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SYNTHESIS OF BIS-AMIDE AND HYDRAZIDE CONTAINING DERIVATIVES OF MALONIC ACID AND THIOPHENOL ADDUCTS OF ACID HYDRAZONES DERIVED FROM 2, 5-DICHLOROANILIDO ACETOHYDRAZIDE

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ABSTRACT

We synthesized a new series of bis-amide and hydrazide-containing derivatives of malonic acid and thiophenol adducts of acid hydrazones by the reaction of 2,5-dichloroanilido acetohydrazide with various carbonyl compounds in 39 to 72% yield. Newly synthesized compounds have been tested for their anti-bacterial activity against gram positive bacteria *S.albus*, *S.aureus* and gram negative bacteria *E.coli* and *Pseudomonas piosineus*. The compound (**3, 7, 14, 16**) shown significant activities and compound (**5, 8, 13, 17**) have shown moderate activity. The same compounds were tested for their anti-fungal activity against *Candida albicans, Aspergillus niger and Alternaria alternata* at concentration of 30 mg/mL using Savored dextrose agar media. The compound (**4, 9, 11, 12**) shown significant activities and compound (**2, 6, 10, 15**) have shown moderate activity against *Candida albicans* and *Aspergillus niger*. All the other compounds did not show significant activity against the fungi at the concentration used.

Key words: Malonic acid, Bis-amides, Acid hydrazides, Hydrazone-thiophenol adducts.

INTRODUCTION

Acid hydrazones and their condensation products possessing an azometine-NHN= CH- proton constitute an important class of compounds for new drug development. In the past several years, numerous compounds with diverse structural features have been reported. Therefore, many researchers have synthesized these compounds as target structures and evaluated their biological activities. Hydrazides, hydrazones and their adducts have displayed diverse range of biological properties such as potential biological activities[1-6], anti-viral[7,8], anti-tuberculosis[9,10], anti-tumor[11,18], anti-fungal[19,20], anti-convulsant[21], anti-helmintic[22],

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anti-malerial[23], anti- Inflammatory[24], anti-cancer[25,26], anti-proliferative[27-29], anti-oxidant[30], agricultural agents[31]. Therapeutic protocols for the treatment of HIV infection are mainly based on the combined use of reverse transcriptase, protease, and more recently, of cell fusion and entry inhibitors. Although drugs targeting reverse transcriptase and protease are in wide use and have shown effectiveness, the rapid emergence of resistant variants, often cross-resistant to the members of a given class, limits the efficacy of existing antiretroviral drugs. Therefore, it is critical to develop new agents directed against alternate sites in the viral life cycle. Moreover, many selectively chloro-substituted organic compounds show peculiar pharmacological and agrochemical properties. The work reported herein was aimed at the preparation of some new thiophenol adducts of acid hydrazones with anticipated biological activities.

