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SYNTHESIS, CHARACTERIZATION AND THEIR ANTIMICROBIAL ACTIVITIES OF 3,5-DIARYL-4,5-DIHYDRO-N-PHENYL-2-PYRAZOLINES

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ABSTRACT

A novel series of N-phenyl pyrazoline derivatives were synthesized, a mixture of substituted ethoxy chalcones **1(a-h)** and phenyl hydrazine (X) was refluxed for 8 hours in ethanol as a solvent in an acidic medium. The completion of reaction was monitored by TLC and then the reaction mixture was cooled to room temperature and poured into ice cold water the products formed are derivatives of 3,5-diaryl-4,5-dihydro-N-phenyl-2-pyrazolines **2(a-h)**. These synthesized compounds were characterized by FT-IR, ¹H NMR, ¹³C NMR, LCMS, HRMS and elemental analysis. The substituted N-phenyl pyrazoline derivatives showed moderate to potent antimicrobial activities.

Keywords: Chalcones, Pyrazolines, antibacterial and antifungal activities.

1. INTRODUCTION

Pyrazoline is the five member heterocyclic ring structure having two nitrogen and three carbon atoms arranged at first and third position. The substituted pyrazoline and its derivatives has been a subject of consistent interest because of the wide range of applications for the synthesis of heterocyclic compounds in the pharmaceutical and agrochemical industries,¹⁻² and also these analogs represent a significant class of nitrogen-fused heterocycles, which are present in many natural products and biologically active compounds.³⁻⁸ Among their range of properties, the compounds containing a pyrazole scaffold have been shown to exhibit HIV-1 reverse transcriptase and IL-1 synthesis inhibition, as well as antihyperglycemic, antibacterial, sedatives, hypnotic, anti-inflammatory, antipyretic and analgesic activity.⁹⁻¹⁴ (Fig. 1)

Moreover, pyrazoline derivatives have their own importance in aromatic heterocyclic family. Applications of these derivatives in chemistry and biology has attracted increasing interest occupy a unique place in field of medicinal chemistry due to their wide range of biological activities, these nitrogen containing heterocyclic compounds showing activities such as antimicrobial antitubercular and antiinflammatory activities.¹⁵

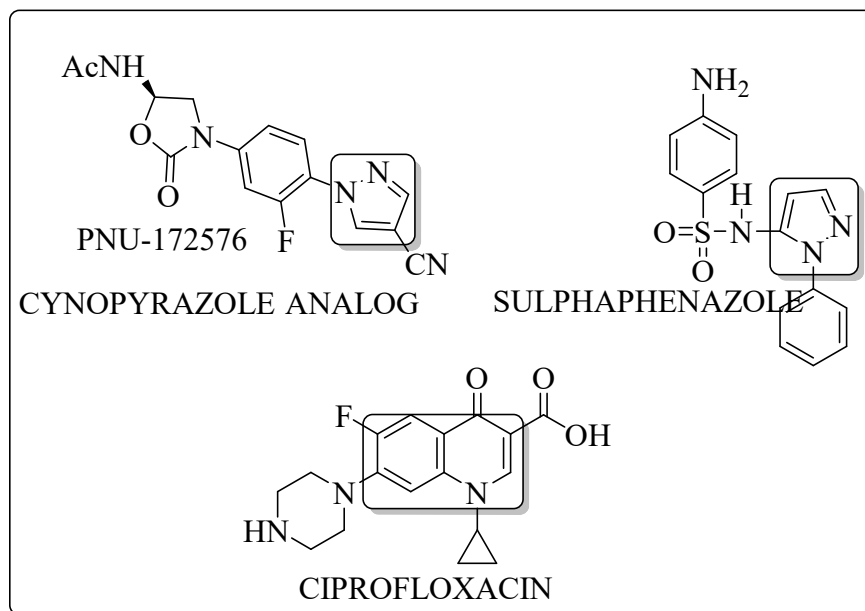


Figure 1 Antimicrobial agents containing Pyrazolines



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2. Material and Methods

Pyrazolines are the well known nitrogen containing heterocyclic compounds, the substituted pyrazoline derivatives in which the substitutions like halogens, electron donating and electron withdrawing groups were occupying the prime position because of their miscellaneous biological applications.

There are numerous synthetic procedures are available for the synthesis of pyrazolines and their derivatives, among them some of them are convenient methods for the synthesis purpose. The simplest and best method for the synthesis of pyrazolines involved the condensation of different derivatives of hydrazine hydrates with α , β -unsaturated carbonyl compounds like chalcones.

In our present investigation, we have synthesized differently substituted derivatives of 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazoline analogues by the reactions of chalcone derivatives with phenyl hydrazine using ethanol as a solvent. These newly synthesized compounds were characterized by IR, ¹H NMR, Mass, and elemental analysis, the newly synthesized phenyl pyrazoline derivatives were screened for antimicrobial analysis.

3.1 Synthesis of 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazolines

In this present work here in we report the synthesis of 3, 5-diaryl-4, 5-dihydro-1-phenyl-2-pyrazolines by treating chalcones with phenyl hydrazine in ethanol in acidic medium.

General method for the synthesis of substituted of 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazolines 2(a-h)

A mixture of substituted ethoxy chalcones (1 mmol) and phenyl hydrazine (3 mmol) was refluxed for 8 hours in ethanol as a solvent in an acidic medium. The completion of reaction was monitored by TLC and then the reaction mixture was cooled to room temperature and poured into ice cold water. The separated solid was filtered, washed several times with cold water and then the mixture of cold ethanol-water to remove impurities, and recrystallized from ethanol.

4. DISCUSSION OF SPECTRA

The IR spectra of the synthesized compounds recorded on a Fourier Transform IR spectrometer (model Shimadzu 8700) in the range of 400-4000 cm⁻¹ using KBr pellets.

