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SYNTHESIS AND ANTI-MICROBIAL, ANTI-OXIDANT AND ANTI-INFLAMMATORY ACTIVITIES OF THIAZOLE-PYRAZOLE BASED PYRIMIDINE DERIVATIVES

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ABSTRACT

A series of thiazole-pyrazole based pyrimidine derivatives **(6a-j)** were synthesized by reacting thiazole chalcones with guanidine hydrochloride. The chemical structures of synthesized compounds were confirmed by IR, ^{1}H NMR, ^{13}C NMR and HRMS. All the title compounds were screened for their anti-microbial (anti-bacterial and anti-fungal), *in vitro* anti-oxidant (DPPH, H_2O_2, NO and SOR) and anti-inflammatory (protein denaturation) activity. The screening data indicated that tested compounds showed significant anti-microbial, anti-oxidant and anti-inflammatory activity.

Keywords: Thiazole, Pyrazole, Pyrimidines, Anti-microbial, Anti-oxidant and Anti-inflammatory.

INTRODUCTION

Progress of organic chemistry especially in the medicinal chemistry branch forever contributes to the growth in multiple fields of science. A common feature of the majority of newly reported organic molecules is the presence of at least one heterocyclic ring. Thus, heterocyclic chemistry is an important tool for search of novel pharmaceutically active molecules with several potent applications. One of the most interesting six-membered heterocyclic ring with heteroatom nitrogen is a pyrimidine. Pyrimidine ring has gained much attention in the field of drug chemistry research to design and discovery of novel physiologically and pharmacologically active compounds. Pyrimidine ring forms a component in several useful drug molecules and are linked with many biological, pharmaceutical and therapeutic activities.[1-2] Annulated pyrimidine derivatives have received great attention in the last decades because they show a wide range of pharmaceutical activities such as *in vitro* activity against unrelated DNA and RNA, viruses including polio herpes, diuretic, antitumor, anti-HIV, and cardiovascular[3].

The literature survey indicated that a wide range of pharmacological activities is exhibited by the compounds encompassing the pyrimidine nucleus. In addition to this, various equivalents of pyrimidine have been shows anti-bacterial[4], anti-fungal[5], anti-inflammatory[6], analgesic[7], anti-viral[8], anti-diabetic[9], anti-convulsant[10], anti-oxidant[11], anti-histaminic[12], herbicidal[13] and anti-cancer activities[14] and many of pyrimidines derivatives are reported to possess potential central nervous system (CNS) depressant properties.[15] It also found that the presence of thiazole and pyrazole nucleus showed potent anti-oxidant and anti-inflammatory activity.[16-21]

Owing to the importance and in continuation of our work on the synthesis of biologically important molecules here we designed and synthesized various thiazole-pyrazole based pyrimidine derivatives (Scheme-1) and evaluated for their anti-microbial, anti-oxidant and anti-inflammatory activity.

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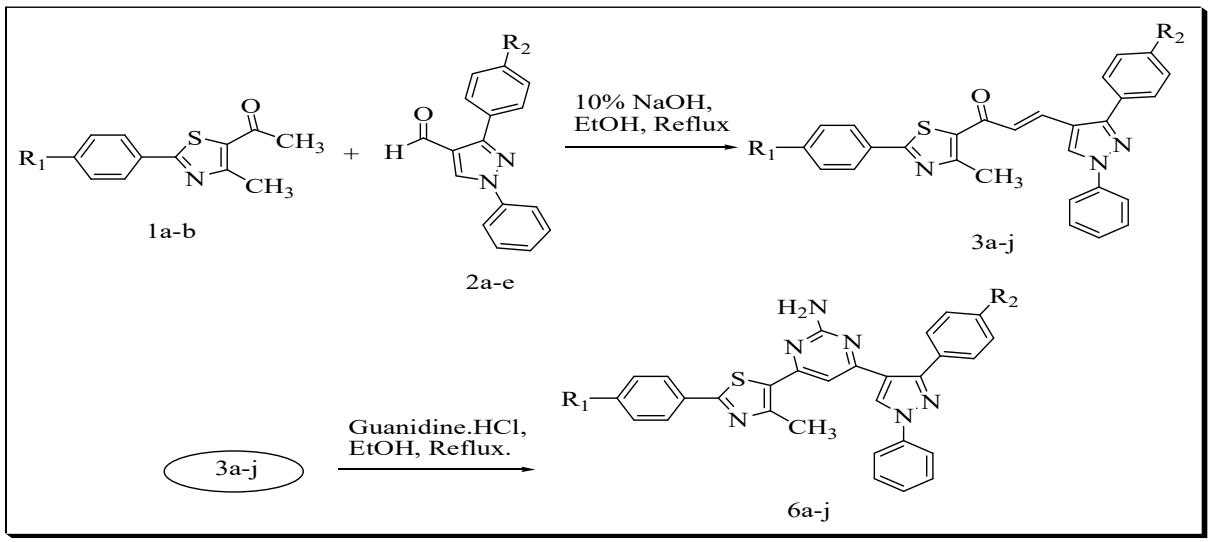
EXPERIMENTAL

