Bull. Chem. Soc. Ethiop. **2024**, 38(3), 775-798. ISSN 1011-3924
© 2024 Chemical Society of Ethiopia and The Authors Printed in Ethiopia
DOI: https://dx.doi.org/10.4314/bcse.v38i3.18 Online ISSN 1726-801X

NEW APPROACHES FOR THE SYNTHESIS OF CHROMENE AND QUINOLINE DERIVATIVES AND THEIR ANTI-PROLIFERATIVE, MORPHOLOGICAL STUDIES

Rafat Milad Mohareb* and Hanan Maged Labib

Department of Chemistry, Faculty of Science, Cairo University, Giza, Egypt

(Received January 16, 2024; Revised February 5, 2024; Accepted February 6, 2024)

ABSTRACT. This work aimed to evaluate the anticancer potential of the novel 5,6,7,8-tetrahydro-4*H*-chromenes against selected six cancer cell lines together with the prostate cancer cell line PC-3. A novel series of substituted 5,6,7,8-tetrahydro-4*H*-chromenes were synthesized through feasible synthetic strategy. The synthetic schemes involve firstly the multi-component reactions of dimedone with the aromatic aldehydes and ethyl acetoacetate to produce the 5,6,7,8-tetrahydro-4*H*-chromenes derivatives. On the other hand, carrying the same reactions using NH_4OAc produced the hexahydroquinoline compounds. Anti-proliferative evaluations and inhibitions for all synthesized compounds toward selected cancer cell lines were carried out and the results revealed that many of them exhibited high inhibitions. Morphology of A549 cell line by the effect of compounds **14f** and **16c** was performed.

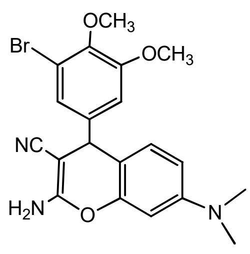
KEY WORDS: Anti-proliferative activity, Chromene derivatives, Morphology, Multi-component reactions

INTRODUCTION

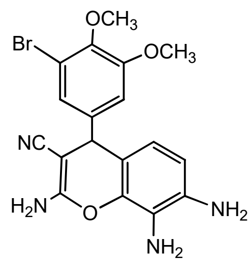
Chromenes are the most important compounds in the drug discovery and production which are bicyclic heterocyclic compounds produced by fusion of a benzene ring with a pyran (Figure 1) [1]. Such a group of compounds are fairly ubiquitous in nature, as these are found in bacteria, fungi, plants and animals [2-5]. In medicinal chemistry, the presence of the chromene moiety within the structure of the compound is responsible for its physiological activities, like antineoplastic, anticoagulant, antihypertensive, β-secretase inhibition, antidepressant, antitrypanosomal, anti-HIV, antidyslipidemic, and antimicrobial [6-8]. Moreover, the chromene derivatives are important class of compounds as anticancer agents, examples of drugs containing chromene moiety is the crolibulin EPC2407 (Figure 1) which is used as vascular disrupting anticancer drug to treat advanced solid tumors, beside it is currently under phase I/II clinical trials [9, 10]. Through Figure 1, the chemotherapeutic agent LY290181 (designed by number 18) which is a notable example of chromene that has been emerged as anticancer agent. Another important drug is the LY290191 which is used to exert its effects by inhibiting the mitosis and microtubules and it is considered as a potent anti-proliferative agent for a variety of cancer cell lines [11, 12]. Cancer is one of the more serious diseases causing death leading either to a solid mass of cells known as a tumor or to non-solid mass such as blood or bone marrow-related cancer in which the growth control is lost in one or more cells [13, 14]. It causes death throughout the world and its treatments involves surgery, chemotherapy, and/or radiotherapy [15, 16]. Despite all drugs available to treat cancer, statistical measurements exhibited that 10 million will die from it and more than18 million new cases appear yearly throughout the globe [17]. Doxorubicin is one of the most common drugs for cancer which is capable for the interaction with DNA causing suspension of cancer and it is known as a good anticancer agent [18-22]. On the other hand, distamycin also has anti-proliferative capability by inhibiting DNA-transcription factors which is considered as a minor groove binder and binding intercalators [23]. In recent years, benzothiazolyl-benz-α-chromene and 3,4-dihydropyrano[*c*]chromene were considered as DNA intercalation agents and as non-intercalating groove binders [24]. The apoptosis and

*Corresponding author. E-mail: raafat_mohareb@cu.edu.eg, raafat_mohareb@yahoo.com
This work is licensed under the Creative Commons Attribution 4.0 International License

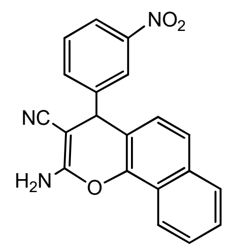
differentiation induced activities of coumarins were extended to several different cell line models *in vitro*, and they appear to be the most promising in terms of cancer treatment [25]. On the other hand, quinoline derivatives play an important role in exhibiting anticancer activity [26-28]. In the light of these facts, and as a continuation of our previous reported work [29, 30], we planned in this work to synthesize a novel series of coumarin analogs and quinoline derivatives through the one-pot multi-component reactions. Moreover, due to the important of fused chromene and quinoline derivatives we report the anti-proliferative activity of the synthesized fused chromene and quinoline compounds where many of the tested compounds exhibited high inhibitions. Such group of compounds were synthesized using dimedone which reacted with ethyl 3-oxobutanoate and aromatic aldehydes to produce 5,6,7,8-tetrahydro-4*H*-chromene-3-carboxylate and 1,4,5,6,7,8-hexahydroquinoline-3-carboxylate derivatives through the use of different catalytic conditions.



MX58151



EPC2407 (Carlobulin)



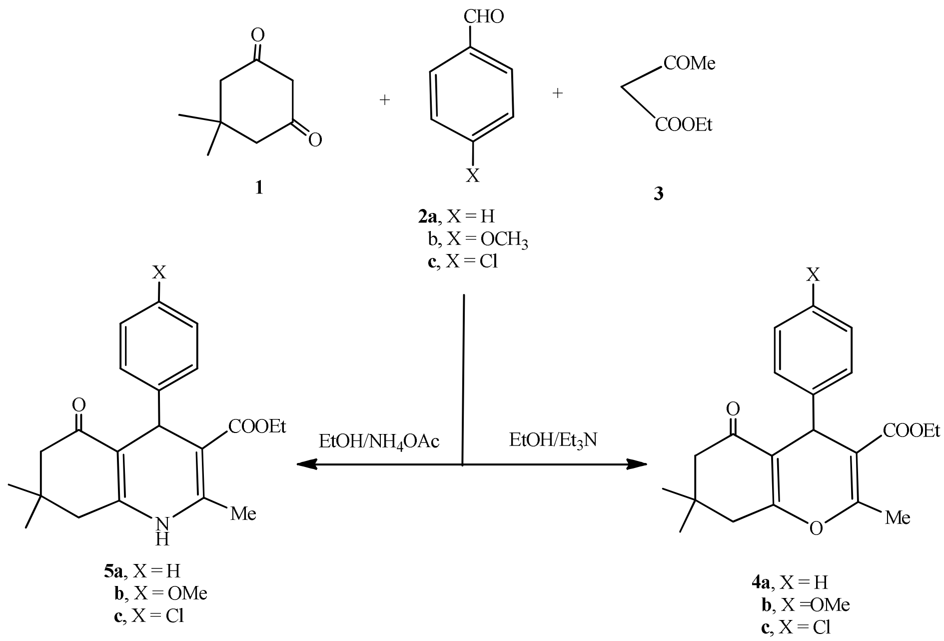
LY290181 (18)

Figure 1. Chemical structures of potential chemotherapeutic chromenes MX58151, EPC2407 (also named crolibulin) and LY290181 (designed by number 18).

RESULTS AND DISCUSSION

The title compounds were synthesized by one-pot multi-component synthetic procedure as shown in Schemes1-4. All the synthesized compounds were established by IR, ^{1}H NMR, ^{13}C NMR and mass spectral data. In the present work we are concerning with the multi-component of dimedone with ethyl 3-oxobutanoate and aromatic aldehydes to produce either 5,6,7,8-tetrahydro-4*H*-chromene-3-carboxylate or 1,4,5,6,7,8-hexahydroquinoline-3-carboxylate. Thus, the multi-component reactions of dimedone with aromatic aldehydes **2a-c** and ethyl 3-oxobutanoate (**3**) in absolute ethanol (50 mL) containing triethylamine gave the 5-oxo-4-aryl-5,6,7,8-tetrahydro-4*H*-chromene-3-carboxylate derivatives **4a-c** (Scheme 1). Their structures were based on their respective analytical and spectral data. Thus, ^{1}H NMR spectrum of **4a** showed (beside the expected signals) the presence of two singlets at δ 1.80 and 2.23 ppm corresponding to two methylene groups, a triplet and a quartet at δ 1.13, 4.23 ppm confirming the methyl and ethyl ester groups, a singlet at δ 6.02 ppm confirming the existence of the pyran *H*-4. Moreover, the ^{13}C NMR spectrum revealed signals at δ 120.3, 122.5, 123.8, 125.4 corresponding to the pyran C-2, C-3, C-5, C-6 and two signals at δ 165.8, 166.2 confirming the presence of two carbonyl groups.

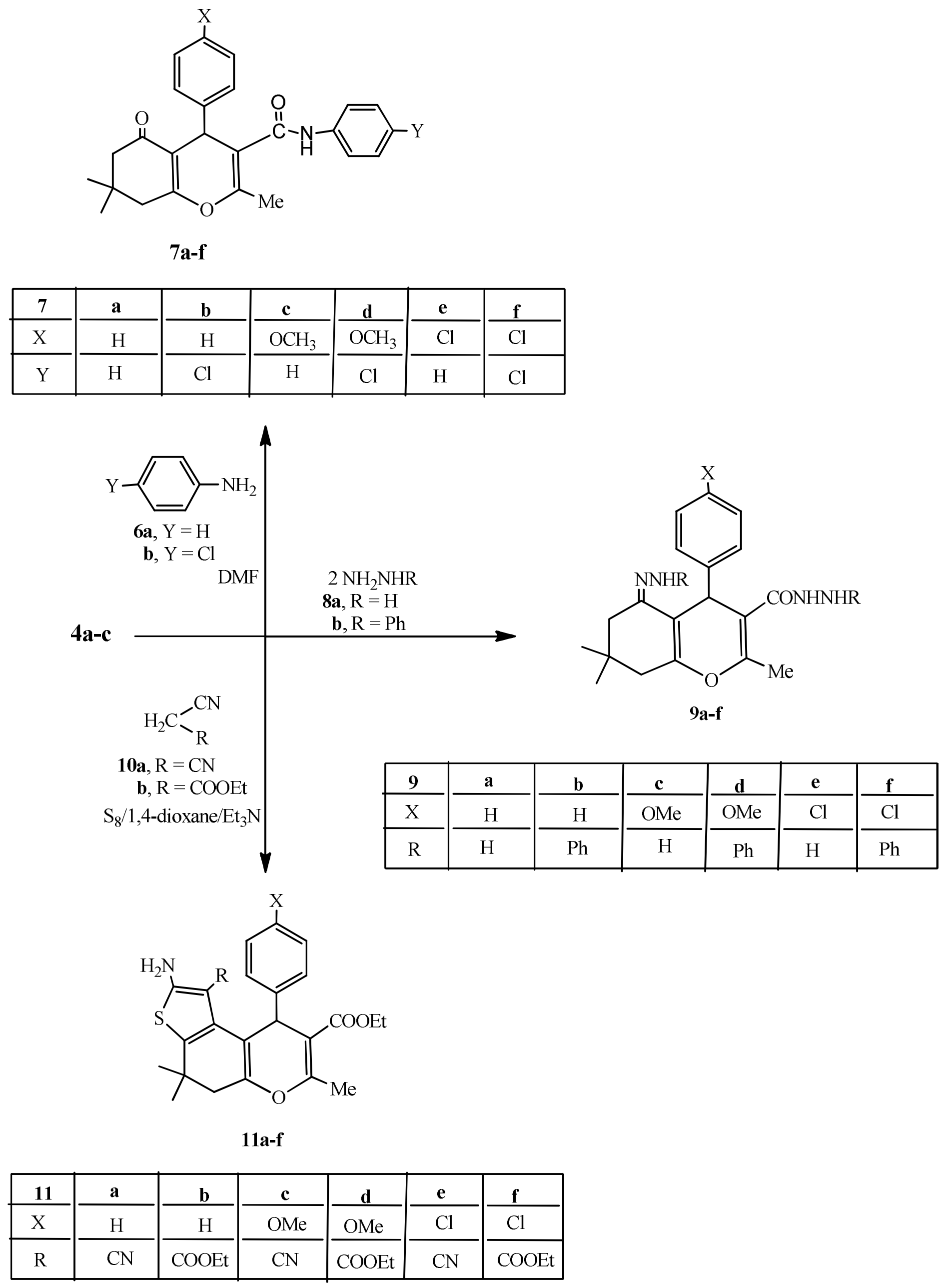
In addition, the multi-component reactions of dimedone with the aromatic aldehydes **2a-c** and ethyl 3-oxobutanoate (**3**) in absolute ethanol (50 mL) containing NH_4OAc as a catalyst gave the 1,4,5,6,7,8-hexahydroquinoline-3-carboxylate compounds **5a-c** (Scheme 1).



Scheme 1. synthesis of compounds **4a-c** and **5a-c**.

Compounds **4a-c** were ready for anilide formation through their reaction with either aminobenzene (**6a**) or 1-amino-4-chlorobenazene (**6b**) in dimethylformamide solution under the reflux conditions to produce the 5,6,7,8-tetrahydro-4*H*-chromene-3-carboxamide compounds **7a-f**, respectively. Their analytical and spectral data were in analogy with their respective structures (see experimental section). Moreover, the reaction of **4a-c** with two-fold of hydrazine hydrate (**8a**) or phenylhydrazine (**8b**) gave the 5-hydrazono-5,6,7,8-tetrahydro-4*H*-chromene-3-carbohydrazide compounds **9a-f**, respectively. Compounds **4a-c** were capable for thiophene formation through the Gewald's thiophene synthesis [31-33] due to the presence of the α-methinocarbonyl moiety. Thus, the reaction of compounds **4a-c** with elemental sulfur and either dicyanomethane (**10a**) or ethyl 2-cyanoacetate (**10b**) gave the 5,9-dihydro-4*H*-thieno[3,2-*f*]chromene-8-carboxylate **11a-f**, respectively (Scheme 2).

