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Design and Synthesis of Novel NSAIDs Class Acting as Anticancer Agents

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Abstract

Several effective anticancer therapeutic drugs containing coumarin nucleus. Thus, some coumarin derivatives 3-22 were prepared. The structures of these compounds established on the basis of spectral data as IR, ^{1}HNMR, ^{13}CNMR and MS. Moreover, the optimization geometries for compounds 3-13,15,17a,18a,19-22 were discussed using PM3 base set. The molecular docking simulations into the active site of COX-2 were performed, and showed that, some compounds 7,8, 11,13, and 20 are suitable inhibitor against COX-2, and can used as anti-cancer drugs. The compounds 7,8, 11,13 and 20 were evaluated on 50µg dose, against Ehrlich solid carcinoma (EAC) tumor model in Swiss albino mice . The activity was assessed using survival time and average increase in body weight, and showed that, these compounds are effective in reducing solid tumor mass EAC cells, and 20 showed highest potency. In silico, The ADMET profiles showed that, these 7,8, 11,13 and 20 compounds are good oral bioavailability, and are CNS active agents without marked health effects observed for rodent toxicity.

Keywords: NSAIDs; Coumarin; Anticancer agent; ADMET; DOCKIN and EAC.

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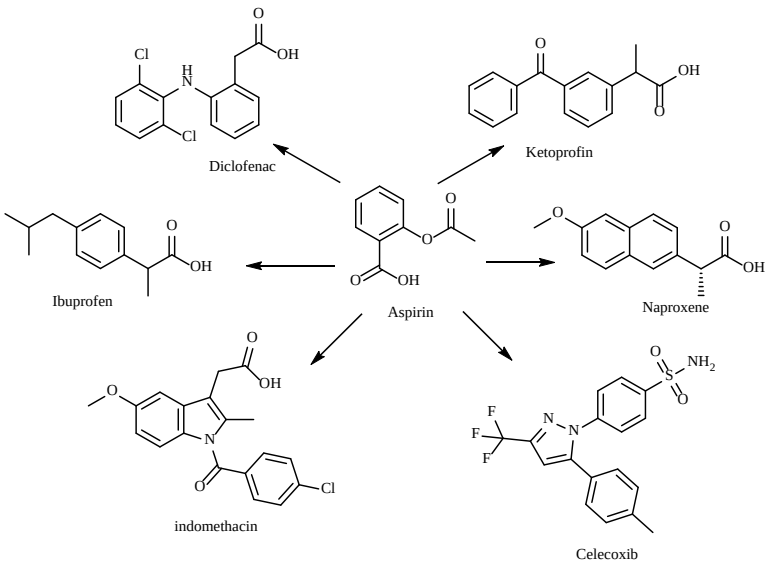
1. Introduction

Cancer is a major public health [1], which known as metastasis [2-4]. The chemotherapy agent is primary strategy for treatment of tumor cell, which depending on poisoned cancer cell [5]. Non-steroidal anti-inflammatory drugs (NSAIDs) are effective in different clinical setting, which widely employed with Patients suffering musculoskeletal and inflammatory diseases [6-9], and act as COX-2 inhibitors, through inhibiting the production of prostaglandins (PGs) [10-12]. The PGS level are increased in various tumor types, the inflammation plays a vital role in formation of tumor cell, and inhibit prostaglandin biosynthesis, which is responsible for the NSAIDs chemo preventive activity of [13-16]. Some famous marketing drugs (Strcture1) are suffering from gastrointestinal drawback [17-19]. Coumarin derivatives are a novel class of inhibitors of the carbonic anhydrase (CA) [20-22]. CA is a metalloenzymes family, which involved providing bicarbonate radical through an important physiological reaction [23-27]. The bicarbonate is important for formation tumor cell, reduction supplies of bicarbonate radical is suggested mechanism action of CA inhibitors (CAIs) [27]. Coumarin nucleus inhibit many isoforms CA I–CA XV, through the hydrolyzed coumarin nucleus to 2-hydroxycinnamic acid, which bind with CA to form enzyme-inhibitor adduct [28-30].



Strcture 1: Chemical strcture of commenly used markting NASIDS

In addition, the hydrophobic amino acid derivatives especially containing amide and thioamide moieties possess diverse biological activities, as anti-inflammatory, anti-tumor and antimicrobial activities [7-9]. Hence, the present work aims to synthesis a novel class series of NSAIDs containing phenylalanine moiety acting as new anticancer agent targeting COX-2. The molecular docking was carried out, to predict the correct binding geometries for each ligand at the active site, followed by molecular modelling to identify the structural features of these new Class.

2. Experimental

Melting points were taken on a Griffin melting point apparatus and are uncorrected. Thin layer chromatography (R_f) for analytical purposes were carried out on silica gel and developed. Benzidine, ninhydrin, and

hydroxamate tests used for detection reactions. The IR spectra of the compounds were recorded on a Perkin–Elmer spectrophotometer model 1430 as potassium bromide pellets and frequencies are reported in cm^{-1}. The NMR spectra were observed on a Varian Genini-300 MHz spectrometer and chemical shifts (δ) are in ppm. The mass spectra were recorded on a mass spectrometer HP model MS–QPL000EX (Shimadzu) at 70 eV. Elemental analyses (C,H,N) were carried out at the Microanalytical Centre of Cairo University, Giza, Egypt.

Swiss albino mice were collected with age form 5 weeks to 7 weeks old, and weighing 18-25 g from the National Cancer Institute (Cairo, Egypt) (NCIE), Cairo. The mice were kept in iron cages with sawdust and straw bedding that were changed once a week regularly. Standard mouse diet (recommended and prepared by the (NCIE) and water were given in adequate amounts.

EAC cells were obtained from a line of EAC cells were obtained from the Cancer Biology Department of the National Cancer Institute (Cairo, Egypt). Body weight was balance by using Sartorious (500.0) g AC-DC CHARGER MODEL:MW79.

Tumor volume was measured by VERNIER CALIPER 150X0.02 MM/6 XL/1000 and viable cell count was determined in a neubauer counting chamber.

2.1. 2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetyl chloride(2)

The starting material (**2**) was prepared as mentioned by elhenawy and his colleagues [5].

2.2. 2-(2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetamido)-3-phenylpropanoic acid (3)

L-phenylalanine(L-Phe.) was dissolved in 1N-NaOH, acid chloride (**2**; 0.01 mol) in acetone was added portion-wise during 1/2h at 10°C, the stirring was continued for additional 3h., the acetone was removed by concentration of the reaction mixture under reduced pressure, water was added, and acidified with 1 N HCl to pH =5. The crude product (**3**) was filtered and re-crystallized from ethanol- water to give compound (**3**) as dark brown crystals. The crude product (**3**) was obtained in 80%yeild R_f = 0.85 (CH2-Phe/MeOH=3/1); M.P. = 90-92C°; IR (KBr cm^{-1}) ν; 3405 cm^{-1}(OH); 3201 cm^{-1}(NH), 3090 (CH-arm.); 2973 cm^{-1}(CH-ali); 1722, 1665 cm^{-1} (C=O); ^{1}H NMR (300 MHz, Chloroform) δ= 10.40(s, 1H, OH-Carboxylic), 8.75(s,1H,OH-arm.), 7.21-6.7 (m,9H, CH-Arm..), 5.55 (s,1H,NH), 4.94(m,1H,CH-Phe), 3.54(s,2H, CH_2-ali.), 2.84 (d,2H,CH_2-Phe); Anal./Calcd. for $C_{20}H_{17}NO_6$ (367):C (65.37%), H(4.63%), N (3.81%). Found: C (65.39); H (4.66); N, (3.81).

2.3. 2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetylisothiocyanate (5)

A mixture of compound (**2**, 0.01 mol) and NH_4SCN (0.01mole) in acetone (10ml.) was stirred for 30 mins., the NH4Cl formed was filtered off, the filtrate containing compound (**5**) was used in other experiment without further purification.

2.4. 2-(2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetamido)-3-phenylpropanoyl chloride (6)

The compound **(3**; 0.01mol) in acetone (10 ml), was heated with $SOCl_2$ (0.01 mol.) for 30 mins. The acid chloride (**6**) formed, which used directly in other experiments.

