1**

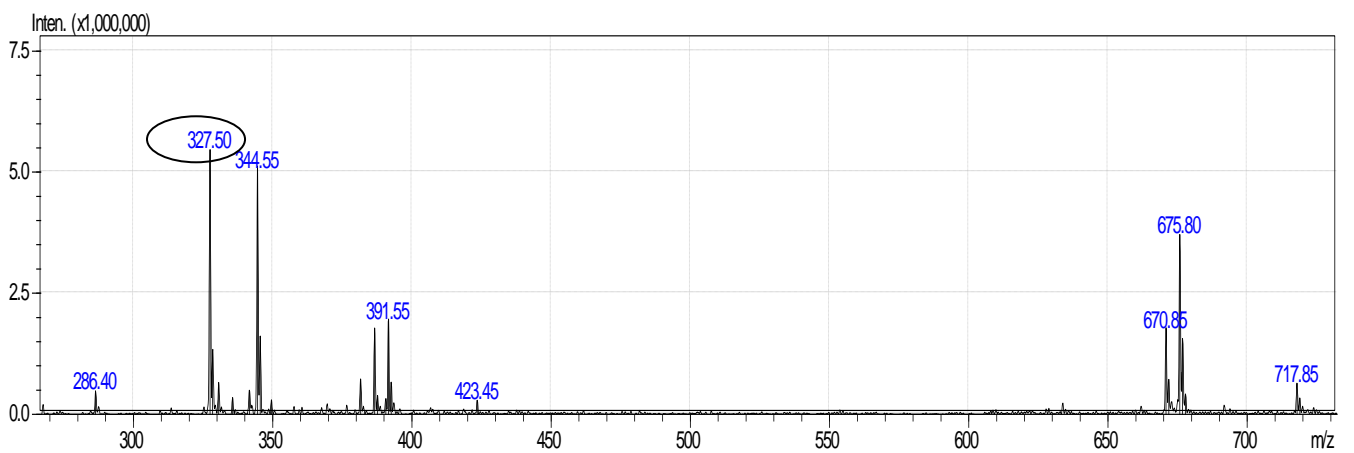


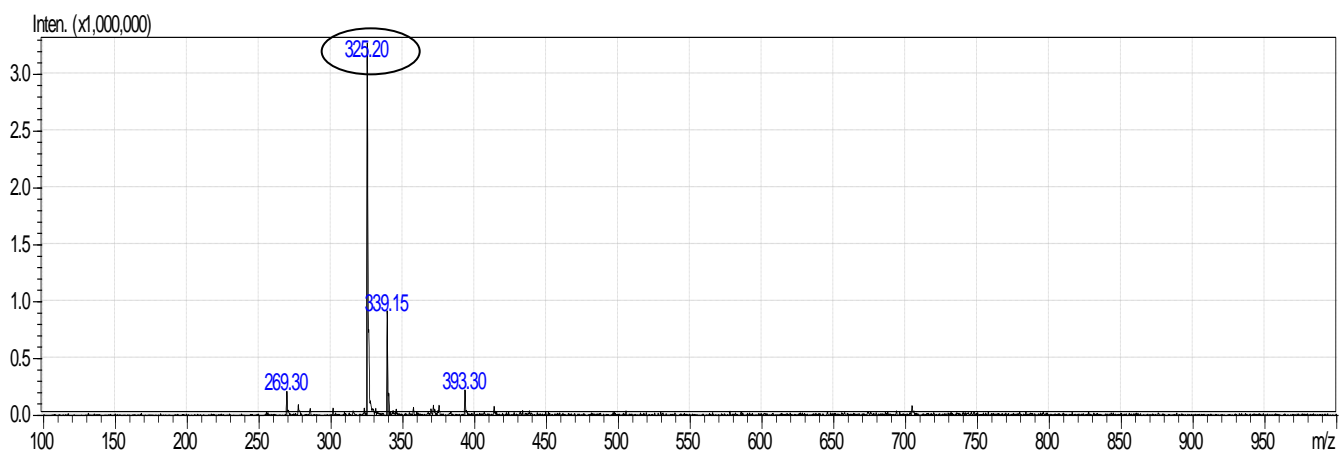
Retention time	Area	Height	Area%	Peak start	Peak end
13.527	697534	732235	21.787	13.003	13.749
26.750	198857	21430	6.211	23.722	27.760

**Figure S10: LC-MS chromatogram of the reaction mixture**



**Figure S11:** Mass spectra for peak 13.567 representing vanillin (152 molecular mass) with 153 m/z  $[M+H]^+$  and and 151 m/z  $[M-H]$ .





**Figure S12:** Mass spectra of peak 26.750 representing eugenol- $\beta$ -D-glucopyranoside (326 molecular mass) with 327 m/z  $[M+H]^+$  and 325 m/z  $[M-H]^-$ . Scan (E+): Positive mode; Scan (E-): Negative mode.