On the other hand, the reaction of either **4a-c** with elemental sulphur and phenylisothiocyanate (**12**) in *p*-dioxane containing a catalytic amount of triethylamine gave the chromeno[5,6-*d*]thiazole-8-carboxylate derivatives **13a-c**, respectively. Compounds **13a-c** reacted with either aminobenzene (**6a**) or 1-amino-4-chlorobenzene (**6b**) in dimethylformamide solution under the reflux conditions to produce the chromeno[5,6-*d*]thiazole-8-carboxamide derivatives **14a-f**, respectively. Moreover, the reaction of **13a-f** with 1,2-diaminobenzene (**15**) in dimethylformamide under the reflux conditions gave the chromeno[5,6-*d*]thiazole-2-thione derivatives **16a-c**, respectively (Scheme 3). The structures of the latter products were based on their respective analytical and spectral data. For example, the ^{1}H NMR spectrum of **16a** revealed the presence of a singlet at δ 2.21 corresponding to the CH_2 moiety, a multiplet at δ 7.25-7.48 ppm due to the presence of the three phenyl groups and a singlet at δ 8.36 ppm due to the NH group. In addition, the ^{13}C NMR spectrum showed a signal at δ 38.9 due to the methyl group, twelve signals at δ 120.2, 120.5, 121.4, 121.7, 121.9, 122.0, 122.6, 122.8, 123.3, 123.9, 124.3, 125.8 according to the three phenyl groups and two signals at δ 172.3 and 180.3 due to the presence of C=N and C=S groups, respectively.

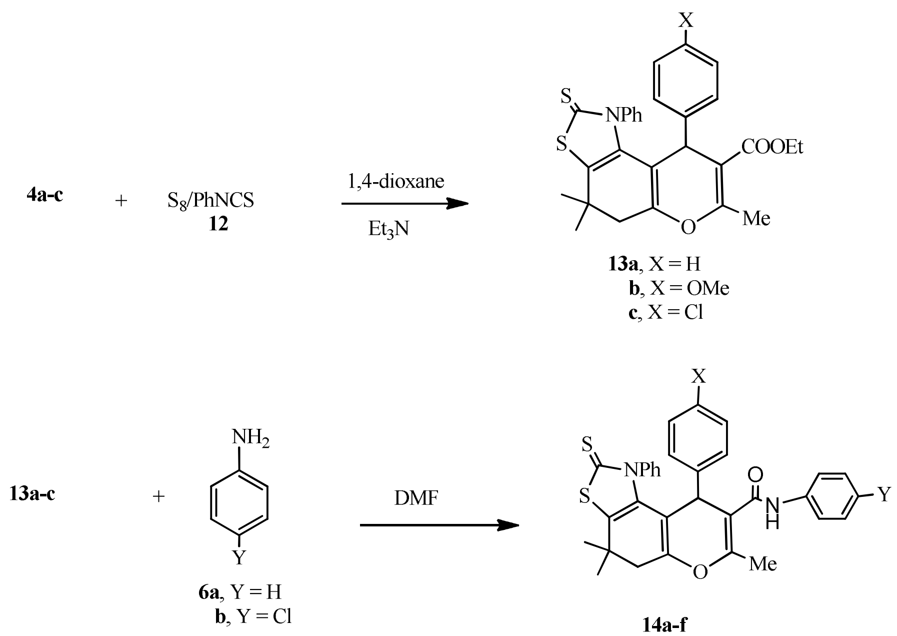


7	a	b	c	d	e	f
X	H	H	OCH_3	OCH_3	Cl	Cl
Y	H	Cl	H	Cl	H	Cl

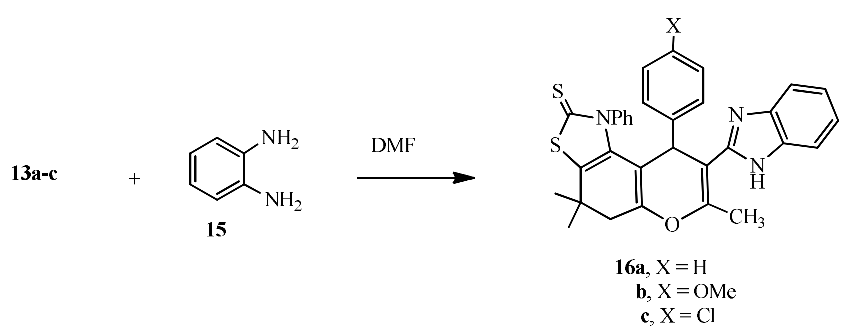
9	a	b	c	d	e	f
X	H	H	OMe	OMe	Cl	Cl
R	H	Ph	H	Ph	H	Ph

11	a	b	c	d	e	f
X	H	H	OMe	OMe	Cl	Cl
R	CN	COOEt	CN	COOEt	CN	COOEt

Scheme 2. Synthesis of compounds **7a-f**; **9a-f** and **11a-f**.

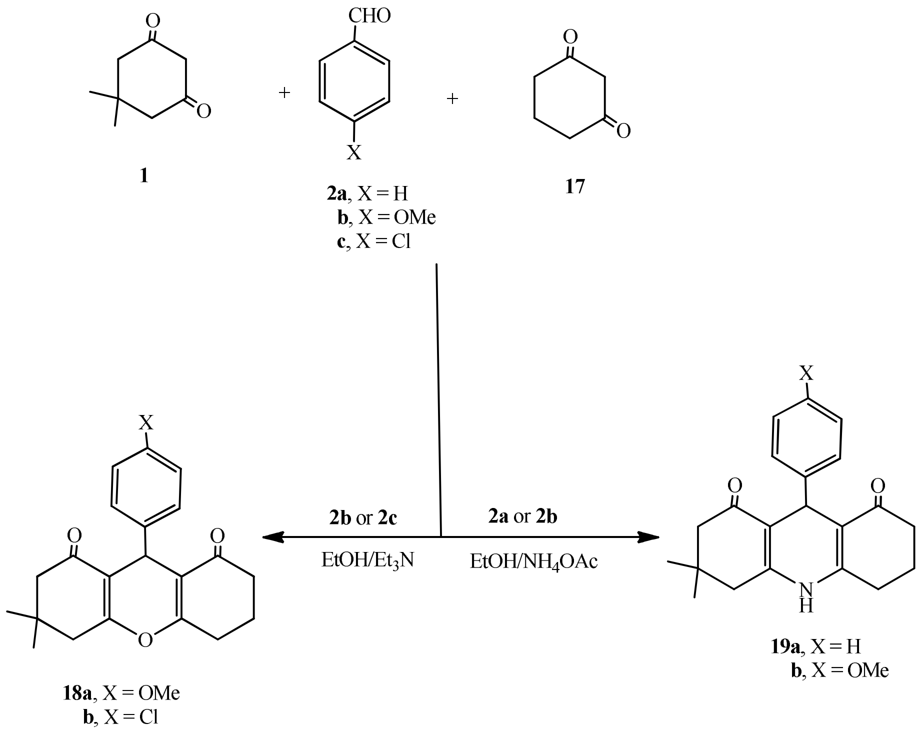


14	a	b	c	d	e	f
X	H	H	OMe	OMe	Cl	Cl
Y	H	Cl	H	Cl	H	Cl



Scheme 3. Synthesis of compounds **13a-c**; **14a-f** and **16a-c**.

The multi-component reactions of dimedone with cyclohexan-1,3-dione and aromatic aldehydes were carried out under two reaction conditions to produce either xanthene or acridine derivatives depending on the nature of the used catalyst. Thus, the reaction of dimedone (**1**) with cyclohexan-1,3-dione and either 4-methoxybenzaldehyde (**2b**) or 4-chlorobenzaldehyde (**2c**) in absolute ethanol containing triethylamine under the reflux conditions gave the xanthene derivatives **18a** and **18b**, respectively. On the other hand, the reaction of dimedone (**1**) with cyclohexan-1,3-dione (**17**) and either benzaldehyde (**2a**) or 4-methoxybenzaldehyde (**2b**) in absolute ethanol containing NH_4OAc gave the acridine derivatives **19a** and **19b**, respectively (Scheme 4). The structures of compounds **18a,b** and **19a,b** were established on the basis of their respective analytical and spectral data (see experimental section).



Scheme 4. Synthesis of compounds **18a,b** and **19a,b**.

Biology

Cell proliferation assay

The IC_{50} values were presented in Table 1 showed that most of the synthesized compounds exhibited potent anti-proliferative activity with IC_{50} values less than 7.0 μM. The applied method using Foretinib as the standard positive control was carried out according to the previously reported work [34-37]. The evaluation of synthesized compounds was carried out on the six cancer cell lines namely A549, HT-29, MKN-45, U87MG, and SMMC-7721 and H460.

IC_{50} values were presented in Table 1 where most of the synthesized compounds exhibited potent anti-proliferative activity with IC_{50} values less than 7.0 μM. In general the nature of substituent whether it is electron attracting as repealing and the nature of the heterocyclic ring has strong influence through inhibitions of the tested compound on the selected cancer cell lines.

Table 1. IC_{50} (μM), inhibitions of the newly synthesized compounds against cancer cell lines *in-vitro* growth inhibitory effects against c-Met enzymatic activity and PC-3.

Compd No.	IC_{50} (μM)									
	A549	H460	HT29	MKN-45	U87MG	SMMC-7721	IC_{50} (nM) c-Met	IC_{50} μM) PC-3	VERO[a] (μM)	SI PC-3[b]
4a	0.36	0.41	0.46	0.63	0.52	0.43	0.37	0.52	57.92	>100
4b	3.21	2.94	3.52	2.73	1.62	2.93	1.80	2.36	25.17	10.66
4c	0.21	0.17	0.16	0.23	0.32	0.27	0.31	0.24	63.28	>100
5a	3.82	2.93	4.51	3.66	4.70	2.85	3.69	1.79	58.30	32.56
5b	5.46	4.33	6.04	5.75	3.96	3.59	4.27	5.02	26.83	5.34
5c	0.24	0.25	0.19	0.35	0.22	0.31	0.27	0.18	64.23	>100
7a	0.25	0.29	0.18	0.35	0.27	0.38	0.17	0.26	56.37	>100
7b	0.21	0.24	0.26	0.31	0.23	0.32	0.28	0.19	63.52	>100
7c	2.73	3.59	4.82	3.29	4.52	4.29	3.39	2.17	36.52	16.83
7d	0.89	0.77	0.59	1.12	1.02	0.86	0.93	0.71	58.93	83.00
7e	0.51	0.62	0.74	0.69	0.51	0.73	0.82	0.38	65.12	>100
7f	0.18	0.19	0.22	0.23	0.33	0.52	0.27	0.32	58.32	>100
9a	1.26	1.23	1.46	2.25	1.38	1.42	1.38	2.82	35.62	12.63
9b	0.65	0.79	0.62	0.48	0.39	0.62	0.72	0.85	44.39	52.22
9c	1.22	2.41	1.73	2.66	2.40	1.38	2.17	2.47	58.69	23.76
9d	6.32	8.53	7.42	6.39	6.27	7.28	6.16	5.72	48.32	8.45
9e	0.38	0.40	0.36	0.53	0.48	0.65	0.38	0.31	64.54	>100
9f	0.45	0.39	0.27	0.27	0.43	0.32	0.26	0.32	58.29	>100
11a	0.23	0.28	0.35	0.27	0.41	0.29	0.37	0.27	56.49	>100
11b	1.18	1.52	1.16	0.86	1.31	0.79	0.62	0.58	68.25	>100
11c	3.55	3.26	2.73	3.90	3.64	5.27	3.82	2.90	48.27	16.44
11d	2.50	3.17	3.92	2.79	3.59	4.16	3.82	2.72	59.35	21.82
11e	0.18	0.19	0.23	0.25	0.30	0.23	0.16	0.18	61.46	>100
11f	0.28	0.37	0.35	0.32	0.37	0.31	0.36	0.29	58.26	>100
13a	0.48	0.52	0.63	0.49	0.58	0.61	0.59	0.49	64.27	>100
13b	0.39	0.42	0.57	0.33	0.51	0.62	0.59	0.29	56.12	>100
13c	0.36	0.26	0.28	0.30	0.22	0.29	0.35	0.32	58.47	>100
14a	0.58	0.48	0.62	0.53	0.69	0.51	0.39	0.40	59.65	>100
14b	0.32	0.34	0.28	0.31	0.26	0.48	0.31	0.26	62.53	>100
14c	2.82	2.59	3.42	2.69	2.26	3.90	3.24	3.19	42.79	13.41
14d	0.92	1.62	0.88	1.15	1.78	1.52	1.41	1.26	60.32	47.87
14e	0.32	0.41	0.29	0.36	0.27	0.28	0.35	0.31	63.44	>100
14f	0.19	0.18	0.23	0.26	0.16	0.22	0.27	0.32	59.27	>100
16a	0.38	0.42	0.28	0.26	0.32	0.42	0.26	0.30	60.20	>100
16b	1.05	1.15	0.85	0.76	1.01	1.34	0.93	0.80	48.66	60.82
16c	0.17	0.28	0.24	0.18	0.25	0.24	0.29	0.31	60.42	>100
18a	4.63	3.69	5.72	3.72	3.91	5.32	2.92	1.49	29.52	19.81
18b	0.20	0.12	0.39	0.28	0.21	0.18	0.25	0.39	60.66	>100
19a	1.28	2.27	2.27	3.55	2.16	2.28	3.17	1.63	38.63	32.70
19b	4.43	4.39	3.25	6.78	4.27	4.36	2.55	3.29	30.72	9.34
Foretinib	0.08	0.18	0.15	0.03	0.90	0.44	**Foretinib** 1.16	**Anibamine** 3.26	-	-

[a]VERO, Monkey Kidney cell line (Cat No-11095–080). [b]Selectivity index (SI) were calculated by IC_{50} values in normal cell line divided by IC_{50} values in PC-3 cancer cell line.

Structure activity relationship

Table 1 demonstrated that many of the synthesized compounds revealed high inhibitions toward the used cancer cell lines. The most cytotoxic compounds were the twenty-five compounds **4a**,

4c, **5c**, **7a**, **7b**, **7d**, **7e**, **7f**, **9b**, **9e**, **19f**,**11a**, **11e**, **11f**, **13a**, **13b**, **13c**, **14a**, **14b**, **14d**, **14e**, **14f**, **16a**, **16c** and **16b** where such compounds showed inhibitions < 1.00 µM. Considering the pyran derivatives **4a-c**, it is clear that compounds **4a** (X = H) and **4b** (X= Cl) showed the highest cytotoxicity. For compound **4b** (X = OCH_3) the presence of the electron donating OCH_3 was responsible for its low inhibitions. On the other hand, the quinoline derivatives **5a-c**, where compound **5a** (X = Cl) exhibited high inhibitions on the six cancer cell lines. It was obvious that the anilide derivatives **7a-f** exhibited high inhibitions on the six cancer cell lines except compound **7c** (X = OCH_3, Y = H) which showed moderate inhibitions. It was surprised that compound **7d** (X = OCH_3, Y = Cl) showed high inhibitions although it contains a methoxy moiety, however, it seemed that the presence of the Cl group together with the anilide moiety enhance the inhibition more than the suspension effect produced by the OCH_3 group. For the chromene-3-carbohydrazide **9a-f** and the thieno[3,2-*f*]chromene-8-carboxylate **11a-f** derivatives, where compounds **9a**, **11a** (X = R =H), **9e**, **11e** (X = Cl, R = H) and **9f**, **11f** (X = Cl, R = Ph) exhibited high inhibitions among the twelve-compounds. Interestingly, the chromeno[5,6-*d*]thiazole-8-carboxylate derivatives **13a-c** and **14a-f** where all compounds exhibited high inhibitions except compound **14c** (X = OCH_3, Y = H) which exhibited moderate inhibitions. For the benzimidazole derivatives **16a-c**, compounds **16a** (X = H) and **16c** (X = Cl) exhibited high inhibitions on the six cancer cell lines. Finely, the xanthenes **18a,b** and the acridine**s 19a,b** where compound **18b** (X = Cl) exhibited the highest inhibitions among the four compounds. It was of great value to note that in most cases the presence of an electron withdrawing group within the structure of the molecule and sulphur containing heterocyclic moiety had a strong impact through the reactivity of the compound. It is of great value to mention that compounds **4a, 4c, 5c, 7a, 7b**, **7d**, **7e**, **7f**, **9b**, **9e**, **9f**, **11a**, **11e**, **11f**, **13a**, **13b**, **13c**, **14a**, **14b**, **14e**, **14f**, **16a** and **18b** exhibited higher inhibitions than the reference foretinib against U87MG cell line. On the other hand compounds **4a**, **4c**, **5c**, **7a**, **7b**, **9f**, **11a**, **11e**, **11f**, **13c**, **14e**, **14f**, **16a**, **16c** and **18b** showed higher inhibitions than the reference foretinib against SMMC-7721 cell line.

