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QSAR study for newly caffeic acid amides with prominent antibacterial and antifungal activity

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ABSTRACT

QSAR can modify the molecular structures for achieving the desired molecule with the proposed property, without experimental measurement. In the current study, we extend a published work that had been investiged the caffeic acid derivatives as antibacterial and antifungal agents. In this report, QSAR and regression analysis were used to predicate the antimicrobial activity of these derivatives. Moreover, the antimicrobial activity for these molecules that was obtained experimentally is compared with the calculated ones. The antimicrobial activity of some of newly postulated caffeic acid derivatives showed a pronounced antimicrobial activity. QSAR and regression equations were useful in predicating the biologic activity of the old and postulated molecules with good validity.

Key Words: QSAR, Antibacterial, antifungal, caffeic acid.

INTRODUCTION

Caffeic acid is one of the most widely distributed hydroxycinnamate and phenylpropanoid metabolites in plant tissues. It is usually found as various simple derivatives including amides, esters, sugar esters, and glycosides [1]. Caffeic acid esters and amide derivatives exhibit a broad spectrum of biological activities, including anti-oxidative properties [2, 3], anti-inflammatory [4], antinociceptive [5], antitumor [6], and potential antimicrobials [7].

QSAR is a mathematical relationship between a biological activity of a molecular system and its geometric and chemical characteristics. QSAR is used to find consistent relationship between biological activity and molecular properties, so that these rules can be used to evaluate the activity of new compounds. The purpose of developing a QSAR model is to reduce the cost of the target designing by modifying the molecular structures for achieving the desired molecule with the proposed property, without experimental measurement [8]. Subsequently, an ideal QSAR model should be capable of accurately predicting the desired property of a newly synthesized or a hypothetical molecule [9]. In the current study, we applied the QSAR and regression analysis for prediction the antimicrobial activity of newly caffeic acid derivatives.

MATREIALS AND METHODS

This work is based on previous investigations of caffeic acid amides. The synthesis and properties of these caffeic acid derivatives were reported earlier [3, 7]. Haung *et al* [3] mentioned the antiplatelet and antioxidative activities for their compounds and did not mention the antimicrobial activity, thus we investigated it herein using QSAR and regression analysis. On the other, Fu *et al* [7], mentioned the antimicrobial activity for their compounds, hence we speculated new derivatives and derived the antimicrobial activity for our speculated ones and compared the calculated activity with the experimental activity for their compounds.

Quantitative structure activity relationship (QSAR)

The descriptors obtained from hyperchem version 8 programs at the semi-empirical theoretical method using AM1 method [10].

Semi-empirical method

The calculation method for commands was placed on the compute menu to semi-empirical quantum mechanics rather than molecular mechanics or ab-initio quantum mechanics. These calculations solve the Schrödinger equation, with certain approximations, to describe the electron properties of atoms and molecules. In semi-empirical method, the calculations can be simplified by calculating the valence electrons only, neglecting the integrals for certain interactions using standard, non-optimized, and electron orbital basis functions. Experimental parameters eliminate the need to calculate certain quantities and to correct for errors resulting from approximations. This method is applicable and appropriate for all atoms in the periodic table, where the variables are saved in the parameter files. The choice remains until one chooses the molecular mechanics or Ab-Initio module. If a file is saved after a semi-empirical calculation, the HIN file will contain the calculated atomic charges [11].

AM1

AM1 is a semi-empirical SCF and a developed MNDO method for chemical calculations [12]. It is useful for molecules containing elements from long rows 1 and 2 of the periodic table, but not transition metals. Together with PM3, AM1 is generally the most accurate semi-empirical method included in Hyperchem., it calculates the electronic properties, optimized geometries, total energy, and heat of formation.

Statistical analysis

Multiregression analysis was used for correlating physicochemical descriptors to the biological activity through QSAR using winks program [13, 14].

RESULTS AND DISCUSSION

Many of caffeic acid amides such as N-(3-Cyano-phenyl)-3-(3,4-dihydroxy-phenyl)-acrylamide, and 3-(3,4-Dihydroxy-phenyl)-N-(3,4-dimethoxy-phenyl)-acrylamide were investigated earlier for their biological activity [3, 7]. Haung *et al* [3] reported the antiplatelet and antioxidative activities for their compounds and did not mention the antimicrobial activity, thus we investigated it herein using QSAR and regression analysis. On the other hand, Fu *et al* [7], mentioned the antimicrobial activity for their compounds, hence we speculated new derivatives and derived the antimicrobial activity for our speculated ones and compared the calculated activity with the experimental activity for their compounds.