Material and Methods

All commercially available chemicals and reagents were purchased from Aldrich, Spectrochem and used without further purification. All the solvents were dried and distilled before use. The melting points of all synthesized pyrimidine derivatives were recorded in an open capillary tube and are uncorrected. The IR spectra of synthesized compounds were recorded on Shimadzu 8400-S FT-IR Spectrophotometer using potassium bromide. The ^{1}H NMR and ^{13}C NMR were recorded in $CDCl_3$ using Bruker 400 MHz NMR spectrometer and chemical shifts are reported as parts per million (ppm) using tetramethyl silane (TMS) as an internal standard. HRMS was recorded on Mass Spectrophotometer. Reactions were monitored using thin-layer chromatography (TLC) carried out on pre-coated aluminium plates. The visualization was achieved under UV light or staining with I_2. Chromatographic separations were achieved on silica gel columns (Merck, 60–120 mesh) using a gradient of hexane/ethyl acetate as eluent.

Synthesis of Thiazole-Pyrazole Based Pyrimidine Derivatives

A mixture of (E)-1-(4-methyl-2-phenylthiazol-5-yl)-3-(1,3-diphenyl-1H-pyrazole-4-yl)prop-2-en-1-one and its derivatives (1mmol) **3a-j** and guanidine hydrochloride (2 mmol) was dissolved under stirring in 15 mL of hot absolute ethanol. A solution of 10% NaOH was added dropwise until pH became basic and the reaction mixture was refluxed for the appropriate time. The progress of the reaction was checked by TLC. After completion of the reaction, the reaction mixture was poured on to crushed ice and stirred for 15-20 minutes and neutralized by using dilute acetic acid, obtained solid was filtered, washed with cold water and dried. The crude product was recrystallized by using ethanol to afford pure titled compound **6a-j**.



Scheme-1: Synthesis of Thiazole-Pyrazole Based Pyrimidine Derivatives

Where,

6a	R_1= H, R_2= H	**6b**	R_1= H, R_2= F
6c	R_1= H, R_2= CH_3	**6d**	R_1= H, R_2= NO_2
6e	R_1= H, R_2= OCH_3	**6f**	R_1= Cl R_2= H
6g	R_1= Cl, R_2= F	**6h**	R_1= Cl, R_2= CH_3
6i	R_1= Cl R_2= NO_2	**6j**	R_1= Cl, R_2= OCH_3

Spectral Data of Representative Compound

4-(3-(4-fluorophenyl)-1-phenyl-1H-pyrazol-4-yl)-6-(4-methyl-2-phenylthiazol-5-yl)pyrimidin-2-amine (6b)

Yield: 73.37%; MW: 504.58; M.P.: 220°C; IR (KBr, cm^{-1}): 3299, 3229, 3093, 2923, 1651, 1215, 755; ^{1}H NMR (400 MHz, $CDCl_3$, δ in ppm): 2.83 (s, 3H, Thy-CH_3), 5.07 (s, 2H, -NH_2), 6.78 (s, 1H, Pyrd-H), 7.06-7.10 (d, 1H, Ar-H), 7.19-7.23 (t, 2H, Ar-H), 7.38-7.39 (t, 1H, Ar-H), 7.47 (t, 1H, Ar-H), 7.52-7.54 (m, 4H,

Ar-H), 7.68-7.71 (m, 2H, Ar-H), 7.79-7.87 (m, 2H, Ar-H), 7.99-8.01 (m, 2H, Ar-H), 8.34 (s, 1H, Pyr-H);^{13}C NMR (400 MHz, $CDCl_3$, δ in ppm): 18.86 (m, Thy-CH_3), 105.81 (m, Pyri-CH), 107.03 (m, C), 117.81 (m, C), 119.44 (s, CH), 124.11 (m, CH), 126.52 (s, CH), 126.91 (m, CH), 127.47 (s, CH), 128.35 (s, CH), 129.63 (m, CH), 130.61 (s, CH), 130.69 (s, CH), 131.09 (m, C), 131.20 (m, C), 132.84 (w, C), 134.64 (m, C), 139.27 (w. C), 153.05 (w, C), 160.47 (w, C), 161.98 (w, C), 164.44 (w, C); HRMS: m/z = 505.16 (M+1).