EXPERIMENTAL

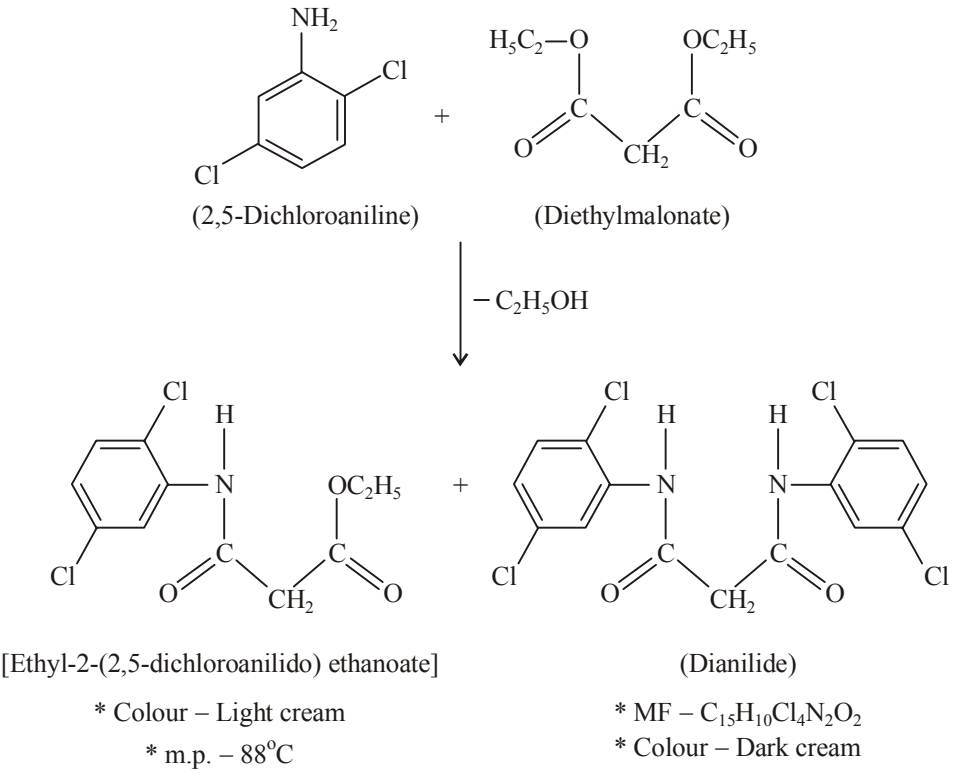
Materials and methods

Anhydrous solvents and all reagents were purchased from, Sigma-Aldrich, B.D.H., Excel-R, Extra pure E. Merk quality, Acros or Carlo Erba. Reactions involving air- or moisture-sensitive compounds were performed under a nitrogen atmosphere using oven-dried glassware and syringes to transfer solutions. Melting points (m.p.) were determined using an electrothermal melting point or a Köfler apparatus and are uncorrected. Infrared (IR) spectra were recorded as thin films or nujol mulls on KBr plates with a Perkin-Elmer-781 IR or 983-spectrophotometer and are expressed in ν (cm^{-1}). Nuclear magnetic resonance spectra (^1H NMR) was determined in DMSO and recorded on a Varian XL-200 (200 MHz) or a Varian VXR-300 (300 MHz). Chemical shifts (δ scale) are reported in parts per million (ppm) downfield from tetramethylsilane (TMS) used as internal standard. Splitting patterns are designated as follows: s, singlet; d, doublet; t, triplet; q, quadruplet; m, multiplet; brs, broad singlet; dd, double doublet. The assignment of exchangeable protons (-OH and -NH) was confirmed by addition of D_2O. Analytical thin-layer chromatography (TLC) was carried out on Merck silica gel, F-254 plates. For flash chromatography Merck Silica gel-60 was used as stationary phase with a particle size 0.040-0.063 mm (230-400 mesh ASTM). Elemental analyses were performed on a Perkin-Elmer-2400 spectrometer, and were within ± 0.7% of the theoretical values.

General procedure for the synthesis of ethyl-2-(2, 5-dichloroanilido) ethanoate (1)

Diethylmalonate (20 mL) and 2, 5-dichloroaniline (10 mL) was refluxed for forty five minutes in a round bottomed flask fitted with an air condenser of such a length (14") that ethanol formed escaped and diethylmalonate flowed back into the flask. Contents were cooled, ethanol (30 mL) was added, when malon-2, 5-dichlorodianilide separated out. It was

filtered under suction. The filtrate was poured on to crushed ice (Ca 160 g) and stirred when ethyl-2-(2, 5-dichloroanilido) ethanoate precipitated as green mass. On recrystallization from aqueous ethanol (50%), ester was obtained as white crystals. Yield: 82%, m. p.: 88°C, M. W.: 276. Anal. Calculation for $C_{11}H_{11}N_1O_3Cl_2$: Found: C 47.7, H 4.0, O 17.2, N 5.1, Cl 25.4, Calcd. C 47.8, H 4.0, O 17.4, N 5.1, Cl 25.7. IR [KBr] ν_{max} cm^{-1} : 1665-1660 [C=O diketone], 1290 [-O- Ester], 760-755 [2,5-disubstituted benzene], 1090 [C-Cl Stretching], 1590, 1520, 1440 [C=C ring stretching], 3150 [N-H Stretching], 3040 [C-H aromatic], 1330-1322 [C-H Stretching]. PMR (DMSO): δ 4.42 (2H, s, CO-CH_2-CO), 4.0 (2H, s, NH_2), 7.4-8.6 (3H, m, Ar-H), 9.2 (1H, s, CO-NH D_2O exchangeable), 10.6 [1H, s, Ar-NH D_2O exchangeable].

