International Research Journal of Pure and Applied Chemistry

Volume 25, Issue 6, Page 41-68, 2024; Article no.IRJPAC.126416
ISSN: 2231-3443, NLM ID: 101647669

Identification of Compounds in the Ethyl Acetate Fraction of *Hyptis suaveolens* Leaves by HPLC-ESI- Q-TOF -MS/ MS Analysis

Mabintou Kalo [a,b*], Amon Diane Marina N'Tamon [c,d], Brahima Soumahoro [e,b*], Ali Sanou [e,b], Philatryho Atché [e,b], Yaya Soro [e,b] and Barthélémy Attioua [f,g]

[a] Sciences and Technology Training and Research Unit, UFR-ST, University Alassane Ouattara, Bouaké, Ivory Coast.
[b] Laboratory of Industrials Synthesis Processes, Environment and New Energies (LAPISEN), National Polytechnic Institute Felix Houphouët-Boigny, Yamoussoukro, Ivory Coast.
[c] Department of Analytical, Mineral and General Chemistry, Food Technology, Felix Houphouët-Boigny University, Abidjan, Ivory Coast.
[d] UFR Pharmaceutical and Biological Science, Felix Houphouët-Boigny University, Abidjan, Ivory Coast.
[e] Department of Agro-food Chemical Engineering, National Polytechnic Institute Felix Houphouët-Boigny, Yamoussoukro, Ivory Coast.
[f] Department of Physics Chemistry, Felix Houphouët-Boigny University, Abidjan, Ivory Coast.
[g] Laboratory of Constitution and Matter Reaction (LCRM), UFR-SSMT, Felix Houphouët Boigny University, Abidjan, Ivory Coast.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: https://doi.org/10.9734/irjpac/2024/v25i6884

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/126416

**Corresponding author: E-mail: kalomabintou@gmail.com, brahima.soumahoro@inphb.ci;*

Cite as: *Kalo, Mabintou, Amon Diane Marina N'Tamon, Brahima Soumahoro, Ali Sanou, Philatryho Atché, Yaya Soro, and Barthélémy Attioua. 2024. "Identification of Compounds in the Ethyl Acetate Fraction of Hyptis Suaveolens Leaves by HPLC-ESI- Q-TOF -MS/ MS Analysis". International Research Journal of Pure and Applied Chemistry 25 (6):41-68. https://doi.org/10.9734/irjpac/2024/v25i6884.*

Original Research Article

Received: 15/09/2024
Accepted: 18/11/2024
Published: 28/11/2024

ABSTRACT

Hyptis suaveolens is a plant of Ivory Coast flora. Preliminary studies have shown that the plant's leaves possess multiple medicinal properties, certainly due to the presence of terpenic and phenolic compounds. Yet, the plant's leaves compounds structures have been fewest elucidated. This study aim to determine the structures of a number of known compounds present in the ethyl acetate fraction obtained from the hydroethanol extract of *Hyptis suaveolens* leaves. Thus, fifteen known compounds structures were identified in ethyl acetate fraction of the plant's leaves by dereplication method. Nine of these compounds, are phenolic compounds and the others are terpenoids. Five of these compounds had not yet been elucidated in the leaves of *Hyptis suaveolens*. All these bioactive molecules in *Hyptis suaveolens* leaves makes this plant an alternative for combating many diseases caused by oxidative stress.

Keywords: Hyptis suaveolens; dereplication; compound structures; ethyl acetate fraction.

1. INTRODUCTION

Hyptis suaveolens is a plant used for these medicinal properties (Tang G and al, 2018). It's used to treat gastrointestinal infections, antirheumatics, respiratory ailments, antisuporifics, colds, indigestion, multiple skin complications, fever, burns, wounds, cramps and abdominal pain (Shah K and al, 2024, Mahesh S and al, 2001, Oliveira MJ and al, 2006). Preliminary analyses have shown that *Hyptis suaveolens* leaves contain alkaloids, terpenoids and phenolic compounds (Goly KRC and al, 2015, Kumar S and al, 2015). In addition, the hydroethanol extract and ethyl acetate fraction of the plant's leaves have shown very good antibacterial (Goly KRC and al, 2015) and antioxidant properties (Soumahoro B and al, 2023). The identification of the structures of the compounds present in the plant leaves extracts and fractions could serve as a database for synthesis and retrosynthesis in the fight against oxidative stress, which is the main cause of diseases. Thus, recent studies on dichloromethane fractions have elucidated the structures of eleven molecules, including five phenolic compounds and six terpene compounds (Soumahoro B and al, 2023, Soumahoro B and al, 2024). However, the molecular structures of the ethyl acetate fractions have not been studied. This study aim to determine a number of compounds structures from ethyl acetate fraction of *Hyptis suaveolens* leaves.

2. MATERIALS AND METHODS

2.1 Materials

2.1.1 Plant material

The *Hyptis suaveolens* leaves collected in July 2017 in Yamoussoukro (6047'18.762'North and 5015'25.9992' West) in Ivoiry Coast center, have been identified by Mr Amani N'Guessan, Institut National Polytechnique Felix HOUPHOUËT-BOIGNY's (INP-HB) botanist. The plant specimen is listed in the CSRS herbarium under number: n° Coll: 18027 / bdcsrs: 65599. After drying in the shade at room temperature (26 to 30°C) for 7 days, the leaves were crushed by an electric grinder (IKA M20, France) and the obtained powder was subdued with screen 0.5 mm mesh. The sieved powder was stored in darkened jar at 4°C until further use.

2.1.2 Experimental equipment

The dereplicative analysis, used an Agilent 1260 Infinity HPLC system coupled to an Agilent 6530 Q-TOF-MS mass spectrometer, equipped with an ESI source. A Sunfire® C18 analytical column (150×2.1 mm; 3.5 µm, Waters) was used.The analyses making in positive ion mode, used purine $C_5H_4N_4$ (ion at m/z 121.050873 g/mol) and phosphagen $C_{18}H_{18}F_{24}N_3O_6P_3$ (ion at m/z 922.009798) as internal locking masses. Scans were acquired at 11000 (at m/z 922) resolution.

2.2 Methods

2.2.1 Sample preparation

Soumahoro and al (2023) method was used to prepare total hydroethanolic extract. 100g of sample crushed material was macerated in 1L of a mixture (70/30: v/v) ethanol/water for 24 hours under magnetic stirrer. Next, the mixture was filtered with wool cotton and N°2 Watman paper. After, obtained filtrate was concentrated at 40°C using a BUCHI 461 rotary evaporator and then freeze-dried to give the total hydroalcoholic extract. This hydroalcoholic extract was fractionated using increasing polarity solvents (hexane, dichloromethane, ethyl acetate, ethanol and water) (Bouamama H and al, 2006). The extract (10 g) was dissolved in 100 mL of water and partitioned respectively with hexane, dichloromethane and ethyl acetate (Soumahoro B and al, 2023). The different organic phases were dried separately over anhydrous sodium sulphate. Then, they were filtered and concentrated under reduced pressure to give hexane (F_{HEX}), dichloromethane (F_{DCM}) and ethyl acetate (F_{AE}) fractions. Thereafter, 5 mg of ethyl acetate fraction was dissolved in 1 mL of analytical methanol then, filtered again using a 0.5 µm filter syringe. Ultimately, 300 µL of this filtrate were stored in a case for the HPLC-QTOF-MS/MS analysis.

2.2.2 Dereplicative analysis of the ethyl acetate fraction

Dereplicative analysis permit a rapid identification of known compounds in a complex mixture (Rivaro P and al, 2024, Jongmin A and al, 2017). This method use High Performance Liquid Chromatography (HPLC) coupled to Tandem Mass Spectrometry (MS/MS or MS2) /Q-TOF (McFarland K, Mulholland DA, 2004).

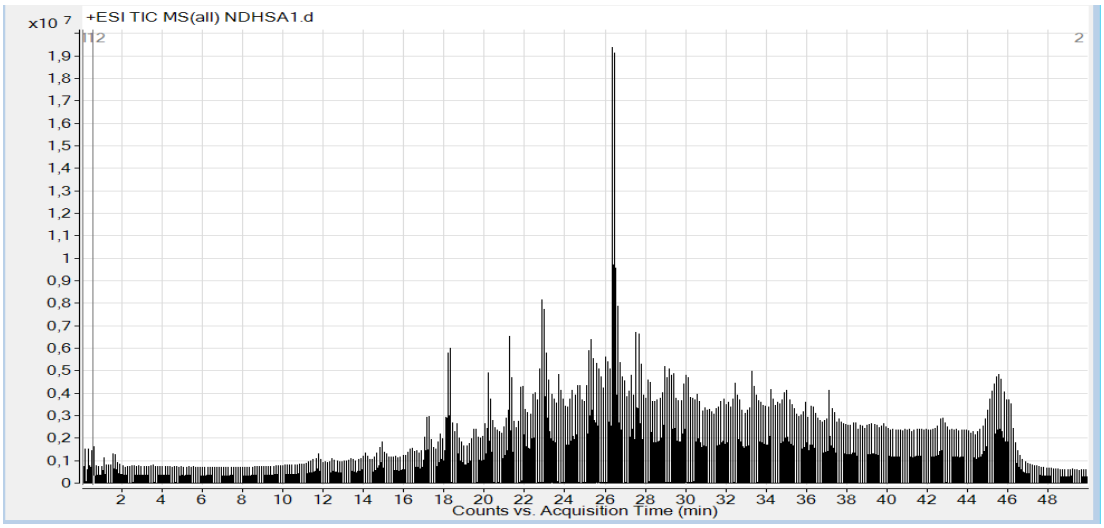


Fig. 1. ESI/MS chromatography profile

Table 1. Formulas suggested

Best	ID Source	Formula	Species	m/z	Score	Diff (ppm)	Score (MFG)	Mass (MFG)	DBE
●	MFG	C20 H30 O2	(M+H)+	303.2318	98,33	-0,24	98,33	302,2246	6
○	MFG	C18 H28 N3 O	(M+H)+	303.2318	89,9	-4,92	89,9	302,2232	6,5
○	MFG	C13 H30 N6 S	(M+H)+	303.2318	83,53	0,58	83,53	302,2253	2
○	MFG	C15 H32 N3 O S	(M+H)+	303.2318	78,37	5,35	78,37	302,2266	1,5
○	MFG	C16 H26 N6	(M+H)+	303.2318	72,65	-9,64	72,65	302,2219	7
○	MFG	C17 H34 O2 S	(M+H)+	303.2318	65,35	10,08	65,35	302,228	1
○	MFG	C19 Cl3 O9 S4	(M+2H)+2	303.3823	46,84	-1,57	46,84	604,7491	18,5
○	MFG	C14 H2 Cl3 N3 O8 S5	(M+2H)+2	303.3823	46,61	1,79	46,61	604,7511	14
○	MFG	C15 Cl3 O14 S3	(M+2H)+2	303.3823	45,56	2,57	45,56	604,7516	14,5
○	MFG	C14 H Cl2 N O12 S5	(M+2H)+2	303.3823	43,98	-3,45	43,98	604,7479	14
○	MFG	C16 H4 Cl3 O9 S5	(M+2H)+2	303.3823	42,77	4,01	42,77	604,7524	13,5
○	MFG	C23 Cl3 O4 S5	(M+2H)+2	303.3823	38,31	-5,7	38,31	604,7466	22,5
○	MFG	C17 Cl3 N4 O5 S5	(M+2H)+2	303.3823	36,77	6,22	36,77	604,7538	18,5

The column (150×2.1 mm; 3.5 µm, Waters) used is a Sunfire® C18 in positive ions mode. It use a flow rate (250 µL/min) and a two-way linear gradient; way A (95-0% H_2O plus 0.1% formic acid) and way B (5-100% ACN) for 30 minutes. Electronic souch ionization (ESI) was set at 320°C, 3.5 kV, and 10 µL/min for the gas flow rate. Purine $C_5H_4N_4$ (ion at m/z 121.050873 g/mol) and phosphagen $C_{18}H_{18}F_{24}N_3O_6P_3$ (ion at m/z 922.009798) were used as internal lock masses. Scans were acquired at a resolution of 11000 (at m/z 922). Sample injection volume was set at 5 µL.

The ethyl acetate fraction analyse use the HPLC-ESI-Q-TOF-MS/MS method. Then, the chromatogram automated integration using MassHunter® (Agilent) Qualitative Analysis B.07.00 software, gave the peaks of the main compounds from this fraction (Fig. 1).

Using the MassHunter software the given peak generates a formulae according to the single molecular ion $[M+H]^+$ (Table 1).

3. RESULTS AND DISCUSSION

3.1 HPLC-MS/Q-TOF Analysis of the Ethyl Acetate Fraction

The HPLC-ESI-Q-TOF-MS/MS analysis of the ethyl acetate fraction of *Hyptis suaveolens* leaves was carried. The aim was to elucidate the compounds structures in this plant leaves.

The ethyl acetate fraction HPLC-MS/Q-TOF chromatographic profiles are shown on Fig. 2.

Fig. 2 shows that the secondary metabolites leave the column in the time interval 1.56-45.58 min.

Table 2 gives the crude formula and corresponding molecular weights with the different scores for each of the main compounds detected.

Table 2 shows that fifteen compounds founded in ethyl acetate fraction (Table 3) have been already identified in Hyptis genus (Tang G and al, 2018, Suárez-Ortiz GA and al, 2017, Mukherjee K and al, 1984).

3.2 The Known Compounds (15) Structures Confirmation

The compounds structures were determined by interpreting ethyl acetate fraction HPLC-ESI-MS/Q-TOF analyses data provided.

These analysis provided mass and fragmentation spectra and crude formulae for several major compound.

3.2.1 Compound 1 structure

The compound 1(retention time: 17.95 min) corresponds to molecular ion $[M+H]^+$ with mass = 177.0545. Its molecular molar mass is therefore 176.084 g/mol and the most probable molecular formula (with the highest score) is $C_{11}H_{12}O_2$ (cal. 176.215). The fragmentation spectrum of compound 1 gives the chromatographic profile SM with the molecular ion (mass = 177) and collision-induced dissociation fragments of the molecular ion (Fig. 3).