4-(4-methyl-2-phenylthiazol-5-yl)-6-(1-phenyl-3-p-tolyl-1H-pyrazol-4-yl)pyrimidin-2-amine (6c)
Yield: 80.0%; MW: 500.62; M.P.: 190ºC-192ºC; IR (KBr, cm^{-1}): 3350, 3290, 3050, 2920, 2850, 1647, 755;^{1}H NMR (400 MHz, $CDCl_3$, δ in ppm): 2.51 (s, 3H, Thy-CH_3), 2.59 (s, 3H, Ar-CH_3), 5.06 (s, 2H, -NH_2), 6.81 (s, 1H, Pyrd-H), 7.06-7.10 (d, 1H, Ar-H), 7.25-7.61 (m, 7 H, Ar-H), 7.70-7.80 (m, 2H, Ar-H), 7.82-8.01 (m, 4H, Ar-H), 8.33 (s, 1H, Pyr-H);HRMS: m/z = 501.18 (M+1).

4-(3-(4-methoxyphenyl)-1-phenyl-1H-pyrazol-4-yl)-6-(4-methyl-2-phenylthiazol-5-yl)pyrimidin-2-amine (6e)
Yield: 87.20 %; MW: 516.62; M.P.: 198ºC; IR (KBr, cm^{-1}): 3327, 3277, 2917, 2887, 1669, 1244, 757;^{1}H NMR (400 MHz, $CDCl_3$, δ in ppm): 2.77 (s, 3H, Thy-CH_3), 3.88 (s, 3H, Ar-CH_3), 5.07 (s, 2H, -NH_2), 6.84 (s, 1H, Pyrd-H), 6.98-7.03 (m, 1H, Ar-H), 7.24-7.39 (m, 2H, Ar-H), 7.42-7.44 (t, 2H, Ar-H), 7.45-7.47 (m, 2H, Ar-H), 7.49-7.51 (d, 2H, Ar-H), 7.51-7.59 (m, 1H, Ar-H), 7.67-7.85 (m, 2H, Ar-H), 7.93-7.97 (m, 2H, Ar-H), 8.60 (s, 1H, Pyr-H);HRMS: m/z = 517.18 (M+1).

4-(2-(4-chlorophenyl)-4-methylthiazol-5-yl)-6-(1,3-diphenyl-1H-pyrazol-4-yl)pyrimidin-2-amine (6f)
Yield: 92.30 %; MW: 521.04; M.P.: 158ºC-160ºC; IR (KBr, cm^{-1}): 3279, 3234, 3067, 2907, 1649, 755, 686;^{1}H NMR (400 MHz, $CDCl_3$, δ in ppm): 2.80 (s, 3H, Thy-CH_3), 5.07 (s, 2H, -NH_2), 6.79 (s, 1H, Pyrd-H), 7.06-7.10 (d, 1H, Ar-H), 7.29-7.54 (m, 3H, Ar-H), 7.64-7.66 (m, 1H, Ar-H), 7.70-7.72 (m, 3H, Ar-H), 7.81-7.89 (d, 1H, Ar-H), 7.92-7.95 (m, 2H, Ar-H), 8.35 (s, 1H, Pyr-H);HRMS: m/z = 521.13 (M+).

4-(2-(4-chlorophenyl)-4-methylthiazol-5-yl)-6-(3-(4-fluorophenyl)-1-phenyl-1H-pyrazol-4-yl) pyrimidin-2-amine (6g)
Yield: 85.66 %;MW: 539.03;M.P.: 178ºC;IR (KBr, cm^{-1}): 3309, 3249, 3066,2923, 1651, 1213,755, 687;^{1}H NMR (400 MHz, $CDCl_3$, δ in ppm): 2.82 (s, 3H, Thy-CH_3), 5.09 (s, 2H, -NH_2), 6.80 (s, 1H, Pyrd-H), 7.05-7.08 (d, 1H, Ar-H), 7.19-7.23 (t, 2H, Ar-H), 7.38-7.44 (t, 1H, Ar-H), 7.45-7.46 (d, 2H, Ar-H), 7.50-7.54 (t, 2H, Ar-H), 7.67-7.71 (m, 2H, Ar-H), 7.79-7.81 (t, 1H, Ar-H), 7.84-7.93 (m, 1H, Ar-H), 7.95-7.95 (d, 1H, Ar-H), 8.34 (s, 1H, Pyr-H);^{13}C NMR (400 MHz, $CDCl_3$, δ in ppm): 18.61 (m, Thy-CH_3), 105.82 (m, Pyri-CH), 106.04 (M, C), 117.77 (w, C), 119.44 (s, -CH), 123.91 (m, -CH), 127.08 (m, -CH), 127.51 (m, -CH), 128.07 (s, -CH), 129.41 (s, -CH), 129.65 (s, -CH), 130.62 (m, C), 130.70 (m, C), 131.33 (w, C), 131.42 (w, C), 134.85 (w, C), 137.27 (w, C), 139.25 (w, C), 153.08 (w, C), 160.43 (w, C), 161.98 (w, C);HRMS: m/z = 539.12 (M+).

