Synthesis & Pharmacological Studies on Novel Pyrimidines Derived from Piperazine Chalcones

Sriramadasu Ushasri[1], Dr. Rahul Raj[2]
1 Research Scholar, Department of Pharmaceutical Chemistry, Glocal University Pharmacy College, Saharanpur, U.P
2 Research Guide, Glocal University Pharmacy College, Saharanpur, U.P

ABSTRACT

Treatment of 1-acetyl-4-(4-hydroxy phenyl) piperazine with aromatic or substituted aromatic aldehydes in presence of methanol and potassium hydroxide, formed 3-substituted phenyl-1--(4-(4- hydroxyl phenyl)- piperzin-1-yl)-Prop-2-en-1-one derivatives (RC-1 to RC-10). These Chalcones were used for condensation with guanidine hydrochloride to obtain pyrimidines. These are assayed for their antibacterial activity against Bacillus pumilus, Bacillus subtilis, Escherichia coli, and Proteus vulgaris; for antifungal activity against Aspergillus niger, Candida albicans strains. Antibacterial assay revealed that Bacillus subtilis and Proteus vulgaris were the most sensitive bacterial strains to compounds RC-2, 3, 4, 7, 10 and in the antifungal assay, compounds RC-1, 3, and 10 were highly effective against Aspergillus niger when compared to other investigated strains. The Pyrimidines were tested for anti Oxidant activity by Nitric Oxide radical scavenging method, measuring UV absorbance of solution of compound in Griess reagent and Super oxide radical method. Both the methods replicated the same results. Especially the compounds RP – 6, 9, 02 and 05 showed significant results.

Key words: 1-acetyl-4-(4-hydroxy phenyl) piperazine, chalcone, pyrimidine, aldehydes, potassium hydroxide, methanol, antibacterial activity, antifungal activity, antioxidant activity.

DOINumber: 10.48047/nq.2022.20.2.NQ22379 **NeuroQuantology2022;20(2):787-799**

INTRODUCTION

Pyrimidine can be regarded as a cyclic amine. Pyrimidine is also known as m- diazine (or) 1, 3-diazine. It is a six membered heterocyclic aromatic compound similar to benzene.

It contains two nitrogen atoms at 1st and 3rd positions. Pyrimidine is present throughout nature in various forms and is the building blocks of numerous natural compounds.

The most commonly recognized pyrimidines are the bases of RNA and DNA; cytosine, thymine or uracil. Pyrimidine and its derivatives have been described with a wide range of biological potential i.e. anticancer, antiviral, antimicrobial, anti-inflammatory, analgesic, anti oxidant and anti malarial etc. Various synthetic aspects indicated that pyrimidine derivatives are easy to synthesize and has diverse biological and chemical applications.

MATERIALS AND METHODS

Antimicrobial Activity

The synthesized compounds were screened for antimicrobial activity against gram-positive bacteria like Bacillus pumilus, Bacillus subtilis and gram negative bacteria like Escherichia coli

and Proteus vulgaris. Similarly the compounds were screened for antifungal activity against fungi like Aspergillus niger and Candida albicans by using cup-borer method. Ciprofloxacin and Miconazole nitrate were used as standard drugs. DMSO was used as solvent.

EXPERIMENTAL

Melting points of the synthesized compounds were determined by open capillary method and are uncorrected. Purity of the compounds was verified on TLC plates coated with silica gel. IR spectra were recorded on thermo Nicolet IR 200 spectrometer using KBr disc method. H^1NMR spectra were recorded on BRUKER amx-400 NMR spectrometer where $CDCl_3$ is used as internal standard. Results of Combustion analysis were found to be within the limits of permissible errors.

Synthesis of Piperazine containing chalcone derivatives

1E.q of 1-acetyl-4-(4-hydroxy phenyl) piperazine was dissolved in 10ml methanol and 2 E.q of ortho or para substituted aromatic aldehyde or unsubstituted aromatic aldehyde was added to it. To this mixture 3 E.q of 40% KOH was added drop wise at room temperature. This mixture was acidified with 1:1 hydrochloric acid and water with constant stirring. The obtained solid was purified and re crystallized by using a mixture of chloroform and n-hexane (6:4).

