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Conventional and microwave assisted synthesis of 2-amino-4,6-diaryl pyrimidine derivatives and their cytotoxic, anti-oxidant activities

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ABSTRACT

Pyrimidine is the parent substance of a large group of heterocyclic compounds and plays a vital role in many biological processes. It is also evident from literature; pyrimidines possess potential anti oxidant activities and cytotoxic activities. Chemoprotection by pyrimidines may be a consequence of their antioxidant properties, mediated via inhibition or induction of metabolic enzymes, by an anti-invasive effect or a reduction in nitric oxide production. Free radicals are formed constantly in human system either as accidental products during metabolism or deliberately during the process of phagocytosis; or due to environmental pollutants, ionizing radiations, ozone, heavy metal poisoning, etc. Therefore, it is worthwhile to synthesize some pyrimidine derivatives by conventional and microwave (Catalyst systems) assisted synthesis methods. The synthesized compounds were purified by recrystallization or by chromatography and are characterized by ^{1}H NMR, ^{13}C NMR and IR analysis. The compounds were tested for their potential cytotoxic activity and antioxidant activities by standard methods. The microwave irradiation method (MWI) is proved to be advantageous with considerable increase in the reaction rate with better yields, after over all observation it is found that pyrimidine derivatives possessing cytotoxic and anti-oxidant activities.

1. Introduction

Nitrogen containing heterocyclic compounds has received considerable attention due to their wide range of pharmacological activity. The increasing importance of pyrimidine and its derivatives as intermediates for the synthesis of biologically active compounds has led to continued development of new simple procedures for their synthesis. Pyrimidine is the parent substance of a large group of heterocyclic compounds and plays a vital role in many biological processes, as found in nucleic acids, several vitamins, co-enzymes and purines, etc. Pyrimidines are considered to be an important precursor because they are integral part of the genetic material *viz*, DNA and RNA as nucleotides and nucleosides but they also impart numerous biological activities such as bactericides, fungicides, vermicides and insecticides. They also found application in agricultural and industrial chemicals. The chemistry of pyrimidines [1] and its derivatives has been studied since past century due to their close pharmacological association with diverse pharmacological properties. Though pyrimidine itself does not exist in nature but substituted pyrimidines are found as part of more complex system and are widely distributed.

These pyrimidine derivatives have been reported to possess a variety of biological activities [2-6], notable among which are the antibacterial [7], anticancer [8], anti-inflamatory [9], antitubercular [10] and analgesic [11] activities.

The chemistry of pyrimidines [12,13] has been discussed previously. Therefore, in the present investigation it has been considered worthwhile to synthesize some new pyrimidine derivatives by conventional and microwave irradiation methods (MWI) (the industrial type of microwave oven used is system catalyst) and comparison between two methods. Therefore a number of pyrimidine derivatives (2,4,6-trisubstituted pyrimidines) were synthesized and in view of varied biological and pharmacological importance. It is felt worthwhile to evaluate them for their possible activities; these compounds therefore were screened for cytotoxic activity and anti-oxidant activities etc.

The synthesized compounds were purified by using recrystallization (ethanol) or by chromatographic (ethyl acetate/hexane mixture) methods. The compounds were characterized by ^{1}H NMR, ^{13}C NMR and IR analysis. The physical properties of the compounds were also included.

2. Experimental

2.1. Instrumentation

Melting point of the synthesized derivatives was determined by digital melting point apparatus (SMP 10). The NMR spectra were recorded on a BRUKER DRX 400 spectrometer at 400 MHz (^{1}H) and at 100 MHz (^{13}C). IR spectra were recorded using Bruker ALPHA FT-IR spectrometer. High resolution mass spectra were obtained on Agilent 6100 Series Single Quadrupole LC/MS.

