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Unexpected Products of the Reaction of Cyanoacetylhydrazones of

Aryl/heteryl Ketones with Hydrazine: A New Route to Aryl/Heteryl Hydrazones, X-ray Structure, and *In vitro* Anti-proliferative Activity against NCI 60-cell Line Panel

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Abstract

A new unexpected synthetic non-catalytic method for the synthesis of novel heteryl hydrazones for basemodification of nucleoside analogs has been developed. Characterizations of the products have been performed using NMR spectroscopy and single crystal x-ray diffraction analysis. Further *in vitro* anti-proliferative potency of the compounds against NCI 60 cell lines has been estimated. The results indicate anti-cancer activity by the compounds against several of the cancer cell lines.

Keywords: Crystal x-ray; hydrazons; heteryl hydrazons; in vitro anti-proliferative activity.

1 Introduction

Hydrazone compounds are versatile materials with important applications including as intermediates in the development novel compounds. The compounds have wide-ranging impact in chemistry and bioscience [1-12]. With appropriate design, synthesis and understanding of their structure-activity relationship, a range of compounds with a diversity of desirable bio-activities can be developed. The materials possess a range of pharmacological and biological properties including antimicrobial, anti-tubercular, analgesic, anti-inflammatory, antiviral, antiplatelet, anticancer, antimalarial, cardioprotective, antihelmintic, anticonvulsant, antiprotozoal [13], antitrypanosomal [14], and antischistosomiasis activity [15]. Hydrazone compounds are associated to ketones and

aldehydes through replacing the carbonyl oxygen with the hydrazinylidene group to produce a category of structures with the formula, $R_1(R_2)C=NNH_2$ [16,

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17]. The compounds possess the (C=N) bond which is conjugated with the lone pair of electrons of the functional nitrogen atom [18]. Combination of the hydrazones with the other functional groups results in the generation of compounds with unique chemical & physical features [19]. In addition, the pharmacological properties may be enhanced by complexation with a metal [20].

Figure 1 shows some examples of anticancer agents comprising hydrazones. Potency against the HI-60 human promyelocytic leukemic cell line has been reported for acylhydrazones such as compound **1**

[21]. Compound 2 is reported to possess in vitro anti-cancer potency against the MCF-7 human breast cancer cell line [22]. The aryl hydrazone derivative
(3) is described to possess an IC₅₀ of 6.7 nM against MCF-7 & MDA-MB 231 breast cancer cell lines

[23]. Other hydrazone derivatives such as compound **4** have a tendency to act against the lung cancer cell line (A549) [24]. The anticancer potency of the thiazolohydrazides (**5**) against prostate cancer has been assessed [25], also acetylpyridine and benzoylpyridine derived hydrazones (**6**, **7**) have been reported as agents against brain tumor [26].

The utility of hydrazones offers an opportunity for improved treatment through site-specific drug release in areas such as tumor tissue. Many researchers are experimenting on ways of generating hydrazones more efficiently, including using heat and chemical catalysts [27]. The importance of these moieties is illustrated by their use in the synthesis of numerous compounds of medicinal interest [28-30]. We have lately reported diverse synthetic methods for the preparation of heterocycles using cyanohydrazones [31-33]. Several derivatives of these ring structures are considered significant as antimetabolites in most biochemical reactions [34-36]. Beside an extensive range of new applications of the functionalized DNA in the biochemistry, attachment of reactive functional groups to nucleic acids is desired for bio-conjugates or further transformations. The introduction of a hydrazide or hydrazone group has been accomplished and the modified DNA was utilized for click chemistry. The hydrazide or the hydrazine functional group is a very useful group as a result of its extraordinary and unambiguous reactivity with various reagents [37]. In the light of this information and in the continuing results of our former research into the synthesis of biologically active heterocyclic compounds [38-41], the present paper reports an efficient and novel way to synthesize a hydrazone derivative with potential to play an essential role as an intermediate in the generation of many compounds. Conceivable clinical applications of these compounds include as anti-inflammatory and anticancer agents, in the prevention of platelet aggregation, and as chelating agents.

2 Results & discussion

2.1 Synthesis

This approach of synthesizing compound **11a-f** varies from other previously synthetic procedures [42]. Upon reacting hydrazine hydrate with 2-cyano-*N*-

(aryl/heteroarylethylidene)acetohydrazide (8a-f), compound 11a-f was formed instead of the pyrazole derivative 12a-f. Significant hydrazinolysis of amide bonds occurs without any catalyst. The reaction proceeds under reflux to give hydrazone 11a-f as a result of cleavage of the amide bond. To our knowledge, this is the first report of production of 2-(1-(aryl ethylidene)hydrazine by this method. The product that was crystallized and characterized via the X-ray diffraction measurement to confirm the structure. The ¹H NMR spectrum of 11c showed CH₃ protons at δ 2.00 ppm and the free NH₂ protons appeared at δ 6.48 ppm.

2.2 Crystal structure

In the crystal, the molecule is essentially planar except for the amine and methyl hydrogen atoms (Figure 2). Neighbouring molecules interact through N-H...N hydrogen bonds with geometry (N2...N1=3.134(5)Å, and N2-H2N...N1=159(4)°) to form dimers. In the dimers, pairs of the hydrogen bonds related by inversion symmetry form rings with geometry $R_2^2(8)$ in graph set notation [43]. The dimers are linked to their neighbours by Br1...N2 contacts with a distance of 3.358(4) Å to form ribbons in the [101] direction.

2.3 In vitro Anti-proliferative activity

The *in vitro* anti-proliferative activity against NCI-60 cell line panel was estimated. The compounds were selected via the National Cancer Institute "NCI", NIH through the Developmental Therapeutic Program for the determination of the *in vitro* anti-proliferative activity. This screen uses human tumor cell lines, representing melanoma, lung, leukemia, ovary, colon, brain, kidney, prostate and breast cancers.