HTRF kinase assay

Materials. c-Met (mesenchymal epithelial transition factor) is a multifunctional transmembrane tyrosine kinase and acts as a receptor for hepatocyte growth factor/Scatter factor (HGF/SF) [38, 39]. The IC_{50} values were presented in Table 1 for c-Met kinase and prostate cancer cell line PC-3 inhibitions.

As indicated from Table 1, all tested compounds displayed potent c-Met enzymatic activity with IC_{50} values ranging from 0.25 to 10.30 nM and potent prostate PC-3 cell line inhibitions with IC_{50} values ranging from 0.16 to 6.16 µM. Compared with foretinib (IC_{50} = 1.16 nM), the twenty-five compounds **4a**, **4c**, **5c**, **7a**, **7b**, **7d**, **7e**, **7f**, **9b**, **9e**, **9f**, **11a**, **11e**, **11f**, **13a**, **13b**, **13c**, **14a**, **14b**, **14d**, **14e**, **14f**, **16a**, **16c** and **18b** showed inhibition < 1.00 µM. Remarkably, all of the synthesized compounds showed anti-proliferation activity higher than the standard Anibamine (IC_{50} = 3.26 µM) except compounds **5b** and **19b**. Analyzing the data indicated in Table 2 showed that compounds **4a**,**4c**, **5c**, **7a**, **7b**, **7e**, **7f**, **9e**, **9f**, **11a**, **11b**, **11e**, **11f**, **13a**, **13b**, **13c**, **14a**, **14b**, **14e**, **14f**, **16a**, **16c** and **18b** with SI > 100.

*Morphological effect of **14f** and **16c** on A549 cell line*

After treatment with doses of **14f** and **16c**, the morphological toward A549 cell line and cellular damage of apoptosis were studied from the cell shrinkage and chromatin condensation, which was visualized using AO/EB staining assay. The results of AO/EBstaining are shown in Figure 2. One can observe that cell penetrable fluorescent dye AO stain stick to the membrane surface of both live and dead cells, whereas EB (another fluorescence dye) allows staining the nuclear DNA in damaged cells [40, 41]. The early apoptotic cells were stained bright green fluorescence with

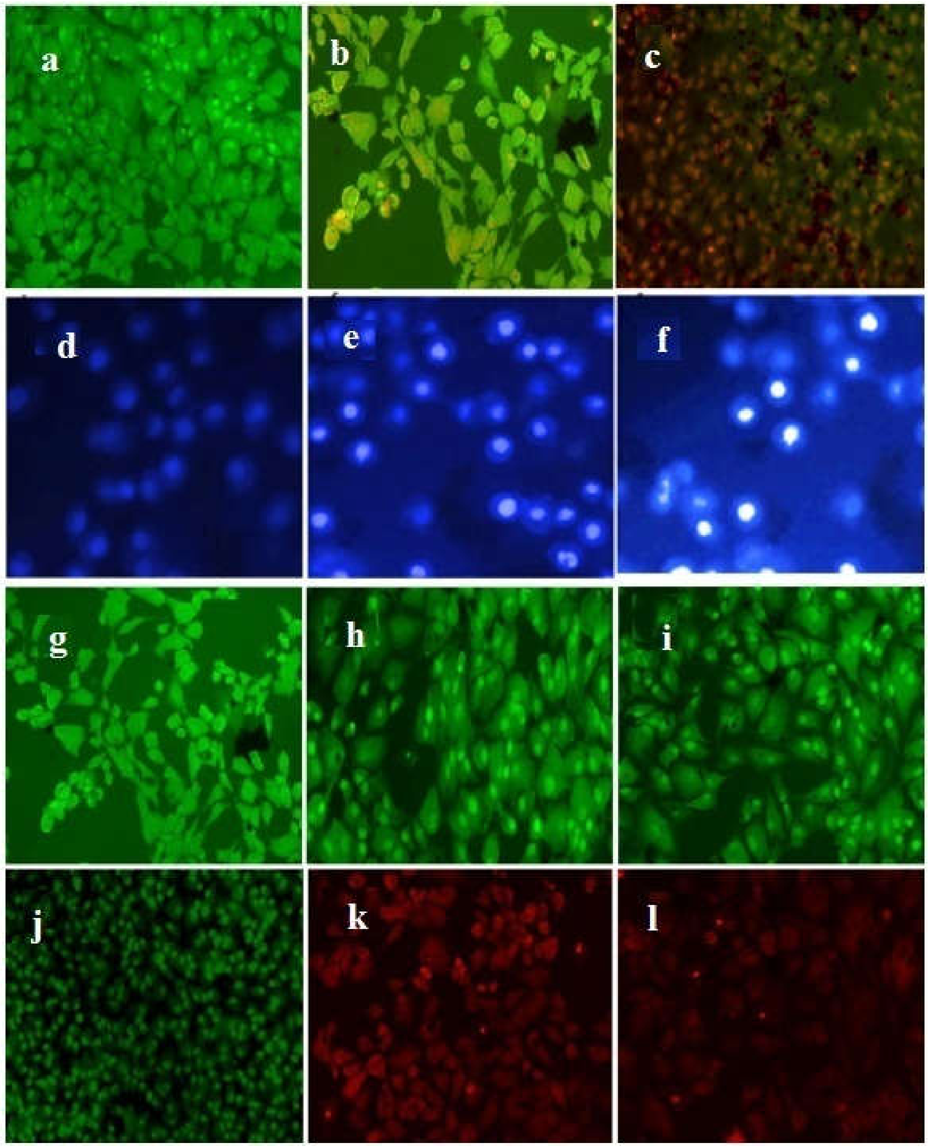


Figure 2. Apoptosis morphology of control (a), **14f** (b) and **16c** (c) against A549 cells, visualized using AO-EB staining method under a florescence microscope. Nuclear morphology of apoptosis of control (d), **14f** (e) and **16c** (f) of A549 treated cells, visualized using Hoechst 33342 staining method under a florescence microscope. Formation of ROS production in A549 cells was monitored using DCFH-CA staining in control (g) **14f** (h) and **16c** (i). Susceptibility of the mitochondrial membrane in A549 cells control (j), **14f** (k) and **16c** (l), visualized using a rhodamine 123 staining method.

condensed chromatin and late apoptotic cells were stained orange fluorescence. The nonviable cells were stained orange to red fluorescence nuclei with no indication of chromatin condensation [42]. In addition, the frequent increase in number of apoptotic cells was observed after treatment with **14f** and **16c** (Figure 2a-l). Among the treatment, **14f** showed complete cell death while stronger chromatin damage was observed compared to **16c**. The complete apoptotic cells of **14f** were clearly exhibited an orange color as reported in Figure 2c. This clearly evidences that the whole cell was damaged and exhibited different morphology compared to the control. The control cells did not show any morphological alteration and both nuclei and cytoplasm fluoresced uniformly green. The results revealed that **14f** and **16c** stimulate cell death through apoptosis where **14f** was found to be able to induce strong apoptosis. Figures 3 and 4 showed the statistical shrinking of A549 cell line by **14f** (Figure 3) and **16c** (Figure 4).

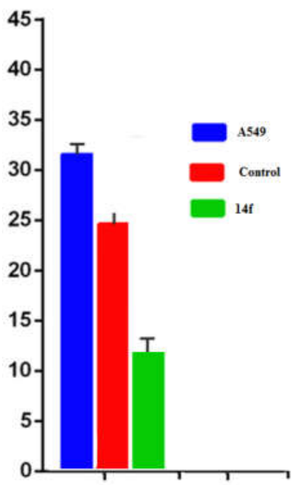


Figure 3. Zone inhibitions by compound **14f** compared with the control.

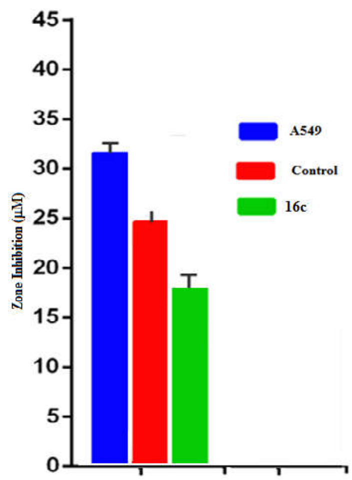


Figure 4. Zone inhibitions by compound **16c** compared with the control.

EXPERIMENTAL

Chemistry

For the synthesized compounds, the melting points were measured in addition; the IR spectra (KBr discs) were recorded on a FITR plus 460 or Pye Unicam SP-1000 spectrophotometer. ^{1}H-NMR spectra were recorded using the Varian Gemini-300 (300 MHZ) (Cairo University) in DMSO-d_6 as solvent using TMS as internal standard and chemical shifts are demonstrated as δ ppm. The molecular weights were determined using the Ex shimadzu instruments for recording m/z values. Elemental analyses CHNS were measured using the Vario El III Elemental CHNS analyzer.

*General procedure for the synthesis of the tetrahydro-4H-chromene-3-carboxylate derivatives **4a-c***

Either phenylcarbinal (1.08 g, 0.01), 4-methoxyphenylcarbinal (1.38 g, 0.01) or 4-chlorophenylcarbinal (1.40 g, 0.01 mol) was added to each of dimedone (1.40 g, 0.01) and ethyl 3-oxobutanoate (1.30 g, 0.01 mol) in absolute ethanol (50 mL) containing piperidine (2.0 mL). The reaction mixture was heated under the reflux conditions for 2 h and the produced solid product after pouring onto ice/water containing a few drops of hydrochloric acid was collected by filtration.

*Ethyl 2,7,7-trimethyl-5-oxo-4-phenyl-5,6,7,8-tetrahydro-4H-chromene-3-carboxylate (**4a**)*. Pale yellow crystals from ethanol, m.p. 173-175 ºC, yield: 2.04 g (60%), IR (ν, cm^{-1}): 3050 (CH-aromatic), 2993, 2899 (methylene, methyl), 1688, 1705 (two carbonyls), 1568 (vinyl bondin+g). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 0.98, 1.08 (s, 6H, two methyl), 1.13 (t, 3H, J = 5.80 Hz, methyl ester), 1.80 (m, 2H, methylene), 2.23 (s, 2H, methylene), 2.70 (s, 3H, methyl), 4.23 (q, 2H, J = 5.80 Hz, methylene ester), 6.02 (s, 1H, pyran H-4), 7.02-7.42 (m, 5H, phenyl), ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.5 (methyl ester), 24.2 (two methyl), 39.5, 42.1 (two methylenes), 38.7 (CH_3), 50.4 (methylene ester), 90.8 (pyran C-4), 120.3, 122.5, 123.8, 125.4 (phenyl), 129.6, 130.4, 134.7, 136.7 (pyran C-2, C-3, C-5, C-6), 165.8, 166.2 (two carbonyls). Anal. cacld for $C_{21}H_{24}O_4$ (340.41): C, 74.09; H, 7.11%. Found: C, 73.86; H, 6.93 %. MS: m/z = 340 M^+ (54%).

*Ethyl 4-(4-methoxyphenyl)-2,7,7-trimethyl-5-oxo-5,6,7,8-tetrahydro-4H-chromene-3-carboxylate (**4b**)*. Pale yellow crystals from EtOH, m.p. 134-136 ºC, yield: 2.40 g (65%), IR (ν, cm^{-1}): 3050 (CH-aromatic), 2991, 2886 (methylene, methyl), 1689, 1705 (two carbonyls), 1568 (vinyl bonding).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 0.96, 1.12 (s, 6H, two methyl), 1.12 (t, 3H, J = 6.72 Hz, methyl ester), 1.85 (m, 2H, methylene), 2.26 (s, 2H, methylene), 2.71 (s, 3H, methyl), 3.73 (s, 3H, methoxy), 4.22 (q, 2H, J = 6.72 Hz, methylene ester), 6.03 (s, 1H, pyran H 4), 7.26-7.58 (m, 4H, phenyl).^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.9 (methyl ester), 24.6 (two methyl), 39.3, 42.5 (two methylenes), 38.8 (methyl), 50.2 (methylene ester), 50.8 (methoxy), 90.6 (pyran C-4), 120.6, 123.2, 124.2, 125.9 (phenyl), 129.6, 130.2, 134.4, 136.2 (pyran C-2, C-3, C-5, C-6), 165.6, 166.8 (two carbonyls). Anal. cacld for $C_{22}H_{26}O_5$ (370.44): C, 71.33; H, 7.07%. Found: C, 71.27; H, 7.04%. MS: m/z = 370 (65%).

*Ethyl 4-(4-chlorophenyl)-2,7,7-trimethyl-5-oxo-5,6,7,8-tetrahydro-4H-chromene-3-carboxylate (**4c**)*. Pale yellow crystals of EtOH, m.p. 127-129 ºC, yield: 2.31 g (62%), IR (ν, cm^{-1}): 3050 (CH-aromatic), 2993, 2899 (methylene, methyl), 1689, 1703 (two carbonyls), 1568 (vinyl bonding). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.08 (s, 6H, two methyl), 1.12 (t, 3H, J = 6.73 Hz, methyl ester),1.83 (m, 2H, methylene), 2.22 (s, 2H, methylene), 2.72 (s, 3H, methyl), 4.21 (q, 2H, J = 6.73 Hz, methylene ester), 6.04 (s, 1H, pyran H-4), 7.25-7.56 (m, 4H, phenyl).^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.8 (methyl ester), 24.2 (two methyls), 39.6, 42.3 (two methylenes), 38.7 (methyl), 50.3 (methylene ester), 90.6 (pyran C-4), 120.1, 122.5, 123.4, 125.8 (phenyl), 129.4, 130.2, 134.7, 136.5 (pyran C-2, C-3, C-5, C-6), 165.9, 166.6 (two carbonyls). Anal. cacld for $C_{21}H_{23}ClO_4$ (374.86): C, 67.29; H, 6.18%. Found: C, 67.47; H, 6.23%. MS: m/z = 374, 376 M^+, M^{+2} (66%).

*General procedure for the synthesis of the hexahydroquinoline-3-carboxylate derivatives **5a-c***

The same procedure described before for the synthesis of **4a-c** was applied but using NH_4OAc as a catalyst instead of Et_3N.