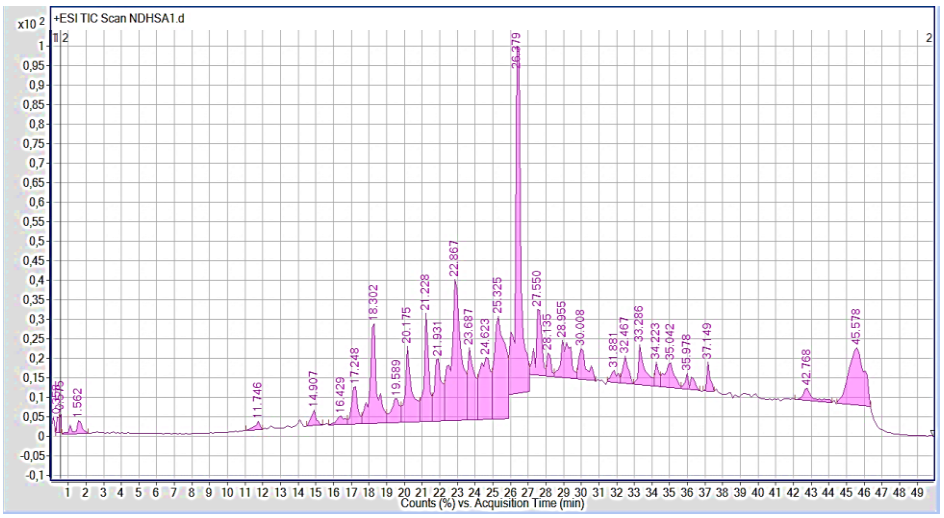


Fig. 2. Ethyl acetate fraction major compounds ESI/MS chromatographic profile

Table 2. Compounds detected in ethyl acetate fraction (FAE1)

PIC number	Retention time (min)	Formula	Molecular mass (g/mol)	Score (%)
1	1,562	$C_{25}H_{34}N_4$	390	98,51
2	11,746	$C_{24}H_{38}O_4$	390	97,15
3	14,907	$C_7H_{14}O_6$	194	86,17
4	16,429	$C_9H_6O_4$	178	86,31
5	17,248	$C_9H_6O_3$	162	98,03
6	17,951	*$C_{11}H_{12}O_2$	176	47,48
7	18,302	$C_{14}H_{12}N_2$	208	87,19
8	19,589	$C_{24}H_{38}O_4$	390	96,28
9	20,175	$C_7H_6O_2$	122	89,6
10	20,994	*$C_{21}H_{28}O_7$	392	50,54
11	21,228	$C_{10}H_{10}O_4$	194	98,83
12	21,814	*$C_{15}H_{10}O_7$	302	98,7
13	21,931	*$C_{21}H_{20}O_{12}$	464	99,49
14	22,165	*$C_9H_8O_3$	164	82,67
15	22,867	$C_9H_6O_3$	162	97,28
16	23,101	*$C_{15}H_{10}O_6$	286	98,49
17	23,687	$C_{30}H_{37}N_{11}O_{10}$	711	97,76
18	24,623	$C_6H_{10}O_3S$	162	83,64
19	25,325	*$C_{16}H_{12}O_6$	300	99,12
20	26,379	$C_8H_4O_3$	148	99,69
21	27,316	*$C_{15}H_{10}O_5$	270	80,07
22	27,55	$C_{13}H_{23}N_{11}$	333	92,08
23	28,135	$C_{20}H_{37}N_{11}O_3$	479	95,52
24	28,955	$C_{18}H_{32}O_2$	280	97,34
25	29,884	*$C_{19}H_{28}O$	272	61,63
26	30,008	$C_{28}H_{24}O_{11}$	536	99,09
27	30,360	*$C_{16}H_{12}O_5$	284	98,53
28	31,881	$C_{23}H_{44}N_6OS$	452	88,24
29	32,467	$C_5H_8O_3S$	148	67,03
30	33,286	$C_9H_{10}O_5$	198	99,59
31	33,520	*$C_{30}H_{46}O_4$	470	91,22
32	34,223	$C_{27}H_{49}NO_9$	531	97,25
33	34.691	*$C_{20}H_{34}O_2$	306	88,25
34	35,042	$C_{24}H_{45}NS$	379	86,93
35	35,978	$C_{18}H_{35}NO$	281	98,31
36	36.330	*$C_{20}H_{32}O$	288	82,67
37	36.40	*$C_{20}H_{30}O$	286	87,94
38	37,149	$C_{24}H_{38}O_4$	390	99,25
39	37,383	*$C_{18}H_{16}O_8$	360	96,53
40	42,768	$C_{44}H_{58}N_2O_3$	662	96,66
41	45,578	NON IDENTIFIE	109	

** Compounds already identified in Hyptis species*

Table 3. Compounds already isolated and detected in the ethyl acetate fraction

Compounds name	Formula	Molecular mass (g/mol)	Retention time (min)	Score (%)
(2E)-1-(2-hydroxyphenyl)-pent-2-en-1-one*	$C_{11}H_{12}O_2$	176,22	17,951	48,47
1-(5-(hydroxy (2-oxotetrahydro-2H-pyran-3-yl) methyl) tetrahydrofuran-2-yl) ethyl 3-(4-hydroxyphenyl) propanoate	$C_{21}H_{28}O_7$	392,45	20,994	50,54
Quercetin	$C_{15}H_{10}O_7$	302,24	21,814	98.7

Compounds name	Formula	Molecular mass (g/mol)	Retention time (min)	Score (%)
Quercetin 3-O-β-D-glucopyranoside	$C_{21}H_{20}O_{12}$	464,38	21,931	99.49
p-Coumaric acid *	$C_9H_8O_3$	164,20	22,165	82.67
Kaempferol *	$C_{15}H_{10}O_6$	286,46	23,101	98,49
Methyl Wogonine *	$C_{16}H_{12}O_6$	300,27	25,325	99,12
Apigenine *	$C_{15}H_{10}O_5$	270,24	27.316	80.07
Genkwanine *	$C_{16}H_{12}O_5$	284,27	30,360	98.53
5α-androst-9(11)-en-12-one	$C_{19}H_{28}O$	272,43	33.106	61,63
1,19α-dihydroxyl-uros-2(3),12-dien-28-oic acid*	$C_{30}H_{46}O_4$	470,69	33,520	91.22
Suaveolol	$C_{20}H_{34}O_2$	306,49	34,691	88.25
(2R,4aS,4bS,10aR)-2,4b,8,8,10a-pentaméthyldecahydro-2H-2,4a-méthanophénanthren-1(4bH)-one	$C_{20}H_{32}O$	288,48	36.330	82.67
Dehydroabietinol*	$C_{20}H_{30}O$	286,46	36.40	87.94
rosamarinic acid	$C_{18}H_{16}O_8$	360,32	37,383	96.53

** Molecules not identified (08) during our previous studies in the dichloromethane fractions of Hyptis suaveolens leaves*

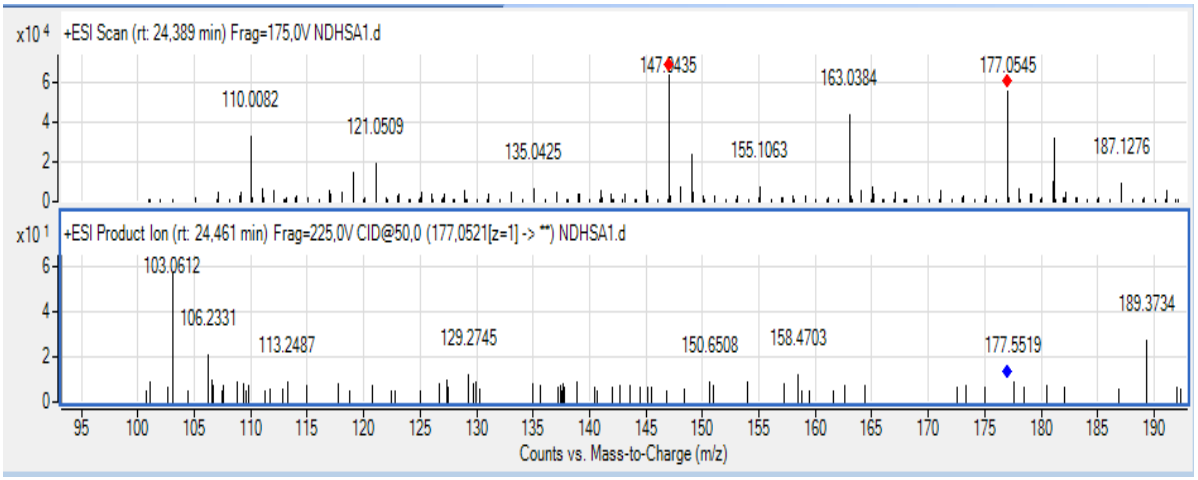


Fig. 3. LC-ESI/MS mass spectrum and ESI/MS fragmentation spectrum of compound 1

This spectrum show major fragment ions at : mass = 158 [M+H-19], mass = 151 [M+H-26], mass = 129 [M+H-19-29], mass = 113 [M+H-19-45], mass = 106 [M+H-19-52], mass = 103 [M+H-19-55].

Among structures proposed by databases (ChemSpider, NIST and PubChem), only (2E)-1-(2-hydroxyphenyl)-pent-2-en-1-one corresponds to the crude formula giving the following structure.

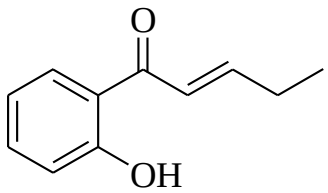
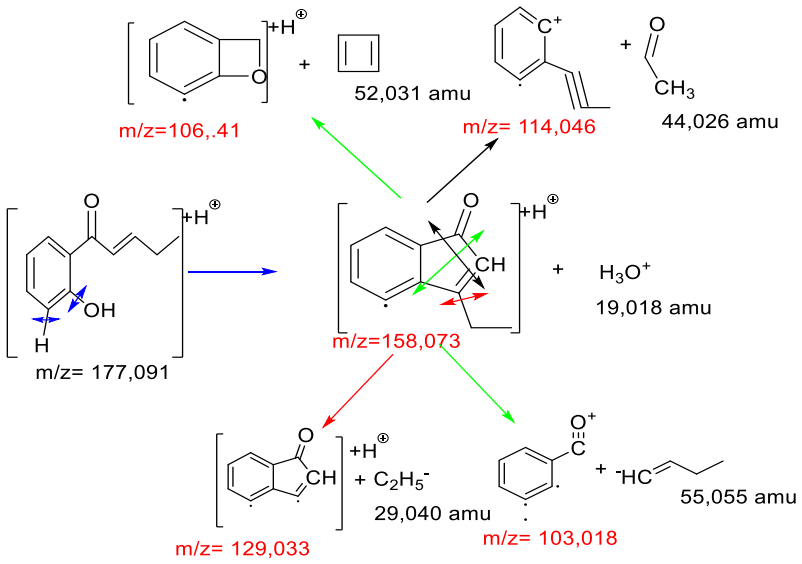


Fig. 4. Structure of compound 1 : ((2E)-1-(2-hydroxyphenyl)-pent-2-en-1-one)

The identified compound fragmentation mode could be like following (Scheme 1)



Scheme 1. Compound 1 fragmentation mode

« Hydronium ion (H_3O^+) on the benzene ring followed by a rearrangement (cyclisation) in accordance with the nitrogen rule (the molar mass of the molecular ion being odd, the molar mass of the fragment resulting from the collusion should also be odd). The fragments with mass = 106 and mass = 103 would be derived from previous fragment by the removal of a cyclobutadiene molecule and a butenyl group respectively following cleavage of the α and γ oxo groups on the (C5) ring. Fragment with mass = 129 may be due to ethyl group departure at the α position of the ethylenic double bond on the (C5) ring. Compound 1 would therefore be (2E)-1-(2-hydroxyphenyl)-pent-2-en-1-one and belongs to the phenolic compound family. This compound has already been isolated in *Hyptis suaveolens* leaves » (Mukherjee K and al, 1984, Soumahoro B and al, 2023, Soumahoro B and al, 2024).

3.2.2 Compound 2 structure

Compound 2, which appeared at 20.859 min, corresponded to the molecular ion $[M+H]^+$ with a mass = 393.1879 and a molecular weight of 392.1835 g/mol. The most probable molecular formula (with the highest score) is $C_{21}H_{28}O_7$ (cal. 392.45) (Fig. 5).

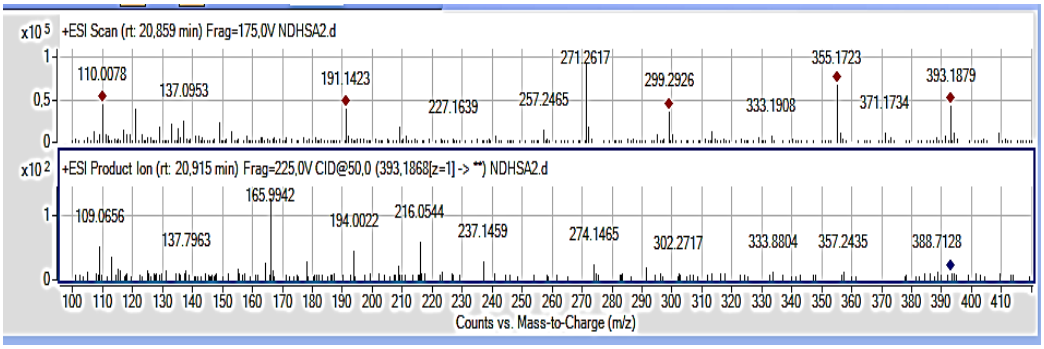


Fig. 5. Compound 2 LC-ESI/MS mass spectrum and ESI/MS fragmentation spectrum

The fragmentation spectrum of compound 2 (Fig. 5) show the presence of characteristic fragments with mass = 216 [M+H-177], mass = 194 [M+H-177-22], mass = 166 [M+H-177-22-28], mass = 109 [M+H-177-22-29-57].

Among structures proposed by databases (ChemSpider, NIST and PubChem), only 1-(5-(hydroxy (2-oxotetrahydro-2H-pyran-3-yl) methyl) tetrahydrofuran-2-yl) ethyl-3-(4-hydroxyphenyl) propanoate corresponds to the crude formula giving the following structure.

