RESEARCH ARTICLE

Synthesis and Characterization of some new Formazan Derivatives from 2-Amino-4-Hydroxy-6-Methyl Pyrimidine and Study the Biological Activity (Anti-Bacteria and Anti-Cancer)

Abdullah Shakir, Shaimaa Adnan*

Department of Chemistry, College of Education, University of Al-Qadisiyah, Al Diwaniyah, Iraq

Received: 22th Dec, 2019; Revised: 24th Jan, 2020; Accepted: 17th Feb, 2020; Available Online: 25th Mar, 2020

ABSTRACT

This study involves a synthesis of some formazan derivatives starting from react chloro acetyl chlorid with 2-amino-4-hydroxy-6-methyl pyrimidine to gate compound (a), (a) react with hydrazine hydrate to give compound (b) also (b) react with 3-4-dimethoxy benzaldehyd to product Schiff base derivative (c) then (c) react with deferent amin derivatives to get formazan derivatives. All these compounds characterized by ^{13}C-NMR, fourier transform infrared spectroscopy (FTIR), ^{1}HMNR. After that, we study the biological activity for all formazan derivatives toward two different kinds of bacteria and anti-cancer.

Keywords: Azo, Formazan, Schiff bases.

International Journal of Pharmaceutical Quality Assurance (2020); DOI: 10.25258/ijpqa.11.1.8

How to cite this article: Shakir A, Adnan S. Synthesis and Characterization of Some New Formazan Derivatives From 2-Amino-4-Hydroxy-6-Methyl Pyrimidine and Study the Biological Activity (Anti-Bacteria and Anti-Cancer). International Journal of Pharmaceutical Quality Assurance. 2020;11(1):53-59.

Source of support: Nil

Conflict of interest: None

INTRODUCTION

Formazan derivatives are organic compounds containing four nitrogen atoms, General formula (-N=N-C=N-N-),[1] Formazan compounds are studied extensively because of applications.[2] It has report to possess wide effected of biological active, such as antiviral, antimicrobial, anti HIV,[3] anti-filamentary,[4] Anticancer,[5] Cyclic formazan derivatives are lithium-specific good indicators of lithium-ion in biological fluids such as blood,[6] Formazan derivatives are used widely to cytotoxicity and evaluate cell viability,[7] Formazan derivatives are used widely as reagents for analysis of trace metal,[8] it's are colored compounds because their π - n* and π - π^* transition, formazan compounds have been generally solids of relatively low melting point although of large the size of the molecules.[9]

MATERIALS

FTIR Spectra (400 -4000 cm-1) in KBr disk were recorded on SHIMADZU FTIR-8400S Fourier transform. ^{13}C-NMR and ^{1}HNMR were recorded on Fourier transformation Bruker spectrometer operating at (500MHz) with (DMSO-d6) measurements were made at Department of Chemistry,kashan University, Iran.

METHODS

Preparation of Compound (a)[10, 11]

Mix equal moles of chloro acetyl chloride compounds (0.0079 mol, 0.6 ml) with the amino compound (2-amino-4-hydroxy-6-methyl bromide) (0.0079 mol, 1 g) in 30 mL of dry benzene at room temperature with stirring for 6 hours The reaction was followed by TLC technique. The precipitation was then filtered and reconstituted with absolute ethyl alcohol. to resulted whit precipitation and molecular weight (202.598) and a percentage (87%) were obtained. The melting point was within range (228–230)^{o}C (R*f* = 0.27).

Preparation Compound (b)[12,13]

Mix equal moles of hydrazine hydrate (0.004 mol, 0.29 mL) with compound (1) at room temperature for 8 hours where the reaction process was followed using TLC technique. The precipitate was then filtered and reconstituted using $CHCl_3$. to resulted whit precipitation and and its melting point was within the range 193–195^{o}C and (R*f* 0.43) (78.1%).

Preparation of Schiff Base Compound (c)[14,15]

Mix equal moles of compound (2) (0.005 m, 1 g) with 3, 4-dimethoxy benzaldihyde (0.005 m, 0.83 g) and reflex at a temperature of(74-78) ^{o}C for 8 hours. where the reaction process was followed using TLC technique,The precipitation was then filtered and reconstituted using methanol . to resulted yellow precipitation and molecular weight of 345.359 and a percentage of 90% were obtained. The melting point was within range (208-212)^{o}C and(R*f* = 0.35).

Preparation of Dizonum salt (d)[16,17]

Dissolve (0.003mol) of the aromatic Amin is dissolved in 5 mL HCl + 8 mL distilled water). The solution is cooled to a

**Author for Correspondence: shaimaa.adnan@qu.edu.iq*

temperature of(0-5 ° C). After that added to 0.003 mol sodium nitrate dissolved in 5 mL distilled water drop by drop the mixture and leave for 20 minutes inside the refrigerator

Preparation of Formazan Derivatives[18,19]

Dissolve (0.003mol) of the Schiff base derivative (c) to the preparation of derivatives from (c1-c8) In (10 mL dioxane + 20 mL ethano).

The dizonium salt produced from the above step is added to the solution of the Schiff base drop by drop in snow bath (0-5)°C. It is left with stirring for a period of 2- 3.5 hours, the reaction process was followed using TLC technique. Obtain color derivatives to be weighed and calculate the percentage and physical properties as shown in Table 1.