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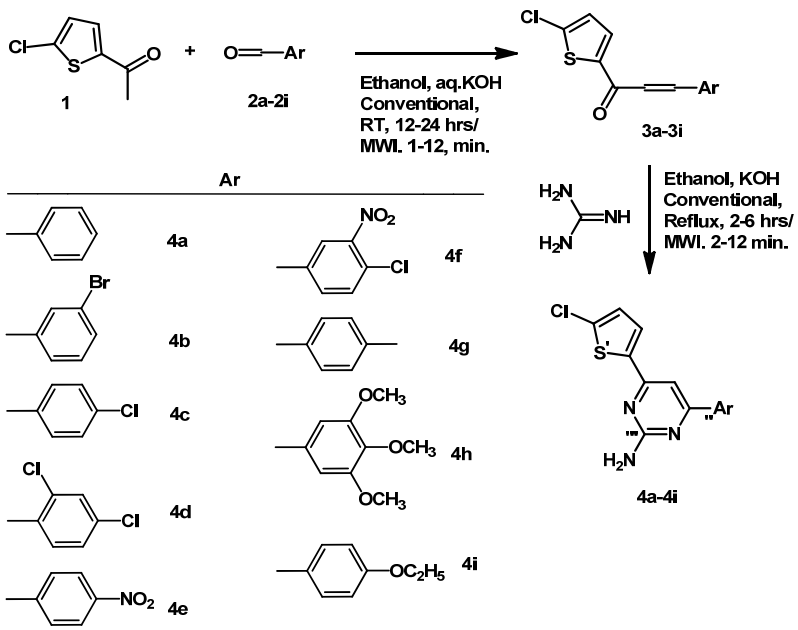

Scheme 1

2.2. General procedure for the synthesis of pyrimidines

2.2.1. Conventional method

The condensation of the chalcones (0.001 mol) with guanidine hydrochloride (0.001 mol) in alkaline medium *viz*, in potassium hydroxide (0.003 mol) in the presence of ethanol (10 mL) at reflux temperatures (2 to 6 hours) resulted the formation of corresponding pyrimidine [14-18] derivatives (Scheme 1). Completion of the reaction was identified by observing on precoated thin layer chromatography (TLC) plates, using ethyl acetate and hexane mixture as mobile phase. The pyrimidine derivatives on purification obtained as fine wine red powder.

2.2.2. Microwave irradiation method

The condensation of the chalcones (0.001 mol) with guanidine hydrochloride (0.001 mol) in alkaline medium *viz*, in potassium hydroxide (0.003 mol) in the presence of ethanol (10 mL), the entire reaction mixture was microwave irradiated at 180 watts for about 2-16 minutes, then kept aside for 2-3 hrs, resulted the formation of corresponding pyrimidine derivatives (Scheme 1) (Catalyst Scientific Microwave Oven, Model: CATA 2R, Range: 140-700 W, Make: Catalyst System, Pune, India). Reaction completion was identified by TLC precoated plates. Ethyl acetate and hexane mixture was used as mobile phase. The pyrimidine derivatives on purification obtained as fine wine red powder.

2.3. Cytotoxic activity

2.3.1. Brine shrimp lethality test (BSLT)

Brine shrimp lethality test have been used as bioassay [19-24] for a variety of toxic substances. This method has also been applied to compounds in order to facilitate the isolation of biologically active compounds. A general bioassay that appears capable of detecting a broad spectrum of bioactivity, present in crude extracts and in synthetic compounds is the brine shrimp lethality bioassay, to study the cytotoxic activity of 2,4,6-trisubstituted pyrimidine derivatives (Compounds **4a-i**).

Brine shrimp lethality bioassay was carried out to investigate the cytotoxicity of the synthesized compounds. Brine shrimps (Artemia salina) were hatched using brine shrimp eggs in a conical shaped vessel (1.0 L), filled with sterile artificial sea water (prepared using sea salt 38 g/L and adjusted to pH = 8.5 using 1.0 N NaOH) under constant aeration for 38 h. After hatching, active nauplii free from egg shells were collected from brighter portion of the chamber and used for the assay. Ten nauplii were drawn through a glass capillary and placed in each vial containing 5 mL of brine solution. In each experiment, test substances whose activities are to be checked were added to the vial according to their concentrations and maintained at room temperature for 24 h under the light and surviving larvae were counted.

Experiments were conducted along with control (vehicle treated), different concentrations (1-5000 µg/mL) of the test substances in a set of three tubes per dose. Replicas should be maintained to get accurate results.

2.3.2. Statistical analysis

The percentage lethality was calculated from the mean survival larvae of compounds treated tubes and control. ED_{50} values were obtained by (best-fit line method) plotting a graph, taking concentration on x-axis and percentage inhibition on y-axis, at 50% of the percentage inhibition the line was drawn from y-axis and aligned with the concentration on x-axis then got the ED_{50} values.