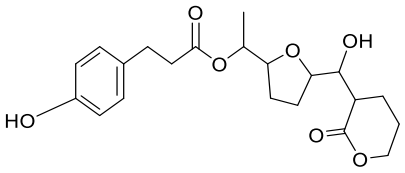
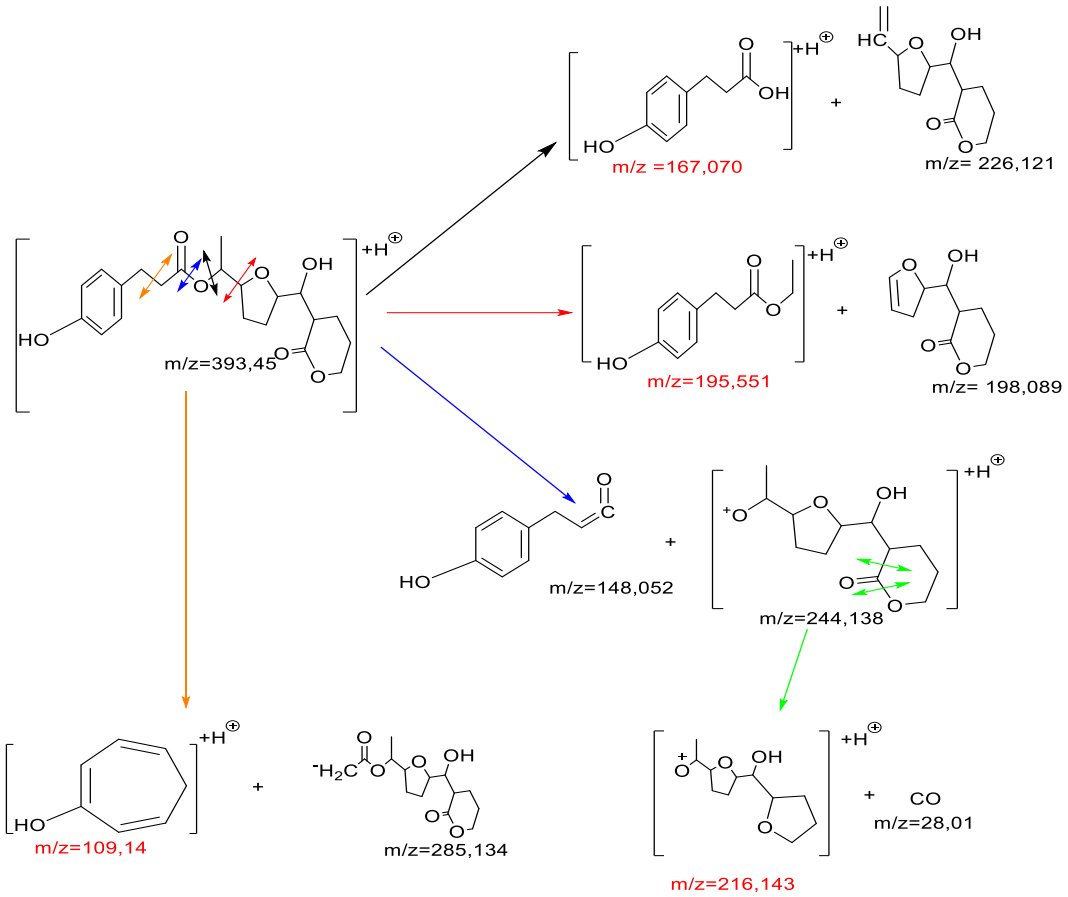


Fig. 6. Structure of compound 2 (1-(5-(hydroxy(2-oxotetrahydro-2H-pyran-3-yl)methyl) tetrahydrofuran-2-yl)ethyl 3-(4-hydroxyphenyl))

The identified compound fragmentation mode could be as following (Scheme 2).



Scheme 2. Compound 2 fragmentation mode

« The basic molecular with mass = 167 (166 on the spectrum following rearrangement according to the nitrogen rule), would be due to Mc Lafferty type rearrangement with cleavage of carbon-oxygen bond at α. Fragment with mass = 195 would result from carbon-carbon bond cleavage at α of the methyl group. Fragment with mass = 109 would rise from the aromatic ring's β-carbon bond cleavage followed by formation of the tropylium ion. Compound 2 would therefore be 1-(5-(hydroxy (2-oxotetrahydro-2H-pyran-3-yl) methyl) tetrahydrofuran-2-yl) ethyl-3-(4-hydroxyphenyl) propanoate and belongs to the phenolic compound family. This compound has already been isolated in *Hyptis brevipes* leaves, species from the same genus as the studied plant » (Suárez-Ortiz GA and al, 2017).

3.2.3 Structure of compound 3

The compound 3 (retention time 21.814 min) corresponds to the molecular ion $[M+H]^+$ with mass = 303.0494. Its molecular molar mass is therefore 302.0427g/mol. the most probable molecular formula (with the highest score) is $C_{15}H_{10}O_7$ (cal. 302.238) (Fig. 7).

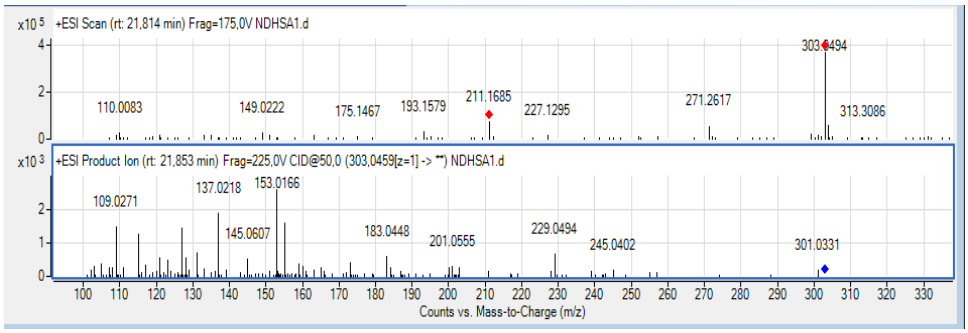


Fig. 7. Compund 3 LC-ESI/MS mass spectrum and ESI/MS spectre de fragmentation spectrm

This spectrum of compound 3 (Fig. 7) show major fragments with mass = 229 [M+H-74], mass = 153 [M+H-150] (base peak), mass = 137 [M+H-166].

Among structures proposed by databases (ChemSpider, NIST and PubChem), only quercetin corresponds to the crude formula giving the following structure.

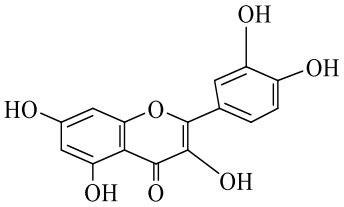
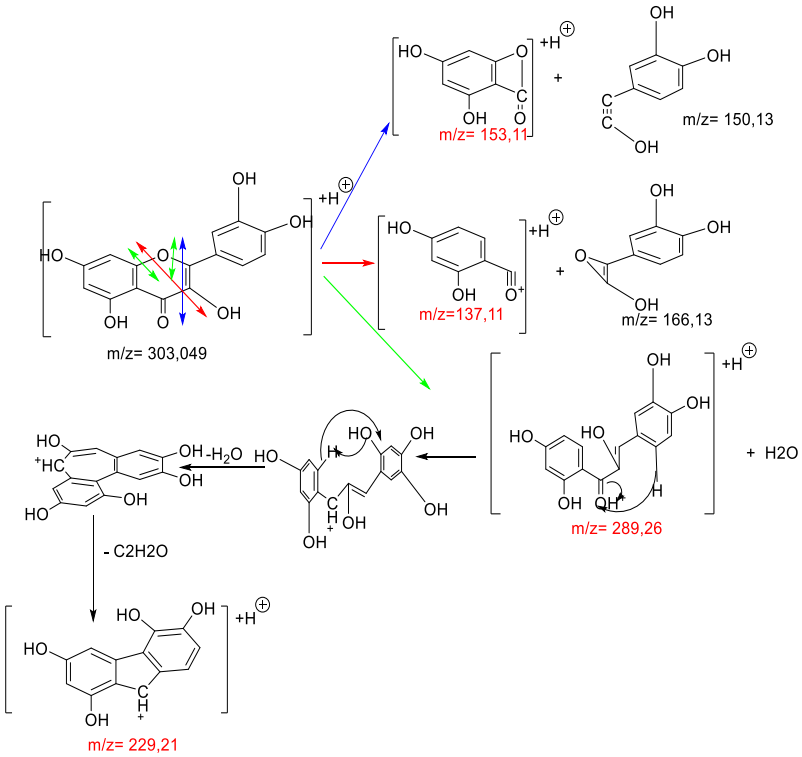


Fig. 8. Structure of compound 3 : quercetin (2-(3,4-dihydroxyphenyl)-3,5,7-trihydroxy-4H-chromen-4-one)

The identified compound fragmentation mode could be as following (Scheme 3).



Scheme 3. Compound 3 fragmentation mode

The base peak with mass = 153 arises from a double cleavage on the intermediate C ring in the γ and β position of its hydroxyl group of carbon-oxygen and carbon-carbon bonds respectively. Also, the fragment with mass = 137 derives from a double cleavage on the intermediate C ring in the α position of the aromatic ring and β position of its hydroxyl group of the carbon-oxygen and carbon-carbon bonds respectively. The fragment with mass = 229, derived from loss of oxo group, followed by cyclisation after elimination of water molecule and ethanone group on the intermediate C ring (Scheme 3). Compound 3 would therefore be quercetin and belongs to flavonoid family. This compound is present in *H. suaveolens* leaves (Tang G and al, 2018).

3.2.4 Compound 4 structure

Compound 4 (retention time: **21.814 min**) corresponds to molecular ion $[M+H]^+$ with mass= 465.1028. Its molecular molar weight is 464.0955g/mol. The most probable molecular formula is $C_{21}H_{20}O_{12}$ (cal. 464.3790) (Fig. 9).

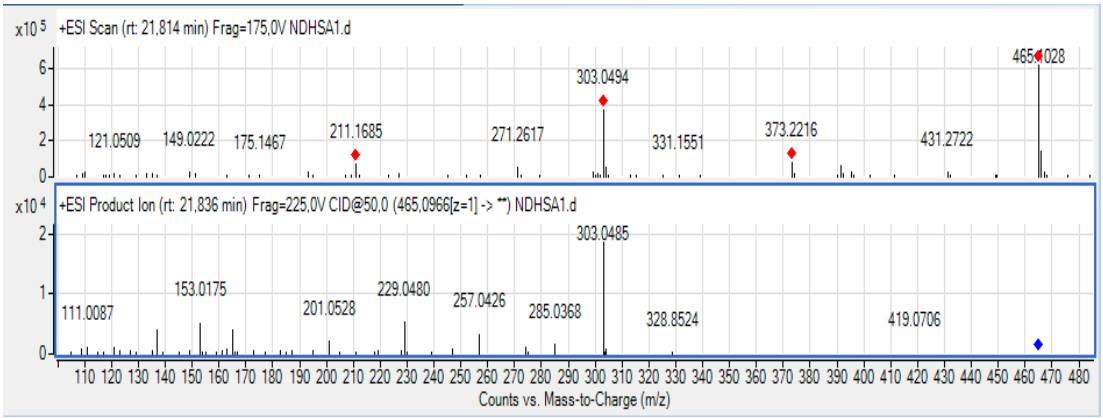


Fig. 9. Compound 4 LC-ESI/MS mass spectrum and ESI/MS spectrum

The spectrum of compound 4 (Fig. 9) show presence of major fragments with mass = 303 [M+H-162] (base peak), mass = 285[M+H-162-18], mass = 257[M+H-162-46], mass = 229[M+H-162-74], mass = 153[M+H-162-150].

Among structures proposed by databases (ChemSpider, NIST and PubChem), only quercetin 3-O-β-D-glucopyranoside corresponds to the crude formula giving the following structure.

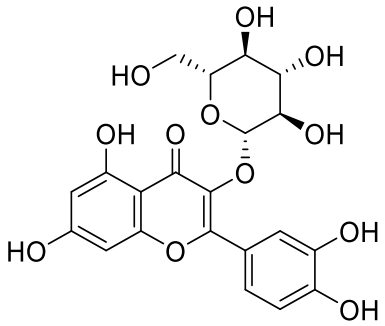
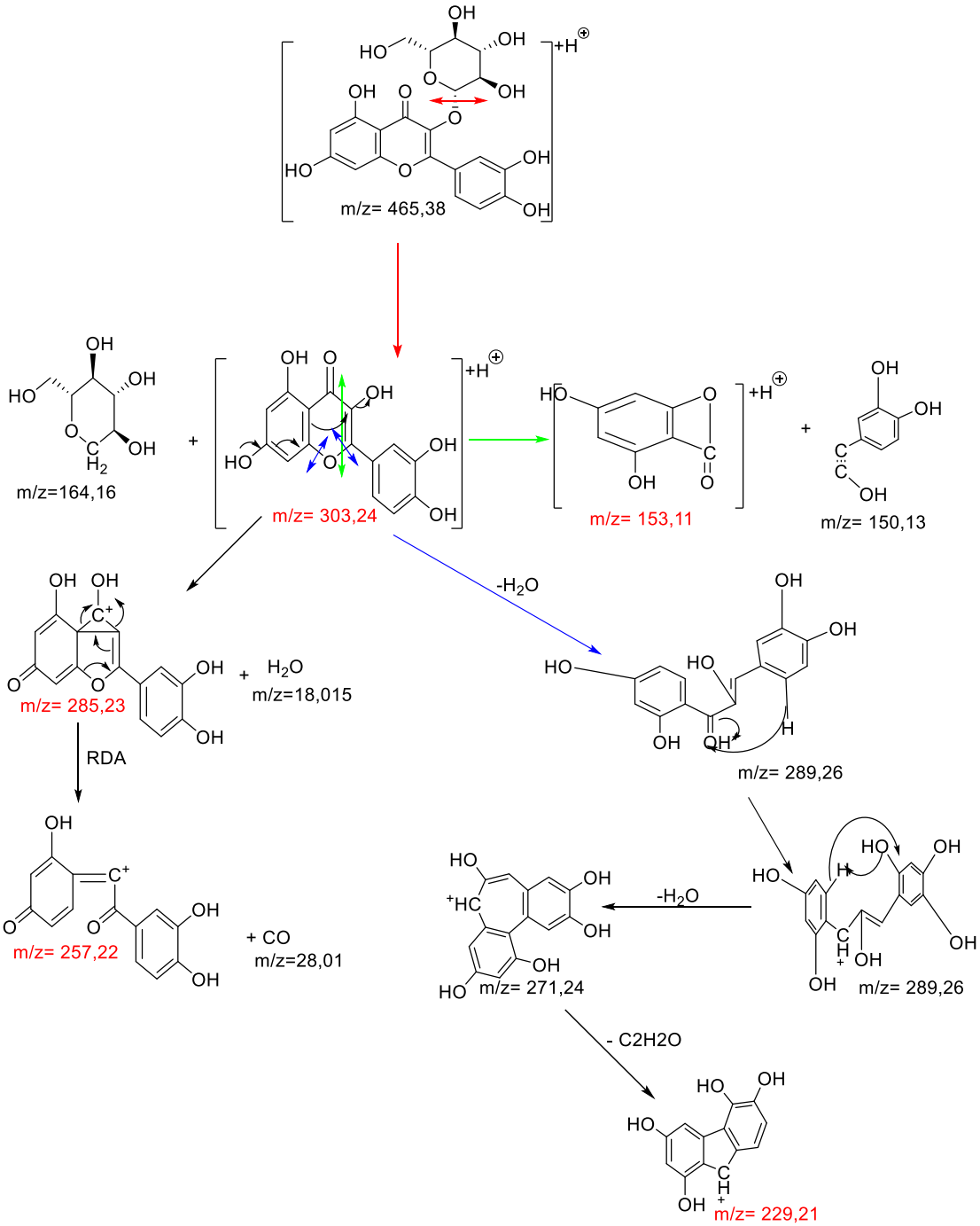


Fig. 10. Structure of compound 4 : quercetin 3-O-β-D-glucopyranoside (2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3-(((2S,3R,4S,5S,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)tetrahydro-2H-pyran-2-yl)oxy)-4H-chromen-4-one)

The identified compound fragmentation mode could be as following (Scheme 4).



