

Isolation and Structural Elucidation of Flavan-3-ols and Phenylacrylaldehydes from the Leaves of *Tapinanthus globiferus* (Loranthaceae) a Host Plant on *Vitex Doniana*Momoh Hassanah^{1*}, Dauda Garba², Yau Jamilu³, Yahaya M. Sani¹, Mohammed I.Sule¹¹Department of Pharmaceutical and Medicinal Chemistry, Ahmadu Bello University, Zaria 810107, Nigeria.²Department of Pharmaceutical and Medicinal Chemistry, University of Abuja, Abuja 900105, Nigeria.³Department of Pharmacology and Therapeutics, Ahmadu Bello University, Zaria 810107, Zaria, Nigeria.**ABSTRACT**

Tapinanthus globiferus is a medicinal plant used in traditional medicine in the treatment of inflammation, diabetes, and infections. The secondary metabolites responsible for its effect have not been thoroughly characterized, despite its ethnomedicinal uses. This study aimed to isolate and characterize secondary metabolites from the leaves of *T. globiferus* on *Vitex Doniana*. The leaves were extracted with 70% aqueous methanol which was then partitioned with n-hexane, chloroform, ethyl acetate, and n-butanol in the order of increasing polarity to give the respective fractions. The isolation of the bioactive compounds from the ethyl acetate and chloroform fractions was done using chromatographic techniques (TLC, column chromatography, and gel filtration). Structural elucidation of the compounds was achieved through spectroscopic analysis such as 1D NMR (¹H, ¹³C, DEPT) and 2D NMR (COSY, HSQC, HMBC). Column chromatography of the ethyl acetate fraction led to the isolation of catechin (Y12) and catechin-3-gallate (Y13). The isolation of 4-methoxyphenylacrylaldehyde (Y17) and 3-hydroxy-4-methoxyphenylacrylaldehyde (Y18) from the chloroform fraction, was done using column chromatography. The identification of the compounds was done by comparing spectral data with literature. The isolation of these compounds for the first time from this plant species is reported herein. Notably, Y17 and Y18 are of significant pharmacological interest, often associated with antimicrobial and anti-diabetic activities. The isolation of catechin and its derivatives scientifically justifies the established anti-inflammatory, bacteriostatic, and antioxidant properties and its traditional use. These findings significantly contribute to the phytochemical profile of the plant and identify key markers for future standardization and pharmacological investigation.

Keywords: Catechin, Gallate, Isolation, Medicinal plant, Spectroscopy, *Tapinanthus globiferus*

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Copyright: © 2026 Hassanah *et al.* This is an open-access article distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.**Introduction**

Medicinal plants have long been valued for their rich diversity of bioactive molecules with therapeutic potential¹ According to Dauda *et al.*¹ a medicinal plant is defined as any plant part containing chemical substances that can be used directly for therapeutic purposes or as precursors for drug development.¹ These plants have been integral to traditional healthcare systems for centuries, with their therapeutic effects attributed to secondary metabolites (i.e. phytochemicals) such as alkaloids, phenolic compounds, steroids, and tannins. Notable examples include morphine, quinine, vincristine, and taxol, which have significant pharmacological applications.^{2,3}

In recent years, scientific interest has surged in exploring medicinal plants for their anti-inflammatory, antimalarial, anticancer, antidiabetic, and antimicrobial properties.^{4,5} This growing research focus highlights their potential as sources of novel drugs, particularly for combating infectious diseases and cancer.

Tapinanthus globiferus (Loranthaceae), a widespread tropical African parasitic flowering plant, has, for example, traditionally been utilized in the management of various conditions like inflammation, bacterial infections, and diabetes.^{6,7} These traditional uses suggest the presence of secondary metabolites with potent antioxidant, antimicrobial, and anti-inflammatory properties. Ethno medicinal investigations indicate its utilization in conventional medication, but the single bioactive chemicals accountable for its pharmacological property are quite poorly explored. While previous phytochemical screening on *T. globiferus* identified the presence of general classes of compounds like alkaloids, flavonoids, tannins, and phenolic acids such as chlorogenic acid and rutin,⁸ there remains a significant gap in the systemic isolation, purification, and full structural elucidation of individual bioactive constituents from *T. globiferus*, particularly from its leaves.⁸ Such phytochemicals have been linked to antioxidant, antimicrobial, and anti-inflammatory properties that suggest a broad spectrum of pharmacological activity.⁹ Systematic isolation, structure elucidation, and contribution of its bioactive compounds to its chemotaxonomy, however, are limited, particularly from the leaves used in traditional medicine. Nuclear Magnetic Resonance (NMR) spectroscopy, specifically 2D techniques such as COSY and HMBC, is a definite pillar of the unambiguous structural assignment of complex natural products, allowing accurate assignment of all the atoms within a molecule.

This study aimed to isolate and characterize bioactive constituents from the leaves of *T. globiferus*. In this study, we report the isolation and identification of bioactive (i.e. catechin, catechin-3-gallate, 4-methoxy-phenyl-acryl aldehyde, and 3-hydroxy-4-methoxy-phenyl-acryl aldehyde) compounds from the leaves of *T. globiferus* using thin-layer chromatography (TLC) and gel filtration chromatographic techniques. The structural elucidation of these compounds not only expands the phytochemical profile of this species but also provides

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purified entities for subsequent *in vitro* and *in vivo* pharmacological investigations, potentially validating their traditional uses and computational predictions. The aim of this study is to isolate and structurally elucidate Flavan-3-ols and Phenylacrylaldehydes from the leaves of *Tapinanthus globiferus* (Loranthaceae) a host plant on *Vitex Doniana*

Materials and Methods

Collection, Identification, and Preparation of Plant Material

T. globiferus leaves were collected from its natural habitat in Hoto (Latitude: 11.978, Longitude: 8.5731), in Kano State, Nigeria. The identification and authentication were done at the Herbarium Unit, Department of Botany, Ahmadu Bello University, Zaria, by Mal. Namadi Sanusi via comparison with herbarium voucher specimen (No. ABU01052). The leaves were air-dried under shade at room temperature and pulverized using a mortar and pestle and subsequently referred to as powdered material.

Chemicals

All organic solvents used for extraction and fractionation—including methanol (analytical grade, purity $\geq 99.8\%$, Sigma-Aldrich, St. Louis, MO, USA), hexane ($\geq 99\%$), chloroform ($\geq 99.9\%$), ethyl acetate ($\geq 99.8\%$), and butanol ($\geq 99.7\%$)—were of laboratory grade obtained from Sigma Aldrich (St. Louis, MO, USA).

