

Synthesis and Spectroscopic Characterisation of Tricyclic Gallate - Ethyl-4-[(3,4,5- trihydroxybenzoyl)(dimethylaminobenzoyl) amino]benzoate

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ABSTRACT

The past years have revealed growing scholarly interest in gallates due to the reported biological efficacy of such compounds especially anti-inflammatory, and antioxidant activities and their usefulness to the field of coordination chemistry as labile ligands. This article reports the synthesis and structural characterisation of a tricyclic gallate ethyl-4-[(3,4,5-trihydroxybenzoyl)amino]benzoate. It was prepared through room temperature condensation of galloyl chloride with ethyl-4-aminobenzoate and 4-dimethylaminobenzaldehyde in moderate yield of 58% under mild conditions. The structural authentication was done by elemental analysis (CHNO), mass spectrometry, Fourier-transform infrared spectroscopy (FTIR) and nuclear magnetic resonance spectra (including ¹H NMR, ¹³C NMR, DEPT 135, COSY, HMQC and HMBC). The spectroscopic data confirmed the molecular structure whose results completely matched with that of the proposed structure. The molecular ion peak was supported by mass spectrometry and extensive NMR characterisation provided structural elucidation with regard to bond connectivity and stereochemistry. Conclusively, an attempt to synthesise and characterise ethyl-4-[(3,4,5-trihydroxybenzoyl)amino]benzoate which is an appealing candidate as a biological probe and a versatile route to many metal complexes used in many extended applications has been successful. The method exhibited offers an expedient and direct route to similar gallate derivatives.

KEYWORDS: Tricyclic gallate, Galloyl chloride, Spectroscopic characterization, Biological activity and Coordination chemistry

I. INTRODUCTION

The numerous biological properties (such as antioxidant, (Gunckel et al., 1998), antimicrobial, (Capasso et al., 2025, Lianget al., 2023) and anticancer (Lianget al., 2017, Locatelliet al., 2013, El-Shahawy et al., 2023) of gallates have made them compounds of interest in the medicinal chemistry field. The incorporation of gallic acid moieties into sophisticated molecular structure has come in to represent a useful method toward creating new therapeutic compounds with greater bioactivity and more attractive pharmacodynamic profiles.

Tricyclic gallate compounds are an exceptionally promising route into drug discovery, this structure presents the extensively documented biological activity of gallate moiety (Lianget al., 2023) with the structural rigidity and increased binding ability of cyclic compounds. The presence of several aromatic rings can enable π - π stacking interactions with biological targets and provide more sites for functional groups modification. Moreover, the design of new organic ligands with multifunctional capabilities remains an essential part of materials science, (Grapeet al., 2022), coordination chemistry and catalysis. (Rushiet al., 2018) gallic acid derivatives have become of considerable interest in view of their

power to bind metals, and their considerable structural diversity (Martyniuk et al., 2024).

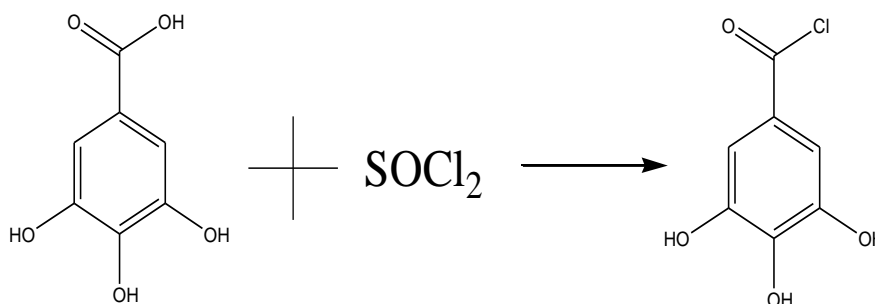
Ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate is a new tricyclic gallate analog having both an electron-withdrawing and electron-donating group. Its substituents (the trihydroxybenzoyl and dimethylaminobenzoyl groups) are incorporated into a tricyclic ring system, providing it with conjugation, which may play a role in its spectroscopic properties and coordination properties. The combination of structures is especially interesting because this combination has the potential to provide two modes of action; the antioxidant potential of the gallate moiety and the electron donating potential of the dimethylamino moiety, which could increase cellular uptake and alter biological activity.

Notwithstanding the favourable biological characteristics of gallate derivatives, little in-depth synthetic methodology and in-depth spectroscopic analysis of complex tricyclic gallate derivatives have been published. Such complex gallate derivatives prove to be rather difficult to produce with regards to regioselectivity, compatibility of functional groups and purification. Improved synthetic methodologies and complete structural elucidation is crucial in the advancement of this type of compounds to practical use in medicinal

chemistry. Synthesis and spectroscopic characterization of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate was achieved and described herein. An improved synthetic methodology is described which circumvents the disadvantages of multi acylation and phenolic oxidation and full structural characterization has been achieved by the use of state-of-the-art elemental analysis, NMR spectrometry, mass spectrometry and IR spectroscopy. The approach used in this paper gives a basis for the synthesis of related tricyclic gallate derivatives and also forms the larger base of harmony of structure-activity in this significant type of bioactive compound.