4-(2-(4-chlorophenyl)-4-methylthiazol-5-yl)-6-(1-phenyl-3-p-tolyl-1H-pyrazol-4-yl)pyrimidin-2-amine (6h)
Yield: 86.89 %; MW: 535.06; M.P.: 160ºC; IR (KBr, cm^{-1}): 3297, 3227, 3062, 2917, 2852, 1647, 755, 687;^{1}H NMR (400 MHz, $CDCl_3$, δ in ppm): 2.17 (s, 3H, Thy-CH_3), 2.58 (s, 3H, Ar-CH_3), 5.06 (s, 2H, -NH_2), 6.80 (s, 1H, Pyrd-H), 7.05-7.09 (d, 1H, Ar-H), 7.19-7.54 (m, 5H, Ar-H), 7.58-7.67 (m, 4H, Ar-H), 7.80-7.94 (m, 3H, Ar-H), 8.33 (s, 1H, Pyr-H);HRMS: m/z = 535.14 (M+).

4-(2-(4-chlorophenyl)-4-methylthiazol-5-yl)-6-(3-(4-methoxyphenyl)-1-phenyl-1H-pyrazol-4-yl) pyrimidin-2-amine (6j)
Yield: 94.11 %; MW: 551.06; M.P.: 196ºC; IR (KBr, cm^{-1}): 3375, 3252, 2918, 2834, 1612, 1245, 755, 689;^{1}H NMR (400 MHz, $CDCl_3$, δ in ppm): 2.78 (s, 3H, Thy-CH_3), 3.85 (s, 3H, Ar-OCH_3), 5.06 (s, 2H, -NH_2), 6.83 (s, 1H, Pyrd-H), 6.99-7.08 (m, 2H, Ar-H), 7.34-7.47 (m, 3H, Ar-H), 7.49-7.59 (m, 2H, Ar-H), 7.63-7.73 (m, 2H, Ar-H), 7.79-7.88 (m, 2H, Ar-H), 7.90-7.95 (m, 2H, Ar-H), 8.32 (s, 1H, Pyr-H);HRMS: m/z = 551.14 (M+).

Biological Activity

Anti-inflammatory Activity

***In vitro* Anti-inflammatory Activity by Protein Denaturation Method**

The reaction mixture (2.5 mL) consisted of 0.1 mL of egg albumin (from fresh hen's egg), 1.4 mL of phosphate-buffered saline (PBS, pH 6.4) and 1 mL of synthetic derivatives (1 mM). A similar volume of Phosphate buffer saline served as control. Then the mixtures were incubated at (37°C ±2) in an incubator for 15 min and then heated at 70°C for 5 min. After cooling, their absorbance was measured at 660 nm by using the vehicle as blank. Diclofenac sodium (1 mM) was used as a reference drug and treated similarly for the determination of absorbance[22]. The protein denaturation % inhibition was calculated by using the following formula. The percentage inhibition of protein denaturation was calculated by using the following formula and results recorded in Table-1.

$$\% \text{ of Inhibition} = 100 \times (Vt / Vc - 1)$$

Where, Vt = Absorbance of the test sample, Vc = absorbance of control.

Anti-oxidant Activity

DPPH Radical Scavenging Activity

The molecule 1, 1-diphenyl-2-picrylhydrazyl (DPPH) is characterized as a stable free radical by the delocalization of the spare electron over the molecule as a whole, so that the DPPH molecule does not dimerize, as would be the case with most other free radicals. The delocalization of electron also gives progress to the deep violet color, characterized by an absorption band in ethanolic solution at about 517 nm. When a solution of DPPH is mixed with that of a substrate (AH) that can donate a hydrogen atom, then this gives rise to the reduced form with the loss of this violet color. To evaluate the anti-oxidant potential through free radical scavenging by the test samples, the change in optical density of DPPH radicals is monitored[23]. Results are recorded in Table-1.