¹H NMR spectra recorded on Bruker 400 MHz NMR spectrometer using CDCl₃ and the chemical shifts (δ) reported are in ppm downfield using tetramethylsilane (TMS) as internal reference. The NMR data is reported as follows: chemical shift, multiplicity (s = single, d = doublet, t = triplet, q = quartet, m = multiplet. The coupling constant (J) is given in Hz.

The mass spectra (MS) recorded on EI-SHIMADZU-GC-MS mass spectrometer. The purity of all newly synthesized derivatives was confirmed by elemental analysis (CHN) using thermo finnigan flash EA 1112 Thermo Finnigan with more than 95%. The melting points were determined open capillary method and are uncorrected.

Scheme-II

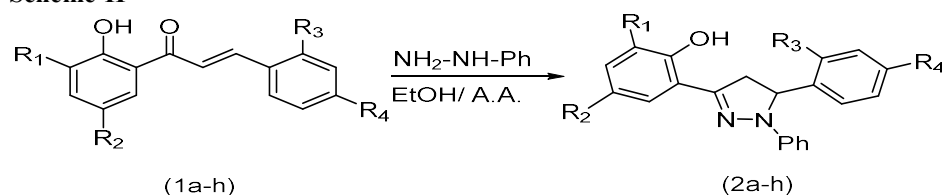


Table-1: Substituted data of synthesized substituted 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazoline derivatives:

Sr. No.	R ₁	R ₂	R ₃	R ₄	Comp	Sr. No.	R ₁	R ₂	R ₃	R ₄	Comp
1	Br	Cl	H	OEt	2a	5	Br	Cl	OEt	H	2e
2	Br	CH ₃	H	OEt	2b	6	Br	CH ₃	OEt	H	2f
3	I	Cl	H	OEt	2c	7	I	Cl	OEt	H	2g
4	I	I	H	OEt	2d	8	I	I	OEt	H	2h



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6.0 EXPERIMENTAL

Synthesis of 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazoline derivatives (2a-h)

The derivatives of 3,5-diaryl-4,5-dihydro-N-phenyl-2-pyrazoline (**2a-h**) were synthesized from the condensation of substituted derivatives of ethoxy chalcones (**1a-h**) (1 mmol) was taken in RBF with phenyl hydrazine (3 mmol) in 10 ml ethanol in an acidic medium using glacial acetic acid mixture was refluxed for 8 hours, after completion of reaction was monitored by TLC.

The reaction mixture was cooled to room temperature and poured into ice cold water. The separated solid product was filtered, washed several times with the mixture of cold ethanol and cold water to remove impurities and recrystallized by using ethanol to afford pure 3,5-diaryl-4,5-dihydro-N-phenyl-2-pyrazoline compounds (**2a-h**) the structures of purely synthesized compounds were confirmed by IR, ¹H NMR and Mass spectral analysis.

Table 2: The physical data of synthesized compounds of N-phenyl-2-Pyrazoline derivatives (2a-h)

Sr. No.	Entry	Molecular formula	Yield In %	Melting Point °C
1	2a	C ₂₃ H ₂₀ BrClN ₂ O ₂	78	152
2	2b	C ₂₄ H ₂₃ BrN ₂ O ₂	70	145
3	2c	C ₂₃ H ₂₀ ClIN ₂ O ₂	71	167
4	2d	C ₂₃ H ₂₀ I ₂ N ₂ O ₂	77	185
5	2e	C ₂₃ H ₂₀ BrClN ₂ O ₂	82	160
6	2f	C ₂₄ H ₂₃ BrN ₂ O ₂	81	170
7	2g	C ₂₃ H ₂₀ ClIN ₂ O ₂	85	186
8	2h	C ₂₃ H ₂₀ I ₂ N ₂ O ₂	80	185

7.0: Result and Discussion

Discussion: Spectral interpretation of 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazolines (2a-h)

IR Spectra of 3,5-diaryl-4,5-dihydro-N-phenyl-2-pyrazolines

The IR spectra of selected products showed the absorption bands in the region of 1590-1600 cm⁻¹ due to -C=N- of pyrazolines, a band in the range between 1370-1355 cm⁻¹ indicates the presence of (-OCH₂CH₃) group. The absorption band in the region 3085-3070 cm⁻¹ is due to -OH stretching. The bands 670-645 cm⁻¹ due to C-Cl stretching, 755-720 cm⁻¹ due to C-Br appears whenever present in the respective compound. All these observations are in agreement with those observed earlier.²¹⁻²⁸

¹H NMR spectra

¹H NMR spectra revealed that the three protons H_A, H_B and H_X attached to C-4 and C-5 carbon atoms of the pyrazol ring gave an ABX spin system and they appeared as doublet of doublet. The methylene proton of pyrazoline ring (H_A, H_B) exhibited a typical ABX spin system with H_X as a doublet of doublet.

The doublets of H_A appeared in the region δ 3.35-4.02 ppm, the doublet of doublet of H_B appeared in the region δ 3.24-3.96 ppm and H_X appeared in the region δ 5.48-5.52. Among these ABX protons H_X is the most deshielded due to close proximity due to benzene ring and they appeared as doublet of doublets.

In the pyrazoline ring as there is the presence of two non-equivalent protons of a methylene group (H_A / H_B) in the range δ 3.35-4.02 ppm and δ 3.24-3.96 ppm, coupled with each other and in turn with the vicinal methine proton (H_X) at δ 5.48-5.52 ppm were present. The rest of the protons of like aliphatic and aromatic were anticipated at respective places.