Scheme 4. Compound 4 fragmentaion mode

The base peak with mass = 303 rise from elimination of the glucosyl group. Fragment with mass = 153 originate from the base peak after a double cleavage on the ring intermediate C at β and α of the hydroxyl group of this ring respectively on carbon-oxygen and carbon-carbon bonds. As same, fragment with mass = 285 is provided from the base peak by a water molecule elimination on the intermediate C ring. As for the fragment with mass = 257, it derived from a Retro Diels-Alder (RDA) mechanism on the intermediate C ring with the carbonyl group loss (Scheme 4). Compound 4 is quercetin 3-O-β-D-glucopyranoside from the flavonoid family. It has been revelated in *Hyptis suaveolens* (Ekow NT and al, 2018).

3.2.5 Structure of compound 5

The compound 5 (retention time: 22.165 min) corresponds to molecular ion $[M+H]^+$ with mass = 165.0901. Its molecular molar mass is therefore 164.0837g/mol. The most probable molecular formula (with the highest score) is $C_{10}H_{12}O_2$ (cal. 164.20) (Fig. 11).

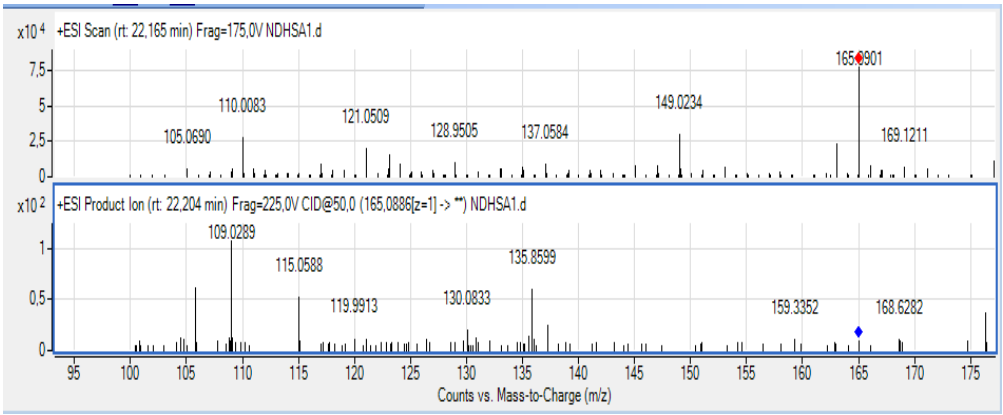


Fig. 11. Compound 5 LC-ESI/MS mass spectrum and ESI/MS spectrum

These spectrums of compound 5 (Fig. 11) prove presence of major fragments with mass= 136 [M+H-29], mass= 120 [M+H-45], mass= 115 [M+H-50], mass= 109 [M+H-56] (base peak). Among structures proposed by databases (ChemSpider, NIST and PubChem), only 4-allyl-2-methoxyphenol corresponds to the crude formula giving the following structure.

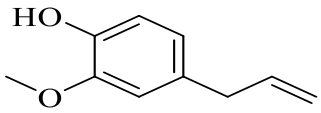
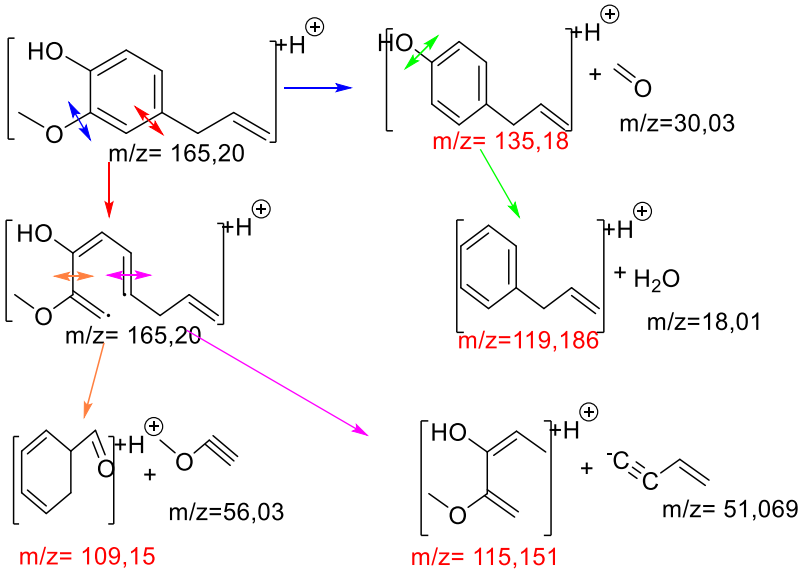


Fig. 12. Structure of compound 5 (4-allyl-2-methoxyphenol)

The identified compound fragmentation mode could be as following (Scheme 5).



Scheme 5. Compound 5 fragmentation mode

The fragment with mass = 135 is thought to derive from methoxyl group loss. Fragment with mass= 119 is thought to have resulted from the previous fragment after dehydration. The major peak with mass= 109 would provided from double cleavage carbon-carbon single bonds in the β and γ positions of the methoxyl group on the aromatic ring. Similarly, fragment with mass= 115 would result to a double cleavage on the ring of the α-single and double bonds of the propenyl group (Scheme 5). Compound 5 would be 4-allyl-2-methoxyphenol from the phenolic compound family. It has been isolated in *H. suaveolens* leaves essential oil harvested in Tanzania (Malele R and al, 2003).

3.2.6 Structure of compound 6

The compound 6 (retention time: 23.101 min) corresponds to molecular ion $[M+H]^+$ with mass = 287.0544. Its molecular molar mass is therefore 286.0477g/mol and the most probable molecular formula (with the highest score) is $C_{15}H_{10}O_6$ (cal. 286.46) (Fig. 13).

Fig. 13. Compound 6 LC-ESI/MS mass spectrum and ESI/MS spectrum

These spectrums of compound 6 (Fig. 13) indicate presence of major fragments with mass= 213[M+H-74], mass= 167[M+H-120], mass= 153[M+H-134] (base peak), mass= 139[M+H-148], mass= 121[M+H-166]. Among structures proposed by databases (ChemSpider, NIST and PubChem), only Kaempferol corresponds to the crude formula giving the following structure.

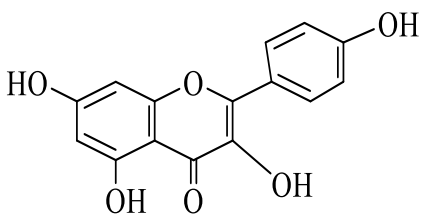
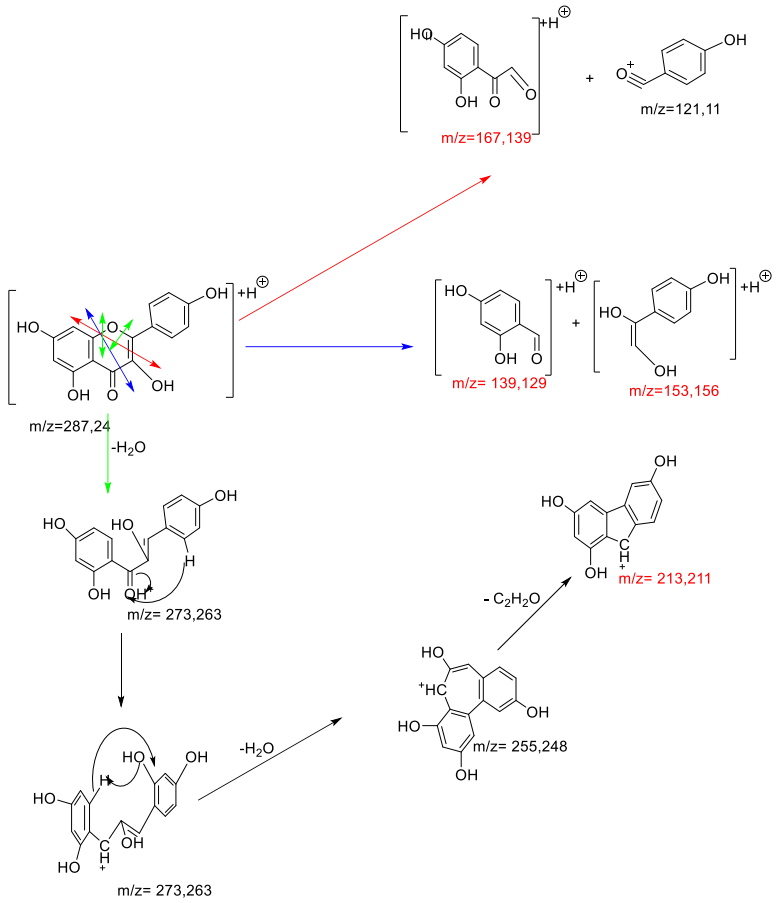


Fig. 14. Structure of compound 6: Kaempferol (3,5,7-trihydroxy-2-(4-hydroxyphenyl)-4H-chromen-4-one)

The identified compound fragmentation mode could be as following (Scheme 6).



Scheme 6. Compound 6 fragmentation mode

The base peak with mass= 153 and the fragment mass= 139 would derivedt from a double cleavage of the carbon-carbon and carbon-oxygen bonds on the intermediate ring respectively at β and α of the adjacent ring to the intermediate (Scheme 6). Compound 6 would be Kaempferol, a flavonoid. It has already been isolated in *Hyptis suaveolens* (Ekow NT and al, 2018).

3.2.7 Structure of compound 7

The compound 7 (retention time: 25.325 min) corresponds to molecular ion $[M+H]^+$ with mass= 301.0706 with a molecular molar mass= 300.0634 g/mol. The most probable (with the highest score) molecular formula is $C_{16}H_{12}O_6$ (cal. 300.2660) (Fig. 15).

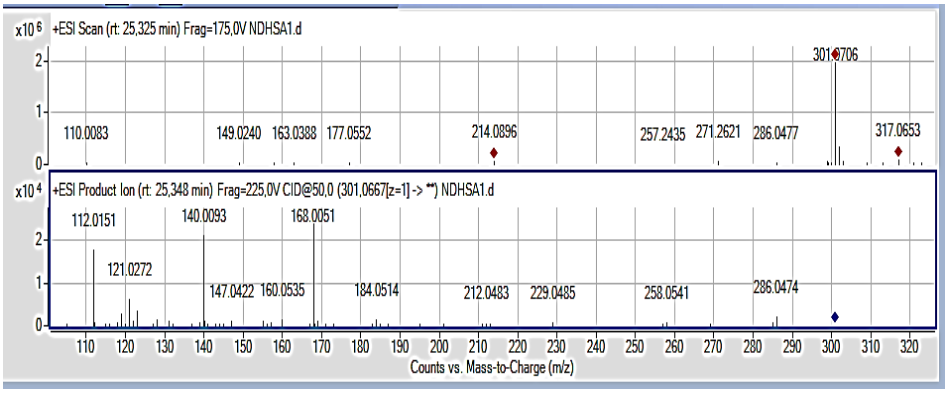


Fig. 15. Compound 7 LC-ESI/MS mass spectrum and ESI/MS spectrum

These spectrums of compound 7 (Fig. 15) indicate major fragments presence with mass= 286 [M+H-15], mass= 168 [M+H-15-118] (base peak), mass= 140 [M+H-15-118-28], mass= 121 [M+H-180], mass= 112 [M+H-189]. Among structures proposed by databases (ChemSpider, NIST and PubChem), only the methyl Wogonine corresponds to the crude formula giving the following structure.