2.5. 2-(2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetamido)-3-phenyl-N-(4-sulfamoylphenyl)propanamide (7).

Sulfanilamide (0.01mol) in acetone with acid chloride (**5**; 0.01 mol), the reaction mixture was continued stirring for additional 3 hrs. at room temperature. After evaporation of the reaction mixture, the resulting crystals was filtered and recrystallized from benzene to give compound (**7**) as brownish red crystals. The compound was obtained in (75) % yield. Rf = 0.96 (CH2-Phe/MeOH=3/1); M.P. = 135-37Cº; IR (KBr cm^{-1}) ν; 3353, cm^{-1} (OH), 3242 cm^{-1} (NH_2 , NH) ,2963 cm^{-1} (CH-ali), 1706 cm^{-1} (C=O),1621 cm^{-1} (CONH amide), 1335,1230 cm^{-1} (SO_2NH_2 and SO_2); MS (m/z, %) 522 (40.28%), Anal./Calcd. for $C_{26}H_{23}N_3O_7S$: C (59.86%), H (4.41%), N (8.05%). Found: C (59.88); H (4.45); N, (8.06).

2.6. Methyl-2-(2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetamido)-3-phenyl-propanoate (8)

The thionyl chloride (0.01 mol.) was added in dropwise during 30 mins, to a stirred cold solution (5 °C) of compound (**3**;0.01 mol) in absolute methanol (30 ml.). The stirring was continued for additional 3 hrs. at (5 °C), the mixture was left for 24 hrs. at room temperature. The mixture was removed under reduced pressure, another portion of abs. methanol (10 ml.) was added and re-evaporated. The solid obtained was recrystallized from benzene to give Compound (**8**) as brown crystal in 90 % yeild, Rf=0.87, (CH2-Phe/MeOH=3/1); M.P.146-48 Cº ,IR (KBr cm^{-1}) ν; 3275 cm^{-1} (NH), 2964 cm^{-1} (CH-ali..), 1721 cm^{-1} (C=O), 1619 cm^{-1} (CONH amide); ^{1}H NMR (300 MHz, Chloroform) δ=8.60(s,1H,OH), 7.686.33(m, 9H, CH-arm..), 5.42 (s,1H,NH), 4.58(m,1H,CH-Phe), 3.74(s,3H.CH_3, OCH_3), 3.32 (s,2H,CH2-ali..), 2.89 (d, 2H,CH2-Phe),13CNMR δ= 172.30,172.18, (2C, C=O),162.64-126.90(13C,C-arm..), 104.44(C-CH2.ali..), 53.84 (C-CH-Phe), 38.06(C-CH2-Phe), 37.63(C,CH_3) MS (m/z, %) 381 (1.26%), Anal./Calcd for $C_{21}H_{19}NO_6$: C(66.12%), H (4.98%), N(3.67%). Found: C (66.13); H (5.02); N, (3.67).

2.7. N-(1-hydrazinyl-1-oxo-3-phenylpropan-2-yl)-2-(7-hydroxy-2-oxo-2H- chromen-4-yl)acetamide (9)

The methyl ester derivative (**8**, 0.01 mol) was stirred in ethanolic hydrazine hydrate solution(0.01mol) for 3hrs, The crude product (**9**) was filtered, washed with water and recrystallized from benzene to give desired compound (**9**) in 90% yield;Rf = 0.80, (CH2-Phe/MeOH=3/1); M.P.230-32 Cº ,IR (KBr cm^{-1}) ν; 3405 cm^{-1} (broad band, OH ,NH and NH_2) ,2962,2931 cm^{-1} (CH-ali.), 1712, cm^{-1} (C=O), 1620 cm^{-1} (CONH amide), ^{1}H NMR (300 MHz, Chloroform) δ=10.94 (s,2H,NH_2), 8.74-(s,1H,OH-Arm.), 7.90(s,1H, NH-Hyd.), 7.52-6.28 (m,9H,CH-arm.), 5.66(s,1H,NH), 4.68 (s,1H,CH-Phe), 3.17 (s,2H,CH_2-ali..), 2.90 (s,2H,CH_2-Phe), MS (m/z, %) 381(1.06%), Anal./Calcd for $C_{20}H_{19}N_3O_5$: C (62.97%), H (4.98%), N(11.01%). Found: C (62.99); H (5.02); N, (11.02).

2.8. N-(1-(2-formylhydrazinyl)-1-oxo-3-phenylpropan-2-yl)-2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetamide (10)

Compound (**9**, 0.01mol) was heated under reflux with formic acid (0.01mol) for 10 hrs. The solid obtained after

cooling was filtered , and recrystallized from benzene to give compound (**10**) as brown crystals Yield: (65%); Rf = 0.83 (CH_2-Phe/MeOH=3/1); M.P.=185-87°C; IR (KBr cm^{-1}) ν; 3411 cm^{-1} (broad band, OH and NH), 2927 cm^{-1} (CH-ali..),1621 cm^{-1} (CONH amide).;^{1}H NMR (300 MHz, Chloroform)δ=9.59(2H,2NH-NHNH), 9.43(s,1H,CH-CHO),8.24(s,1H,OH-arm.),7.66-6.60(m,9H,CH-arm.),5.45(s,1H,NH), 4.78 (s,1H,CH-Phe), 3.57 (s,2H,CH_2-ali..), 3.16 (d,2H, CH_2-Phe); Anal./Calcd. for $C_{21}H_{19}N_3O_6$ (409): C (61.59%), H (4.64%), N (10.26%). Found: C (61.61); H (4.68); N, (10.26).

2.9. N-(1-(2-acetylhydrazinyl)-1-oxo-3-phenylpropan-2-yl)-2-(7-hydroxy-2-oxo-2H-chromen-5-yl)acetamide (11)

Compound (**9**, 0.01 mole) was refluxed with glacial acetic acid (20 ml.) for 3hrs., The solid obtained (**11**) after cooling was filtered and recrystallized from benzene to give compound (**11**) as brown crystals ,Yield: (70 %); Rf = 0.80 °C., (CH_2-Phe/MeOH=3/1); M.P.=213-15°C; IR (KBr cm^{-1}) ν; 3402 cm^{-1} (broad band, OH and NH), 2959cm^{-1} cm^{-1} (CH-ali..), 1714 cm^{-1} (C=O), 1617cm^{-1} (CONH amide);^{1}H NMR (300 MHz, Chloroform) δ= 10.59(s, 2H,NHNH), 9.36 (s, 1H, OH-arm.), 8.02-6.20 (m, 9H, CH-arm.) 5.43 (s, H, NH), 4.07 (1H,CH-L-Phe), 3.47 (s,2H,CH_2-ali..), 3.00(2H-CH2-L-Phe)2.06(s,3H,CH_3); MS (m/z, %) 423 (67.96); Anal./Calcd. for $C_{22}H_{21}N_3O_6$: C (62.41%),H(4.96%), N(9.92%). Found: C(62.41%), H (5.00%), N(9.93%).

2.10. N-(1-(5-(1-amino-2-phenylethyl)-1,3,4-oxadiazol-2-yl)-2-phenylethyl)-2-(7-hydroxy-2-oxo-2H chromen-5-yl)acetamide (12)

Compound (**9**; 0.01 mol), L-phe. (0.01 mol) were dissolved in concentrated sulfuric acid(10 ml); the reaction mixture was stirred for 6h. at 80°C. The reaction mixture was cooled, poured into crushed ice, and neutralized with sodium carbonate to PH=7. The crude product (**12**) was filtered, washed with water, dried and the grey crystal was recrystallized by ethanol to obtain the pure product (**14**), as white crystal, in Yield: (85 %); Rf=0.89; (CH_2-Phe/MeOH=3/1); M.P.=155-57°C; IR (KBr cm-1) ν; 3408 cm^{-1} (broad band, OH ,NH and NH_2), 2958,2927cm^{-1} (CH-ali.), 1714, cm^{-1} (C=O), 1620 cm^{-1} (CONH amide); MS (m/z, %) 509 (019%), Anal./Calcd. for $C_{29}H_{26}N_4O_5$: C(68.22 %), H (5.09 %), N (10.98 %). Found: C (68.22); H (5.13); N, (10.97).