*Ethyl 2,7,7-trimethyl-5-oxo-4-phenyl-1,4,5,6,7,8-hexahydroquinoline-3-carboxylate (**5a**)*. Pale yellow crystals from EtOH, m.p. 180-182 ºC, yield: 1.96 g (58%), IR (ν, cm^{-1}): 3477-3362 (imino), 3053 (CH-aromatic), 2991, 2894 (methylene, methyl), 1689, 1702 (two carbonyls), 1564 (vinyl bonding). ^{1}H NMR(DMSO-d_6, 300 MHz): δ = 1.02, 1.08 (s, 6H, two methyls), 1.12 (t, 3H, J = 7.11 Hz, methyl ester),1.80 (m, 2H, methylene), 2.22 (s, 2H, methylene), 2.72 (s, 3H, methyl), 4.23 (q, 2H, J = 7.11 Hz, methylene ester), 6.04 (s, 1H, pyran H 4), 7.23-7.48 (m, 5H, phenyl), 8.29 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.7 (methyl ester), 24.2 (two methyls), 39.2, 42.4 (two methylenes), 38.5 (methyl), 50.2 (methylene ester), 91.4

(pyridine C-4), 120.8, 122.1, 124.2, 125.1 (phenyl), 129.6, 130.5, 134.5, 136.3 (pyridine C-2, C-3, C-5, C-6), 165.8, 166.2 (two carbonyls). Anal. cacld for $C_{21}H_{25}NO_3$ (339.43): C, 74.31; H, 7.42; N, 4.13%. Found: C, 74.38; H, 7.52; N, 4.26%. MS: *m/z* = 339 M^+ (64%).

*Ethyl 4-(4-methoxyphenyl)-2,7,7-trimethyl-5-oxo-1,4,5,6,7,8-hexahydro-quinoline-3-carboxylate (**5b**)*. Yellow crystals from *p*-dioxane, m.p. 210-212 ºC, yield: 1.91 g (52%), IR (ν, cm^{-1}): 3484-3352 (imino), 3050 (CH-aromatic), 2975, 2886 (methylene, methyl), 1689, 1702 (two carbonyls), 1564 (vinyl bonding).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.12 (s, 6H, two methyl), 1.14 (t, 3H, J = 6.55 Hz, methyl ester),1.82 (m, 2H, methylene), 2.28 (s, 2H, methylene), 2.73 (s, 3H, methyl), 3.72 (s, 3H, methoxy), 4.22 (q, 2H, J = 6.55 Hz, methylene ester), 6.03 (s, 1H, pyran H 4), 7.26-7.68 (m, 4H, phenyl), 8.27 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.7 (methyl ester), 24.2 (two methyls), 39.5, 42.6 (two methylenes), 38.4 (methyl), 50.3 (methylene ester), 50.6 (methoxy), 90.5 (pyridine C-4), 120.7, 123.1, 124.2, 125.7 (phenyl), 129.6, 130.2, 133.9, 135.8 (pyridine C-2, C-3, C-5, C-6), 165.4, 166.2 (two carbonyls). Anal. cacld for $C_{22}H_{27}NO_4$ (369.45): C, 71.52; H, 7.37; N, 3.79%. Found: C, 71.37; H, 7.41; N, 3.82%. MS: *m/z* = 369 M^+ (78%).

*Ethyl 4-(4-chlorophenyl)-2,7,7-trimethyl-5-oxo-1,4,5,6,7,8-hexahydroquinoline-3-carboxylate (**5c**)*. Pale yellow crystals from EtOH, m.p. 196-198 ºC, yield: 2.49 g (67%), IR (ν, cm^{-1}): 3477-3362 (imino), 3050 (CH-aromatic), 2991, 2879 (methylene, methyl), 1688, 1701 (two carbonyls), 1565 (vinyl bonding). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.09 (s, 6H, two methyl), 1.13 (t, 3H, J = 5.23 Hz, methyl ester), 1.83 (m, 2H, methylene), 2.26 (s, 2H, methylene), 2.71 (s, 3H, methyl), 4.23 (q, 2H, J = 5.23 Hz, methylene ester), 6.03 (s, 1H, pyridine H-4), 7.27-7.64 (m, 4H, phenyl), 8.39 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.4 (methyl ester), 24.5 (two methyl), 39.3, 42.6 (two methylenes), 38.9 (methyl), 50.2 (methylene ester), 90.5 (pyridine C-4), 120.4, 122.7, 123.6, 125.6 (phenyl), 129.2, 130.5, 134.1, 136.3 (pyridine C-2, C-3, C-5, C-6), 165.5, 166.9 (two carbonyls). Anal. cacld for $C_{21}H_{24}ClO_3$ (373.87): C, 67.46; H, 6.47; N, 3.75%. Found: C, 67.52; H, 6.42; N, 4.04 %. MS: *m/z* = 373, 375 M^+, M^{+2} (80%).

*General procedure for the synthesis of the 5,6,7,8-tetrahydro-4H-chromene-3-carboxamide derivatives 7**a-f***

Either phenylamine (0.94 g, 0.01 mol) or 4-chlorophenylamine (1.27 g, 0.01 mol) was added to a solution of either **4a** (3.40 g, 0.01 mol), **4b** (3.70 g, 0.01 mol) or **4c** (3.74 g, 0.01 mol) in dimethylformamide (40 mL). The whole reaction mixture was heated under the reflux conditions for 2 h then the working up was carried out in a similar manner like the synthesis of **4a-c**.

*2,7,7-Trimethyl-5-oxo-N,4-diphenyl-5,6,7,8-tetrahydro-4H-chromene-3-carboxamide (7**a**)*. Pale yellow crystals from EtOH, m.p. 145-147 ºC, yield: 1.36 g (61%), IR (ν, cm^{-1}): 3497-3332 (imino), 3053 (CH-aromatic), 2991, 2896 (methylene, methyl), 1688, 1701 (two carbonyls), 1563 (vinyl bonding). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.06 (s, 6H, two methyl), 1.83 (m, 2H, methylene), 2.22 (s, 2H, methylene), 2.76 (s, 3H, methyl), 6.05 (s, 1H, pyran H 4), 7.24-7.46 (m, 10H, two phenyls), 8.32 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.2 (two methyl), 39.6, 42.7 (two methylenes), 38.8 (methyl), 120.2, 120.6, 121.4, 122.1, 123.0, 123.6, 124.8, 125.5 (two phenyls), 130.6, 134.2, 136.7, 138.5 (pyran C-2, C-3, C-5, C-6), 165.5, 166.6 (two carbonyls). Anal. cacld for $C_{25}H_{25}NO_3$ (387.47): C, 77.49; H, 6.50; N, 3.61%. Found: C, 77.60; H, 6.41; N, 3.79%. MS: *m/z* = 387 M^+ (64%).

*N-(4-Chlorophenyl)-2,7,7-trimethyl-5-oxo-4-phenyl-5,6,7,8-tetrahydro-4H-chromene-3-carbox-amidee (7**b**)*. Yellow crystals from EtOH, m.p. 172-174 ºC, yield: 1.36 g (61%), IR (ν, cm^{-1}): 3483-3351 (imino), 3053 (CH-aromatic), 2994, 2893 (methylene, methyl), 1689, 1702 (two carbonyls), 1560 (vinyl bonding). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.08 (s, 6H, two methyl), 1.85 (m, 2H, methylene), 2.21 (s, 2H, methylene), 2.78 (s, 3H, methyl), 6.07 (s, 1H, pyran H 4), 7.22-7.56 (m, 9H, two phenyl), 8.34 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.4 (two methyls), 39.6, 42.9 (two methylene), 38.9 (methyl), 120.4, 120.8, 121.3, 121.6, 123.3, 123.9, 124.3, 125.8 (two phenyls), 130.1, 134.2, 136.7, 138.4 (pyran C-2, C-3, C-5, C-6), 165.9, 166.7 (two carbonyls). Anal. cacld for $C_{25}H_{24}ClNO_3$ (421.92): C, 71.17; H, 5.73; N, 3.32%. Found: C, 71.27; H, 5.94; N, 3.46%. MS: *m/z* = 421, 423 M^+, M^{+2} (70%).

*4-(4-Methoxyphenyl)-2,7,7-trimethyl-5-oxo-N-phenyl-5,6,7,8-tetrahydro-4H-chromene-3-carboxamide (7**c**)*. White crystals from ethanol, m.p. 177-179 °C, yield: 2.71 g (65%), IR (ν, cm^{-1}): 3474-3327 (imino), 3053 (CH-aromatic), 2987, 2873 (methylene, methyl), 1689, 1702 (two carbonyls), 1561 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.08 (s, 6H, two methyls), 1.84 (m, 2H, methylene), 2.21 (s, 2H, methylene), 2.73 (s, 3H, methyl), 3.69 (s, 3H, methoxy), 6.07 (s, 1H, pyran H 4), 7.26-7.54 (m, 9H, two phenyl), 8.36 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.2 (two methyl), 39.6, 42.7 (two methylene), 38.8 (methyl), 50.6 (methoxy), 120.3, 120.9, 121.3, 122.6, 123.2, 123.9, 124.2, 125.8 (two phenyls), 131.6, 134.2, 136.7, 138.5 (pyran C-2, C-3, C-5, C-6), 165.8, 166.9 (two carbonyls). Anal. cacld for $C_{26}H_{27}NO_4$ (417.50): C, 74.80; H, 6.52; N, 3.35%. Found: C, 75.02; H, 6.70; N, 3.46%. MS: *m/z* = 417 M^+ (64%).

*N-(4-Chlorophenyl)-4-(4-methoxyphenyl)-2,7,7-trimethyl-5-oxo-5,6,7,8-tetrahydro-4H-chromene-3-carboxamide (7**d**)*. White crystals from EtOH, m.p. 145-147 °C, yield: 2.48 g (55 %), IR (ν, cm^{-1}): 3483-3342 (imino), 3056 (CH-aromatic), 2983, 2870 (methylene, methyl), 1689, 1701 (two carbonyls), 1565 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.04, 1.05 (s, 6H, two methyls), 1.83 (m, 2H, methylene), 2.23 (s, 2H, methylene), 2.76 (s, 3H, methyl), 3.71 (s, 3H, methoxy), 6.09 (s, 1H, pyran H 4), 7.23-7.57 (m, 8H, two phenyls), 8.38 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.5 (two Methyl), 39.6, 42.9 (two methylene), 38.3 (methyl), 50.8 (methoxy), 120.1, 120.5, 121.2, 122.4, 123.5, 123.7, 124.4, 125.6 (two phenyls), 130.3, 133.6, 136.5, 138.7 (pyran C-2, C-3, C-5, C-6), 165.6, 166.7 (two carbonyls). Anal. cacld for $C_{26}H_{26}ClNO_4$ (451.94): C, 69.10 H, 5.80; N, 3.10%. Found: C, 69.24; H, 5.69; N, 3.25%. MS: *m/z* = 451, 452 M^+ (80%).

*4-(4-Chlorophenyl)-2,7,7-trimethyl-5-oxo-N-phenyl-5,6,7,8-tetrahydro-4H-chromene-3-carboxamide (7**e**)*. White crystals from EtOH, m.p. 210-212 °C, yield: 2.73 g (65%), IR (ν, cm^{-1}): 3473-3328 (imino), 3054 (CH-aromatic), 2983, 2870 (methylene, methyl), 1688, 1701 (two carbonyls), 1562 (vinyl bonding).^1H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.06 (s, 6H, two methyls), 1.81 (m, 2H, methylene), 2.27 (s, 2H, methylene), 2.78 (s, 3H, methyl), 6.09 (s, 1H, pyran H-4), 7.25-7.59 (m, 9H, two phenyls), 8.36 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.7 (two Methyl), 39.9, 42.8 (two methylenes), 38.6 (methyl), 120.3, 120.8, 121.6, 122.8, 123.2, 123.3, 124.5, 125.9 (two phenyls), 130.7, 133.8, 136.2, 138.4 (pyran C-2, C-3, C-5, C-6), 165.6, 166.9 (two carbonyls). Anal. cacld for $C_{25}H_{24}ClNO_3$ (421.92): C, 71.17; H, 5.73; N, 3.32%. Found: C, 71.26; H, 5.59; N, 3.18%. MS: *m/z* = 421, 423 M^+, M^{+2} (68%).

*N,4-bis(4-Chlorophenyl)-2,7,7-trimethyl-5-oxo-5,6,7,8-tetrahydro-4H-chromene-3-carboxamide (7**f**)*. White crystals from ethanol, m.p. 177-179 °C, yield: 2.86 g (63%), IR (ν, cm^{-1}): 3484-3327 (imino), 3056 (CH-aromatic), 2983, 2873 (methylene, methyl), 1689, 1704 (two carbonyls), 1562 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.08 (s, 6H, two methyl), 1.83 (m, 2H, methylene), 2.28 (s, 2H, methylene), 2.59 (s, 3H, methyl), 6.07 (s, 1H, pyran H-4), 7.23-7.64 (m, 8H, two phenyl), 8.38 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.7 (two methyls), 39.9, 42.8 (two methylene), 38.8 (two methyls), 120.3, 120.6, 121.9, 122.4, 123.2, 123.6, 124.1, 125.4 (two phenyls), 130.2, 133.6, 136.5, 138.7 (pyran C-2, C-3, C-5, C-6), 165.8, 166.9 (two carbonyls). Anal. cacld for $C_{25}H_{23}Cl_2NO_3$ (456.36): C, 65.80; H, 5.08; N, 3.07%. Found: C, 65.77; H, 5.25; N, 3.25%. MS: *m/z* = 456, 458 M^+, M^{+2} (75%).

*General procedure for thesynthesisofthe 5-hydrazono-5,6,7,8-tetrahydro-4H-chromene-3-carbohydrazide derivatives **9a-f***

1,2-Diaminobenzene (1.08 g, 0.01 mol) was added to a solution of either **4a** (3.40 g, 0.01 mol), **4b** (3.70 g, 0.01 mol) or **4c** (3.74 g, 0.01 mol) in dimethylformamide (40 mL). The whole reaction mixture was heated under the reflux conditions for 2 h then the working up was carried out in a similar manner as previously described for the synthesis of **4a-c**.

*5-Hydrazineylidene-2,7,7-trimethyl-4-phenyl-5,6,7,8-tetrahydro-4H-chromene-3-carbohydrazide (**9a**)*. White crystals from EtOH, m.p. 148-150 ºC, yield: 1.79 g (58%), IR (ν, cm^{-1}): 3462-3337 (imino), 3053 (CH-aromatic), 2986, 2871 (methylene, methyl), 1687 (carbonyl), 1565 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.06 (s, 6H, two methyls), 1.86 (m, 2H, methylene), 2.25 (s, 2H, methylene), 2.54 (s, 3H, methyl), 4.88, 5.04 (2s, 4H, D_2O exchangeable, amino), 6.07 (s, 1H, pyran H 4), 7.23-7.64 (m, 5H, phenyl), 8.38 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyl), 39.7, 42.8 (two methylenes), 38.8 (methyl), 121.39, 122.6, 123.4, 125.4 (phenyl), 130.2, 133.4, 136.7, 138.1 (pyran C-2, C-3, C-5, C-6), 165.9 (carbonyl). Anal. cacld for $C_{19}H_{24}N_4O_2$ (340.43): C, 67.04; H, 7.11; N, 16.46%. Found: C, 67.18; H, 7.04; N, 16.25%. MS: *m/z* = 340 M^+ (68%).