It is well known that the work of Fu *et al* was based on constructing and preparing the chemical compounds and testing each one individually as antimicrobial agent, in which trials and errors method was followed. Because the preparation of caffeic acid amides is very expensive, tedious, time consuming and require lengthy procedures. Accordingly, QSAR equations using physicochemical parameters can help in this situation. In the current study, QSAR equations have been elaborated to predict new caffeic acid amides with potential antimicrobial activity. In our work, the data obtained from QSAR are based on their chemical structures of caffeic acid amides (scheme 1). The physicochemical properties (descriptors) of the investigated chemical compounds are illustrated in table 1. These descriptors include the area, volume, hydration energy, logarithm of partition coefficient, high occupied molecular orbital, low unoccupied molecular orbital, difference between LUMO, dipole moment on X directions dmx, net dipole moment, gradient charge by K Cal/mole angstrom on carbon atom [$C_{(9)}$], oxygen atom [$O_{(10)}$], charge on oxygen atom [$O_{(12)}$], charge on nitrogen atom [$N_{(20)}$], [$O_{(9)}$], charge on keto oxygen [$O_{(10)}$], and charge on nitrogen atom [$N_{(12)}$].

The descriptors obtained from hyperchem at semiemperical theoretical method [13]. Fruitful descriptors are gained using multi-regression statistical calculations in winks program [17] feeding with these descriptors together with the biological activities previously measured. It is noted that, the data obtained from multi-regression calculated by winks include equations used for calculating the biological activity (antibacterial and Antifungal) of the compounds in concern as well as focusing on the most chief descriptors affecting the biological activity. Accordingly, four equations had been obtained from multi-regression statistical calculations. Equation one is concerned with calculating the ability of the caffeic acid derivatives to act as antibacterial agent against *B. subtilis*.

Equation one: *B. subtilis* = -767.6 ^{10}O + 678.6 ^{12}O + 868.9 ^{20}N -1829.4 LUMO +14.0 Total Dipole -0.29 Volume + 15.21 Dipole X + 8.85 Log p +1916.68 HOMO +0.37 Heat of Formation -1882.35 (HOMO –LUMO) +572.66

The other three equations that have been developed and shown below (equation two to four) are concerned with calculating the ability of the caffeic acid derivatives to act as antimicrobial agent against *P. fluorescence*, *S. aureus* and *C. albicans* respectively.

Equation two: *P. fluorescence* = 67.36 ^{9}C + 51.88661 ^{10}O +191.75 ^{11}O +81.64 ^{12}O + 134.67 ^{20}N -23.25 LUMO -3.74 Total Dipole -0.002 Volume -4.183156 Dipole X -1.20 Log p -0.04 Surface Area -0.95 Hydration Energy +3.736 HOMO -0.016 Total Energy -0.0075 Heat of Formation +75.36

Equation three: *S. aureus* =101.85 ^{9}C + 59.89 ^{11}O +124.40 ^{12}O +89.12 ^{20}N +227.33 LUMO +1.42 Total Dipole + 0.054 Volume + 3.009 Dipole X +1.19 Log p -0.045 Surface -221.43 HOMO -0.015 Total Energy + 0.023 Heat of Formation +0.023 MW -1.576176 Polarization +228.62 (HOMO –LUMO)+123.25

