Anti-bacterial and Anti-leishmanial Studies of 4, 6-diarylpyrimidin-2-amines

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Summary: Seven new chalcones along with nine already reported ones were synthesized from aryl aldehydes and substituted acetophenones by Claisen-Schmidt condensation. Each chalcone was treated with guanidine hydrochloride followed by oxidation with H_2O_2 which yielded substituted 4, 6-diarylpyrimidin-2-amines with good to excellent yields. The titled compounds were characterized by spectroscopic techniques (NMR, IR, MS) and elemental analysis. All the compounds were screened for the first time for anti-bacterial and anti-leishmanial activities which exhibited marked biological activity. Compound **4** showed significant activity against *E. coli* and *S. aureus*. Compounds **7**, **8**, **10**, **12**, **15** and **16** exhibited IC_{50} values comparable to the standard drug Amphotericin B in anti-leishmanial studies.

Introduction

Chalcones (1,3-diaryl-2-propen-1-ones) are widely used as intermediates in synthesis of heterocyclic compounds [1-5]. They are generally synthesized by the Claisen-Schmidt condensation of aryl aldehydes and substituted acetophenones using bases, such as hydroxides of sodium, potassium, barium; aluminium oxide, hydrotalcite [6], zeolite [7], potassium phosphate [8], hexadecyltri-methylammonium bromide [9] etc.

Pyrimidine scaffold being an integral part of DNA and RNA plays a vital role in several biological processes and have considerable chemical and pharmacological importance. The pyrimidine ring system is found in antibiotic, antibacterial, cardiovascular as well as agrochemical and veterinary products [10]. Recent investigations reveal that pyrimidine analogs act as anticancer [11] antifolate [12], antiviral [13], antimycobacterial [14], anti-inflammatory, antiallergic [15], anti-HIV [16], anti-HCV (Hepatitis C Virus) [17] and analgesic [18] agents.

Several species of *Leishmania* cause an infectious disease leishmaniasis [19]. The World Health Organization has identified the various forms of leishmaniasis, among some other infectious diseases, as major and increasing public health problem, particularly in developing countries [20-22] Unfortunately, efforts directed towards the discovery of new drugs and vaccines against leishmaniasis are lethargic [23, 24] Thus, the need for the development of new, broad spectrum, cost-effective and safe drugs for the treatment of leishmaniasis is very important.

Keeping in view the threat of leishmaniasis to the world, and reported antibacterial activities of pyrimidines, we evaluated the synthesized compounds as anti-leishmanial as well as anti-bacterial agents.

2-Aminopyrimidines and their related compounds, in particular, are known as potent drugs including trimethoprim and daraprim, Gleevec, sulphadiazine and inhibitors of Adenosine, CDK, CB2, VEGFR [25] and several others.

Results and Discussion

Each chalcone was synthesized by treating aromatic aldehyde and substituted acetophenone in MeOH by gradual addition of 10% NaOH solution at ambient temperature. The chalcones synthesized were washed with cold MeOH followed by cold distilled water which gave pure compounds.

To get title compounds, each chalcone was refluxed together with guanidine hydrochloride in the presence of 50% aqueous KOH solution and portion wise addition of 30% aqueous H_2O_2 solution as shown in Scheme I [26].

The crude product was recrystallised with distilled water. The pure compound was dried and kept in vacuum desiccator over P_2O_5/KOH. This method proved to be efficient and gave up to 70% yield. All the compounds were characterized by IR, MS and ^{1}HNMR spectral techniques.