The service of the NCI screening prioritises structures having a mode of action behaves as drugs on the bases of the computer-aided design (CAD). The ability of the submitted structures to bring the assortment to the collection of the NCI small molecules is essential to select them for the program of screening.

The compounds were assigned NCI codes NSC D-839209, NSC D-839207, NSC D-832401, NSC D-839205 & NSC D-839208 signifying the chemo type of this study. It was estimated at initial 10 μ M one dose percent inhibition assay. The results's expression is represented as growth percent for the estimated compound on each cell line. The results are shown in figures (3-6) & table 1 indicate the lowest cell growth promotion for selected compounds.

The lowest cell growth promotion for compound 11a was against breast cancer T-47D (GP = 65.06%), renal cancer CAKI-1 (GP = 82.64%), CNS cancer SNB-75 (GP= 89.80%), melanoma UACC-62 (GP = 90.26%), and NCI-H522 of the non-small cell lung cancer (GP = 92.83 %). Also the lowest cell growth promotion for compound 11b was against breast cancer T-47D (GP = 82.48%), Renal cancer CAKI-1 (GP = 83.73%), non-small cell lung cancer HOP-62 (GP = 85.60 %), ovarian cancer SK-OV-3 (GP = 86.08%), CNS cancer SNB-75 (GP= 88.65%), and melanoma UACC-62 (GP = 90.79%). The lowest cell growth promotion for compound 11c was against leukemia HL-60(TB) cell line (GP = 89.92 %), and non-small cell lung cancer EKVX (GP = 94.48 %), colon cancer HCT-15 (GP = 98.40 %), CNS cancer SNB-19 (GP = 93.98 %), renal cancer UO-31 (GP = 91.59%), and breast cancer MCF7 (GP = 92.75%). The screening results thus show that hydrazone exhibited anti-cancer activity at 10 µM concentration against several of the cancer cell lines tested.

The lowest cell growth promotion for compound **11d** was against ovarian cancer cell line (GP = 72.47%), renal cancer (GP = 82.79%), breast cancer MDA-MB-231/ATCC (GP = 84.34%), CNS cancer SNB-19 (GP = 95.36%), non-small cell lung cancer HOP-62 (GP = 95.43%), & melanoma UACC-62 (GP = 98.53%).

Moreover, compound **11f** reveals the lowest cell growth promotion against breast cancer T-47D (GP = 70.32%), non-small cell lung cancer HOP-62 (GP =

82.54%), renal Cancer CAKI-1 (GP = 86.84%), ovarian cancer SK-OV-3 (GP = 89.75%), CNS cancer SNB-75 (GP = 91.07%), melanoma SK-MEL28 (GP = 94.95%).



Figure.1: Structures of anticancer agents comprising hydrazones



Scheme 1: Synthetic route for compound 11a-f



Figure 2: Molecule's ORTEP representation of compound 11c in the crystal

Table 1

Crystal data & structure refinement for compound 11c	
Formula	C8H9BrN2
Formula weight	213.08
Temperature	296(2) K
Wavelength	1.54184 Å
Crystal system	Monoclinic
Space group	P21/c
a	9.5991(6) Å
b	14.1926(9) Å
c	6.3214(4) Å
b	92.143(5)°
Volume Z	860.60(9) Å ³ 4
Density (calculated)	1.645 Mg/m ³
Absorption coefficient F(000)	5.982 mm ⁻¹ 424
Crystal size Theta range for data collection	0.309 x 0.132 x 0.132 mm ³ 4.610 to 76.052°.
Index ranges	$-11 \le h \le 12, -15 \le k \le 17, -7 \le l \le 7$
Reflections collected	3734
Independent reflections	1769 [R(int) = 0.0394]
Completeness to theta = 67.684°	99.8 %
Refinement method Data / restraints / parameters	Full-matrix least-squares on F ² 1769 / 0 / 109
Goodness-of-fit on F^{2}	1.120
Final R indices [I>2 σ (I)]	R1 = 0.0530, wR2 = 0.1647
R indices (all data)	R1 = 0.0639, wR2 = 0.1823
Largest diff. peak and hole	1.067 and -0.664 e.Å ⁻³

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Developmental Therapeutics Program		NSC: D-839209/1	Conc: 1.00E-5 Molar	Test Date: Aug 22, 2022	
One Dose Mean Graph		Experiment ID: 2208	Experiment ID: 22080547		
Panel/Cell Line	Growth Percent	Mean Growth	Percent - Growth Per	cent	
Leukemia		1		1 1	
CCRF-CEM	110.24	1 1		1 1	
K-562	126.07			1 1	
MOLT-4	122.08			1 1	
RPM1-8226	105.99			1 1	
Non-Small Cell Lung Cancer	100.13			1 1	
A549/ATCC	105.98				
EKVX	102.31				
HOP-62	95.80				
NCI-H228	117,10				
NCI-H23	98.51				
NCI-H322M	109.03				
NCI-H522	92.83				
Colon Cancer					
COLO 205	110.07			1 1	
HCC-2998	101.74		- F	1 1	
HCT-15	100.47		1-	1 1	
HT29	108.72		-	1 1	
KM12	100.25		-	1 1	
CNS Cancer	105.01				
SF-208	104.72				
SF-295	104.07			1 1	
SF-539	100.13		E	1 1	
SNB-75	89.80			1 1	
U251	99.24		-		
Melanoma	112.00		_	1 1	
MALME-3M	103.80			1 1	
M 1-4	104.45			1 1	
MDA-MB-435	101.34			1 1	
SK-MEL-28	99.55			1 1	
SK-MEL-5	102.47			1 1	
UACC-257	107.49	1 1	-	1 1	
Ovarian Cancer	90.26			1 1	
IGROV1	104.85			1 1	
OVCAR-3	115.92			1 1	
OVCAR-5	99.85			1 1	
OVCAR-8	103.26			1 1	
SK-OV-3	99.93		-	1 1	
786-0	101.16		-	1 1	
A498	96.95			1 1	
ACHN	109.51		-	1 1	
CAKI-1 BYE 202	105.44			1 1	
SN12C	96.04			1 1	
TK-10	101.00		-	1 1	
UO-31	99,92			1 1	
PC-3	140.58	1 1		1 1	
DU-145	109.25		-	1 1	
Breast Cancer	00.03				
MDA-MB-231/ATCC	91.55		an annual state of the state of		
HS 578T	105.99				
BT-549	110.65				
MDA-MB-468	108.72		-		
Mean	104.82	1 1			
Range	75.52	1 1		1 1	
	450	400 50			