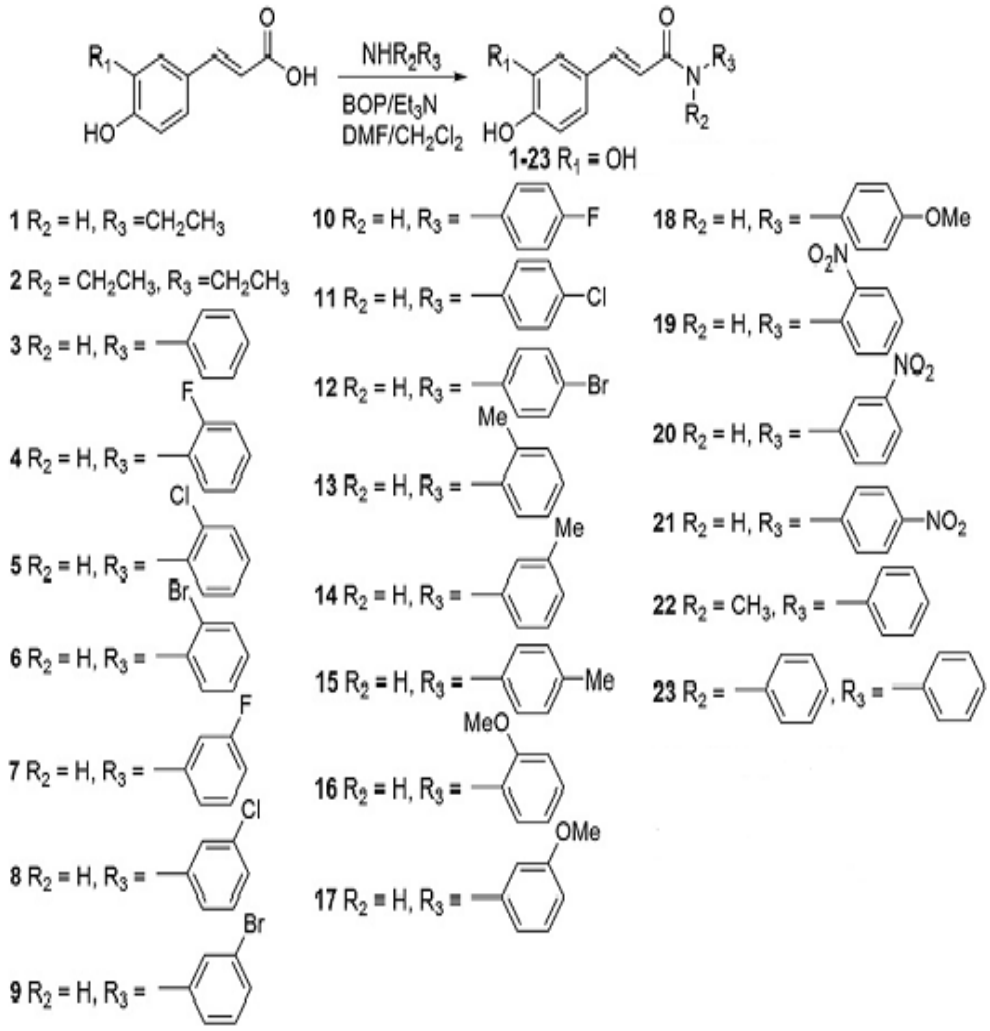
Equation four: *C. albicans* =-135.63 ^{10}O -350.77 ^{11}O -80.89 ^{20}N -54.64 LUMO -6.83 Total Dipole-0.07 Volume -4.11 Dipole X -1.82 Log p -0.002 Surface Area -0.13 Hydration Energy +22.18 HOMO -0.014 Total Energy +0.09 Heat of Formation +282.25

The degree of the validity of the four equations obtained from multi-regression statistical calculations was measured via different tools. One of such is based on calculating the biological activity and applying our proposed equations. The data obtained are monitored with that obtained from the work of Fu *et al*. (9) and tabulated in table 2 for comparison purposes. Reading such table, one can easily notice that the great concordance between the results obtained experimentally by Fu *et al* and that calculated by using our equations. As shown from the results presented in table four, the value of R is close to unity reflecting more validity of the proposed equations. Through reading the data in table four especially the F and P-values (Table 3), one can touch the highly proximity of calculated values to the experimentally measured biological activities.

Based on the experimental results of Fu et al, (Table 2), caffeic acid anilides 10–12, 15, 18 and 21 exhibit excellent activities against *B. subtilis*, whereas caffeic acid fatty amides 1 and 2 obviously showed much lower inhibition. This indicated that anilides had better antibacterial activities than fatty amides. Caffeic acid secondary amides 2, 22 and 23 had low activities, proving that H–N is important for the activities. Although caffeic acid anilides exhibit better antibacterial activity against *B. subtilis*, their MICs differ significantly. One rule could be found: anilides with a single substitute at the p-positions of the benzene ring exhibit the most powerful inhibitory activities, whereas anilides with a substituent at o- or m-positions show less potent activities. From this fact, it is reasoned that the p-position is an essential point for the antibacterial activities of anilides, and substituents at the o- or m-positions decrease their inhibition. For example, when a methyl is introduced at p-position to form 15, the activity is the most significant. However, when a methyl was introduced at o-position (or m-position) to form 13 (or 14), the activity decreases significantly. Replacement of methyl group of (15) at p-position by a bromide (12) resulted in the decreases of activity. Similarly, substitution of a chloride (11) at p-position for a methoxy group (18) also led to a decrease of inhibitory activity. This suggested that compounds with electron donating groups at p-position showed better inhibitory activities than those with electron-withdrawing groups. For more extensive evidences, the substitution of methoxy group at p-position (18) by a fluoride or nitro (10 or 21) produced some decrease in activity [7].