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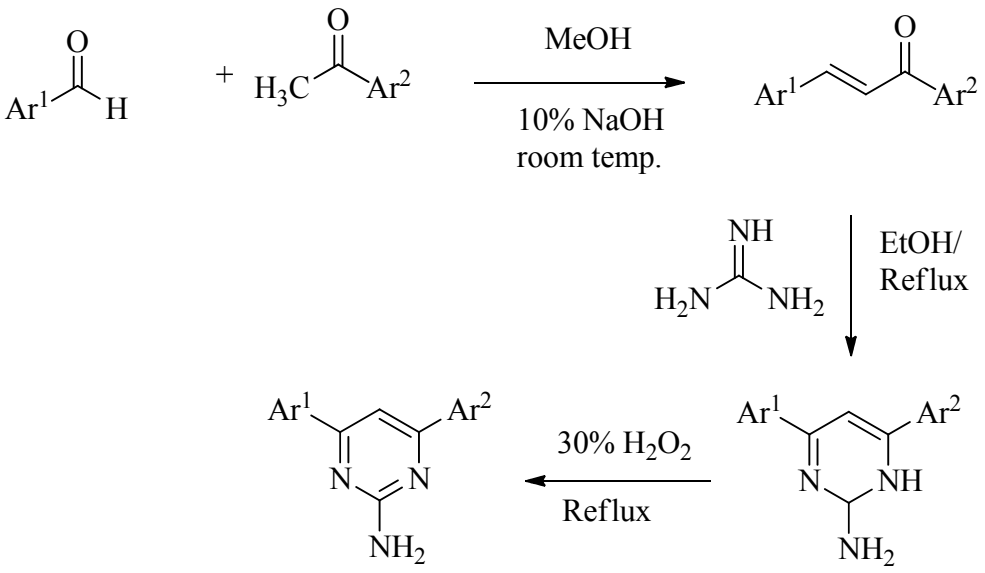

1-16

Compounds	Ar^1	Ar^2	Compounds	Ar^1	Ar^2
1	4-chlorophenyl	3, 4-dichlorophenyl	9	4-chlorophenyl [8, 28, 29]	4-chlorophenyl
2	4-chlorophenyl	3,4-dimethoxyphenyl	10	4-chlorophenyl [30]	4-bromophenyl
3	4-chlorophenyl	2,3,4-trichlorophenyl	11	4-chlorophenyl [28, 31]	Phenyl
4	4-chlorophenyl	2-4-dichlorophenyl	12	4-chlorophenyl [32]	furan-2-yl
5	4-fluorophenyl	naphthalen-2-yl	13	4-chlorophenyl [33]	naphthalen-2-yl
6	2,4-dichlorophenyl	3,4-dichlorophenyl	14	4-chlorophenyl [34]	1H-pyrrol-2-yl
7	2,4-dichlorophenyl	3,4-dimethoxyphenyl	15	4-fluorophenyl [35]	Phenyl
8	4-fluorophenyl [27]	furan-2-yl	16	2,4-dichlorophenyl [36]	furan-2-yl

Scheme 1

The IR spectra showed characteristic peaks of NH_2 at 3450-3425, 3285-3200 and 1655-1630 cm^{-1}. A pyrimidine skeletal peak at ~1550 cm^{-1} appeared for all the pyrimidines synthesized. In ^1H NMR spectra, NH_2 protons gave a broad peak in 4.98-5.17 ppm range. Another characteristic peak appeared at almost 7.3 ppm as a singlet was assigned to H-5. However, in case of some compounds, this peak was not individually differentiated. It is observed that the aromatic protons of Ar^1 at positions 2 and 6 appeared at higher values than those at positions 3 and 5 where present. Each signal is assigned in the Experimental. Some 4, 6-diarylpyrimidine-2-amines have been reported by Chinese researchers using different synthetic route which included two compounds **9** and **15** [28]. Compounds **9** and **11** were also synthesized by Pore et al (2006) using K_3PO_4 as an efficient catalyst [8]. Crystal structure of compound **9** was reported by Wang et al in 2005 [29].

The compounds **1-16** were subjected to anti-bacterial and anti-leishmanial studies. Some compounds exhibited moderate activity against *B. subtilis, E. coli* and *S. aureus*. Particularly, compound **11** showed significant activity against *E. coli* and *S. aureus,* and to less extent, against *B. subtilis.* The results of anti-bacterial activity are summarized in Table-1.

Table-1: Anti-bacterial activity (Zone of inhibition in mm).

Compounds	*B. Subtilis*	*E. Coli*	*S. Aureus*	Compounds	*B. Subtilis*	*E. Coli*	*S. Aureus*
1	**13**	**12**	**14**	**9**	**15**	**13**	**15**
2	**-**	**15**	**-**	**10**	**12**	**13**	**-**
3	**-**	**12**	**-**	**11**	**21**	**25**	**27**
4	**-**	**16**	**-**	**12**	**13**	**11**	**-**
5	**-**	**13**	**14**	**13**	**15**	**12**	**15**
6	**-**	**16**	**13**	**14**	**13**	**15**	**-**
7	**13**	**17**	**11**	**15**	**12**	**15**	**-**
8	**-**	**17**	**-**	**16**	**-**	**-**	**-**
Std. Drug Imipenem	**30**	**35**	**30**	**Std. Drug Imipenem**	**30**	**35**	**30**

All the compounds were also tested for their anti-leishmanial activity using *Leishmania major* promastigotes as parasites for *in vitro* screening. The results are summarized in Table-2.

