PREPARATION, IDENTIFICATION AND STUDY THE BIOLOGICAL ACTIVITY OF NOVEL HETEROCYCLIC COMPOUNDS DERIVED FROM AZO–CHALCONE

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Abstract: *This work describes the preparation and spectroscopic investigation of new heterocyclic compounds derived from the 4-aminoantipyrene moiety. The compound (R1)3-((1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)diaz-enyl)-4-hydroxybenzaldehyde was prepared by reacting 4-aminoantipyrene with 4-Hydroxybenzaldehyde according to the cold condition at (0-5) Celsius. This represents the starting point to create novel azo-chalcone compounds with a new nucleus as the alpha-beta unsaturated group, and the formation of compound ($R_{2\text{-}6}$) via acetophenone derivatives using ethanol as a solvent under a basic medium, and for synthesis of various compounds of 4-((5-((Z)-3-argio-3-oxoprop-1-en-1-yl)-2-hydroxyphenyl)diazenyl)-1,5-dimethyl-2-phenyl-1,2-dihydro-3H-pyrazol-3-one, using the cyclization of these compounds via (urea and hydrazine hydrate) to forming five and six novel heterocyclic rings of 4-((5-(2-amino)-4-Argyo-6H-1,3-oxazine-6-yl)-2-hydroxyphenyl(diazenyl)-1,5-dimethyl-2-phenyl-1,2-dihydro-3H-pyrazole-3-one [$R_{7,\ 11}$], and 4-((5-(5-(4-argiophenyl)-4,5-dihydro-1H-pyrazol-4-yl)-2-hydroxyphenyl)diazenyl)-1,5-dimethyl-2-phenyl-1,2-dihydro-3H-pyrazole-3-one [$R_{12,16}$], respectively. Some diagnostic measurements have been used to characterize these molecules, such as melting point, thin-layer chromatography, measuring Nuclear Magnetic Resonance, Spectroscopy in the Infrared, and Mass Spectrometry, and determining the effectiveness of some of these compounds by diagnosing their effectiveness against types of pathogenic bacteria.*

Keywords: *Chalcone, Azo-Chalcone, 4-Amino Antipyrene, Biological Activity*
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1. Introduction

Chalcone is one of the carbonyl compounds and among the most widespread organic compounds in biological and organic chemistry [1, 2] and it is a source for building important compounds in the industrial and medical fields [3]. α-β unsaturated compounds contain a carbonyl group that exists in a replaced state with an unsaturated group (C=C) [4]. The double bond (C-C) and the bond (CO-O) are separated by a single bond [5], and the two bonds are in an alternating state. The coupling between the two groups represents the possibility of spreading charges across the four atoms, which gives stability to the chain system due to the presence of resonance [6].

Because of the presence of the keto ethylenic moiety, CO-CH=CH-, chalcones and their derivatives are regarded valuable. Moieties in the domain of synthetic and heterocyclic organic chemistry [7] were created via Claisen-Schmidt synthesis [8] by condensation acetophenones together with benzaldehyde substitutes [9].

Chalcones consider α-β unsaturated compounds that dissolve in organic solvents but not in water. Kastanek first used chalcone terminology in 1899, when he conducted preliminary experiments in preparing natural colored compounds [10]. Chalcone is a basic chemical scaffold found in many plant products such as fruits, tea, and vegetables [8]. Chalcone has an extrinsic spectrum of biological activity [11] and occupies an important place in biochemistry and medicine because it is anti-inflammatory [12, 13]. It is antibacterial, antiviral [14], anti-tuberculous [15], antitumor

[16, 17], anti-HIV [18], anti-fungal, antioxidant [19] and anti-ulcer agents [20].

Azo-chalcone dyes, common in vegetables, plants, and fruits, are compounds with systems linked to heterocyclic or degradable aromatic rings [21]. These a major source for the synthesis of new organic compounds, such as heterocyclic compounds, which may have promising biological activity [22].

2. Experimental part

2.1. Material and methods.

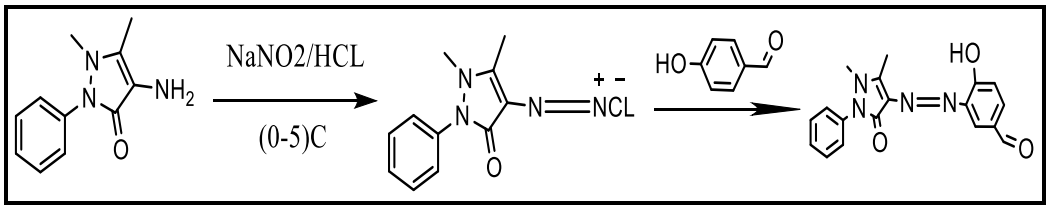
The Smp.30 device, produced by the British company STURART, was employed to measure melting temperatures to the prepared compounds. The chemicals used were purchased from BHD and Fluka and Sigma Aldrich. The infrared spectra were measured using IR Affinity-S 1, a Shimadzu type device at a wavenumber of (4000-600 cm-1).

^{1}H-NMR using America Bruker 400MHz, TMS was used internal standard; DMSO-d^6 was used as the solvent. The mass spectrums were also observed via (GCMS-QP 2010 plus), Japanese origin, a Shimadzu type device. The reaction was followed using thin -layer chromatography (TLC) was carried out in silica gel (120 mash) coated plates (1×10). The media for microorganisms was sterilized using an autoclave device equipped from a Spanish company, the dishes were developed in an incubator device, and testing was done in the laboratories of the Microbiology Division in the College of Science.

2.2. General procedure for synthesis of 3-((1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)diazenyl)-4-hydroxybenzaldehyde (R1) [23].

A (0.01 mol, 2.03 g) of 4-amino antipyrin was dissolved in mixture of 7 ml concentrated hydrochloric acid (37%) with (7 ml) of (H_2O) in an ice bath while constant stirring until the temperature reacted (0-5 ºC). In another bowl, 1.6 g of sodium nitrite was dissolved in 8 ml H_2O. The sodium nitrate solution was added in batches to the first solution with continuous stirring (1-2ml) for each batch. In this case, it is necessary to maintain the temperature of the ice bath so that the temperature does not rise above 5 ºC. The solution formed diazonium salt in the ice bath solution- was prepared (0.1g) from 4-hydroxy benzaldehyde and dissolved it in 2 ml of diluted (10% NaOH) by adding 5 ml of H_2O. The last solution was stirred and then added to 2 ml of diazonium salt.