2.11. N-(1-(2-carbamothioylhydrazinyl)1-oxo-3-phenylpropan-2-yl)-2-(7-hydroxy-2-oxo-2H- chromen-4-yl)acetamide (13)

The hydrazide derivative (**9**; 0.01 mol.) was reacted with ammonium thiocyanate (0.01mol) in concentrated hydrochloric acid (20 ml). The reaction mixture was concentrated, and cool. The separated solid (**13**) was filtered, washed with water, and recrystallized from ethanol to give compound (**13**) as brown crystals; Yield: 80%; Rf = 0.83(CH2-Phe/MeOH=3/1); M.P.=165-67°C, ; IR (KBr $_{cm-1}$) ν; 3405$_{cm-1}$ (broad band, OH ,NH and NH2) ,2960,2930 cm^{-1} (CH-ali.), 1698 cm^{-1}(C=O), 1622 cm^{-1}(CONH amide) 1239 cm^{-1} (C=S); ^{1}H NMR (300 MHz, Chloroform) δ= 9.01(s,1H,OH), 8.95(s,2H,NH2),7.43-6.33 (m,11H,(2H,NH-NHNH)+(9H,CH-arm.),5.63(s,1H,NH), 4.97(m,1H,CH-Phe), 3.19(s,2H, CH_2-ali..) , 2.90 (d,2H,CH_2-Phe); Anal./Calcd. for $C_{21}H_{20}N_4O_5S$(440): C(57.25 %), H(4.54%),N(12.72%). Found: C (57.26); H (4.58); N, (12.72).

2.12. 2(7-hydroxy-2-oxo-2H-chromen-4-yl)-N-(2-phenyl-1-(5-thioxo-4,5-dihydro-1,3,4-oxadiazol-2-yl)-

ethyl)acetamide(15)

Dissolve compounds (**9**; 0.01mol) in alcoholic KOH, carbon disulfide (0.01 mol) was added to the reaction mixture. The reaction mixture was removed under reduced pressure; the residual salt was treated with water. The filtrate was neutralized to pH= 6 using dil. HCl, the residual material (**15**) was filtered, washed with water, dried, crystallized by ethanol as brown crystal; in Yield 80 %; Rf = 0.88 (CH2-Phe/MeOH=3/1); M.P.=218-20°C; IR (KBr cm^{-1}) ν; 3412 cm^{-1}(broad band, OH and NH) ,2960_{cm-1} (CH-ali.), 1620 cm^{-1}(CONH amide), 1239 cm^{-1} (C=S); ^{1}H NMR (300 MHz,Chloroform) δ= 8.62 (s,1H,OH), 7.66-6.47 (m,10H(1H,NH-oxadiazolthione) + (9H,CH-arm.), 5.47 (s,1H,NH),4.83 (m,1H,CH-Phe),3.60 (s,2H,CH_2-ali.), 3.20 (d, 2H,CH_2-Phe) ; Anal./Calcd. for C21H17N3O5S (423): C(59.56%), H(4.01%), N(10.63%). Found: C (59.57); H (4.05); N, (9.92).

2.13. General procedures for synthesis N-(1-(2-aryylidenehydrazinyl)-1-oxo-3-phenyl -propan-2-yl)-2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetamide (17-c)

The compound (**9**, 0.01mol) in glacial acetic acid (10 ml.) was heated with aromatic aldehydes (0.01 mole), the reaction mixtures were heated under reflux for 6-8 hrs. The solid products (**17a-c**) obtained after cooling, collected by filtration and recrystallized from ethanol to give compounds (**17a-c**),

2.13.1. N-(1-(2-(2,4-dihydroxybenzylidene)hydrazinyl)-1-oxo-3-phenyl-propan-2-yl)-2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetamide (17a)

Brownish red crystal; Yield 90 %; Rf = 0.85(CH2-Phe/MeOH=3/1); M.P.=85-87°C; IR (KBr $_{cm-1}$) ν; 4434 cm^{-1}(broad band, OH), 3353,3270 cm^{-1}(NH) , 2921 $_{cm-1}$ (CH-ali.), 1657 cm^{-1}(C=O), 1597 cm^{-1}(CONH amide); ^{1}H NMR (300 MHz, Chloroform) δ= 10.11-10.3 (t,3H,OH), 8.10 (s,1H,CH-N CH), 7.44-6.64 (m,13H,CH-arm.), 6.41(1H,NH-NHNCHR),5.44(s,1H,NH-arm.), 4.85 (m,1H,CH-Phe), 3.07 (s,2H,CH_2-ali.), 2.90 (d,2H,CH_2-Phe); Anal./Calcd. for $C_{27}H_{23}N_3O_7$(501) : C (64.67 %), H (4.59%), N(8.38%). Found: C(64.67); H (4.62); N, (8.38).

2.13.2. N-(1-(2-(4-(dimethylamino)benzylidene)hydrazinyl)-1-oxo-3-phenyl-propan-2-yl)-2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetamide (17b)

Brown crystal; Yield 73 %; Rf = 0.90 (CH_2-Phe/MeOH=3/1); M.P.=170-72°C; IR (KBr cm^{-1}) ν; 3385 cm^{-1}(broad band, OH, NH), 2931cm^{-1}(CH-ali.), 1662 cm^{-1}(C=O), 1601cm^{-1}(CONH amide) ^{1}H NMR (300 MHz, Chloroform) δ= 10.82 (s,1H,OH), 8.19 (s,1H,CH-NCH),7.53-6.71(m,14H,CH-arm.),6.38(s,1H,NH-NHNCH), 5.57(s, 1H,NH-arm.), 4.56 (s,2H,CH_2.Phe.), 3.35 (s,2H,CH2-ali.), 3.00(s,6H,2CH_3-N(CH_3)2), 2.84 (2H,CH_2-Phe); Anal./Calcd. for $C_{29}H_{28}N_4O_5$ (512): C (67.69 %), H (5.46%), N (10.93%). Found: C (67.96); H (5.51); N, (10.93)

2.13.3. 2-(7-hydroxy-2-oxo-2H-chromen-4-yl)-N-(1-(2-(4-methox-ybenzyl-idene)hydrazinyl)-1-oxo-3-phenylpropan-2-yl) acetamide (17c)

Brown crystal; Yield 65 %; Rf = 0.87 (CH_2-Phe/MeOH=3/1); M.P.=210-12°C; IR (KBr cm^{-1}) ν; 3391 cm^{-1}

^{1}(broad band,OH ,NH) , 2957,2926 cm^{-1}(CH-ali.), 1698 cm^{-1}(C=O), ,1620 cm^{-1}(CONH amide);),; ^{1}H NMR (300 MHz, Chloroform); δ= 10.57 (s,1H,OH), 8.32 (s,1H,CH-N-CH), 7.60-6.65 (m,14H,CH-arm.), 5.86 (1H,NH-NHNCHR),5.48(s,1H,NH-arm.),5.06 (s,2H,CH_2. Phe.), 3.78(s,3H,CH_3-OCH_3), 3.19 (s,2H,CH_2-ali.), 3.17 (m,1H, CH-Phe), 2.96 (d,2H,CH_2- Phe) MS (m/z, %) m/z 499 (46.51%) ,Anal./Calcd.for $C_{28}H_{25}N_3O_6$: C (67.33%), H (5.01%), N (8.41%). Found: C (67.33); H (5.04); N, (8.40).

2.14. General procedures for synthesis of 4-(2-((1-(4-acetyl-5-aryl-4,5-dihydro-1,3,4-oxadiazol-2-yl)-2-phenylethyl)amino)-2-oxoethyl)-2-oxo-2H-chromen-7-ylacetate (18a-c):

The compounds (**17a-c**; 0.01 mol) were heated under reflux with acetic anhydride (15ml) for 10 hrs. The excess of acetic anhydride was decomposed with water (10ml), the reaction mixture was stirred for 30 mins. The separated solids were filtered, washed with water, dried and recrystallized from ethanol to give compounds (**18 a-c**), respectively.