RESULTS AND DISCUSSION

Compound (a) :- 2-chloro-N-(4-hydroxy-6-methylpyrimidin-2-yl)acetamide

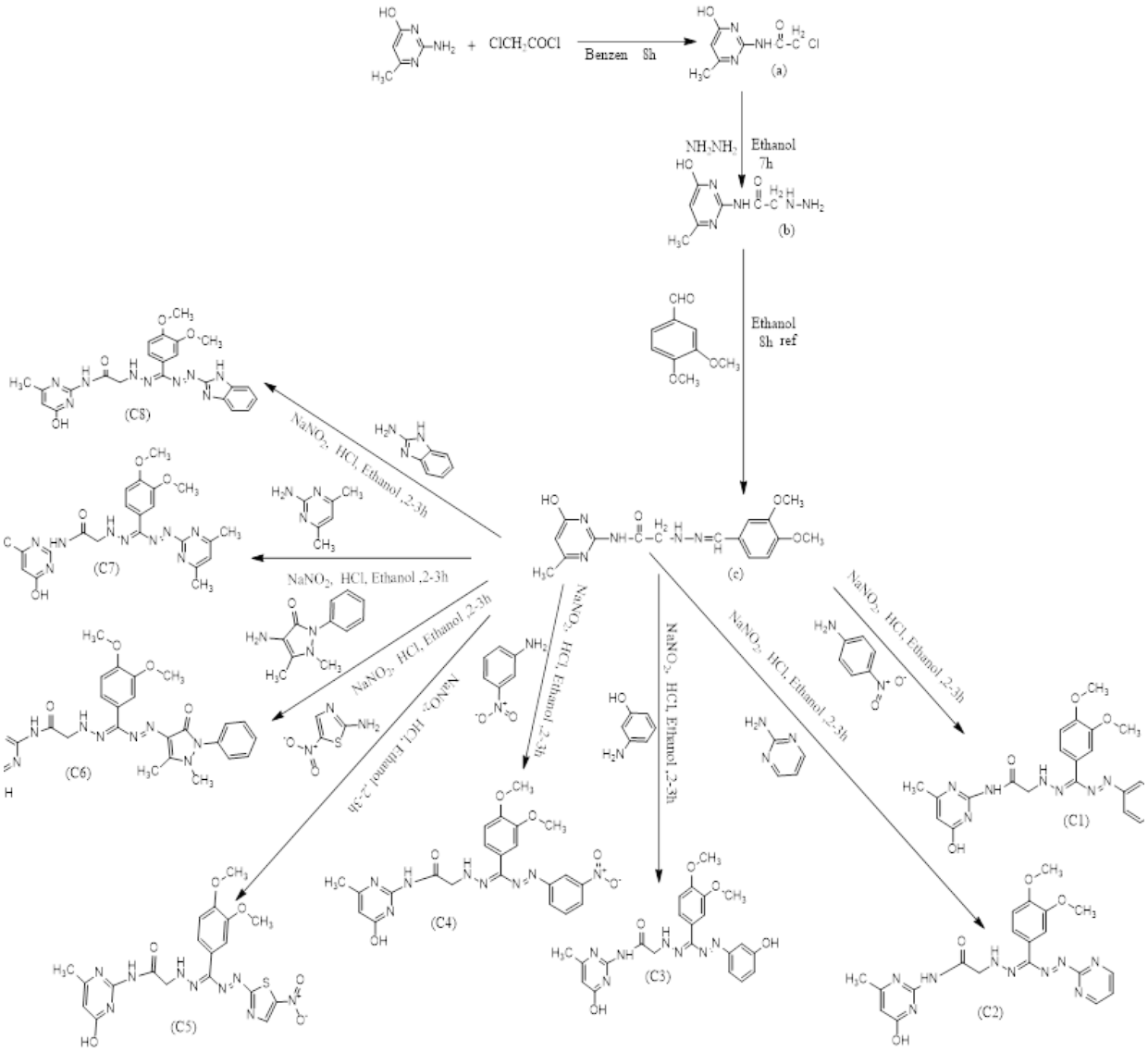
FT-IR spectrum data for derivative (a) show band at 3386 cm^{-1} for (O – H), 3278 cm^{-1} for (N – H), 3070 cm^{-1} for (Ar – H), 2839 cm^{-1} for (C- H) in (CH_3), 1712 cm^{-1} for (C=O),1642 cm^{-1} for (C=N), 1582 cm^{-1} for (C=C) aromatic .

Compound (b) :- 2-hydrazinyl-N-(4-hydroxy-6-methylpyrimidin-2-yl)acetamide

FT-IR spectrum data for derivative (b) show band at (3332-3178) cm^{-1} for (O – H) and (N – H), 3070 for (Ar – H), 2926 cm^{-1} for (C- H) in (CH_3), 1700 cm^{-1} for (C=O), 1666 cm^{-1} for (C=N), 1604 cm^{-1} for (C=C) aromatic

Compound (c):- 2-(2-(3,5-dimethoxybenzylidene) hydrazinyl)-N-(4-hydroxy-6-methylpyrimidin-2-yl) acetamide

FT-IR spectrum data for derivative (c) show band at 3394 cm^{-1} for (O – H), (3278-3178) cm^{-1} for two kind of (N – H), 3078 for (Ar – H), 2968cm^{-1} for (C- H) in CH_3, 1750 cm^{-1} for (C=O), 1620 cm^{-1} for (C=N), 1600 cm^{-1} for (C=C) . ^{1}HMNR spectrum data of compound (c) show 2.50ppm (DMSO), 10PPm (s, 1H, OH), 2ppm (s,3H, CH_2) Figure 1, 2.4ppm (S,6H, OCH_3),3.9ppm (s,



Scheme 1: Synthesis of some formazan derivatives

2H, CH_2),7.4-7,9ppm (m, 4H, Ar-H), 8.2PPm (s,1H,N=CH). The C13–NMR spectrum data (DMSO) compound (c) show :23.76ppm (C_{13}), 26.24ppm (C_{14},C_{15}), 55ppm (C_6), 160ppm (C_5),129ppm(C_7),145PPm (C_4), 143 (C_1),136(C_{14},C_{15}), 117-134ppm (C_{Arom}), 39ppm DMSO .

