Microwave Green Synthesis of Pyrazolo-Pyrimidine Derivatives and Biological Profile Evaluation

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Abstract **-** Pyrazolo-Pyrimidine structural moieties are the crucial elements for the preparation of aimed drugs which are biologically various numerous importance features indirectly encourages budding scientists to synthesis other pyrazoles-pyrimidine derivatives with similar behavior but a better medicinal action including antimicrobial, anti-tumor, anti-inflammatory applications. Nitrogen containing heterocyclic cyclic synthetic organic researchers have focused towards green methods of synthesis by avoiding hazardous reaction steps which depend on organic solvents by the replacement of eco-friendly solvents. Microwave green synthesis creates a new stair in heterocyclic synthesis. Desire molecules were obtained the reaction between 1,3 di-ketones and hydrazine hydrates and urea/ thiourea with different aromatic aldehyde derivatives by using ethanol medium. New structures are being confirms with the support analytical techniques of elemental analysis(CHN) and spectral analytical data: FTIR, Proton(^{1}H) and Carbon (^{13}C) NMR spectra. All new moieties were screened for biological potential analysis: All the proposed products show moderate to excellent antimicrobial, activity against different gram +ve (st-aureus, bacillus-subtillis) and Gram –ve (E-coli) microbial strains. Meanwhile anti-fungal evaluation as regards fungus infection to man kinds and pets.

Index Terms: Pyrazolo PyrimidineMoeties,1,3, diketones, Hydrazine hydrates, Urea/thiourea.

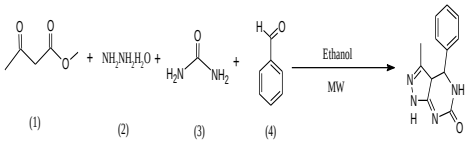
1. INTRODUCTION: Mainly nitrogen heterocycles are widely occupied the medicinal as well as in pharmaceutical field because they have given significant contribution to drug synthesis. They are naturally found in nucleic acid, vitamins, antibiotics, enzymes, hormones etc. Among Pyrazolo-pyrimidines have received special attention because, Pyrazolopyrimidines found to be the structural analogous of biogenic purine class compounds, having high impact in the field of pharmaceutical and biotechnological sciences with vast spectrum of biological activities. Enormous pharmacological activities like mitotic, CNS stimulant, analgesic, anti-inflammatory, antifungal, antibacterial and anticancer activity have been reviewed for Pyrazolopyrimidines. Pyrazolopyrimidines moieties have very frequently appeared in recent discoveries with new variety of medicinal and pharmaceutical applications like antibacterial, antifungal, antiviral, antitubercolosis, antimalarial, antidepressant, applications. In order to discover new leading compounds with high antibacterial and antifungal activity in new eco-friendly protocol suitable for Pyrazolopyrimidines derivatives. It was possible to synthesize new structures in simple step reactions ideology implementation in green method of organic synthesis are acceptable which are indirectly nullify the hazardous pollutants and intermediate of reaction.

2. Experimental Details: 2.1 ***Chemicals and apparatus*****:** Commonly selected chemicals are analytical grade used for preparation. MP are determined by Bucchi-450 melting point apparatus. Infrared spectra were reported using KBr pellets on BRUKER-FTIR spectrometry. The ^{1}H (proton) NMR notes (300 MHz) and ^{13}C (carbon) NMR notes (75 MHz) spectra recorded in a Varian XL300 spectrometry. chemical shifts (j) were record in ppm relatives to TMS (tetramethylsilane), and multiple peaks are named as s singlet (s), broad singlet (bs), doublet (d), triplet (t), quartet (q), and multiplet (m). Usually reactions progress was monitored with TLC [Thin layer chromatography] carried out on silica gel (Merck) plates (pre-coated), Spots are detect using (254 and 366 nm) in UV Chamber.

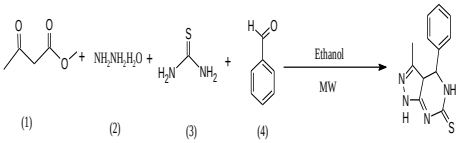
2.2 General Procedure for the synthesis of Pyrazolo-pyrimidine derivatives.