The quintet for -CH₃ of ethoxy group appeared in the region δ 1.23-1.31 ppm and the triplet of -CH₂ for ethoxy group appeared in the region δ 3.96-4.05 ppm. The compilation of signals multiplet of aromatic hydrogen appeared in the region δ 6.80-7.61 ppm and the exchangeable proton of ortho hydroxyl group appeared as a 1H singlet in the region δ 10.14-11.35 ppm. While the protons belonging to the aromatic ring and aliphatic protons were observed at expected regions. The desire peaks were observed as per the earlier findings of literature values.²⁹⁻³⁴



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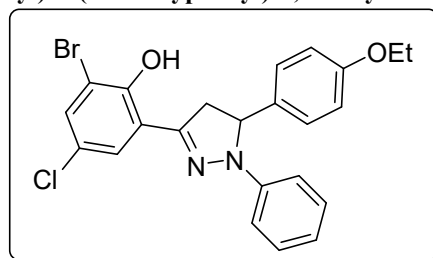
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Mass spectra

The mass spectra of synthesized products (N-Phenyl-2-pyrazolines) are also in agreement with their molecular formulae weights.

Spectral data of synthesized compounds

1. 3-(3-bromo-5-chloro-2-hydroxyphenyl)-5-(4-ethoxyphenyl)-4, 5-dihydro -1-phenyl-2-pyrazolines (2a):



IR (KBR cm^{-1}): 3383, 1610, 1355, 809, 505 cm^{-1} ;

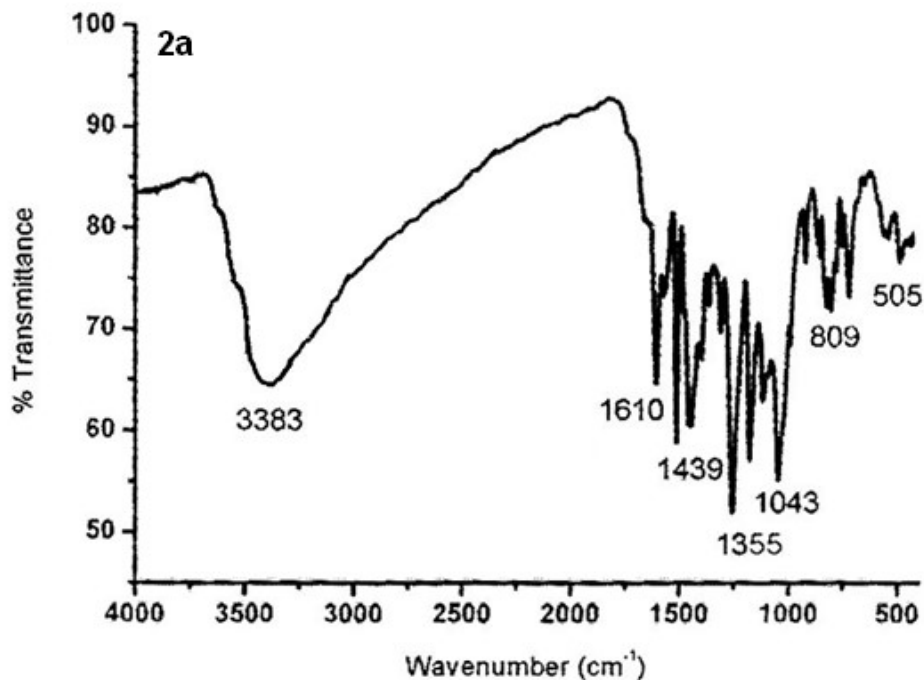
$^1\text{H NMR}$ (400 MHz, δ 10.14 (s, 1H, OH), 6.83-7.45 (m, 11H, Ar-H), 5.48- (CDCl₃, ppm) 5.53 (t, 1H, H_X), 4.02-4.07 (q, 2H, -OCH₂), 3.98-4.02 dd, 1H, H_A), 3.94-3.99 (dd, 1H, H_B), 1.27-1.30 (t, 3H).

$^{13}\text{C-NMR}$ (CDCl₃): δ 162.2, 156.1, 150.9, 143.5, 135.9, 134.8, 130.3, 130.1, 129.9, 129.8, 126.1, 125.9, 123.6, 121.3, 117.3, 117.2, 115.8, 115.4, 115.3, 65.2, 60.6, 40.6, 14.4;

LCMS (m/z): 471.

CHN analysis: Calculated for C₂₃H₂₀BrClN₂O₂: C, 58.56; H, 4.27; N, 5.94; found C, 58.65; H, 4.23; N, 4.89.