Figure 3: The data of the anti-cancer screening displayed for compound 11a



Figure 4: The data of the anti-cancer screening displayed for compound 11b

UNEXPECTED PRODUCTS OF THE REACTION OF CYANOACETYLHYDRAZONES OF..



Figure 5: The data of the anti-cancer screening displayed for compound 11c



Figure 6: The data of the anti-cancer screening displayed for compound 11d

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One Dose Mean Graph Experiment ID: 2000547 Report Date: Sep 20. Pane ICCell Line Growth Percent Mean Growth Percent - Growth Percent Citted State 133.450 133.450 Non-Small Coll Line 133.450 133.450 Citted State 133.450 133.450 Model 133.450 140.750 Model 133.450 140.750 Model 133.450 140.750 Model 130.550 140.750 Model 130.550 140.750 Model 100.552 100.552 Model 100.553 100.552 <	Developmental Ther	apeutics Program	NSC: D-839208 / 1	Conc: 1.00E-5 Molar	Test Date: Aug 22, 2022
Panel Cell Line Growth Percent Mean Growth Percent - Growth Percent With as a company of the second of the	One Dose Mean Graph		Experiment ID: 2208	Report Date: Sep 26, 202	
Leukemia Cit. do Cit. B Kit.	Panel/Cell Line	Growth Percent	Mean Growth	Percent - Growth Per	cent
Current - Lindian 1334 650 Hereit - 1320 Hereit - 13200 Hereit - 13200 Hereit - 13200 Herei	Leukemia		1 1		
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OVCAR-3 OVCAR	lopov1	103.43	1 1		1 1
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ACHN ACHN RXF 303 BY12C BC 303 BT 303	A498	98.69	1 1	-	1 1
RXP1*103 107.96 SN12C 06.30 TU-13 106.67 Postate Cancer 101.47 Postate Cancer 102.46 Breast Cancer 102.47 Postate Cancer 00.40 Breast Cancer 00.43 MCF7 00.43 MCF7 00.43 MCF7 00.43 MCF7 00.43 MCF7 00.43 MCF7 00.43 MDA-MB-409 100.32 MDA-MB-409 104.75 Detta 34.4.43 Range 76.20	ACHN	95.56	1 1		1 1
Sh12C 18.30 TK-10 106.60 UO-31 101.47 PC-3 112.62 DU-145 112.62 MDA-MB-231/ATCC 00.40 BT-440 108.35 T-47D 70.32 MDA-MB-68 118.90 Delta 34.43 Range 75.20	CAKI-1 BYE 303	80.84	1 1		1 1
TK:10 108.80 Proch3 Cancer 101.47 Proch3 Cancer 134.76 Broth Corr 34.30 MCF7 03.43 MCF7 04.43 MCF7 05.32 MCF7 104.76 Detta 34.43 Range 76.20	SN12C	88.36	1 1		1 1
UO-31 101.47 Postata Cancer 134.78 DU-145 Presst Caoer 93.43 MDA-MB-231/ATCC 90.40 HS 678T 108.18 B 7.470 70.32 MDA-MB-468 119.90 Mean 104.76 Range 75.20	TK-10	105.60	1 1		1 1
Prostate Cancer Prostate Cancer DU-46 DU-	UO-31	101.47	1 1		1 1
DU346 Breast Cancer 1129 MCF7M -231/ATCC 9040 HS 576T 9040 BT-540 106.32 MDA-MB-458 106.32 MDA-MB-458 104.75 Deta 34.43 Range 76.20	Prostate Cancer		1 1		1 1
Breast Caroer 112.03 MGF7 03.43 MG54M1-231/ATCC 00.40 BT-540 108.35 T-470 70.32 MDA-MB-68 118.96 Mean 104.75 Delta 34.43 Range 75.20	DUL 145	134.76	1 1		1 1
MCF7.ms-231/ATCC 93.43 MCF7.ms-231/ATCC 98.19 HS 87540 106.35 BT 540 106.35 MDA.MB-408 104.76 Delta 34.43 Range 76.20	Breast Cancer	112.02			1 1
MDA-MB-231/ATCC 00.40 H0 576 T 470 108.15 T 470 170.32 MDA-MB-468 104.75 Delta 34.43 Range 75.20	MCF7	93.43	1 1		1 1
HS 578T 09.10 BT-540 106.32 T/D/D 118.96 MDA-MB-468 118.96 Mean 104.76 Detta 34.43 Range 76.20	MDA-MB-231/ATCC	90.40	1 1		1 1
B1-10 190.32 MDA-MB-409 118.90 Detta 34.43 Range 76.20	HS 578T	98.19	1 1		1 1
MDA-MB-409 118.06 Mean 104.75 Detta 34.43 Range 76.20	B1-049	108.35	1 1		1 1
Mean 104.75 Deta 34.43 Range 75.20	MDA-MB-468	118.96	1 1		1 1
Mean 104.75 Delta 34.45 Range 76.20	 An and a second sec second second sec		1 1		1 1
Range 75.20	Mean	104.75			1 1
	Range	75 20			
	country of				1 1
150 100 50 0 -50 -100 -1		150	100 50	0 -50	-100 -150