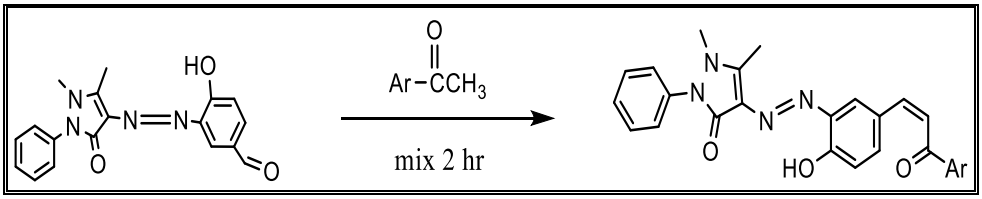
Observing the formation of dye, the solution leaves for 15 min in an ice bath to settle. The crystals of the dye thickly separated, filtered and recrystallized with ethanol: water at ratio 1:2 orang needle crystals separated at melting point (168-169 ºC), (monitored via TLC). The solvent system (4:1) benzene:MeOH. RF was 6.5.



4-aminoantipyrin (R1)

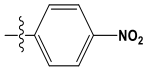
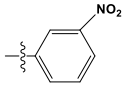
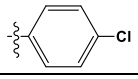
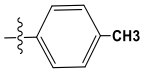
2.3. Preparation of 4-((5-((Z)-3-argio-3-oxoprop-1-en-1-yl)-2-hydroxyphenyl)-diazenyl)-1,5-dimethyl-2-phenyl-1,2-dihydro-3H-pyrazol-3-one (R_{2-6}) [24]

In a 100 ml round-bottomed flask (0.004 mol) of the aldehyde derivative ($\mathbf{R_1}$) was placed and 10 ml of ethanol with (0.004 mol) of acetophenone derivatives dissolved in 10 ml of ethanol were added to it. The reaction mixture was stirred with a magnetic stirrer for 2 hours and the reaction was noted (TLC), the solvent system was benzene:methanol (4:1). After heating was completed, the mixture was cooled and 15 ml of chilled water was poured in. Then it was filtered and recrystallized with ethanol. The azo-chalcone compound ($\mathbf{R_{2-6}}$) was recognized. The physical data of these compounds are listed in Table 1.



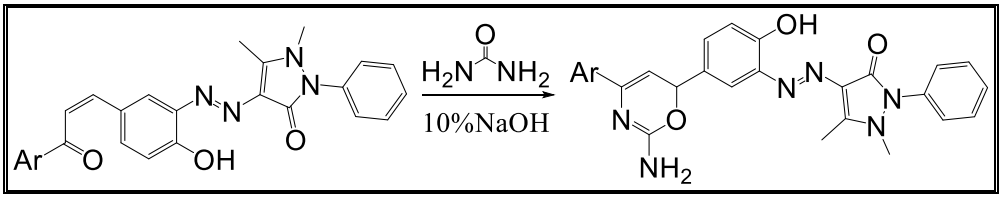
(R_1) (R_{2-6})

Table 1. Physical data of compounds (**R_{2-6}**)

Comp No.	Ar	Molecular formula and m.wt g/mol	M.P. (°C)	Yield %	R_f	Colour
R_2		**$C_{25}H_{19}N_5O_5$ 469**	**222-223**	**85**	**0.625**	**Brown powder**
R_3		**$C_{25}H_{19}N_5O_5$ 469**	**178-180**	**82**	**0.542**	**Brown powder**
R_4		**$C_{25}H_{19}N4O_3CL$ 458**	**-229 228**	**73.5**	**0.43**	**Brown powder**
R_5		**$C_{25}H_{19}N_4O_3CL$ 458**	**-110 108**	**74**	**0.66**	**White powder**
R_6		**$C_{26}H_{22}N_4O_3CL$ 473**	**-228 227**	**90**	**0.71**	**White powder**

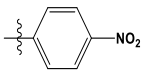
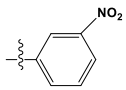
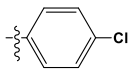
2.4. Preparation of 4-((5-(2-amino-4-argio-6H-1,3-oxazin-6-yl)-2-hydroxy-phenyl)diazenyl)-1,5-dimethyl-2-phenyl-1,2-dihydro-3H-pyrazol-3-one(R7-11) [25].

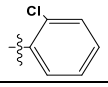
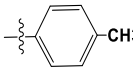
A mixture of 0.02 mol of chalcone derivatives (R_{2-6}) with (0.02 mol, 1.2 g) of urea in (10%) NaOH was dissolved, the mixture was stirred with a magnetic stirrer for (3-4 hours), 20 ml of cold water was poured into the reaction mixture and stirring was continued for another hour. Then the reaction mixture was left in the refrigerator for (24 hours). Then drops of hydrochloric acid were added to the equation, the resulting precipitate was filtered and recrystallized from methanol. Physical data of compounds (R7-11) are given in Table 2.



(R_{2-6}) (R_{7-11})

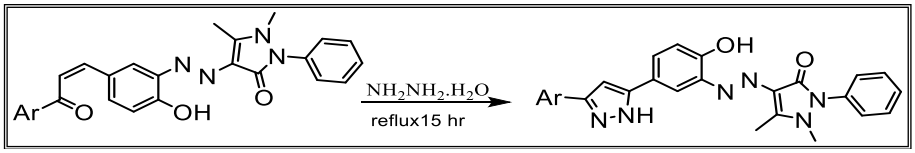
Table 2. Physical data of compound (R_{7-11})

Comp. No	Ar	Molecular formula and m.wt, g/mol	M.P. (°C)	Yield %	R_f	Colour
R_7		**$C_{26}H_{22}N_5O_5$ 512**	**180-182**	**81**	**0.45**	**Orange powder**
R_8		**$C_{26}H_{22}N_5O_5$ 512**	**72-73**	**85**	**0.55**	**White powder**
R_9		**$C_{26}H_{22}N6O_3CL$ 501**	**-164 163**	**75**	**0.6**	**Yellow powder**

R_{10}		**$C_{26}H_{22}N_6O_3CL$ 501**	**86-88**	**88**	**0.44**	**Yellow powder**
R_{11}		**$C_{27}H_{25}N_6O_3$ 481**	**212-213**	**90**	**0.62**	**White powder**

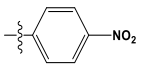
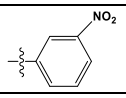
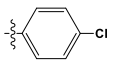
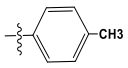
2.5. Preparation of 4-((5-(3-argio-1H-pyrazol-5-yl)-2-hydroxyphenyl)diazenyl)-1,5-dimethyl-2-phenyl-1,2-dihydro-3H-pyrazol-3-one (R_{12-16}) [26] .