Extraction and Partitioning

The powdered material (2.0 kg) was extracted using cold maceration method with 10 L of aqueous methanol (70%) for 72 hours with occasional shaking. The extract was evaporated *in vacuo* using a rotary evaporator (BUCHI Rotavapor, Switzerland) at 40°C to yield crude methanol extract (CME, 72.0 g). About 71.20 g of the CME was subjected to partitioning successively with hexane, chloroform, ethyl acetate, and n-butanol in order of increasing polarity. This yielded the corresponding fractions: hexane fraction (22.0 g), chloroform fraction (11.0 g), ethyl acetate fraction (10.20 g), and n-butanol fraction (18.60 g) respectively, which were stored inside a desiccator. The ethyl acetate and chloroform fractions were subjected to column chromatographic analysis based on preliminary TLC profiles.

Isolation of compounds from *T. globiferus* Fractions

Column chromatographic analysis of ethyl acetate fraction

The ethyl acetate fraction (8.0 g) was laden on 8.0 g silica gel (60-120 mesh size) and chromatographed on a pre-packed column with 400 g silica gel (75 cm \times 3.5 cm diameter). The column was eluted under gradient elution conditions starting with 100% chloroform, followed by mixtures of chloroform: ethyl acetate with ratios increasing in polarity by the addition of 5% ethyl acetate till ethyl acetate: methanol at a ratio of 85:15. The column was finally washed with methanol 100%. A total of 80 fractions of 100 mL each was collected and pooled together based on similarity in TLC profile into a total of 12 major fractions labelled EAF1-EAF12. Sephadex LH-20 gel filtration of collection EAF9 with methanol as an eluting solvent led to the isolation of compound Y12 (9.0 mg). Fraction EAF11 was further purified on Sephadex LH-20 with methanol as an eluting solvent to obtain compound Y13 (10.5 mg).

Column chromatographic analysis of chloroform fraction

Chloroform fraction (9.0 g) was adsorbed onto 9.0 g of silica gel (60-120 mesh size), which was then chromatographed in a 75 cm \times 3.5 cm column pre-packed with 350 g of silica gel. Gradient elution was used to run the column, starting with 100% chloroform followed by chloroform:methanol mixtures in a ratio of increasing polarity adding 5% chloroform until a solvent system ratio of 70:30 was used, and the column was then washed with 100% methanol. A total of 140 collections of 100 mL each were made, and aliquots with similar TLC profiles were pooled together to obtain 20 major fractions coded CF1-CF20. Gel filtration and repeated gel filtration of collection CF12 over Sephadex LH-20 with methanol as an eluting solvent led to the

isolation of compound Y17 (10 mg Gel filtration and repeated gel filtration of collection CF18 over Sephadex LH-20 yielded Y18, an amorphous yellow substance with single spots (11.5 mg).

Characterization of compounds

The purity of all the isolated compounds was evaluated by thin layer chromatography (TLC) on silica gel F254 aluminium sheets (Merck, Germany). The isolated compounds were individually tested for physical properties such as color and appearance. In addition to this, a Chemical test was performed to check the purity of the isolated compounds by applying ferric chloride test and Shinoda test.¹⁰ The melting point test was performed on the isolated compounds using a Gallenkamp electro-thermal melting point apparatus (UK).

Spectroscopic Analysis

¹H-NMR and ¹³C-NMR (1D and 2D) spectra were done in *DMOS-d₆* with NMR spectrometer and were recorded on a Bruker DRX 300, 400 MHz (Bruker Bio Spin, Rheinstetten, Germany) at the Department of Chemistry, University of Pretoria, Faculty of Natural Resources, South Africa. The chemical shifts (δ) were reported in parts per million (ppm) relative to tetramethylsilane (TMS) as an internal standard, and the coupling constants (*J* values) were reported in hertz (Hz). For compound Y12, HMBC experiment was not performed due to sufficient structural information which was obtained from ¹H NMR, ¹³C-NMR, DEPT, and COSY experiments, which unambiguously established the structure when Y12 was compared with available literature data. Spectroscopic data for known compounds were compared with literature values for confirmation.

Results and Discussions

Structure elucidation of the isolated compounds viz. Y12, Y13, Y17, and Y18

The crude methanol extract was obtained through the cold maceration method using 70% methanol, which was fractionated sequentially using four different solvents to give n-hexane, chloroform, ethyl acetate, and n-butanol fractions in order of increasing polarity. The compounds catechin (Y12) and catechin-3-gallate (Y13) were isolated from the ethyl acetate fraction through column chromatography (CC) separation and Sephadex LH-20. The n-butanol fraction was separated with CC on silica gel and Sephadex LH-20 to give two compounds, 4-methoxy phenyl acryl aldehyde (Y14) and 3-hydroxy-4-methoxy phenyl acryl aldehyde (Y15). Four (4) compounds were suggested based on the NMR spectral of the isolated compounds Y12, Y13, Y17 (Figure S1-S19), and Y18 (Table 1-7) resulting from the elution of compounds in the chromatographic studies of ethyl acetate and n-butanol fractions.

Compound Y12 was obtained from the column chromatographic separation of the ethyl acetate fraction as a brown amorphous substance with a melting point of 179-182°C suggestive of a flavan-3-ol nucleus reported for a catechin.¹¹ A green coloration was observed with the ferric chloride reagent, indicating the presence of a free hydroxyl functional group in the nucleus.¹⁰

The proton NMR spectrum of compound Y12 revealed the presence of five (5) aromatic methine proton signals. Two meta-coupled protons at 5.89 (1H, d, *J* = 2.0 Hz) and 5.72 (1H, d, *J* = 2.0 Hz) were assigned to protons at H-6 and H-8 for a 5, 7-hydroxyl of the A ring in an AB ring system.^{11, 12} The ¹H NMR of compound Y12 displayed proton signals at 6.89 (1H, brs), 6.68 (1H, d, *J* = 2.4 Hz) and 6.66 (1H, brs) for a 1', 3', 4'-trisubstituted aromatic ring of an ABX rings system.^{10, 12} Other proton signal were also observed at 4.0 (1H, d, *J* = 4.4 Hz), 4.67 (1H, d, *J* = 4.4 Hz), which were assigned to two methine protons at H-3 and H-2,^{10, 13} suggesting that the flavan structure possesses a trans-2, 3 stereochemistry.^{10, 13}