Hydrogen Peroxide (H_2O_2) Radical Scavenging Activity

Human beings are exposed to H_2O_2 indirectly via the environment nearly about 0.28 mg/kg/day with intake mostly from leaf crops. Hydrogen peroxide may enter the human body through inhalation of vapor or mist and through eye or skin contact. H_2O_2 is quickly decomposed into oxygen and water molecule and this may produce hydroxyl radicals (OH) that can initiate lipid peroxidation and cause DNA damage in the body[24]. Results are recorded in Table-1.

NO Radical Scavenging Activity

NO is formed in biological tissues by specific NO synthases, which metabolizes arginine to citrulline with the formation of NO via a five-electron oxidative reaction. The sodium nitroprusside is known to decompose in aqueous solution at physiological pH (7.2) producing NO. Under aerobic conditions, NO reacts with oxygen to produce nitrate and nitrite (stable products), which can be quantitatively determined using Griess reagent[25]. Results are recorded in Table-1.

SOR Scavenging Activity

SOR radical scavenging activity was carried out as per the reported method. The mixture consisting of 1mL of nitro blue tetrazolium (NBT) solution (156 mM NBT in phosphate buffer, pH 7.4), 1 mL NADH solution (468 mM NADH in phosphate buffer, pH 7.4), and 1 mL of synthetic compound (1 mM) solution was mixed. The reaction was started by adding 1 mL of phenazinemethosulfate (PMS) solution (60 mM PMS in phosphate buffer, pH 7.4) to the above mixture. The reaction mixture was incubated for 5 minutes at 25°C and the absorbance was measured at 560 nm against the blank sample and compared with standards and percentage inhibition was calculated using the same formula as above. Decreased absorbance indicates increased SOR scavenging activity[26]. Results are recorded in Table-1.

Anti-microbial Activity

Anti-bacterial Activity and Anti-fungal Activity (Agar Well Diffusion Method)

The solution of different synthesized compounds under test at a concentration of 1 mg/ml of DMSO solvent was poured in the well of bacteria seeded agar plates. These plates were incubated at 370°C for 24 hours.

The activity was reported by measuring the diameter of the zone of inhibition in mm concerning standard anti-bacterial agent Tetracycline and anti-fungal agent Nystatin. The solution without compound i.e. only DMSO was used as control[27]. Results are recorded in Table-2.

Table-1: Anti-inflammatory and antioxidant activity of synthesized compounds (6a-j)

S. No.	Compound No./ Code	Anti-inflammatory Activity % inhibition (1mM)	Anti-oxidant Activity % Inhibition (1mM)			
			DPPH	H_2O_2	NO	SOR
1	6a	65	30	43	25	37
2	6b	92	45	59	10	55
3	6c	62	25	50	23	46
4	6d	47	22	46	42	40
5	6e	87	41	55	37	49
6	6f	51	25	40	33	36
7	6g	83	35	28	18	25
8	6h	56	28	25	27	21
9	6i	40	16	33	45	28
10	6j	78	37	40	38	12
11	Diclofenac Sodium	90.21	--	--	--	--
12	Ascorbic Acid	--	42.98	--	42.63	89.13
13	BHT	--	--	88.42	--	--

Table-2: Anti-microbial Activity of the Synthesized Compound (6a-j)

S. No.	Compound	Anti-bacterial Activity Zone of Inhibition (mm)								Anti-fungal Activity Zone of Inhibition (mm)		
		Gram-positive Pathogens				Gram-negative Pathogens						
--	--	Micrococcus luteus	Bacillus megaterium	Staphylococcus aureus	Bacillus cereus	Salmonella typhi	Escherichia coli	Pseudomonas aerogenosa	Salmonella abony	Candida albicans	Saccharomyces cerevisiae	Aspergillusniger
1	6a	11	11	19	21	08	14	12	17	18	11	07
2	6b	10	16	14	12	06	10	10	12	13	11	09
3	6c	09	12	08	09	05	12	11	16	06	18	08
4	6d	08	15	11	09	10	16	14	08	10	18	11
5	6e	08	12	09	11	05	15	13	11	12	13	10
6	6f	09	12	16	12	16	10	11	10	17	12	08
7	6g	10	16	11	10	08	07	10	12	14	18	09
8	6h	08	17	12	08	17	09	11	18	19	12	12
9	6i	07	16	06	05	09	12	10	12	14	11	10
10	6j	11	17	09	05	08	15	10	12	18	10	19
DMSO	--	--	--	--	--	--	--	--	--	--	--	--
Tetracyclin	--	25	20	30	25	22	20	33	21	--	--	--
Nystatin	--	--	--	--	--	--	--	--	--	24	20	25