IR spectra of:- 2a



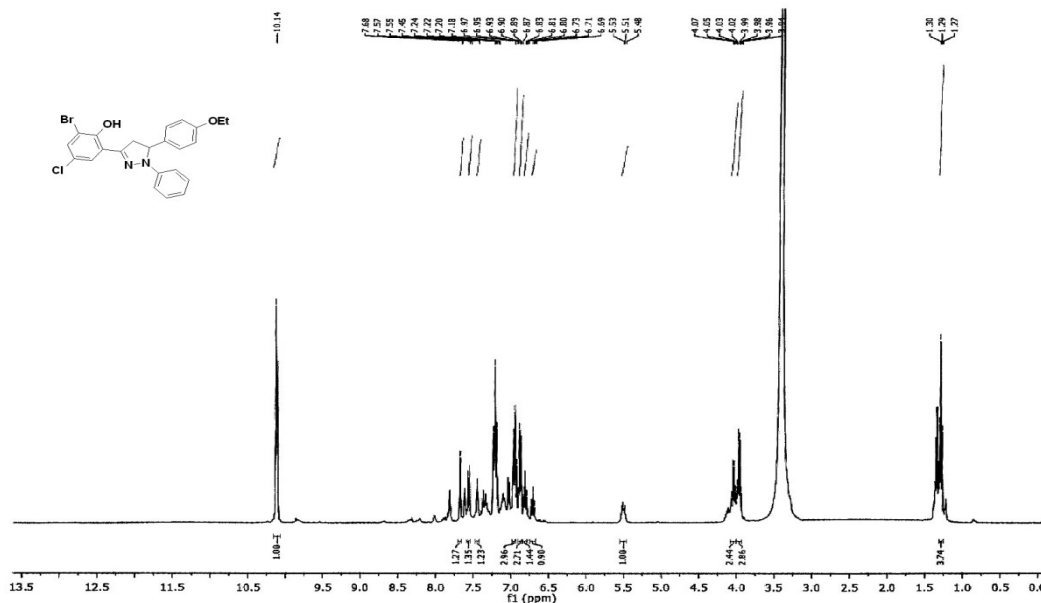


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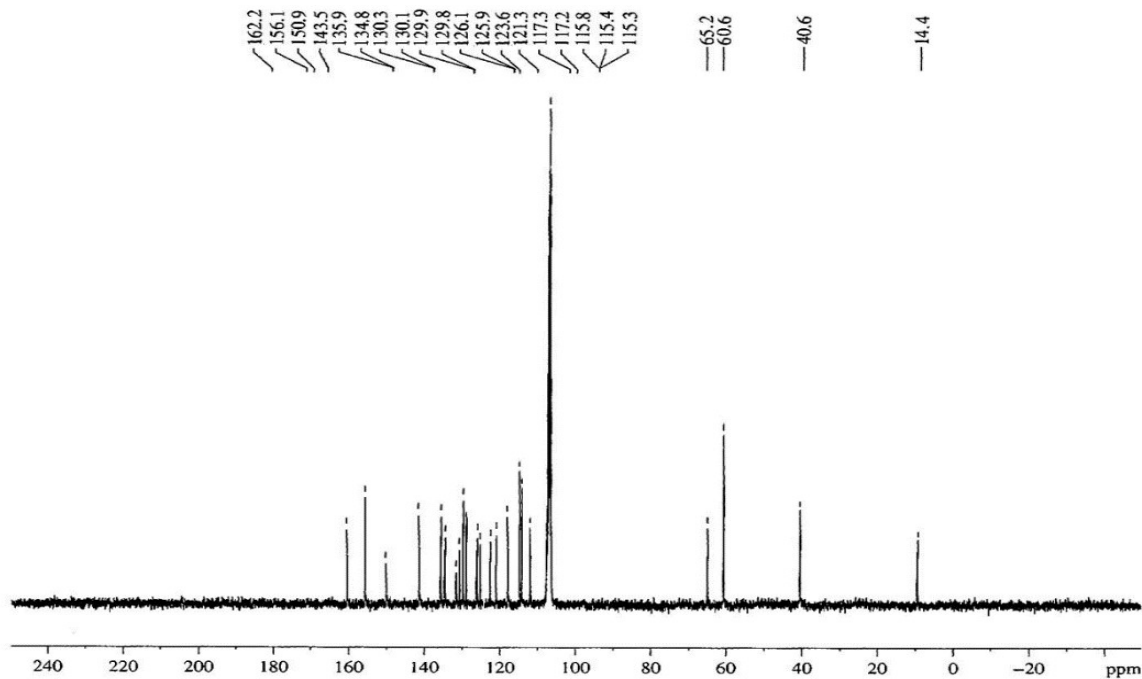


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¹H-NMR spectra of: 2a



¹³C-NMR spectra of: 2a



LCMS spectra of: 2a



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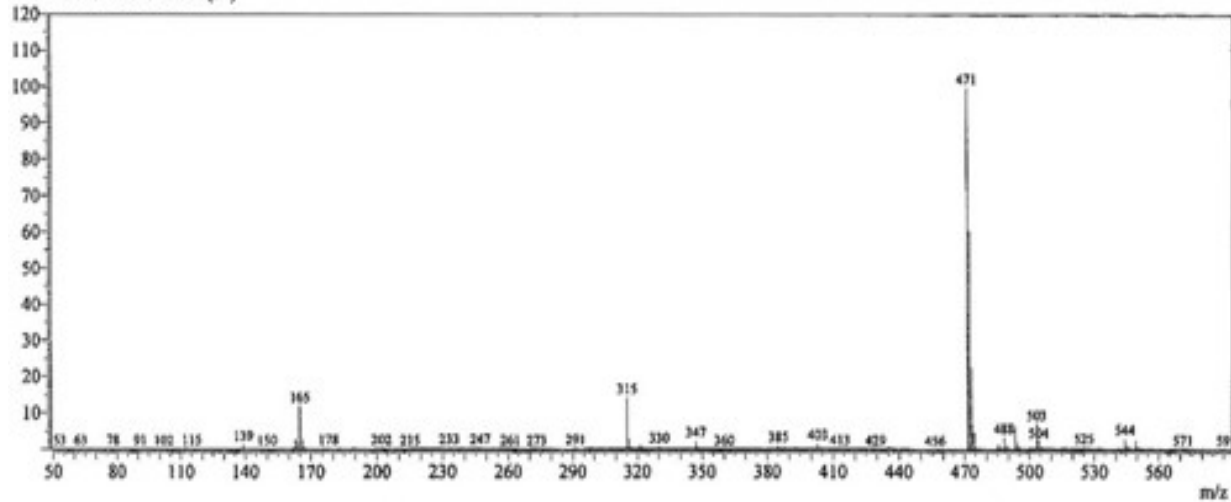
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LCMS-2010A DATA REPORT SHIMADZU

User : Admin
 Sample : 2a
 Inj. Volume : 5.000
 Data Name : E:\Data\AXPH-APCI-POS1.qld
 Method Name : C:\LCMSsolution\User\Method\esi.qlm

MS Spectrum

Line#1 R.Time:0.765(Scan#:46) Positive
 MassPeaks:370 BasePeak:471.30(15473457)
 RawMode:Single 0.765(46)
 BG Mode:Peak Start 0.560(34)



MS Peak Table

Peak#	R.Time	I.Time	F.Time	Area	Height	A/H	Mark	%Total	Name	Base m/z	Base Int.
1	0.765	0.560	1.143	678285523	37263095	18.20		100.00		471.30	15473457
				678285523	37263095			100.00			