Fig.1. Scheme of piperazine containing chalcone derivatives

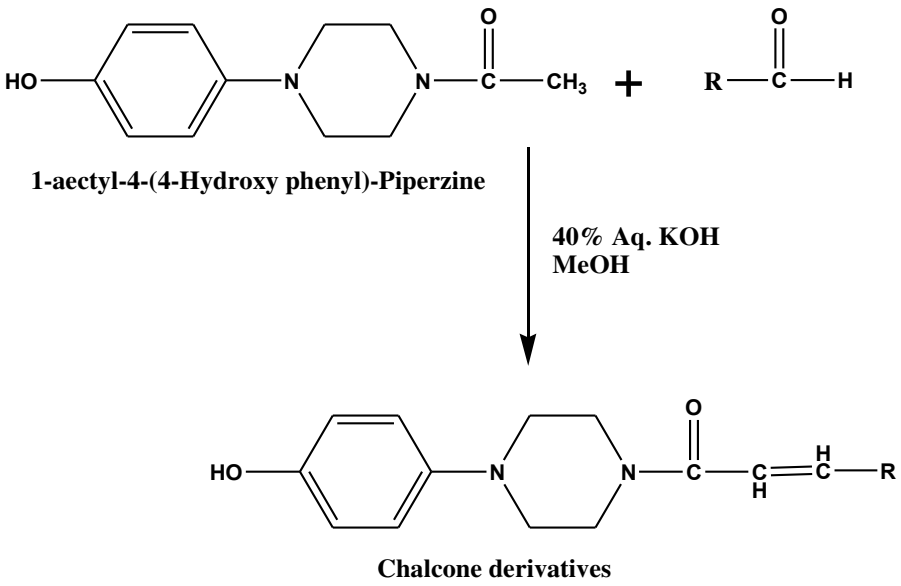


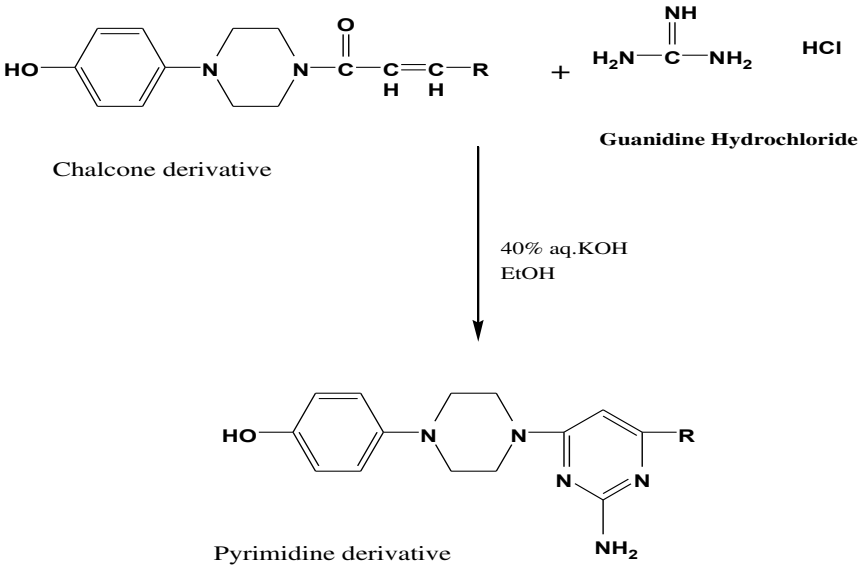
Table.1. Compound code containing derivatives

Compound code	R
RC-1	Phenyl
RC-2	4-cholro phenyl
RC-3	4- fluoro phenyl
RC-4	4- nitro phenyl
RC-5	4- methoxy phenyl

RC-6	2- ethoxy-4- hydroxy phenyl
RC-7	2,4- dimethoxy phenyl
RC-8	N,N- dimethyl amino phenyl
RC-9	Napthyl
RC-10	2,4-dichloro phenyl

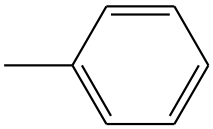
Synthesis of pyrimidine from chalcones:-

A mixture of chalcone of 1-acetyl-4-(4-hydroxy phenyl) piperazine (1 E.q.) and guanidine hydrochloride (3 E.q.) in absolute ethanol (10 ml), with KOH as base, were refluxed on a water bath for 72 hours. After that the solvent was completely evaporated and the residue was poured into ice cold water, the precipitated solid was collected by filtration and crystallized from suitable solvent to give the pyrimidine derivative.