RESULTS AND DISCUSSION

The synthesis of desired thiazole-pyrazole based pyrimidine derivatives **(6a-j)** was achieved by reacting thiazole-pyrazole chalcones with guanidine hydrochloride in ethanol (Scheme-1). The synthesis of thiazole-

pyrazole chalcones **(3a-j)** was carried out by the Claisen-Schmidt condensation reaction of substituted 1-(4-methyl-2-phenylthiazol-5-yl) ethanone and substituted pyrazole aldehyde in presence of 10% NaOH in ethyl alcohol as solvent under reflux condition. The chemical structures of all the synthesized compounds were confirmed by IR, ^{1}H NMR, ^{13}C NMR and HRMS.

Biological Activity

Anti-inflammatory Activity

All the newly synthesized thiazole-pyrazole based pyrimidine derivatives **(6a-j)** were evaluated for their *in vitro* anti-inflammatory properties against protein denaturation of egg albumin and results are illustrated in Table-1. It was found that most of the compounds were found to possess good to moderate anti-inflammatory properties. Among the compounds screened, compounds **6b, 6e, 6g** and **6j** demonstrated significant inhibition of protein denaturation compared to the Diclofenac sodium, a standard anti-inflammatory drug at 1 mM concentration whereas, compounds **6a, 6c** and **6h** displayed moderate inhibition of protein denaturation. On the other hand, all other compounds exhibited weak inhibition of heat-induced egg albumin denaturation (Fig.-1). Structure-activity relationship (SAR) study reveals that, electron-donating groups like methoxy or fluro group on phenyl ring increases anti-inflammatory activity while donating group like nitro group decreases the activity.

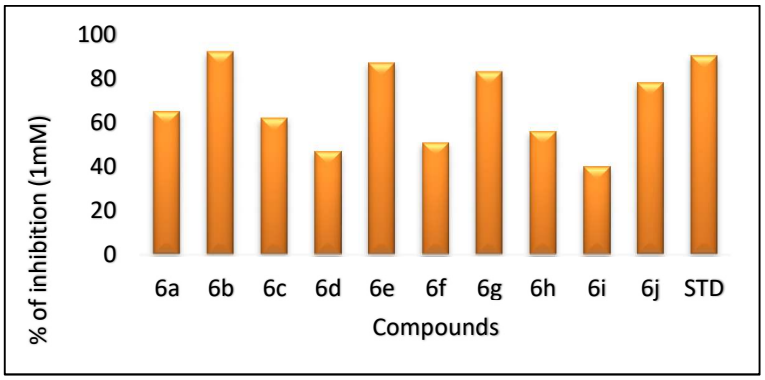


Fig.-1: Comparison of Anti-inflammatory Activity of Compounds 6a-j with Standard Drug Diclofenac Sodium

Anti-oxidant Activity

All the synthesized thiazole-pyrazole based pyrimidine derivatives **(6a-j)** were further subjected for evaluation of anti-oxidant properties against ROS such as DPPH, H_2O_2, NO and SOR radicals using ascorbic acid and BHT as a reference standard and the results are enumerated in Table-1. All the compounds demonstrated excellent to moderate scavenging activity against DPPH and NO radical and moderate to weak scavenging activity against H_2O_2 and SOR. The anti-oxidant activity results revealed that the compounds **6b, 6e, 6j** and **6g** exhibited significant inhibition of DPPH radicals. On the other hand, all other remaining compounds were found to possess moderate DPPH radical scavenging activity compared to the ascorbic acid, a reference drug. Compounds **6i, 6d, 6j** and **6e** exhibited significant inhibition of NO radicals compared to the standard drug ascorbic acid. However, remaining compounds were moderate NO radical scavengers (Fig.-2). Structure-activity relationship (SAR) study reveals that, presence of electron-donating groups like methoxy or fluro group on phenyl ring exhibited significant inhibition of DPPH, H_2O_2, NO and SOR radicals, while donating group like nitro group exhibited weak inhibition.