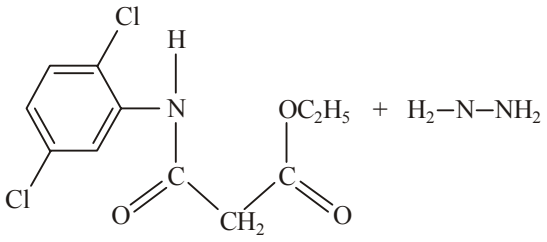


Scheme 1

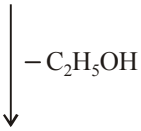
Synthesis of 2-(2, 5-dichloroanilido) acetohydrazide (2)

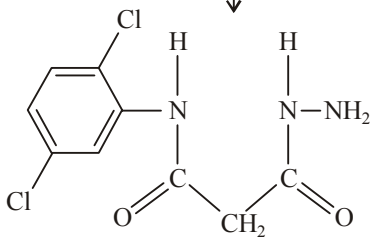
Ethyl-2-(2,5-dichloroanilido) ethanoate (9.54 gm; 0.03 mol), ethanol (10 mL) and hydrazine hydrate (15 mL; 80%) were mixed together and stirred for forty five minutes. There were evolution of heat and reaction was spontaneous after 30 minutes, 2-(2, 5-

dichloroanilido) acetohydrazide was filtered under suction and recrystallised from ethanol in silver white crystals. Yield; 82%, m.p. = 172°C, MW 262: Analytical calculation for $C_9H_9N_3O_2Cl_2$: Calculated ; N 09.04 ,C 41.32, O 10.33, Cl 15.28, Found; N 09.01, C 41.30, O 10.31, Cl 15.27; IR [KBr] ν_{max} cm^{-1}: 3160 [N-H Stretching], 3048 [C-H aromatic], 1660 [C=O diketone], 1430 [C-Cl aromatic], 1595, 1520, 1445 [C=C ring stretching]. NMR Spectra (δ DMSO): 2.44 (2H, s, CH_2), 3.2 (3H, s, CH_3), 4.22-4.32 (1H, t, N-H), 7.2-7.6 (3H, m, ArH).



Ethyl-2-(2,5-dichloroanilido) ethanoate





2-(2,5-dichloroanilido) acetohydrazide

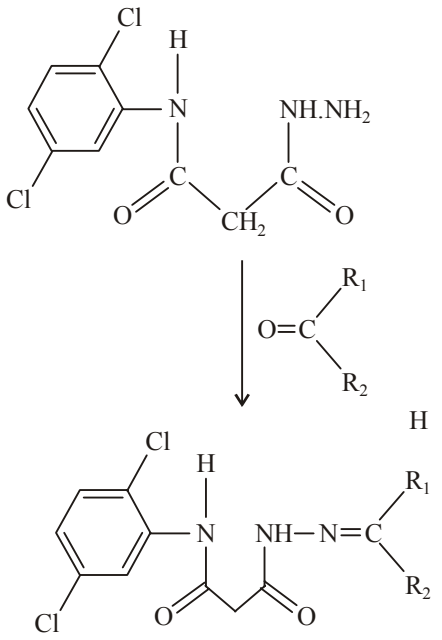
* Colour – Pale yellow

Scheme 2

Synthesis of new acid hydrazones (3)