In a solution prepared from (0.0015 mole) chalcone derivatives (**R_{2-6}**) dissolved in 30 ml absolute ethanol and added (0.0015 mol, 0.048 g) hydrazine hydrate with few drops from glacial acetic acid, the reaction of mixture was refluxed for (15 hour), the resultant precipitate is filtered and washed by water then recrystallized via ethanol monitored by (TLC) the solvent system 4:1 benzen:MeOH. Physical properties of compounds (R_{12-16}) are listed in Table 3.



(**R_{2-6}**) (**R_{12-16}**)

Table (3) Physical data of compound (R_{12-16})

Comp No.	Ar	Molecular formula and m.wt; g/mol	M.P(^{0} c)	Yield %	R_f	Colour
R_{12}		**$C_{26}H_{22}N_6O_4S$ 512**	**220-221**	**72**	**0.63**	**Yellow powder**
R_{13}		**$C_{26}H_{22}N_5O_5S$ 512**	**76-77**	**74.5**	**0.65**	**Wight powder**
R_{14}		**$C_{26}H_{22}N6O_3SCL$ 501**	**-167 165**	**65**	**0.45**	**Yellow powder**
R_{15}		**$C_{26}H_{22}N_6O_3SCL$ 501**	**96-97**	**55**	**0.35**	**Brawn powder**
R_{16}		**$C_{27}H_{25}N_6O_3S$ 481**	**175-177**	**80**	**0.62**	**White powder**

2.6. Mass spectra study [**27]**

(0.01) g of the sample was dissolved in (5 ml) of ethanol, the device injector was set (2) microliters of sample by using a capillary column of the type (inert cap 1) is a non-polar column bonded 100% dimethylpoly siloxane (DMSO) with a length (30) meters, the carrier gas was helium at a flow rate of (30 ml / min).

At 50°, the oven temperature program was initiated with split ratio of 1:2, this temperature was continued for 2 minutes, after which the temperature was raised at a rate of 25°C per minute until reaching a temperature of 200°C, and then held for 1 minute with a total holding time of 7 minute. The mass spectra were recorded with a range of m / z 900 -30 with an energy of 27 ev.

2.7. The Biological study [28, 29]

Preparation of the agar-Mueller-Hinton medium used in bacterial growth: agar-weld fusion method was used action biological effectiveness of some synthesized chemical compounds **(R_1, R_2, R_5, R_8, R_9, R_{14}, R_{16})** , where (38g) molar "Hinton Agar" was dissolved via distilled boiled water, the acid function of a culture medium was neutralized to (pH = 7). The resulting solution was placed in an autoclave under pressure (150 Pounds/inj) at

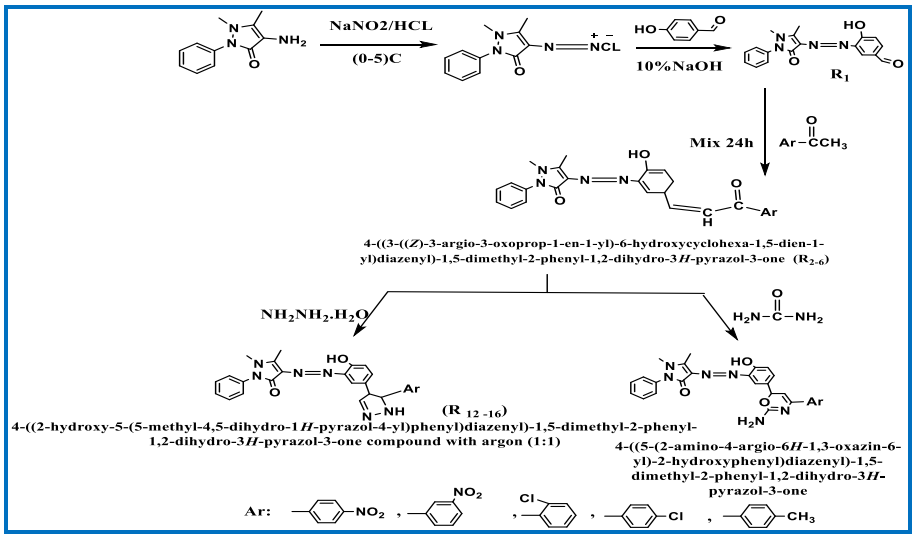
(121°C) after the temperature decreased to about (50°C), then the resulting solution was poured into a glass dish with a thickness of (1-0.5 cm) for each. After that, the solution is left in the incubator for 24 hours until it solidifies.

Preparation of selected compounds. These compounds (R_1, R_2, R_5, R_8, R_9, R_{14}, R_{16}) are prepared by dissolving them using a solvent other than water, which is dimethyl sulfoxide, at a concentration of 200 mg/1ml and 500 mg/1ml.

The bacterial isolates used. In the current research, four kinds of bacteria were identified: Gram-positive bacteria called ***Staphylococcus aurous*** and Gram-negative bacteria (***Proteus mirabilis, Ecoli, Morganella morgani***) and the uses of these types in applying the effectiveness through the metric cylinder method with a cork borer. The process of making holes and spreading certain bacteria on the surface of the dish was performed using sterile cotton. Leave the dishes at a temperature of (37) for (20 minutes) dishes are inserted at (372) for (20min).

3. Result and discussion

The research included the preparation of azo-chalcone derivatives by reacting the Diaz onium salt with 4-hydroxybenzaldehyde to form the aromatic aldehyde derivative, which reacts via Claisen–Schmidt condensation with acetophenone derivatives. The other step involved cyclizing the azo-chalcone derivatives with cyclizing agents such as urea and hydrazine **[30]** according to the Scheme 1.