A solution of Ethyl aceto acetate/Ethyl cyanoacetate (20 M mol) was made to treat with hydrazine hydrate (20 M mol), Urea/thiourea and different benzaldehyde derivatives in Round bottom flask and Ethanol solvent (25-30 ml) was added to it. The reaction mixture was kept in Microwave oven and run it for the reaction. The progress of the reaction was monitored by TLC (Pet Ether: Ethyl acetate: 8:2). After completion of the reaction, the mixture (colour changed) contained the crude product, was filtered off. The catalyst was separated from the crude by washing with hot water for 3-4 times (approx. 50 ml). After the separation, it was recrystallized by ethanol to get pure product.

Scheme-1



Scheme-2



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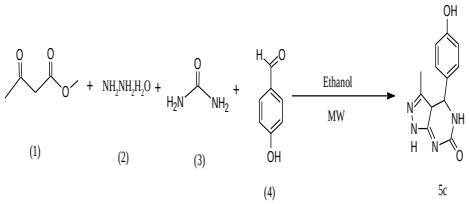

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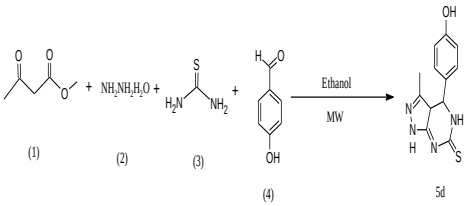
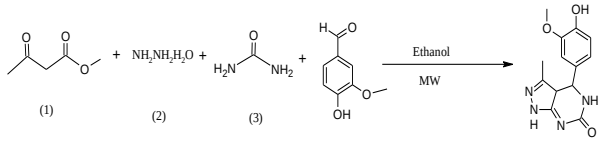
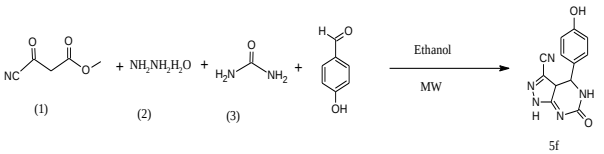


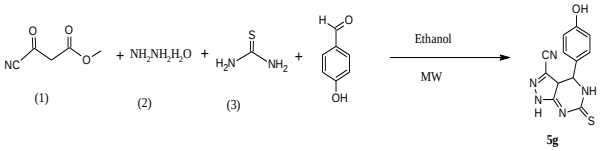
Scheme-5



Scheme-6



Scheme-7



Schematic Diagram: Aseries of Pyrazolo-pyrimidine derivatives 5a-g

3-methyl-4-phenyl-1,3a,4,5-tetrahydro-6*H* pyrazolo[3,4-*d*] pyrimidin-6-one: 5a: Yellow, M. P.-218–220°C, yield (81.00%). IR (KBr) νmax/ cm−1 3326 (NH), 1768 (C=O). ^{1}H-NMR **($CDCl_3$)** , 400 MHz, δppm): 3.80 (s, 3H, OCH3), 6.82 (d, 2H, J=9.0Hz, ArH), 7.00 (d, 1H, J=4.8Hz, pyrimidine), 7.12 (t, 1H, ArH), 7.33–7.46 (m, 5H, ArH), 7.62 (d, 2H, J=9.0 Hz, ArH), 7.54 (d, 2H, J=8.4 Hz, ArH), 8.11 (d, 2H, J=8.3 Hz, ArH), 8.48 (d, 1H, J=4.8 Hz, pyrimidine), 9.39 (s, 1H, NH), 10.05 (s, 1H). ^{13}C-NMR ($CDCl_3$) 100 MHz, δppm): 55.5 (C, OCH_3), 87.6(C,C_3Pyrazolopyrimidine),106.0(C,C_6Pyrazolopyrimidine), 114.7, 118.2, 120.8, 123.7, 126.7, 129.6, 129.8,129.9(14C,Ar),134.8(C,C3apyrazolopyrimidine) , 137.8, 142.8, 145.7 (3C,Ar), 147.7 (C,C7 –pyrazolopyrimidine), 163.2 (C=S).