2.14.1. 4-(2-((1-(4-acetyl-5-(2,4-dihydroxyphenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-2-phenylethyl)amino)-2-oxoethyl)-2-oxo-2H-chromen-7-yl acetate (18a)

Brown crystal; Yield: 85 %; Rf = 0.90 (CH_2-Phe/MeOH=3/1); M.P.=221-23°C; IR (KBr cm^{-1}) ν; 3411cm^{-1}(NH) ,2959,2929 $_{cm-1}$,(CH-ali.), 1621 cm^{-1}(CONH amide) and; ^{1}H NMR (300 MHz, Chloroform) δ= 8.03-6.54 (m, 14H, (1H,NH-arm.) +(13H,CH-arm.), 4.38(m,1H,CH-Phe), 3.02(s,2H,CH2.ali.), 2.82 (m, 8H,(6H,2(CH_3)-$N(CH_3)_2$) + (2H,CH_2-Phe), 2.25 ,2.06 (s,6H,$_2CH_3$-$COCH_3$);Anal./Calcd. for $C_{31}H_{27}N_3O9$(585): C (63.58%), H (4.61%), N (7.17%). Found: C (63.55); H (4.61); N, (7.18).

2.14.2. 4-(2-(1-(4-acetyl-5-(4-methoxyphenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-2-phenylethylamino)-2-oxoethyl)-2-oxo-2H-chromen-7-yl acetate(18b)

Brown crystal; Yield: 80 %; Rf = 0.86 (CH2-Phe/MeOH=3/1); M.P.=90-92°C; IR (KBr cm^{-1}) ν; 3198 cm^{-1}(NH) , 2927cm^{-1}(CH-ali.), 1764,1684 cm^{-1}(C=O), 1604 cm^{-1}(CONH amide); MS (m/z, %) 596 (12.19%),Anal./Calcd. for $C_{33}H_{32}N_4O_7$: C (66.44%), H (9.39%). Found: C (66.43); H (5.41); N, (9.36).

2.14.3. 4-(2-(1-(4-acetyl-5-(4-chlorophenyl)-4,5-dihydro-1,3,4-oxadiazol-2-yl)-2-phenylethylamino)-2-oxoethyl)-2-oxo-2H-chromen-7-ylacetate (18c)

Brown crystal; Yield: 76 %; Rf = 0.89 (CH_2-Phe/MeOH=3/1); M.P.=220-22°C; IR (KBr cm^{-1}) ν; 3220 cm^{-1}(NH) , 2961cm^{-1}(CH-ali.), 1624 cm^{-1}(CONH amide); MS (m/z, %) 583 (0.42%) Anal./Calcd. for $C_{32}H_{29}N_3O_8$: C (65.86%), H(4.97%), N (7.20). Found: C (65.86); H (5.01); N, (7.21).

2.15. 4-(2-((1-(2-acetylhydrazinyl)-1-oxo-3-phenylpropan-2-yl)amino)-2-oxoethyl)-2-oxo-2H-chromen-7-yl acetate (19)

Compound (**9**; 0.01mol) was heated with acetic anhydride (5 ml) for 3 hrs., the catalytic amount of AcONa was added. The solid obtained (**19**) after cooling, filtered, washed with pet. ether (60/80°C) and recrystallized from

abs. ethanol to give compound (**19**) , as brown crystals; Yield: 65 %; Rf =0.80(CH_2-Phe/MeOH=3/1); M.P.=185-87°C; IR (KBr cm^{-1}) ν 3323 cm^{-1}(NH), 2961cm^{-1} (CH-ali.), 1767 cm^{-1} (C=O),1619cm^{-1} (CONH amide; ^{1}H NMR (300 MHz, Chloroform) δ= 9.24,8.32 (s,2H,NH-NHNH), 7.62-6.55 (m,9H,CH-arm.), 6.54 (s,1H,NH-arm.), 4.81 (m,1H,CH-phe.), 3.23 (s,2H,CH2-ali..), 2.84 (d, 2H,CH_2-Phe), 2.27,1.99(s,6H,2CH_3-$COCH_3$); MS (m/z,%) 465 (1.61%) Anal./Calcd.for $C_{24}H_{23}N_3O_7$:C (61.91%), H (4.94%), N (9.02%). Found: C (61.93); H (4.98) ; N, (9.03).

2.16. 2-(3-(2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetyl)thioureido)-3-phenyl-propanoic acid (20)

L-phe. (0.01) was stirred with compound (**5**) for 6hrs. Then the reaction mixture poured onto ice path. The solid obtained was recrystallized from ethanol to give compound (**20**) as white crystal; Yield:(85 %); Rf = 0.86 (CH_2-Phe/MeOH=3/1); M.P.=238-40°C.; IR (KBr cm^{-1}) ν; 3404cm^{-1}(broad band, OH ,NH) ,2960cm^{-1}(CH-ali.), 1698 cm^{-1}(C=O), 1620 cm^{-1}(CONH amide), 1238 cm^{-1}(C=S).; ^{1}H NMR (300 MHz, Chloroform) 10.28(1H, CONHCS), 10.12 (s,1H,OH-COOH),9.41 (s,1H,OHphenyl), 7.47-6.27 (m,9H,CH-arm.), 5.47 (s,1H,NH-CSNH), 4.70 (m,1H,CH-Phe), 3.03 (s , 2H,CH2-ali.), 2.80 (d, 2H,CH_2- Phe); MS (m/z, %) 427 M+1 (6.81%),Anal./Calcd. for $C_{21}H_{18}N_2O_6S$: C(59.14%), H(4.22%), N(6.57%). Found: C (59.15); H (4.25); N, (6.57).

2.17. Methyl2-(3-(2-(7-hydroxy-2-oxo-2H-chromen-4-yl)acetyl)thioureido)-3-phenylpropanoate (21)

The acid (**20**, 3.98 g., 0.01 mol.) was allowed to react with thionyl chloride (0.01 mol) in presence of methanol, using the technique described in the preparation of compounds (**8**) to give compound (**21**) which was recrystallized from ethanol, as brown crystal; Yield: 3.50 g (85 %); Rf = 0.89 (CH_2-Phe/MeOH=3/1);M.P.=195-97°C; ν; 3428cm^{-1}(broad band, OH and NH), 2963cm^{-1}(CH-ali.), 1622cm^{-1} (CONH amide); ^{1}H NMR (300 MHz, Chloroform) 10.34(s,1H,CONHCS), 9.45 (s,1H,OHPhenyl), 7.99-6.11 (m,9H,Ar-H.), 5.40(s,1H,NHCS), 3.92(s,3H, CH_3,OCH_3), 3.16 (s,2H, CH_2-ali.); MS(m/z,%) 440 (16.18%) Anal./Calcd. for $C_{22}H_{20}N_2O_6S$: C(60.00%), H(4.54%), N (6.36%). Found: C (59.99); H(4.58); N(6.36).

2.18. N-(1-hydrazinyl-1-oxo-3-phenylpropan-2-ylcarbamothioyl)-2-(7-hydroxy-2-oxo-2H-chromen-4-yl) acetamide (22)

The compound (**21** ; 0.01 mol.) was reacted with alcoholic hydrazine hydrate (0.01 mol), the remaining procedures as which described in the preparation of the compound (**9**) which was recrystallized from ethanol, as brown crystal; Yield: (89 %); Rf = 0. 94 (CH_2-Phe/MeOH=3/1); M.P= 290-92°C; IR (KBr cm^{-1}ν; 3408cm^{-1}(broad band, OH, NH and NH_2), 2960cm^{-1}(CH-ali.), 1622$cm^{-1}$$cm^{-1}$ (CONH amide), 1239 cm^{-1}(C=S); ^{1}H NMR (300 MHz, Chloroform) 11.08(s,1H,NH,CONHCS), 9.82(s,1H.OH), 7.24-6.78 (m,10H,(1H,NH-NHNH2) + (9H,CH-arm.), 5.59 (s,H, NH-CSNH),5.69(s,2H,NH2), 3.97(s,1H,CH-Phe), 3.07(2H,CH2-ali.), 2.79 (2H,CH2-Phe); MS (m/z, %) 440 (0.07%) Anal. /Calcd. for C21H20N4O5S: C (57.25%), H(4.54%),N (12.72%). Found: C (57.26); H (4.58); N, (12.72).