Figure 7: The data of the anti-cancer screening displayed for compound 11f

Table 2

Antitumor determintions of the compounds using human tumor cell lines at a dose of 10 $\mu M.$

Panel/Cell line					
	11a	11b	11c	11d	11f
Leukemia					
CCRF-CEM	116.24	131.48	98.74	118.05	133.45
HL-60(TB)	137.23	121.81	89.92	123.80	126.20
K-562	126.07	106.18	115.30	129.22	145.52
MOLT-4	122.08	132.88	103.21	137.01	126.70
RPMI-8226	105.99	112.48	108.60	116.54	111.44
SR	100.13	104.82	92.41	117.25	107.61

Non-Small Cell Lung Cancer

A549/ATCC	105.98	103.27	96.54	108.43	101.45	
EKVX	102.31	110.00	94.48	119.46	102.98	
HOP-62	95.80	85.60	111.98	95.43	82.54	
HOP-92	126.93	119.14	96.44	135.97	122.21	
NCI-H226	117.10	109.13	106.26	116.64	123.58	
NCI-H23	98.51	101.42	95.93	105.11	100.22	
NCI-H322M	109.03	111.05	104.61	114.35	108.32	
NCI-H460	105.27	102.10	105.94	107.35	105.68	
NCI-H522	92.83	98.13	101.41	102.05	95.19	
Colon Cancer						
COLO 205	116.67	108.27	119.55	115.79	112.93	
HCC-2998	101.74	106.48	106.30	104.94	106.20	
HCT-116	106.36	110.00	102.32	108.56	107.50	
HCT-15	100.47	102.61	98.40	104.72	101.51	
HT29	108.72	107.77	115.36	111.58	105.35	
KM12	100.25	101.93	110.22	110.40	102.34	
SW-620	105.01	100.53	101.96	101.10	108.72	
CNS Cancer						
SF-268	104.72	99.57	105.39	111.58	114.28	
SF-295	104.07	99.44	95.15	106.82	96.51	
SF-539	100.13	95.20	100.51	100.93	95.98	
SNB-19	99.61	97.09	93.98	95.36	94.97	
SNB-75	89.80	88.65	114.44	99.59	91.07	
U251	99.24	100.46	103.69	103.82	97.63	
Melanoma						
LOX IMVI	112.80	107.03	NT	106.32	106.36	
MALME-3M	103.80	100.09	102.06	106.89	106.92	
M14	104.45	100.37	110.25	107.72	102.90	
MDA-MB-435	101.34	101.06	105.36	104.74	103.17	
SK-MEL-2	109.90	115.25	109.59	115.64	108.00	
SK-MEL-28	99.55	95.86	104.46	103.77	94.95	

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	SK-MEL-5	102.47	103.01	101.36	104.64	102.01	
	UACC-257	107.49	102.96	111.42	106.60	106.38	
	UACC-62	90.26	90.79	101.32	98.53	95.49	
	Ovarian Cancer						
	IGROV1	104.85	107.69	106.44	106.16	103.43	
	OVCAR-3	115.92	101.96	117.98	111.90	117.24	
	OVCAR-4	98.85	101.45	112.72	103.48	99.28	
	OVCAR-5	99.71	99.17	111.17	99.86	102.08	
	OVCAR-8	103.26	103.23	105.37	104.21	102.21	
	NCI/ADR-RES	NT	NT	101.41	NT	NT	
	SK-OV-3	99.93	86.08	140.24	72.47	89.75	
	Renal Cancer						
	786-0	101.16	104.95	95.45	105.07	102.83	
	A498	96.95	98.79	NT	97.78	96.69	
	ACHN	109.51	105.88	110.28	105.55	95.56	
	CAKI-1	82.64	83.73	102.07	82.79	86.84	
	RXF 393	105.44	114.34	115.51	125.31	107.66	
	SN12C	96.04	88.60	100.24	91.44	88.36	
	TK-10	101.00	109.20	117.84	116.34	105.60	
	UO-31	99.92	91.87	91.59	107.69	101.47	
	Prostate Cancer						
	PC-3	140.58	137.16	100.71	151.80	134.76	
	DU-145	109.25	107.07	105.22	109.97	112.82	
	Breast Cancer						
	MCF7	98.03	96.76	92.75	107.68	93.43	
	MDA-MB- 231/ATCC	91.55	87.14	105.79	84.34	90.40	
	HS 578T	105.99	96.76	107.21	104.43	98.19	
	BT-549	119.65	125.77	102.78	128.99	108.35	
	T-47D	65.06	82.48	106.40	92.56	70.32	
(*NT: Not tested)	MDA-MB-468	108.72	116.03	110.38	123.92	118.96	

3 Experimental

3.1 Chemical Methods. Monitoring of the reaction progress was performed with visualization under Ultra violet light using TLC through pre-coated silica gel 60 F₂₄₅aluminium plates. The melting points were measured using a Stuart SMP30 and were uncorrected. The measurements of the spectral data of the synthesized compounds were performed in Cairo University, Ain Shams University, & National Research Centre, Egypt. The ¹H NMR spectra were measured on a Bruker Fourier 400 & 500 (at 400 & 500 MHz, respectively) at 300 K.

3.2. Synthesis & crystallization

3.2.1. General procedure for synthesizing compounds 10a-e:

The substituted cyanoacetohydrazides **10** were furnished upon react the 2-cyanoacetohydrazide **8** (0.01 mol) with the acetophenone derivatives **9** (0.01 mol) for 5-10 minutes under reflux in ethyl alcohol (20 mL). The precipitate was filtered, and then recrystallized using ethyl alcohol.

Compounds **10a** [44], **10e** [45] & **10f** [46, 47] were previously synthesized and reported in literature.