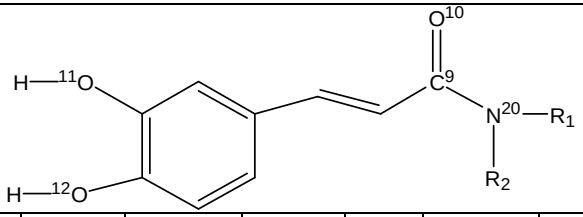
In view of the aforementioned discussion and according to the facts obtained from applying Hyperchem programs, the descriptors of the newly postulated structures are examined (Figure 1) and such data are presented in table 4 &5. Taking into account these data and applying our equations obtained from Hyperchem, the biological activity of these compounds is calculated and illustrated (table 6). These compounds are speculated taking into account that they having caffeic acid derivatives and the rest of their structures are completed by active sites complementing the best descriptors obtained from our Hyperchem investigation. Thus the newly caffeic acid derivatives remain to be synthesized and investigated experimentally for their

antimicrobial properties. Finally, our data may be exhibited a potential interest for investigators attempting to find new antimicrobial agents.



Scheme: 1[7]

Table (1): Calculated descriptors by HyperChem for the caffeic acid derivatives presented in scheme one. [13]



SN	^{9}C	^{10}O	^{11}O	^{12}O	^{20}N	LUMO	Total Dipole	Volume	Dipole X	Log p	Surface	HE	HOMO	Total Energy	Heat of Formation	MW	POLARI	(HOMO – LUMO)
1	-0.0127	0.008	0.0068	0.002	0.007	-0.270	0.756	739.1	-0.193	-0.94	509.2	-16.8	-8.434	-101.177	-40.39	207.23	22.3	-8.164
2	-0.017	0.005	-0.00192	-0.006	-0.005	-0.225	1.100	819	-0.961	-0.35	358	-13.5	-8.32	-112.60	-39.06	235.28	25.8	-8.095
3	-0.011	-0.007	0.00211	-0.015	-0.001	-0.251	2.650	775	-2.35	-0.79	407	-18.8	-8.423	-120.00	-26.07	255.27	28.1	-8.172
4	-0.013	0.024	-0.00779	0.005	0.031	-0.286	2.848	787	-1.007	-1.39	406	-17.8	-8.597	-137.32	-66.18	273.26	28.03	-8.311
5	-0.012	-0.004	-0.01103	0.006	-0.001	-0.255	1.981	821	0.231	-1.01	429	-17.5	-8.607	-133.22	-26.24	289.75	30.05	-8.352
6	-0.009	0.002	0.00752	-0.009	0.006	-0.269	2.095	839	0.350	-0.74	438	-17.5	-8.623	-132.47	-13.69	334.17	30.74	-8.354
7	-0.001	0.009	-0.00332	0.004	0.006	-0.364	1.254	790	0.239	-1.4	419	-17.8	-8.573	-137.32	-67.50	273.26	28.03	-8.209
8	0.007	0.004	-0.01072	-0.015	0.041	-0.35	0.976	822	0.214	-1.1	442	-17.4	-8.560	-133.23	-29.38	289.72	30.05	-8.207
9	0.024	0.005	-0.005	-0.008	0.013	-0.358	1.100	843	0.300	-0.74	451	-17.7	-8.569	-132.47	-17.31	334.17	30.74	-8.211
10	-0.009	-0.002	0.003	0.009	0.008	-0.368	0.962	790	0.089	-1.4	421	-17.9	-8.501	-137.32	-67.62	273.23	28.03	-8.133
11	0.014	0.002	-0.002	-0.006	-0.014	-0.368	0.749	823	0.095	-1.01	445	-17.8	-8.503	-132.23	-29.76	289.72	30.05	-8.135
12	0.004	0.016	-0.013	0.002	0.009	-0.389	1.016	842	0.149	-0.74	454	-17.8	-8.528	-132.47	-18.00	334.17	30.74	-8.139
13	0.029	-0.009	-0.022	0.009	-0.012	-0.239	1.298	825	-0.239	-0.63	429	-16.8	-8.576	-125.72	-27.76	269.3	30	-8.337
14	0.003	0.004	-0.011	-0.003	-0.007	-0.264	1.136	831	-0.170	-0.63	447	-16.9	-8.402	-125.72	-30.09	269	30	-8.137
15	0.011	0.012	-0.012	-0.009	0.005	-0.263	1.218	832	-0.148	-0.63	453	-16.9	-8.335	-125.72	-30.23	269	30	-8.072
16	-0.013	-0.013	-0.022	0.048	-0.013	-0.298	0.396	846	0.022	-1.78	389	21.6	-8.643	-137.50	-71.16	285.3	30.6	-8.345
17	-0.002	0.002	-0.008	0.028	0.0005	-0.262	1.984	855	-0.244	-1.78	465	-19.5	-8.410706	-137.48	-60.07	285.3	30.6	-8.149
18	0.008	0.004	0.008	0.003	-0.009	-0.439	2.670	883	-0.714	-1.78	453	-19.8	-8.631	-137.37	6.201	285.3	30.6	-8.192
19	-0.003	-0.002	0.001	0.004	0.009	-0.385	2.931	829	0.474	-5.47	434	-21.0	-8.670	-150.37	77.46	300.27	30	-8.294
20	-0.001	0.004	-0.005	0.008	0.009	-0.483	2.770	840	0.589	-5.47	426	-23	-8.705	150.38	76.4	300.27	30	-8.222
21	-0.0013	-0.004	-0.004	-0.011	-0.004	-1.017	5.990	838	0.968	-5.47	463	-23.1	-8.886	-150.53	-20.24	300.27	30	-7.869
22	-0.009	-0.003	0.013	0.0001	0.002	-0.507	2.578	841	-1.161	-0.54	408	-15.6	-8.742	-125.60	43.53	269.3	29	-8.23489
23	0.0071	-0.006	-0.006	-0.001	0.028	-0.242	0.968	975	-0.708	-0.05	457	-17.0	-8.158	-150.20	21.30	331.37	37.78	-7.915