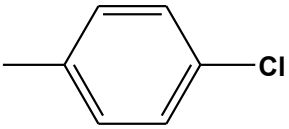


Chalcone moiety attached:

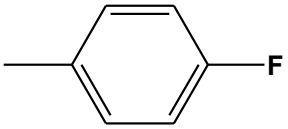
S.NO.	SAMPLE CODE	R
1.	RP-1	Phenyl
2.	RP-2	4-chloro phenyl
3.	RP-3	4-Fluoro phenyl
4.	RP-4	4′-nitro phenyl
5.	RP-5	4′-methoxy phenyl
6.	RP-6	3-ethoxy-4-hydroxy phenyl
7.	RP-7	2,4-diimethoxy phenyl
8.	RP-8	N,N-dimethyl amino phenyl
9.	RP-9	Napthyl
10.	RP-10	2,4-dichloro phenyl



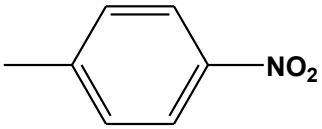
RP 1



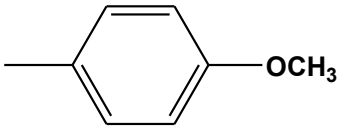
RP 2



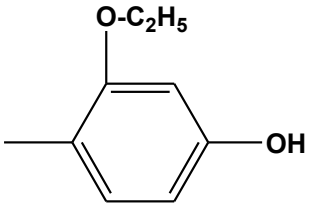
RP 3



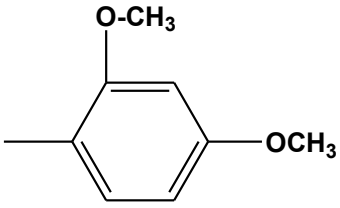
RP 4



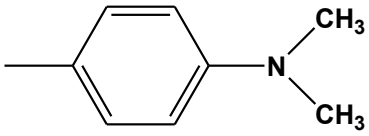
RP 5



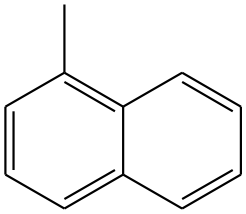
RP 6



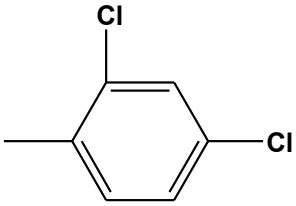
RP 7



RP 8



RP 9



RP 10

Spectral details

RP-1: 4-(4-(2- amino-6- phenyl pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 72%; mp 110 -112^{0}C; **IR (KBr) vcm^{-1}** 3346.29(-NH, str),1620.76 (-C=N, str), 1509.08(-C=C-, str); **H^1 NMR (ppm)** 1.246 & 2.993(8H, Piperzinyl protons), 3.763(2H, s, -NH_2), 6.004 & 8.472 (10H, aromatic protons), 7.068(1H, s, -C-5H), 10.420(1H, s, phenolic OH).

RP-2: 4-(4-(2- amino-6-(4-chloro phenyl) pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 71%; mp 117 -119^{0}C; **IR (KBr) vcm^{-1}** 3432.48(-NH, str), 1634.34(-C=N, str), 1452.10(-C=C-, str); **H^1 NMR (ppm)**1.26 & 2.863(8H, Piperzinyl protons), 3.653(2H, s, -NH_2), 7.811(1H, s, -C-5H), 10.516(1H, s, phenolic OH).

RP-3: 4-(4-(2- amino-6-(4- fluoro phenyl) pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 70.12%; mp 128 -130^{0}C; **IR (KBr) vcm^{-1}** 3433.72(-NH, str), 1636.63(-C=N, str), 1512.44(-C=C-, str); **H^1 NMR (ppm)** 1.289 & 2.902(8H, Piperzinyl protons), 3.563(2H, s, -NH_2), 7.088(1H, s, -C-5H), 9.976(1H, s, phenolic OH).

RP-4: 4-(4-(2- amino-6-(4- nitro phenyl) pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 64.5%; mp 124 -126^{0}C; **IR (KBr) vcm^{-1}** 3428.75(-NH, str), 1642.28(-C=N, str), 1541.41(-C=C-, str); **H^1 NMR (ppm)** 1.241 & 2.514(8H, Piperzinyl protons), 3.315(2H, s, -NH_2), 7.313(1H, s, -C-5H), 6.814 & 8.403(9H, aromatic protons), 10.214(1H, s, phenolic OH).

RP-5: 4-(4-(2- amino-6-(4- methoxy phenyl) pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 69%; mp 155 -157^{0}C; **IR (KBr) vcm^{-1}** 3437.75(-NH, str), 1641.11(-C=N, str), 1459.27(-C=C-, str); **H^1 NMR (ppm)** 1.248 & 2.511(8H, Piperzinyl protons), 3.305(2H, s, -NH_2), 7.306(1H, s, -C-5H), 9.894(1H, s, phenolic OH).

RP-6: 4-(4-(2- amino-6-(3-ethoxy-4- hydroxy phenyl) pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 64.5%; mp 137 -139^{0}C; **IR (KBr) vcm^{-1}** 3435.06(-NH, str), 1640.24(-C=N, str), 1456.07(-C=C-, str); **H^1 NMR (ppm)** 1.248 & 2.312(8H, Piperzinyl protons), 3.301(2H, s, -NH_2), 7.114(1H, s, -C-5H), 10.127(1H, s, phenolic OH).