II. EXPERIMENTALS

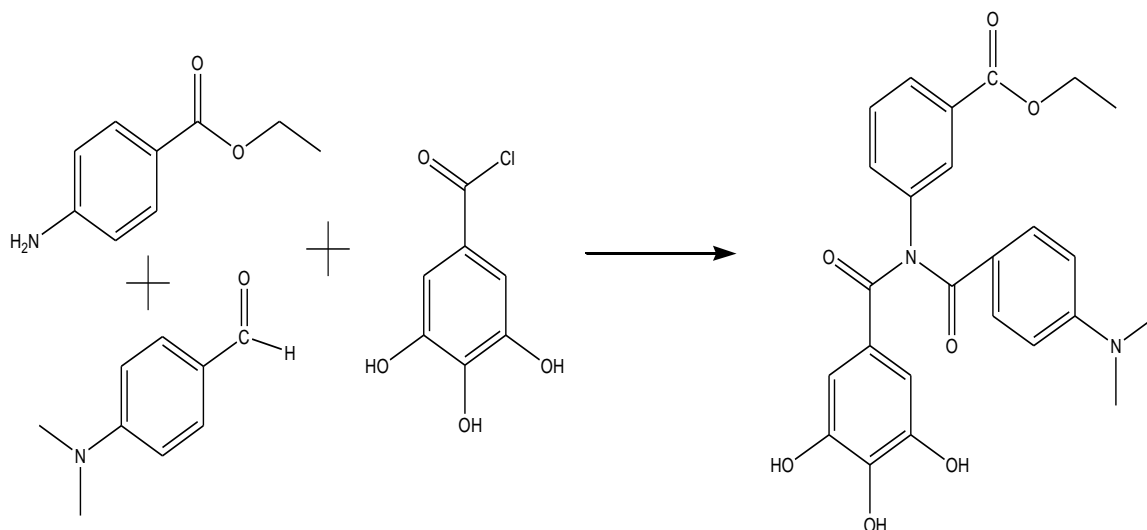
Galloyl chloride was synthesised following the procedure reported by Human and Mills (1946). 0.1 mole (18.8g) of gallic acid was dissolved in 100 cm³ of benzene and 0.1 mole (7.33 cm³) of thionyl chloride (SOCl₂) added to it followed by adding 0.1 mole (8.16 cm³) of pyridine. The resultant mixture was refluxed for 2 hours at 100 °C. The precipitate formed was filtered and washed repeatedly with methanol and dried at room temperature. The reaction scheme is as shown in Equation below



Synthesis of Ethyl-4-[(3,4,5 -trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (EDAB)

The synthesised galloyl chloride was used as a precursor for the synthesis of the ligand ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (EDAB). A solution containing 0.83g (5 mmols) of ethyl-4-aminobenzoate was dissolved in 40 cm³ of methanol and added to 40 cm³ methanolic solution containing 0.99g (5 mmols) of

4-dimethylamino benzaldehyde with stirring. The green solution formed was reacted with a solution containing 0.94g (5 mmols) of galloyl chloride with stirring. A bright yellow solution formed was left for 7 days after which a bright yellow solid formed was recovered and washed repeatedly with water and air-dried. The reaction scheme is shown below



The dry product was characterised by spectroscopic characterisation including elemental analysis, mass spectrometry, and NMR spectroscopy and FT-IR.

The dry product was characterised by spectroscopic characterisation including elemental analysis, mass spectrometry, carbon -13 and proton NMR spectroscopy, COSY, DEPT 135, HMQC, HMBC and Fourier transform infrared spectroscopy.

III. RESULTS AND DISCUSSION

Galloyl chloride ($C_7H_5ClO_4$) was synthesised via the method stated above as a white powdery solid with a melting point of $442\text{ }^\circ\text{C}$ in high yield of 93% and used as a starting material for the synthesis of EDAB. This reaction pathway was chosen because the reaction of acid chlorides with either amines or alkanols is one of the easiest routes to gallates.

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No.	Weight [mg]	Name	N Area	C Area	H Area	N [%]	C [%]	H [%]	O [%]	Date	Time
18	2.4230	Sample-8	5 092	45 880	13 046	0.22	44.70	2.609	33.782	08.05.2024	14..
19	3.2850	Sample-1	7 485	74 065	18 538	6.05	64.65	5.121	24.239	08.05.2024	14..
20	2.3230	Sample-9	7 285	74 165	18 616	5.55	64.61	6.370	24.323	08.05.2024	15..