^{9}C: charge on carbon atom 9; ^{10}O: charge on oxygen atom10; ^{11}O; charge on oxygen atom 11; ^{12}O: charge on oxygen atom 12; ^{20}N: charge on nitrogen atom 20; LUMO: low unoccupied molecular orbital; HOMO: high occupied molecular orbital; H.E: hydration energy; Log P: Log of calculated octanol-water partition coefficient; MW: molecular weight; dmx (dipole x): dipole moment in X direction.

Table (2): The Antimicrobial Activities against *B. subtilis, P. fluorescence, S. aureus* and *C. albicans* of caffeic acid amides as determined theoretically (by equations one to four) and experimentally as reported earlier [7].

Biological Activity	Antimicrobial Activity							
	B. subtilis		*P. fluorescence*		*S. aureus*		*C. albicans*	
Compound	Exper.	Calcul.	Exper	Calcul	Exper	Calcul	Exper	Calcul
1	50.00	49.20	50	49.3	50.0	49.3	50.0	51.2
2	50.00	18.90	50	52.4	50.0	52.4	50.0	49.7
3	28.40	33.60	50	42.0	41.5	42.0	50.0	49.3
4	50.00	43.10	50	49.4	50.0	49.4	33.3	34.3
5	45.80	53.60	41.2	48.2	50.0	48.2	42.8	40.7
6	49.50	49.30	42.2	50.4	50.0	50.4	27.5	31.1
7	46.70	24.00	50	49.8	50.0	49.8	50.0	47.2
8	50.00	49.40	50	49.6	49.2	49.6	50.0	49.2
9	50.00	34.30	50	51.8	50.0	51.8	50.0	47.3
10	7.95	33.70	50	49.9	50.0	49.9	50.0	50.7
11	6.25	6.70	50	47.0	46.8	47.0	50.0	54.7
12	3.89	25.30	50	49.8	50.0	49.8	50.0	52.7
13	35.00	37.30	42.5	49.2	50.0	49.2	50.0	51.3
14	40.20	24.30	38.8	46.8	45.6	46.8	50.0	50.6
15	1.18	27.40	42.2	48.3	50.0	48.3	50.0	49.5
16	39.80	19.70	13.2	48.9	47.4	48.9	50.0	50.9
17	50.00	35.70	50	49.9	50.0	49.9	44.7	42.0
18	3.12	12.00	50	49.8	50.0	49.8	42.8	42.3
19	50.00	64.10	50	50.3	50.0	50.3	50.0	49.8
20	50.00	51.30	50	46.9	50.0	46.9	50.0	51.6
21	15.50	16.10	50	49.2	50.0	49.2	49.4	52.0
22	50.00	45.70	50	50.9	50.0	50.9	50.0	47.2
23	50.00	45.90	50	49.4	50.0	49.4	50.0	48.4

Table (3): Regression analysis reflecting the validity of the proposed four equations.