Compound(C_1):- (E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)-1-(4-nitrophenyl)formazan

FT-IR spectrum data for derivative (C_1) show band at 3340 cm^{-1} for (O – H), 3224 cm^{-1} for (N – H), 3078 for (Ar – H), 2931 cm-1 for (C- H) in (CH_3), 1728 cm-1 for (C=O), 1600 cm-1 for (C=N), 1596 cm-1 for (C=C) aromatic . ^{1}HMNR spectrum data of derivative (C_1) show 2.43ppm (DMSO) 10.5PPm (s, 1H, OH), 1.22ppm (s,3H, CH_3), 3.2ppm, (s,6H, OCH_3),3.9ppm (s, 2H, CH_2),6.90,6.92ppm (s, 2H, NH), 7.1, 7.7ppm (m, 8H, Ar-H) . The C13–NMR spectrum data (DMSO) derivative (C_1) show :26.7ppm (C_{18}), 28.6ppm ($C_{19,}$ C_{20}), 29.7ppm (C_6), 161ppm (C_5),154ppm(C_4),148ppm (C_3), 139ppm (C_{17}), 129ppm (C_7), 137ppm (C_{14}), 100-130ppm (C_{Arom}), 39ppm (DMSO) Figure 2.

Compound(C_2):- (E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)-1-(pyrimidin-2-yl)formazan

FT-IR spectrum data for derivative (C_2) show band at 3425 cm-1 for (O – H), 3224 cm-1 for (N – H), 3332 cm^{-1} for (Ar – H), 2931 cm-1 for (C- H) in (CH_3), 1735 cm-1 for (C=O), 1697 cm-1 for (C=N), 1581 cm-1 for (C=C) aromatic . ^{1}HMNR spectrum data of derivative (C_2) show 2.50ppm (DMSO) 10.3 ppm (s, H, OH), 2. 4ppm (s,3H, CH_3), 3.3ppm (s,6H, OCH), 3.9ppm (s, 2H, CH_2), 6.6, 6.8 ppm (s, 2H, NH), 7.1- 7.8 ppm (m,7H, Ar-H) . The C13–NMR spectrum data (DMSO) derivative (C_2) show : 22.1ppm (C_{17}), 26ppm (C_{18}), 28ppm (C_6), 164ppm (C_5),159ppm(C_4), 129ppm (C_7), 155ppm (C_{14}), 151ppm (C_3), 147ppm ($C_{10,}$ C_{11}),100 ppm -138PPm (C_{Arm}) See Figure 3 and 4.

Compoundc_3:- (E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)-1-(3-hydroxyphenyl)formazan

FT-IR spectrum data for derivative (c_3) show band at 3425 cm-1 for (O – H), 3276 cm^{-1} for (N – H), 3132 cm^{-1} for (Ar – H), 2958 cm^{-1} for (C- H) in(CH_3), 1797 cm^{-1} for (C=O), 1604 cm^{-1} for (C=N), 1419 cm^{-1} for (C=C) aromatic . ^{1}HMNR spectrum data of derivative (c_3) show 10 ppm (s, 2H, OH), 1.2 ppm (s,3H, CH_3), 2.0ppm (s,6H, OCH), 3ppm (s, 2H, CH_2), 6.4PPm, 6.8 ppm (s, 2H, NH), 6.9-8.3 ppm (m,8H, Ar-H) . The C13–NMR spectrum data (DMSO) derivative (c_3) show : 26ppm (C_{20}), 28ppm (C_{22}, C_{23}), 37ppm (C_6), 163ppm (C_5),158ppm(C_4), 129ppm (C_7), 147ppm (C_9, C_{10}), 151ppm ($C_{3,}$ C_{16}), 101 ppm -137PPm (C_{Arm}) .

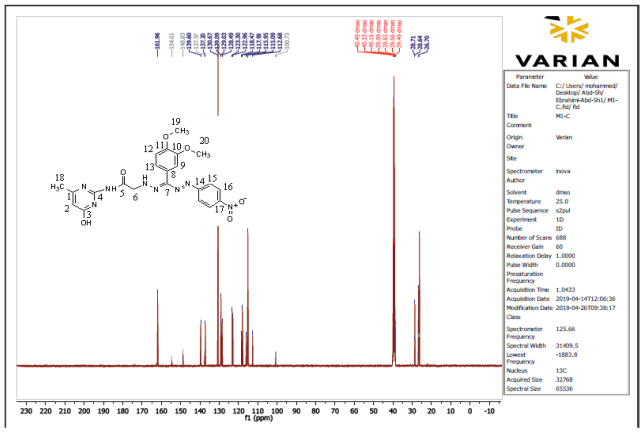


Figure 2: ^{13}C–NMR spectrum of compound (c_1)