Figure I.: Result of Elemental Analysis of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

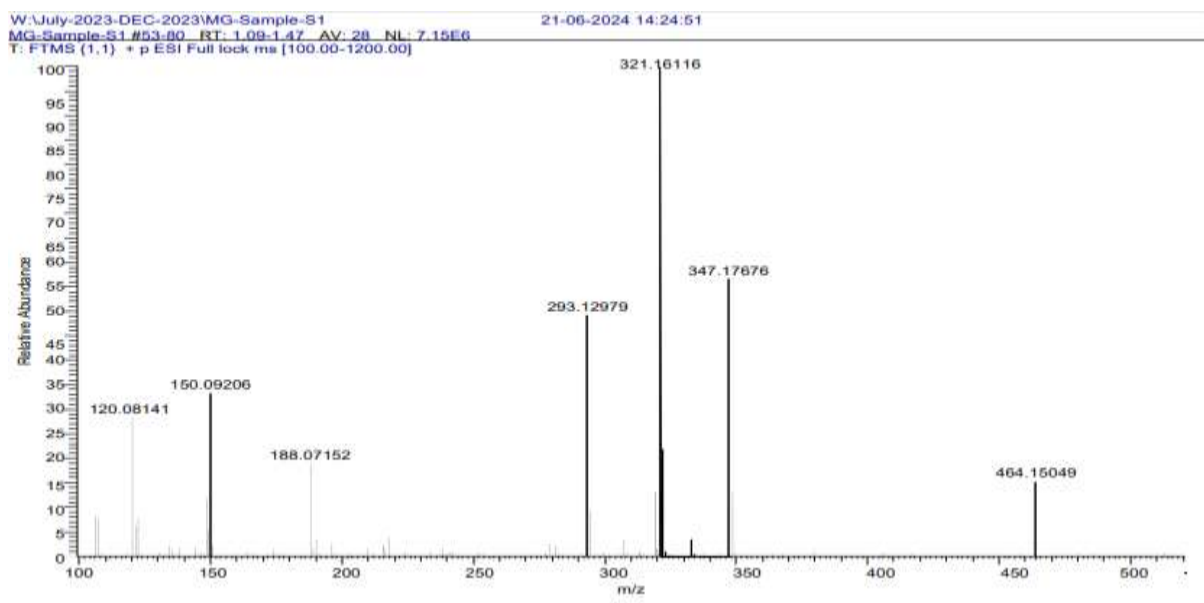


Figure 2: Mass Spectrum of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

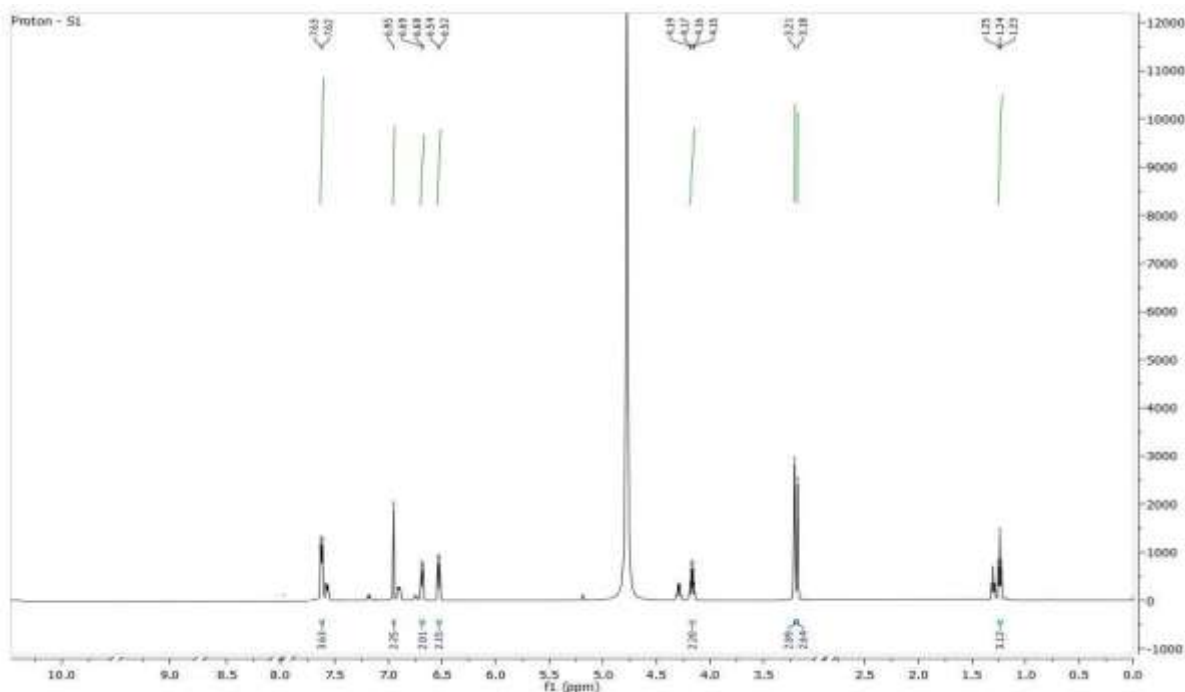


Figure 3: Proton NMR Spectrum of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