Scheme 1. Synthesis of compounds ($R_{1\text{-}16}$)

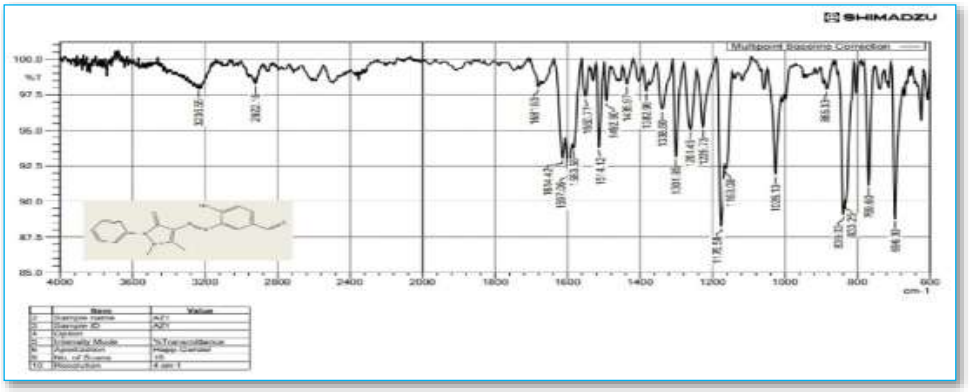


Fig. 1. FTIR spectra of (R_1)

The aromatic aldehyde derivatives (R_1) was identified by FTIR, where it gave,1436 cm^{-1} for the N=N azo group and aldehyde carbonyl group an absorption band appeared at 1681 cm-

1, an absorption band for OH- group of phenol appeared a peak 3236 cm^{-1} for aldehyde stretch **[31] (**Fig. 1). However, the H^1-NMR spectra for (R1) compounds as shown up: 2.44ppm(s,CH_3,3H at CH_3) 3.15ppm(s,CH_3,3H-N-CH_3), 7.35 ppm (s,1H,phenyl ring), 7.37ppm (m,5H,for phenyl ring), 7.71ppm(d.2H.phenyl ring),9.44ppm(s,1H,C=O).9.82ppm(s,1H.OH phenol), as displayed in Fig. 2.

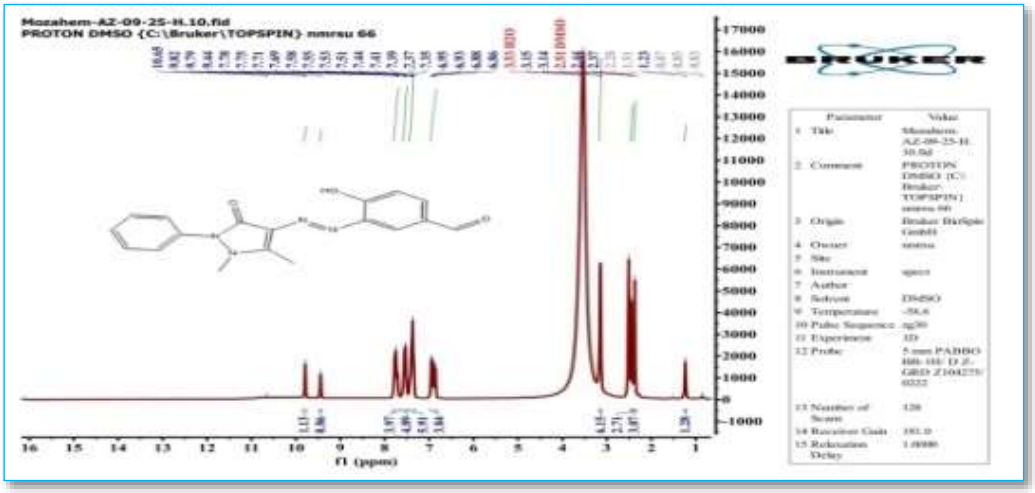


Fig. 2. ^{1}H-NMR spectra of (R_1)

Table 4. FTIR spectra of compounds (R_{2-6})

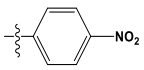
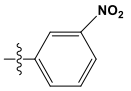
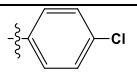
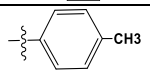
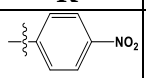
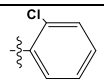
Comp No	Ar	IR.KBr, v(cm^{-1})						
		N=N	C=C	OH	C-H aromatic	C=O	C==C	Others
R_2		1452	1608	------	3048	1654	1413-1579	N-O 1308 Sym 1509 Assy
R_3		1421	1601	3580	3012	1689	1421-1577	N-O 1136 Sym 1499Assy
R_4		1509	1601	3580	3010	1650	1455-1576	C-Cl 746
R_5		1449	1601	3567	3050	1677	1421-1576	C-Cl 700
R_6		1454	1605	3582	3049	1618	1416-1579	

Table 5. The ^{1}H-NMR spectra data were indicated

Comp. No	R	^{1}H-NMR (ppm) DMSA-d^6
R_2		2.42 (5,3H, for CH_3), 3.14 (S, 3H, for N-CH_3) 6.82- 6.85 (m, CH, 1H for aromatic ring), 7.38-7.52(m, CH,1H for aromatic ring), 7.67-(d, CH,2H for methylene group), 9.95 (S, 1H,OH - phenol).
R_5		2.43(s,3H, for CH_3),3.13 (s, 3H, for N-CH_3),6.8 (s,1H,1H for 1-ethylene),7.31-7.75(m,9H,1H for aromatic ring),9.44 (s, 1H, OH-phenol).

As for the compounds (**R** $_{2-6}$), their infrared spectrum showed a clear band at (1509-1421) cm^{-1} for N=N azo-group, and carbonyl group at (1689-1618) cm^{-1}. C-H aromatic at (3050-3010) cm^{-1} and strong bands related at C=C Alkene at (1608-1601) cm^{-1}.

Then, all spectroscopic information about FTIR and ^{1}H-NMR as shown in Tables 4 and 5, respectively, and Fig. 3 and 4.