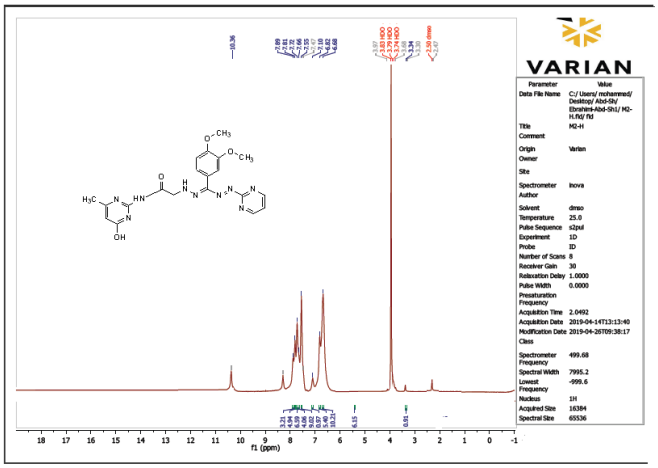


Figure 3: ^{1}H–NMR spectrum of compound (c_2)

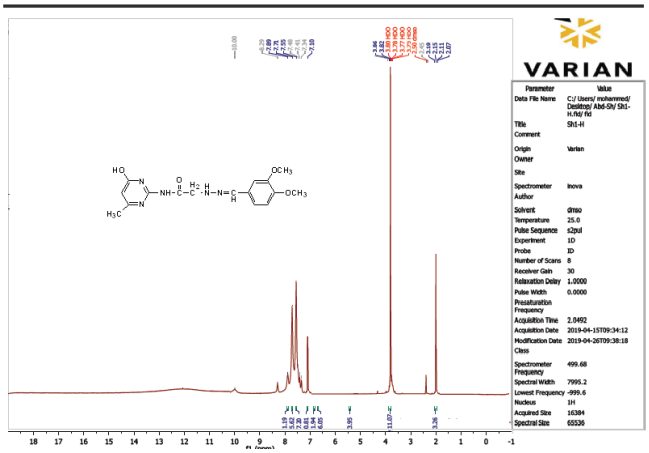


Figure 1: ^{1}H–NMR spectrum of compound (c)

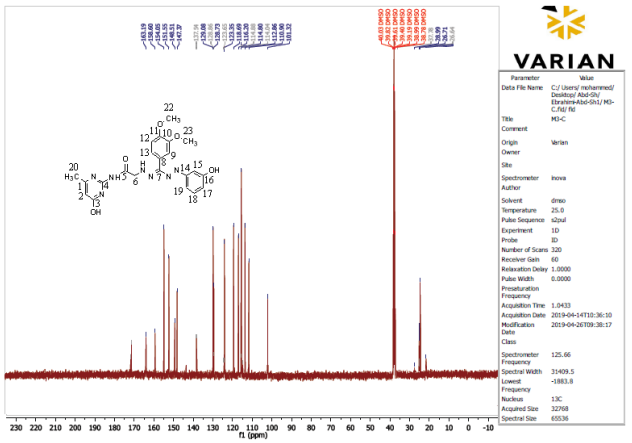


Figure 4: ^{13}C–NMR spectrum of compound (c_3)

Compoundc_4 :- (E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)-1-(3-nitrophenyl)formazan

FT-IR spectrum data for derivative (c_4) show band at 3440 cm-1 for (O – H), 3261 cm-1 for (N – H), 3078 cm-1 for (Ar – H), 2931 cm-1 for (C- H for CH_3), 1735 cm-1 for (C=O), 1581 cm-1 for (C=N), 1512 cm-1 for (C=C) aromatic . ^{1}HMNR spectrum data of derivative (c_4) show 10ppm (s, H, OH), 2ppm (s,3H, CH_3), 3.3ppm (s,6H, OCH), 3.9ppm (s, 2H, CH_2), 5.3ppm, 6.5 ppm (s, 2H, NH), 6.8ppm – 8.1 ppm (m,8H, Ar-H) . The C13–NMR spectrum data (DMSO) derivative (c_4) show : 20ppm (C_{19}), 26ppm, 29ppm ($C_{20,}$ C_{21}), 33ppm (C_6), 162ppm (C_5),159ppm(C_4), 129ppm (C_7), 155ppm (C_{14}), 130ppm (C_{16}), 138ppm ($C_{10,}$ C_{11}),100 ppm-129ppm (C_{Arm}), 39ppm (DMSO) .

Compoundc_5:- (E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)-1-(5-nitrothiazol-2-yl)formazan

FT-IR spectrum data for derivative (c_5) show band at 3409 cm-1 for (O – H), 3224 cm-1 for (N – H), 3032 cm-1 for (Ar – H), 2839 cm-1 for (C- H for CH_3), 1560 cm-1 for (C=O), 1581 cm-1 for (C=N), 1481 cm-1 for (C=C) aromatic . ^{1}HMNR spectrum data of derivative (c_5) show 10.7ppm (s, 1H, OH), 1.2ppm (s,3H, CH_3), 2ppm (s,6H, OCH), 3.8ppm (s, 2H, CH_2), 8.1ppm (s, 1H, thiazol), 5.3, 6.7 ppm (s, 2H, NH), 6.9ppm – 7.4 ppm (m,4H, Ar-H) . The C13–NMR spectrum data (DMSO) derivative (c_5) show : 21ppm (C_{17}), 26.1ppm ($C_{18,}$ C_{19}), 26.6 ppm (C_6), 160ppm (C_5),145ppm(C_4), 129ppm (C_7), 143ppm (C_{14}), 147ppm ($C_{10,}$ C_{11}),136.3ppm(C_3), 117 ppm-129ppm (C_{Arm}), 39ppm (DMSO) See Figure 5 and 6.