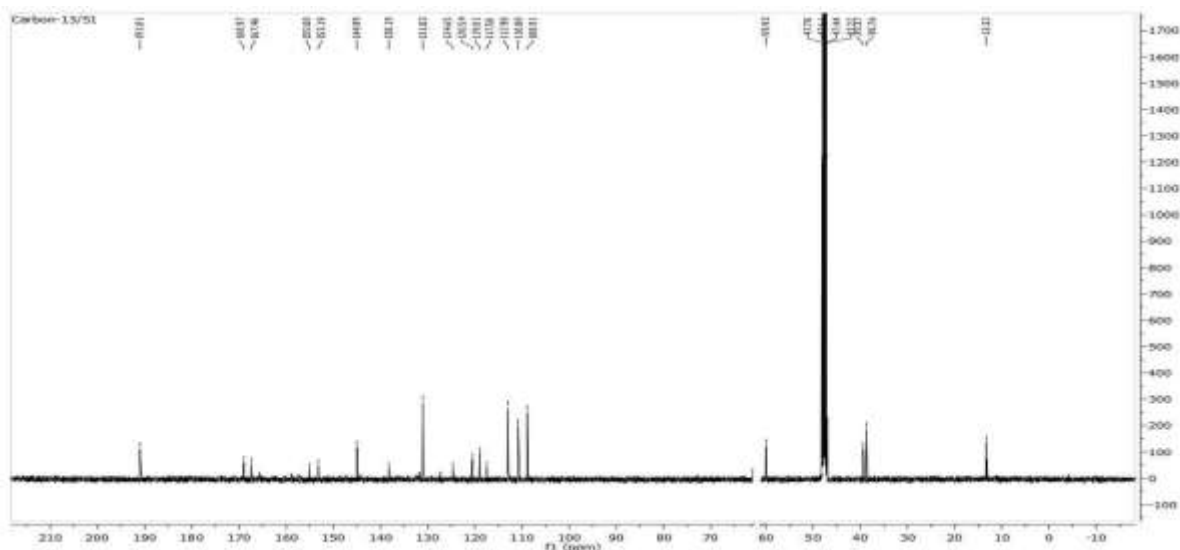


Figure 4: Carbon-13 NMR spectrum of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

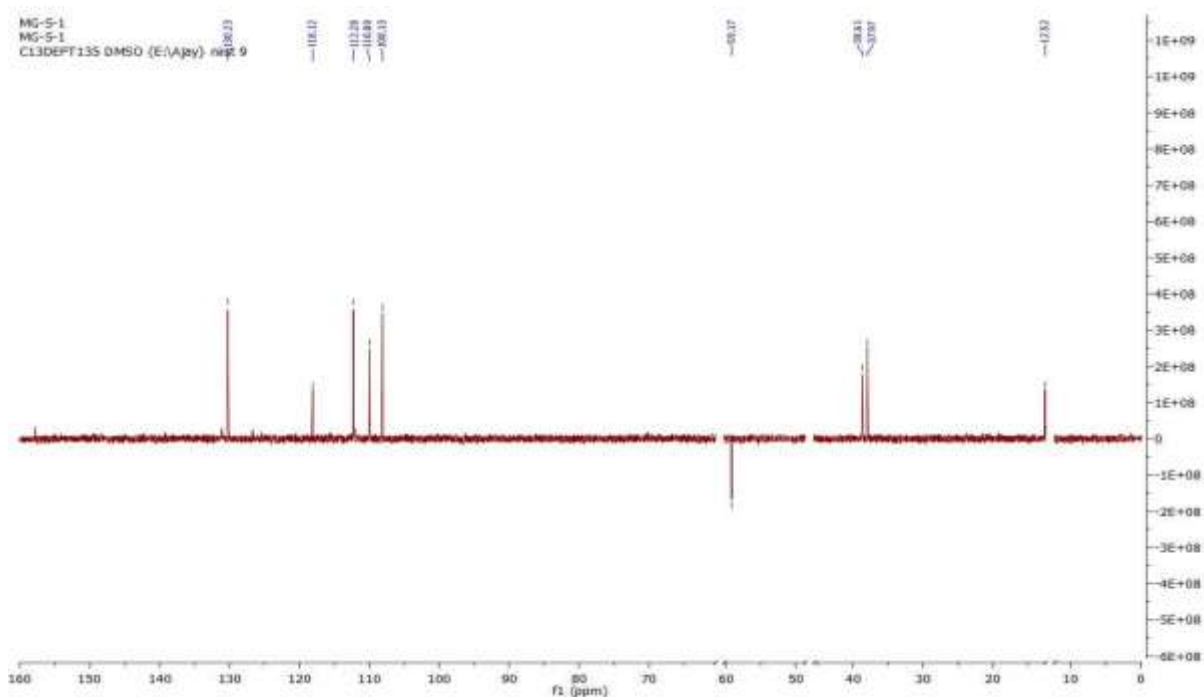


Figure 5: DEPT 135 NMR spectrum of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