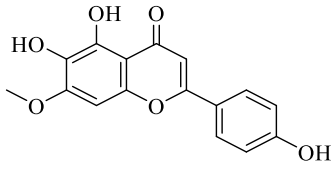
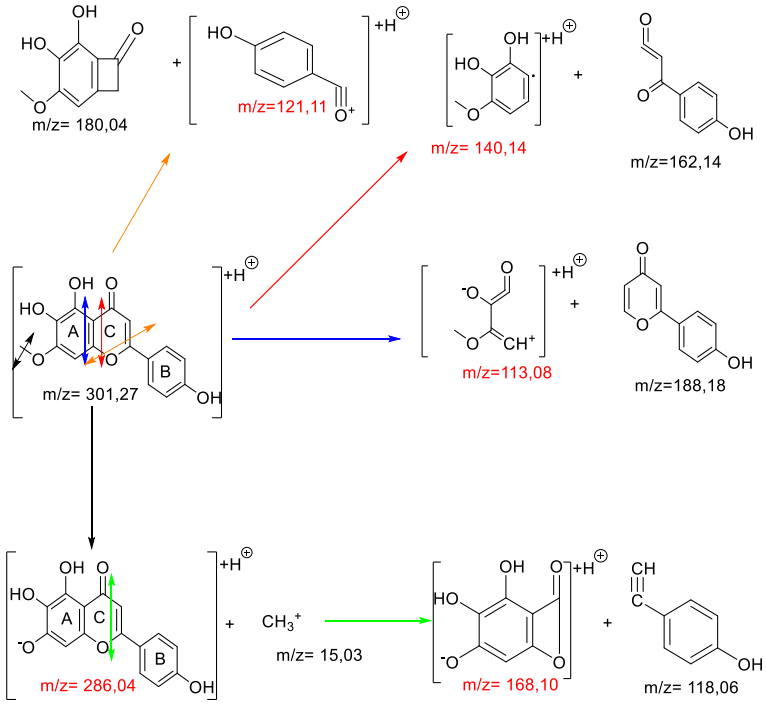


Fig. 16. Structure of compound 7: methyl Wogonine (5,6-dihydroxy-2-(4-hydroxyphenyl)-7-methoxy-4H-chromen-4-one)

The identified compound fragmentation mode could be as following (Scheme 7).



Scheme 7. Compound 7 fragmentation mode

The fragment with mass= 286 arises from a methyl group loss. The base peak with mass= 168 is resulted from the previous fragment following of carbon-carbon and carbon-oxygen bonds cleavage on the intermediate C ring in the β position of contiguous ring (A). Fragment with mass= 112 derives from a double cleavage of carbon-carbon bonds at α of the intermediate C ring on ring (A). Fragment with mass= 140 results from (RDA)-type cleavage on the intermediate C ring of carbon-carbon and carbon-oxygen bonds in the α position of the contiguous ring (A). As for the fragment with mass= 121, it is due to carbon-carbon and carbon-oxygen bonds cleavage respectively at the γ and β positions on adjacent ring (Scheme 7). Compound 7 is methyl Wogonin of flavonoid family. It has already been described in *Hyptis suaveolens* plant (Tang G and al, 2018).

3.2.8 Compound 8 structure

The compound 8 (retention time: 27.316 min) corresponds to molecular ion $[M+H]^+$ with mass = 271.0627 with a molecular mass of 270.0528 g/mol. The most probable molecular formula is $C_{15}H_{10}O_5$ (cal. 270.24) (Fig. 17).

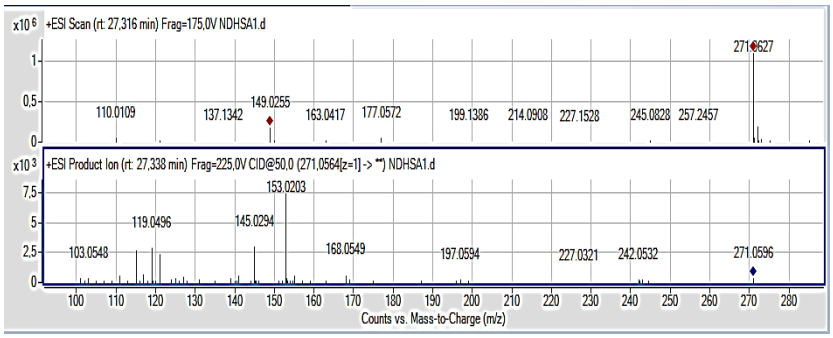


Fig. 17. Compound 8 LC-ESI/MS mass spectrum and ESI/MS spectrum

These spectrums of compound 8 (Fig. 17) show major fragments presence with mass = 153[M+H-118] (base peak), mass = 145[M+H-126], mass = 119[M+H-152]. Among structures proposed by databases (ChemSpider, NIST and PubChem), only Apigenin corresponds to the crude formula giving the following structure.

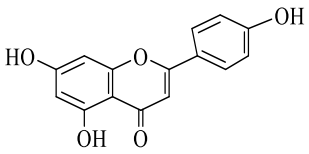
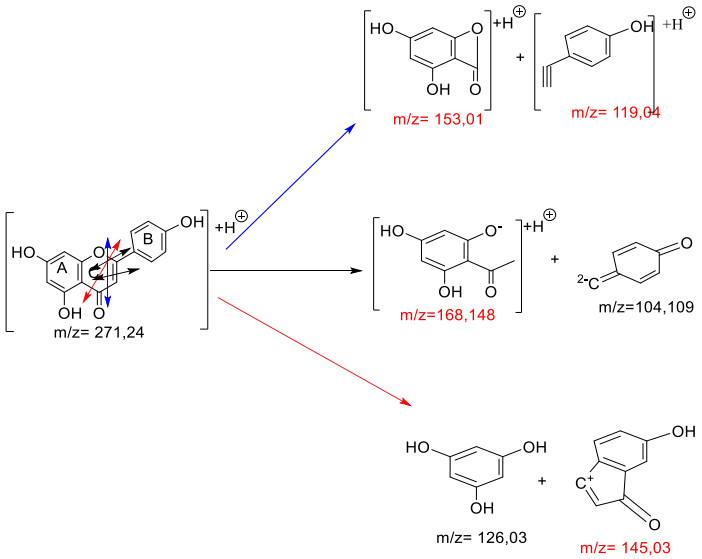


Fig. 18. Structure of compound 8 : Apigenin (5,7-dihydroxy-2-(4-hydroxyphenyl)-4H-chromen-4-one)

The identified compound fragmentation mode could be as following (Scheme 8).



Scheme 8. Compound 8 fragmentation mode

The base peak with mass = 153 results from carbon-carbon and carbon-oxygen bonds cleavage on the ring intermediate C in β of the contiguous ring (A). Fragment with mass = 168 results from carbon-carbon and carbon-oxygen bonds cleavage on the intermediate C ring respectively at γ and β of the contiguous ring (A). As for the fragment with mass = 145, it is due to the cleavage of the carbon-carbon and carbon-oxygen bonds respectively at α and β of the contiguous ring (A) (Scheme 8). Compound 8 would be Apigenin, a flavonoid. It has been described in the studied plant (Ekow NT and al, 2018).

3.2.9 Compound 9 structure

The compound 9 (retention time: 30.360 min) corresponds to molecular ion $[M+H]^+$ with mass = 285.0763. Its molecular molar mass is therefore 284.0685 g/mol and the most probable molecular formula (with the highest score) is C16H12O5 (cal. 284.2700) (Fig. 19).

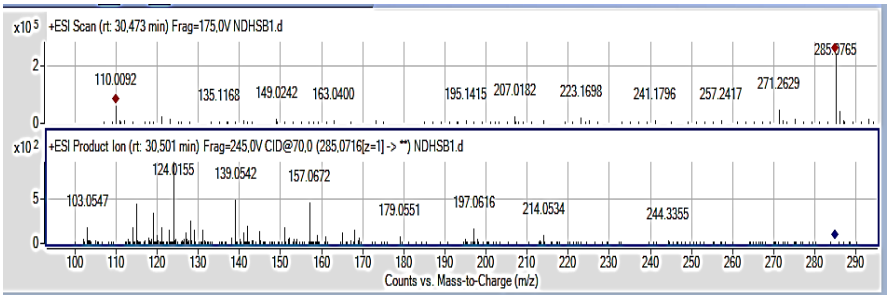


Fig. 19. Compound 9 LC-ESI/MS mass spectrum and ESI/MS spectrum

These spectrums of compound 9 (Fig. 19) show major fragments presence at mass= 197 [M+H-88], mass=157 [M+H-128], mass=139 [M+H-146], mass=124 [M+H-161] (base peak). Among structures proposed by databases (ChemSpider, NIST and PubChem), only Genkwanine corresponds to the crude formula giving the following structure.

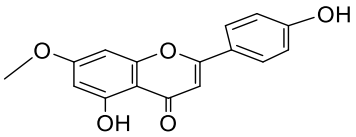
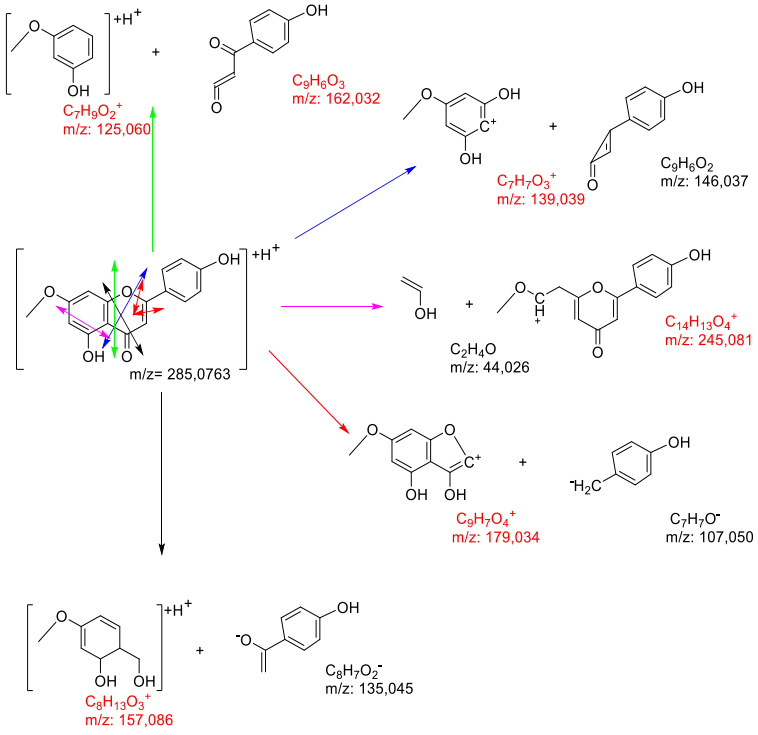


Fig. 20. Structure of compound 9: Genkwanine (5-hydroxy-2-(4-hydroxyphenyl)-7-methoxy-4H-chromen-4-one)

The identified compound fragmentation mode could be as following (Scheme 8).



Scheme 9. Compound 9 fragmentation

The major peak with mass= 125, arises from the carbon-carbon and carbon-oxygen bonds cleavage on the intermediate C ring at α of the contiguous ring (A). Fragment with mass= 139 is due to carbon-carbon and carbon-oxygen bonds breaking on the intermediate C ring respectively at α and β of the contiguous ring (A). Fragment mass= 245 results from single carbon-carbon bonds breakage at α of the hydroxyl group and at β of the methoxyl group, on the aromatic ring. Fragment with mass= 179 results from carbon-carbon and carbon-oxygen bonds cleavage on the intermediate C ring in β of the monosubstituted ring. Similarly, fragment with mass= 157 results from carbon-carbon and carbon-oxygen bonds cleavage at β and α respectively of the methoxylated ring on the intermediate C ring (Scheme 9). Compound 9 is Genkwanine, a flavonoid. It has been isolated in *H. suaveolens* leaves (Ekow NT and al, 2018).

3.2.10 Compound 10 structure

The compound 10 (retention time 33.106 min) corresponds to molecular ion $[M+H]^+$ with mass= 273.2210. Its molecular molar mass is therefore 272.2140 g/mol and the most probable molecular formula (with the highest score) is $C_{19}H_{28}O$ (cal. 272.43) (Fig. 21).

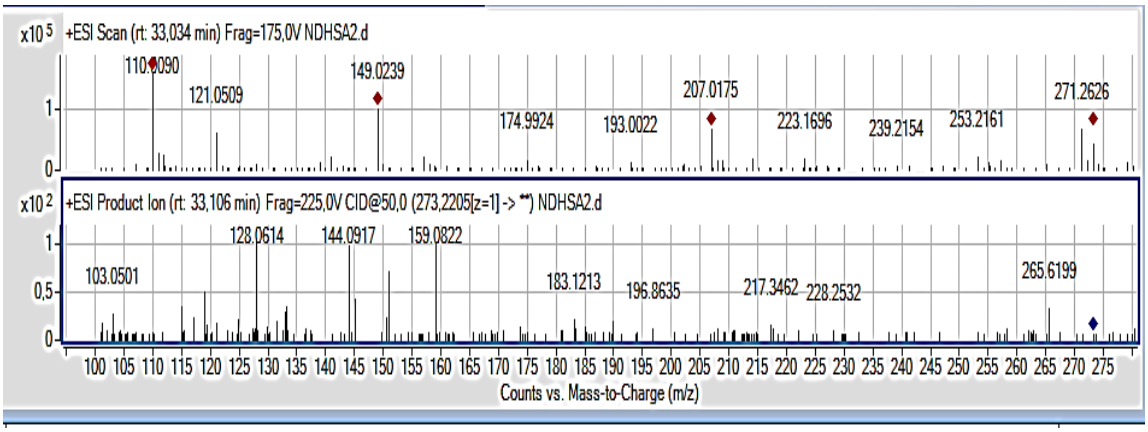


Fig. 21. Compound 10 LC-ESI/MS mass spectrum and ESI/MS fragmentation spectrum

These spectrums of compound 10 (Fig. 21) indicate major fragments presence with mass= 159[M+H-114] (base peak), mass= 144[M+H-129], mass= 128[M+H-145], mass= 115[M+H-158], mass= 103[M+H-114-56]. Among structures proposed by databases (ChemSpider, NIST and PubChem), only 5α-androst-9(11)-en-12-one corresponds to the crude formula giving the following structure.

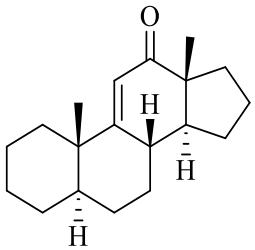
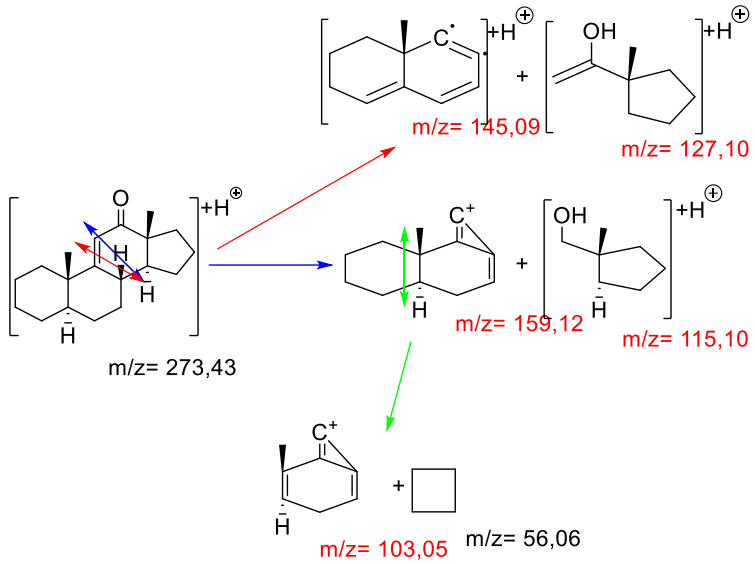


Fig. 22. Structure of compound 10 : 5α-androst-9(11)-en-12-one ((5R,8S,10S,13R,14S)-10,13-dimethyl-1,2,3,4,5,6,7,8,10,13,14,15,16,17-tetradecahydro-12H-cyclopenta[a]phenanthren-12-one)

The identified compound fragmentation mode could be as following (Scheme 10).