Compoundc_6:- (E,E)-3-(3,4-dimethoxyphenyl)-1-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)formazan

FT-IR spectrum data for derivativ (c_6) show band at 3425 cm-1 for (O – H), 3217 cm-1 for (N – H), 3124 cm-1 for (Ar – H), 2931 cm-1 for (C- H for CH_3), 1720 cm-1 for (C=O), 1581 cm-1 for (C=N), 1419 cm-1 for (C=C) aromatic . ^{1}HMNR spectrum data of derivative (c_6) show 10ppm (s, H, OH), 2.1ppm (s,3H, CH_3 in pyrazoline derivative), 3.6ppm, 3.7ppm (s,6H, OCH), 3.97ppm (s, 2H, CH_2), 6.77ppm, 6.79 ppm (s, 2H, NH), 2.5 ppm (s,3H, in pyrazoline derivative) . The C13–NMR spectrum data (DMSO) derivative (c_6) show : 20ppm (C_{23}), 26.21ppm, 26.62ppm, 29ppm ($C_{24,}$ C_{25}), 29.32ppm (C_{26}), 29.72ppm (C_{27}) 33.62ppm (C_6), 162ppm (C_5),149ppm(C_4), 139ppm (C_{14}), 155ppm (C_{15}), 137ppm (C_{22}),100 ppm-130ppm (C_{Arm}), 39ppm (DMSO) See Figure 7 and 8.

Compoundc_7 :- (E,E)-3-(3,4-dimethoxyphenyl)-1-(4,6-dimethylpyrimidin-2-yl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)formazan

FT-IR spectrum data for derivative (c_7) show band at 3440 cm-1 for (O – H), 3240 cm-1 for (N – H), 3132 cm-1 for (Ar – H), 2839 cm-1 for (C- H for CH_3), 1720 cm-1 for (C=O), 1581 cm-1 for (C=N), 1419 cm-1 for (C=C) aromatic . ^{1}HMNR spectrum data of derivative (c_7) show 10.9ppm (s, 1H, OH), 1.91ppm,1.97ppm, 2.1ppm (s,9H, CH_3), 3.6ppm (s,6H, OCH), 3.7ppm (s, 2H, CH_2), 6.5ppm, 6.7 ppm (s, 2H, NH), 6.8ppm – 7.9 ppm (m,4H, Ar-H) . The C13–NMR spectrum data (DMSO) derivative (c_7) show : 22ppm (C_{18}), 26.65ppm, 26.23ppm (C_{21}), 28.6 ppm, 28.7 ppm (C_{19}, C_{20}), 30.9 ppm (C_6), 167ppm (C_5),153ppm(C_4), 129ppm (C_7), 152ppm (C_{14}), 141ppm ($C_{10,}$ C_{11}),121 ppm -135ppm (C_{Arm}), 39ppm (DMSO) See Figure 9.

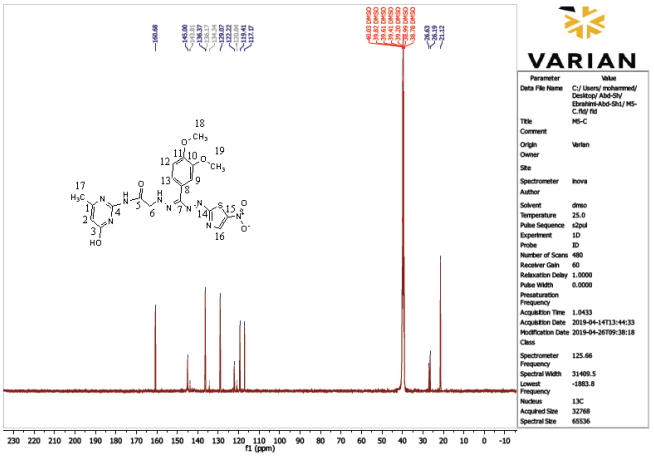


Figure 6: ^{13}C–NMR spectrum of compound (c_5)

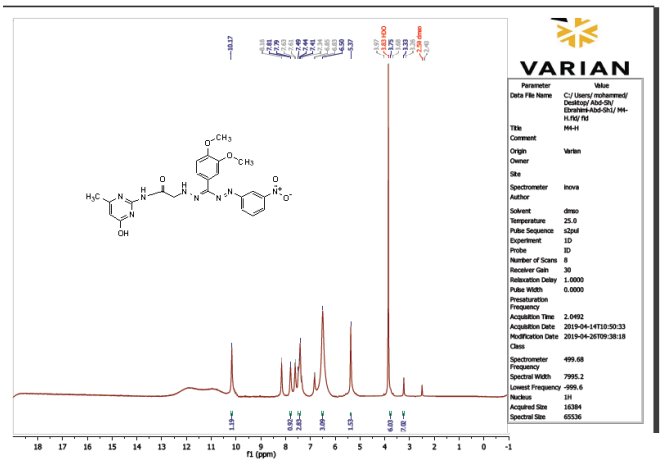


Figure 5: ^{1}H–NMR spectrum of compound (c_4)