RP-7: 4-(4-(2- amino-6-(3,4-dimethoxy phenyl pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 62%; mp 145 -148^{0}C; **IR (KBr) vcm^{-1}** 3361.86(-NH, str), 1622.10(-C=N, str), 1532.86(-C=C-, str); **H^1 NMR (ppm)** 1.244 & 2.516(8H, Piperzinyl protons), 3.304(2H, s, -NH_2), 6.648 & 8.317(8H, aromatic protons),7.132(1H, s, -C-5H), 10.416(1H, s, phenolic OH).

RP-8: 4-(4-(2- amino-6-(4-(dimethyl amino) phenyl) pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 67.4%; mp 135 -137^{0}C; **IR (KBr) vcm^{-1}** 3320.75(-NH, str), 1621.52(-C=N, str), 1516.19(-C=C-, str); **H^1 NMR (ppm)**

2.513 & 2.913(8H, Piperzinyl protons), 3.359(2H, s, $-NH_2$), 7.357(1H, s, -C-5H), 6.746 & 7.357(9H, aromatic protons), 9.465(1H, s, phenolic OH).

RP-9: 4-(4-(2- amino-6-(napth-1-yl) pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 61.5%; mp 244 -246^{0}C; **IR (KBr) vcm^{-1}** 3431.44(-NH, str), 1688.19(-C=N, str), 1507.69(-C=C-, str); **H^1 NMR (ppm)** 1.241 & 2.518(8H, Piperzinyl protons), 3.314(2H, s, $-NH_2$), 7.798(1H, s, -C-5H), 7.117 & 9.171(12H, aromatic protons), 10.431(1H, s, phenolic OH).

RP-10: 4-(4-(2- amino-6-(2,4-dicholro phenyl) pyrimidin-4-yl)- piperzin-1-yl) Phenol; Yield 65.7%; mp 135 -137^{0}C; **IR (KBr) vcm^{-1}** 3384.31(-NH, str), 1619.84(-C=N, str), 1569.43(-C=C-, str); **H^1 NMR (ppm)** 1.218 & 2.576(8H, Piperzinyl protons), 3.375(2H, s, $-NH_2$), 7.623(1H, s, -C-5H), 6.056 & 7.944(8H, aromatic protons), 9.571(1H, s, phenolic OH).

RESULTS & DISCUSSION

Anti Bacterial Activity of Pyrimidines:

Compound code	Zone of inhibition (in mm)			
	B.subtilis		E.coli	
	500 µg/ml	1000 µg/ml	500 µg/ml	1000 µg/ml
RP-1	9	14	-	11
RP-2	13	16	-	14
RP-3	16	18	-	10
RP-4	11	14	-	11
RP-5	13	15	-	10
RP-6	12	15	-	9
RP-7	13	17	-	11
RP-8	10	12	-	-
RP-9	12	16	-	9
RP-10	14	20	-	17
Sparfloxacin	22	25	26	28
Control	-	-	-	-

(-) no zone of inhibition

From the above results it is evident that synthesized pyrimidines (RP-1 to RP-10) showed significant gram positive antibacterial activity at both 0.5% (500µg/ml) and 1%(1000µg/ml) concentration levels, but a poor gram negative effect that too at 1%(1000µg/ml) when compared with standard drug Sparfloxacin. In particular compounds like., RP-2 & 10 showed maximum activity against because of presence of chlorine substituent on the aromatic rings.

Anti Fungal Activity of Pyrimidines

Compound code	Zone of inhibition (in mm)	
	Aspergillus niger	*Candida albicans*
RP-1	12	-
RP-2	10	-
RP-3	-	-
RP-4	14	10
RP-5	10	-
RP-6	-	-
RP-7	18	14
RP-8	17	14
RP-9	18	13
RP-10	16	14
Miconazole nitrate	30	27
Control	-	-

(-) no zone of inhibition

Statistical Analysis:-

From the above results it is evident that synthesized pyrimidines (RP-7 to RP-9) showed significant antifungal activity.