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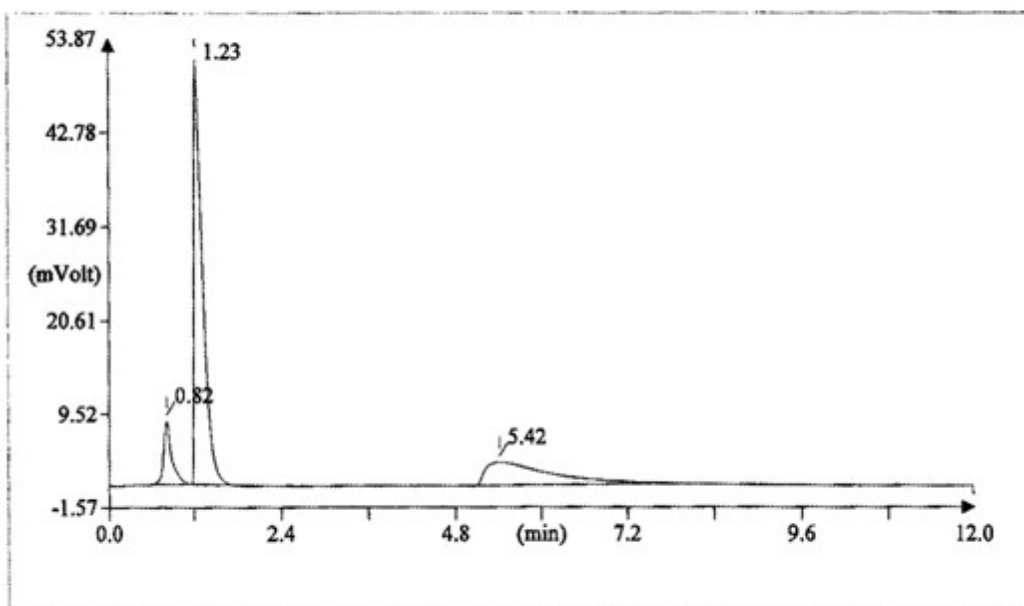


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CHN analysis spectra of: 2a

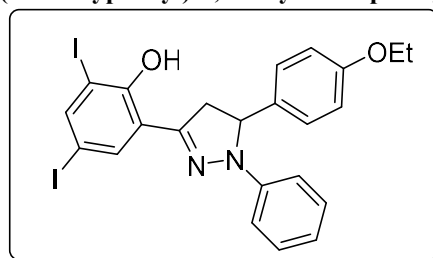
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Chromatogram filename: UNK-25102017-1.dat
Sample weight: 1.312



Element Name	Element %	Ret. Time
Nitrogen	4.89	0.82
Carbon	58.65	1.23
Hydrogen	4.23	5.42

2. 3-(2-hydroxy-3,4-diiodo-phenyl)-5-(4-ethoxyphenyl)-4,5-dihydro-N-phenyl-2-pyrazoline (2d):



IR (KBR cm⁻¹): 3090, 1595, 1330, 780, 620 cm⁻¹

¹H NMR(CDCl₃): δ 11.73 (s, 1H, OH), 7.03 - 7.63 (m, 11H, Ar-H),
(400 MHz, ppm) 5.63-5.65 (t, 1H, H_X), 4.04-4.17 (q, 2H, -OCH₂),
3.96-4.00 (dd, 1H, H_A), 3.64-3.65 (dd, 1H, H_B),



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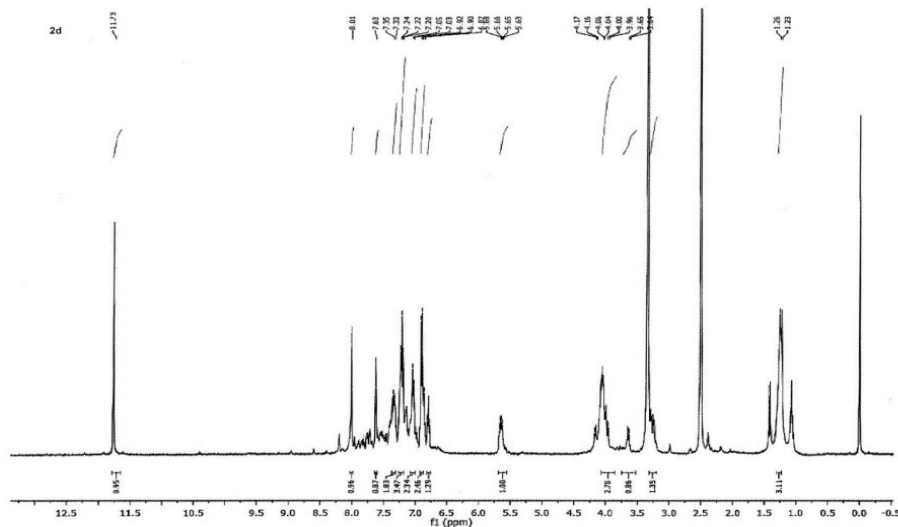
1.23-1.26 (t, 3H).

¹³C NMR(CDCl₃) : 157.6, 157.4, 151.1, 143.9, 142.9, 134.2, 133.2, 130.3, 129.9, 129.8, 120.5, 121.9, 121.8, 116.7, 117.7, 117.6, 115.3, 115.2, 88.2, 83.8, 65.1, 40.2, 14.6.