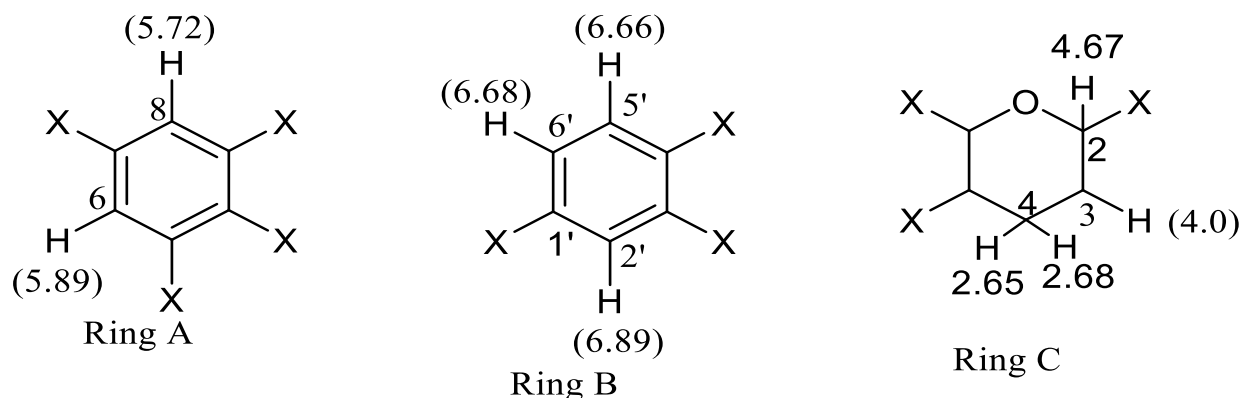
This was supported by the coupling constant (*J* = 4.4 Hz, H-3) as opposed to epicatechin (*J* < 1 Hz)^{11, 13}. Two proton signals were seen at 2.65 (1H, d, *J* = 2.0 Hz; β) and 2.68 (1H, d, *J* = 2.0 Hz; α) due to methylene protons at H-4, suggesting a methylene proton in the central ring C (Figure 1).

Table 1: Summary of 1D and 2D spectral data for compound Y12 (DMOS-d₆, 400 MHz).

Position	δ_H Y12 (J in Hz, DMOS-d ₆)	δ_C Y12 (ppm)	COSY	DEPT
2	4.67 (1H, d, J= 4.4 Hz)	78.51	H-3	CH
3	4.0 (1H, d, J= 4.4 Hz)	65.37	H-2, H-4	CH
4	2.68 (1H, d, J = 2.0Hz; H _a) 2.65 (1H, d, J = 2.0Hz; H _b)	28.67	H-3	CH ₂
5	-	156.99	-	-
6	5.89 (1H, d, J = 2.0 Hz)	95.52	H-8	CH
7	-	156.69	-	-
8	5.72 (1H, d, J = 2.0 Hz)	94.53	H-6	CH
9	-	156.24	-	-
10	-	98.94	-	-
1'	-	131.07	-	-
2'	6.89 (1H, brs)	115.21	H-6'	CH
3'	-	144.96	-	-
4'	-	144.90	-	-
5'	6.66 (1H, brs)	115.35	-	CH
6'	6.68 (1H, d, J = 2.4)	118.41	H-2'	CH

Table 2: Summary of 1D spectral data of compound Y13 (DMOS-d₆, 400 MHz).

Position	δ_H Y13 (J in Hz, DMOS-d ₆)	δ_C Y13 (ppm)	DEPT
2	5.0 (1H, s)	76.92	CH
3	5.35 (1H, s)	68.07	CH
4	2.94 (1H, dd, J = 4.4, 17.2 Hz, α) 2.68 (1H, d, J = 16.8 Hz, β)	26.11	CH ₂
5	-	156.08	-
6	5.94 (1H, d, J = 2.0 Hz)	95.98	CH
7	-	156.94	-
8	5.83 (1H, d, J = 2.0 Hz)	94.79	CH
9	-	157.0	-
10	-	97.73	-
1'	-	129.83	-
2'	6.84 (1H, d, J = 1.6 Hz)	114.72	CH
3'	-	145.19	-
4'	-	145.16	-
5'	6.65 (1H, d, J = 8.4 Hz)	115.52	CH
6'	6.75 (1H, dd, J = 1.6, 8.0 Hz)	118.02	CH
1''	-	119.65	-
2''/6''	6.82 (2H, s)	109.04	CH
3''/5''	-	145.58	-
4''	-	139.01	-
C=O	-	165.63	-

Fig. 1: Fragments from ^1H NMR of Y12Table 3: ^1H - ^1H COSY and HMBC correlation data of compound Y13 (DMOS- d_6 , 400 MHz)

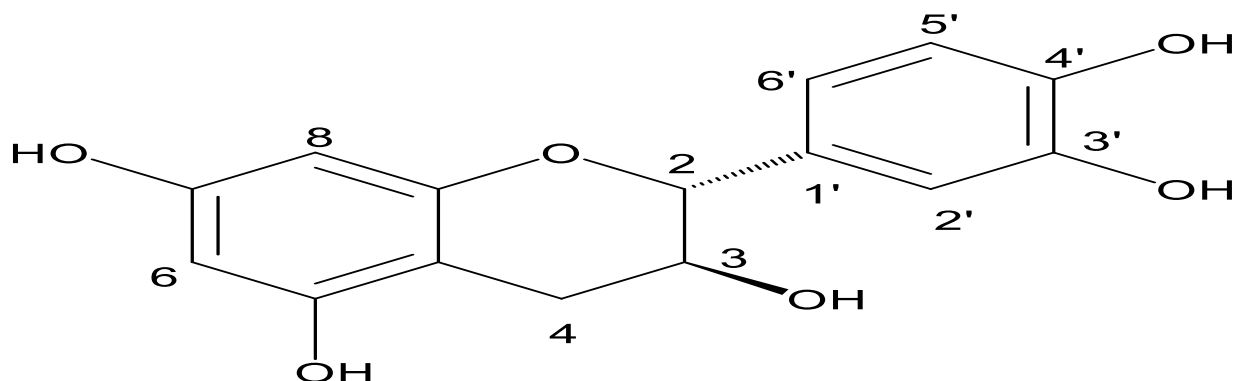
Protons	^1H - ^1H COSY	HMBC correlations	
		$^2J(\text{H}\rightarrow\text{C})$	$^3J(\text{H}\rightarrow\text{C})$
H-2	H-3	C-3, C-1'	C-2', C-6'
H-3	H-2, H-4	C-1''	C-10
H-4	H-3	C-3, C-10	C-2, C-5
H-6	H-8	C-5	C-8, C-10
H-8	H-6	C-9	C-10
H-2'	H-6'	C-1'	C-2, C-6', C-4'
H-5'	H-6'	-	C-1', C-3'
H-6'	H-5', H-2'	-	C-2, C-2'
H-2''/6''	-	C-1'', C-3''/5''	C-4'', C-1'''