*2,7,7-Trimethyl-N',4-diphenyl-5-(2-phenylhydrazineylidene)-5,6,7,8-tetrahydro-4H-chromene-3-carbohydrazide (**9b**)*. White crystals from EtOH, m.p. 141-143 ºC, yield: 3.44 g (70%), IR (ν, cm^{-1}): 3487-3340 (imino), 3055 (CH-aromatic), 2989, 2876 (methylene, methyl), 1687 (carbonyl), 1565 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.08 (s, 6H, two methyl), 1.83 (m, 2H, methylene), 2.28 (s, 2H, methylene), 2.56 (s, 3H, methyl), 6.07 (s, 1H, pyran H-4), 7.24-7.58 (m, 15H, three phenyl), 8.29, 8.38, 8.50(3s, 3H, D_2O exchangeable, three imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.5 (two methyls), 39.5, 42.9 (two methylenes), 38.5 (methyl), 120.2, 120.5, 121.4, 122.6, 122.8, 123.2, 123.5, 124.2, 124.5, 124.7, 125.7, 125.8 (three phenyls), 130.2, 133.8, 136.2, 138.3 (pyran C-2, C-3, C-5, C-6), 166.3 (carbonyl). Anal. cacld for $C_{31}H_{32}N_4O_2$ (492.62): C, 75.58; H, 6.55; N, 11.37%. Found: C, 75.37; H, 6.72; N, 11.49%. MS: *m/z* = 492 M^+ (85%).

*5-Hydrazineylidene-4-(4-methoxyphenyl)-2,7,7-trimethyl-5,6,7,8-tetrahydro-4H-chromene-3-carbohydrazide (**9c**)*. Yellow crystals from EtOH, m.p. 112-114 ºC, yield: 2.30 g (62%), IR (ν, cm^{-1}): 3492-3318 (NH), 3053 (CH-aromatic), 2988, 2871 (methylene, methyl), 1688 (carbonyl), 1562 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.07 (s, 6H, two methyls), 1.87 (m, 2H, methylene), 2.24 (s, 2H, methylene), 2.58 (s, 3H, methyl), 3.71 (s, 3H, methoxy), 4.89, 5.16 (2s, 4H, D_2O exchangeable, amino), 6.05 (s, 1H, pyran H 4), 7.24-7.54 (m, 4H, phenyl), 8.36 (s, 1H, D_2O exchangeable, NH). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyl), 39.7, 42.8 (two methylenes), 38.8 (methyl), 50.6 (methoxy), 120.5, 121.8, 123.6, 124.8 (phenyl), 130.6, 133.6, 136.5, 138.3 (pyran C-2, C-3, C-5, C-6), 165.8 (carbonyl). Anal. cacld for $C_{20}H_{26}N_4O_3$ (370.45): C, 64.84; H, 7.07; N, 15.12%. Found: C, 64.57; H, 7.22; N, 15.39%. MS: *m/z* = 370 M^+ (55%).

*4-(4-Methoxyphenyl)-2,7,7-trimethyl-N'-phenyl-5-(2-phenylhydrazineylidene)-5,6,7,8-tetrahydro-4H-chromene-3-carbohydrazide (**9d**)*. Yellow crystals from p-dioxane, m.p. 180-182 ºC, yield: 3.13 g (60%), IR (ν, cm^{-1}): 3484-3348 (imino), 3055 (CH-aromatic), 2989, 2879 (methylene, methyl), 1686 (carbonyl), 1563 (vinyl bonding).^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.07 (s, 6H, two methyls), 1.86 (m, 2H, methylene), 2.23 (s, 2H, methylene), 2.58 (s, 3H, methyl), 3.69 (s, 3H, methoxy), 6.05 (s, 1H, pyran H 4), 7.24-7.58 (m, 14H, three phenyl), 8.31, 8.38, 8.53 (3s, 3H, D_2O exchangeable, three imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyl), 39.7, 42.6 ($2CH_2$), 38.8 (methyl), 50.6 (methoxy), 120.1, 120.4, 121.3, 121.7, 122.4, 123.6, 123.2, 124.5, 124.3, 124.9, 125.4, 125.6 (three phenyls), 130.1, 133.3, 136.5, 138.2 (pyran C-2, C-3, C-5, C-6), 166.5 (carbonyl). Anal. cacld for $C_{32}H_{34}N_4O_2$ (522.65): C, 73.54; H, 6.56; N, 10.72%. Found: C, 75.48; H, 6.48; N, 10.93%. MS: *m/z* = 522 M^+ (75%).

4-(4-Chlorophenyl)-5-hydrazineylidene-2,7,7-trimethyl-5,6,7,8-tetrahydro-4H-chromene-3-carbohydrazide ***(9e)***. Yellow crystals from EtOH, m.p. 108-110 ºC, yield: 2.31 g (62%), IR (ν, cm^{-1}): 3475-3331 (imino), 3053 (CH-aromatic), 2985, 2878 (methylene, methyl), 1688 (carbonyl), 1560 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.09 (s, 6H, two methyls), 1.83 (m, 2H, methylene), 2.24 (s, 2H, methylene), 2.61 (s, 3H, methyl), 4.87, 5.18 (2s, 4H, D_2O exchangeable, amino), 6.05 (s, 1H, pyran H 4), 7.23-7.62 (m, 4H, phenyl), 8.38 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.5 (two methyls), 39.5, 42.6 (two methylenes), 38.9 (methyl), 120.8, 123.1, 124.2, 125.7 (phenyl), 130.1, 133.6, 136.5, 138.3 (pyran C-2, C-3, C-5, C-6), 165.8 (carbonyl). Anal. cacld for $C_{19}H_{23}ClN_4O_2$ (374.87): C, 60.88; H, 6.18; N, 14.95%. Found: C, 60.94; H, 6.03; N, 15.19%. MS: *m/z* = 374, 376 M^+, M^{+2}(70%).

4-(4-Chlorophenyl)-2,7,7-trimethyl-N'-phenyl-5-(2-phenylhydrazineylidene)-5,6,7,8-tetrahydro-4H-chromene-3-carbohydrazide ***(9f)***. Pale yellow crystals from 1,4-dioxane, m.p. 105-107 ºC, yield: 3.31 g (62%), IR (ν, cm^{-1}): 3489-3340 (imino), 3055 (CH-aromatic), 2986, 2873 (methylene, methyl), 1687 (carbonyl), 1562 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.01, 1.06 (s, 6H, two methyls), 1.85 (m, 2H, methylene), 2.29 (s, 2H, methylene), 2.56 (s, 3H, methyl), 6.07 (s, 1H, pyran H 4), 7.25-7.63 (m, 14H, three phenyls), 8.26, 8.38, 8.52(3s, 3H, D_2O exchangeable, three imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.8 (two methyl), 39.5, 42.9 (two methylenes), 38.7 (methyl), 120.4, 120.8, 121.5, 122.3, 122.6, 123.2, 123.7, 124.2, 124.7, 124.9, 125.2, 125.9 (three phenyls), 130.4, 133.6, 136.2, 138.0 (pyran C-2, C-3, C-5, C-6), 166.6 (carbonyl). Anal. cacld for $C_{31}H_{31}ClN_4O_2$ (527.06): C, 70.64; H, 5.93; N, 10.63%. Found: C, 70.74; H, 6.15; N, 10.80%. MS: *m/z* = 526, 528 M^+, M^{+2} (70%).

General procedure for the synthesis of thieno[3,2-f]chromene-8-carboxylate derivatives ***11a-f***

Elemental sulfur (0.32 g, 0.01 mol) and either dicyanomethane (0.66 g, 0.01 mol) or ethyl 2-cyanoacetate (1.07 g, 0.01 mol) were added to a solution of either **4a** (3.40 g, 0.01 mol), **4b** (3.70 g, 0.01 mol) or **4c** (3.74 g, 0.01 mol) in *p*-dioxane (40 mL) containing triethylamine (2.0 mL). The whole reaction mixture was heated under the reflux conditions for 2 h then the working up was carried out in a similar manner previously described for the synthesis of **4a-c**.

Ethyl 2-amino-1-cyano-4,4,7-trimethyl-9-phenyl-5,9-dihydro-4H-thieno[3,2-f]chromene-8-carboxylate ***(11a)***. Orange crystals from AcOH, m.p. 138-140 ºC, yield: 2.73 g (65%), IR (ν, cm^{-1}): 3481-3324 (amino), 3054 (CH-aromatic), 2986, 2873 (methylene, methyl), 2220 (cyano), 1689 (carbonyl), 1565 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.05 (s, 6H, two methyls), 1.13 (t, 3H, J = 7.20 Hz, ester methyl), 2.26 (s, 2H, methylene), 2.53 (s, 3H, methyl), 4.22 (q, 2H, J = 7.20 Hz, ester methylene), 4.89 (s, 2H, D_2O exchangeable, amino), 6.06 (s, 1H, pyran H 4), 7.26-7.55 (m, 5H, phenyl). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.6 (ester methyl), 24.6 (two methyls), 42.8 (methylene), 50.2 (ester methylene), 38.7 (methyl), 116.9 (cyano), 121.1, 122.5, 123.3, 125.4 (phenyl), 130.2, 132.6, 133.5, 137.7, 138.2, 139.7, 140.2, 142.5 (pyran C-2, C-3, C-5, C-6, thiophene C), 166.6 (carbonyl). Anal. cacld for $C_{24}H_{24}N_2O_3S$ (420.53): C, 68.55; H, 5.75; N, 6.66; S, 7.62%. Found: C, 68.73; H, 5.82; N, 6.75; S, 7.53%. MS: *m/z* = 420 M^+ (79%).

Diethyl 2-amino-4,4,7-trimethyl-9-phenyl-5,9-dihydro-4H-thieno[3,2-f]chromene-1,8-dicarboxylate ***(11b)***. Orange crystals from AcOH, m.p. 172-174 ºC, yield: 2.33 g (50%), IR (ν, cm^{-1}): 3469-3328 (amino), 3054 (CH-aromatic), 2984, 2876 (methylene, methyl), 1689,1688 (two carbonyls), 1565 (vinyl bonding).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.07 (s, 6H, two methyls), 1.12, 1.30 (2t, 6H, J = 7.16, 5.49Hz, two ester methyls), 2.26 (s, 2H, methylene), 2.53 (s, 3H, methyl), 4.20, 4.22 (2q, 4H, J = 7.16, 5.49 Hz, two ester methylenes), 4.85 (s, 2H, D_2O exchangeable, amino), 6.06 (s, 1H, pyran H 4), 7.24-7.57 (m, 5H, phenyl). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.4, 16.6 (two ester methyl), 24.6 (two methyls), 42.8 (methylene), 38.7 (methyl), 50.2, 50.4 (two ester methylenes), 121.2, 122.3, 123.6, 125.1 (phenyl), 130.4, 132.6, 133.5, 137.5, 138.1, 139.7, 140.2, 142.7 (pyran C-2, C-3, C-5, C-6, thiophene C), 166.3, 166.3 (two carbonyls). Anal. cacld for $C_{26}H_{29}NO_5S$ (467.58): C, 66.79; H, 6.25; N, 3.00; S, 6.86%. Found: C, 66.84; H, 6.41; N, 3.29; S, 7.12%. MS: m/z = 467M$^+$ (79%).

*Ethyl 2-amino-1-cyano-9-(4-methoxyphenyl)-4,4,7-trimethyl-5,9-dihydro-4H-thieno[3,2-f]chromene-8-carboxylate (**11c**)*. Orange crystals from AcOH, m.p. 201-203 ºC, yield: 2.83 g (62%), IR (ν, cm^{-1}): 3496-3331 (amino), 3056 (CH-aromatic), 2989, 2876 (methylene, methyl), 2222 (cyano), 1687 (carbonyl), 1562 (vinyl bonding). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.08 (s, 6H, two methyls), 1.12 (t, 3H, J = 6.88 Hz, ester methyl), 2.26 (s, 2H, methylene), 2.56 (s, 3H, methyl), 3.70 (s, 3H, methoxy), 4.23 (q, 2H, J = 6.88 Hz, ester methylene), 4.87 (s, 2H, D_2O exchangeable, amino), 6.05 (s, 1H, pyran H-4), 7.24-7.62 (m, 4H, phenyl). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.3 (ester methyl), 24.4 (two methyls), 42.9 (methylene), 38.9 (methyl), 50.2 (ester methylene), 50.6 (methoxy), 117.0 (cyano), 121.3, 122.6, 123.1, 125.8 (phenyl), 130.4, 132.8, 133.5, 137.4, 138.3, 139.3, 140.8, 142.6 (pyran C-2, C-3, C-5, C-6, thiophene C), 166.9 (carbonyl). Anal. cacld for $C_{25}H_{26}N_2O_4S$ (450.55): C, 66.65; H, 5.82; N, 6.22; S, 7.12%. Found: C, 66.80; H, 6.16; N, 6.42; S, 7.23%. MS: m/z = 450 M$^+$ (58%).

*Diethyl 2-amino-9-(4-methoxyphenyl)-4,4,7-trimethyl-5,9-dihydro-4H-thieno[3,2-f]chromene-1,8-dicarboxylate (**11d**)*. Orange crystals from AcOH, m.p. 196-198 ºC, yield: 2.63 g (55%), IR (ν, cm^{-1}): 3484-3317 (amino), 3056 (CH-aromatic), 2985, 2878 (methylene, methyl), 1689, 1688 (two carbonyls), 1567 (vinyl bonding). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.04, 1.08 (s, 6H, two methyls), 1.13, 1.31 (2t, 6H, J = 7.12, 6.53Hz, two ester methyls), 2.24 (s, 2H, CH_2), 2.53 (s, 3H, methyl), 3.73 (s, 3H, OCH_3), 4.21, 4.24 (2q, 4H, J = 7.12, 6.53Hz, two ester methylenes), 4.88 (s, 2H, D_2O exchangeable, amino), 6.07 (s, 1H, pyran H 4), 7.25-7.59 (m, 4H, phenyl). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.5, 16.8 (two ester methyls), 24.8 (two methyls), 42.8 (methylene), 38.6 (methyl), 50.3, 50.5 (two ester methylenes), 50.9 (methoxy), 121.0, 121.6, 123.8, 125.6 (phenyl), 130.1, 132.5, 133.5, 137.8, 138.3, 139.9, 140.2, 142.6 (pyran C-2, C-3, C-5, C-6, thiophene C), 165.4, 166.8 (two carbonyls). Anal. cacld for $C_{27}H_{31}NO_5S$ (497.61): C, 65.17; H, 6.28; N, 2.81; S, 6.44%. Found: C, 65.27; H, 6.36; N, 3.16; S, 7.32%. MS: m/z = 497M$^+$ (84%).