It was observed that compounds **7**, **8**, **10, 12**, **15** and **16** exhibited IC_{50} values in the range close to the standard drug Amphotericin B. It may be considered that activity of compounds **8**, **12** and **16** is due to furan ring present in the molecule.

Table-2: Anti-leishmanial activity of the synthesized compounds (IC_{50})

Compounds	IC_{50} µg/mL	Compounds	IC_{50} µg/mL
1	**0.80 ± 0.07**	**9**	**0.85 ± 0.05**
2	**0.76 ± 0.26**	**10**	**0.68 ± 0.06**
3	**0.78 ± 0.12**	**11**	**0.88 ± 0.40**
4	**0.79 ± 0.12**	**12**	**0.69 ± 0.34**
5	**0.81 ± 0.45**	**13**	**0.79 ± 0.03**
6	**0.84 ± 0.35**	**14**	**0.93 ± 0.98**
7	**0.67 ± 0.23**	**15**	**0.68 ± 0.56**
8	**0.67 ± 0.05**	**16**	**0.69 ± 0.17**
Standard Drug (Amphotericin B)	**0.56 ± 0.20**	**DMSO as –ve control**	**0.99 ± 0.58**

Experimental

All 1, 3-diaryl prop-2-en-1-ones and their corresponding pyrimidines were prepared according to the reported procedures [37, 26]. The progress of the reaction was monitored by TLC. All the products were purified through recrystalization and purity of the compounds was checked by thin layer chromatography (TLC) performed on silica gel G coated plate of 0.25 mm thickness. Melting points are uncorrected and were measured on a standard GallenKamp apparatus. The ^{1}HNMR spectra were recorded at 298 K on a Bruker/ XWIN-NMR (400 MHz) spectrometer with TMS as an internal reference. MS data were collected on a JEOL MSRoute mass spectrometer. Elemental analysis was carried out using a Perkin Elmer 2400-CHN Analyzer. IR spectra were recorded on Perkin Elmer FTIR 750 spectrophotometer.

Typical Experimental Procedure for Preparation of 4,6-diarylpyrimidin-2-amines

4-(4-Chlorophenyl)-6-(3,4-dichlorophenyl)-pyrimidin-2-amine **(1)**

3-(4-Chlorophenyl)-1-(3,4-dichlorophenyl)-prop-2-en-1-one (3.17 g, 9.08 mmol), guanidine hydrochloride (1.3 g, 1.5 mmol), ethanol (20mL) and 50% aqueous KOH solution (4 mL,) were mixed together, then heated up and stirred at reflux temperature for 1 hr. Under the same conditions, 30% aqueous H_2O_2 (3.1 mL, 27.3 mmol) was added to the above mixture in small portions over a period of I hr. The ethanol was removed under reduced pressure in a rotary evaporator and distilled water (~20mL) was added to the residue. The product was easily isolated as precipitates and was washed repeatedly with pure water. The crude product was recrystallized from ethanol and was dried finally in a vacuum desiccator over P_2O_5/ KOH.

Compound **1**: (48%, Cream colored solid), mp 230-232 ºC. IR (KBr) υ_{max} cm^{-1}: 3445, 3285 and 1649 (NH_2), 1568 (pyrimidine skeletal), 755 ($C_{Ar}H$); ^{1}H NMR (400 MHz, $CDCl_3$) δ: 5.14 (2H, br. s, NH_2), 7.36 (1H, s, H-5), 7.45 (2H, d, *J*=8.5 Hz, ^{Ar1}H-3+^{Ar1}H-5), 7.54 (2H, d, *J*=8.4 Hz, ^{Ar1}H-2+^{Ar1}H-6), 7.87 (1H, dd, *J*=2.0, 3.5 Hz, ^{Ar2}H-5), 7.99 (1H, d, *J*=8.9 Hz, ^{Ar2}H-6), 8.17 (1H, d, *J*=2.0 Hz, ^{Ar2}H-2). MS m/z: 349 (M^+, 100%), 351 (97%), 353 (31%). Anal. calc. for $C_{16}H_{10}Cl_3N_3$; C, 54.81; H, 2.87; N, 11.98; Found: C, 54.80; H, 2.88; N, 11.96.