C. Anti-Oxidant Activity of Pyrimidines:

Nitric oxide radical scavenging activity:

CALCULATION:

$$\%\text{INHIBITION} = \frac{\text{ABSORBANCE(BLANK)} - \text{ABSORBANCE(SAMPLE)}}{\text{ABSORBANCE(BLANK)}} \times 100$$

Table- 4.1 - % Scavenging activity of Pyrimidine derivatives by Nitric oxide method (RP 01, 03, 06 and 07)

S.No	C0NCENTRATIONS	CONTROL	STANDARD	RP 01	RP 03	RP 06	RP 07
1.	25	6.78	23.08	5.45	7.87	9.34	7.65
2.	50	6.78	34.64	9.39	13.21	19.34	14.31
3.	75	6.78	45.17	12.77	16.43	23.65	16.98

4.	100	6.78	59.29	16.66	23.58	36.98	22.95
5.	125	6.78	67.43	21.76	34.16	46.33	35.78
6.	150	6.78	79.97	29.65	42.98	55.89	43.06
7.	175	6.78	87.76	31.55	55.67	65.32	55.72
8.	200	6.78	99.3	36.58	59.06	75.67	59.68

Fig: 4.1- % Scavenging activity of Pyrimidine derivatives by Nitric oxide method (RP 01, 03, 06 and 07)

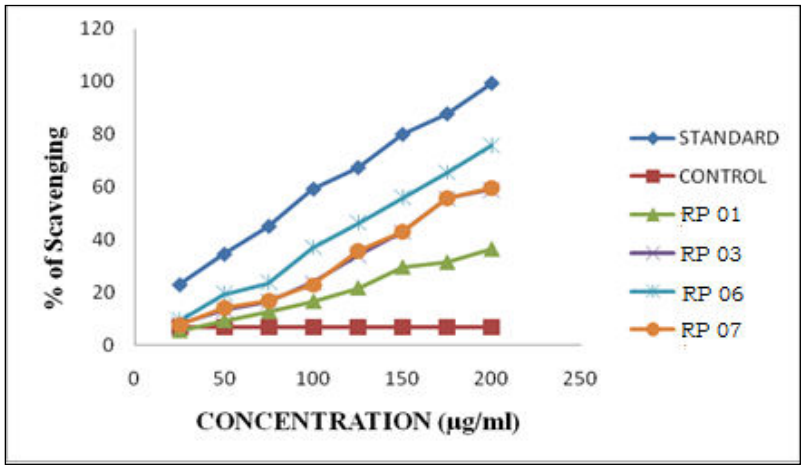


Table- 4.2 - % Scavenging activity of Pyrimidine derivatives by Nitric oxide method (RP 04 and RP 08)

S.No	C0NCENTRATIONS	CONTROL	STANDARD	RP 04	RP 08
1	25	6.78	23.08	4.55	4.65
2	50	6.78	34.64	7.01	7.34
3	75	6.78	45.17	10.54	10.83
4	100	6.78	59.29	15.59	12.54
5	125	6.78	67.43	19.34	18.43
6	150	6.78	79.97	21.76	22.75
7	175	6.78	87.76	26.77	28.01
8	200	6.78	99.3	29.33	31.25

Fig: 4.2 - Scavenging activity of Pyrimidine derivatives by Nitric oxide method

(RP 04 and RP 08)