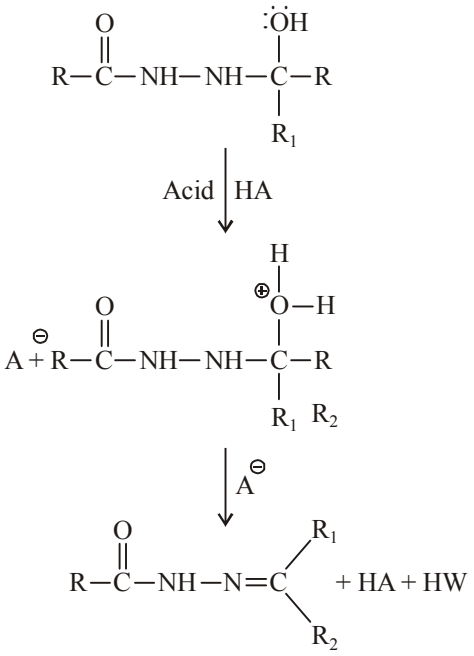
2-(2,5-dichloroanilido) acetohydrazide (.001 mol) and (.001 mol) of aromatic aldehyde or ketone (carbonyl compound) dissolve in absolute alcohol and added 2-drops of conc. H_2SO_4 and stirred for 20-25 minutes. It was filtered under suction and recrystallised from hot ethanol. Synthetic strategy has been out lined in **Scheme 1, 2 & 3**. Mechanism for the formation of acid hydrazones is given in chart-I. IR absorption band (cm^{-1}): 3150 (N-H stretching), 2960-2970 (C–H aliphatic), 1665-1660 (C=O Ketone), 785-780 (C-Cl

Stretching), 760-755 (2, 5-disubstituted benzene), NMR spectra (δ DMSO), 2.25 (2 H, s, CH_2), 4.21 (1 H, s, NH), 6.95–7.2 (10 H, m, ArH).



(Acid hydrazones)

Scheme 3



(Acid hydrazone)

Mechanism of new acid hydrazones

Chart 1

Biological evaluation

Anti-bacterial activity

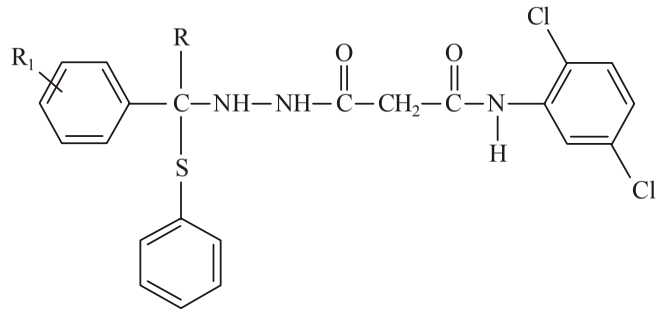
Newly synthesized thiophenol adducts of acid hydrazones were screened for their anti-bacterial activity against the gram positive bacteria *S. albus, S. aureus* and gram negative bacteria *E. Coli* and *Pseudomonas piosineus* by agar plate disc diffusion method at 30 µg/mL concentration. *Ampicillin* and *tetracycline* were used as a reference compounds. The compound (**3, 7, 14, 16**) shown significant activities and compound (**5, 8, 13, 17**) have shown moderate activity.

Anti-fungal activity

The same compounds were tested for their antifungal activity against *Candida albicans, Aspergillus Niger* and *Alternaria alternata* at concentration of 30 mg/mL using Savored dextrose agar media. The compound (**4, 9, 11, 12**) shown significant activities and

compound (**2, 6, 10, 15**) have shown moderate activity against *Candida albicans* and *Aspergillus niger*. All the other compounds did not show significant activity against the fungi at the concentration used.

Table 1: Reaction conditions for the formation of thiophenol adducts of acid hydrazones



(i) Quantity of acid hydrazone = 0.001 mol.

(ii) Quantity of thiophenol = 0.110 g (0.001 mol)

(iii) Hours of heating = 12 hours.