*Ethyl 2-amino-9-(4-chlorophenyl)-1-cyano-4,4,7-trimethyl-5,9-dihydro-4H-thieno[3,2-f]chromene-8-carboxylate (**11e**)*. Orange crystals from AcOH, m.p. 171-173 ºC, yield: 2.63 g (58%), IR (ν, cm^{-1}): 3496-3341 (amino), 3056 (CH-aromatic), 2983, 2878 (methylene, methyl), 2220 (cyano), 1688 (carbonyl), 1563 (vinyl bonding).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.06 (s, 6H, two methyls), 1.12 (t, 3H, J = 6.73 Hz, ester methyl), 2.28 (s, 2H, methylene), 2.55 (s, 3H, methyl), 4.22 (q, 2H, J = 6.73 Hz, ester methylene), 4.88 (s, 2H, D_2O exchangeable, NH_2), 6.08 (s, 1H, pyran H 4), 7.22-7.64 (m, 4H, phenyl). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.3 (ester methyl), 24.4 (two methyls), 42.6 (methylene), 50.2 (ester methylene), 38.7 (methyl), 90.8 (pyran C-4), 116.9 (cyano), 121.1, 122.5, 123.6, 125.6 (phenyl), 130.2, 132.8, 133.5, 137.4, 138.3, 139.7, 140.5, 142.2 (pyran C-2, C-3, C-5, C-6, thiophene C), 166.4 (carbonyl). Anal. cacld for $C_{24}H_{23}ClN_2O_3S$ (454.97): C, 63.36; H, 5.10; N, 6.16; S, 7.05%. Found: C, 63.47; H, 4.96; N, 6.25; S, 7.17 %. MS: m/z = 454, 456 M$^+$, M^{+2} (83%).

*Diethyl 2-amino-9-(4-chlorophenyl)-4,4,7-trimethyl-5,9-dihydro-4H-thieno[3,2-f]chromene-1,8-dicarboxylate (**11f**)*. Pale brown crystals from AcOH, m.p. 210-212 ºC, yield: 3.31 g (66%), IR

(ν, cm^{-1}): 3474-3322 (amino, imino), 3055 (CH-aromatic), 2985, 2878 (methylene, methyl), 1689, 1687 (two carbonyl), 1567 (vinyl bonding). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.06 (s, 6H, two methyl), 1.16, 1.34 (2t, 6H, J = 6.44, 6.03Hz, two ester methyls), 2.25 (s, 2H, methylene), 2.56 (s, 3H, methyl), 4.21, 4.24 (2q, 4H, J = 6.44, 6.03 Hz, two ester methyls), 4.88 (s, 2H, D_2O exchangeable, amino), 6.05 (s, 1H, pyran H 4), 7.23-7.64 (m, 4H, phenyl). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.5, 16.8 (two ester methyls), 24.6 (two methyls), 42.5 (methylene), 38.6 (methyl), 50.4, 50.7 (two ester methyls), 90.8 (pyran C-4), 121.3, 121.5, 123.7, 125.2 (phenyl), 130.6, 132.8, 133.8, 137.6, 138.2, 139.5, 140.3, 142.6 (pyran C-2, C-3, C-5, C-6, thiophene C), 165.9, 166.4 (two carbonyls). Anal. cacld for $C_{26}H_{28}ClNO_5S$ (502.02): C, 62.21; H, 5.62; N, 2.79; S, 6.39%. Found: C, 62.39; H, 5.71; N, 2.86; S, 6.50%. MS: m/z = 502, 504 M^+, M^{+2} (84%).

*General procedure for the synthesis of the chromeno[5,6-d]thiazole-8-carboxylate derivatives **13a-c***

Elemental sulphur (0.32 g, 0.01 mol) and phenylisothiocyanate (1.30 g, 0.01 mol) were added (1.08 g, 0.01 mol) to a solution of **4a** (3.40 g, 0.01 mol), **4b** (3.70 g, 0.01 mol) or **4c** (3.74 g, 0.01 mol) in 1,4-dioxane (40 mL) containing triethylamine (2 mL). The whole reaction mixture was heated under the reflux conditions for two hours then the working up was carried out in a similar manner as previously described for the synthesis of **4a-c**.

*Ethyl 4,4,7-trimethyl-1,9-diphenyl-2-thioxo-1,4,5,9-tetrahydro-2H-chromeno[5,6-d]thiazole-8-carboxylate (**13a**)*. Pale brown crystals from AcOH, m.p.180-182 ºC, yield: 3.31 g (66%), IR (ν, cm^{-1}): 3474-3322 (amino), 3055 (CH-aromatic), 2985, 2878 (methylene, methyl), 1689 (carbonyl), 1567 (vinyl bonding), 1207 (thiocarbonyl).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.06 (s, 6H, two methyls), 1.16 (t, 3H, J = 6.03 Hz, ester methyl), 2.25 (s, 2H, methylene), 2.56 (s, 3H, methyl), 4.21, 4.24 (q, 2H, J = 6.03 Hz, ester methylene), 6.05 (s, 1H, pyran H-4), 7.23-7.64 (m, 10H, two phenyls). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.2 (ester methyl), 24.6 (two methyls), 42.5 (methylene), 38.6 (methyl), 50.4 (ester methylene), 90.8 (pyran C-4), 121.3, 121.5, 122.1, 122.5, 122.8, 123.7, 124.2, 125.2 (two phenyls), 130.6, 132.8, 137.6, 139.5, 140.3, 142.6 (pyran C-2, C-3, C-5, C-6, thaizole C), 165.9, 166.4 (carbonyl), 180.2 (thiocarbonyl). Anal. cacld for $C_{28}H_{27}NO_3S_2$ (489.65): C, 68.68; H, 5.56; N, 2.86; S, 13.10%. Found: C, 68.72; H, 5.64; N, 2.72; S, 13.23%. MS: m/z = 489 M^+ (78%).

*Ethyl 9-(4-methoxyphenyl)-4,4,7-trimethyl-1-phenyl-2-thioxo-1,4,5,9-tetrahydro-2H-chromeno [5,6-d]thiazole-8-carboxylate (**13b**)*. Pale orange crystals from EtOH, m.p. 168-170 ºC, yield: 3.01 g (58%), IR (ν,cm^{-1}): 3055 (CH-aromatic), 2985, 2878 (methylene, methyl), 1689 (carbonyl), 1567 (vinyl bonding), 1205 (thiocarbonyl).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.08 (s, 6H, $2CH_3$), 1.16 (t, 3H, J = 5.84 Hz, ester methyl), 2.24 (s, 2H, methylene), 2.58 (s, 3H, methyl), 4.23 (q, 2H, J = 5.84Hz, ester methylene), 3.69 (s, 3H, methoxy), 6.05 (s, 1H, pyran H 4), 7.25-7.54 (m, 9H, two phenyls). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.5 (ester methyl), 24.3 (two methyls), 42.5 (methylene), 38.8 (methyl), 50.1 (ester methylene), 50.6 (methoxy), 90.5 (pyran C-4), 120.3, 120.5, 121.1, 121.4, 122.0, 122.7, 123.8, 125.4 (two phenyls), 130.8, 133.5, 137.6, 138.7, 140.3, 142.2 (pyran C-2, C-3, C-5, C-6 and thiazole C), 166.3 (carbonyl), 180.3 (thiocarbonyl). Anal. cacld for $C_{29}H_{29}NO_4S_2$ (519.67): C, 67.03; H, 5.63; N, 2.70; S, 12.34%. Found: C, 66.84; H, 5.73; N, 2.65; S, 12.52%. MS: m/z = 519 M^+ (68%).

*Ethyl 9-(4-chlorophenyl)-4,4,7-trimethyl-1-phenyl-2-thioxo-1,4,5,9-tetrahydro-2H-chromeno [5,6-d]thiazole-8-carboxylate (**13c**)*. Pale brown crystals from AcOH, m.p. 220-222 ºC, yield: 2.88 g (55%), IR (ν, cm^{-1}): 3055 (CH-aromatic), 2985, 2878 (methyl, methylene), 1688 (carbonyl), 1567 (vinyl bonding), 1209 (thiocarbonyl).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.07 (s, 6H, two methyls), 1.13 (t, 3H, J = 6.44 Hz, ester methyl), 2.25 (s, 2H, methylene), 2.58 (s, 3H, methyl), 4.22 (q, 2H, J = 6.44Hz, ester methylene), 6.06 (s, 1H, pyran H-4), 7.23-7.68 (m, 9H, two phenyls). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 16.5 (ester methyl), 24.6 (two methyls), 42.5 (methylene), 38.6 (methyl), 50.4 (ester methylene), 90.8 (pyran C-4), 120.2, 120.6, 121.3, 121.5, 122.3, 122.5, 123.7, 125.2 (two phenyls), 130.6, 132.8, 137.6, 139.5, 140.3, 142.6 (pyran C-2, C-3, C-5, C-6, thaizole C), 166.2 (carbonyl), 1801.3 (thiocarbonyl). Anal. cacld for $C_{28}H_{26}ClNO_3S_2$ (524.09): C, 64.17; H, 5.00; N, 2.67; S, 12.23%. Found: C, 64.33; H, 5.26; N, 2.75; S, 12.40%. MS: *m/z* = 524, 526 M^+, M^{+2} (60%).

*General procedure for the synthesis of the chromeno[5,6-d]thiazole-8-carboxamide derivatives **14a-f***

Either aminobenzene (0.93 g, 0.01 mol) or 1-amino 4-chlorobenzene (1.27 g, 0.01 mol) was added to a solution of either **13a** (4.89 g, 0.01 mol), **13b** (5.19 g, 0.0l mol) or **13c** (5.24 g, 0.01 mol) in dimethylformamide (40 mL). The whole reaction mixture was heated under the reflux conditions for two hours then poured onto ice/water mixture containing a few drops of HCl and the produced solid product was collected by filtration.

*4,4,7-Trimethyl-N,1,9-triphenyl-2-thioxo-1,3a,4,5,9,9b-hexahydro-2H-chromeno[5,6-d]thiazole-8-carboxamide (**14a**)*. Pale yellow crystals from EtOH, m.p. 196-198 ºC, yield: 3.16 g (60%), IR (ν, cm^{-1}): 3487-3330 (imino), 3053 (CH-aromatic), 2991, 2896 (methylene, methyl), 1688 (carbonyl), 1562 (vinyl bonding), 1205 (thiocarbonyl). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.07 (s, 6H, two methyls), 2.24 (s, 2H, methylene), 2.78 (s, 3H, methyl), 6.07 (s, 1H, pyran H 4), 7.23-7.56 (m, 15H, three phenyls), 8.34 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.5 (two methyls), 342.8 (methylene), 38.8 (methyl), 120.1, 120.5, 120.8, 121.4, 121.6, 122.1, 122.5, 123.0, 123.4, 123.6, 124.8, 125.5 (three phenyls), 130.3, 134.5, 136.2, 138.6, 138.3, 140.2 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 166.4 (carbonyl), 181.3 (thiocarbonyl). Anal. cacld for $C_{32}H_{28}N_2O_2S_2$ (536.71): C, 71.61; H, 5.26; N, 5.22; S, 11.95%. Found: C, 71.42; H, 5.30; N, 5.39; S, 11.73%. MS: *m/z* = 536 M^+ (75%).

*N-(4-Chlorophenyl)-4,4,7-trimethyl-1,9-diphenyl-2-thioxo-1,4,5,9-tetrahydro-2H-chromeno [5,6-d]thiazole-8-carboxamide (**14b**)*. Pale yellow crystals from *p*-dioxane, m.p. 175-177 ºC, yield: 3.31 g (58%), IR (ν, cm^{-1}): 3469-3327 (imino), 3055 (CH-aromatic), 2993, 2893 (methylene, methyl), 1687 (carbonyl), 1560 (vinyl bonding), 1207 (thiocarbonyl). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.08 (s, 6H, two methyls), 2.23 (s, 2H, methylene), 2.75 (s, 3H, methyl), 6.07 (s, 1H, pyran H-4), 7.24-7.63 (m, 14H, three phenyls), 8.34 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyls), 42.6 (methylene), 38.7 (methyl), 120.3, 120.8, 120.9, 121.6, 121.7, 122.1, 122.8, 123.4, 123.8, 124.3, 124.5, 125.9 (three phenyls), 130.2, 134.7, 136.2, 138.4, 139.1, 140.4 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 166.7 (carbonyl), 181.3 (thiocarbonyl). Anal. cacld for $C_{32}H_{27}ClN_2O_2S_2$ (571.12): C, 67.29; H, 4.77; N, 4.90; S, 11.23%. Found: C, 67.41; H, 4.54; N, 5.15; S, 11.46%. MS: *m/z* = 571, 573 M^+, M^{+2} (82%).

*9-(4-Methoxyphenyl)-4,4,7-trimethyl-N,1-diphenyl-2-thioxo-1,4,5,9-tetrahydro-2H-chromeno-[5,6-d]thiazole-8-carboxamide (**14c**)*. Pale yellow crystals from EtOH, m.p. 205-207 ºC, yield: 3.56 g (63%), IR (ν, cm^{-1}): 3463-3325 (imino), 3053 (CH-aromatic), 2991, 2896 (methlene, methyl), 1688 (carbonyl), 1562 (vinyl bonding), 1207 (thiocarbonyl). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.08 (s, 6H, two methyls), 2.26 (s, 2H, methylene), 2.77 (s, 3H, methyl), 3.59 (s, 3H, methoxy), 6.09 (s, 1H, pyran H- 4), 7.24-7.63 (m, 14H, three phenyls), 8.38 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyl), 42.6 (methylene), 38.8 (methyl), 50.7 (methoxy), 120.1, 120.5, 120.8, 121.4, 121.6, 122.3, 122.5, 122.9, 123.2, 123.7, 124.3, 125.5 (three phenyls), 130.2, 133.6, 135.7, 138.1, 138.3, 140.8 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 166.5 (carbonyl), 180.7 (thiocarbonyl). Anal. cacld for $C_{33}H_{30}N_2O_3S_2$

(566.73): C, 69.94; H, 5.34; N, 4.94; S, 11.31%. Found: C, 70.21; H, 5.45; N, 5.14; S, 11.28%. MS: *m/z* = 566M$^+$ (75%).

*N-(4-Chlorophenyl)-9-(4-methoxyphenyl)-4,4,7-trimethyl-1-phenyl-2-thioxo-1,4,5,9-tetrahydro-2H-chromeno[5,6-d]thiazole-8-carboxamide **(14d)***. Pale yellow crystals from EtOH, m.p. 165-167 ºC, yield: 2.88 g (48%), IR (ν, cm^{-1}): 3474-3331 (imino), 3053 (CH-aromatic), 2977, 2873 (methylene, methyl), 1688 (carbonyl), 1564 (vinyl bonding), 1205 (thiocarbonyl). ^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.06 (s, 6H, two methyls), 2.28 (s, 2H, methylene), 2.76 (s, 3H, methyl), 3.57 (s, 3H, methoxy), 6.06 (s, 1H, pyran H- 4), 7.26-7.65 (m, 13H, three phenyls), 8.37 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.4 (two methyls), 42.8 (methylene), 38.8 (methyl), 50.7 (methoxy), 120.2, 120.4, 120.8, 121.2, 121.8, 122.1, 122.7, 122.9, 123.5, 123.6, 124.6, 125.8 (three phenyls), 130.5, 133.8, 135.4, 138.2, 138.6, 140.5 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 166.3 (carbonyl), 180.5 (thiocarbonyl). Anal. cacld for $C_{33}H_{29}ClN_2O_3S_2$ (601.18): C, 65.93; H, 4.86; N, 4.66; S, 10.67%. Found: C, 65.73; H, 4.76; N, 4.72; S, 10.80%. MS: *m/z* = 601, 603 M$^+$, M^{+2} (86%).