2.19. Pharmacological activity

Swiss albino mice were divided into 26 groups (n=5). All the groups were injected with Ehrlich Ascites Carcinoma (EAC) cells (0.2 ml of 2×106 cells/mouse) intraperitoneally and intramuscularly in the thigh of each recipient mouse except the normal group, This was taken as day zero, the normal saline and tween (0.2 ml /mouse/day) were administered to normal and EAC control groups, the compounds (**7,8, 11,13, and 20**) and standard drug Dose (50 µg/kg/day) were administered in groups (4,5) respectively for 9 days intraperitoneally, after the administration of last dose followed by 24 hrs. Fasting 3 mice form each group was sacrificed for the study of antitumor activity.

2.20.1. Drug Preparation: Preparation and Administration of Doses

Solution of tested compounds (**7,8, 11,13, and 20**) were prepared in normal saline. The required volume was emulsified in distilled water by using Twin 80 (0.5% of the total volume). The emulsion was prepared in such a way that the required daily dose was contained in 0.2mL of the emulsion. 0.2mL of this emulsion was administered to each mouse intraperitoneally, daily from day 0 to 9.

2.20.2. Tumor Transplantation

Ehrlich's Ascites Carcinoma was maintained by serial transplantation from tumor bearing Swiss Albino mice. EAC cells were obtained from donor mice (Swiss albino) of 18–20 g body weight and suspended in sterile isotonic saline. A fixed number of viable cells usually (2×106 cells/20 g body weight) were injected intramuscularly in the thigh of each recipient mouse [31].

2.20.3. Body Weight

Animals were weighted on every other day throughout the period of the experiment.

2.20.4. Solid Tumor Volume and Tumor Growth Inhibition

Antitumor effects for the different treatments were evaluated by tumor growth inhibition. Tumors were measured individually using a caliper. Tumor volume was determined by the following equation [32]:

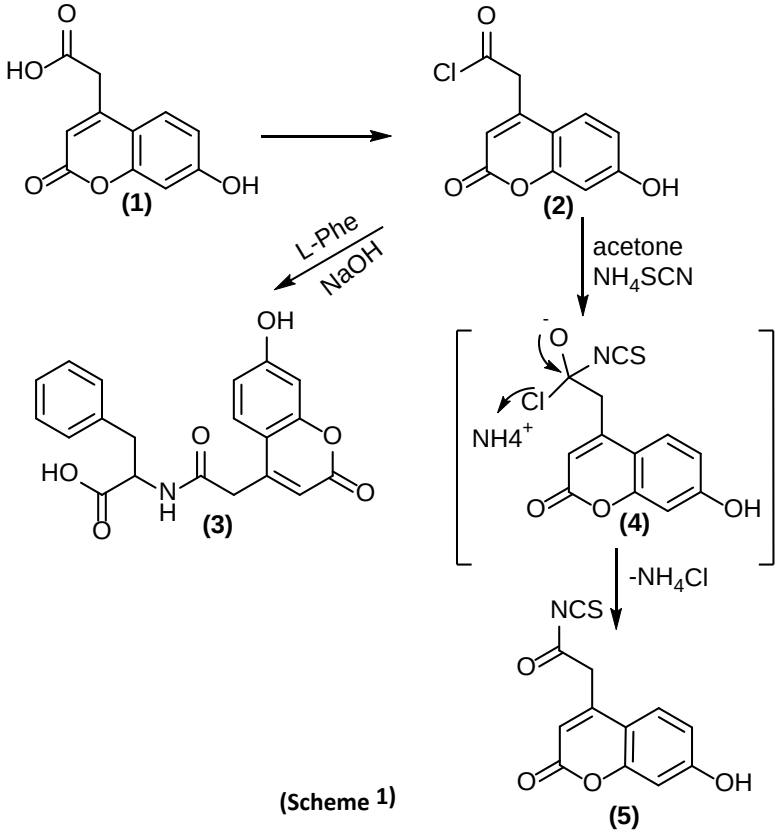
$$Tumor\ Volume = (Width^2 \times length)/2$$

The percent tumor growth inhibition was calculated on day13 by comparing the average values of treated groupsthat of tumor bearing control group. Tumor growth in saline. Treated control animals was taken to be 100% [33].

3. Result Discussion

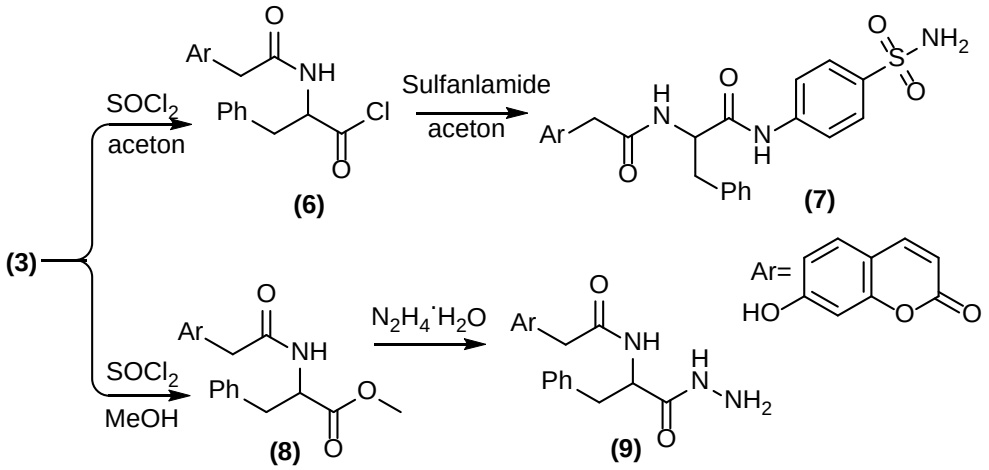
3.1. Chemistry

The synthetic routes to obtain the target compounds **1-22** were depicted in (Schemes 1-6). The starting material **2** was prepared as mentioned by elhenawy et al [30] (Scheme 1), which reacted with L-phenyl alanine (L-Phe.), and L-Phe. derivative **3** was formed. The presence COOH proton (δH=10.40 ppm) for 1HNMR data, characterized the structure. The isothiocyanate derivative **5** was prepared by the reaction of acid chloride **2** with ammonium thiocyanate via intermediate **4** (**Scheme** 1).



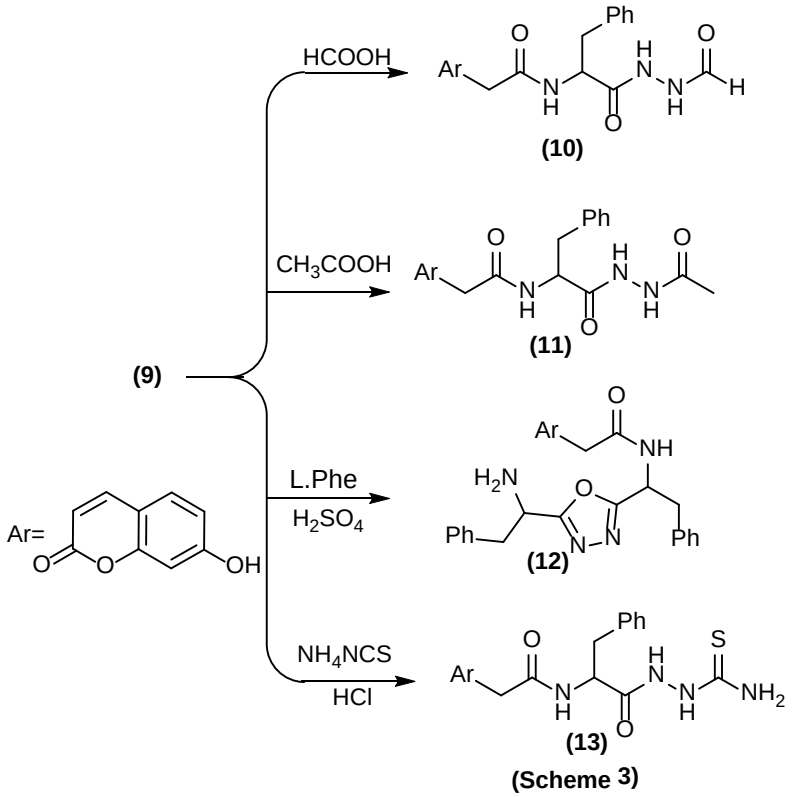
(Scheme 1)

The free acid derivative 3 undergoing SN^2 mechanism, and give the corresponding acid chloride 6. The sulphanilamide was reacted with compound **6**, to give the sulfanilamide derivative **7**, which supported by IR data, due to the appearance of characteristic sulfonamide bands at (1335,1230 cm^{-1}). Disappearance of characteristic peak (δH=10.40) for COOH proton of compound **3**, it is evidence for esterified the acid **3** to corresponding methyl ester derivative **8,** and was hydrazonlized to corresponding hydrazide derivative **9** (Scheme 2).