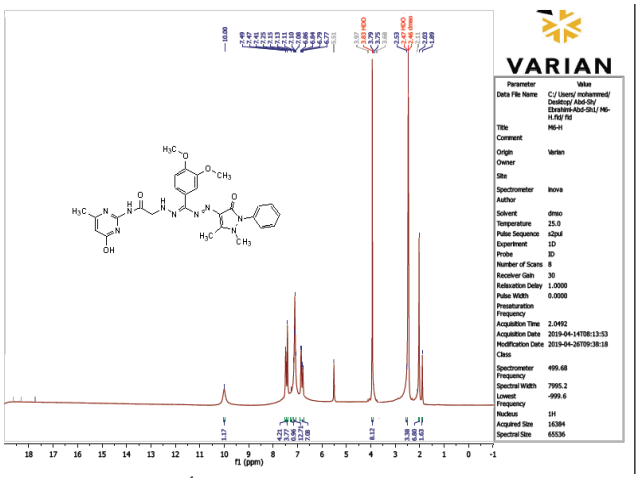


Figure 7: ^{1}H–NMR spectrum of compound (c_6)

Compound c_8:- (E,E)-1-(1H-benzo[d]imidazol-2-yl)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)formazan

FT-IR spectrum data for derivative (c_8) show band at 3487 cm^{-1} for (O – H), 3217 cm^{-1} for (N – H), 3116 cm^{-1} for (Ar – H), 2908 cm^{-1} for (C- H in CH_3), 1735 cm^{-1} for (C=O), 1620 cm^{-1} for (C=N), 1504 cm^{-1} for (C=C) aromatic . ^{1}HMNR spectrum data of compound (c_8) show 10ppm (s, 1H, OH), 2ppm, 3.3, 3.6 (s,3H, CH_3), 3.7ppm (s,6H, OCH), 3.7ppm (s, 2H, CH_2), 5.3ppm,6.6ppm (s, 3H, NH), 6.8ppm – 8.33 ppm (m,9H, Ar-H) . The C13–NMR spectrum data (DMSO) derivative (c_5) show : 23ppm (C_{21}), 26ppm, 29ppm ($C_{22,}$ C_{23}), 37 ppm (C_6), 164ppm (C_5),163ppm(C_4), 161ppm (C_{14}), 155ppm ($C_{15,}C_{20}$), 129ppm (C_7), 149ppm (C_3), 137ppm ($C_{10,}C_{11}$), 100 ppm-130ppm (C_{Arm}), 39ppm (DMSO) .

BIOLOGICAL ACTIVITY

in the above studies it can be conclusion that the prepare compounds reduce significant antibacterial effectiveness against bacteria staphylococcus aurous and Escherichia coli. The compounds that show good activity are (2,3,6) against (staphylococcus aurous), and compound that show good activity are (1,2,6) against (Escherichia coli), the results of the antibacterial activity are shown in the Figure 10.

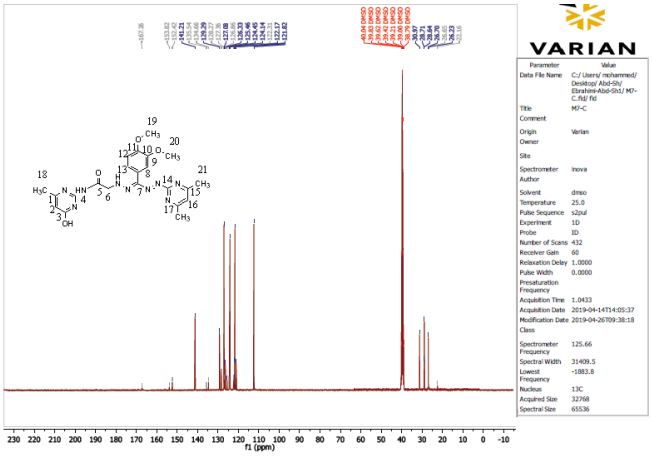


Figure 8: ^{13}C–NMR spectrum of compound (c_7)

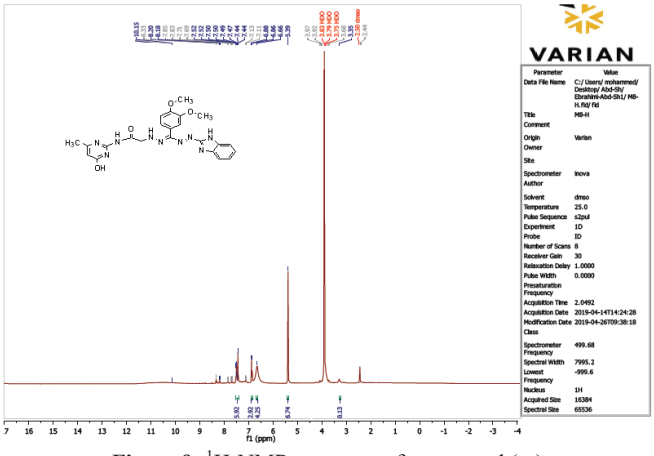


Figure 9: ^{1}H–NMR spectrum of compound (c_8)

The biological activity of the prepared compounds was investigated and the most effective compound (2) The drug was studied on two types of cells: MCF-7 and WRL The study showed that this type of compounds was effective with good pharmacologic effect (Figure 11). At concentration 400, we found a response rate of 61% in MCF-7 cells versus 21% in WRL cells and table (1-2) showing the effect of the compound on cells versus concentrations Which was developed

The IC50 values for the cells were also calculated and there was a significant difference between the values of the IC50 in the WRL and MCF-7 cells as shown in the Figure 11.