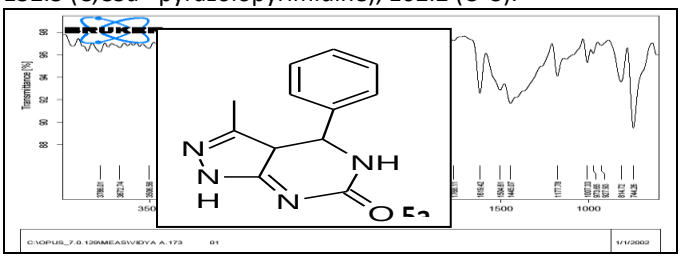
3-methyl-4-phenyl-1,3a,4,5-tetrahydro-6*H*pyrazolo [3,4-*d*] pyrimidine-6-thione: 5b: Yellow, m.p. 219–221°C, yield (74.00%). IR (KBr) νmax/ cm−1 3339 (NH), 1760 (C=S). ^{1}H-NMR **($CDCl_3$,)** 400 MHz, δppm): 2.45 (s, 3H, CH_3), 3.6 (s, 3H, OCH_3), 6.77 (d, 2H, J=8.9 Hz, ArH), 6.66 (d, 1H, J=4.7 Hz), 7.06 (t, 1H, ArH), 7.26 (d, 2H, J=8.3 Hz, ArH), 7.28 (t, 2H, ArH), 7.56 (d, 2H, J=8.9 Hz, ArH), 7.72 (d, 2H, J=7.6 Hz, ArH), 8.10 (d, 2H, J=8.1 Hz, ArH), 8.33 (d, 1H, J=4.7Hz), 9.33 (s, 1H, NH), 10.01 (s, 1H, NH).^{13}C-NMR ($CDCl_3$) 100 MHz, δ ppm): 21.5(C,CH_3),55.6(C,OCH3),86.9(C,C_3–pyrazolo pyrimidine), 107.1(C,C_6 pyrazolopyrimidine), 113.9, 119.1, 120.1, 123.5, 127.5, 129.0, 129.4, 129.6 (14C,Ar), 133.8 (C,C3a–pyrazolopyrimidine), 138.8, 142.2, 146.4 (3C, Ar), 163.1 (C=S).

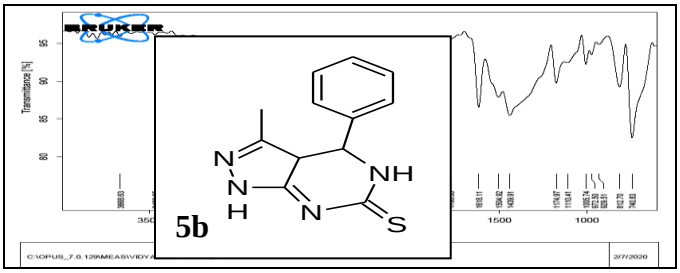
4-(4-hydroxyphenyl)-3-methyl-1,3a,4,5-tetrahydro-6*H*-pyrazolo[3,4-*d*] pyrimidin-6-one: 5c: Yellow, m.p. 206–208°C, yield (76.00%). IR (KBr) vmax/ cm−1 3451 (NH), 1606 (C=O). ^{1}H-NMR **($CDCl_3$)**, 400 MHz, δ ppm): 3.80 (s, 3H, OCH_3), 3.91 (s, 3H, OCH3), 6.87 (d, 2H, J=8.9 Hz, ArH), 6.89 (d, 1H, J=4.8 Hz), 7.05 (d, 2H, J=8.8 Hz, ArH), 7.11 (t, 1H, ArH), 7.37 (t, 2H, ArH), 7.60 (d, 2H, J=8.9 Hz, ArH), 7.72 (d, 2H, J=7.6 Hz, ArH), 8.18 (d, 2H, J=8.8 Hz, ArH), 8.40 (d, 1H, J=4.8 Hz), 9.36 (s, 1H, NH), 10.02 (s, 1H, NH). ^{13}C-NMR **($CDCl_3$)**, 100 MHz, δ ppm): 55.5 (C, OCH3), 55.6 (C, OCH3), 87.4 (C,C3 pyrazolopyrimidine),106.4(C,C6 pyrazolopyrimidine), 113.9, 114.2, 119.0, 120.0, 122.4, 123.5, 128.9, 131.3 (14C,Ar), 134.0 (C,C3a– pyrazolopyrimidine), 162.2 (C,Ar), 163.2 (C=O).