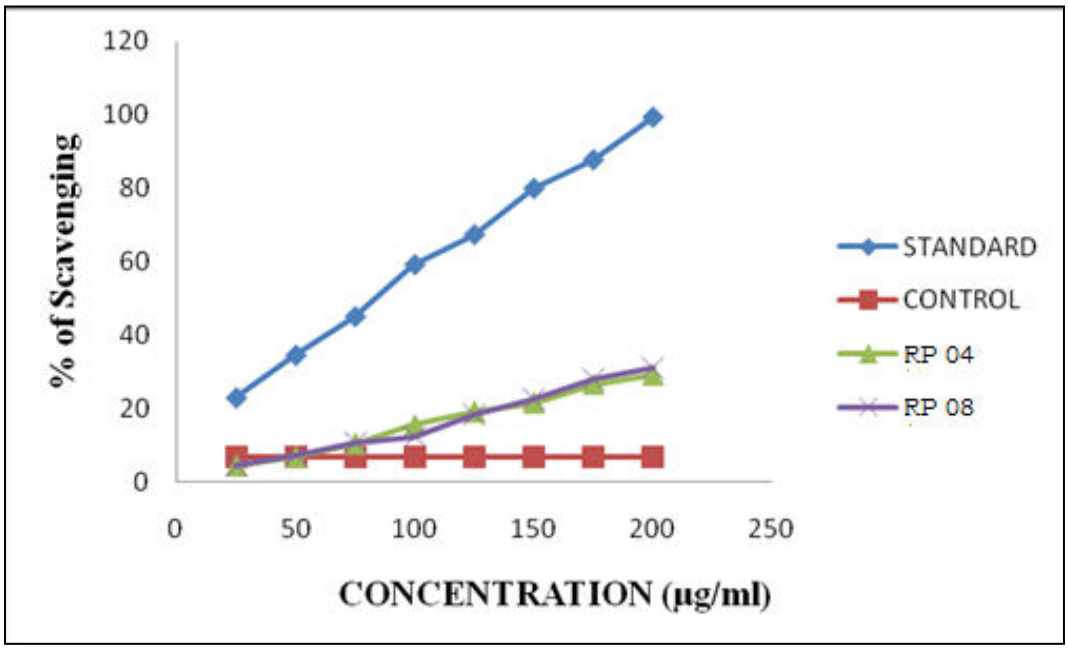
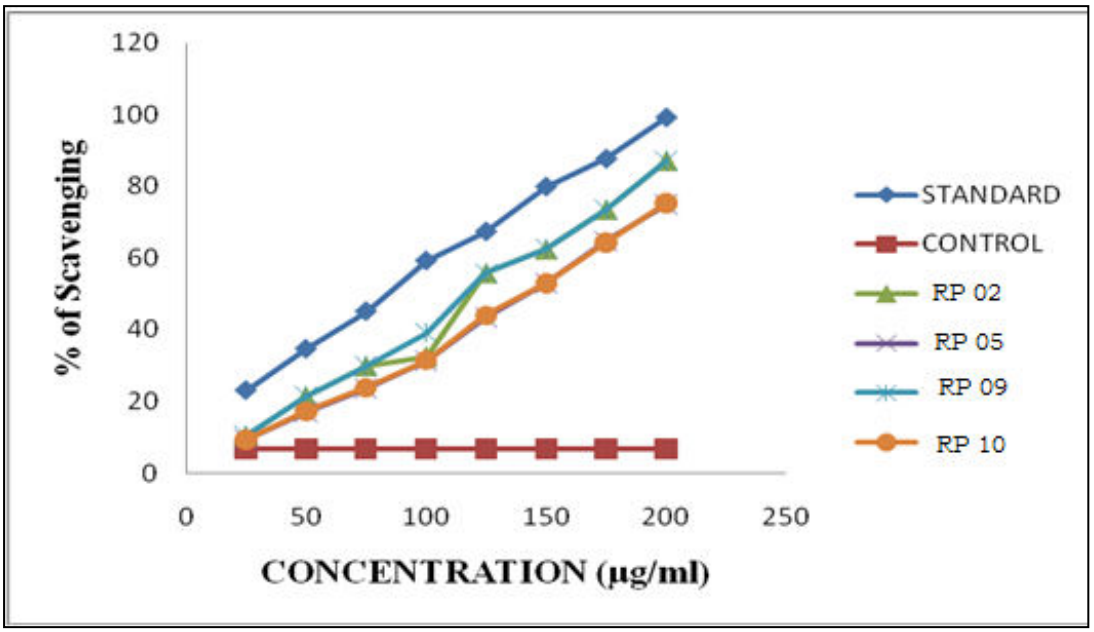


Table- 4.3 - % Scavenging activity of Pyrimidine derivatives by Nitric oxide method

(RP 02, 05, 09, 10)

S.No.	C0NCENTRATIONS	STANDARD	RP 02	RP 05	RP 09	RP 10
1	0	0	0	0	0	0
2	25	23.08	15.98	16.72	10.43	9.21
3	50	34.64	27.09	28.04	21.4	16.98
4	75	45.17	38.23	34.76	29.87	23.67
5	100	59.29	49.76	48.76	32.56	31.34
6	125	67.43	59.05	60.54	55.74	43.45
7	150	79.97	68.67	68.54	62.32	52.81
8	175	87.76	76.96	76.51	73.44	64.67
9	200	99.3	86.54	85.68	87.07	74.98

Fig: 4.3 - % Scavenging activity of Pyrimidine derivatives by Nitric oxide method (RP 02, 05, 09, 10)



SUPER OXIDE RADICAL METHOD)

Table- 4.4 - % Scavenging activity of Pyrimidine derivatives by Super oxide radical method (RP 01, 03, 06 and 07)

S.No.	C0NCENTRATIONS	STANDARD	CONTROL	RP 01	RP 03	RP 06	RP 07
1	25	25.65	7.93	7.43	7.87	10.45	7.65
2	50	34.64	7.93	10.2	13.67	21.64	12.75
3	75	45.17	7.93	13.03	16.73	27.9	15.78
4	100	59.29	7.93	16.85	24.6	38.76	22.14
5	125	67.43	7.93	22.43	34.98	47.43	34.9
6	150	79.97	7.93	28.34	43.32	56.45	42.27
7	175	87.76	7.93	32.64	52.39	65.75	55.01
8	200	99.3	7.93	38.03	60.46	78.34	58.95

Fig- 4.4 - % Scavenging activity of Pyrimidine derivatives by Super oxide radical method (RP 01, 03, 06 and 07)