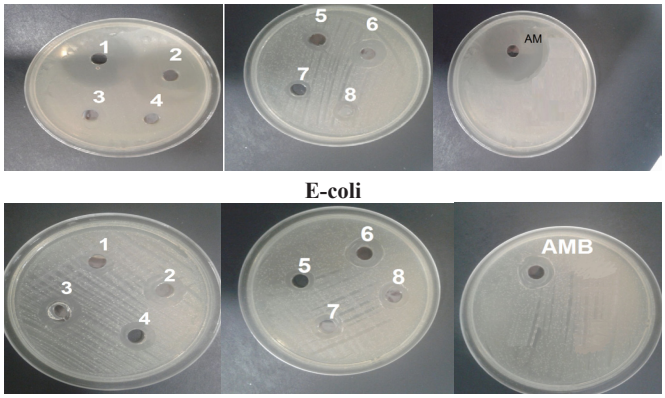


Figure 10: Activity of staphylococcus aurous) against inhibition

Table 1: Show Biological activity for compounds (1-8)

Comp No	*E.Coli*	*Mm*	*Staph aureus*	*Mm*
1	+++	28	+	7
2	+++	31	+++	25.5
3	-	0	++	17
4	-	0	+	9
5	+	5.2	+	6.5
6	++	13	++	12
7	+	7.5	+	3
8	+	9	+	5
Ampicillin	+++	29	+++	23

=-No inhibition active = in active,+= (5-10)mm =slightly active, ++= (11-20)mm moderately +++ = More than 20, good active

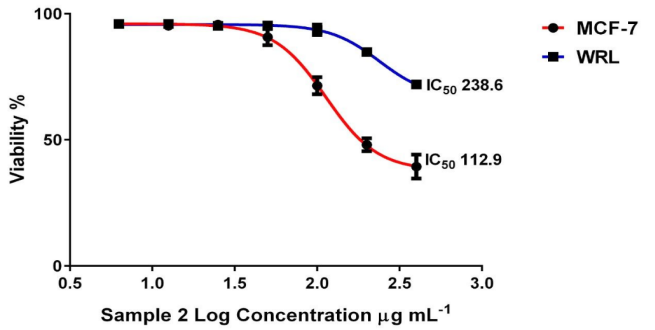


Figure 11: biological activity of the prepared compounds on two types of cells: MCF-7 and WRL

Table 2: Showing the effect of the compound on cells versus concentrations Which was developed

WRL		A	B	C	D	E	F	G
		400	200	100	50	25	12.5	6.25
		Y	Y	Y	Y	Y	Y	Y
1	Number of values	3	3	3	3	3	3	3
2								
3								
4	Mean	71.95	84.80	93.60	95.33	95.22	95.95	95.95
5	Std. Deviation	0.8128	1.205	2.100	1.183	0.8209	1.028	0.2003
6	Std. Error of Mean	0.4693	0.6955	1.212	0.6828	0.4739	0.5937	0.1157

MCF-7		A	B	C	D	E	F	G
		400	200	100	50	25	12.5	6.25
		Y	Y	Y	Y	Y	Y	Y
1	Number of values	3	3	3	3	3	3	3
2								
3								
4	Mean	39.35	48.03	71.49	90.70	95.72	95.18	95.95
5	Std. Deviation	4.783	2.554	3.399	3.183	0.8099	1.280	0.5306
6	Std. Error of Mean	2.762	1.475	1.962	1.838	0.4676	0.7393	0.3063

Table 3: Physical and analytical data of compounds (1-8)

Comp	*Nam of compound*	*M.F* *M.wat*	*m.p*	*Rf*	*Colour*	*%*
1	((E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl) amino)-2-oxoethyl)-1-(4-nitrophenyl)formazan	$C_{22}H_{22}N_8O_6$ 494.47	169	0.31	Yellow	62
2	(E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl) amino)-2-oxoethyl)-1-(pyrimidin-2-yl)formazan	$C_{20}H_{21}N_9O_4$ 451.45	204	0.16	Yellow	64
3	(E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl) amino)-2-oxoethyl)-1-(3-hydroxyphenyl)formazan	$C_{22}H_{23}N_7O_5$ 465.47	201	0.27	Pink	72
4	((E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl) amino)-2-oxoethyl)-1-(3-nitrophenyl)formazan	$C_{22}H_{22}N_8O_6$ 494.47	192	0.43	Yellow	62
5	((E,E)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl) amino)-2-oxoethyl)-1-(5-nitrothiazol-2-yl)formazan	$C_{19}H_{19}N_9O_6S$ 501.48	215	0.25	Brown red	69
6	((E,E)-3-(3,4-dimethoxyphenyl)-1-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)formazan	$C_{27}H_{29}N_9O_5$ 559.59	183	0.28	Yellow	63
7	((E,E)-3-(3,4-dimethoxyphenyl)-1-(4,6-dimethylpyrimidin-2-yl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)formazan	$C_{22}H_{25}N_9O_4$ 479.50	220	0.12	Yellow	61
8	((E,E)-1-(1H-benzo[d]imidazol-2-yl)-3-(3,4-dimethoxyphenyl)-5-(2-((4-hydroxy-6-methylpyrimidin-2-yl)amino)-2-oxoethyl)formazan	$C_{23}H_{23}N_9O_4$ 489.50	176	0.22	Brown	67