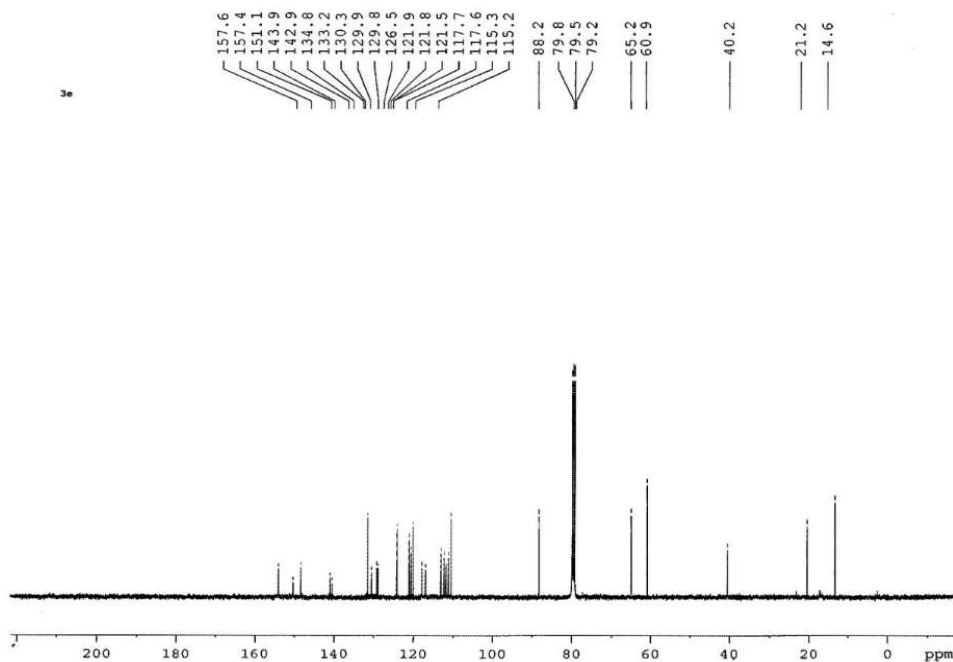
M. S. (m/z) : 609 [M - 1]

CHN analysis : Calculated for C₂₃H₂₀I₂N₂O₂, C, 45.27; H, 3.30; N, 4.59.
Found: 45.30; H, 3.27; N, 4.55.

¹H-NMR spectra of : 2d



¹³C-NMR spectra of : 2d





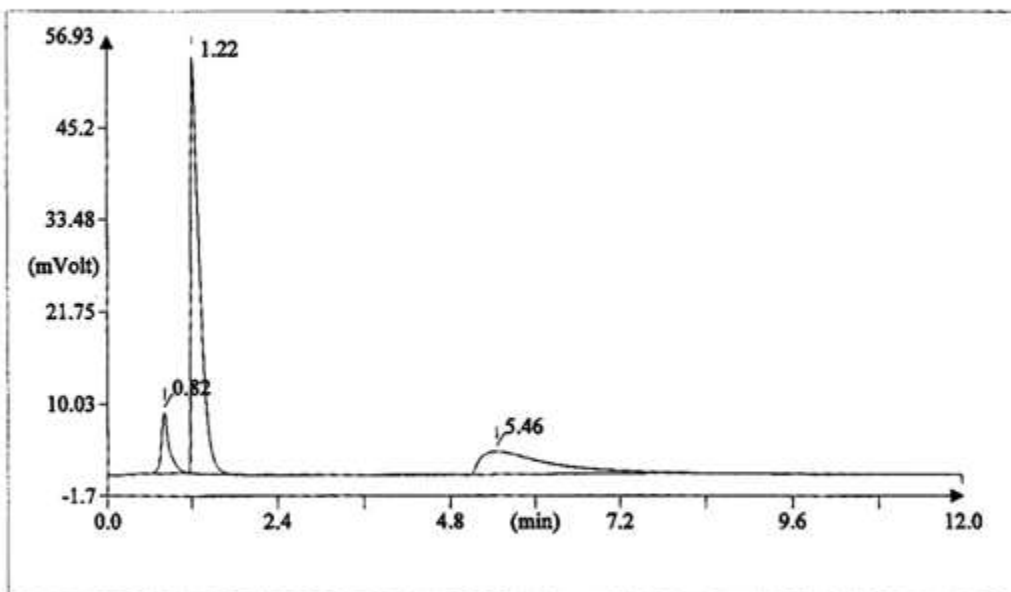
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Chromatogram filename: UNK-25102017-2.dat
Sample weight: 1.326



Element Name	Element %	Ret. Time
Nitrogen	4.55	0.82
Carbon	45.30	1.22
Hydrogen	3.27	5.46

8.0 Antimicrobial activities

The microbes are found all over around us, the presences of microbes on our body are more than microbial cells presents in our body cells. They found in water streams and on the rocks, on smartphone screen, on bikes, on cars, on the laptops and on the food as well in the food. The way the microbes are having the bad status but among them some are neutral and are essential for the living organisms.

The microorganisms are excessively small in size and we cannot see with the naked eyes they are the bacteria, fungi, and viruses. In our day today life we come across two types of microbes or bacteria they are resident bacteria and transient bacteria. The resident bacteria are the part of our body as they reside on skin and gut and acting as the first line of defense against the dangerous transient bacteria. The transient bacteria are the temporary bacteria we can come across by touching the door handle, on sofa sheets, bed sheets or in crowd when somebody sneezes nearby us.

The existence of resident bacteria the body will prevent from settling in andcausing an infection from transient bacteria. The treatments of diseases caused by microorganisms by using the chemicals to prevent or to kill the microorganisms are called the antimicrobial agents. However, some microorganisms are the essential part of our day today life the dairy products such as cheese, butter are impossible without microorganisms due to fermentation process by the microorganisms it gives that products the characteristics texture and test, the tart taste of butter is only due to the microorganisms only. The drinks we enjoy are dueto the yeasts



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we get the alcohol in the beverages like beer and wines. The plant needs bacteria from the soil to grow properly.