Comp.	Acid hydrazones	Quantity of acid hydrazone (g)	Adducts		m.p. (°C)	Yield (%)	Formula Weight	Molecular formula	Colour
			R_1	R_2					
1.	Benzaldehyde-2-(2, 5-dichloroanilido) acetohydrazone	0.539	H	Ph	272	69	539	$C_{28}H_{26}O_2N_3Cl_2S_1$	White
2.	Vanilline-2-(2,5-dichloroanilido) acetohydrazone	0.586	H	Ph(OMe (3), OH (4))	261	72	586	$C_{27}H_{29}O_2N_3Cl_2S_1$	White
3.	5-chloro salicyl-aldehyde-2-(2,5-dichloroanilido) acetohydrazone	0.591	H	Ph(OH (2), Ol (5))	238	56	590.5	$C_{28}H_{26}O_3N_3Cl_3S_1$	White

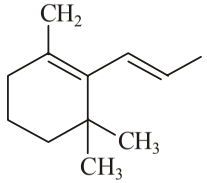
Cont…

Comp.	Acid hydrazones	Quantity of acid hydrazone (g)	Adducts		m.p. (°C)	Yield (%)	Formula Weight	Molecular formula	Colour
			R_1	R_2					
4.	5-Bromo salicylaldehyde-2-(2,5-dichloroanilido) acetohydrazone	0.619	H	Ph(OH (2), Br (5))	258	70	619	$C_{28}H_{26}O_2N_3Cl_2BrS_1$	Silver White
5.	2-Nitro vanilline-2-(2,5-dichloroanilido) acetohydrazone	0.631	H	Ph(NO_2 (2), OCH_3 (3), OH (4))	244	60	631	$C_{29}H_{28}O_6N_4Cl_2S_1$	Cream
6.	O-Nitrobenzaldehyde 2-(2,5-dichloroanilido) acetohydrazone	0.585	H	Ph – NO_2 (2)	235	64	585	$C_{28}H_{26}O_4N_4Cl_2S_1$	White
7.	2-Nitro-5-bromo vanilline-2-(2,5-dichloroanilido) acetohydrazone	0.710	H	Ph(NO_2 (2), OMe (3), OH (4), Br (5))	259	55	710	$C_{29}H_{27}O_6N_4Cl_2BrS_1$	Cream
8.	3, 5-dichloro-2-hydroxy benzaldehyde-2-(2,5-dichloroanilido) acetohydrazone	0.625	H	Ph(OH (2), Cl (3), Cl (5))	253	62	625	$C_{28}H_{25}O_3N_3Cl_4S_1$	White
9.	3-Nitro-6-hydroxy acetophenone-2-(2,5-dichloroanilido) acetohydrazone	0.615	Me	Ph(NO_2 (3), OH (6))	248	57	615	$C_{29}H_{28}O_5N_4Cl_2S_1$	Cream

Cont…

Comp.	Acid hydrazones	Quantity of acid hydrazone (g)	Adducts		m.p. (°C)	Yield (%)	Formula Weight	Molecular formula	Colour
			R_1	R_2					
10.	Acetone-2-(2,5-dichloroanilido) acetohydrazone	0.491	Me	Me	235	41	491	$C_{24}H_{26}O_2N_3Cl_2S_1$	Cream
11.	2-Chlorobenzalde-hyde-2-(2,5-dichloroanilido) acetohydrazone	0.575	H	Ph – Cl (2)	240	49	574.5	$C_{28}H_{26}O_2N_3Cl_3S_1$	White
12.	4-NN-Bis-2'-cyanoethylamino benzaldehyde-2-(2,5-dichloroanilido) acetohydrazone	0.660	H	Ph–N–($CH_2 - CH_2 - CH$)	251	52	660	$C_{34}H_{33}O_2N_6Cl_2S_1$	Light brown
13.	2-Methyl-4-N-N-Bis-2'-cyanoethyl aminobenzaldehyde (2,5-dichloroanilido) acetohydrazone	0.675	H	Ph$\langle^{CH_3\ (2)}_{N(CH_2-CH_2-CN)_2\ (4)}$	258	55	675	$C_{35}H_{36}O_2N_6Cl_2S_1$	Brown
14.	2-Methoxy-4-N-N-bis-2'-cyanoethylamino benzaldehyde (2,5-dichloroanilido) acetohydrazone	0.691	H	Ph$\langle^{OCH_3\ (2)}_{N(CH_2-CH_2-CN)_2\ (4)}$	238	63	691	$C_{35}H_{36}O_3N_6Cl_2S_1$	Brown