4-(4-Chlorophenyl)-6-(3,4-dimethoxy-phenyl)pyrimidin-2-amine **(2)**

The experimental procedure was similar to that described for compound **1** starting from 3-(4-Chlorophenyl)-1-(3,4-dimethoxyphenyl)prop-2-en-1-one.

Compound **2**: (60%, white amorphous solid), mp 210-212 ºC. IR (KBr) υ_{max} cm^{-1}: 3445, 3285 and 1645 (NH_2), 1555 (pyrimidine skeletal), 768 ($C_{Ar}H$ ^{1}H NMR (400 MHz, $CDCl_3$) δ: 3.94 (3H, s, OCH_3), 3.99 (3H, s, OCH_3), 5.10 (2H, br. s, NH_2), 6.94 (1H, d, ^{Ar2}H-5), 7.37 (1H, s, H-5), 7.44 (2H, d, *J*=8.5 Hz, ^{Ar1}H-3 + ^{Ar1}H-5), 7.61 (1H, dd, *J*=1.4, 2.4 Hz, ^{Ar2}H-2), 7.68 (1H, d, *J*=1.7 Hz, ^{Ar2}H-6), 7.99 (2H, d, *J*=8.5 Hz, ^{Ar1}H-2 + ^{Ar1}H-6). MS m/z: 341 (M^+, 100%), 343 (32%). Anal. calc. for $C_{18}H_{16}ClN_3O_2$; C, 63.25; H, 4.72; N, 12.29; Found: C, 63.25; H, 4.73; N, 12.28.

4-(4-Chlorophenyl)-6-(2,3,4-trichlorophenyl)pyrimidin-2-amine **(3)**

The experimental procedure was similar to that described for compound **1** starting from 3-(4-Chlorophenyl)-1-(2,3, 4-trichlorophenyl)prop-2-en-1-one.

Compound **3**: (70%, Pale yellow amorphous solid), mp 281-283 ºC. IR (KBr) υ_{max} cm^{-1}: 3450, 3290 and 1655 (NH_2), 1550 (pyrimidine skeletal), 762 ($C_{Ar}H$); ^{1}H NMR (400 MHz, $CDCl_3$) δ: 5.16 (2H, br. s, NH_2), 7.42 (1H, d, *J*=4.7 Hz, ^{Ar2}H-5), 7.45 (1H, s, H-5), 7.48 (2H, d, *J*=8.4 Hz, ^{Ar1}H-3 + ^{Ar1}H-5), 7.63 (1H, d, *J*=2.1 Hz, ^{Ar2}H-6), 7.96 (2H, d, *J*=8.6 Hz, ^{Ar1}H-2 + ^{Ar1} H-6). MS m/z: 383 (M^+, 78%) 385 (100%), 386 (17%), 387 (48%). Anal. calc. for $C_{16}H_9Cl_4N_3$; C, 49.90; H, 2.36; N, 10.91; Found: C, 49.90; H, 2.36; N, 10.90.

4-(4-Chlorophenyl)-6-(2,4-dichlorophenyl)pyrimidin-2-amine **(4)**

The experimental procedure was similar to that described for compound **1** starting from 3-(4-

chlorophenyl)-1-(2,4-dichlorophenyl)prop-2-en-1-one.

Compound **4**: (50%, white crystalline solid), mp 177-179 ºC. IR (KBr) υ_{max} cm^{-1}: 3435, 3275 and 1635 (NH_2), 1550 (pyrimidine skeletal), 760 ($C_{Ar}H$); ^{1}H NMR (400 MHz, $CDCl_3$) δ: 5.14 (2H, br. s, NH_2), 7.33 (1H, s, ^{Ar2}H-3), 7.36 (1H, dd, *J*=1.8, 2.5 Hz, ^{Ar2}H-3), 7.44 (2H, d, *J*=8.2 Hz, ^{Ar1}H-3 + ^{Ar1}H-5), 7.50 (1H, d, *J*=1.9 Hz, H-5), 7.97 (2H, d, *J*=8.5 Hz, ^{Ar1}H-2 + ^{Ar1}H-6). MS m/z: 349 (M^+, 100%), 351 (97%), 353 (31%). Anal. calc. for $C_{16}H_{10}Cl_3N_3$; C, 54.81; H, 2.87; N, 11.98; Found: C, 54.80; H, 2.88; N, 11.96.