The ^{13}C NMR spectra revealed the presence of 15 carbon signals¹⁰. ^{13}C signals at δ_{C} 156.99 (C-5), 156.69 (C-7), 144.96 (C-3'), and 144.90 (C-4') were due to oxygenated aromatic carbons.^{10, 12} Additionally, carbon signals were observed at δ_{C} 95.52 (C-6), 94.53 (C-8), 115.21 (C-2'), 115.35 (C-5'), and 118.41 (C-6') due to aromatic methine carbon atoms. Other carbon signals at δ_{C} 156.24 (C-9), 98.94 (C-10), and 131.07 (C-1') all show the presence of quaternary carbon atoms. Carbon signals at δ_{C} 78.51 (C-2) and 65.37 (C-3) were due to methine protons, and 28.67 (C-4) was due to the hetero cyclic C-ring of the molecule.

The DEPT-135 NMR spectra (400 MHz; DMOS- d_6) showed the presence of seven (7) methine carbon atoms and one methylene carbon

atom. The assignments of these carbons and the benzylic moieties within the compounds were achieved using 2D experiments.

The attachments between protons to their respective carbons were established with the use of the (HSQC). The ^1H - ^1H COSY spectrum was used to confirm the substitution patterns of the aromatic rings present in compound Y12, viz.; 1 tetra-substituted, 1 tri-substituted, and a hetero cyclic central ring. Visible correlations were those between H-6 and H-8, H-2' and H-6', H-3 and H-2, and H-3 and H-4. By comparing the 1D and 2D NMR spectroscopy data obtained for compound Y12 with literature values reported by Bila *et al.*¹⁰ the structural elucidation of compound Y12 was achieved, confirming its identity and consistency with previously characterized compounds (Figure 2).

Fig. 2: Chemical structure of compound Y12 (Catechin). Chemical formula: $\text{C}_{15}\text{H}_{14}\text{O}_6$

Compound Y13 was isolated as a brown amorphous substance suggestive of a flavan-3-*o*-glycoside nucleus reported for a catechin-3-

gallate.¹⁴ A pink coloration was observed when subjected to the Shinoda test, indicating that the compound has a flavonoid nucleus.¹¹

Table 4: Summary of 1D spectral data of compound Y17 (DMOS-d₆, 400 MHz).

Position	δ_H Y17 (J in Hz, DMOS-d ₆)	δ_C Y17 (ppm)	DEPT
1		127.91	-
2/6	7.65 (2H, d, $J = 8.0$ Hz)	128.86	CH
3/5	6.94 (2H, d, $J = 8.0$ Hz)	114.52	CH
4	-	160.48	-
7	7.78 (1H, brs)	139.63	CH
8	6.42	128.49	CH
9	10.10 (1H, brs)	-	-
OCH ₃	3.78 (3H, s)	55.67	CH ₃
C=O		192.26	-

Table 5: ¹H-¹H COSY and HMBC correlation data of compound Y17 (DMOS-d₆, 400 MHz)

Protons	¹ H- ¹ H COSY	HMBC correlations	
		² J (H→C)	³ J (H→C)
H-2/H-6	H-3, H-5	C-1, C-3/C-5	C-4
H-3/H-5	H-2, H-6	C-4	C-1
H-7		C-1	
H-8			C-1
H-9			C-7
OCH ₃		C-4	

Table 6: 1D spectral data of compound Y18 (DMOS-d₆, 400 MHz).

Position	δ_H Y18 (J in Hz, DMOS-d ₆)	δ_C Y18(ppm)	DEPT
1		128.05	-
2	6.93 (1H, brs)	114.43	CH
3	-	156.9	-
4	-	160.41	-
5	7.42 (1H, d, $J = 8.0$ Hz)	129.51	CH
6	8.24 (1H, d, $J = 8.0$ Hz)	128.68	CH
7	8.18 (1H, brs)	134.37	CH
8	7.62	128.48	CH
9	10.60 (1H, brs, COH)	-	-
OCH ₃	3.77	55.68	CH ₃
CO		195.65	-

The proton NMR spectrum of compound Y13 revealed the presence of six (6) aromatic methine proton signals. Two meta-coupled protons at δ_H 5.94 (1H, d, $J = 2.0$ Hz) and 5.83 (1H, d, $J = 2.0$ Hz) were assigned to proton at H-6 and H-8 for a 5, 7-dihydroxyl of the A ring in an AB ring system.^{10, 12, 14} The ¹H NMR of compound Y13 displayed proton signals at δ_H 6.84 (1H, d, $J = 1.6$ Hz), 6.75 (1H, dd, $J = 1.6, 8.0$ Hz) and 6.65 (1H, d, $J = 8.4$ Hz) for a 1', 3', 4'-trisubstituted aromatic ring of an ABX ring system.^{10, 12, 15} Additionally, one aromatic proton signal was observed at δ_H 6.82 (2H, s) assigned to the tetra-substituted ring (Figure 3).¹⁴ Other proton signals were also observed at 5.0 (1H, s), 5.35 (1H, s) which were assigned to two methine protons at H-2 and H-3,¹³ suggesting that the flavan structure possesses a trans-2, 3 stereochemistry.¹⁴ Two proton signals were seen at 2.68 (1H, d, $J = 16.8$ Hz; β) and 2.94 (1H, d, $J = 17.2$ Hz; α) due to methylene protons at H-4, suggesting a methylene proton in the central ring C (Figure 3).

The ¹³C NMR spectra revealed the presence of 20 carbon signals which are consistent with a catechin gallate.¹⁴ The carbon signal at δ_C 165.63 (C-1'') was assigned to the carbonyl carbon of the ring D. The carbon signals at δ_C 156.08 (C-5), 156.94 (C-7), 145.16 (C-3'), 145.19 (C-4'), 145.58 (C-3''/5''), and 139.01 (C-4'') were due to oxygenated aromatic carbons^{10, 12, 14} Additionally, carbon signals were observed at δ_C 95.98 (C-6), 94.79 (C-8), 114.72 (C-2'), 115.52 (C-5'), and 118.02 (C-6'') due to aromatic methine carbon atoms. Other carbon signals at δ_C 156.24 (C-9), 98.94 (C-10), 131.07 (C-1'), and 109.04 (C-1'') all show the presence of quaternary carbon atoms. Carbon signals at δ_C 76.92 (C-2) and 68.07 (C-3) were due to methine protons, and 23.95 (C-4) of the hetero cyclic C-ring of the molecule.