	R-Square	p-value	F
Equation 1 (*B. subtilis*)	0.657	0.148	1.914
Equation 2 (*P. fluorescence*)	0.955	0.003	9.97
Equation 3 (*S .aureus*)	0.961	0.006	9.34
Equation 4 (*C. albicans*)	0.902	0.004	6.37

Where F, P and R are respectively the degree of freedom, the significance and regression coefficient.

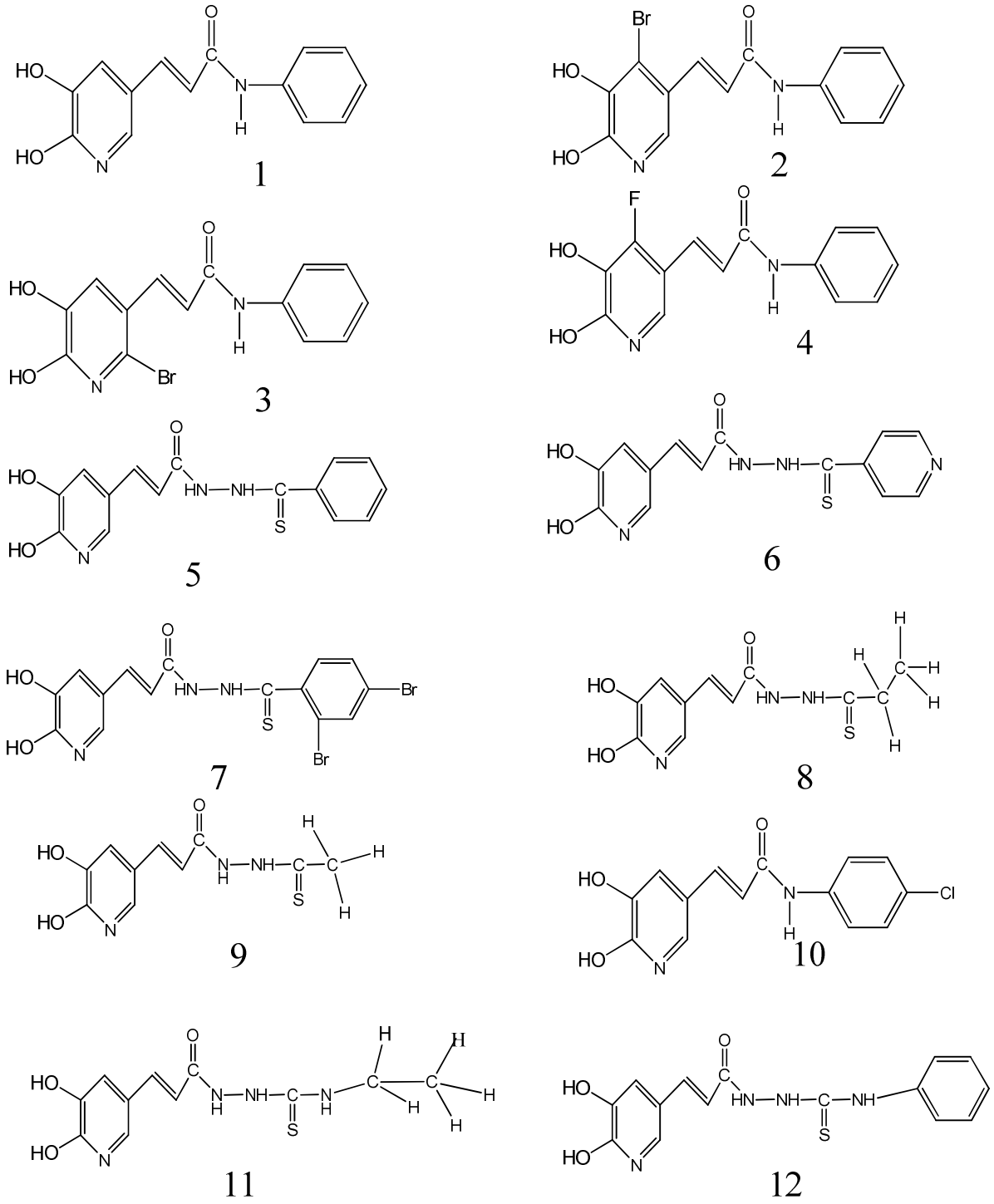


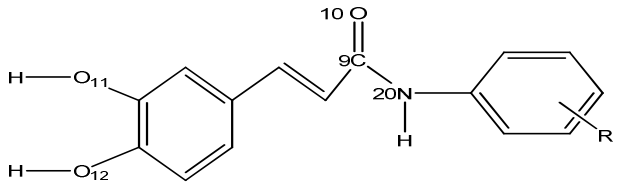
Figure (1): structure of the newly speculated molecules based on moiety of caffeic acid presented by Fu *et al* [7].