4-(4-Fluorophenyl)-6-(naphthalen-2-yl)pyrimidin-2-amine (**5**)

The experimental procedure was similar to that described for compound **1** starting from 3-(4-Fluorophenyl)-1-(naphthalen-2-yl)prop-2-en-1-one.

Compound **5**: (48%, white powder), mp 190-192 ºC. IR (KBr) υ_{max} cm^{-1}: 3435, 3205 and 1635 (NH_2), 1564 (pyrimidine skeletal), 755($C_{Ar}H$). ^{1}H NMR (400 MHz, $CDCl_3$) δ: 5.15 (2H, br. s, NH_2), 7.16-7.20 (2H, m, , ^{Ar1}H-3 + ^{Ar1}H-5), 7.50-7.56 (2H, m, ArH), 7.86-7.89 (1H, m, ArH), 7.93-7.97 (2H, m, ArH), 8.08-8.15 (4H, m, ArH), 8.56 (1H, s, ^{Ar2}H-1). MS m/z: 315 (M^+, 100%). Anal. calc. for $C_{20}H_{14}FN_3$; C, 76.18; H, 4.47; N, 13.33; Found: C, 76.20; H, 4.47; N, 13.32.

4-(2,4-Dichlorophenyl)-6-(3,4-dichlorophenyl)pyrimidin-2-amine (**6**)

The experimental procedure was similar to that described for compound **1** starting from 3-(2,4-Dichlorophenyl)-1-(3,4-dichlorophenyl)prop-2-en-1-one.

Compound **6**: (42%, yellow solid), mp 185-187 ˚C. ν_{max} (KBr) 3368, 3225, 1610, 1582 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 5.17 (2H, br. s, NH_2), 7.31 (1H, s, ^{Ar1}H-3), 7.36 (1H, d, *J*=8.5 Hz, ^{Ar1}H-5), 7.51 (1H, d, *J*=8.0 Hz, ^{Ar2}H-5), 7.61 (1H, s, H-5), 7.84 (1H, s, ^{Ar2}H-2), 8.16 (1H, d, *J*=5.6 Hz, ^{Ar1}H-6). MS m/z: 383 (M^+, 78%), 385 (100%), 387 (48%). Anal. calc. for $C_{16}H_9Cl_4N_3$; C, 49.90; H, 2.36; N, 10.91; Found: C, 49.92; H, 2.36; N, 10.90.

4-(2,4-Dichlorophenyl)-6-(3,4-dimethoxyphenyl)pyrimidin-2-amine) (**7**)

The experimental procedure was similar to that described for compound **1** starting from 3-(2,4-dichlorophenyl)-1-(3,4-dimethoxyphenyl)prop-2-en-1-one.

Compound 7: (52%, pale yellow solid), mp 195-197 ˚C. ν_{max} (KBr) 3378, 3300, 1590, 1575 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 3.81 (6H, s, 2xOMe), 4.98 (2H, br. s, NH_2), 6.78 (1H, m, ^{Ar2}H-5), 6.98 (1H, d, *J*=8.0 Hz, ^{Ar2}H-6), 7.08 (1H, s, ^{Ar2}H-2), 7.14 1H, d, *J*=8.0 Hz, ^{Ar1}H-5), 7.32 (1H, s, ^{Ar1}H-3), 7.40 (1H, s, H-5), 7.57 (1H, d, *J*=8.4 Hz). MS m/z: 375 (M^+, 100%), 377 (20%), 379 (64%). Anal. calc. for $C_{18}H_{15}Cl_2N_3O_2$; C, 57.46; H, 4.02; N, 11.17; Found: C, 57.47; H, 4.01; N, 11.18.

4-(4-Fluorophenyl)-6-(furan-2-yl)pyrimidin-2-amine (**8**)

The experimental procedure was similar to that described for compound **1** starting from 3-(4-Fluorophenyl)-1-(furan-2-yl)prop-2-en-1-one. We have already reported the crystal structure of the title compound [27].