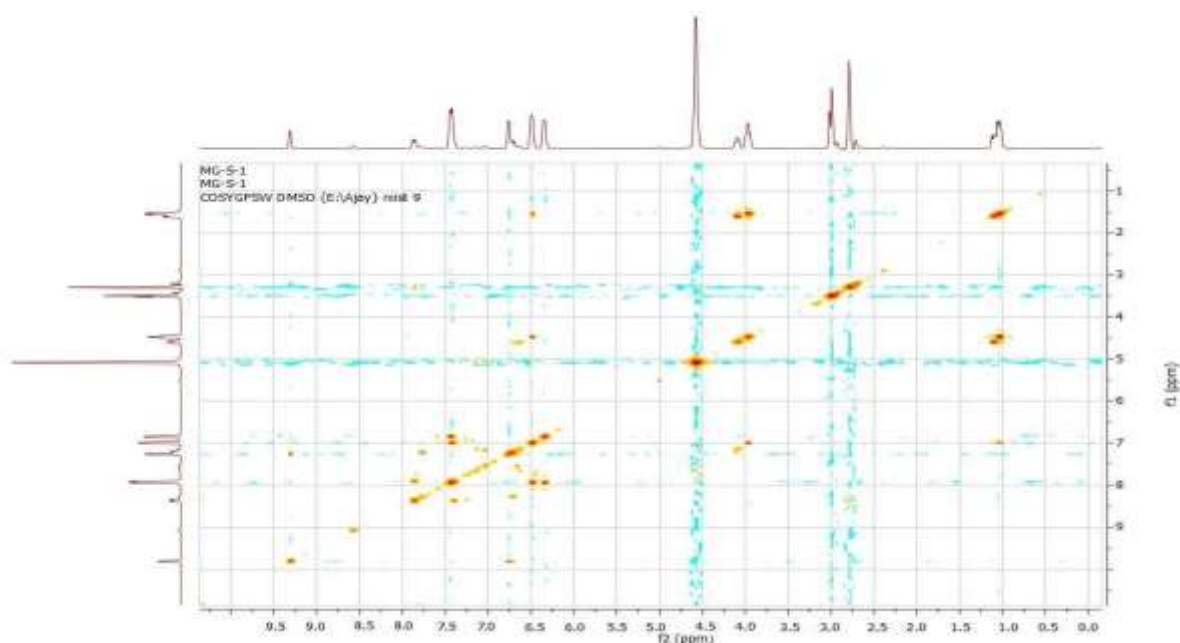


Figure 6: COSY NMR spectrum of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

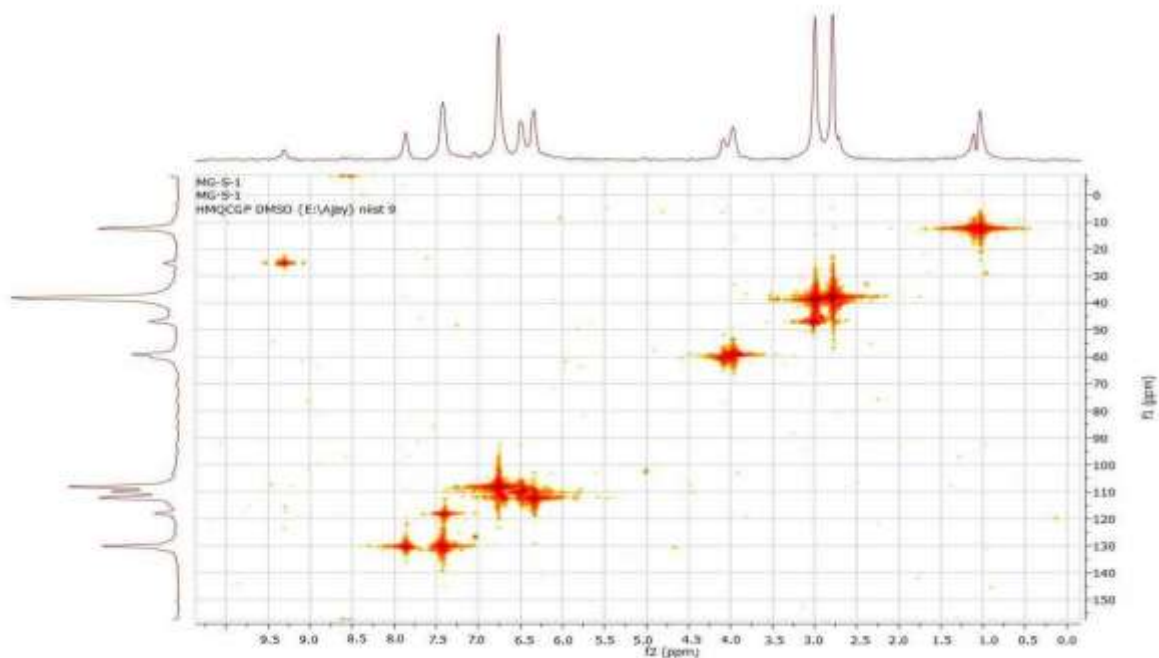


Figure 7: HMOC NMR spectrum of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