The DEPT-135 NMR spectra of compound Y13 (400 MHz; DMOS-d₆) showed the presence of eight methine carbon atoms and one methylene carbon atom. The assignments of these carbons and the benzylic moieties within the compounds were achieved using 2D experiments.

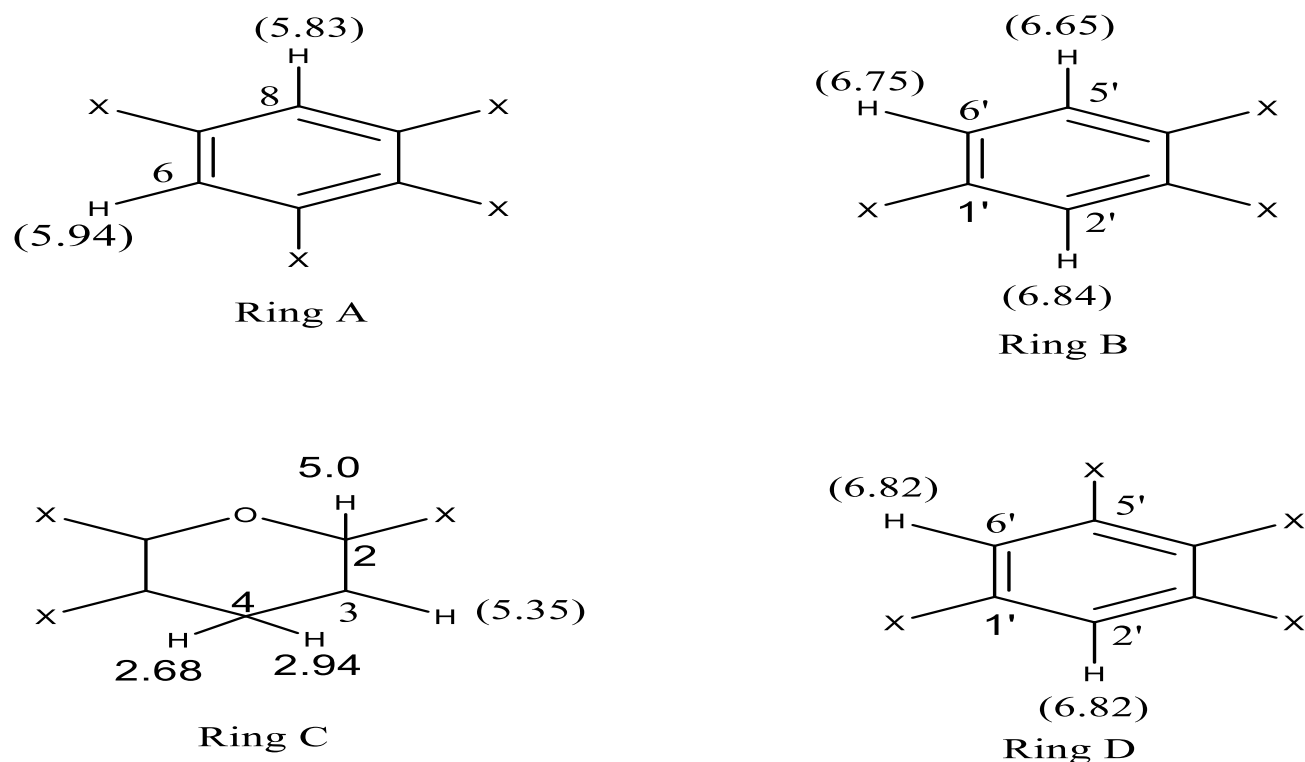


Fig. 3: Fragments from ^1H NMR of Y13

The attachments between protons to their respective carbons were established with the use of the (HSQC). The ^1H - ^1H COSY spectrum was used to confirm the substitution patterns of the aromatic rings present in compound Y13, viz.; 1 tetra-substituted, 1 tri-substituted, and a hetero cyclic central ring. Visible correlations were those between H-6 and H-8, H-2' and H-6', H-6' and H-5', H-3 and H-2, and H-3 and H-4.

In the HMBC experiment of compound Y13, the *meta*-coupled protons of the A ring at H-6 (δ_{H} 5.94; δ_{C} 95.98 from HSQC) and H-8 (δ_{H} 5.83; δ_{C} 94.79 from HSQC) exhibited a common 3J correlation to an aromatic quaternary carbon at δ_{C} 97.73 (C-10) from the HMBC spectrum. The proton at H-6 also has a 2J correlation with another oxygenated quaternary carbon at δ_{C} 156.08 (C-5) and a 3J correlation to an aromatic methine carbon at δ_{C} 94.79 (C-8). The proton at H-8 also has a 2J correlation with aromatic quaternary carbon at δ_{C} 157.0 (C-9).

For the *meta*-coupled protons for the B ring at H-2' (δ_{H} 6.84; δ_{C} 114.72 from HSQC) and H-6' (δ_{H} 6.75; δ_{C} 118.02 from HSQC), a common 3J correlation to a hetero cyclic methine carbon at δ_{C} 76.92 (C-2) and an aromatic methine carbon at δ_{C} 114.72 (C-2'). Also, the proton at H-2' has a 2J correlation to a quaternary aromatic carbon at δ_{C} 129.83 (C-1') was exhibited.