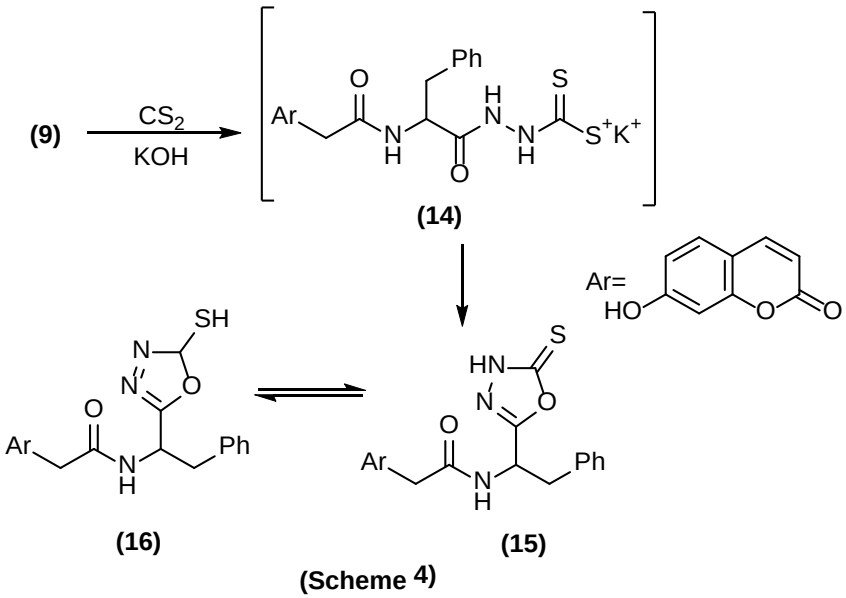
(Scheme **2)**

The compound **10** was obtained by formylation of hydrazide **9**, which acylated with acetic anhydride, which gave 2-(7-hydroxy-2-oxo-2H-chromen-4-yl)-N-(1-(5-methyl-1,3,4-oxadiazol-2-yl)-2-phenyl-ethyl)acetamide (**11)** in 70% yield. The compound **9** undergoing cyclization reaction with L-Phe in presence Conc. H_2SO_4, and gaveN-(4-(5-(1-amino-2-phenyl-ethyl)-1,3,4-oxadiazol-2-yl)phenyl)-2-(7-hydroxy-2-oxo-2H-chromen-4-yl) acetamide (**12**). The compound **9** was reacted with NH_4SCN in catalytic amount of HCl to give thiourea derivative **13** in 80% yield (Scheme 3).



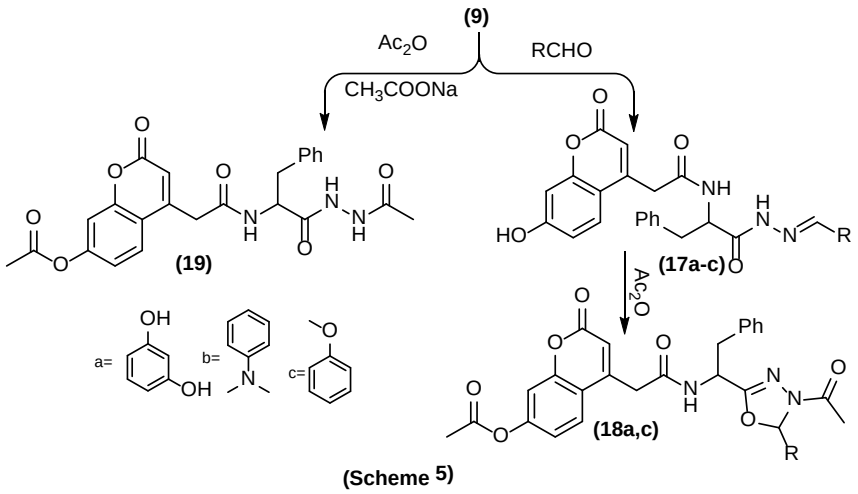
(Scheme 3)

The CS_2 was reacted with compound **9**, and led to formation thion **15** or thiol **16** derivatives via salt formation intermediate **14**, the disappearance peak of thiol group in spectral data led to preferred the formation of thion derivative **15** in 75 % yield (Scheme 4).



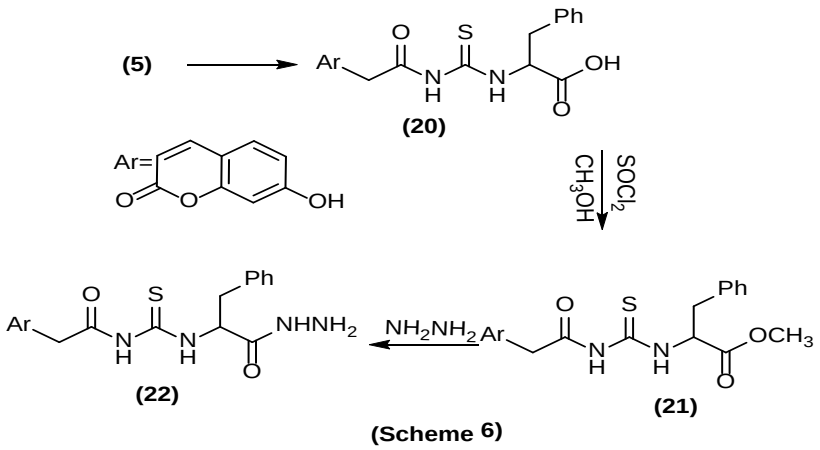
(Scheme 4)

The compound **9** was condensed with different aromatic aldehydes, and afforded the corresponding Schiff¨s bases **17a-c**, which undergoing cyclization reaction with acetic anhydride to led formation oxadizole derivatives **18a-c,**). The disappearance of CH═N and OH phenyl peaks of ^{1}HNMR spectrum for Schiff¨s bases **17a-c**, confirmed its structures**18a-c** (Scheme5). The 4-(2-(1-(2-acetyl-hydrazinyl)-1-oxo-3-phenylpropan-2-ylamino)-2-oxoethyl)-2-oxo-2H-chromen-7-ylacetate (**19**) was obtained by acylated with acetic anhydride with fused sodium acetate, which confirmed with disappearance OH aromatic proton (δH=8.74) for hydrazide **9** (Scheme 5).



(Scheme 5)

Thiourido derivative **20** was prepared by coupling of compound **5** with L-Phe, the Thiourido free acid **20** was methylated with thionyl chloride via formation acid chloride intermediate to give methyl ester derivative **21**, the disappearance of peak (δH=10.12) of COOH proton for acid **20**, and appearance of 3H proton Of OCH_3 at (δH=3.92), which confirmed the proposed molecular structure **21**. The methyl ester derivative **21** was hydrazonolysis and exhibited hydrazide derivative **22** in good yield 89%(Scheme 6).



(Scheme 6)

3.2. Pharmacology

3.2.1. Ehrlich Tumor (Solid)

The antitumor activity of tested compounds (**7,8, 11,13,** and **20**) were assayed by observation of various parameters like Body weight of animals, tumor volume, inhibition tumor growth [35,36] **.**

3.2.2. The body weights

The average weight loss of tested compounds (**7,8, 11,13,** and **20**) were observed against untreated (table 1). The data showed that, the compounds (**20**) showed higher average weight loss (20.5±0.25) upon reference drug (DOX) mice group (21.85±0.25), the rest tested members have lower observed average weight change between untreated groups (table1).

Table 1: Average weights and tumor inhibition of ehrlich solid tumor after treatment compounds (**7,8, 11,13 and 20**).