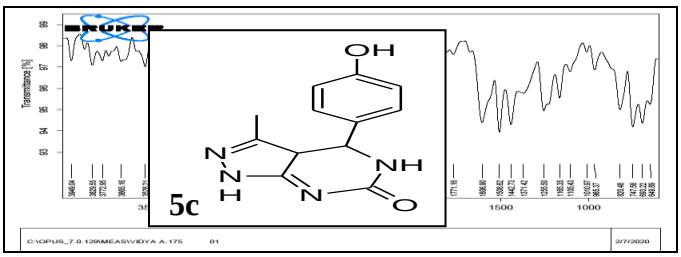
4-(2-hydroxyphenyl)-3-methyl-1,3a-dihydro-6H-pyrazolo[3,4-d] pyrimidine-6-thione: 5d: Yellow, m.p. 252–253°C, yield (88.00%); IR (KBr) vmax/ cm−1 3391 (NH), 1591 (C=S).^{1}H-NMR **($CDCl_3$)**, 400 MHz, δppm): 3.71 (s, 3H, OCH_3), 6.78 (d, 2H, J=9.0 Hz, ArH), 6.54 (d, 1H, J=4.7 Hz, pyrimidine), 7.33 (t, 1H, ArH), 7.35 (t, 2H, ArH), 7.57 (d, 4H, J=8.8 Hz, ArH), 7.77 (d, 2H, J=8.6 Hz, ArH), 8.45 (d, 2H, J=8.7 Hz, ArH), 8.52 (d, 1H, J=4.7 Hz, pyrimidine), 9.44 (s, 1H, NH), 9.99 (s, 1H, NH).^{13}C-NMR **($CDCl_3$)** 100 MHz, δppm): 55.6 (C,OCH3), 88.0 (C,C3 –pyrazolopyrimidine), 107.0 (C,C6 -pyrazolopyrimidine), 114.0, 119.2, 120.1, 123.7, 129.1, 129.1, 130.6, 131.8 (14C, Ar), 163.2 (C=S)

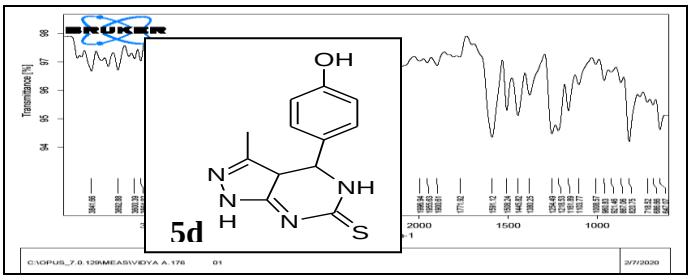
4-(4-hydroxy-3-methoxyphenyl)-3-methyl-1,3a,4,5-tetrahydro-6*H*-pyrazolo[3,4-*d*]pyrimidine6one 5e:Yellow m.p. 278–280°C, yield (84.00%); IR (KBr) vmax/ cm−1 3295 (NH), 1616 (C=S). 1 H-NMR (**DMSO-d6**, 400 MHz, δppm): 3.71 (s, 3H, OCH3), 6.79 (d, 2H, J=9.0 Hz, ArH), 7.22 (t, 1H, ArH), 7.36 (t, 2H, ArH), 7.45 (d, 1H, J=4.8 Hz), 7.56 (d, 2H, J=9.0 Hz, ArH), 7.71 (d, 2H, J=7.6Hz, ArH), 7.90 (d, 2H, J =8.7Hz, ArH), 8.21 (d, 2H, J=8.7Hz, ArH), 8.72 (d, 1H, J=4.8 Hz), 9.28 (s, 1H, NH), 10.04 (s, 1H, NH). 13C-NMR **(DMSO-d6**, 100 MHz, δppm): 55.7 (C,OCH3), 87.6 (C,C3–pyrazolopyrimidine),106.9(C,C6pyrazolopyrimidine), 114.4, 119.1, 120.5, 123.3, 129.4, 129.8, 131.0, 131.6 (14C,Ar), 133.7 (C,C3a–pyrazolopyrimidine), 163.7 (C=S).