REFERENCE

1. Türkoğlu G, Cinar ME. Experimental and computational studies on the absorption properties of novel formazan derivatives. Turkish Journal of Chemistry. 2017 Nov 10; 41(5):710-727.
2. YAĞLIOĞLU AŞ, ŞENÖZ H. Synthesis of novel 5-substituted phenyl-3-(p-isopropylphenyl)-1-phenylformazan and their biological activities. Turkish Journal of Chemistry. 2017 Dec 20;41(6):883-891.
3. Vujasinović I, Paravić-Radičević A, Mlinarić-Majerski K, Brajša K, Bertoša B. Synthesis and biological validation of novel pyrazole derivatives with anticancer activity guided by 3D-QSAR analysis. Bioorganic & medicinal chemistry. 2012 Mar 15;20(6):2101-2110.
4. Mohammed OA, Dahham OS. Synthesis, Characterization, and Study of Antibacterial Activity of Some New Formazan Dyes Derivatives, Derived from 2-Mercapto Benzoxazole. InIOP Conference Series: Materials Science and Engineering 2018 Dec (Vol. 454, No. 1, p. 012015). IOP Publishing.
5. Aljamali NM, Kashash DR, Microbial Test Of Formazan Compounds Against Types Of Bacteria. Pak. J. Biotechnol. 2018; 15(1): p. 175-179.
6. Khattab T, Haggag KM. Synthesis and spectral properties of symmetrical and asymmetrical 3-cyano-1, 5-diarylformazan dyestuffs for dyeing polyester fabrics. Egyptian Journal of Chemistry. 2017 Dec 1;60(Conference Issue (The 8th International

Conference of The Textile Research Division (ICTRD 2017), National Research Centre, Cairo 12622, Egypt.)):33-40.
7. Mariappan G, Korim R, Joshi NM, Alam F, Hazarika R, Kumar D, Uriah T. Synthesis and biological evaluation of formazan derivatives. Journal of advanced pharmaceutical technology & research. 2010 Oct;1(4):396.
8. V Petunin P, R Valiev R, G Kalinin R, E Trusova M, V Zhdankin V, S Postnikov P. General and simple method for the synthesis of 3-nitroformazan using arenediazonium tosylates. Current Organic Synthesis. 2016 Aug 1;13(4):623-628.
9. Al-Araji YH, Shneine JK, Ahmed AA. Chemistry of formazan. Int. J. Res. Pharm. Chem. 2015;5:41-76.
10. Abbady MA, Abdel-Hafez SH. Organic selenium compounds. Part I: Synthesis and application of some new diaryl-selenides and selenones containing amino acid moieties. Phosphorus, Sulfur, and Silicon and the Related Elements. 2000 May 1;160(1):121-139.
11. Pattan SR, Dighe NS, Nirmal SA, Merekar AN, Laware RB, Shinde HV, Musmade DS. Synthesis and biological evaluation of some substituted amino thiazole derivatives. Asian journal of research in chemistry. 2009;2(2):196-201.
12. Sonwane SK, Srivastava SD, Srivastava SK. Synthesis and antimicrobial activity of 2-(2′-arylidene-hydrazino-acetyl-amino)-4-phenyl-1, 3-thiazoles and 2-[2′-{4″-substituted-aryl-3″-chloro-2″-oxo-azetidine}-acetyl-amino]-4-phenyl-1, 3-thiazoles. 2008.
13. Wasfy AA. Fused heterocycles. Part I. Synthesis of some annelated 1, 2, 4-triazole systems from [4-(1 H-benzimidazol-2-yl)-phthalazin-1-yl] hydrazine. Journal of Chemical Research. 2003 Aug;2003(8):457-458.
14. Manikandan S, Thirunarayanan G. Synthesis, Spectral Characterization of Some Novel Schiff's base and its Antibacterial Studies. International Journal of Applied Engineering Research. 2018;13(13):11077-11086.
15. Mohamed SS, Al-Sadawi IA, Gbaj MA, Alsabri SG, Elmaki N, Bensaber S, Hermann A, Gbaj A. Microwave assisted synthesis and antimicrobial evaluation of symmetrical 1, 2-Phenylene-diamine Schiff's base derivatives. Pharm Pharmacol Int J. 2018;6(5):344-348.
16. Albasha M. Synthesis, Characterization of New Azo Compounds and Their Biological Evaluation. 2018; 6: 16-24.
17. Kantar C, Baltas N, Karaoglu SA, Sasmaz S. Some azo dyes containing eugenol and guaiacol, synthesis, antioxidant capacity, urease inhibitory properties and anti-helicobacter pylori activity. Revue Roumaine De Chimie. 2018 Mar 1;63(3):189-197.
18. Joseyphus RS, Shiju C, Joseph J, Dhanaraj CJ, Bright KC. Synthesis and characterization of Schiff base metal complexes derived from imidazole-2-carboxaldehyde with L-phenylalanine. Pharm Chem. 2015;7(6):265-270.
19. Bhosale JD, Shirolkar AR, Pete UD, Zade CM, Mahajan DP, Hadole CD, Pawar SD, Patil UD, Dabur R, Bendre RS. Synthesis, characterization and biological activities of novel substituted formazans of 3, 4-dimethyl-1H-pyrrole-2-carbohydrazide derivatives. Journal of Pharmacy Research. 2013 Jul 1;7(7):582-587.