3.2.1.1. N'-(1-(4-bromophenyl)ethylidene)-2-cyanoacetohydrazide (10c)

Compound **10c** was afforded as a colorless crystals (93 %), 186-188 °C; ¹H-NMR (400 MHz, DMSO-*d*₆):

δ 2.26 (*s*, 3H, CH₃), 4.2 (s, 2H, CH₂), 7.61 (*d*, 2H, *J* = 16 Hz, 2CH), 7.75 (*d*, 2H, *J* =8 Hz, 2CH), 11.01 (*s*, 1H, NH). Analysis calculated for C₁₁H₁₀BrN₃O (280.12): C, 47.16; H, 3.60; Br, 28.52; N, 15.00. Found: C, 47.15; H, 3.60; Br, 28.51; N, 15.00.

3.2.1.2. 2-cyano-N'-(1-(2-

methoxyphenyl)ethylidene)acetohydrazide (10d)

Compound **10b** was afforded as colorless crystals (92 %), ¹H-NMR (500 MHz, DMSO- d_6): δ 2.13 (s, 3H,

CH₃), 3.77-3.87 (m, 3H, OCH₃), 4.07 (s, 2H, CH₂), 6.92-7.03 (*m*, 3H, 3CH), 7.33-7.35 (*m*, 1H, 1CH), 10.90 (*s*, 1H, NH). ¹³C-NMR (500 MHz, DMSO- *d*₆) δ (ppm): 18.39, 25.27, 56.07, 112.21, 116.65, 121.44, 129.83, 130.89, 131.44, 155.93, 156.04, 166.14. Analysis calculated for C₁₂H₁₃N₃O₂ (231.25): C, 62.33; H, 5.67; N, 18.17. Found: C, 62.31; H, 5.66; N, 18.15.

3.2.2. General procedure for synthesizing compounds 11a-e:

A mixture of the N-(1-(aryl/heteroaryl)ethylidene)-2-cyanoacetohydrazide (10) (0.01 mol), hydrazine hydrate (0.01 mol) was allowed to reflux for 3 h in ethyl alcohol (10 mL). Some solvent was allowed evaporate and the solid product was filtered and then re-crystallized utilizing ethyl alcohol.

3.2.2.1. 1-(1-phenylethylidene)hydrazine (11a)

Compound **11a** was afforded as a yellow crystals (81 %), 120-121° C;IR (KBr, cm⁻¹): υ 3054 (ArCH), 1567 (C=C). ¹H-NMR (400 MHz, DMSO-*d*₆): δ 2.2 (*s*, 3H, CH₃), 3.3 (*s*, 2H, NH₂), 7.45–7.48 (*d*, 3H, CH), 7.95–7.99 (*d*, 2H, CH). ¹³C-NMR (400 MHz, DMSO-d6) δ (ppm): 14.6, 126.4, 128.4, 129.7, 137.8, 157.2. Analysis calculated for C₈H₁₀N₂ (134.18): C, 71.61; H, 7.51; N, 20.88. Found: C, 71.59; H, 7.50; N, 20.87.

3.2.2.2. 2-(1-(3-aminophenyl)ethylidene)hydrazine (11b)

Compound **11b** was afforded as a yellow crystals (78 %), 85-86°C;¹H-NMR (400 MHz, DMSO-*d*₆): δ 1.9 (*s*, 3H, CH₃), 4.9 (*s*, 2H, NH₂), 6.3 (*s*, 2H, NH₂), 6.4 (*d*, 1H, CH), 6.8 (*d*, 1H, CH), 6.96-6.99 (*m*, 2H, 2CH). ¹³C-NMR (400 MHz, DMSO-d6) δ (ppm): 11.4, 110.5, 112.95, 112.98, 128.3, 140.4, 142.8, 148.2. Analysis calculated for C₈H₁₁N₃ (149.19): C, 64.40; H, 7.43; N, 28.16. Found: C, 64.40; H, 7.42; N, 28.15.

3.2.2.3. 2-(1-(4-bromophenyl)ethylidene)hydrazine (11c)

Compound **11c** was afforded as a buff crystals (70 %), 77-78° C;¹ H-NMR (400 MHz, DMSO-*d*6): δ 2.00 (*s*, 3H, CH₃), 6.48 (*s*, 2H, NH₂), 7.46–7.49 (*d*, 2H, 2CH), 7.57–7.55 (*d*, 2H, 2CH). ¹³C-NMR (400 MHz, DMSO-d6) δ (ppm): 11.1, 120.0, 126.7, 128.5, 130.9, 131.4, 139.0, 140.7. Analysis calculated for C₈H9BrN₂ (213.07): C, 45.09; H4.26; Br, 37.50; N, 13.15 %. Found: C, 45.09; H4.25; Br, 37.50; N, 13.14 %.

3.2.2.4. 1-(1-(2-

methoxyphenyl)ethylidene)hydrazine (11d)

Compound **11d** was afforded as a pink solid (76 %), >300° C;¹ H NMR (400 MHz, DMSO-*d*₆): δ not dissolved properly in the solvent. IR (KBr, cm⁻¹): υ 3171 (ArCH), 2343, 2050, 1980, 1542 (C=C), 1154 (C-O). Analysis calculated for C9H₁₂N₂O (164.2): C, 65.83; H, 7.37; N, 17.06. Found: C, 65.81; H, 7.36; N, 17.05.

3.2.2.5. 1-(1-(thiophen-2-yl)ethylidene)hydrazine (11e)

Compound **11e** was afforded as a yellow crystals (70 %), ¹H-NMR (400 MHz, DMSO-*d*₆): δ 2.03 (*s*, 3H, CH₃), 6.17 (*s*, 2H, NH₂), 6.96-6.98 (t, 1H, CH), 7.09-7.11 (d, 1H, CH), 7.29-7.30 (d,1H, CH). Analysis calculated for C6H₈N₂S (140.21): C, 51.40; H, 5.75; N, 19.98; S, 22.87%. Found: C, 51.40; H, 5.74; N, 19.97; S, 22.86%.