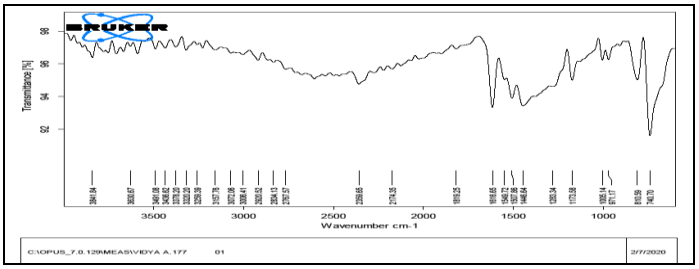
4-(4-hydroxyphenyl)-6-oxo-3a,4,5,6-tetrahydro-1*H*-pyrazolo[3,4-*d*]pyrimidine-3-carbonitrile:5f:Yellow crystals, m.p. 237–239°C, yield (67.00%); IR (KBr) vmax/ cm−1 3380 (NH), 1606 (C=S). 1 H-NMR **(DMSO-d6**, 400 MHz, δppm): 3.71 (s, 3H, OCH3), 6.95 (d, 2H, J=9.0 Hz, ArH), 7.16 (t, 1H, ArH), 7.38 (d, 1H, J=4.9 Hz), 7.35 (d, 2H, J=7.6 Hz, ArH), 7.52 (t, 2H, ArH), 7.54 (d, 2H, J=9.0 Hz, ArH), 7.79 (d, 2H, J=8.6 Hz, ArH), 8.35 (d, 2H, J=8.9 Hz, ArH), 8.79 (d, 1H, J=4.8 Hz), 9.33 (s, 1H, NH), 10.05 (s, 1H, NH). 13C-NMR (**DMSO-d_6**), 100 MHz,δppm):55.4(C,OCH3),86.8(C,C_3pyrazolopyrimidine), 108.5 (C,C6 -pyrazolopyrimidine), 114.5, 115.8, 115.9, 118.7, 119.5, 123.6, 126.5, 129.1 (14C, Ar), 132.3 (C,C3a– pyrazolopyrimidine), 162.2 (C=S).