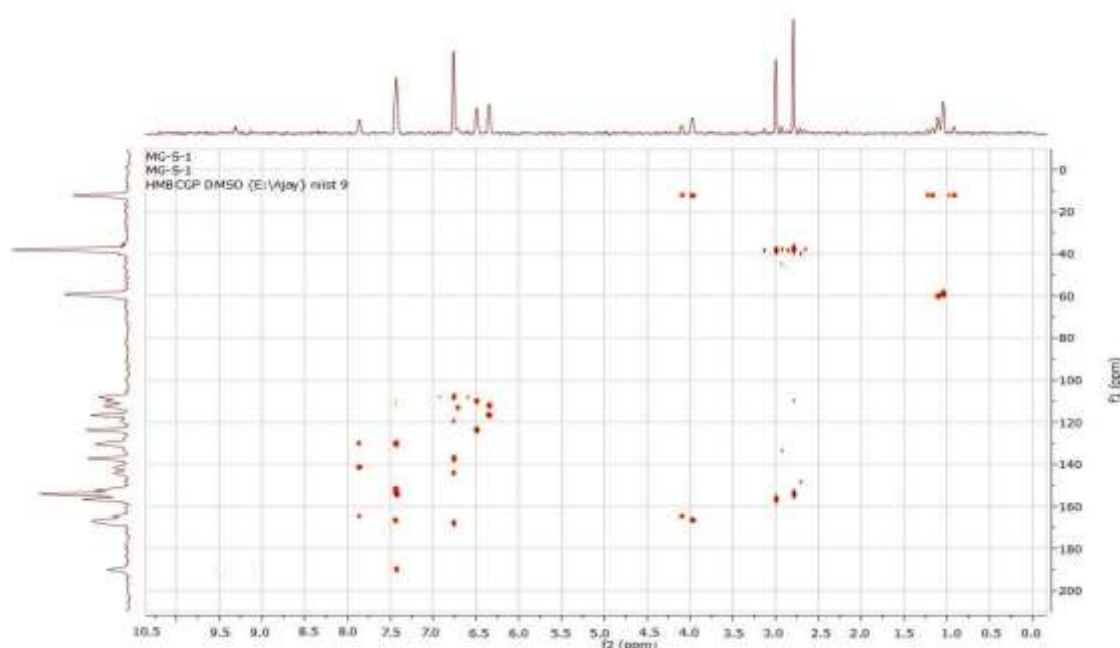


Figure 8: HMBC NMR spectrum of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

Sample 1

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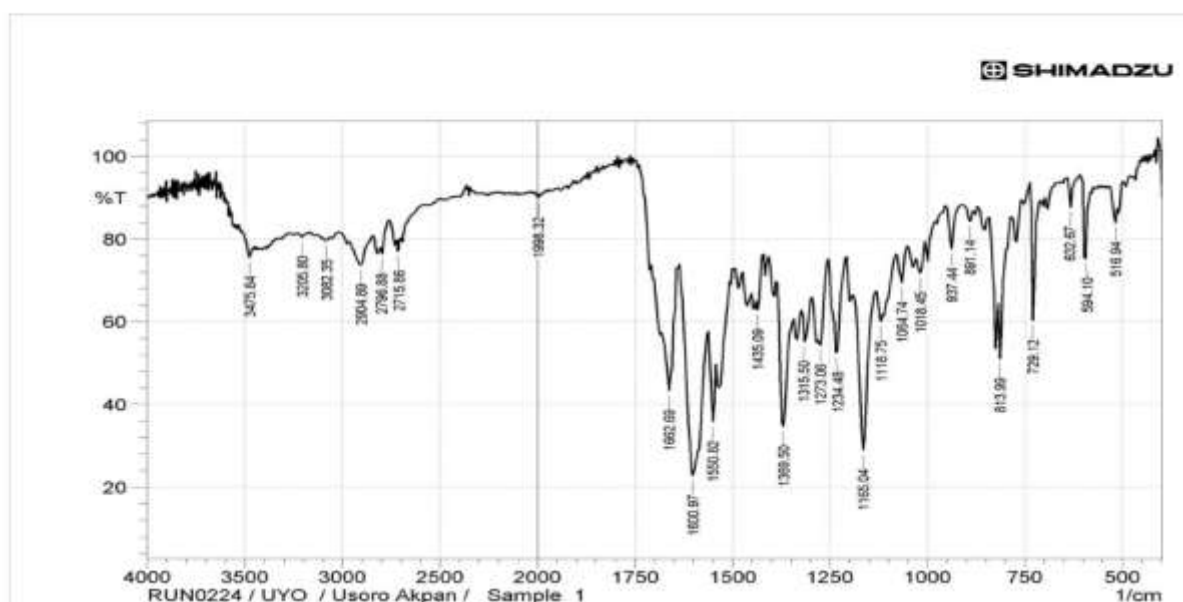