Compound **8**: (58%, cream colored powder), mp 240-242 ºC. IR (KBr) υ_{max} cm^{-1}: 3425, 3255 and 1625 (NH_2), 1560 (pyrimidine skeletal), 765($C_{Ar}H$), 591(furan ring def.); ^{1}HNMR ($CDCl_3$, 400 MHz): δ 5.07(2H, br. s, NH_2), 6.55(1H, dd, *J*= 3.0, 2.0 Hz, ^{Ar2}H-4), 7.15(1H, dd, *J*=3.5, 3.2 Hz, ^{Ar2}H-3), 7.36(1H, s, H-5) 7.57(1H, dd, *J*=3.0, 2.0 Hz, ^{Ar2}H-5), 8.02-8.06(4H, m, 4×^{Ar1}H); MS m/z: 255 (M^+, 100%). Anal. calc. for $C_{14}H_9FN_3O$; C, 54.92; H, 2.96; N, 13.73; Found: C, 54.91; H, 2.96; N, 13.74.

Biological Activities

(a)Anti-bacterial Activity

All the synthesized compounds (dissolved in DMSO) were subjected to anti-bacterial screening for determining the zone of inhibition by cup diffusion method. The Petri plates were inoculated in cultures of fungus on potato dextrose agar medium. Plates were incubated at 37 ˚C for 24 hours for bacteria. After inoculation, the diameter of clear zone of inhibition surrounding the sample was taken as a measure of the inhibitory power of the sample against the particular test organism [38].

(b)Anti-leishmanial Activity

Anti-leishmanial activity of the title compounds was carried out on the pre-established culture of L. major. Parasites were cultured in medium M199 with 10% foetal bovine serum; 25 mM of HEPES, and 0.22 mg of penicillin and

streptomycin respectively at 24 ˚C in a shaking incubator. 1 mg of each compound was dissolved in 1 mL of DMSO and as a positive control 1 mg of Amphotericin B was also dissolved in 1 mL of DMSO. Parasites at log phase were centrifuged at 3000 rpm for 3 minutes. Parasites were diluted in fresh culture medium to a final density of $2x10^6$ cells/mL. In 96-well plates, 180 µl of medium was added in different wells. 20 µl of the experimental compound was added in medium and serially diluted. 100 µl of parasite culture was added in all wells. In negative controls, DMSO was serially diluted in medium while the positive control contained varying concentrations of standard anti-leishmanial compound i.e. Amphotericin B. The plates were incubated for 72 hours at 24 ˚C. The culture was examined microscopically on an improved Neubauer counting chamber and IC_{50} values of compounds possessing anti-leishmanial activity were calculated. All the assays were run in duplicate [39].