Cont…

Comp.	Acid hydrazones	Quantity of acid hydrazone (g)	Adducts		m.p. (°C)	Yield (%)	Formula Weight	Molecular formula	Colour
			R_1	R_2					
15.	Acetophenone-2-(2,5-dichloroanilido) acetohydrazone	0.553	Me	Ph	244	61	553	$C_{29}H_{28}O_2N_3Cl_2S_1$	White
16.	Salicylaldehyde-2-(2,5-dichloroanilido) aceto hydrazone	0.556	H	Ph – OH (2)	247	54	556	$C_{28}H_{27}O_3N_3Cl_2S_1$	White
17.	Anisicaldehyde-2-(2,5-dichloroanilido) acetohydrazone	0.570	H	Ph – OCH_3 (2)	240	58	570	$C_{29}H_{29}O_3N_3Cl_2S_1$	Yellow
18.	β-Ionone-2-(2,5-dichloroanilido) acetohydrazone	0.627	Me		256	39	627	$C_{34}H_{42}O_2N_3Cl_2S_1$	Buff

Solvent for crystallization-ethanol

RESULTS AND DISCUSSION

Thiophenol adducts of various acid hydrazones have been synthesized by the reaction of 2-(2,5-dichloroanilido) acetohydrazide with various carbonyl compounds in 39 to 72% yield. Hydrazone-thiophenol adducts are white, brown and yellow colour solids, having high melting points. The structure of all the compounds are confirmed by IR, PMR and Mass spectral data and are further supported by correct elemental analysis. Newly synthesized compounds have been tested for their antibacterial activity against gram positive bacteria *S. albus, S. aureus* and gram negative bacteria *E. Coli* and *Pseudomonas piosineus*. The compound (**3, 7, 14, 16**) shown significant activities and compound (**5, 8, 13, 17**) have shown moderate activity. The same compounds were tested for their antifungal activity against *Candida albicans, Aspergillus niger* and *Alternaria alternata* at concentration of 30 mg/mL using savored dextrose agar media. The compound (**4, 9, 11, 12**) shown significant activities and compound (**2, 6, 10, 15**) have shown moderate activity against *Candida albicans and Aspergillus Niger*. All the other compounds did not show significant activity against the fungi at the concentration used.

CONCLUSION

Newly synthesized compounds have been tested for their antibacterial activity against gram positive bacteria *S. albus, S. aureus* and gram negative bacteria *E.coli and Pseudomonas piosineus* by agar plate disc diffusion method at 30 µg/mL concentration. *Ampicillin and tetracycline* were used as a reference compounds. The compound (**3, 7, 14, 16**) shown significant activities and compound (**5, 8, 13, 17**) have shown moderate activity. The same compounds were tested for their antifungal activity against *Candida albicans, Aspergillus niger and Alternaria alternata* at concentration of 30 *albicans and Aspergillus niger*. All the other compounds did not show significant activity mg/mL using Savored dextrose agar media. The compound (**4, 9, 11, 12**) shown significant activities and compound (**2, 6, 10, 15**) *have* shown moderate activity against *Candida* against the fungi at the concentration used.