*9-(4-Chlorophenyl)-4,4,7-trimethyl-N,1-diphenyl-2-thioxo-1,4,5,9-tetrahydro-2H-chromeno-[5,6-d]thiazole-8-carboxamide **(14e)***. Pale brown crystals from EtOH, m.p. 198-200 ºC, yield: 3.65 g (65%), IR (ν, cm^{-1}): 3469-3321 (imino), 3053 (CH-aromatic), 2987, 2863 (methylene, methyl), 1688 (carbonyl), 1564 (vinyl bonding), 1207 (thiocarbonyl).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.07 (s, 6H, two methyls), 2.27 (s, 2H, methylene), 2.78 (s, 3H, methyl), 6.06 (s, 1H, pyran H-4), 7.24-7.67 (m, 14H, three phenyls), 8.38 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.8 (two methyls), 42.6 (methylene), 38.8 (methyl), 120.1, 120.5, 121.1, 121.3, 121.7, 122.3, 122.5, 122.9, 123.5, 123.3, 124.4, 125.9 (three phenyls), 130.2, 133.7, 135.2, 138.2, 138.7, 140.8 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 166.6 (carbonyl), 180.3 (thiocarbonyl). Anal. cacld for $C_{32}H_{27}ClN_2O_2S_2$ (571.15): C, 67.29; H, 4.77; N, 4.90; S, 11.23%. Found: C, 67.31; H, 4.59; N, 4.83; S, 11.42%. MS: *m/z* = 571, 573 M$^+$. M^{+2} (75%).

*N,9-Bis(4-chlorophenyl)-4,4,7-trimethyl-1-phenyl-2-thioxo-1,4,5,9-tetrahydro-2H-chromeno-[5,6-d]thiazole-8-carboxamide **(14f)***. Pale brown crystals from EtOH, m.p. 179-181 ºC, yield: 3.32 g (55%), IR (ν, cm^{-1}): 3480-3343 (imino), 3056 (CH-aromatic), 2989, 2861 (methylene, methylene), 1687 (carbonyl), 1564 (vinyl bonding), 1205 (thiocarbonyl). ^{1}H NMR(DMSO-d_6, 300 MHz): δ = 1.03, 1.07 (s, 6H, two methyls), 2.25 (s, 2H, methylene), 2.74 (s, 3H, methyl), 6.05 (s, 1H, pyran H-4), 7.22-7.63 (m, 13H, three phenyl), 8.37 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyls), 42.4 (methylene), 38.8 (methyl), 120.1, 120.3, 121.0, 121.5, 122.1, 122.5, 122.8, 122.6, 123.7, 123.2, 124.5, 125.6 (three phenyls), 130.4, 133.4, 135.6, 138.5, 138.9, 140.6 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 166.7 (carbonyl), 180.5 (thiocarbonyl). Anal. cacld for $C_{32}H_{26}Cl_2N_2O_2S_2$ (605.59): C, 63.47; H, 4.33; N, 4.63; S, 10.59%. Found: C, 63.58; H, 4.71; N, 4.52; S, 10.63%. MS: *m/z* = 605, 607 M$^+$, M^{+2} (75%).

*General procedure for the synthesis of the 8-(1H-benzo[d]imidazol-2-yl)- 2H-chromeno[5,6-d]thiazole-2-thione derivatives **16a-c***

1,2-Diaminoaniline (1.08 g, 0.01 mol) was added to a solution of either **13a** (4.89 g, 0.01 mol), **13b** (5.19 g, 0.0l mol) or **13c** (5.24 g, 0.01 mol) in dimethylformamide (40 mL). The reaction mixture was heated under the reflux conditions for 3 h then poured onto ice/water mixture and the produced solid product was collected by filtration.

*8-(1H-Benzo[d]imidazol-2-yl)-4,4,7-trimethyl-1,9-diphenyl-1,3a,4,5,9,9b-hexahydro-2H-chromeno[5,6-d]thiazole-2-thione **(16a)***. Yellow crystals from EtOH, m.p. 205-207 ºC, yield: 3.53 g (66%), IR (ν, cm^{-1}): 3469-3335 (imino), 3053 (CH-aromatic), 2996, 2896 (methylene, methyl), 1560 (vinyl bonding), 1203 (thiocarbonyl).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.07 (s, 6H, two methyls), 2.21 (s, 2H, methylene), 2.79 (s, 3H, methyl), 6.06 (s, 1H, pyran H-4), 7.25-7.48 (m, 14H, three phenyls), 8.36 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyls), 42.9 (methylene), 38.9 (methyl), 120.2, 120.5, 121.4, 121.7, 121.9, 122.0, 122.6, 122.8, 123.3, 123.9, 124.3, 125.8 (three phenyls), 130.2, 133.6, 134.9, 138.2, 138.6, 140.8 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 172.3 (C=N), 180.3 (thiocarbonyl). Anal. cacld for $C_{32}H_{27}N_3OS_2$ (533.71): C, 72.02; H, 5.10; N, 7.87; S, 12.01%. Found: C, 71.28; H, 5.24; N, 7.93; S, 12.17%. MS: *m/z* = 533M$^+$ (80%).

*8-(1H-Benzo[d]imidazol-2-yl)-9-(4-methoxyphenyl)-4,4,7-trimethyl-1-phenyl-1,3a,4,5,9,9b-hexa hydro-2H-chromeno[5,6-d]thiazole-2-thione **(16b)***. Yellow crystals from p-dioxane, m.p. 182-184 ºC, yield: 2.87 g (50%), IR (ν, cm^{-1}): 3483-3328 (imino), 3056 (CH-aromatic), 2987, 2876 (methylene, methyl), 1560 (vinyl bonding), 1205 (thiocarbonyl).^1H NMR (DMSO-d_6, 300 MHz): δ = 1.05, 1.08 (s, 6H, two methyls), 2.25 (s, 2H, methylene), 2.68 (s, 3H, methyl), 3.73 (s, 3H, methoxy), 6.04 (s, 1H, pyran H-4), 7.23-7.52 (m, 13H, three phenyls), 8.34 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyls), 42.9 (two methylene), 38.9 (methyl), 50.3 (methoxy), 119.8, 120.3, 120.8, 121.4, 121.6, 122.2, 122.7 122.8, 123.1, 123.6, 124.3, 125.7 (three phenyls),130.4, 133.2, 135.3, 138.8, 139.2, 140.5 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 172.6 (C=N), 180.2 (thiocarbonyl). Anal. cacld for $C_{33}H_{29}N_3O_2S_2$ (563.73): C, 70.31; H, 5.19; N, 7.45; S, 11.37%. Found: C, 70.25; H, 5.24; N, 7.52; S, 11.50%. MS: *m/z* = 563M$^+$ (77%).

*8-(1H-Benzo[d]imidazol-2-yl)-9-(4-chlorophenyl)-4,4,7-trimethyl-1-phenyl-1,4,5,9-tetrahydro-2H-chromeno[5,6-d]thiazole-2-thione **(16c)***. Yellow crystals from p-dioxane, m.p. 148-150 ºC, yield: 2.84 g (50%), IR (ν, cm^{-1}): 3483-3362 (imino), 3055 (CH-aromatic), 2986, 2890 (methylene, methyl), 1563 (vinyl bonding), 1206 (C=S).^1H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.08 (s, 6H, two methyl), 2.28 (s, 2H, methylene), 2.77 (s, 3H, methyl), 6.08 (s, 1H, pyran H-4), 7.24-7.53 (m, 13H, three phenyls), 8.39 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.7 (two methyl), 43.5 (methylene), 38.9 (methyl), 120.1, 120.4, 120.8, 121.3, 121.7, 122.1, 122.4, 122.8, 123.1, 123.4, 124.2, 125.5 (three phenyls), 130.1, 133.6, 134.8, 138.1, 138.5, 140.8 (pyran C-2, C-3, C-5, C-6, thiazole C-4, C-5), 172.6 (C=N), 180.4 (thiocarbonyl). Anal. cacld for $C_{32}H_{26}ClN_3OS_2$ (568.15): C, 67.65; H, 4.61; N, 7.40; S, 11.29%. Found: C, 67.72; H, 4.81; N, 7.24; S, 11.40%. MS: *m/z* = 568, 570 M$^+$, M^{+2} (75%).

*General procedure for the synthesis of the xanthene derivatives **18a,b***

The same experimental procedure that was used for the synthesis of **4a-c** was carried out but using dimedone (1.40 g, 0.01), cyclohexan-1,3-dione (1.12 g, 0.01 mol) and 4-methoxybenzaldehyde (1.38 g, 0.01) or 4-chlorobenzaldehyde (1.40 g, 0.01 mol) instead of the reagents previously described for **4a-c**.

*9-(4-Methoxyphenyl)-3,3-dimethyl-3,4,5,6,7,9-hexahydro-1H-xanthene-1,8(2H)-dione **(18a)***. White crystals from EtOH, m.p. 110-112 ºC, yield: 2.60 g (74%), IR (ν, cm^{-1}): 3057 (CH-aromatic), 2994, 2893 (methylene, methyl), 1689, 1704 (two carbonyls), 1560 (vinyl bonding). ^1H NMR (DMSO-d_6, 300 MHz): δ = 1.02, 1.08 (s, 6H, two methyls), 1.85-1.95 (m, 6H, three methylenes), 2.21-2.43 (m, 4H, two methylenes), 3.69 (s, 3H, methoxy), 6.07 (s, 1H, pyran H-4), 7.27-7.53 (m, 4H, phenyl). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.6 (two methyls), 26.7, 30.4, 33.2, 35.6, 39.6, 42.9 (five methylenes), 50.8 (methoxy), 96.3 (pyran C-4), 120.8, 121.3, 123.3, 124.3 (phenyl), 130.1, 134.6, 136.3, 138.2 (pyran C-2, C-3, C-5, C-6), 165.5, 166.8 (two carbonyls). Anal. cacld for $C_{22}H_{24}O_4$ (352.43): C, 74.98; H, 6.86%. Found: C, 74.87; H, 6.64% MS: *m/z* = 352 M$^+$ (70%).

9-(4-Chlorophenyl)-3,3-dimethyl-3,4,5,6,7,9-hexahydro-1H-xanthene-1,8(2H)-dione ***(18b)***. White crystals from p-dioxane, m.p. 158-160 °C, yield: 2.73 g (75%), IR (ν, cm^{-1}): 3055 (CH-aromatic), 2994, 2893 (methylene, methyl), 1696, 1702 (two carbonyls), 1560 (vinyl bonding).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.06 (s, 6H, two methyls), 1.83-1.98 (m, 6H, two methylenes), 2.25-2.46 (m, 4H, two methylenes), 6.08 (s, 1H, pyran H-4), 7.27-7.56 (m, 4H, phenyl). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.8 (two methyls), 26.2, 30.3, 34.7, 35.8, 39.9, 42.8 (five methylenes), 96.8 (pyran C-4), 120.8, 122.8, 123.5, 124.6 (phenyl), 130.5, 134.3, 136.5, 138.6 (pyran C-2, C-3, C-5, C-6), 166.2, 166.7 (two carbonyls). Anal. cacld for $C_{21}H_{21}ClO_3$ (356.85): C, 70.68; H, 5.93%. Found: C, 70.83; H, 6.17%. MS: *m/z* = 356, 358 M^+, M^{+2} (70%).

General procedure for the synthesis of the acridine derivatives ***19a,b***

The same experimental procedure that was used for the synthesis of **18a,b** was carried out but using NH_4OAc (2.0 g) instead of Et_3N.

3,3-Dimethyl-9-phenyl-3,4,6,7,9,10-hexahydroacridine-1,8(2H,5H)-dione ***(19a)***. Pale yellow crystals from *p*-dioxane, m.p. 197-199 °C, yield: 2.18 g (68 %), IR (ν, cm^{-1}): 3054 (CH-aromatic), 2994, 2892 (CH_2, CH_3), 1697, 1703 (two carbonyls), 1563 (vinyl bonding).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.04, 1.07 (s, 6H, two methyls), 1.85-1.97 (m, 6H, three methylenes), 2.23-2.44 (m, 4H, two methylenes), 6.13 (s, 1H, pyridine H 4), 7.25-7.49 (m, 5H, phenyl), 8.40 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.8 (two methyls), 26.2, 30.3, 34.7, 35.8, 39.9, 42.8 (five methylenes), 98.3 (pyridine C-4), 120.8, 122.8, 123.5, 124.6 (phenyl), 130.5, 136.3, 138.5, 140.1 (pyridine C-2, C-3, C-5, C-6), 166.2, 166.7 (two carbonyls). Anal. cacld for $C_{21}H_{23}NO_2$ (321.42): C, 78.47; H, 7.21; N, 4.36%. Found: C, 78.62; H, 7.39; N, 4.52%. MS: *m/z* = 321 M^+ (70%).

9-(4-Methoxyphenyl)-3,3-dimethyl-3,4,6,7,9,10-hexahydroacridine-1,8(2H,5H)-dione ***(19b)***. Pale yellow crystals from p-dioxane, m.p. 166-168 °C, yield: 3.58 g (65%), IR (ν, cm^{-1}): 3056 (CH-aromatic), 2994, 2890 (methylene, methyl), 1695, 1702 (two carbonyls), 1561 (vinyl bonding).^{1}H NMR (DMSO-d_6, 300 MHz): δ = 1.03, 1.05 (s, 6H, two methyls), 1.87-1.95 (m, 6H, three methylenes), 2.23-2.46 (m, 4H, two methylenes), 3.72 (s, 3H, methoxy), 6.12 (s, 1H, pyridine H-4), 7.25-7.49 (m, 4H, phenyl), 8.40 (s, 1H, D_2O exchangeable, imino). ^{13}C NMR (DMSO-d_6, 75 MHz) δ: 24.5 (two methyl), 26.6, 30.4, 34.6, 35.8, 39.5, 42.7 (five methylenes), 98.6 (pyridine C-4), 120.5, 122.6, 123.8, 124.5 (phenyl), 130.3, 134.5, 136.2, 138.8 (pyridine C-2, C-3, C-5, C-6), 166.3, 166.7 (two carbonyls). Anal. cacld for $C_{22}H_{25}NO_3$ (351.45): C, 75.19; H, 7.17; N, 3.99%. Found: C, 75.38; H, 7.21; N, 4.25%. MS: *m/z* = 351 M^+ (78%).

Method of cell proliferation assay

The anti-proliferative activities of the newly synthesized compounds (Table 1) were evaluated against the six cancer cell lines A549, HT-29, MKN-45, U87MG, and SMMC-7721 and H460 using the standard MTT assay *in vitro*, with foretinib as the positive control. Supplemented with 10% fetal bovine serum (FBS) the cancer cell lines were cultured in minimum essential medium (MEM). To each well of 96-well plate and incubated in 5% CO_2 at 37 °C for 24 h approximate 4 x 103 cells, suspended in MEM medium, were plated. The cell cultures were continued for 72 h and the compounds tested at the indicated final concentrations were added to the culture medium. Fresh MTT was added to each well at a terminal concentration of 5 □g/mL, and incubated with cells at 37 °C for 4 h. With an ELISA reader, the formazan crystals were dissolved in 100 □L of DMSO each well, and the absorbency at 492 nM (for absorbance of MTT formazan) and 630 nM (for the reference wavelength) was measured. The results expressed as IC_{50} (inhibitory concentration 50 %) calculated by using the Bacus Laboratories Incorporated Slide Scanner (Bliss) software.