Bacterial diseases

Bacteria are microscopic, single-cell organisms and found everywhere and anywhere, they found in each climate and places on the earth. Among them some are airborne while some bacteria found in water or soil. Bacteria are present in three basic shapes rod-shaped called as bacilli, spherical means cocci, and the last one in helical shape that is spirilla. These are classified as gram-positive or gram-negative bacteria. Some of the deadly diseases like anthrax, typhoid fever, plague, tuberculosis, leprosy, gangrene, tetanus, gonorrhea are controlled and some of them irradiated but still we are facing illness due to pneumonia, meningitis, and food poisoning due to the harmful bacteria and several of them are fatal.

The medicinal chemistry is boon for fighting the various bacterial diseases. Still, we need new antimicrobial drugs because some microbes have developed resistant towards the antimicrobial agents.

Fungal diseases

We the human beings on the earth are surrounded by millions of different fungi. These are present in soil, plants and trees outdoors on indoor surfaces and on animal skins. A few of the fungi are not dangerous to mankind but some are harmful to human beings. Here we highlight some fungal diseases, Aspergillosis caused by fungi *Aspergillus* and people get affected by lung diseases or weaken immune systems. Blastomycosis caused by fungus *Blastomyces*, are present in the humid soil or nearby humid areas.

Candidiasis caused by the yeast it occurs in the mouth and throat, vagina, or the bloodstream. The fungal disease like athlete's foot is the infection of the skin between the toes caused by the fungus *Trichophyton*, Mold allergies generally caused by mold spores and ringworm is a skin infection caused by the fungus *Trichophyton*. The fungi causes eye infections the eye infections are rare but can develop after an eye injury or some times after surgery. Due to fungi three different illnesses are facing they are poisonings, parasitic infections, and allergies.

Now a day the chemists are engaged in developing the new drugs to fight bacterial and fungal infections. At present there is a great scope for discovery of innovative drugs having better remedial actions against the diseases caused by bacteria and fungi. These newly synthesized drugs are tested for the antimicrobial activities.

8.2.3 Antimicrobial assay of the compounds

As in the past and current reviews of literatures the synthesized derivatives of pyrazoline like 3-(3-bromo-5-chloro-2-hydroxyphenyl)-5-(4-ethoxyphenyl)-4,5-dihydro-1-phenyl-2-pyrazolines (**2a**) and 3-(2-hydroxy-3,4-diiodo-phenyl)-5-(4-ethoxyphenyl)-4,5-dihydro-1-phenyl-2-pyrazoline (**2d**) are acting as potentially useful bactericidal and fungicidal agents.³⁵⁻⁴⁰

The selected derivatives of pyrazolines (**2a-h**) were screened for the antibacterial activity gram(+ve) bacteria *Staphylococcus aureus*, *Bacillus subtilis* and against gram(-ve) bacteria *Escherichia coli* (E. coli), *Salmonella typhi* by using Agar cup method the assay was conducted by using the Nutrient Agar, here we have used Penicillin as standard drug for reference.⁴¹⁻⁴²



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Table 2: Antimicrobial activity of synthesized 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazolines (2a-h)

Sr. No	Entry	Antibacterial activity (Zone of Inhibition in mm)				Antifungal activity (Zone of Inhibition in mm)			
		A	B	C	D	E	F	G	H
1	2a	20	21	20	15	-ve	-ve	-ve	-ve
2	2b	18	14	15	14	-ve	-ve	-ve	RG
3	2c	21	12	12	13	-ve	-ve	RG	RG
4	2d	17	11	14	13	-ve	-ve	-ve	-ve
5	2e	13	11	12	12	-ve	-ve	RG	RG
6	2f	14	09	20	18	-ve	-ve	-ve	RG
7	2g	19	11	22	18	-ve	-ve	RG	-ve
8	2h	16	12	18	10	-ve	-ve	-ve	RG
+ve Control DMSO		-ve	-ve	-ve	-ve	+ve	+ve	+ve	+ve
Penicilline		12	20	34	22	X	X	X	X
-ve Control (Griseofulvin)		X	X	X	X	-ve	-ve	-ve	-ve

Zone of Inhibition in mm)

A = Escherichia coli, B = Salmonella typhi, C = Staphylococcus aureus, D = Bacillus subtilis E = Aspergillus niger, F =penicillium chrysogenum, G = Fusarium moneliforme, H = Aspergillus flavus -- = No Antibacterial activity, RG = Reduced Growth (Moderate Activity)

-ve = Growth (Antifungal Activity Observed), X = Not Applicable

9.0 Conclusion

In this work, we have synthesized 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazoline **2(a-h)** derivatives using simple experimental procedure with high yields, relatively short reaction time, easilywork up and low cost. These synthesized derivatives of pyrazolines were screened for their antibacterial and antifungal activities. 3, 5-diaryl-4, 5-dihydro-N-phenyl-2-pyrazoline derivatives are showing moderate to potent antimicrobial activities. The result got through the synthesis and characterizations of derivatives of pyrazolines and their antifungal and antibacterial activities are useful for drug designing and their medicinal applications.

References

1. Katritzky A R, Rees C W, Scriven E F, Comprehensive Heterocyclic Chemistry II, **1996**, 3, 1.
2. Katritzky A R, Wang M, Zhang S, Voronkov M V, Steel P J, J. Org. Chem., **2001**, 66, 20, 6787-6791.
3. Kamalakar G, Komura K, Sugi Y, Ind. Eng. Chem. Res., **2006**, 45, 18, 6118-6126.
4. Weingaertner H, Franck E U, Angew. Chem. Int. Ed Engl., **2005**, 44, 18, 2672-2692.
5. Sheldon R, Chem. Commun., **2001**, 23, 2399-2407.
6. Zhao H, Malhotra S V, Aldrichim Acta., **2002**, 35, 3, 75-83.
7. Wasserscheid P, Keim W, Angew. Chem. Int. Ed., **2000**, 39, 3772-3789.
8. Welton T, Chem. Rev., **1999**, 99, 8, 2071-2084.
9. Kees K L, Fitzgerald J J, Steiner K E, Mattes J F, Mihan B, Tosi T, Mondoro D, McCaleb M L, J. Med. Chem., **1996**, 39, 20, 3920-3928.