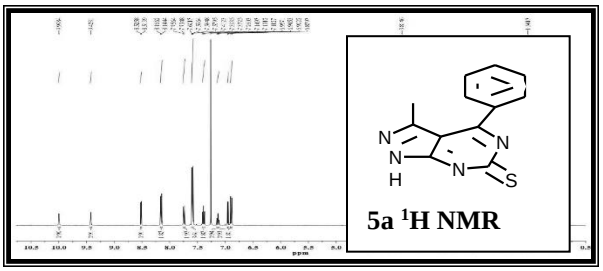


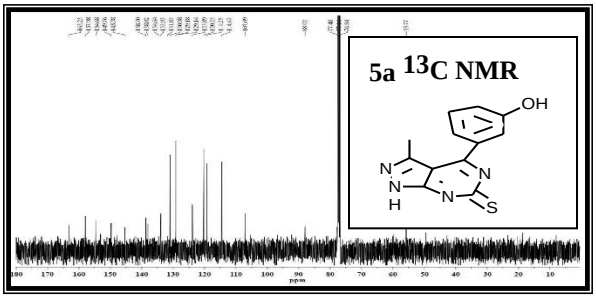


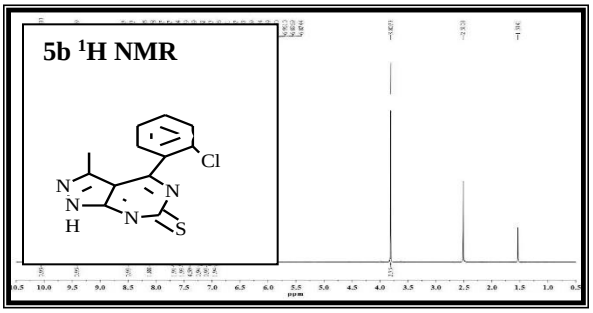


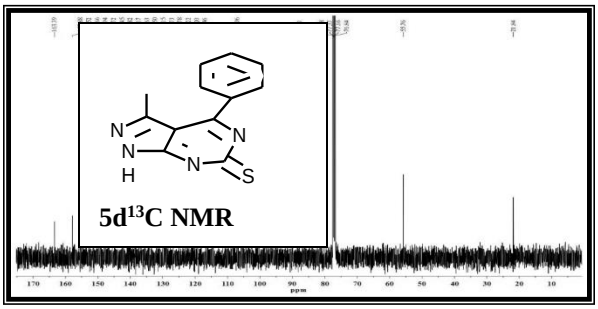












RESULTS AND DISCUSSION

The synthetic route used to synthesize the target Pyrazolo-pyrimidine derivatives (**5a-g**) is outlined in Scheme [1-7]. Ethyl acetoacetate/Ethyl cyanoacetate, hydrazine hydrate, Urea/thiourea and Aromatic aldehydes were treated via one-pot multi-component synthesis to get target compounds Pyrazolo-pyrimidine-ones[49-50]. The superior performance of the microwave reaction, in terms of yield and reaction time, could result from the higher temperature and pressure achieved. The substrate scope for the one-pot reaction was then explored, with variations of substituents at the pyrazolo[3,4-d] pyrimidinone core.

Spectral data (IR, ^{1}H NMR, and mass) of the newly synthesized compounds 5**a-g** were in full agreement with the proposed structures. In the ^{1}H NMR spectra of **4** and **5**, the C=CH proton displayed more downfield signal in the range δ 10.18 to 10.25. Besides this, C_5-H of the pyrazole ring resonates at around δ 7.51 to 7.63. The IR spectra of **5a** showed a characteristic absorption band around 1,674 to 1,682 cm^{-1} that was assigned to the C=O stretching, while the two absorptions bands around 1,304 to 1,335 and 1,149 to 1,165 cm^{-1} which further supported the proposed structures of newly synthesized compounds displayed the SO_2 stretching.

Table-1: Reaction of Pyrazolo-pyrimidine derivatives 5a-g by Microwave method

Samples	Microwave method	Convention method	Yield MW in (%)	M.P.
5a	3min	30 min	81.00	218-220 °C
5b	3min	45 min	78.00	21-221 °C
5c	3min	30 min	76.00	206-208 °C
5d	3min	30 min	87.00	252-253 °C
5e	3min	45 min	79.00	278-280 °C
5f	3min	40 min	82.00	237-239 °C
5g	3min	50 min	81.0	233-225 °C

In vitro antibacterial and antifungal activity.

Biological investigation optimized will describe in this work all compounds were evaluated in vitro antibacterial activity assessed against the four pathogenic (gram positive and negative) (St-aureus, Salmonella typhi, Streptococci and E-coli) bacterial strains by agar well diffusion method (Table 2). Gentamicin was used as reference drug. Most of the titled derivatives in the series (*5a-g*) showed moderate

to good antibacterial activity in that (5b) compound shows (zone of inhibition 18 mm) against st-aureus were found to be better than standard drug gentamycin (zone of inhibition 16 mm). While the other titled moieties (5a-g) were found to possess good antibacterial activity.

Comp.	*E-coli*	*Salmonella typhi*	*St. aureus*	*Streptococci*	Gentamicin
5a	15 ± 0.2	17 ± 0.3	19 ± 0.3	1 7± 0.3	16 ± 0.3
5b	14 ± 0.1	17 ± 0.1	20 ± 0.1	1 6± 0.1	16 ± 0.3
5c	18± 0.2	16 ± 0.9	19 ± 0.3	1 8± 0.2	16 ± 0.3
5d	19 ± 0.2	13 ± 0.3	20 ± 0.2	1 5± 0.0	16 ± 0.3
5e	18 ± 0.2	17 ± 0.2	18 ± 0.2	18 ± 0.2	16 ± 0.3
5f	14 ± 0.1	17 ± 0.1	20 ± 0.1	1 6± 0.1	16 ± 0.3
5g	14 ± 0.1	17 ± 0.1	20 ± 0.1	1 6± 0.1	16 ± 0.3

in vitro antifungal activity against fungal *Candida albicans* (MTCC227), S,*Cerevisiae* (MTCC170) by same method. Fluconazole used as the reference drug. Most of the titled compounds in series (**5a-g**) showed good antifungal activity. Compound 5**g** was found to be as effective standard drug, zone of inhibition16mmagainst C.*albicans*.Against *S.cerevisiae*, 5**a** (zone of inhibition 28 mm) were found to be the most effective and were better than the standard drug fluconazole (zone of inhibition 24 mm), while the other compound (5**d**) were found to possess good antifungal activity. Interestingly, the titled compound (5**a**) showed multifold reduction in the MIC values against *S. cerevisiae*, making the new derivatives attractive for further evaluation.