Treatment	*Body weight of animal on days (g)*			*day 13*			
	day (0)	*Day 5*	*day 9*	*Avg. body wt.(g)*	*Avg. tumor volume*	*%Tumor growth inhibition*	*Mortality*
normal	20.2 ± 0.25^{e}	21.6 ± 0.25^{ed}	22.8 ± 0.25^{b}	23.2 ± 0.16^{a}	-	-	0/5
untreated	20.9 ± 0.25^{e}	21.4 ± 0.25^{e}	22.2 ± 0.25^{c}	23.0 ± 0.07^{ed}	1.8 ± 0.09^{a}	-	0/5
Dox	23 ± 0.25^{b}	22.4 ± 0.07^{c}	21.85 ± 0.25^{d}	20.4 ± 0.25^{f}	0.4 ± 0.09^{e}	79.04	0/5
7	21.9 ± 0.07^{d}	22.8 ± 0.12^{c}	23.2 ± 0.25^{b}	23.5 ± 0.16^{cd}	1.5 ± 0.09^{ab}	56.6	0/5
8	23.5 ± 0.25^{c}	23.8 ± 0.12^{c}	22.5 ± 0.25^{c}	-	1.6 ± 0.09^{d}	40.7	1/5
11	23.6 ± 0.25^{c}	23.0 ± 0.25^{b}	23.7 ± 0.25^{b}	25.7 ± 0.16^{c}	1.6 ± 0.09^{ab}	13.9	2/5
13	24.4 ± 0.25^{c}	23.5 ± 0.12^{b}	23.3 ± 0.25^{b}	24.6 ± 0.16^{b}	1.7 ± 0.09^{cd}	31.1	0/5
20	20.5 ± 0.25^{f}	21.2 ± 0.25^{e}	22.9 ± 0.07^{b}	20.5 ± 0.25^{f}	0.5 ± 0.09^{e}	70.5	0/5

Values are expressed as Mean ± SEM and, $p \leq 0.05$ indicates the level of statistical significance as compared with control. Treatments show highly significant deference at pr>0.0001, the letters (a-f) represents statistically significant

3.2.3. Solid Tumor Volume:

The average tumor volume calculated on day 13 of tested compounds (**7,8, 11,13,** and **20**) in compared with untreated control (table 1), and reference drug (Dox). From (table1), all members were exhibited decreasing average tumor volume in order **20<7<8<13<20** in range (~0.5-1.7).

3.2.1. Structure activity relationship

The antitumor activity of tested compounds (**7,8, 11,13,** and **20**) were examined in vitro of mice, using the following parameters, body weight of animals, tumor volume, and tumor growth inhibition. The above data showed that, the member containing thiourea *L-phenylalanine methyl ester* moiety **20** showed highest inhibition potency (70%), the compounds have sulfonamide **7** and amino acid methyl ester fragments **8** showed moderate inhibition potency (56.6 and 40 %), respectively, the other members showed lower inhibition potency, (table1).

3.3. Molecular Modeling studies

3.3.1. Conformational analysis

In trying to achieve better insight of the molecular structure, due to presence chiral center for the synthesized compounds (**3-13,15,17a,18a,19-22**), which led to existence optical isomerism's for its compounds (**3-13,15,17a,18a,19-22**). The optimization geometry was performed in vacuo with PM3 semi-empirical *Hamiltonian* molecular orbital calculation MOPAC7 package [34], the conformational analysis of the target compounds have been performed to prefer most stable form of stereoisomers using the PM3 (calculations in vacuo, bond dipole option for electrostatics, PolakeRibiere algorithm, RMS gradient of 0.01 kcal/ mol) for all synthesized compounds (**3-13,15,17a,18a,19-22**, (table 2). In addition, the computed molecular parameters, total energy, electronic energy, heat of formation, the highest occupied molecular orbital (HOMO) energies, the lowest unoccupied molecular orbital (LUMO) energies and the dipole moment for studied compounds were calculated (table 2). The calculated molecular parameters (table 2) showed, *(L)* forms have most stable structures (**3-13,15,17a,18a, 19-22**) which may be explained by slightly reducing calculated energy for (L) forms over (D) forms for (**3-13,15,17a,18a,19-22**). The benzocoumarine rings arranged in or coplanar modes with L-Phe moieties (**3-13,15,17a,18a,19-22**), (table2).

3.3.2. Docking studies

COX protein are known with two isoforms: COX-1 and COX-2. The COX-1 produced prostaglandins, and expressed in most tissues; and COX-2, is responsible for the increasing production of prostaglandins during process of inflammation, which is induced by endotoxins, cytokines and mitogens in inflammatory cells [35]. Recently, analysis the X-ray of COX-2 active site showed that, the negative charge of the tetrahedral intermediate was stabilized through coordinated COOH-arachidonic with Tyr385and Ser-530, as well as the action of NSAIDs[36]. So, Tyr-385 and Ser-530 have importance structural and functional site for chelating ligand [37]. Molecular docking of the synthesized compounds **7,8,11,13** and **20** into the active site of COX-2 performed, in order to discover biological data on a structural basis, through rationalized ligand–protein

interaction behavior. All calculations for docking experiment preformed with MOE 2015.10 [38]. The tested compounds were evaluated in silico (docking), using X-ray crystal structures and COX2 (ID: 1PXX) [34] complexes with reference inhibitor.

Table 2: The optimized calculations energies for most stable steroisomer at PM3 molecular orbital for (**3-13,15, 17a,18a,19-22**):

Cpd	E	HF	IP	HOMO	LUMO	μ
3	-134034.73	-168.80	8.96	-8.96	-1.09	5.56
6	-130228.95	-121.66	9.16	-9.16	-0.87	4.52
7	-137635.72	-183.02	9.07	-9.03	-1.21	4.07
8	-144153.28	-157.35	9.65	-9.65	-1.30	9.07
9	-151554.33	-209.68	9.18	-9.18	-1.13	7.76
10	-123068.74	-44.05	9.20	-9.20	-1.22	1.81
11	-130459.84	-86.10	9.19	-9.20	-1.02	6.93
12	-128821.32	-33.25	8.95	-8.93	-1.10	8.59
13	-136217.69	-118.38	9.20	-9.20	-1.09	3.04
15	-149838.89	1.78	9.21	-9.21	-1.05	4.85
17a	-164621.2	-115.77	8.98	-8.98	-1.01	4.60
18a	-150486.83	-49.54	8.18	-8.17	-1.20	2.07
19	-149196.3	-90.62	8.78	-8.78	-1.05	7.28
20	-146045.95	-41.43	8.85	-8.85	-0.81	8.62
21	-177024.5	-134.22	9.29	-9.29	-1.32	4.16
22	-174357.33	-106.50	9.36	-9.36	-1.08	5.13

E: Total energy (kcal/mol)., *HF*: Heat of formation (kcal/mol),*IP*: Ionization potential energy(kcal/mol), *HOMO*: Highest Occupied Molecular Orbital(eV), *LUMO*: Lowest Occupied Molecular Orbital(eV), μ*: Dipole moment*(*Deby)*.

The tested compounds docked into COX-2active site. The active site of the enzyme was defined to include residues within a 10.0 Å radius to any of the inhibitor atoms. MOE scoring function of the most stable docking model for tested compounds applied to evaluate the binding affinities between the inhibitors with (COX) active site, (Table 3). The minimization energy preformed for complexes (inhibitor-active site) with anMMFF94 force field [39] until the gradient convergence reached to 0.05 kcal/mol. The most active compound **20** docked successfully into the COX-2 active site, and compared with reference inhibitor (diclofenac). the compound **20** was exhibited highest binding score -15.13 Kcal/mol. (table 3).

3.3.3. Quantum Structures activity relationships

In order to get a deeper insight into the nature and type of interactions of docked compounds **7,8,11,13** and **20,** the complexes between each compound and COX-2 receptor were visualized. The H bond interactions playing an important role in the structure and function of biological molecules. The current ligand-receptor

interactions were analyzed on the basis of H bonding. In order to reduce the complexity, hydrophobic and π-cation interactions (>6Å) are not shown in (Figure 1).

Table 3: Docking energy scores (kcal/mol) derived from the MOE for new isolated ligands (**7,8,11,13,20**).