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10. Genin M J, Biles C, Keiser B J, Poppe S M, Swaney S M, Tarpley W G, Yagi Y, Romero D L, J. Med.Chem., **2000**, 43, 5, 1034-1040.
11. Manfredini S, Bazzanini R, Baraldi P G, Guarneri M, Simoni D, Marongiu M E, Pani A, Tramontano E, Colla P L, J. Med. Chem., **1992**, 35, 5, 917-924.
12. Jungheim L N, Tetrahedron Lett., **1989**, 30, 1889-1892.
13. Dannhardt G, Laufer S, Curr. Med. Chem., **2000**, 7, 11, 1101-1112.
14. Penning T D, Talley J J, Bertenshaw S R, Carter J S, Collins P W, Docter S, Graneto M J, Lee L F, Malecha J W, Miyashiro J M, Rogers R S, Rogier D J, Yu S S, Anderson G D, Burton E G, Cogburn J N, Gregory S A, Koboldt C M, Perkins W E, Seibert K, Veenhuizen A W, Zhang Y Y, Isakson P C, J. Med. Chem., **1997**, 40, 9, 1347-1365.
15. Salyens A A, Amiable C F, Antimicrob Agents Chemother, **1997**, 41, 11, 2321-2325.
16. Karrouchi, Khalid, Ramli, Youssef, Ramli, Youssef, Taoufik, J. Mabkhot, Yahia, Al-Aizari, Faiz, Ansar, M'Hammed, Molecules – vol. 23, **2018**.
17. Ebenezer O, Shapi M, Tuszynski JA. Biomedicines. **2022**, 10(5):1124. doi: 10.3390/biomedicines10051124. PMID: 35625859; PMCID: PMC9139179.
18. Ajay Kumar., et al. Acta Scientific Pharmaceutical Sciences, 5.11, **2021**, 59-69.
19. Mathew AT, Chandran M, Elias G et.al. International Journal of Research and Review. **2021**; 8(10): 174-183. DOI: <https://doi.org/10.52403/ijrr.20211022>.
20. Pramod P. Kattimani, Ravindra R. Kamble, Aravind R. Nesaragi, Mahadevappa Y. Kariduraganavar, Shrinivas D. Joshi, Suneel S. Dodamani & Sunil S. Jalalpure, **2021**, SyntheticCommunications, 51:20, 31253140, DOI: 10.1080/00397911.2021.1964530.
21. Khode S V, Maddi P, Aragade M, Palkar P K, Ronad S, Mammedesai A H, Thippeswamy M, Sayanarayana D, Eur. J. Med. Chem., **2009**, 44, 4, 1682-1688.
22. Mokle S S, Vibhute A Y, Khansole S V, Zandge S B, Vibhute Y B, Research J. Pharm, Bio, Chem, Sci., **2010**, 1, 3, 631-638.
23. Chimeni F, Fioravanti R, Balasco A, Manna F, Chemeni P, Secci D, Rossi F, Turini P, Ortuso F, Alcaro S, Cardia M C, Eur. J. Med. Chem., **2008**, 43, 2262-2267.
24. EI-Sabbagh O I, Baraka M M, Ibrahim S M, Pannecouque C, Andrei G, Snoeck R, Balzarini J, Rashad A A, Eur. J. Med. Chem., **2009**, 44, 3746-3753.
25. Sohar P, Csampai A, Perjesi P, Arkivoc, **2003**, V, 114.
26. Sohar P, Perjesi P, Tornroos K W, Husebye S, Vertes A, Vankoand G Y, Bozak R E, J. Mol. Struct., **2000**, 524, 297.
27. Hassan S Y, J. Braz. Soc., **2011**, 22, 7, 1286-1298.
28. Bajia B, Srivastava Y K, E-Journal Chem., **2007**, 4, 2, 187-191.
29. Kabli R A, Khalaf A A, Zimaity M T, Khalil A M, Kaddah A M, Airifaie H A, J. Ind. Chem. Soc., **1991**, 68, 47.
30. Joshi M G, Wadodkar K N, Ind. J. Chem.,**1981**, 20B, 1090.
31. Basaif S A, Albar H A, Faidallah H M, Ind. J. Hetetrocyl. Chem., **1995**, 5, 121.
32. Mishriky N, Asaad F M, Ibrahim Y A, Girgis A S, Pharmazie,**1996**, 51, 544.
33. Holla B S, Mithun M M, Karegoudar A P, Phosphorus, Sulfur and Silicon., **2006**,181, 1427-1436.
34. Konda S G, Ph.D., Thesis Submitted to S.R.T.M. University, Nanded, India, **2009**.
35. Simon N, Larsen M, Thomas F, Kristian B, Schnning, Kromann, Hasse, J.Med. Chem., **2005**, 48, 7, 2667-2677.
36. Chikhalia K, Patel H, Mayank J, Vashib, Dhaval B, ARKIVOC xiii, **2008**,189-197.
37. Babasaheb P B, Sachin A P, Balaji L K, Shivraj H N, Chandrahase N K, a. Eur. J. Med. Chem., **2010**, 45, 2629.
38. Tanvir H, Hamid L S, Muhammad Z R, Muhammad M Y, Masood P, Eur.J. Med. Chem., **2009**, 44, 4654.
39. Bondock S, Fadaly W, Metwally M A, European Journal of MedicinalChemistry, **2010**, 45, 9, 3692-3701.
40. Shah N N, Ziauddin H M, Zameera M, Hingole S S, Baseer M A, J. Chem.Pharm. Res., **2010**, 2, 6, 441-445
41. Barry A L, Biol Abstr., **1976**, 64, 180, 25183.
42. In: Microbiological assays and tests, Indian Pharmacopoeia, Ministry of Health and Family Welfare, The Controller of Publications, New Delhi., A100 (**1996**).