For the *ortho*-coupled protons at H-6' (δ_{H} 6.75; δ_{C} 118.02 from HSQC) and H-5' (δ_{H} 6.65; δ_{C} 115.52 from HSQC), hydrogen H-5' has 3J correlations to a carbon at δ_{C} 129.83 (C-1') and 145.19 (C-3'). The proton for the ring D at H-2'/6'' (δ_{H} 6.82; δ_{C} 109.04 from HSQC) has a 2J correlation to a quaternary aromatic carbon at δ_{C} 119.65 (C-1'') and an oxygenated aromatic carbon at δ_{C} 145.58 (C-3''/5''). The proton at H-2'/6'' also shows a 3J correlation to an oxygenated aromatic carbon at δ_{C} 139.01 (C-4'') as well as a carbonyl carbon at δ_{C} 165.63 (C-1''') confirming the presence of a carbonyl group on the D ring. The proton at H-3 (δ_{H} 5.352, δ_{C} 68.07 HSQC) shows a 2J correlation to a quaternary carbon at δ_{C} 97.73 (C-10) and a quaternary carbonyl carbon of ring D at δ_{C} 165.63 (C-1''') confirming the placement of the gallate at C-3 of the central ring C. Other HMBC correlations observed were at H-2 (δ_{H} 5.0, δ_{C} 76.92 from HSQC), which has a 2J correlation with carbons at δ_{C} 68.07 (C-3) and 129.83 (C-1'). It also

exhibited a 3J correlation with aromatic methine carbon at δ_{C} 114.72 (C-2') and 118.02 (C-6') of the B ring.

Figure 4, which depicts the chemical structure of Y13 (catechin-3-gallate), illustrates the ester linkage at the C-3 position of the catechin moiety with gallic acid. This structural feature is significant as it often enhances the biological activity, particularly the antioxidant capacity, compared to the parent catechin.¹⁴

Based on its physical properties, chemical tests, spectral analysis, and comparison with literature,¹⁴ the compound was identified as catechin gallate (Figure 4).

Compound Y17 was isolated as an off-white amorphous substance suggestive of a coumaric acid nucleus.¹⁶ The proton NMR spectrum of compound 17, revealed the presence of four (4) aromatic methine protons signal. Two *ortho* coupled protons at δ_{H} 7.65 (2H, d, $J = 8.0$ Hz) and 6.94 (2H, d, $J = 8.0$ Hz) were assigned to proton at H-2/6 and H-3/5 for a 1, 4 di-substituted aromatic of the A ring in an AA BB' ring system.^{12, 16} The ^1H NMR of compound Y17 displayed proton signals at δ_{H} 7.78 (1H, brs) and 6.42 (1H, brs) for an olefinic system in a *trans* configuration. Additionally, one proton signal was observed at δ_{H} 10.10 (1H, s) assigned to an aldehyde proton of CO-H. Other proton signal was also observed at 3.78 (3H, s) which was due to a methoxy proton as shown in fragments A and B (Figure 5).

The ^{13}C NMR spectra revealed the presence of 8 carbons signals which are consistent with a family of acryl aldehyde or coumaric acid. The carbon signal at δ_{C} 192.26 (C-9) was assigned to the carbonyl carbon of the molecule for an aldehyde group. The carbon signals at δ_{C} 160.48 (C-4) is due to methylated oxygenated aromatic carbon.¹⁶ Additionally, carbon signals were observed at δ_{C} 114.52 (C-3/5) and 128.86 (C-2/6) were due to aromatic methine carbon atoms. Other carbon signal at δ_{C} 128.49 (C-8) and 139.63 (C-7) were due to olefinic carbons. Additionally, signal at δ_{C} 127.91 (C-1) was due to quaternary carbon atom and 55.67 (C-1') was due to aromatic methoxy carbon atom of the molecule.

The DEPT-135 NMR spectra of compound Y17 (400 MHz; DMOS- d_6) showed the presence of four methine carbon atoms and one methoxy carbon atom. The assignments of these carbons and the benzylic moiety within the compounds were achieved using 2D experiments.

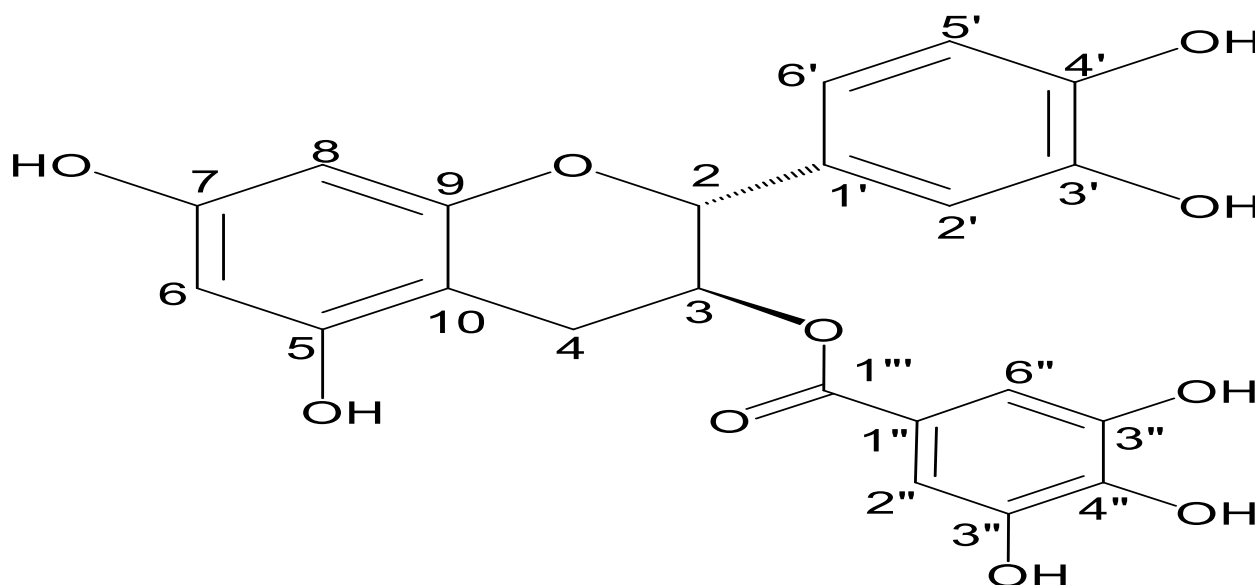


Fig. 4: Chemical structure of compound Y13 (Catechin-3-gallate). Chemical formula: $C_{22}H_{18}O_{10}$