Cpd.	dG	Int.	H.B.	Eele	Evdw
7	-14.58	34.92	-93.88	-3.59	1.27
8	-13.53	84.77	-51.65	-4.10	1.73
9	-14.52	-14.36	-50.41	1.74	1.05
11	-14.81	85.84	-87.83	-4.38	0.72
13	-14.13	68.04	-102.71	-3.87	1.56
15	-12.16	62.39	-44.47	-2.3	3.44
20	-15.13	80.23	-69.3	-1.45	0.97

d.G.: free binding energy of the ligand from a given conformer, Int.: affinity binding energy of hydrogen bond interaction with receptor, H.B.: Hydrogen bonding energy between protein and ligand.Eele: the electrostatic interaction with the receptor, Evdw: van der Waals energies between the ligand and the receptor.

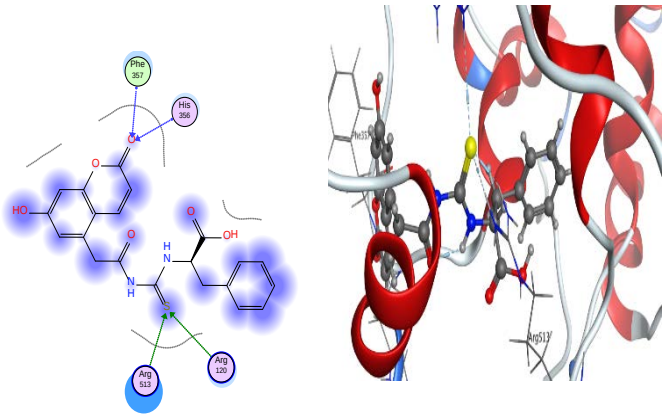


Figure1: The most active compound 20 were Docked into the active site of COX-2, using MOE tool, H- bonds are in blue.

The highest binding score member is **20,** exhibited important two hydrogen interactions with binding site through L-phe.-OMe fragment, and stabilized in binding pocket by adjusting its phenyl ring of L-Phe and coumarin ring in Parallel mode with Tyr-385. The compound 20 was stabilized with itself by arranged of phenyl ring with coumarin ring in coplanar position, (Figure.1). Furthermore, The results obtained clearly revealed that, the amino acid residues close to the reference molecule are mostly the same as observed in the tested compounds (Figure 1). The higher binding process interaction observed in **20** with COX-2, indicated that, the compounds **20** act as selective inhibitors against COX-2, this may be explained, the presence of hydrophobic amino acid in the synthesized compounds.

3.3.4. ADMET Profile

Oral bioavailability was considered to play an important role in the development of bioactive molecules as therapeutic agents. Many potential therapeutic agents fail to reach the clinic, because of their ADMET Factors. Therefore, a computational study for prediction of ADMET properties of the molecules (**3-13,15,17a,18a,19-22**) were performed, by the determination of topological polar surface area (TPSA), a calculated percent absorption (%ABS)[40], and Lipinski rules [41]. In addition, the total polar surface area (TPSA) is another key property linked to drug bioavailability, the passively absorbed molecules with (TPSA>140) have low oral bioavailability [42]. All calculated descriptors were performed using MOE Package [38], and their results were disclosed in (Table 4). Our results revealed that, the CLogP (factor of the lipophilicity [43] were less than 5.0, hydrogen bond acceptors between (7-10), hydrogen bond donors between (1-5), these data showed these compounds fulfill Lipinski's rule. Also, the absorption percent in ranged between (~ 50-86%). The HOMO and LUMO of a molecule play important roles in intermolecular interactions [44], through the interaction between the HOMO of the drug with the LUMO of the receptor and vise versa. The interactions were stabilized inversely with energy gap between the interacting orbitals. Increasing HOMO energy and decreasing LUMO energy in the drug molecules led to enhancement stabilizing interactions, and hence, binding to the receptor.

Table 4: Pharmacokinetic parameters important for good oral bioavailability of compounds (**3-13,15, 17a,18a,19-22**):

CPD	HBD	HBA	LogP	V	TPSA	%ABS	Log S	ΔE
3	2	7	2.63	0	101.93	73.83	-4.67	8.39
6	3	10	1.88	0	139.89	60.73	-5.45	8.29
7	3	8	4.80	0	141.34	60.24	-6.64	7.50
8	4	9	1.64	0	133.83	62.83	-4.74	8.32
9	3	10	1.88	0	139.89	60.73	-5.45	8.29
10	5	10	2.07	0	164.89	52.11	-6.49	8.43
11	2	7	2.63	0	101.93	73.83	-4.67	8.39
12	5	8	1.22	0	130.75	63.89	-4.53	8.53
13	3	8	2.52	0	146.05	61.14	-6.40	8.25
15	5	10	3.90	0	157.55	86.73	-7.96	7.09
17a	3	10	2.05	0	139.90	52.48	-6.80	7.76
18a	4	10	1.75	1	160.13	55.12	-7.47	7.97
19	3	8	4.97	0	141.34	50.67	-7.88	7.09
20	3	8	2.52	0	146.05	69.14	-6.40	8.25
21	5	10	2.24	1	164.89	98.53	-6.30	8.53
22	4	9	4.93	1	137.32	61.85	-6.27	8.38

TPSA: Polar surface area (A°2),%ABS: Absorption percentage, Vol: Volume (A3), HBA: Number of hydrogen bond acceptor, HBD: Number of hydrogen bond donor, V: Number of violation from Lipinski's rule of five., Log P: Calculated lipophilicity., Log S: Solubility parameter, ΔE: Energy Gaps(eV).

3.3.5. Prediction of blood–brain barrier permeability

Blood–brain barrier (BBB) permeability, is a one of the most important challenges in the pharmacology of CNS active drugs. Many drugs have limited usage and fail to pass the clinical trials, due to failure penetration of CNS . In silico (Table 5), the pharmakinetic parameters were calculated for most active compound 20, using ADME-T algorthim, and defined human intestinal absorption (HIA) model [45,46], which predicted that, the compounds should be able to transported across the intestinal epithelium, which probably have high affinity binding to the plasma proteins, and may be passed through the blood-brain barrier, and it is necessary for ability drug transported throughout the body. In general, these data (table 5) suggested that, no marked health effects observed for rodent toxicity profiles, among the most active compound **20**, its compound is a good ability transport against *BBB*, good activity for CNS can be used as a good oral bioavailability.

Table 5: The prediction of blood brain barrier most active compounds **20**.

ADME-Tox	20
LogBBB (Blood-brain barrier.)	0.048
PPB% (Plasma protein binding)	94.36
LD50 rat/mouse(mg kg^{-1}, oral)	500/590
LD50 rat/mouse(mg kg^{-1}, intraperitoneal)	310/360
LD50 mouse(mg kg^{-1}, intravenous)	32
LD50 mouse(mg kg^{-1}, subcutaneous)	350
Ames test (genotoxicity, %)	0.32
Prob. of blood effect	0.42
Prob. of cardiovascular System	0.84
Prob. of gastrointestinal System	0.90
Prob. of kidney effect	0.82
Prob. of liver effect	0.41
Prob. of lung effect	0.52

4. Conclusion

The present work aims to synthesis some novel NSAIDs containing coumarin nucleus. The synthesized compounds were characterized by different spectral data (IR, ^{1}HNMR). The antitumor activity of tested compounds (**7,8, 11,13,19b and 20**) were examined in vitro against Ehrlich solid carcinoma (EAC) tumor model in Swiss albino mice on dose 50µg , using the following parameters, body weight of animals, tumor volume, and tumor growth inhibition. The data showed that, introducing thiourea free acid fragment **20** enhancements antitumor activity than reference drug (DOX). The optimization geometries for compounds containing L-Phe., showed that, the *L*- isomers are most stable form for isolated compounds **3-13,15,17a,18a,19-22**. The molecular docking simulations into the active site of COX-2 showed that, some

compounds (**7,8, 11,13, and 20**) suitable inhibitor against COX-2, and can used as New class of NSAIDs. The ADMET profiles in silico showed that, these compound are good oral bioavailability, and the most active compound **20** is CNS active agent without marked health effects observed for rodent toxicity. On the light of these data, we think that, this compound may be used new class NSAIDs as candidate anticancer drug with some modification of this structure.

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