3.2.2.5. 1-(1-(pyridin-2-yl)ethylidene)hydrazine (11f)

Compound **11f** was afforded as a yellow crystals (54 %), >300°C;IR (KBr, cm⁻¹): υ 3240, 3125 (NH₂), 3023 (ArCH), 1533 (C=C). ¹H-NMR (400 MHz, DMSO*d*₆): δ 2.2 (*s*, 3H, CH₃), 5.9 (*s*, 2H, NH₂), 7.2 (*t*, 1H, CH), 7.68-7.69 (*t*, 1H, CH), 8.46-8.47 (d, 1H, 1CH), 9.34 (*d*, 1H, 1CH). ¹³C-NMR (400 MHz, DMSO-d6) δ (ppm): 10.1, 151.0, 151.2, 151.5, 167.1. Analysis calculated for C7H9N3 (135.17): C, 62.20; H, 6.71; N, 31.09. Found: C, 62.19; H, 6.70; N, 31.08.

3.3 Determination of the crystal structure

Collection of the Single crystal XRD data were performed on an Agilent SuperNova Dual Atlas diffractometer via a mirror mono-chromator utilizing Cu ($\lambda = 1.5418$ Å) radiation at ambient temperature. Utilizing SHELXS the crystal structure was solved

[48] and then refined utilizing SHELXL [49]. Refinement of the non-hydrogen atoms carried out with the anisotropic displacement parameters. In idealized positions the hydrogen atoms were inserted, and a riding model was utilized with Uiso set at 1.2 or 1.5 times the value of Ueq for the atom to which they are bonded. The crystal and the refinement data are accomplished in table 2. The deposition of the crystal structure has been performed in the Cambridge Structural Database under reference CCDC 2087302.

3.4 In vitro Anti-proliferative activity

Primary anti-cancer assays were performed consistent with the NCI's protocol [50-54]. The compound was added at single concentration and then the incubation of the cell culture was carried out for forty eight hours. Sulforhodamine B (SRB), a protein binding dye, was utilized to identify the endpoints (SRB). The compound's results are expressed as the percent growth of the treated cells comparable to the un-treated cells of the control (Figure 3). Range of growth (%) indicated the highest & the lowest growth that found for several cancer cell lines in refer to the sensitivity against the cell lines at the primary single high dose $(10^{-5}M)$.

4 Conclusions

2-(1-(Aryl/heteroaryl)ethylidene)hydrazines have been obtained by *non-catalytic* hydrazinolysis of amide. The reaction was performed using hydrazine monohydrate to cause amide bond-cleavage under reflux to yield the product. The compounds have been identified utilizing spectroscopic and single crystal X-ray diffraction measurements. Investigations of the *in vitro* anti-tumor activity of

the products have been performed. The results indicate that the compounds exhibit anticancer activity against a variety of the cancer cell lines.

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References

- [1] Zhu, D.; Lv, L.; Li, C. C.; Ung, S.; Gao, J.; Li, C. J. Umpolung of carbonyl groups as alkyl organometallic reagent surrogates for palladium-catalyzed allylic alkylation. Angew Chem Int. Ed. Engl. 2018; 57: 16520-16524.
- [2] Liu W, Twilton J, Wei B, Lee M, Hopkins MN, Bacsa J, Stahl SS, Davies HML. Copper-Catalyzed oxidation of hydrazones to diazo compounds using oxygen as the terminal oxidant. ACS Catal. 2021; 11(5): 2676–2683.
 [3] Rahim F, Zaman K, Taha M, Ullah H, Ghufran
- [3] Rahim F, Zaman K, Taha M, Ullah H, Ghufran M, Wadood A, Rehman W, Uddin N, Shah SAA, Sajid M, Nawaz F, Khan KM. Synthesis, in vitro alpha-glucosidase inhibitory potential of benzimidazole bearing bis-Schiff bases and their molecular docking study. Bioorg. Chem. 2020; 94: 103394.
- [4] Chuit C, Corriu RJ, Reye C, Young JC. Reactivity of penta- and hexacoordinate silicon compounds and their role as reaction intermediates. Chem Rev. 1993; 93(4): 1371-1448.
- [5] Elgemeie, GH, Alkhursani, SA, Mohamed, RA. New synthetic strategies for acyclic and cyclic pyrimidinethione nucleosides and their analogues. Nucleosides Nucleotides. 2019; 38: 12–87.
- [6] Mohamed-Ezzat RA, Elgemeie GH, Jones PG. Crystal structures of (*E*)-2-amino-4-methylsulfanyl-6-oxo-1-(1-phenylethylideneamino)-1,6-dihydro pyrimidine-5-carbonitrile and (*E*)-2amino-4-methylsulfanyl-6-oxo-1-[1-(pyridin-2yl)ethylideneamino]-1,6-dihydropyrimidine-5-

carbonitrile. Acta Cryst. 2021; E77: 547-550.