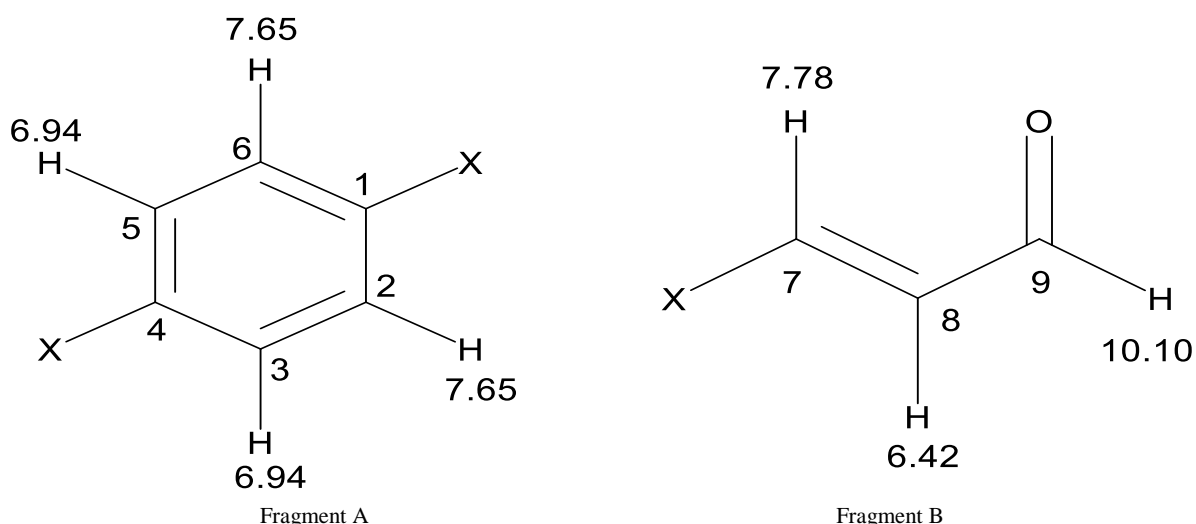


Fig. 5: Fragments from 1H NMR of Y17

The attachments between protons to their respective carbons were established with the use of the (HSQC). The 1H - 1H COSY spectrum was used to confirm the substitution patterns of the aromatic ring present in compound Y17, viz. 1 di-substituted ring. Visible correlations were those between H-6 and H-5.

In the HMBC experiment of compound Y17, the ortho-coupled protons at H-2/6 (δ_H 7.65; δ_C 128.86 from HSQC) and H-3/5 (δ_H 6.94; δ_C 114.52 from HSQC), proton H-2/6 has a 3J correlation to a carbon at δ_C 160.48 (C-4) and has a 2J correlation to a quaternary aromatic carbon at δ_C 127.91 (C-1) and an aromatic methine carbon at δ_C 114.52 (C-3/5). The ortho proton at H-3/5 also shows a 3J correlation to an aromatic methine carbon at δ_C 127.91 (C-1) as well as a 2J correlation to quaternary aromatic carbon at δ_C 160.48 (C-4). The trans olefinic protons at H-7 (δ_H 7.78; δ_C 139.63 from HSQC) and H-8 (δ_H 6.42; δ_C 128.49 from HSQC), proton H-7 has a 2J correlation to a quaternary aromatic carbon at δ_C 127.91 (C-1) and proton at H-8 has a 3J correlation with δ_C 127.91 (C-1), confirming the presence of the attachment of the fragment B. The proton at H-1' (δ_H 3.78, δ_C 55.67 HSQC) shows a 2J correlation to a quaternary carbon at δ_C 160.48 (C-4) confirming the placement of the methoxy at C-4 of the fragment A. Based on its physical properties, and spectral analysis, the compound was identified as 4-methoxy phenyl acryl aldehyde.¹⁶ (Figure 6).

Compound Y18 was obtained from the column chromatographic separation of the chloroform fraction as a white amorphous substance suggestive of a coumaric acid.¹⁶

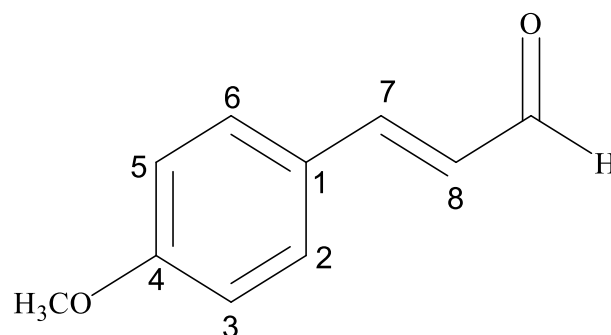


Fig. 6: Chemical structure of compound Y17 (4-methoxy phenyl acryl aldehyde). Chemical formula: $C_{10}H_{10}O_2$

The proton NMR spectrum of compound Y18 revealed the presence of three (3) aromatic methine proton signals. One ortho-coupled proton at δ_H 8.24 (1H, d, J = 8.0 Hz) and 7.42 (1H, d, J = 8.0 Hz) was assigned to protons at H-6 and H-5, and a proton at δ_H 6.93 (1H, d, J = 8.0 Hz;

H-2) for a 1, 3, 4-trisubstituted aromatic of the A ring in an ABX ring system.^{12, 16} The ¹H NMR of compound Y18 displayed proton signals at δ_H 8.18 (1H, brs) and 7.62 (1H, brs) for an olefinic system in a trans configuration. Additionally, one proton signal was observed at δ_H

10.60 (1H, s) assigned to an aldehyde proton of CO-H. Another proton signal was also observed at 3.77 (3H, s), which was due to a methoxy proton as shown in fragments A and B (Figure 7).

Table 7: ¹H-¹H COSY and HMBC correlation data of compound Y18 (DMOS-d₆, 400 MHz)

Protons	¹ H- ¹ H COSY	HMBC correlations	
		² J(H→C)	³ J(H→C)
H-2		C-1	C-3
H-5	H-6	C-6	
H-6	H-5	C-5	C-7
H-7			C-6
H-8		C-7	C-1
H-9			C-7
OCH ₃		C-4	C-3