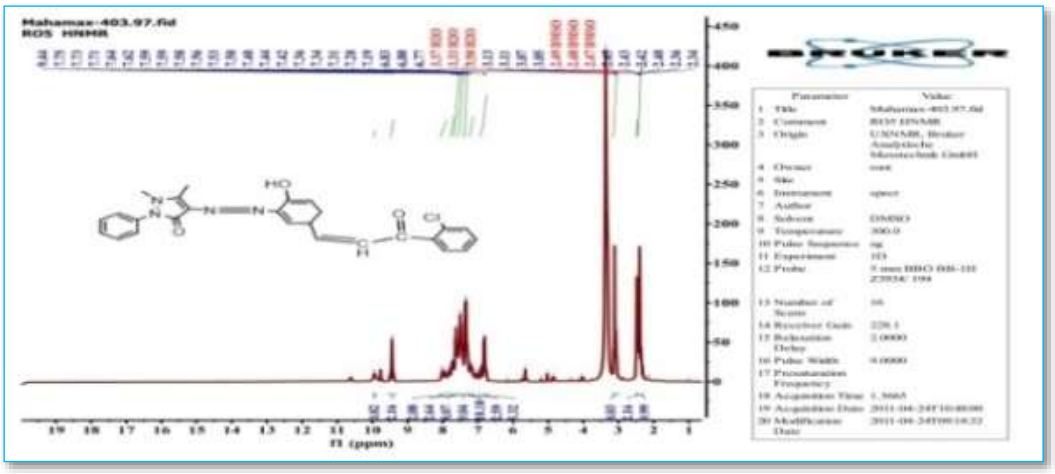


Fig. 3. ^{1}H-NMR spectra of (R_2)

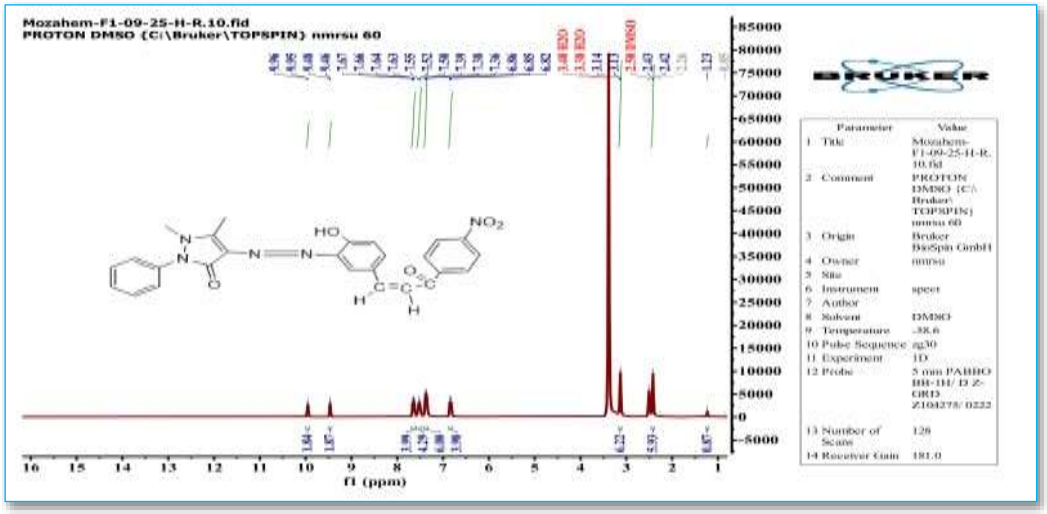
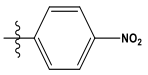
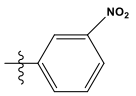
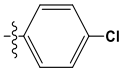
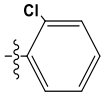
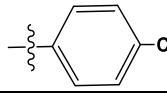


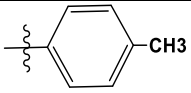
Fig. 4. ^{1}H-NMR spectra of (R_5)

Table 6. FTIR spectra of compounds (R_{7-11})

Comp. No	Ar	IRN cm^{-1}					
		N=N	C=C	C=N	C-H	NH2	Others
R_7		1436	1611	1596	3033	3209	N-O 1341 Sym 1491 Assy.
R_8		1429	1602	1576	3088	3256	N-O 1335 Sym 1470 Assy.
R_9		1458	1616	1597	3033	3216	C-Cl 746
R_{10}		1456	1595	1576	2163	3566	C-Cl 763
R_{11}		1457	1602	1577	--------	3546	

Urea was used to convert chalcone group in compound (**R2-6**) to 1,3-oxazine compounds (**R7-11**) in 10% NaOH via cyclization chalcone group with stir the mixture magnetically for 3-4 hours. These compounds were characterized by FTIR and showed the following main absorption bands (1429 -1458) cm^{-1} for N=N, (2163-3088) cm^{-1} for C=C, and Clear bands appear for the amine group (NH_2) at (3209-3566) cm^{-1}. All spectroscopic information is shown in Tables 6 and 7.

Table 7. The ^{1}H-NMR spectra data.

Comp. No	R	H^1-NMR(PPM)-DMSO-d^6
R_{11}		2.51ppm (s, 3H, forCH_3), 3.13 (S,3H, for N-CH_3), 6.83 (S, 2H , 2H for NH_2), 6.86-7.67 (m,8H , CH for benzene ring), 9.47 (S, ,1H ,OH- phenol)