Table (4): Calculated physicochemical descriptors of newly speculated chemical compounds (structure is shown in figure one).

SN	^{9}C	^{10}O	^{11}O	^{12}O	^{20}N	LUMO	Total Dipole	Volume	Dipole X	Log p	surface	H E	HOMO	TOTAL ENERGY	Heat of Formation	MW	POLARI	(HOMO-LUMO)
1	0.013	0.017	-0.005	-0.012	0.001	-0.490	3.679	733.4	3.618	-0.7	239.86	-17.42	-8.90	-122	-17.5	-256.26	-27.41	-8.50
2	0.021	-0.004	-0.021	-0.002	0.002	-0.410	3.066	786	2.851	-0.65	280.6	-16.7	-9.04	-135	-8.6	335	30	8.99
3	0.005	-0.003	-0.017	-0.002	-0.001	-0.415	3.769	794	2.492	0.74	277.5	-17.04	-8.98	-135	-9.2	335	30	-8.56
4	-0.004	0.007	-0.020	0.025	0.004	-0.471	3.226	744	3.052	-1.3	257	-17.03	-9.12	-140	-60.0	274.3	27.3	-8.56
5	0.040	-0.007	-0.001	-0.008	-0.004	-1.103	0.726	906	-0.029	-029	239	-21.78	-9.13	-160	60.4	333.34	33.75	-8.01
6	0.021	0.002	-0.001	0.0043	-0.001	-1.157	1.485	883	-0.513	-1.16	232.7	-23.4	-9.18	-145	115.8	316.3	33.13	-8.03
7	0.020	0.007	-0.003	-0.009	0.004	-1.123	1.344	1010	-0.161	-0.42	310.23	-21.00	-9.15	-167	118.1	473.14	39.01	-8.03
8	0.001	0.016	0.001	-0.009	-0.008	-0.981	2.722	785	2.471	-0.21	172.16	-14.66	-9.01	-123	61.5	267.3	27.85	-8.0
9	0.009	-0.0071	0.006	0.010	0.002	-0.979	2.626	730	2.396	-0.84	178.4	-24.4	-8.99	-118	67.5	253.3	26.02	8.01
10	0.009	-0.006	-0.010	-0.001	-0.001	-0.861	4.691	780	4.496	0.09	261.21	-21.75	-9.13	-155	98.5	349.8	35.77	-8.27
11	-0.001	0.001	-0.009	0.002	-0.001	-0.104	4.762	853	4.562	-0.18	249.5	-16.05	-8.82	-132	33.5	282.3	29.2	-8.72
12	0.013	0.013	-0.007	0.005	-0.004	-0.897	4.278	902	4.259	-0.1	300	-27.0	-8.45	-150	45.4	330.36	35.19	-7.54

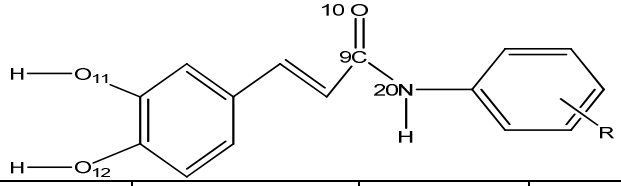
^{9}C: charge on carbon atom 9; ^{10}O: charge on oxygen atom10; ^{11}O; charge on oxygen atom 11; ^{12}O: charge on oxygen atom 12; ^{20}N: charge on nitrogen atom 20; LUMO: low unoccupied molecular orbital; HOMO: high occupied molecular orbital; H.E: hydration energy; Log P: Log of calculated octanol-water partition coefficient; MW: molecular weight; dmx (dipole x): dipole moment in X direction; dm (total dipole): polarization magnitude of dipole moment.