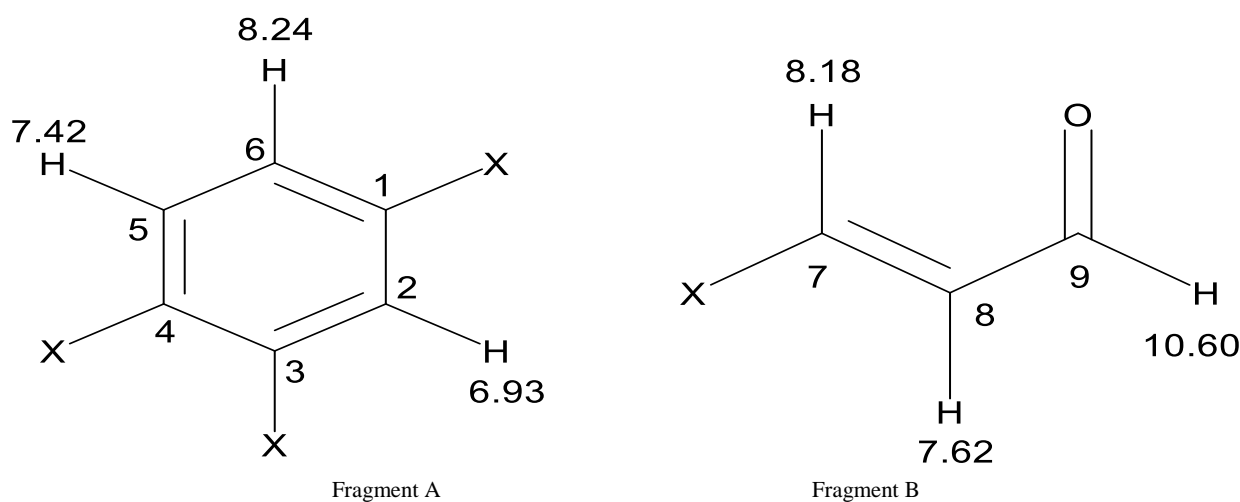


Fig. 7: Fragments from ¹H NMR of Y18

The ¹³C NMR spectra revealed the presence of 10 carbon signals which are consistent with a family of acryl aldehyde or coumaric acid. The carbon signal at δ_C 195.65 (C-9) was assigned to the carbonyl carbon of the molecule for an aldehyde group. The carbon signal at δ_C 160.41 (C-4) is due to methylated oxygenated aromatic carbon,¹⁶ and 156.9 (C-3) was due to the oxygenated aromatic carbon atom. Additionally, carbon signals were observed at δ_C 114.43 (C-2), 128.68 (C-6), and 129.51 (C-5) due to aromatic methine carbon atoms. Other carbon signals at δ_C 128.48 (C-8) and 134.37 (C-7) were due to olefinic carbons. Additionally, the signal at δ_C 128.05 (C-1) was due to the quaternary carbon atom, and 55.68 (C-1') was due to the aromatic methoxy carbon atom of the molecule.

The DEPT-135 NMR spectra of compound Y18 (400 MHz; DMOS-d₆) showed the presence of five methine carbon atoms and one methoxy carbon atom. The assignments of these carbons and the benzylic moiety within the compounds were achieved using 2D experiments.

The attachments between protons to their respective carbons were established with the use of the (HSQC). The ¹H-¹H COSY spectrum was used to confirm the substitution patterns of the aromatic ring present in compound Y18, viz.; 1 tri-substituted ring. Visible correlations were those between H-6 and H-5.

In the HMBC experiment of compound Y18, the ortho-coupled protons at H-6 (δ_H 8.24; δ_C 128.68 from HSQC) and H-5 (δ_H 7.42; δ_C 129.51 from HSQC), proton H-6 has a ³J correlation to a carbon at δ_C 134.37 (C-7) and has a ²J correlation to an aromatic methine carbon at δ_C 129.51 (C-5). The ortho proton at H-5 also shows a ²J correlation to

an aromatic methine carbon at δ_C 128.68 (C-6). The trans olefinic protons at H-7 (δ_H 8.18; δ_C 134.37 from HSQC) and H-8 (δ_H 7.62; δ_C 128.48 from HSQC), proton H-7 has a ²J correlation to a quaternary aromatic carbon at δ_C 127.91 (C-1), and proton at H-8 has a ²J correlation to an aromatic methine carbon at δ_C 134.37 (C-7) and also shows a ³J correlation at δ_C 127.91 (C-1), confirming the presence of the attachment of fragment B. The proton at H-1' (δ_H 3.77, δ_C 55.68 HSQC) shows a ²J correlation to a quaternary carbon at δ_C 160.41 (C-4), confirming the placement of the methoxy at C-4 of the fragment A, and also a ³J correlation to an oxygenated carbon atom at δ_C 156.91 (C-3).

Based on its physical properties, and spectral analysis, the compound was identified as 3-hydroxy-4-methoxy acryl aldehyde (Figure 8).¹⁶ The isolation of the phenylacrylaldehydes Y17 and Y18, though not novel from a global phytochemical perspective, is reported for the first time from *Tapinanthus globiferus*. This finding expands the chemotaxonomic profile of the genus *Tapinanthus* and supports its traditional use in managing infections and inflammation. Cinnamaldehyde derivatives are well-documented for their antimicrobial, anti-inflammatory, and antidiabetic properties.¹⁶ Their presence in *T. globiferus* provides a plausible chemical basis for its ethnomedicinal applications and warrants further targeted pharmacological evaluation.

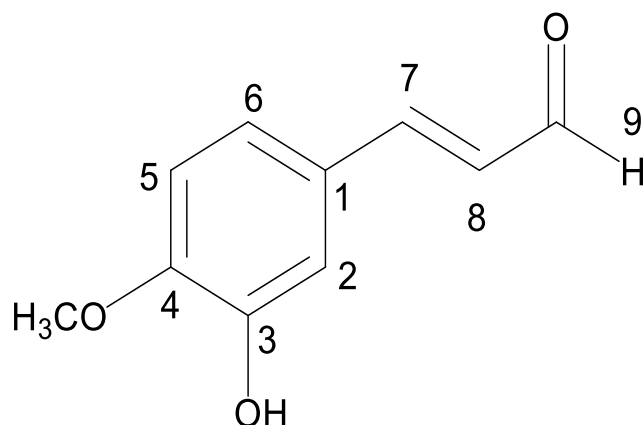


Fig. 8: Chemical Structure of Y18 (3-hydroxy-4-methoxy acryl aldehyde)

Conclusion

This study led to the isolation and characterization of four bioactive compounds—catechin (Y12), catechin-3-gallate (Y13), 4-methoxyphenylacrylaldehyde (Y17), and 3-hydroxy-4-methoxyphenylacrylaldehyde (Y18)—from *Tapinanthus globiferus* leaves using a combination of chromatographic and spectroscopic techniques. This is the first report of these specific phenylacrylaldehydes in the species, contributing to its phytochemical inventory. The presence of known antioxidant and anti-inflammatory activities of flavan-3-ols and cinnamaldehyde corroborates the use of the plant in folk medicine for treating inflammation and infections. These findings endorse the species as a lead source of bioactive compounds and call for further pharmacological studies to validate its therapeutic potential geared towards conducting *in vivo* trials to investigate efficacy and safety, semi-synthetic modification of the new phenylacrylaldehydes with a view to improving bioactivity, and bioprospecting of other species of *Tapinanthus* to seek out further new compounds with potential synergy.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article are original and that any liability for claims relating to the content of this article will be borne by them.

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