Anti-microbial Activity

Newly synthesized thiazole-pyrazole based pyrimidine derivatives (**6a-j)** were screened for their anti-bacterial and anti-fungal activity by agar well diffusion method at inhibitory concentration 1mg/ml compared with standard drug anti-bacterial agent tetracycline and anti-fungal agent nystatin. The diameter in millimeter of zone of inhibition around each disk was measured by scale and the observed data of anti-bacterial and anti-fungal activities of all screened compounds and the standard reference drugs are displayed in Table-2. Anti-bacterial activity results reveal that the compound **6a** showed significant activity,

compounds **6b, 6d, 6g, 6h, 6i** and **6j** showed moderate activity and compounds **6c, 6e** and **6f** showed weak activity comparable with standard drug Tetracyclin against tested Gram-positive pathogens (*Micrococcus luteus, Bacillus megaterium, Staphylococcus aureus and Bacillus cereus*) and graphically represented in Fig.-3. Compounds **6a** and **6h** showed significant activity, compounds **6c, 6d, 6e, 6f** and **6i** displayed moderate activity and compounds **6b, 6g** and **6i** showed weak activity comparable with standard drug Tetracyclin against tested Gram-negative pathogens *(Salmonella typhi, Escherichia coli, Pseudomonas aerogenosa and Salmonella abony)*and graphically represented in Fig.-4.

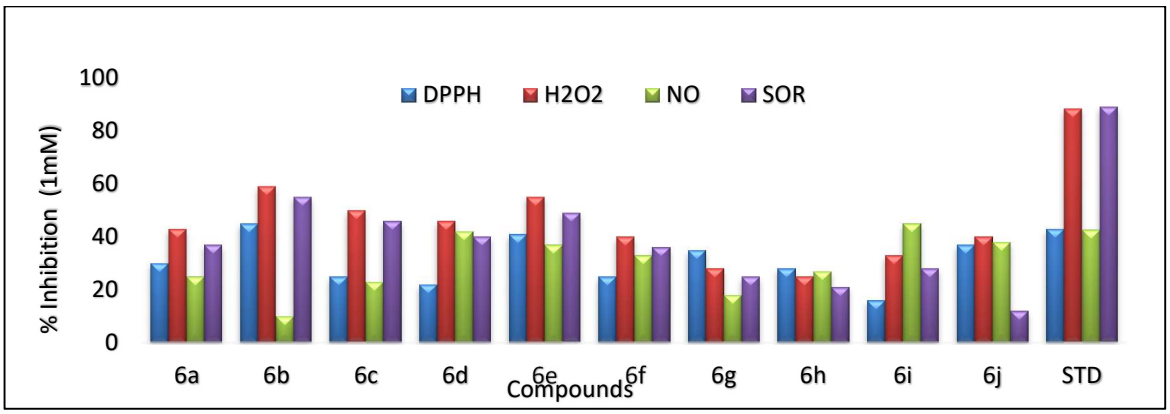


Fig.-2: Comparison of DPPH, NO and SOR Radical Scavenging Activities of Compounds 6a-j with Ascorbic Acid and H_2O_2 with BHT

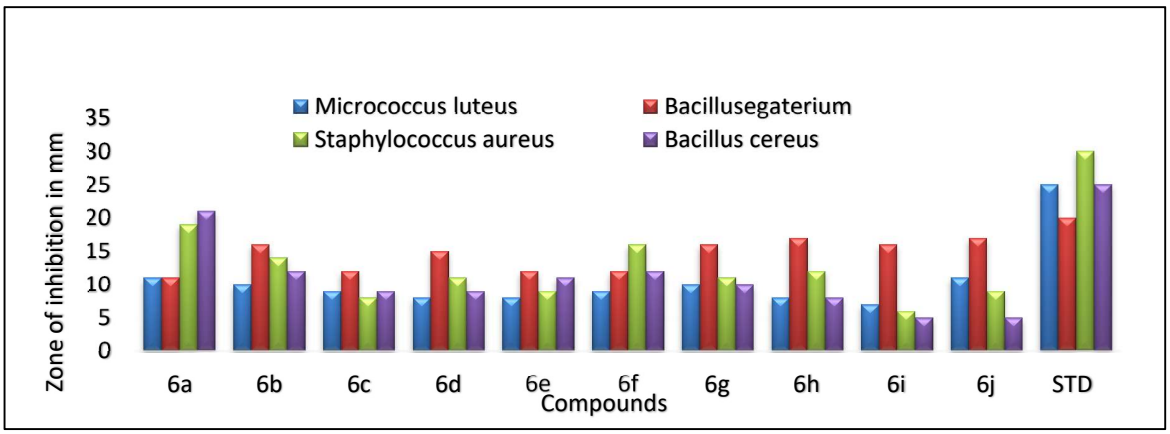


Fig.-3: Comparison of Anti-bacterial Activity of Compounds 6a-j against Gram-positive Pathogen with Standard Drug Tetracyclin