Table (5): Calculated physicochemical descriptors of twelve caffeic acid amides and their chemical structure are presented earlier by Haung *et al*. [3]



SN	R	^{9}C	^{10}O	^{11}O	^{12}O	^{20}N	LUMO	Total Dipole	Volume	Dipole X	Log p	surface	H E	HOMO	TOTAL ENERGY	Heat of Formation	MW	POLARI	(HOMO-LUMO)
1	2OH	-0.003	0.001	-0.010	0.013	0.021	-0.38	4.13	766.6	3.9	-1.8	271.3	-21.6	-8.67	-132	-72.5	271.3	28.8	-8.3
2	3OH	0.004	0.016	-0.003	0.007	0.022	-0.42	3.04	768.2	2.34	-1.81	271.3	-22.9	-8.72	-132	-72.0	271.3	28.8	-8.3
3	4OH	0.007	0.013	-0.002	0.009	0.003	-0.4	2.55	764.1	2.2	-1.81	271.3	-22.8	-8.71	-132	-72.6	271.3	28.8	-8.3
4	2Cl	0.006	-0.003	-0.007	-0.003	0.003	-0.38	3.3	744.5	2.94	-0.79	251.6	-16.04	-8.70	-120	-28.1	255.3	28.11	-8.3
5	3Cl	-0.010	0.014	-0.005	0.008	0.002	-0.49	2.96	789.3	2.16	-1.0	289.0	-15.7	-8.76	-133	-35.7	289.7	30.1	-8.26
6	4Cl	0.013	0.031	-0.013	-0.012	0.007	-0.49	2.96	785.7	2.16	-1.0	290.2	-15.7	-8.74	-133	-35.7	289.7	30	-8.24
7	2 Br	0.015	0.028	0.001	-0.003	0.013	-0.55	2.39	806.6	1.97	-0.74	294.9	-15.6	-8.72	-132	-24.0	334.17	30.7	-8.17
8	3Br	0.007	-0.025	-0.022	-0.0012	0.005	-0.54	2	808.4	1.58	-0.74	298.2	-15.7	-8.77	-133	-24.3	334.17	30.74	-8.2
9	4Br	0.019	-0.007	-0.009	-0.010	0.011	-0.54	2.74	803.8	1.87	-0.74	299.9	-15.7	-8.757	-133	-24.3	334.17	30.74	-8.20
10	3CN	0.013	-0.001	-0.026	-0.005	0.024	-0.81	1	802.9	-0.52	-1.06	311.9	-20.3	-8.837	-132	3.3	280.3	30	-8.01
11	2-CO2Et	-0.232	0.015	0.314	0.342	0.091	-0.54	1.65	961.2	1.34	-1.03	363.4	-18.5	-8.78	-160	-86.0	327.3	34.4	-8.23
12	3,4 dimethoxy	0.019	-0.004	-0.010	-0.007	0.005	-0.33	3.62	845.7	3.11	-1.48	326.6	-13.9	-8.68	-131	-54.1	283.3	31.8	-8.35

Table (6): The calculated biological activity generated using the predictable four equations which are concerned with the descriptors of the speculated chemical compounds (Table 4) and non-speculated compounds (Table 5)

		Antimicrobial Activity			
Speculated Molecules		***B. subtilis***	***P. fluorescence***	***S. aureus***	***C. albicans***
1		49.22	49.28	49.28	51.16
2		287.31	132.64	132.64	22.94
3		-16142.60	2036.54	2036.54	28.28
4		110.31	65.28	65.28	21.98
5		-70.29	85.60	85.60	25.99
6		-315.38	28.97	28.97	135.39
7		-0.23	56.87	56.87	87.51
8		-36.63	56.22	56.22	75.47
9		28.38	73.53	73.53	59.25
10		-30059.60	3736.27	3736.27	67.22
11		167.58	66.10	66.10	37.92
12		175.03	77.35	77.35	-9.21
Non-Speculated Molecules[3]					
1a	2OH	89.34	77.05	77.05	43.62
2a	3OH	163.26	67.07	67.07	16.07
3a	4OH	93.82	62.45	62.45	26.65
4a	2Cl	42.04	63.90	63.90	31.46
5a	3Cl	88.20	69.67	69.67	28.29
6a	4Cl	63.16	62.42	62.42	32.46
7a	2 Br	31.16	62.69	62.69	33.06
8a	3Br	48.77	63.61	63.61	35.19
9a	4Br	28.88	63.92	63.92	53.71
10a	3CN	-4.91	53.75	53.75	82.99
11a	2-CO2Et	261.15	102.82	102.82	-87.44
12a	3,4 dimethoxy	82.43	63.69	63.69	15.58

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