- [7] Elgemeie, GH, Salah, AM, Abbas, NS, Hussein, HA, Mohamed, RA, Pyrimidine non-nucleoside analogs: A direct synthesis of a novel class of *N*substituted amino and *N*-sulfonamide derivatives of pyrimidines, Nucleosides Nucleotides. 2017; 36: 213-223. https://doi.org/10.1080/15257770.2016.1257808.
- [8] Mohamady S, Kralt B, Samwel SK, Taylor SD. Efficient One-Pot, Two-Component Modular Synthesis of 3,5-disubstituted pyrazoles. ACS Omega 2018: 3(11): 15566–15574.
- [9] Coogan NT, Chimes MA, Raftery J, Mocilac P, Denecke MA. Regioselective synthesis of Vshaped Bistriazinyl-phenanthrolines. J. Org. Chem. 2015; 80(17): 8684-8693.
 [10] Tripathi RK, Ayyannan SR. Design, Synthesis,
- [10] Tripathi RK, Ayyannan SR. Design, Synthesis, and Evaluation of 2-Amino-6nitrobenzothiazole-derived hydrazones as MAO inhibitors: Role of the Methylene Spacer Group. ChemMedChem. 2016; 11(14): 1551-1567.
- [11] Souza LW, Squitieri RA, Dimirjian CA, Hodur BM, Nickerson LA, Penrod CN, Cordova J, Fettinger JC, Shaw JT. Enantioselective synthesis of indolines, benzodihydrothiophenes, and indanes by C–H insertion of Donor/Donor Carbenes. Angew. Chem. Int. Ed. 2018; 57(46): 15213-15216.
- [12] Ma XD, Yang SQ, Gu SX, He QQ, Chen FE, De Clercq E, Balzarini J, Pannecouque C. Synthesis and anti-HIV activity of aryl-2-[(4cyanophenyl)amino]-4-pyrimidinone hydrazones as potent non-nucleoside reverse transcriptase inhibitors. ChemMedChem. 2011; 6(12): 2225-2232.
- [13] Rollas S, Küçükgüzel SG. Biological activities of hydrazone derivatives. Molecules. 2007; 12: 1910-1939.
- [14] Narang R, Narasimhan B, Sharma S. A review on biological activities and chemical synthesis of hydrazide derivatives. Curr Med Chem. 2012; 19: 569-612.
- [15] Negi VJ, Sharma AK, Negi JS, Ra V. Biological activities of hydrazone derivatives in the new millennium. Int J Pharm Chem. 2012; 4: 100-109.
- [16] Uppal G, Bala S, Kamboj S, Saini M. Therapeutic review exploring antimicrobial potential of hydrazones as promising lead. Der Pharma Chem. 2011; 3: 250-268.
- [17] Verma G, Marella A, Shaquiquzzaman M, Akhtar M, Ali MR, Alam MM. A review exploring biological activities of hydrazones. J Pharm Bioallied Sci. 2014; 6(2): 69-80.
- [18] Corey EJ, Enders D. Applications of N, Ndimethylhydrazones to synthesis. Use in efficient, positionally and stereochemically selective C=C bond formation, oxidative hydrolysis of carbonyl compounds. Tetrahedron Lett. 1976; 17: 3-6.
- [19] Xavier AJ, Thakur M, Marie JM. Synthesis and spectral characterisation of hydrazone based 14membered octaaza macrocyclic Ni (II) complexes. J Chem Pharm Res. 2012; 4: 986-90.
- [20] Banerjee S, Mondal S, Chakraborty W, Sen S, Gachhui R, Butcher RJ, et al. Syntheses, X-ray crystal structures, DNA binding, oxidative cleavage and antimicrobial studies of two Cu (II)

hydrazone complees. Polyhedron. 2009; 28: 2785-2793.

- [21] Cui Z, Li Y, Ling Y, Huang J, Cui J, Wang R, et al. New class of potent antitumor acylhydrazone derivatives containing furan. Eur J Med Chem. 2010; 45: 5576-5584.
- [22] Al-Said MS, Bashandy MS, Al-Qasoumi SI, Ghorab MM. Anti-breast cancer activity of some novel 1,2-dihydropyridine, thiophene and thiazole derivatives. Eur J Med Chem. 2011; 46: 137-141.
- [23] Vogel S, Kaufmann D, Pojarová M, Müller C, Pfaller T, Kühne S, et al. Aroyl hydrazones of 2phenylindole-3-carbaldehydes as novel antimitotic agents. Bioorg Med Chem. 2008; 16: 6436-6447.
- [24] Zheng LW, Wu LL, Zhao BX, Dong WL, Miao JY. Synthesis of novel substituted pyrazole-5carbohydrazide hydrazone derivatives and discovery of a potent apoptosis inducer in A549 lung cancer cells. Bioorg Med Chem. 2009; 17: 1957-1962.
- [25] Gürsoy E, Güzeldemirci NU. Synthesis and primary cytotoxicity evaluation of new imidazo[2,1-b] thiazole derivatives. Eur J Med Chem. 2007; 42: 320-326.
- [26] Despaigne AA, Parrilha GL, Izidoro JB, da Costa PR, dos Santos RG, Piro OE, et al. 2-Acetylpyridine-and 2-benzoylpyridine-derived hydrazones and their gallium (III) complexes are highly cytotoxic to glioma cells. Eur J Med Chem. 2012; 50: 163-172.
- [27] Wahbeh J, Milkowski S. The Use of Hydrazones for Biomedical Applications. SLAS Technology 2019; 24(2): 161-168.
 [28] Kim J, Khoo J, Lee J, Shin W, Choi S. Process for the preparation of pyrrolo[2,3-c]pyridine derivatives or pharmaceutically acceptable salts thereof. PCT Int. Appl. 2014; WO 2014189238 A1 20141127.
- [29] Basawaraj R, Channamma M, Sangmeshwar W. Synthesis and antitubercular activity of 4thiazolidinone derivatives incorporating benzofuran moiety. Indian J. Heterocycl. Chem. 2014; 24(1): 59-66.
- [30] Chen F, Ma X, Gu S. Preparation of diaryl pyrimidinone hydrazone derivative for an antitumor and anti-HIV agents. Faming Zhuanli Shenqing 2011; CN 102153517 A 20110817.
- [31] Elgemeie, GH, Mohamed, RA, Hussein, HA, Jones, PG. Crystal structure of N-(2-amino-5cyano-4-methylsulfanyl-6-oxo-1,6dihydropyrimidin-1-yl)-4-bromobenzenesulfonamide dimethylformamide monosolvate, Acta Crystallogr E Crystallogr Commun. 2015; 71: 1322-1324. https://doi.org/10.1107/S2056989015018903.
- [32] Élgemeie ĞH, Salah, AM, Mohamed, RA, Jones, PG. Crystal structure of (*E*)-2-amino-4methylsulfanyl-6-oxo-1-{[(thiophen-2yl)methyl-idene]amino}-1,6-dihydropyrimidine-5-carbo-nitrile, Acta Crystallogr E Crystallogr Commun. 2015; 71: 1319–1321.
- [33] Azzam RA, Elgemeie, GH, Osman RR. Synthesis of novel pyrido[2,1-*b*]benzothiazole and *N*-substituted 2- pyridylbenzothiazole

derivatives showing remarkable fluorescence and biological activities. J. Mol. Structure. 2020; 1201: 127194.