2.4. Antioxidant activity

Free radicals are formed constantly in human system either as accidental products during metabolism or deliberately during the process of phagocytosis; or due to environmental pollutants, ionizing radiations, ozone, heavy metal poisoning, cigarette smoking and chronic alcohol intake. Free radicals being highly reactive can oxidize biomolecules leading to tissue injury and cell death.

Table 1. Comparative reaction time and percentage yield of pyrimidine derivatives by conventional and microwave irradiation methods.

Compound No	Reaction time		Yield (%)	
	Conventional (hr)	MWI (min)	Conventional	MWI
4a	4.5	5.5	35	48
4b	4.5	4.5	32	47
4c	5.5	5.0	42	56
4d	5.0	4.5	49	56
4e	5.5	6.0	34	43
4f	5.0	4.5	41	44
4g	5.0	5.0	39	52
4h	5.0	6.0	34	45
4i	6.0	6.5	39	51

Table 2. Melting points and elemental analysis of pyrimidine derivatives.

Compound No	R_f value	M.p., °C	Elemental analysis, %	
			Calculated	Found
4a	0.62	120 ± 2	C: 58.39 H: 3.47 N: 14.59	C: 58.42 H: 3.45 N: 14.57
4b	0.59	138 ± 2	C: 45.82 H: 2.45 N: 11.45	C: 45.85 H: 2.47 N: 11.47
4c	0.64	130 ± 2	C: 52.14 H: 2.79 N: 13.03	C: 52.11 H: 2.82 N :13.00
4d	0.60	145 ± 2	C: 47.11 H: 2.24 N: 11.77	C: 47.09 H: 2.12 N: 11.79
4e	0.54	142 ± 2	C: 50.49 H: 2.70 N: 16.83	C: 50.52 H: 2.68 N: 16.85
4f	0.56	160 ± 2	C: 47.75 H: 2.17 N: 15.25	C: 45.77 H: 2.14 N: 16.85
4g	0.63	136 ± 2	C: 59.66 H: 3.77 N: 13.92	C: 59.69 H: 3.99 N: 13.90
4h	0.51	98 ± 2	C: 53.99 H: 4.23 N: 11.11	C: 53.97 H: 4.27 N: 11.14
4i	0.57	127 ± 2	C: 57.86 H: 4.21 N: 12.65	C: 57.89 H: 4.23 N: 12.62

In the present study *in vitro* antioxidant activity is done by nitro blue tetrazolium (NBT). The IC_{50} values of chalcones tested for their antioxidant activity. Solvent used in the test for compounds was DMSO (dimethylsulphoxide).

2.4.1. Superoxide free-radical scavenging activity (Riboflavin photo reduction method (NBT))

Superoxide scavenging activity of the compounds was determined by Mc Cord and Fridovich method [25], which depends on light induced superoxide generation by riboflavin and corresponding reduction of nitro blue tetrazolium (NBT). The assay mixture contained EDTA (Ethylenediamine tetra-acetic acid) solution (6.6 mM) containing NaCN (3.0 µg), riboflavin (2.0 µM), NBT (50.0 µM), test substances and phosphate buffer (67.0 mM, pH = 7.8) in a final volume of 3.0 mL. The absorbance at 560 nm were measured before and 15 minutes after illumination. All tests were run in triplicate and mean values were used to calculate percentage scavenging ability and IC_{50} values were calculated (using linear regression analysis). The inhibitory effects of samples on the generation of superoxide anions were estimated by the equation 1.

$$\text{Percentage Inhibition} = [(A_0-A_1) \times 100]/A_0 \quad (1)$$

where A_0 is the absorbance with no addition of sample, A_1 is the absorbance with addition of sample. To study superoxide free radical scavenging activity of 2,4,6-trisubstituted pyrimidine derivatives (**4a-i**).

3. Results and discussion

In the present study, we have performed the synthesis of pyrimidine derivatives by conventional and microwave irradiation method in Scheme 1 but to reduce the reaction time, it was decided to synthesize the compounds by microwave irradiation, which can be more effective, faster and energy efficient in addition; we have compared those with others that were obtained via conventional heating methods and results were mentioned in Table 1-3. The compounds have been confirmed by spectral data, elemental analysis. The synthesized compounds were evaluated by various pharmacological methods like cytotoxic activity and anti-oxidant activities, pharmacological activities, based upon the type of substitution present on aromatic ring-b, the compounds are showing moderate activity or more activity. The procedure and results of both the activities are given below and the compounds **4a**, **4f** and **4g** proven to be more potent for cytotoxic activity, the compounds **4c**, **4d**, **4f** and **4h** possessing good anti-oxidant activity.