Scheme 10. Compound 10 fragmentation mode

The base peak with mass= 159 derives from the double carbon-carbon bond cleavage between the unsaturation and the carbonyl group and that between the (C5) ring and the saturated (C6) ring, on the unsaturated ring. The fragment with mass= 103 would resulte from the base peak by cyclobutane loss following the carbon-carbon bonds adjacent to the second ring cleavage at (C6) on the saturated ring. Fragments with mass= 145 (144 on the spectrum) and mass= 127 (128 on the spectrum) would derive from carbon-carbon bonds contiguous to the intermediate ring at (C6) breakage on the unsaturated ring (Scheme 10). Compound 10 is 5α-androst-9(11)-en-12-one, from the terpene and sterol family. It has been elucidate in the plant leaves (Edeoga H and al, 2006).

3.2.11 Compound 11 structure

The compound 11 (retention time: 33.520 min) corresponds to molecular ion $[M+H]^+$ with mass = 471.346. Its molecular molar mass is therefore 470.340 g/mol and the most probable molecular formula is $C_{30}H_{46}O_4$ (cal. 470.69) (Fig. 23).

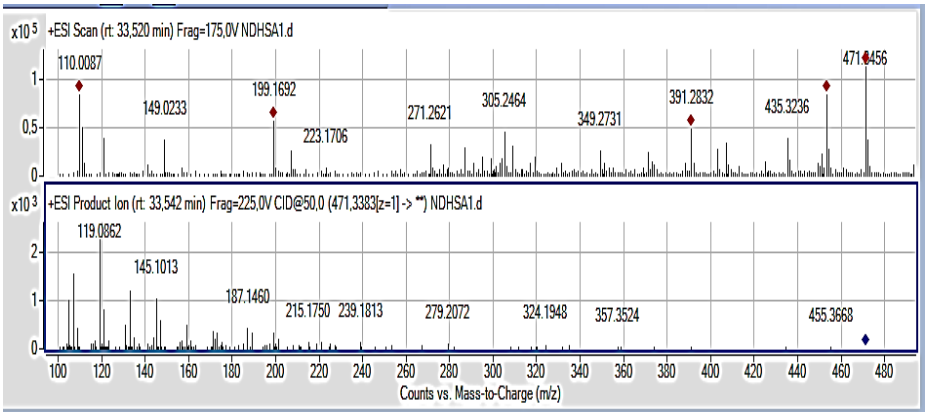


Fig. 23. Compound 11 LC-ESI/MS mass spectrum and ESI/MS fragmentation spectrum

These spectrums of compound 11 (Fig. 23) indicate major fragments presence at : mass= 455[M+H-16], mass= 357[M+H-16-98], mass= 187[M+H-16-268], mass= 157[M+H-16-298], mass= 145[M+H-16-310], mass= 119[M+H-16-336]. Among structures proposed by databases (ChemSpider, NIST and PubChem), only 1,19α-dihydroxyl-uros-2(3),12-dien-28-oic acid corresponds to the crude formula giving the following structure.

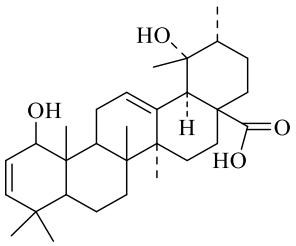
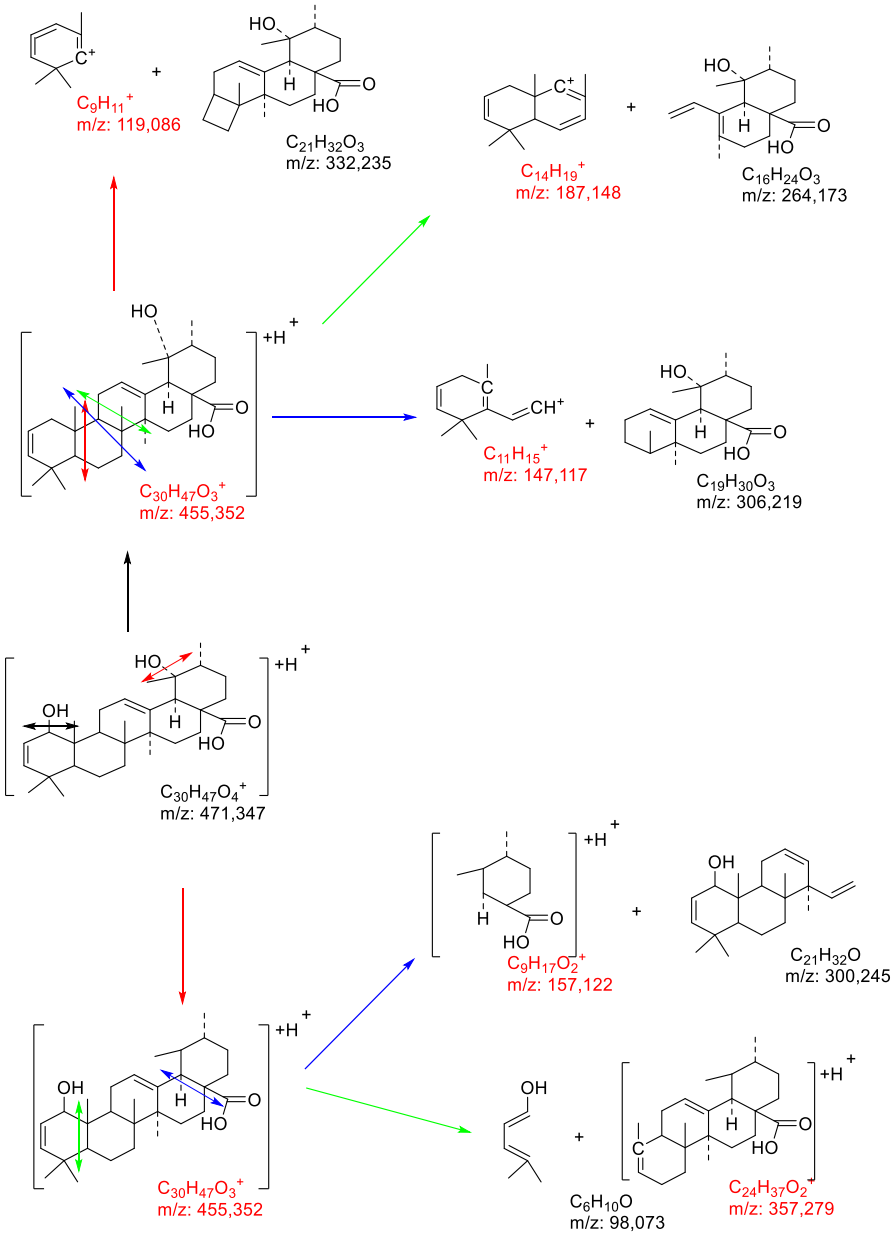


Fig. 24. Structure of compound 11 : 1,19α-dihydroxyl-uros-2(3),12-dien-28-oic acid ((1R,2R,6aS,14bR)-1,12-dihydroxy-1,2,6a,6b,9,9,12a-heptamethyl-1,3,4,5,6,6a,6b,7,8,8a,9,12,12a,12b,13,14b-hexadecahydropicene-4a(2H)-carboxylic acid)

The identified compound fragmentation mode could be as following (Scheme 11).



Scheme 11. Compound 11 fragmentation mode

The fragment with mass= 455 is obtained after deshydatation on a the peripheral rings. Those fragments are resulted from this fragment. The major peak with mass= 119 and fragments with mass= 187 and mass= 147, are thought to have derived from dehydroxylation on the peripheral unsaturated ring followed by a double carbon-carbon bonds cleavage respectively contiguous to the unsaturated ring, between the two unsaturated rings and in γ of the extreme unsaturated ring and finally bonds contiguous to the saturated ring intermediate to unsaturated rings. As for fragments with mass= 357 and mass= 157, its rise from dehydration on peripheral saturated ring followed by a double carbon-carbon bonds scission respectively on peripheral α-unsaturated ring of adjacent ring and on the α-saturated ring of the extreme saturated ring (Scheme 11). Compound 11 would therefore be 1,19α-dihydroxyl-uros-2(3),12-dien-28-oic acid, a molecular of terpenoids family. This compound has been already isolated in H. suaveolens leaves (Raja Rao KV and al, 1990, Aspinall G and al, 1991).

3.2.12 Compound 12 structure

The compound 12 (retention time: 34.691 min) corresponds to molecular ion $[M+H]^+$ with mass= 307.262. Its molecular molar mass is therefore 306.256 g/mol and the most probable molecular formula (with the highest score) is $C_{20}H_{34}O_2$ (cal. 306.49) (Fig. 25).

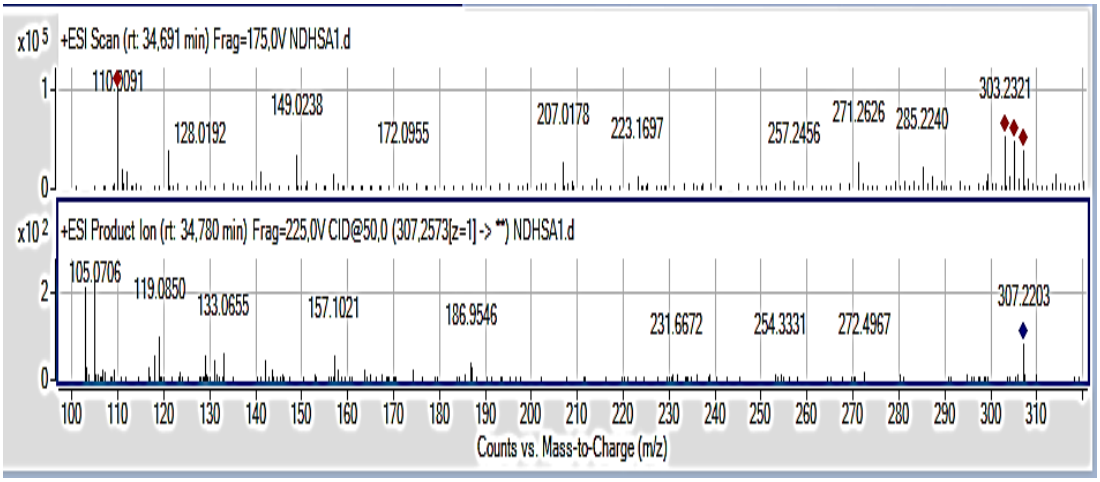


Fig. 25. Compound 12 LC-ESI/MS mass spectrum and ESI/MS fragmentation spectrum

These spectrums of compound 12 (Fig. 25) is show major fragments presence at mass= 187[M+H-18-102], mass= 157[M+H-18-132], mass= 133[M+H-18-156], mass= 119[M+H-18-170], mass= 105[M+H-18-184] (base peak). Among structures proposed by databases (ChemSpider, NIST and PubChem), only Suaveolol corresponds to the crude formula giving the following structure.

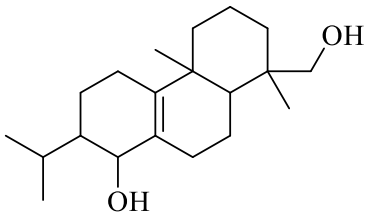
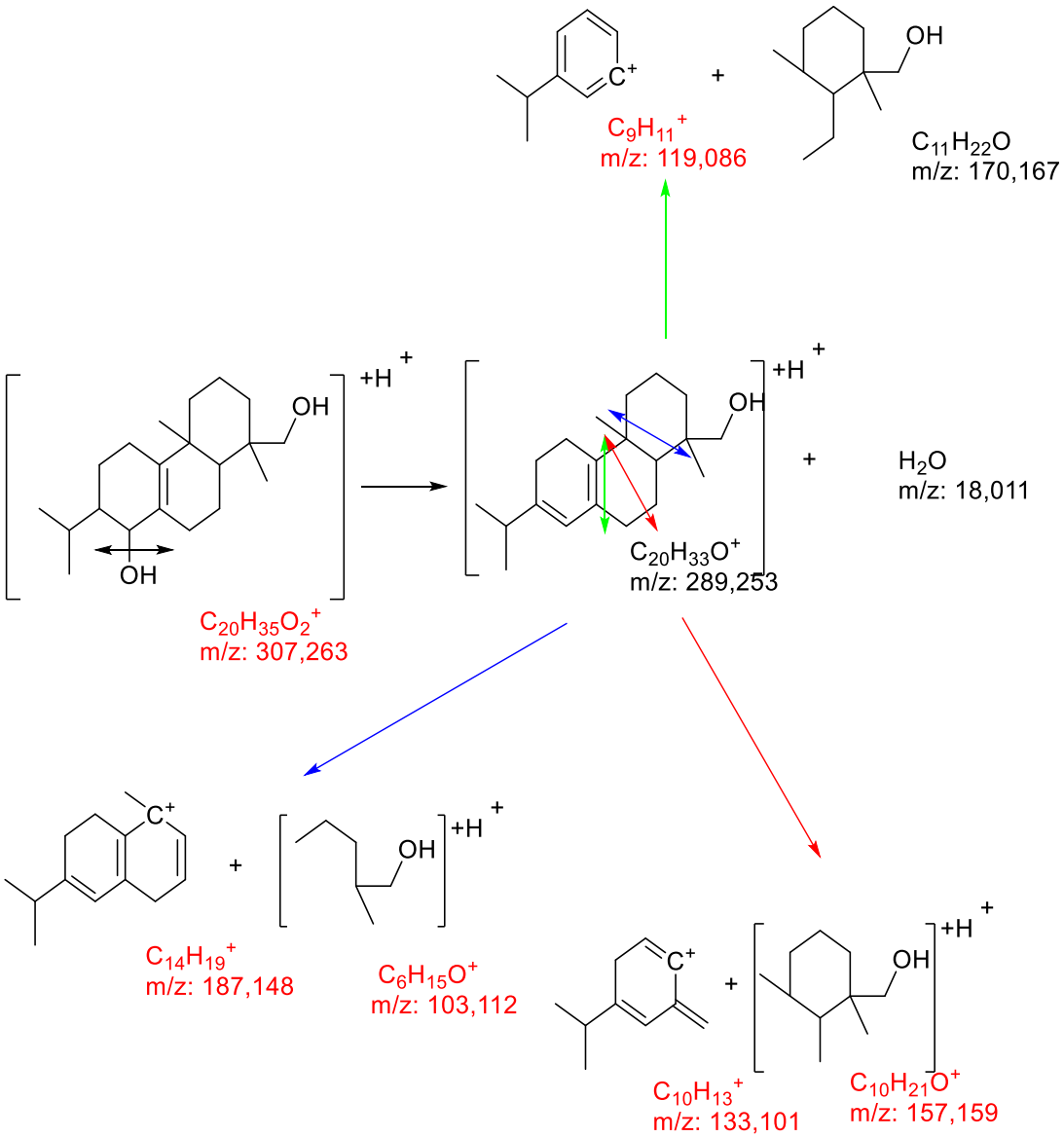


Fig. 26. Structure of compound 12: Suaveolol (8-(hydroxymethyl)-2-isopropyl-4b,8-dimethyl-1,2,3,4,4b,5,6,7,8,8a,9,10-dodecahydrophenanthren-1-ol)

The identified compound fragmentation mode could be as following (Scheme 12).