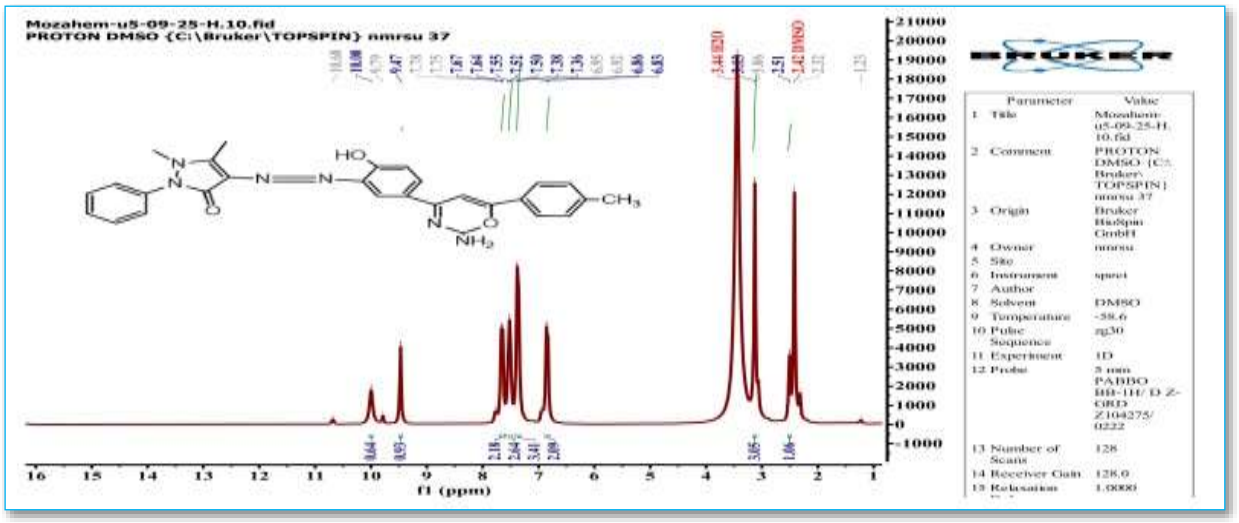
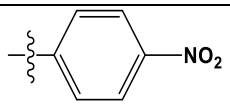
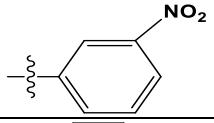
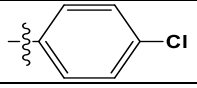
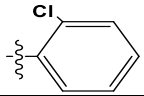
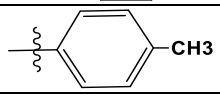


Fig. 5. ^{1}H-NMR spectra to sample (R11)

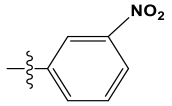
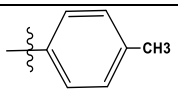
Table 8. FTIR spectra of compounds (R_{12-16})

Comp No	Ar	N=N	C=C	C-H Aliphatic	C=N	N-H	Other
R12		1456	1591	2617	1631	3400	N-O 1373 Sym 1456 Assy
R13		1514	1598	2858	1643	3325	N-O 1377 Sym 1475 Assy
R14		1454	1577	2922	1602	3398	C-Cl/750
R15		1444	1593	2400	1593	3375	C-Cl/750
R16		1498	1579	2953	1600	3325	

Hydrazine hydrate was used to convert chalcone group in compounds (**R2-6**) to pyrazole compounds (**R12-16**) in absolute ethanol and reflexed the mixture for (15 hour) with few drops Glacial acetic acid. These compounds characterized by FTIR and showed the

following main absorption bands (1444-1556) cm^{-1} for N=N, (1577-1598) for C=C, (2400-2617) cm^{-1} for C-H Aliphatic, (1593-1643) cm^{-1} for C=N, and (3325-3400) cm-1 for N-H. All spectroscopic data appeared in Tables 8 and 9.

Table 9. The ^{1}H-NMR spectra data were indicated

Comp No	R	H^1-NMR (ppm)-DMSO-d^6
R_{13}		2.07 (s,3H , for CH_3), 3.86 (s, 3H for N-CH_3), 6.80 - 7.63 (m, 11H, CH for aromatic ring), 8.07 (s,NH,1H for pyrazole ring), 8.41 (s,1H,OH - phenol).
R_{16}		2.43(s,3H , for CH3),3.17 s, 3H for N-CH3),6.83(s,1H,CH for pyrazole ring),7.19-7.46(m,12H,CH for aromatic ring),9.8(s,1H,OH for phenol

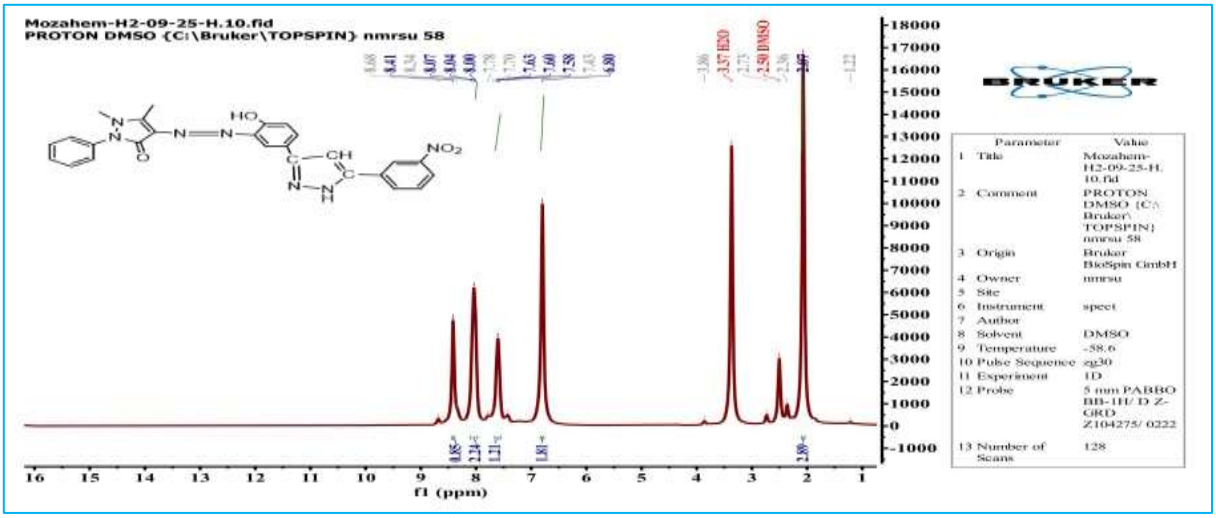


Fig. 6. ^{1}H-NMR spectra to sample (R_{13})

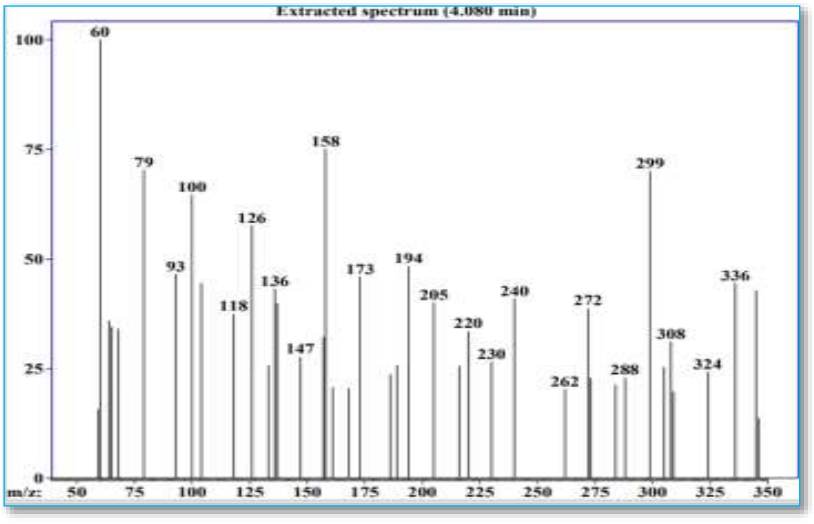


Fig. 7. GCMS of (R_1)