3.1. Cytotoxic activity

Brine shrimp lethality test have been used as bioassay for variety of toxic substances. All the 2,4,6-tri substituted pyrimidine derivatives (**4a-i**), were tested for cytotoxic activity by the Brine shrimp lethality test (BSLT) bioassay method, and all the compounds were found to possess cytotoxic activity. DMSO was used as a solvent. Among them few compounds showed a dose dependent cytotoxic activity at concentrations of (**4a**) 4.13 µg/mL, (**4c**) 4.97 µg/mL, (**4f**) 4.13 µg/mL, (**4g**) 3.61 µg/mL and (**4i**) 36.61 µg/mL.

Table 3. Characterization data of the pyrimidine derivatives is given below.

Compound No	IR (cm^{-1}), ^1H NMR, ^{13}C NMR, Mass
4a	IR (ν_{max}, cm^{-1}): 3432 (NH_2), 1627 (C=N), 1408 (C=C), 771 (C-S). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 5.78 (2H, d, C-2-NH_2), 6.24 (1H, d, *J* = 4 Hz, C-4'-H), 6.31 (1H, d, *J* = 4.2 Hz, C-3'-H), 6.79 (1H, t, *J* = 3.2 Hz, C- 4''-H), 7.12 (2H, d, *J* = 8 Hz, C-3'' and 5''-H), 7.69 (1H, s, C-5-H), 7.83 (2H, d, *J* = 8.8 Hz, C-2'' and 6''-H).
4b	IR (ν_{max}, cm^{-1}): 3394 (NH_2), 1673 (C=N), 1621 (C=C), 775 (C-S), 858 (C-Br). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 6.87 (2H, s, C-2-NH_2), 7.28 (1H, d, *J* = 4 Hz, C-4'-H), 7.51 (1H, t, *J* = 7.6 Hz, C- 5''-H), 7.71 (1H, d, *J* = 4.6 Hz, C-3'-H), 7.77 (1H, s, C-5-H), 8.06 (1H, d, *J* = 3.6 Hz, C-4''-H), 8.21 (1H, d, *J* = 8 Hz, C-6''-H), 8.41 (1H, s, C-2''-H).
4c	IR (ν_{max}, cm^{-1}): 3421 (NH_2), 1628 (C=N), 1567 (C=C), 812 (C-Cl), 772 (C-S). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 6.83 (2H, s, C-2-NH_2), 7.27 (1H, d, *J* = 4 Hz, C-4'-H), 7.61 (2H, d, *J* = 7.4 Hz, C-3'' and 5''-H), 7.74 (1H, s, C-5-H), 8.03 (2H, d, *J* = 4 Hz, C-2'' and 6''-H), 8.23 (1H, d, *J* = 8.2 Hz, C-3'-H). ^{13}C NMR ($CDCl_3$, 100 MHz, δ, ppm): 101.28 (C-5), 126.16 (C-3'), 127.36 (C-5'), 128.36 (C-2'' and 6''), 128.97 (C-3'' and 5''), 134.50 (C-4'), 135.81 (C-4''), 136.80 (C-2'), 141.51 (C-4), 159.99 (C-6), 163.25 (C-1''), 164.86 (C-2).
4d	IR (ν_{max}, cm^{-1}): 3406 (NH_2), 1627 (C=N), 1567 (C=C), 816 (C-Cl), 760 (C-S). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 6.93 (2H, s, C-2-NH_2), 7.11 (1H, d, *J* = 4 Hz, C-4'-H), 7.23 (1H, d, *J* = 4 Hz, C- 6''-H), 7.31 (1H, s, C-5-H), 7.57 (1H, d, *J* = 3 Hz, C-4''-H), 7.77 (1H, d, *J* = 4 Hz, C- 3'-H), 7.86 (1H, d, *J* = 8 Hz, C-3''-H).
4e	IR (ν_{max}, cm^{-1}): 3404 (NH_2), 1606 (C=N), 1568 (C=C), 1519 (Ar-NO_2), 771 (C-S). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 6.51 (2H, s, C-2-NH_2), 6.95 (1H, d, *J* = 4 Hz, C-4'-H), 7.12 (1H, d, *J* = 8.2 Hz, C-2''-H), 7.45 (1H, s, C-5-H), 7.73 (1H, d, *J* = 7.8 Hz, C-6''-H), 7.81 (1H, d, *J* = 4 Hz, C-3'-H), 8.12 (1H, d, *J* = 8 Hz, C-5''-H), 8.44 (1H, d, *J* = 7.4 Hz, C-3''-H).