Scheme 12. Compound 12 fragmentation mode

The molecular peak with mass= 307 on the spectrum would be due to presence of unsaturation in the structure. All fragments are obtained after dehydration of the unsaturated ring. The major fragment with mass= 103 and the fragment with mass= 187, rise from the α-carbon bonds of the adjacent ring cleavage on the saturated ring. Fragment with mass= 119 is derive to α-carbon bonds of the unsaturation cleavage on the intermediate ring. Fragments with mass= 157 and mass= 133, result from the double carbon-carbon bonds cleavage on the intermediate ring in the α and β positions of other two rings (Scheme 12). Compound 12 is Suaveolol belonging to terpenoids family. It's already describe in literature (Manchand PS and al, 1974).

3.2.13 Compound 13 structure

The compound 13 (retention time: 36.092min) corresponds to molecular ion $[M+H]^+$ with mass= 287.237. Its molecular molar mass is therefore 286.230 g/mol and the most probable molecular formula is $C_{20}H_{30}O$ (cal. 286.46) (Fig. 27).

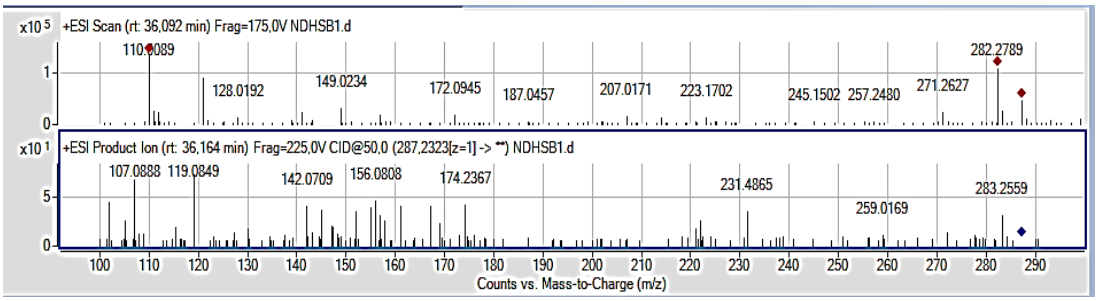


Fig. 27. Compound 13 LC-ESI/MS mass spectrum and ESI/MS fragmentation spectrum

Theses spectrums of compound 13 (Fig. 27) point major fragments presence at : mass = 231[M+H-56], mass= 174[M+H-113], mass= 156[M+H-131], mass= 142[M+H-145], mass= 119[M+H-168] (base peak) and mass= 107[M+H-180]. Among structures proposed by databases (ChemSpider, NIST and PubChem), only dehydroabietinol corresponds to the crude formula giving the following structure.

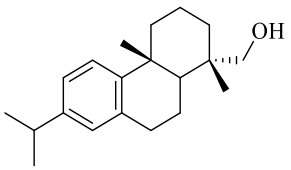
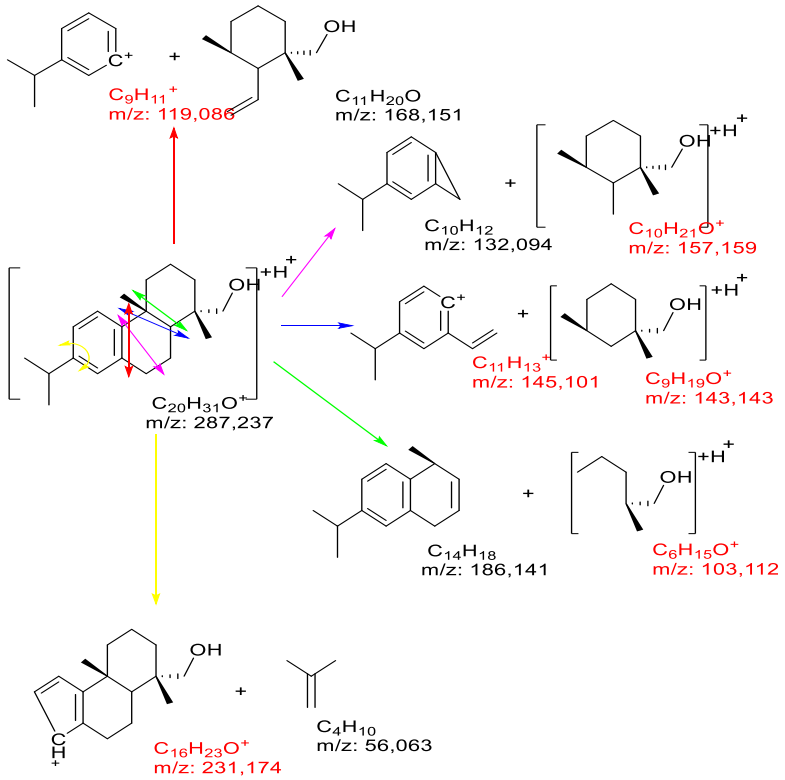


Fig. 28. Structure of compound 13: dehydroabietinol (((1S,4aR)-7-isopropyl-1,4a-dimethyl-1,2,3,4,4a,9,10,10a-octahydrophenanthren-1-yl)methanol)

The described compound fragmentation mode could be as following (Scheme 13).



Scheme 13. Compound 13 fragmentation mode

The major fragment with mass= 119 would be due to carbon-carbon bonds adjacent to the aromatic ring cleavage on the intermediate ring. Fragment with mass= 103 would derive from carbon-carbon bonds in α of the adjacent ring cleavage on the saturated ring. As for the fragment with mass= 157, it would rise from the double carbon-carbon bonds cleavage on the intermediate ring in the α and β positions of the other two rings. Fragment with mass= 143 (142 on the spectrum), would result from carbon-carbon bonds cleavage on the intermediate ring in α position of the adjacent saturated ring. Fragment with mass= 231 would originate from β-carbon bonds of the isopropyl group break on the aromatic ring (Scheme 13). Compound 13 would be dehydroabietinol from terpenoids family. This compound is reported in the literature (Misra TN SR and al, 1983).

3.2.14 Compound 14 structure

The compound 14 (retention time: 36.330 min) corresponds to molecular ion $[M+H]^+$ with mass= 289.252. Its molecular molar mass is therefore 288.245 g/mol and the most probable molecular formula is $C_{20}H_{32}O$ (cal. 288.48) (Fig. 29).

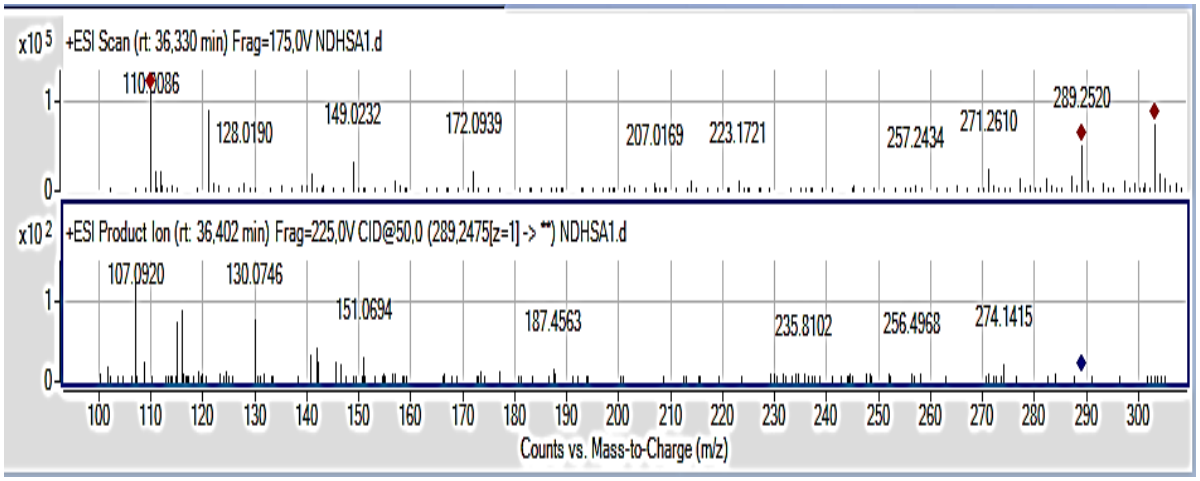


Fig. 29. Compound 14 LC-ESI/MS mass spectrum and ESI/MS fragmentation spectrum

These spectrums of compound 14 (Fig. 29) denote major fragments with mass= 274[M+H-15], mass= 151[M+H-138], mass=130[M+H-159], mass= 107[M+H-172] (base peak). Among structures proposed by databases (ChemSpider, NIST and PubChem), only (2R, 4aS, 4bS, 10aR)-2, 4b, 8, 8, 10a-pentamethyldecahydro-2H-2, 4a-methanophenanthrene-1(4bH)-one corresponds to the crude formula giving the following structure.

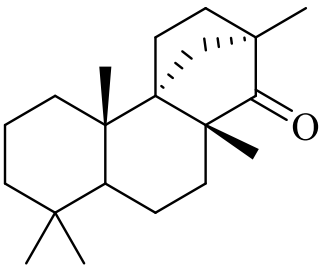
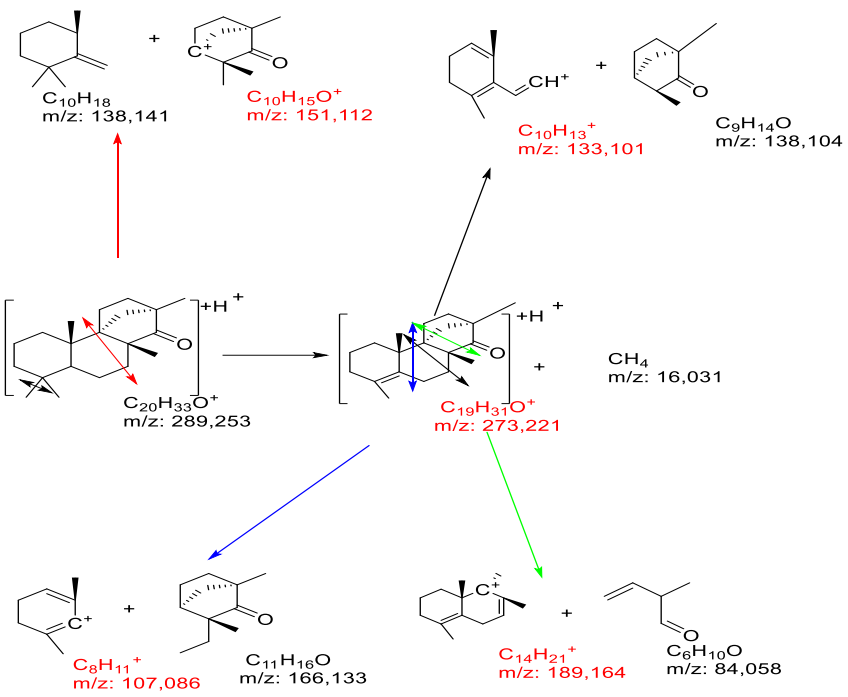


Fig. 30. Structure of Compound 14 ((2R,4aS,4bS,10aR)-2,4b,8,8,10a-pentamethyldecahydro-2H-2,4a-methanophenanthrene-1(4bH)-one)

The identified compound fragmentation mode could be as following (Scheme 14).



Scheme 14. Compound 14 fragmentation

The fragment with mass = 151 would be due to carbon-carbon bonds scission on the intermediate ring in α and β of two extreme rings. Fragment with mass = 273 (274 on the spectrum), would derive from one of the two methyl groups loss on peripheral cyclohexane. The other major fragments are originated from the fragment with mass = 273. The most major fragment with mass = 107 would rise from the intermediate ring bonds adjacent to the unsaturated ring cleavage. Fragment with mass = 133 would result from carbon-carbon bonds cleavage on the intermediate ring at α of carbonyl ring. Fragment with mass = 189, would derive from α-carbon bonds of the intermediate ring break on the carbonyl ring (Scheme 14). Compound 14 could be (2R,4aS,4bS,10aR)-2,4b,8,8,10a-pentamethyldecahydro-2H-2,4a methanophenanthrene -1(4bH)-one of terpenoids family. This compound is described in the literature (Chukwujekwu J and al, 2005).

3.2.15 Compound 15 structure

The compound 15 (retention time: 37.322 min) corresponds to molecular ion $[M+H]^+$ with mass =361.091. Its molecular molar mass is therefore 360.085 g/mol and the most probable molecular formula (with the highest score) is $C_{18}H_{16}O_8$ (cal. 360.32) (Fig. 31).