Figure 9: FTIR spectrum of ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate (S1)

Spectroscopic Characterisation of Ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate

The reaction of galloyl chloride with ethyl-4-aminobenzoate and 4-dimethylaminobenzaldehyde afforded EDAB in good yield of 58 %. EDAB is stable and has a melting point of 335 °C. The results of

spectroscopic characterization are shown as Figure 1-9. The elemental analysis result (Figure 1) of the compound with chemical formula $C_{25}H_{24}N_2O_7$ and molecular weight of 464.47; showed the calculated and (experimental) percentage compositions to be: C, 64.65 (64.65), H, 5.21 (5.121), N, 6.03 (6.05) and O, 24.11 (24.239). The calculated and the experimental values are in good agreement. The molecular mass of the compound was confirmed by the result of mass spectrometry (Figure 2) with a molecular ion peak at 464.46 m/z.

The proton NMR in DMSO (δ ppm) spectrum (Figure 3) of the compound ethyl-4-[(3,4,5 trihydroxybenzoyl)(dimethylaminobenzoyl)amino] benzoate showed methyl signal at δ_H 1.24 (3H, t, $J=7.1$ Hz), which was assigned to the protons at H-16. Two singlet signals were observed at δ_H 3.18 and δ_H 3.21 which were assigned to the deshielding protons of the two methyl groups attached to nitrogen atom (H-24 and H-25). One deshielding proton signal at δ_H 4.17 (2H, q, $J=7.1$ Hz) which was assigned to proton at H-15 and aromatic protons signal at δ_H 6.53 (1H, d, $J=8.4$ Hz, H-12 and H-10), at the adjacent side, signal resonated at δ_H 7.62 (1H, d, $J=7.4$ Hz, H-9 and H-13) were observed. Another singlet signal resonated at δ_H 6.95 (1H, s,) and was attributed to the aromatic protons H-3 and H-7 while the signal which occurred at δ_H 6.69 (2H, d, $J=8.4$ Hz) was assigned to the aromatic protons H-22 and H-20 which resonated with peak signals of H-23 and H-19. The ^{13}C NMR (125 MHz, MeOD) spectrum (Figure 4) showed 19 carbon signals, of which 6 carbon signals appearing in the same chemical environment resonated with high peak signals summing up to 25 carbon signals, the solvent (MeOD) signal occurred at δ_C 47.44. The carbon signals of the carbonyl carbons resonated at δ_C 191.01, δ_C 168.97 and 167.46, which were attributed to carbon atoms at C-17, C-1 and C-14 respectively due to the deshielding effect of heteroatoms, nitrogen and oxygen, while the aromatic carbon signals resonated between δ_C 155.0 and δ_C 108.9. Carbon signals showing deshielding effect due to heteroatom was observed at δ_C 59.9 (C-15) while a methyl carbon signal was observed at δ_C 13.3 (C-16). The DEPT 135 spectrum (Figure 5) showed the presence of eight C-H carbon atoms around δ_C 130.23 and 108.13

one CH_2 at δ_C 59.17 (C-15) carbon atom and three CH_3 carbon atom signals δ_C 12.33 (C-16), δ_C 37.97 and δ_C 38.61 (C-24/C-25). The signal at 47.10 is the solvent (MeOD) methyl carbon peak. The 1H - 1H coupling COSY spectrum (Figure 6) showed a correlation between the methylene proton at δ_H 4.17 (2H, q, $J=7.1$ Hz, H-15) with δ_H 1.24 (3H, t, $J=7.1$ Hz, H-16) and the proton at δ_H 6.53 (1H, d, $J=8.4$ Hz, H-12 and H-10) with δ_H 7.62 (1H, d, $J=8.2$ Hz, H-9 and H-13). Signals at δ_H 3.18 and 3.21 (H-24/H-25) as well as proton at δ_H 6.95 (H-3 and H-7) were not coupled to any other proton (no neighbouring proton).