4f	IR (ν_{max}, cm^{-1}): 3393 (NH_2), 1632 (C=N), 1571 (C=C), 1533 (Ar-NO_2), 768 (C-S). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 6.97 (2H, s, C-2-NH_2), 7.28 (1H, d, *J* = 4 Hz, C-4'-H), 7.87 (1H, s, C-5-H), 7.97 (1H, d, *J* = 8 Hz, C-5''-H), 8.04 (1H, d, *J* = 3.8 Hz, C-3'-H), 8.51 (1H, d, *J* = 8.4 Hz, C-6''-H), 8.83 (1H, s, C-2''-H).
4g	IR (ν_{max}, cm^{-1}): 3400 (NH_2), 1619 (C=N), 1568 (C=C), 801 (C-Cl), 756 (C-S). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 1.58 (3H, s, C-4''-CH_3), 6.95 (1H, d, *J* = 4.2 Hz, C-4'-H) , 7.05 (1H, d, *J* = 4.5 Hz, C-3'-H), 7.22 (1H, s, C-5-H), 7.29 (2H, d, NH_2), 7.72 (2H, m, C-3''and 5''-H), 8.03 (2H, m, C-2'' and 6''-H). ^{13}C NMR ($CDCl_3$, 100 MHz, δ, ppm): 21.38 (C-4''-CH_3), 101.33 (C-5), 125.9 (C-4), 126.97 (C-2'' and 6'') 127.29 (C-3'), 129.47 (C-3'' and 5''), 134.13 (C-5'), 134.60 (C-6), 140.94 (C-4'), 141.82, (C-4''), 159.63 (C-1''), 163.26 (C-2'), 166.15 (C-2). MS (*m/z*, %): 302.3 $[M+H, 100]^+$.
4h	IR (ν_{max}, cm^{-1}): 3384 (NH_2), 3004 (C-H-CH_3), 1590 (C=N), 1572 (C=C), 1226 (C-O-C), 770 (C-S). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 3.71 (3H, d, C-4''-OCH_3), 3.92 (6H, d, C-3'' and 5''- OCH_3), 6.76 (2H, s, C-2-NH_2), 7.21 (1H, d, *J* = 4 Hz, C-4'-H), 7.48 (2H, s, C-2'' and 6''-H), 7.67 (1H, s, C-5-H), 8.02 (1H, d, *J* = 4.2 Hz, C-3'-H).
4i	IR (ν_{max}, cm^{-1}): 3395 (NH_2), 1636 (C=N), 1602 (C=C), 1227 (C-O-C), 771 (C-S). ^1H NMR ($CDCl_3$, 400 MHz, δ, ppm): 1.38 (3H, t, C-4''-CH_3), 4.12 (2H, d, C- 4''- OCH_2), 6.69 (2H, s, C-2-NH_2), 7.05 (2H, d, *J* = 8.2 Hz, C-3'' and 5'-H), 7.25 (1H, d, *J* = 4 Hz, C-4'-H), 7.64 (1H, s, C-5-H), 7.99 (1H, d, *J* = 4.2 Hz, C-3'-H), 8.16 (2H, d, *J* = 8.6 Hz, C-2'' and 6''-H).

The remaining compounds exhibited less activity when compared with the above mentioned compounds at various concentration levels. Podophyllotoxin was used as a standard drug for BSLT assay method. By comparing the results; compounds **4a**, **4f** and **4g** found to be the best among all the tested compounds. The results and complete data of test present in Table 4.

The potency of the pyrimidine derivatives was estimated by ED_{50} values. Few of the pyrimidine derivatives showed good percentage inhibition but their ED_{50} values were more. Hence they were less potent among the tested compounds with respect to ED_{50} values.

Table 4. Cytotoxic activity of pyrimidine derivatives using Brine shrimp lethality test (DMSO was used as a solvent).