Samples	**Diameter of zone of inhibition in mm (MIC).**			
Strains	***Candida albicans***	**Fluconazole (Reference)**	***Saccharomyces Cerevisiae***	**Fluconazole (Reference)**
5a	12 ± 0.43 (400)	16 ± 0.45 (40)	28 ± 0.15 (0.04)	24 ± 0.50 (40)
5b	12 ± 0.56 (400)	16 ± 0.45 (40)	16 ± 0.38 (40)	24 ± 0.50 (40)
5c	12 ± 0.54 (400)	16 ± 0.45 (40)	16 ± 0.40 (40)	24 ± 0.50 (40)
5d	14 ± 0.14 (400)	16 ± 0.45 (40)	20 ± 0.45 (4.0)	24 ± 0.50 (40)
5e	12 ± 0.32 (400)	16 ± 0.45 (40)	18 ± 0.16 (40)	24 ± 0.50 (40)
5f	12 ± 0.32 (400)	16 ± 0.45 (40)	16 ± 0.27 (40)	24 ± 0.50 (40)
5g	16 ± 0.27 (40)	16 ± 0.45 (40)	18 ± 0.48 (40)	24 ± 0.50 (40)

In-vitro antifungal activity and MIC of compound **5a-g** using agar well diffusion method MIC(µg/ml) minimum inhibitory concentration. Hyphen denotes no bacterial growth. The concentration 4.0 mg/ml. the valves, including the diameter of the well (8 mm) i.e. the three replicates.

CONCLUSION:

All the compounds completed their reaction time at 3 minutes under microwave irradiation while in convention it took above 30 minutes. It concludes that microwave method is an efficient one for the green synthesis in organic chemistry. All the tested compounds showed moderate antifungal activity against *C. albicans*, while two compounds showed activity better than the reference drug against *S. cerevisiae*. In short, the reported compounds showed remarkable potential as antimicrobial agents and warranted further investigation of their mechanism of actions and binding site. The studies regarding these aspects are being planned with the help of a prospective collaborator.

REFERENCES

[1] Mohammad, A., Int J Bioorganic Chemistry, 230, 146-52, 2017.

[2] Amir, M., Ja Indian J. Chem, 46, 1014-1019, 2007.

[3] Trilok, C., Neha, G., Suman, L. and Saxena, S. S., Eur J. Med Chem, 45,1772- 1776, 2010.

[4] Asif, H. and AjmalActa Pharm.,59, 223-233, 2009.

[5] Mymoona, A., Asif, H. and Bismillah, A., Eur J. Med Chem, 2372-2378, 2009.

[6] Chao, J-S., Huia, P.X. and Shuo, L., J. Chinese Chem.Soc, 52, 539-544 539, 2005.

[7] Srinivas, K., Srinivas, U., Bhanuprakash, K. and Harakis., Eur J Med Chem, 41: 1240-1246, 2006.

[8] Mudasir, B. R., Rayees, M. H. and Abdul, R., J. Chem. Sci, 122(2), 177–182, 2010.

[9] Nanjunda, S., Swamy, S., Basppa, P. B., Prabhuswamy, Doreswamy., EurJMedChem, : 531-538.3, 2006.

[10] Hui, X. P., Chu, C. H., Zangzu, Wang Q, and Zhang, Q., Indian J. Chem, 41B: 2176, 2002.

[11] Chen, Q., Zhu, X. L., Jiang, L.L., Liu, Z. M. and Yang, G. F., Eur J Med Chem,43(3):595-603, 2008.

[12] Singh, S., Sharma, P. K., Dudhe, R., An International Journal of Pharmaceutical Sciences, 2(4) 131-141, 2011.

[13] Pacher, P., Nivorozhkin, A. and Szabó, C., Pharmacological Reviews, 58 (1), 87– 114, 2006.

[14] WHO Model Formulary 08, The American Society of Health-System Pharmacists. Retrieved, 2016.