The 1H - ^{13}C HMQC spectrum (Figure 7) indicated that sp^3 proton δ_H 1.24 (3H, t, $J=7.1$ Hz, H-16) showed a direct bonding to δ_C 13.32 (C-16), δ_H 3.18 correlated to δ_C 38.8 while δ_H 3.21 is bonded to δ_C 39.4 (H-24/H-25 bonded to C-24/C-25), and sp^2 proton δ_H 4.17 (2H, q, $J=7.1$ Hz, H-15) is directly bonded to δ_C 59.93 (C-15). The aromatic protons (H-3 and H-7) δ_H 6.95 (1H, s) directly bonded to aromatic carbons (C-3 and C-7) δ_C 110.0 respectively, (H-10 and H-12) δ_H 6.53 bonded correlated to δ_C 112.28 (C-10 and C-12) and δ_H 7.62 (H-9 and H-13) bonded to δ_C 130.0 (C-9 and C-13) respectively. The HMBC spectrum (Figure 8) showed a correlation between the proton at δ_H 6.95 (1H, s, H-3 and H-7) and the carbon atom at δ_C 168.97 (C-1). Also, signals at δ_H 1.24 (3H, t, $J=6.7$ Hz, H-16) showed correlation with carbon signal at δ_C 59.93 (C-15) and δ_H 4.17 (2H, q, $J=6.7$ Hz, H-15) correlated with δ_C 13. (C-16) and 167.46 (C-14).

The FTIR spectrum (Figure 8) of EDAB (S1) in KBr showed O-H stretch of phenol at 3475.84 cm^{-1} , aromatic C-H stretch at 3205.80 cm^{-1} , C-H stretch aliphatic at 3082.35 cm^{-1} , C=O stretch at 1662.69 cm^{-1} , C=C aromatic at 1600.97 cm^{-1} , C-N stretch at 1273.06 cm^{-1} , O-H in plane bending phenol 1435.09 cm^{-1} , C-H bending aromatic at 813.99 cm^{-1} . It also showed the typical ester absorptions at 1662.69 cm^{-1} , 1369.50 cm^{-1} and 1165.04 cm^{-1} which correspond to C=O stretch, C-C-O stretch and C-O-C stretch respectively (the three peaks rule for ester). The synthesis of the compound EDAB was confirmed based on the above spectroscopic data. The proposed structure of the ligand ethyl-4-[(3,4,5 trihydroxybenzoyl)(dimethylaminobenzoyl)amino] benzoate is shown in Figure 4.1.

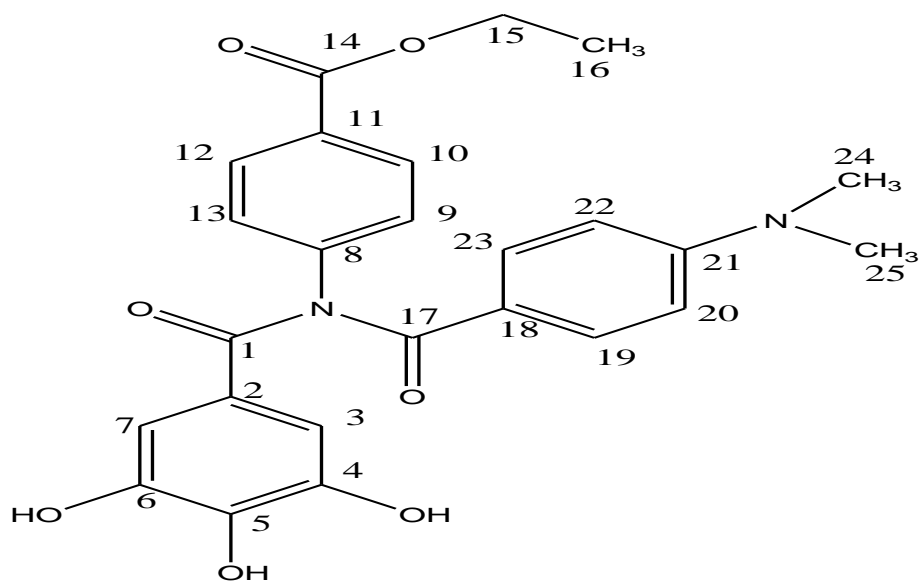


Figure 9: Proposed Structure of Ethyl-4-[(3,4,5-trihydroxybenzoyl)(dimethylaminobenzoyl)amino]benzoate

IV. CONCLUSION

The chemical synthesis and full spectroscopic analysis of the new tricyclic gallate derivative, ethyl- 4- [(3,4,5-trihydroxybenzoyl) (dimethylaminobenzoyl)amino] benzoate (EDAB), have been described. The synthesis occurred via an efficient but mild pathway and the structure of the compound was confirmed by elemental analysis, FTIR, mass spectrometry, and multi-dimensional NMR (^1H , ^{13}C , DEPT-135, COSY, HMQC, and HMBC). These data validate the proposed structure and show the unique electronic and stereochemical features which arise due to the conjugated tricyclic skeleton and neighboring electron-deficient and electron-receiving substituents.

The existence of both dimethylaminobenzoyl and galloyl groups in the tricyclic scaffold implies higher possibilities to the biological activity and the capacity to coordinate with metal ions. It follows that the article contributes to the currently expanding body of knowledge regarding gallate-based ligands and forms the basis of future studies of the use of such ligands in coordination chemistry, catalysis, and medicinal chemistry. Also, the synthetic method used could be easily modified for the preparation of structural analogues thus enabling further investigation of structure-activity relationships of this potentially highly useful class of compounds.

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