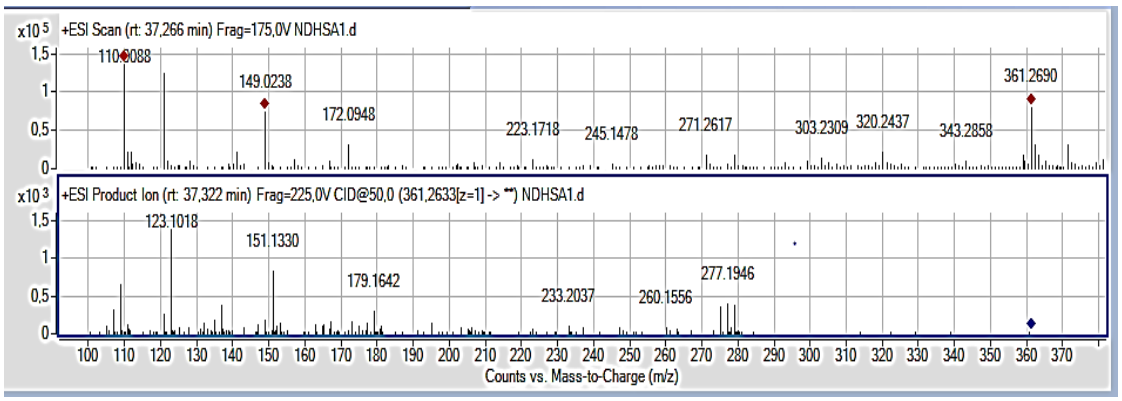


Fig. 31. Compound 15 LC-ESI/MS mass spectrum and ESI/MS fragmentation spectrum

These spectrums of compound 17 (Fig. 31) prove major fragments presence with mass= 277[M+H-84], mass= 179[M+H-182], mass = 151[M+H-210], mass = 123[M+H-238] (base peak). Among structures proposed by databases (ChemSpider, NIST and PubChem), only rosamarinic acid corresponds to the crude formula giving the following structure.

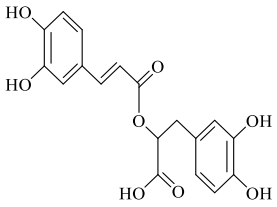
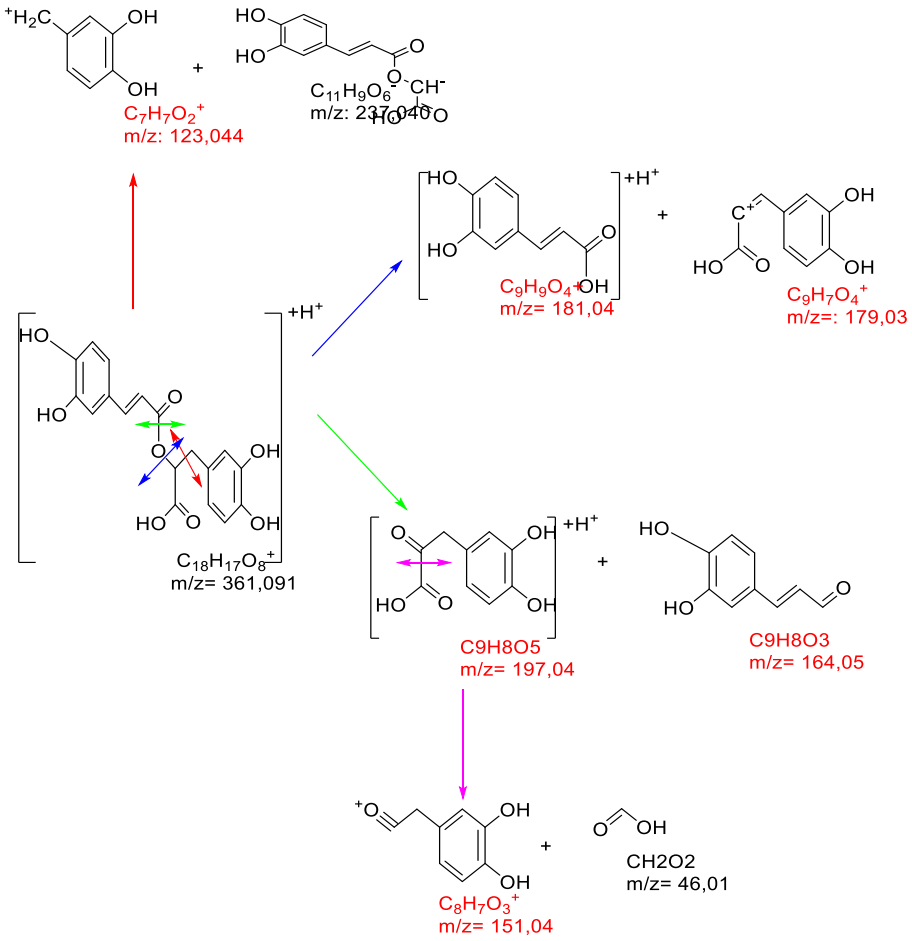


Fig. 32. Structure of Compound 15: rosamarinic acid ((E)-3-(3,4-dihydroxyphenyl)-2-((3-(3,4-dihydroxyphenyl)acryloyl)oxy)propanoic acid)

The identified compound fragmentation mode could be as following (Scheme 15).



Scheme 15. Compound 15 fragmentation mode

The most abundant peak with mass= 123 would derives from carbon-carbon bond cleavage at β-position of carboxyl group. Fragment with mass= 179 would rise from carbon-oxygen bond cleavage at β-position of carboxyl group. Fragment with mass= 163 would result to carbon-oxygen bond cleavage at γ of the carboxylic group. Fragment with mass= 151, would link from carbon-carbon and carbon-oxygen bonds cleavage respectively at α and γ of the carboxylic group (Scheme 15). Compound 15 is hence Rosamarinic acid, belonging to phenolic family. It has already been reported (Prawatsri S and al, 2013, Lautie E and al, 2008).

4. CONCLUSION

This study carried out on ethyl acetate fraction of *Hyptis suaveolens* leaves revealed several compound (around 40). Many of the molecular formulas proposed have been first identified in the Hyptis genus. Still, fifteen structures are already identified in the genus. Among these fifteen, eight have'nt been identified in our previous studies. Moreover, these eight new molecules are phenolic compound. Nine of the fifteen elucidated structures are phenolics compouds and six are terpenoids. All these bioactive compounds in *Hyptis suaveolens* leaves makes this plant an alternative for combating many diseases caused by oxidative stress.

ACKNOWLEDGEMENT

The technical inputs of Dr Dibi Jacques Konan of organic chemistry and natural substances (LCOSN), UFR-SSMT, University of Felix Houphouët-Boigny are acknowledged.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

Aspinall, G., Capek, P., Carpenter, R., Gowda, D. C., & Szafranek, J. (1991). A novel L fuco-4-O-methyl-D-glucurono-D-xylan from *Hyptis suaveolens*. *Carbohydrate Research, 214*(1), 107–113.

Bouamama, H., Noël, T., Villard, J., Benharref, A., & Jana, M. (2006, March). Antimicrobial activities of the leaf extracts of two Moroccan *Cistus* L. species. *Journal of Ethnopharmacology, 104*, 104–107. Available at: https://linkinghub.elsevier.com/retrieve/pii/S0378874105006033

Chukwujekwu, J., Smith, P., Coombes, P., Mulholland, D. A., & Staden, J. V. (2005). Antiplasmodial diterpenoid from the leaves of *Hyptis suaveolens*. *Journal of Ethnopharmacology, 102*(2), 295–297.

Edeoga, H., Omosun, G., & Uche, L. (2006). Chemical composition of *Hyptis suaveolens* and *Ocimum gratissimum* hybrids from Nigeria. *African Journal of Biotechnology, 5*(10), 892–895.

Ekow, N. T., Dzobo, K., Adu, F., Chirikure, S., Wonkam, A., & Dandara, C. (2018). Studies on the chemical compositions of *Hyptis suaveolens* (L.) Poit. *Journal of Ethnopharmacology, 58*.

Goly, K. R. C., Soro, Y., Dadie, A., Kassi, A. B., & Djé, M. (2015). Antibacterial activity of essential oils and extracts from the leaves of *Hyptis suaveolens* and *Lippia multiflora* on multi-resistant bacteria. *Rasayan Journal of Chemistry, 8*(4), 396–403. Available at: http://www.rasayanjournal.com, http://www.rasayanjournal.co.in

Goly, K. R. C., Soro, Y., Dadie, A., Kassi, A. B., & Djé, M. (2015). Antibacterial activity of essential oils and extracts from the leaves of *Hyptis suaveolens* and *Lippia multiflora* on multi-resistant bacteria. *Rasayan Journal of Chemistry, 8*(4), 396–403.

Jongmin, A., Hee-Sung, C., & Young-Won, C. J. K. (2017). Dereplication-guided isolation of new phenylpropanoid-, substituted diglycosides from *Cistanche salsa* and their inhibitory activity on NO production in macrophage. *Molecules, 22*, 1138.

Kumar, S., Nandha, & Thampi, N. (2015). Phytochemical screening and characterization of the bioactive compounds from the leaves of *Hyptis suaveolens* and *Spathodea campanulata*. *Journal of Chemical and Pharmaceutical Research, 7*(7), 840–850. Available at: www.jocpr.com

Lautie, E., Quintero, R., Fliniaux, M. A., & With, V. M.-L. (2008). Selection methodology with scoring system: Application to Mexican plants producing podophyllotoxin related lignans. *Journal of Ethnopharmacology, 120*(3), 402–412.

Mahesh, S., Chatterjee, A., & Pakrashi, S. C. (2001). *The Treatise on Indian Medicinal Plants* (Vol. 1). PID, New Delhi.

Malele, R., Mutayabarwa, C., Mwangi, J. W., & Thoithi, G. (2003). Essential oil of *Hyptis suaveolens* (L.) Poit. from Tanzania: Composition and antifungal activity. *Journal of Essential Oil Research, 15*(6), 438–440.

Manchand, P. S., White, J. D., Fayos, J., & Clardy, J. (1974). Chemical constituents of tropical plants. Structures of suaveolic acid and suaveolol. *Journal of Organic Chemistry, 39*(15), 2306–2308.

Misra, T. N. S. R., & U. (1983). A natural triterpene acid from *Hyptis suaveolens*. *Phytochemistry, 22*(11), 2557–2558.

Mukherjee, K., Mukherjee, R., & Ghosh, P. (1984). Chemistry of *Hyptis suaveolens*: A pentacyclic triterpene. *Journal of Natural Products, 47*(2), 377–378.

Oliveira, M. J., Campos, I. F. P., Oliveira, C. B. A., Santos, M. R., Souza, P. S., Santos, S. C., et al. (2005). Influence of growth phase on the essential oil composition of *Hyptis suaveolens*. *Biochemical Systematics and Ecology, 33*(3), 275–285.

Prawatsri, S., Suksamrarn, A., Chindaduang, A., & Rukachaisirikul, T. (2013, August). Abietane diterpenes from *Hyptis suaveolens*. *Chemistry & Biodiversity, 10*(8), 1494–1500. Available at: http://doi.wiley.com/10.1002/cbdv.201200213

Rao, R. K. V., Rao, L. J. M., & Prakasa Rao, N. S. (1990). An A-ring contracted triterpenoid from *Hyptis suaveolens*. *Phytochemistry*.

Rivaro, P., Vivado, D., Ianni, C., Salis, A., Parodi, A., & Millo, E. (2024, January 6). A new approach to characterize siderophore-type ligands in seawater by solid phase synthesis and SPE-HPLC-ESI-MS / MS analysis. *Journal of Marine Science and Engineering, 12*(1), 110.

Shah, K., Sharma, G. P., & Sagar, R. (2024, December). An arrow in the quiver: Evaluating the performance of *Hyptis suaveolens* (L.) Poit. in different light levels. *Ecological Processes, 13*(1), 1–8.

Soumahoro, B., Bohui, G. S., Kalo, M., Kanaté, L., Attioua, B., & Soro, Y. (2024). Compound identification by HPLC-ESI-Q-TOF-MS / MS analysis of the dichloromethane fraction of *Hyptis suaveolens* leaves after extraction of the essential oil. *Science, 12*(1), 1–14.

Soumahoro, B., Bohui, G. S., Kalo, M., Kanaté, L., Attioua, B., & Soro, Y. (2024). Compound identification by HPLC-ESI-Q-TOF-MS/MS analysis of the dichloromethane fraction of *Hyptis suaveolens* leaves after extraction of the essential oil. *Science, 12*(1), 1–4.

Soumahoro, B., Konan, K. T., Benjamin , A. B., & Soro, Y. (2023). Comparative study of *Hyptis suaveolens* leaves antioxidant activity before and after essential oil extraction. *International Journal of Chemical Studies, 11*(5), 27–33.

Soumahoro, B., Kone, K. P. F. O., Douhore, G. Y. T., & Soro, Y. (2023). Identification of compounds by HPLC-ESI-Q-TOF-MS / MS analysis of the dichloromethane fraction from the hydroalcoholic extract of *Hyptis suaveolens* leaves before extraction of the essential oil. *Journal of Materials and Environmental Science, 14*(12), 1529–1550.

Soumahoro, B., Kone, K. P., Douhore, G. Y., & Soro, Y. (2023). Identification of compounds by HPLC-ESI-Q-TOF-MS/MS analysis of the dichloromethane fraction from the hydroalcoholic extract of *Hyptis suaveolens* leaves before extraction of the essential oil. *Journal of Materials and Environmental Science, 14*(12), 1529–1550.

Suárez-Ortiz, G. A., Cerda-García-Rojas, C. M., Fragoso-Serrano, M., & Pereda-Miranda, R. (2017, January 27). Complementarity of DFT calculations, NMR anisotropy, and ECD for the configurational analysis of brevipolides K-O from *Hyptis brevipes*. *Journal of Natural Products, 80*(1), 181–189. Available at: https://pubs.acs.org/doi/10.1021/acs.jnatprod.6b00953

Tang, G., Xile, L., Xue, G., Xiaojing, L., Xiudi, L., & Ji, D. W. A. S. (2018). Studies on the chemical compositions of *Hyptis suaveolens* (L.) Poit. *Journal of the Serbian Chemical Society, 83*, 1–15.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/126416