- [34] Elgemeie, GH, Mohamed-Ezzat, RA. New Strategies Targeting Cancer Metabolism: Anticancer Drugs, Synthetic Analogues and Antitumor Agents. Edited by. Elgemeie, GH, Mohamed-Ezzat, RA. New Strategies Targeting Cancer Metabolism: Anticancer Drugs, Synthetic Analogues and Antitumor Agents, Elsevier, 2022, pp. 303-392, ISBN 9780128217832.
- [35] Elgemeie, GH, Mohamed-Ezzat, RA, Chapter 10 - Synthetic strategies for antimetabolite analogs in our laboratory, Editor(s): Elgemeie, GH, Mohamed-Ezzat, RA, New Strategies Targeting Cancer Metabolism, Elsevier, 2022, pp. 547-611, ISBN 9780128217832.
- [36] Mohamed-Ezzat RA, Kariuki, BM, Azzam RA. Morpholin-4-ium [5-cyano-6-(4-methylphenyl)-4-(morpholin-4-yl)pyrimidin-2-yl](phenylsulfonyl)amide. IUCrData 2022; 7 (11): x221033.
 [37] Raindlová V, Pohl R, Šanda M, Hocek M.
- [37] Raindlová V, Pohl R, Sanda M, Hocek M. Direct polymerase synthesis of reactive aldehyde-functionalized DNA and its conjugation and staining with hydrazines. Angew. Chem. 2010; 122: 1082-1084.
- [38] Metwally NH, Elgemeie GH, Jones PG. Crystal structure of ethyl 2-(3-amino-5-oxo-2-tosyl-2,5dihydro-1H-pyrazol-1-yl)acetate. Acta Cryst. 2021; E77: 615-617.
- [39] Elgemeie, GH, Mohamed RA. Microwave chemistry: Synthesis of purine and pyrimidine nucleosides using microwave radiation. J. Carbohydr. Chem. 2019; 38: 1-47.
- [40] Mohamed-Ezzat, RA, Kariuki, BM, Azzam, R A. Synthesis and crystal structure of *N*-(5-acetyl-4-methyl-pyrimidin-2-yl)benzene-sulfonamide. Acta Cryst. 2023; E79 (Pt 4): 331–334.
- [41] Metwally NM, Élgemeie GH, Jones PG. Crystal structure of 2-{[5-amino-1-(phenylsulfonyl)-1H_pyrazol-3-yl]oxy}-1-(4-methylphenyl)ethan-1-one. Acta Cryst. 2021; E77: 1054-1057.
- [42] Newkome GR, Fishel DL. Preparation of hydrazones: Acetophenone hydrazine. Org. Synth. 1970; 50: 102.
- [43] Bernstein J, Davis RE, Shimoni L, Chang NL. Patterns in hydrogen bonding: functionality and graph set analysis in crystals. Angew. Chem., Int. Ed. 1995; 34: 1555-1573.
- [44] Reinecke MG, Woodrow TA, Brown ES. Pyrazolo[3,4-c]pyridazines from hydrazine and aminothiophenecarboxylates. J. Org. Chem. 1992; 57 (3): 1018-1021.
- [45] Al-Awadi NA, Elnagdi MH, Mathew T, Abdel Khalik M. Pyrolysis of aminonitriles, cyanohydrazones, and cyanoacetamides. Part II. Elimination reactions of arylacetylhydrazone, arylcyanoacetylhydrazone, and substituted cyanoacetamides. Int. J. Chem. Kinet. 1996; 28(10): 749-754.
- [46] Khidre RE, El-Gogary SR, Mostafa MS. Design, synthesis, and antimicrobial evaluation of some novel pyridine, coumarin, and thiazole derivatives. J. Heterocycl. Chem. 2017; 54(4): 2511-2519.

- [47] Abdel-Wahab, BF, Abdelbasset FA, Awad GEA, El-Hiti, GA. Lett Drug Des Discov. 2017; 14(11): 1316-1323.
- [48] Sheldrick GM. A Short History of SHELX. Acta Crystallogr. A Found. Crystallogr. 2008; 64: 112–122.
- [49] Sheldrick GM. Crystal Structure Refinement with SHELXL. Acta Crystallogr. C Struct. Chem. 2015; 71: 3–8.
- [50] http://dtp.nci.nih.gov.
- [51] Boyd MR, Paull KD. Some practical considerations and applications of thenational cancer institute in vitro anticancer drug discovery screen, Drug Dev.Res. 1995; 34: 91-109.
- screen, Drug Dev.Res. 1995; 34: 91-109.
 [52] Monks A, Scudiero D, Skehan P, Shoemaker R, Paull K, Vistica D, Hose C, Langley J, Cronise P, Vaigro-Wolff A, Gray-Goodrich M, Campbell H, Mayo J, Boyd M. Feasibility of a high-flux anticancer drug screen using a diverse panel of cultured human tumor cell lines, J. Nat. Cancer Inst. 1991; 83(11): 757-766.
- [53] Shoemaker RH. The NCI60 human tumor cell line anticancer drug screen, Nat. Rev. Cancer. 2006; 6: 813-823.
- [54] Boyd MR, Teicher BA. (Ed.), Cancer Drug Discovery and Development, 2, Humana Press, 1997, pp. 23-43.