[16] Elie, R., Rüther, E., Farr, I., Emilien, G and Salinas, E., J Clin Psychiatry. 60 (8), 536–44, 1999.

[17] Mirza, N. R., Rodgers, Journal of Pharmacology and Experimental Therapeutics., 316 (3):1291–9, 2006.

[18] Feely, M., Boyland, P., Picar Gent, J. P., European Journal of Pharmacology. 164 (2), 1989.

[19] Lucy, H., Baleeiro,T., Fabiana, D., Savion, G., Benjamin, S., Antonio, G. and Thomas, R., Human Psychopharmacology: Clinical and Experimental, 29, (3), 266–273, 2014.

[20] Holla, B. S., Mahalinga, M., Karthikeyan, M. S, Akberali and N. S, Bioorg. Med. Chem., 14, 2040-2047, 2006.

[21] Bakavoli, M., Bagherzadeh, G., Vaseghifa,r M., Shiri, A., Pordel, M., Ma Pordeli, P. and Araghi, A., Bioorg. Med. Chem., 45, 647-650, 2010.

[22] Karthikeyan, Bioorg. Med, 2006. [23] Cheng, C. C., and Robins, R. K. L., Eur. J. Org. Chem.,21, 1240-56, 2007.

[26] Ameur R.; Anis R.; Abderrahim B.S.; J.; Turkish Journal of Chemistry. 38, 210-221, 2014.

[29]. A.S., Grewal, K. Kumar, S. Redhu, S. Bhardwaj, Int. Res. J. Pharm. App. Sci., 2013, 3, 278-285.

[30]. P. Lidstrom, J. Tierney, B. Wathey, J. Westmana, Tetrahedron, 2001, 37, 9225- 9283.

[31]. M. Lancaster. Principles of sustainable and green chemistry. Handbook of green chemistry,2002p10-27

[32]. M. Kidwai, R. Venkataraman, B. Dave, Green Chem., 2001, 3, 278-279.

[33]. R.S. Varma, R.K. Saini, R. Dahiya, Tetrahedron Lett., 1997, 38, 7823-7824.

[34]. A. Loupy, Microwaves in Organic Synthesis, Wiley-VCH: Weinheim, 2002, p 68.

[35]. Hassan, A.S.; Hafez, T.S.; Osman, S.A.; Ali, M.M. Synthesis and in vitro cytotoxic activity of novel pyrazolo [1, 5-a] pyrimidine and related Schiff bases. Turk. J. Chem. 2015, 39, 1102-1113.

[36]. Hassan, A.S.; Mady, M.F.; Awad, H.M.; Hafez, T.S. Synthesis and antitumor activity of some new pyrazolo [1, 5-a] pyrimidine. Chin. Chem. Lett. 2017, 28, 388-393.

[37]. Hassan, A.S.; Masoud, D.M.; Sroor, F.M.; Askar, A.A. Synthesis and biological evaluation of pyrazolo [1, 5-a] pyrimidine-3-carboxamide as antimicrobial agents. Med. Chem. Res. 2017, 26, 2909-2919.

[38]. Hassan, A.S.; Moustafa, H.M. Synthesis and in vitro anticancer activity of pyrazolo[1,5-a] pyrimidinesandpyrazolo[3,4d][1,2,3]triazines.Synth. Commun. 2017, 47, 1963-1972.

[39.] Hassan, A.S.; Awad, H.M.; Magd-El-Din, A.A.; Hafez, T.S. Synthesis and in vitro antitumor evaluation of novel Schiff bases. Med. Chem. Res. 2018, 27, 915-927.

[40] El-Naggar, M.; Hassan, A.S.; Awad, H.M.; Mady, Design, synthesis and antitumor evaluation of novel pyrazolo-pyrimidines and pyrazoloquinazolines. Molecules 2018, 23, 1249. DOI: 10.3390/molecules23061249.

[41] Kordik, C.P.; Luo, C.; Zanoni, B.C.; Lovenberg, T.W.; Wilson, S.J.; Vaidya, A.H.; Crooke, J.J.; Rosenthal, D.I.; Reitz, A.B. Aminopyrazoles with high affinity for the human neuropeptide Y5 receptor. Bioorg. Med. Chem. Lett. 2001, 11, 2283-2286.