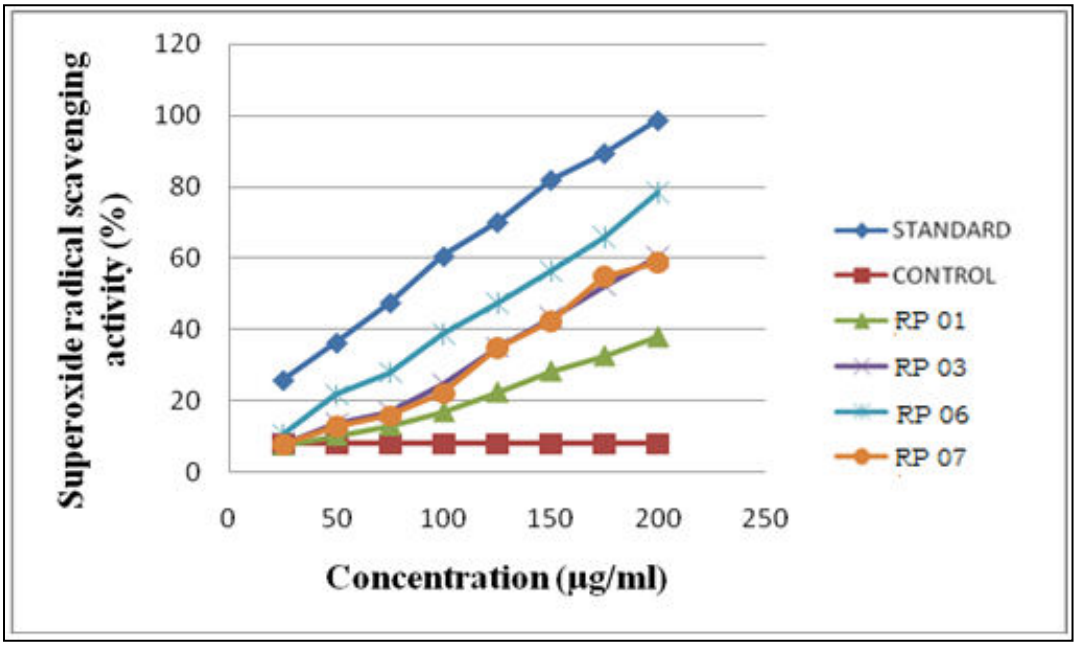


Table- 4.5 - % Scavenging activity of Pyrimidine derivatives by Super oxide radical method (RP 04 and 08)

S.No.	C0NCENTRATIONS	STANDARD	CONTROL	RP 04	RP 08
1	25	25.65	7.93	7.65	8.9
2	50	34.64	7.93	10.9	12.34
3	75	45.17	7.93	12.85	14.9
4	100	59.29	7.93	17.32	19.57
5	125	67.43	7.93	21.66	23.75
6	150	79.97	7.93	26.03	27.84
7	175	87.76	7.93	30.31	30.42
8	200	99.3	7.93	32.45	32.56

Fig - 4.5 - % Scavenging activity of Pyrimidine derivatives by Super oxide radical method (RP 04 and 08)