Compound No	Compounds	ED_{50}, µg/mL
4a	Phenyl	4.13
4b	3''-bromo phenyl	49.47
4c	4''-chloro phenyl	4.97
4d	2,4''-dichloro phenyl	43.52
4e	4''-nitro phenyl	45.39
4f	4''-chloro-3''-nitro phenyl	4.13
4g	4''-methyl phenyl	3.61
4h	3'',4'',5''-trimethoxyphenyl	159.98
4i	4''-ethoxyphenyl	36.61
Standard	Podophyllotoxin	3.69

3.2. Antioxidant activity

The i*n vitro* antioxidant activity and scavenging effects of the 2,4,6-trisubstituted pyrimidine derivatives (**4a-4i**), were evaluated by using different reactive species assay containing NBT-superoxide free-radical scavenging activity. The potency of the pyrimidine derivatives was estimated by IC_{50} values. The IC_{50} values of pyrimidine derivatives synthesized in the present study were given in Table 5 and Figure 1.

Table 5. Percentage inhibition of superoxide radicals using NBT-riboflavin photo reduction method.

Compound No	Quantity (µg/mL)			
	25 µg/mL	50 µg/mL	100 µg/mL	IC_{50} µg/mL
4a	20.14	27.40	36.81	>100
4b	17.77	38.60	13.64	>100
4c	28.41	30.69	30.98	68.92
4d	18.54	16.12	12.36	53.10
4e	26.01	29.88	57.63	82.54
4f	17.22	15.59	41.39	79.43
4g	21.60	26.28	35.20	>100
4h	15.03	12.14	7.510	79.40
4i	18.80	19.50	24.37	87.30
Gallic acid	31.21 (0.25 µg/mL)	40 (0.5 µg/mL)	59.83 (0.75 µg/mL)	0.61

3.3. NBT-superoxide radical scavenging activity

All the 2,4,6-tri substituted pyrimidine derivatives (**4a-i**), were found to scavenge the superoxides generated by photo reduction of riboflavin. Among them, compounds **4c**, **4d**, **4f** and **4h** showed a dose dependent inhibition of superoxide radicals at concentrations of 25, 50 and 100 µg/mL. The remaining compounds exhibited less activity when compared to the above compounds at similar concentration levels and presented in Table 5 and Figure 1.

Gallic acid, the known antioxidant was employed in the study as a standard drug for comparing the results, at concentrations of 0.25, 0.50 and 0.75 µg/mL; compound **4c** appears to be the best among all the tested compounds. Few of the pyrimidine derivatives showed good percentage inhibition

but their IC_{50} values were more. Hence they were less potent among the tested compounds with respect to IC_{50} values.

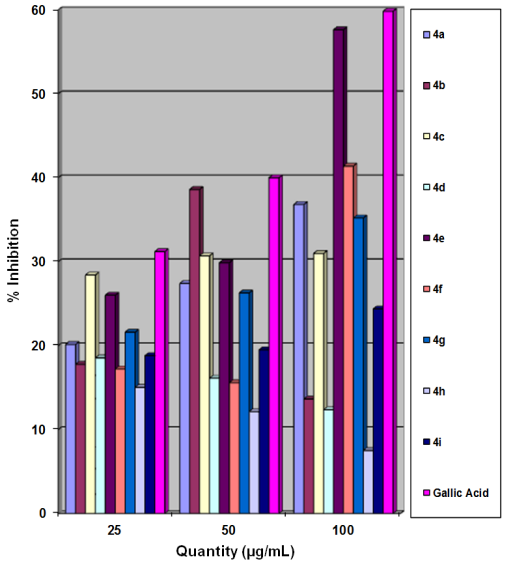

Figure 1. Percentage inhibition of superoxide radicals using NBT-riboflavin photo reduction method.

4. Conclusion

All the synthesized nine compounds (**4a-i**) were purified by recrystallization or by column chromatography. The identification of compounds was established by single spot TLC, melting point and by spectral analysis involving IR, ^{1}H NMR, ^{13}C NMR, Mass and elemental analysis. Since pyrimidines were widely reported to possess cytotoxic activity and antioxidant activities etc. All the pyrimidine derivatives were evaluated for the above mentioned activities.

From the BSLT bioassay it was proven that most of the pyrimidine derivatives are potent and possessing cytotoxic activity. And the compounds also exhibited promising antioxidant activity.