Results of mass spectrometry study:

Using mass spectrometry is considered an important analytical tool **[32]** as a measure the mass-to-charge ratio involving one or more molecules present in the sample. The use of mass spectrometry to calculate the exact molecular weight and identify unknown compounds by determining their molecular weight, and for the purpose of proving the structural formula **[33]** of the prepared

compounds, the compounds (**R_1, R_4, R_5, R_{10}**) were chosen and a gas-mass chromatography was measured for them, where the fractionation showed their exact molecular weights, which are equal to the molecular weights calculated theoretically. Where the compound (R_1) gave the mass/charge ratio (M/Z) at (336) with a main fragmentation bund base peak (60) and the compound (R_{13}) gave (M/Z)=(495) with a base peak (45) at abundance 100%, the compound (R_6) also gave the mass ratio (M/Z) at (452) with a base peak at (107) abandunce 100%, while the compound (**R_8**) gave a ratio (M/Z) at (524) a main or base peak at (44). So, these ratios are considered identical to the molecular weights calculated theoretically, in addition to the other broken units whose masses are listed in Table 10, and the spectral shapes (Fig. 7, 8, and 9) they describe the mass spectrometry of the compounds (**R_1) (R_{13})** and **(R_6)**, respectively.

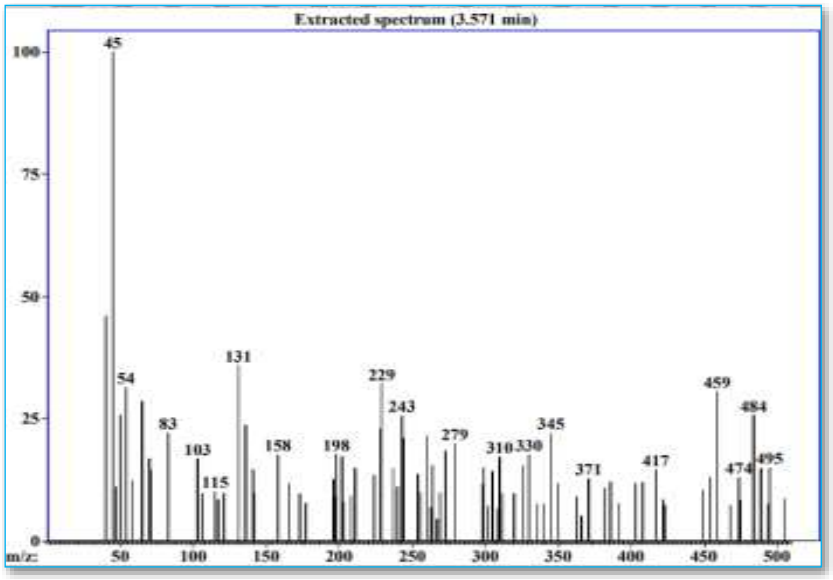


Fig. 8. GCMS of (R_{13})

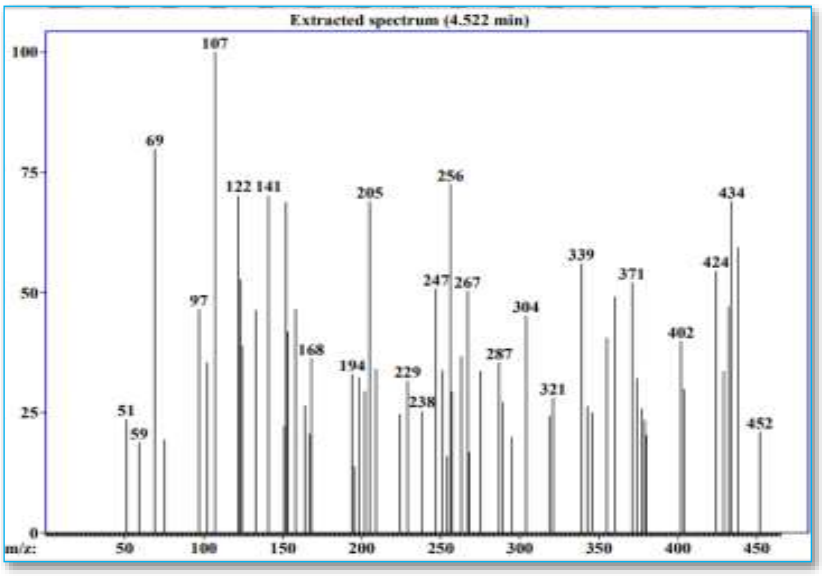
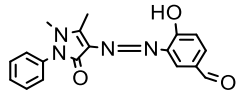
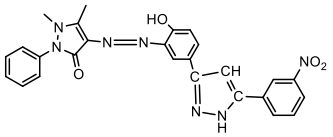
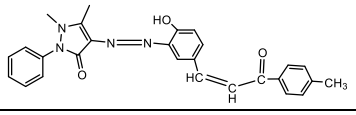
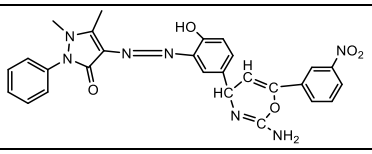