HTRF kinase assay

Materials. Foretinib has been utilized as a positive control for the HTRF kinase activity and results were expressed as IC_{50} (Table 1). By utilizing anibamine (as a reference drug) in the MTT assay the anti-proliferative action of novel heterocyclic compounds towards the human prostatic cancer PC-3 cell line were evaluated. With different concentration of these proteins namely, hemoglobin, lactoferrin, and lipocalin for 24 h the MTT assay was used to determine the cytotoxic activities. The surrounding DMEM medium was removed and 0.1 µg/mL of MTT treatment (MP Biomedical, USA) in the DMEM media for approximately 4 h was done in order to determine the cell viability. The temperature of 37 °C in 5% CO_2 incubator was maintained during the measurements. Compared with the untreated control samples the formazan crystals were further dissolved in the dissolving buffer and the absorbance of the same was read at 570 nm using an ELISA plate reader and the final readings were recorded. To assess cell viability the MTT assay has been widely been used and the enzymatic reduction of 3-[4,5-dimethylthiazole-2-yl]-2,5-diphenyltetrazolium bromide (MTT) to MTT-formazan was catalyzed by mitochondrial succinate dehydrogenase. Through the MTT assay there is a colorimetric reaction that can easily be measured from cell monolayers that have been plated in 35 mm dishes or multiwell plates. Cell cultures are incubated for 2 h in culture medium or in a Krebs–Hensleit–HEPES buffer (115 µM NaCl, 5 µM KCl, 1 µM KH_2PO_4, 1.2 µM $MgSO_4$, 2 µM $CaCl_2$, and 25 µM HEPES at pH 7.4) containing 0.5 µg ml^{-1} MTT. The incubation buffer is removed and the blue MTT–formazan product is extracted with acidified isopropyl alcohol (0.04 N HCl) after two hours. The absorbance of the formazan solution is read spectrophotometrically at 570 nm after 30 min extraction at room temperature.

CONCLUSION

The target molecules either chromene or quinoline derivatives were synthesized using dimedone. The produced compounds utilized for the synthesis of fused pyran, pyridine and thiazole derivatives. The anti-proliferative activities of the newly synthesized compounds were evaluated against selected six cancer cell lines. Further studies of the synthesized molecules toward tyrosine kinase c-Met and the cytotoxicity of the target molecules against the human prostatic cancer PC-3 were done. The cytotoxic effect of compounds **14f** and **16c** on A549 cell lines was scanned and showed high effects and these studies through this work supply the field for future studies.

REFERENCES

1. Pratap, R.; Ram, V.J. Natural and synthetic chromenes, fused chromenes, and versatility of dihydrobenzo[h]chromenes in organic synthesis. *Chem. Rev.* **2014**, 114, 10476–10526.
2. Masters, K.S.; Brase, S. Xanthones from fungi, lichens, and bacteria: the naturalproducts and their synthesis. *Chem. Rev.* **2012**, 112, 3717–3776.
3. Gebhardt, P.; Dornberger, K.; Gollmick, F.A.; Grafe, U.; Hartl, A.; Gorls, H.; Schlegel, B.; Hertweck, C. Quercinol, an anti-inflammatory chromene from the wood-rotting fungus Daedaleaquercina (Oak Mazegill). *Bioorg. Med. Chem. Lett.* **2007**, 17, 2558–2560.
4. Jang, K.H.; Lee, B.H.; Choi, B.W.; Lee, H.S.; Shin, J. Chromenes from the brown alga Sargassumsiliquastrum. *J. Nat. Prod.* **2005**, 68, 716–723.
5. Moore, B.P. Coumarin-like substances from Australian termites. *Nature* **1962**, 195, 1101–1102.
6. Nikpassand, M.; Fekri, L.; Badri, H.; Asadpour, L. Synthesis and antimicrobial activity of mono, bis and tris 2-amino-4*H* Chromenes. *Lett. Org. Chem.* **2015**, 12, 685–692.
7. Rao, N.K.; Rao, T.N.; Parvatamma, B.; Devi, K.P.; Setty, S.C. Multicomponentone pot synthesis and characterization of derivatives of 2-amino-7,7-dimethyl-5-oxo-4-phenyl-

5,6,7,8-tetrahydro-4*H*-chromene-3-carbonitrile and study of antimicrobial activity. *Bull. Chem. Soc. Ethiop.* **2018**, 32, 133–138.
8. Crops, F.; Stahmann, M.A.; Huebner, C.F.; Link, K.P. Studies on the hemorrhagic sweet clover disease: V. Identification and synthesis of the hemorrhagic agent. *J. Biol. Chem.* **1941**, 138, 513–527.
9. Cai, S.X.; Drewe, J.; Kemnitzer, W. Discovery of 4-aryl-4*H*-chromenes as potentapoptosis inducers using a cell- and caspase-based anti-cancer screening apoptosisprogram (ASAP): SAR studies and the identification of novel vascular disruptingagents. *Anti. Cancer Agents Med. Chem.* **2009**, 9, 437–456.
10. Patil, S.A.; Patil, R.; Pfeffer, L.M.; Miller, D.D. Chromenes: potential new chemotherapeutic agents for cancer. *Future Med. Chem.* **2013**, 5, 1647–1660.
11. Wood, D.L.; Panda, D.; Wiernicki, T.R.; Wilson, L.; Jordan, M.A.; Singh, J.P. Inhibition of mitosis and microtubule function through direct tubulin binding by a novel antiproliferative naphthopyran LY290181. *Mol. Pharmacol.* **1997**, 52, 437–444.
12. Kumar, D.; Sharma, P.; Singh, H.; Nepali, K.; Gupta, G.K.; Jain, S.K.; Ntie-Kang, F. The value of pyrans as anticancer scaffolds in medicinal chemistry. *RSC Adv.* **2017**, 7, 36977–36999.
13. Kumar, A.; Jaitak, V. Natural products as multidrug resistance modulators in cancer. *Eur. J. Med. Chem.* **2019**, 176, 268–291.
14. Clapp, A.; Murphy, A.I..; Ascherman, J.A..; Rohde, C.H. Mastectomy: Weighing the risks of delayed chemotherapy, radiotherapy, and hormonal therapy. *J. Plastic, Reconst. Aesthetic Surg.* **2023**, 89, doi: 10.1016/j.bjps.2023.11.040.
15. Zhao, W.; Ke, S.; Cai, X.; Zuo, Z.; Shi, W.; Qiu, H.; Cai, G.; Gong, Y.; Wu, Y.; Ruan, S.; Chen, Y. Radiotherapy plus camrelizumab and irinotecan for oligometastatic esophageal squamous cell carcinoma patients after first-line immunotherapy plus chemotherapy failure: An open-label, single-arm, phase II trial. *Radiother. Oncol* **2023**, 184, 109679.
16. Bray, F.; Ferlay, J.; Soerjomataram, I.; Siegel, R.L.; Torre, L.A.; Jemal, A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries, CA. *Cancer J. Clin.* **2018**, 68, 394–424.
17. Brana, M.F.; Cacho, M.; Gradillas, A.; de Pascual-Teresa, B.; Ramos, A. Intercalators as anticancer drugs. *Curr. Pharm. Des.* **2005**, 7, 1745–1780.
18. Avendano, C.; Menendez, J.C. *Medicinal Chemistry of Anticancer Drugs*, 2nd ed., Elsevier Science: Amsterdam; **2015**.
19. Agudelo, D.; Bourassa, P.; Berube, G.; Tajmir-Riahi, H.A.A. Review on the binding of anticancer drug doxorubicin with DNA and RNA: structural models and antitumor activity. *J. Photochem. Photobiol. B Biol.* **2016**, 158, 274–279.
20. Ross, W.E.; Bradley, M.O. DNA double-strand breaks in mammalian cells after exposure to intercalating agents, BBA Sect. *Nucleic Acids Protein Synth.* **1981**, 654, 129–134.
21. D'Incalci, M.; Sessa, C. DNA minor groove binding ligands: a new class of anticancer agents. *Expet. Opin. Invest. Drugs* **1997**, 6, 875–884.
22. Sibirtsev, V.S.; Garabadzhiu, A.V. Spectral study of the interaction of DNA with benzothiazolyl-benz-α-chromene. *Biochem.* **2007**, 72, 901–909.
23. Dehkordi, M.F.; Dehghan, G.; Mahdavi, M.; Feizi, M.A.H. DNA binding study of dihydropyrano[3, 4-*c*]chromene derivative by some spectroscopic techniques. *J. Rep. Pharm. Sci.* **2016**, 5, 80–88.
24. Hać, A.; Milaş, D.; Dziubek, F.; Łomeć, M.; Antosiewicz, A.H.; Żamojć, K. Searching for relationships between the structure of selected simple coumarins, their antioxidant capacity and anticancer properties. *J. Mol. Struct.* **2023**, 1294, 136477.
25. Jeleń, M.; Młodawska, B.M.; Korlacki, R. Anticancer activities of tetra-, penta-, and hexacyclic phenothiazines modified with quinoline moiety. *J. Mol. Struct.* **2023**, 1287, 135700.
26. Walle, T.V.; Cools, L.; Mangelinckx, S.; D'hooghe, M. Recent contributions of quinolines to antimalarial and anticancer drug discovery research. *Eur. J. Med. Chem.* **2021**, 226, 113865.

27. Zhang, Y.P.; He, Q.; Zhou, X.H.; Liu, G.H.; Yue, A.Q.; Gao, C.Y.; Zhao, J.Z.; Du, W.J.; Yan, S.P. In situ reduction synthesis of quinoline-based copper(I) complexes: "Self-activating" chemical nuclease, antioxidation and anticancer activity. *J. Mol. Struct.* **2023**, 1292, 136090.
28. Mohareb, R.M.; Helal, M.E.; Mayhoub, A.E.; Abdallah, A.E. Multi-component synthesis of pyrazolo[1,5-a]quinoline, thiazole and thiophene derivatives as cytotoxic agents. *Bull. Chem. Soc. Ethiop.* **2023**, 37, 1251–1538.
29. Mohareb, R.M.; Helal, M.H.E.; Mayhoub, A.E.; Abdallah, A.E.M. Multi-component synthesis of pyrazlo[1,5-*a*]quinoline, thiazole and thiophene derivatives as cytotoxic agents. *Bull. Chem. Soc. Ethiop.* **2023**, 37, 1251–1538.
30. Alwan, E.S.; Mohareb, R.M. Synthesis of biologically active xanthenes, chromene, thiazole, thiophene, pyrazole and isoxazole derivatives from camphor. *Bull. Chem. Soc. Ethiop*. **2023**, 37, 1539–1552.
31. Mohareb, R.M.; Abdallah, A.E.M. New approaches for the synthesis of pyrazole, thiophene, thieno[2,3-b]pyridine, and thiazole derivatives together with their anti-tumor evaluations. *Med. Chem. Res.* **2014**, 23, 564–579.
32. Mohareb, R.M.; Wardakhan, W.W.; Hamid, F.I. Synthesis and cytotoxicity of fused thiophene and pyrazole derivatives derived from 2-N-acetyl-3-cyano-4,5,6,7-tetrahydrobenzo-[*b*]thiophene. *Med. Chem. Res.* **2015**, 24, 2043–2054
33. Mohareb, R.M.; Zaki M.Y.; Abas, N.S. Synthesis, anti-inflammatory and anti-ulcer evaluations of thiazole, thiophene, pyridine and pyran derivatives derived from androstenedione. *Steroids* **2015**, 98, 80–91.
34. Liu, L.; Siegmund, A.; Xi, N.; Kaplan-Lefko, P.; Rex, K.; Chen, A.; Lin, J.; Moriguchi, J.; Berry, L.; Huang, L.Y.; Teffera,Y.; Yang, Y.J.; Zhang,Y.H.; Bellon, S.F.; Lee, M.; Shimanovich, R.; Bak, A.; Dominguez, C.; Norman, M.H.; Harmange, J.C.; Dussault,I.; Kim, T.S. Discovery of a potent, selective, and orallybioavailable c-Met inhibitor: 1-(2-hydroxy-2-methylpropyl)-*N*-(5-(7-methoxyquinolin-4-yloxy)pyridin-2-yl)-5-methyl-3-oxo2-phenyl-2,3-dihydro-1*H*-pyrazole-4-carboxamide (AMG 458). *J. Med. Chem.* **2008**, 51, 3688–3691.
35. Peach, M.L.; Tan, N.; Tan, N.; Choyke, S.J.; Giubellino,A.; Athauda, G.; Burke, T.R.; Nicklaus, M.C.; Bottaro, D.P. Directed discovery of agents targeting the met tyrosine kinase domain by virtual screening. *J. Med. Chem.* **2009**, 52, 943–951.
36. Bacco, F.D.; Luraghi, P.; Medico, E.; Reato, G.; Girolami, F.; Perera, T.; Gabriele, P.; Comoglio, P.M.; Boccaccio, C. *J. Natl. Cancer Inst.* **2011**, 103, 645–661.
37. Knudsen, B.S.; Gmyrek, G.A.; Inra, J.; Scherr, D.S.; Vaughan, E.D.; Nanus, D.M.; Kattan, M.W.; Gerald, W.L.; Woude, G.F. High expression of the Met receptor in prostate cancermetastasis to bone. *Urology* **2002**, 60, 1113–1117.
38. Humphrey, P.A.; Zhu, X.; Zarnegar, R.; Swanson, P.E.; Ratliff, T.L.; Vollmer, R.T.; Day, M.L. Hepatocyte growth factor and its receptor (c-MET) in prostatic carcinoma. *Am. J. Pathol.* **1995**, 147, 386–396.
39. Rubin, J.S.; Bottaro, D.P.; Aaronson, S.A. Hepatocyte growth factor/scatter factor and its receptor, the c-met proto-oncogene product. *Biochim. Biophys. Acta* **1993**, 1155, 357–371.
40. Zhao, R.; Cai, K.; Yang, J.J.; Zhou, Q.; Cao, W.; Xiang, J.; Shen, Y.H.; Cheng, L.L.; Zang, W.D.; Lin, Y.; Yuan, Y.Y.; Xu, W.; Tao, H.; Zhao, S.M.; Zhao, J.Y. Nuclear ATR lysine-tyrosylation protects against heart failure by activating DNA damage response. *Cell Reports* **2023**, 42, 112400.
41. Liu, L.; Simon, M.; Muggiolu, G.; Vilotte, F.; Antoine, M.; Caron, J.; Kantor, G.; Barberet, P.; Seznec, H.; Audoin, B. Changes in intra-nuclear mechanics in response to DNA damaging agents revealed by time-domain Brillouin micro-spectroscopy. *Photoacoustics* **2022**, 27, 100385.
42. Rathke, C.; Barckmann, B.; Burkhard, S.; Raja, S.J.; Roote, J.; Pohl, R.R. Distinct functions of Mst77F and protamines in nuclear shaping and chromatin condensation during *Drosophila spermiogenesis*. *Eur. J. Cell Biol.* **2010**, 89, 326–338.