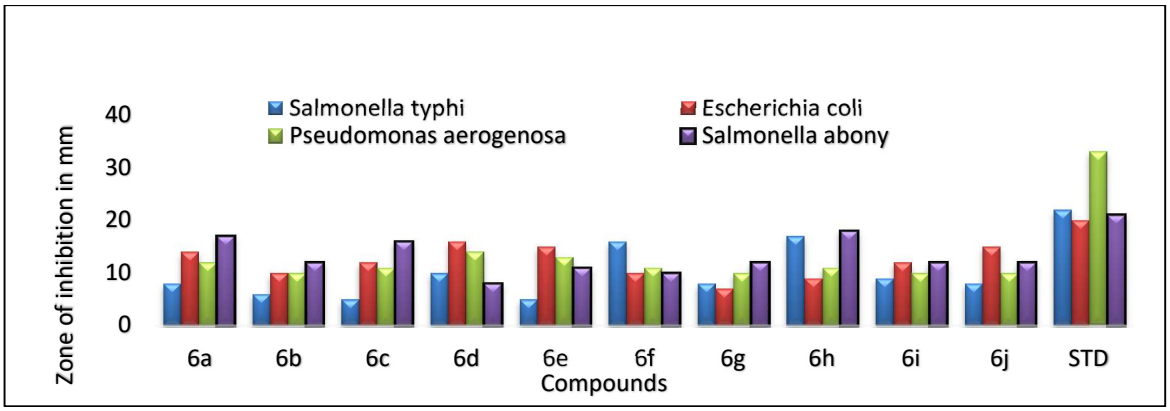


Fig.-4: Comparison of Anti-bacterial Activity of Compounds **6a-j** against Gram-negative Pathogen with Standard Drug Tetracyclin

Anti-fungal activity results reveal that the compound **6j** showed significant activity, compounds **6a, 6c, 6d, 6f, 6g** and **6h** moderate activity and compounds **6b, 6e** and **6i** showed weak activity comparable with standard drug Nystatin against fungal pathogens **(***Candida albicans, Saccharomyces cerevisiae, Aspergillusniger*). Anti-fungal activity reports of screened compounds graphically represented in Fig.-5.

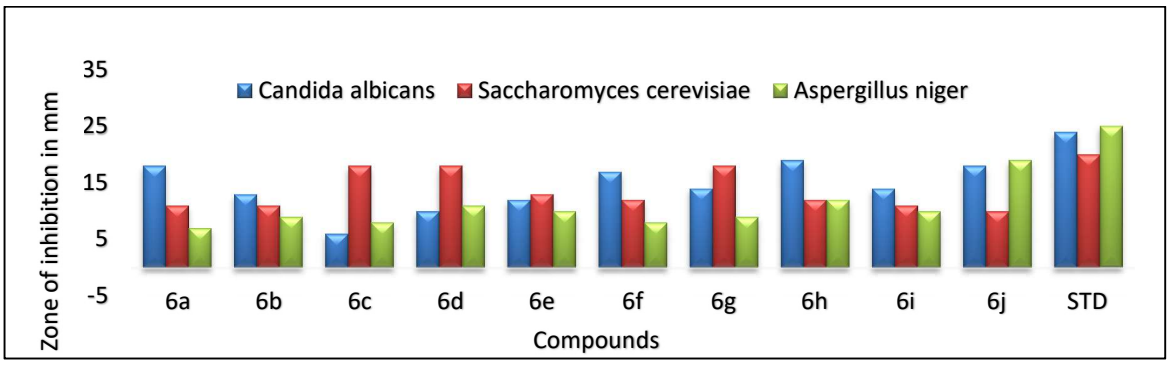


Fig.-5: Comparison of Anti-fungal Activity of Compounds 6a-j against Fungal Pathogen with Standard Drug Nystatin

CONCLUSION

In conclusion, a series of thiazole-pyrazole based pyrimidine derivatives **(6a-j)** were prepared and evaluated for their *in vitro* anti-inflammatory, anti-oxidant, anti-bacterial and anti-fungal properties. Among the compounds screened most of the compounds showed significant anti-inflammatory, anti-oxidant, anti-bacterial and anti-fungal activity. Compounds **6b, 6e, 6g** and **6j** exhibited prominent inhibition of protein denaturation compared to the reference drug Diclofenac sodium. Compounds **6b, 6e, 6j** and **6g** were found to be significant DPPH radical scavengers. Compounds **6i, 6d, 6j** and **6e** were significant inhibitors of NO radicals. Compound **6a** exhibited significant activity comparable with standard drug Tetracyclin against tested Gram-positive pathogens. Compounds **6a** and **6h** showed significant activity compared with standard drug Tetracyclin against tested Gram-negative pathogens and compound **6j** exhibited potent activity comparable with standard drug Nystatin against fungal pathogens.

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