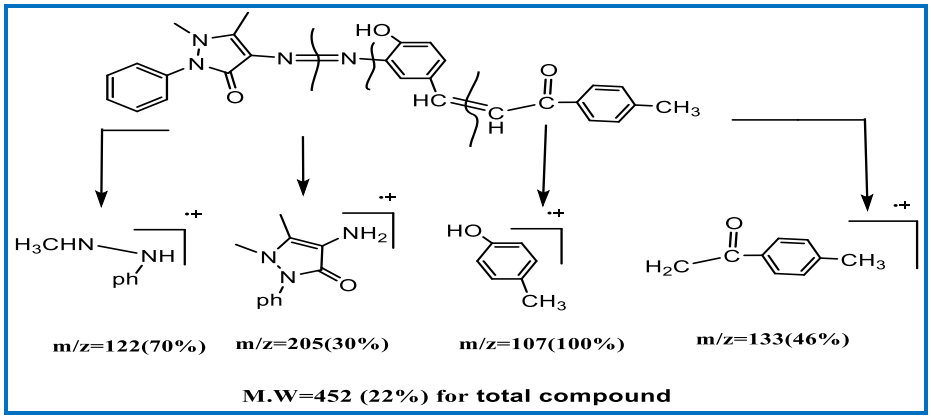
Fig. 9. GCMS of (R6)

Table 10. The mass spectra of the prepared compounds

Comp. No	Structure	m/z value	Base peak

R_1		**336**	**60**
R_{13}		**495**	**45**
R_6		**452**	**107**
R_8		**524**	**44**

Some of the breakdowns of compounds can be interpreted hypothetically and as in the following Scheme 2: example the compound (R_6).



Scheme 2. Proposed fragmentation pattern of compound (R_6)

Antimicrobial activity outcome:

The bacterial activity of some laboratory-prepared compounds (**R_1, R_2, R_5, R_8, R_9, R_{14}, R_{16})** was evaluated Each compound is prepared against one type of bacteria isolated and identified, namely gram-Negative bacteria, there are three types howeve isolated Including ***(Proteus Mirabili, Morganella Morgani, Ecoli***) and ***staphylococcus aurousis*** gram positive bacteria The tested compounds showed high inactivation activity, and some of them were moderate, at a concentration of 500 mg/1ml, while the 200 mg/1ml concentration of the prepared compounds did not give any significant activity. Note that a pilot study was conducted at concentrations less than 200 mg/1ml as mentioned in the literature, and it did not give any significant effectiveness.

The highest effectiveness was observed for the two compounds **(R_{14}, R_{16})**, as they gave an inhibition diameter of (35) (31) and respectively with ***Morganella Morgani*** bacteria and the diameter of inhibition (30) with ***E.coli*** and the Table 11 shows the sensitivity of the bacteria to some of the prepared compound and Fig. 10 below illustrate this.

Table 11. Activity results for the compounds against Bacteria

Comp. No	The symbol of the compound on the plate	Inhibitory Zone (mm) at cosentration 200 mg/1ml	Inhibitory Zone (mm)at concentration 500 mg/1ml			
			Staph aureu	***E.coli***	***Morganella Mirabilis***	***Proteus Mirabilis***
R1	**1**	----------	----------	----------	----------	----------

R2	**6**	----------	----------	----------	**20**	----------
R5	**5**	----------	**22**	**15**	**15**	**20**
R8	**10**	----------	**28**	----------	----------	----------
R9	**9**	----------	----------	----------	----------	----------
R14	**7**	----------	**30**	**30**	**35**	----------
R16	**8**	----------	**25**	**30**	**31**	----------

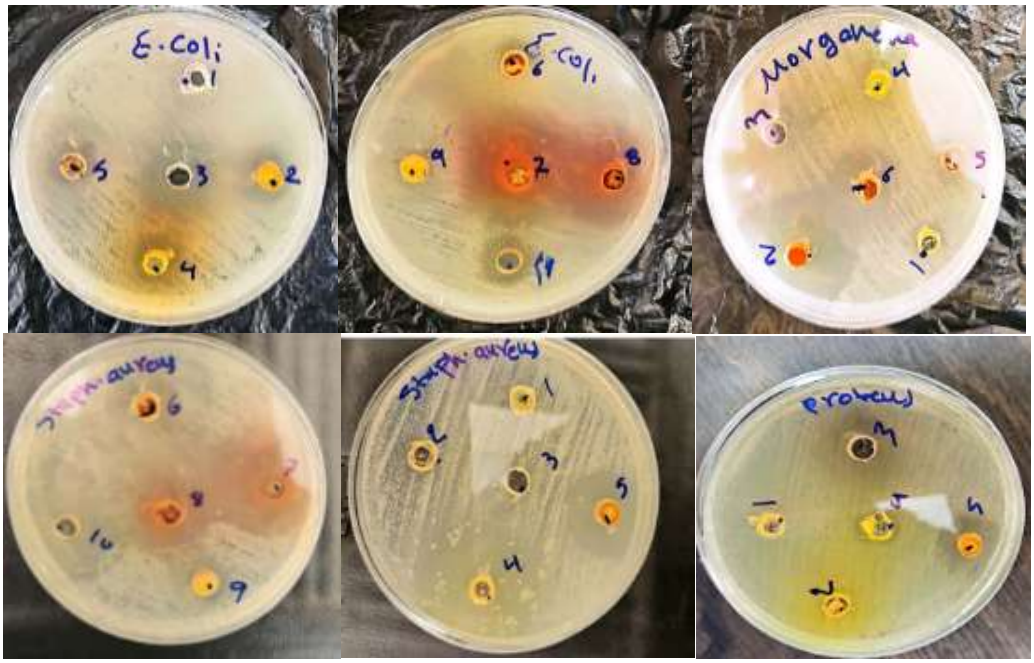


Fig. 10. The anti-bacterial activity of prepared compounds

4. Conclusion

New azo-chalcone derivatives were prepared as heterocyclic compounds and investigated by several important spectroscopic measurements. The biological activity of some of them was also studied, and compounds with high inhibitory activity or strong inhibition were identified. For example, two compounds (**R_{14}, R_{16}**) showed good results against two types of bacteria ***Morganella Morgani*** bacteria and ***E.col***i, respectively, which cause diseases in humans. In light of the phenomenon of bacterial resistance to many antibiotics, these compounds can be considered as alternatives for treatment if they are combined with antibiotics to increase their effectiveness; especially when binding to these compounds, synergy can occur between them and achieve good results. The importance of the research is the use of synthetic or semi-synthetic alternative drugs, since they are used to treat infections caused by these bacteria, such as urinary tract infections, wound infections and burns. Therefore, we recommend that histological and anatomical studies be carried out on the prepared compounds so that they can be used as promising alternatives. We recommend further study of azo-chalcone compounds and preparation of other new lines of them.

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