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References

1. S. R. Sandler and W. Karo, "Organic Functional Group Preparations," p. 372. Academic, New York, 1972.
2. A. Levai, *Arkivoc* **part (ix)**, 344 (2005).
3. B. C. Ranu, S. S. Dey, and S. Samanta, *Arkivoc* **part (iii)**, 44 (2005).
4. D. N. Dhar, "The chemistry of chalcones and related compounds." Wiley-Interscience, New York, N.Y, 1981.
5. J. R. Dimmock, D. W. Elias, M. A. Beazely, N. M. Kandepu, and . *Curr. Med. Chem.* **6**, 1125–1149 (1999).
6. M. J. Liment, A. Corma, S. Iborra, and J. Primo, *J. Catal.* **151**, 60 (1995).
7. A. Guida, D. T. Lhouty, F., P. Figueras, and P. Geneste, *Appl Catal., A* **164**, 251 (1997).
8. D. M. Pore, U. V. Desai, T. S. Thopate, and P. P. Wadgoankar, *Russian J. Org. Chem.* **43**, 1088 (2007).
9. T. Hussain, H. L. Siddiqui, M. Zia-ur-Rehman, M. M. Yasinzai, and M. Parvez, *Eur. J. Med. Chem.* **44**, 4654–4660 (2009).
10. H. Parveen, F. Hayat, A. Salahuddin, and A. Azam, *Eur. J. Med. Chem.* **45**, 3497 (2010).
11. Y. Miyazaki, S. Matsunaga, J. Tang, Y. Maeda, M. Nakano, R. J. Philippe, M. Shibahara, W. Liu, H. Sato, L. Wang, and R. T. Notle, *Bioorg. Med. Chem. Lett.* **15**, 2203 (2005).
12. A. Gangjee, E. Elzein, S. F. Queener, and J. J. McGuire, *J. Med. Chem.* **41**, 1409 (1998).
13. M. S. Rao, N. Ehso, C. Sergeant, and R. Dembinski, *J. Org. Chem.* **68**, 6788 (2003).
14. L. Ballell, A. F. Robert, G. A. C. Chung, and R. J. Young, *Bioorg. Med. Chem. Lett.* **17**, 1736 (2007).
15. G. A. Breault and J. E. Pease, *Chem. Abstr.* **132**, 194385 (2000).
16. V. Malik, P. Singh, and S. Kumar, *Tetrahedron* **62**, 5944 (2006).
17. V. N. S. V. Chamakura, K. S. Ramasamy, J. L. Girardet, E. Gunic, V. Lai, W. Zhong, H. An, and Z. Hong, *Bioorg. Med. Chem. Lett.* **35**, 25 (2007).
18. S. M. Sondhi, N. Singh, M. Johar, and A. Kumar, *Bioorg. Med. Chem.* **13**, 6158 (2005).
19. E. C. Grisard, M. Steindel, J. J. Shaw, E. A. Y. Yshikawa, C. J. Carvalho-Pinto, I. Eger-Mangrich, H. K. Toma, A. J. Romanha, and D. A. Campbell, *Acta Trop.* **74**, 89 (2000).
20. M. I. Choudhary, A. Adhikari, Samreen, Atta-Ur-Rahman, *Journal of the Chemical Society of Pakistan*, **32**, 799 (2010).
21. F. Modabber, *in* "Tropical disease research progress 1991-92", p. 77. World Health Organization, Geneva, Switzerland, 1993.
22. F. R. Opperdoes, *J. Bioenerg. Biomembr.* **26**, 145 (1994).
23. S. L. Croft, *Parasitology* **114**, 83 (1997).
24. H. D. Engers, R. Bergquist, and F. Moddaber, *Dev. Biol. Stand.* **87**, 73 (1996).
25. M. D. Hill and M. M., *Chem. Eur. J.* **14**, 6836 (2008).
26. L. Varga, T. Nagy, I. Kovesdi, J. Benet-Buchholz, G. Dorman, L. Urge, and F. Darvas, *Tetrahedron* **59**, 655 (2003).
27. M. H. Bukhari, H. L. Siddiqui, M. A. Chaudhary, T. Hussain, and M. Parvez, *Acta Cryst E* **64**, o963 (2008).
28. Q. Zhuang, Han, H. X., Wang, S., Tu, S., Rong, L., *Synthetic Communications* **39**, 516 (2009).
29. S. Wang, C. Meades, G. Wood, A. Osnowski, S. Anderson, R. Yuill, M. Thomas, M. Mezna, W. Jackson, C. Midgley, G. Griffiths, I. Fleming, S. Green, I. McNae, S.-Y. Wu, C. McInnes, D.

Zheleva, M. D. Walkinshaw, and P. M. Fischer, *J. Med. Chem.* **47**, 1662 (2004).
30. M. Ghannoum, K. Abu Elteen, and N. R. El-Rayyes, *Microbios* **60**, 23 (1989).
31. M. Gopalakrishnan, J. Thanusu, and V. Kanagarajan, *Chem. Hetero. Comp.* **45**, 1539 (2009).
32. V. Desai, C. M. Desai, D. Patel, and A. G. Mehta, *J. Inst.Chem.* **77**, 104 (2005).
33. H. S. Patel, V. K. Patel, and B. C. Dixit, *Oriental J. Chem.* **17**, 411 (2001).
34. M. N. Narule and J. S. Meshram, *Oriental J. Chem.* **22**, 387 (2006).
35. V. Kanagarajan, J. Thanusu, and M. Gopalakrishnan, *Eur. J. Med. Chem.* **45**, 1583 (2010).
36. Y. R. Prasad, P. V. Rao, P. R. Kumar, and S. H. Latha, *Int. J. Chem. Sci.* **6**, 333 (2008).
37. B. S. Furniss, A. J. Hannaford, P. W. G. Smith, and A. R. Tatchell, *in* "Vogel's Textbook of Practical Organic Chemistry", p. 1034. Longman, New York, 1989.
38. N. S. C. Gaulejac, Y. Glories, and N. Vivas, *Food Res. Int.* **32**, 327 (1999).
39. S. A. Ali, J. Iqbal, and M. M. Yasinzai, *Methods Cell Sci.* **19**, 107 (1997).