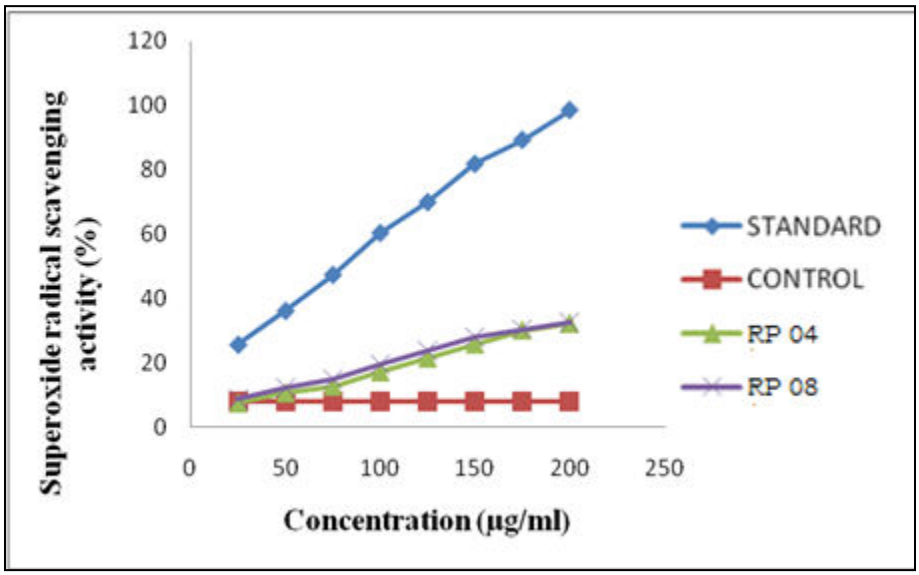
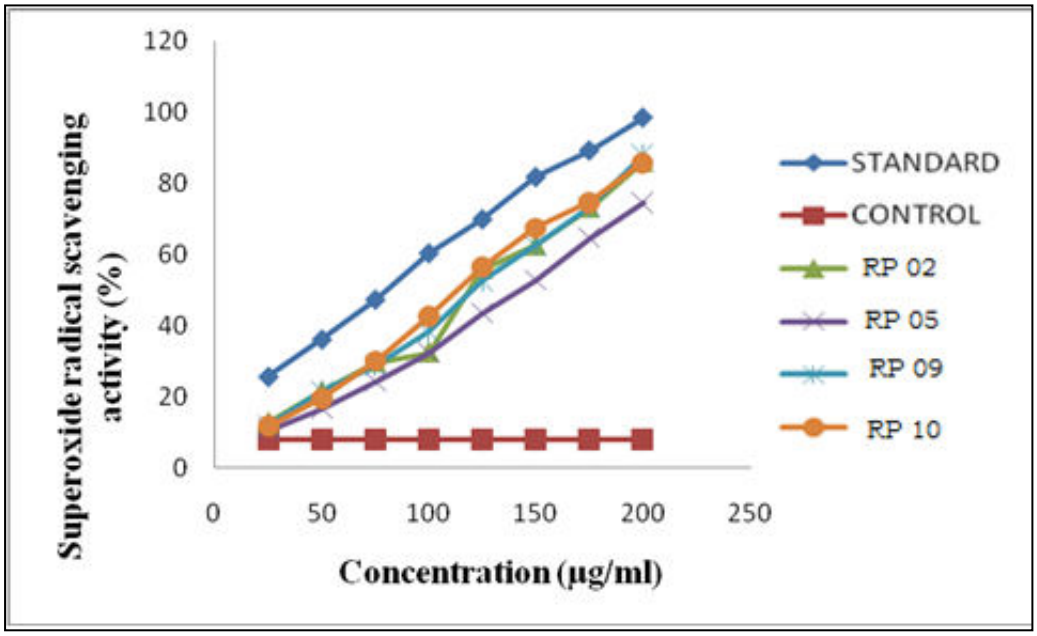


Table- 4.6 - % Scavenging activity of Pyrimidine derivatives by Super oxide radical method (RP 02, 05, 09, 10)

S.No.	CONCENTRATIONS	STANDARD	CONTROL	RP 02	RP 05	RP 09	RP 10
1	25	25.65	7.93	11.97	11.67	12.76	10.45
2	50	34.64	7.93	21.54	19.54	21.58	16.73
3	75	45.17	7.93	28.95	30.18	29.87	24.24
4	100	59.29	7.93	38.38	42.56	32.56	32.36
5	125	67.43	7.93	52.81	56.65	56.05	43.45
6	150	79.97	7.93	62.65	67.54	62.76	52.81
7	175	87.76	7.93	73.46	74.76	73.41	64.67
8	200	99.3	7.93	88.21	85.98	86.34	74.63

Fig - 4.6 - % Scavenging activity of Pyrimidine derivatives by Super oxide radical method (RP 02, 05, 09, 10)



Statistical Analysis:

The compounds were tested for anti Oxidant activity by Nitric Oxide radical scavenging method, measuring UV absorbance of solution of compound in Griess reagent and Super oxide radical method. Both the methods replicated the same results. Especially the compounds RP – 6, 9, 02 and 05 showed significant results.

REFERENCE

1. J.A.Joule, K.Mills, 'Hetero cyclic chemistry', published by Blackwell science Ltd., 2000, Chapter 9, 170-186.
2. Y.M. Lin, Y. Zhou, M. Flavin, L.M. Zhou, W. Nie and F.C. Chen, "Chalcones and flavonoids as anti-tuberculosis agents", Bioorganic & Medicinal Chemistry, Vol. 10, 2002, pp. 2795-2802.
3. B. Bandgar, S. Gawande, R. Bodade, N. Gawande and C. Khobragade, "Synthesis and biological evaluation of a novel series of pyrazolechalcones as anti-inflammatory, antioxidant and antimicrobial agents", Bioorganic & Medicinal Chemistry, Vol. 17, 2009, pp. 8168-8173.
4. M.L. Go, X. Wu and X.L. Liu, "Chalcones: an update on cytotoxic and chemoprotective properties", Current Medicinal Chemistry, Vol. 12, 2005, pp. 483-499.
5. Leusink, A.J.; Noltes, J.G. (1966). "Reaction of organotin hydrides with α,β-unsaturated ketones". *Tetrahedron Letters* 7 (20): 2221.