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References

[1]. Kidwai, M.; Saxena, S.; Rastogi, S.; Venkataramanan, R. *Anti-Infect. Agents Med Chem.* **2003**, *2(4)*, 269-286.
[2]. Shah, V. H.; Trivedi, A. R.; Dodiya, D. K.; Ravat, N. R. *Arkivoc* **2008**, *11*, 131-141.
[3]. Kamal, A.; Reddy, K. L.; Devaiah, V.; Shankaraiah, N.; Kumar, M. S.; Reddy, G. S. K. *Lett. Drug Des. Discovery* **2005**, *2*, 55-61.
[4]. Huang, J.; Li, H.; Li, J.; Jiang, H.; Zhu, J.; Chen, T.; Liu, J. *Molecules* **2009**, *14*, 785-797.
[5]. Kau, B.; Pathak, P.; Kaur, R. *Arkivoc* **2006**, *6*, 160-171.
[6]. Narule, M. N.; Meshram, J. S. *Int. J. Chem. Sci.* **2007**, *5(1)*, 310-318.
[7]. Amir, M.; Aggrawal, R.; Javed, S. A. *Orient .J Chem.* **2004**, *20(3)*, 477-480.
[8]. Singh, P.; Kaur, J.; Paul, K. *Indian J. Chem.* **2008**, *47B*, 291-296.
[9]. Sondhi, S. M.; Jain, S.; Dwivedi, A. D.; Shukla, R.; Raghubir, R. *Indian J. Chem.* **2008**, *47B*, 136-143.
[10]. Desai. K. R.; Chikhalia, K. H.; Patel, R. B.; Desai, P. S. *Indian J. Chem.* **2006**, *45B*, 773-778.
[11]. Sondhi, S. M.; Dinodia, M.; Rani, R.; Shukla, R.; Raghubir, R. *Indian. J. Chem.* **2009**, *49B*, 273-281.
[12]. Brown, D. J. *Rev. Pure Appl. Chem.* **1953**, *3*, 115-116.
[13]. Brown, D. J. The pyridines, A. Weissberger, Interscience, New York, 1962, pp. 183-210.
[14]. Reddy, C. S.; Nagaraj, A. *J. Heterocyclic. Chem*, **2007**, *44(5)*, 1181-1185.
[15]. Suryawanshi, S. N.; Bhat, B. A.; Susmita, P.; Naveen, C.; Suman, G. *Eur. J. Med. Chem.* **2007**, *42*, 1211-1217.
[16]. Sunduru, N.; Agarwal, A.; Katiyar, S. B.; Nishi; Goyal, N.; Gupta, S.; Chauhan, P. M. S. *Bioorg. Med. Chem*. **2006**, *14*, 7706-7715.
[17]. Akbar, M.; Naser, F.; Golnar, K.; Neda, F. *Synth. React. Inorg., Met.-Org., Nano-Met. Chem* **2007**, *37*, 279-282.
[18]. Shujang, T.; Fung, F.; Chunbao, M.; Hong, J.; Youjian, F.; Daqing S.; Xiangshan, W. *Tetrahedron Lett.* **2003**, *44*, 6153-6156.
[19]. Meyer, B. N.; Ferringi, N. R.; Putnam, J. E.; Jacobsen, L. B.; Nicholas, D.; McLaughlin, J. L. *Planta Med.* **1982**, *45,* 31-34.
[20]. Michael, A. S., Thompson, C. G.; Abramovitz, M. *Science* **1956**, *123*, 464-464.
[21]. Vanhaeke, P.; Persoone, G.; Claus, C.; Sorgeloos, P. *Ecotoxicol. Environ. Saf.* **1981**, *5*, 382-387.
[22]. Harwing, J.; Scott, P. *Appl. Microbiol*. **1971**, *21*, 1011-1016.
[23]. McLaughlin, J. L.; Chang, C. J.; Smith, D. L. *American Chemical Society Symp. Series 534, Am. Chem. Soc*. Washington, D. C, 1993, 112-137.
[24]. Sleet, R. B.; Brendel, K. *Ecotoxicol. Environ. Saf.* **1983**, *7*, 435-446.
[25]. Lamaison, J. L.; Ptitjean-Freytet, C.; Carnet, A. *Pharm. Acta Helv*. **1991**, *66*, 185-188.