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REFERENCES

1. V. M. Rahman, S. Mukhtar, W. H. Ansari and G. Lemiere, Eur. J. Med. Chem., **40**, 173-184 (2005).
2. J. R. Dimmock, S. C. Vashishtha and J. P. Stables, Eur. J. Med. Chem., **35**, 241-248 (2000).
3. R. Yapia, M. P. La Mara and G. H. Massieu, Biochem. Pharmacol., **16**, 1211-1218 (1967).
4. G. Sava, L. Perissin, L. Lassiani and G. Zabucchi, Chem. Biol. Interact., **53**, 37-43 (1985).
5. O. O. Ajani, C. A. Obafemi, O. C. Nwinyi and D. A. Akinpelu, Bioorg. Med. Chem., **18**, 214-21 (2010).
6. N. V. Bhagavan, Medical Biochemistry, Elsevier Science B. V., Amsterdam, The Netherlands, **17**, (2002) pp. 331-363.
7. M. G. Saulnier, U. Velaprthi and K. Zimmermann, in Progress in Heterocyclic Synthesis, G. Gribble Ed., Elsevier Science B. V., Amsterdam, The Netherlands, **16**, (2005) pp. 228-271.
8. A. Faroumadi, Z. Kiano and F. Soltani, Farmaco., **58**, 1073-1076 (2003).
9. M. R. Holdiness, Tubercle., **68**, 301-309 (1987).
10. E. I. Short, Tubercle., **43**, 33-42 (1962).
11. V. V. Bokharev, A. A. Ghidaspov and E. V. Peresedova, Chem. Heterocycl. Comp., **42**, 1096-1106 (2006).
12. R. P. Fernando, P. I. Maia, S. R. Leite, V. M. Deflon, A. A. Batista, D. N. Sato, S. G. Franzblau and C. Q. Leite, Eur. J. Med. Chem., **45**, 1898-1905 (2010).
13. Z. Brzozowski and F. S. Czewski, Eur. J. Med. Chem., **37**, 709-720 (2002).
14. A. Sherif and F. Rostom, Bioorg. Med. Chem., **18**, 2767-2776 (2010).
15. R. M. Mohareb and A. A. Mohamed, Molecules, **15**, 3602-3617 (2010).
16. R. M. Mohareb, E. E. El-Arab and K. A. El-Sharkaway, Sci. Pharm., **77**, 355-365 (2009).
17. Hoda Z. Shams, Rafat M. Mohareb, Maher H. Helal and Amira E. Mahmoud, Molecules, **16**, 52-73 (2011).

18. Rafat M. Mohareb, Daisy H. Fleita and Ola K. Sakka, Molecules, **16**, 16-27 (2011).

19. W. W. Wardakhan, H. Z. Shams and H. E. Moustafa, Phosph. Sulf. Silicon, **180**, 1815-1827 (2005).

20. E. Pinto, M.-J. R. P. Queiroz, L. A. Vale-Silva, J. F. Oliveira, A. Begouin, J.-M. Begouin and G. Kirsch, Med. Chem., **16**, 8172-8177 (2008).

21. P. Vicini, F. Zani, P. Cozzini and I. Doytchinova, Eur. J. Med. Chem., **37**, 553-564 (2002).

22. C. F. R. Isabel, C. C. Ricardo, M. E. Letícia and R. P. Q. Maria-Joao, Bioorg. Med. Chem. Lett., **14**, 5831-5833 (2004).

23. P. Melnyk, V. Leroux, C. Serghergert and P. Grellier, Design, Bioorg. Med. Chem. Lett., **16**, 31-35 (2006).

24. G. Dannhardt, W. Kiefer, G. Krämer, S. Maehrlein, U. Nowe and B. Fiebich, Eur. J. Med. Chem., **35**, 499-510 (2000).

25. L. W. Zheng, L. L. Wu, B. X. Zhao, W. L. Dong and Y. J. Miao, Bioorg. Med. Chem., **17**, 1957-1962 (2009).

26. Y. L Xia, F. Chuan-Dong, B. X. Zhao, J. Zhao, D. S. Shin and J. Y. Miaom, Eur. J. Med. Chem., **43**, 2347-2353 (2008).

27. L. Brault, E. Migianu, A. Néguesque, E. Battaglia, D. Bagrel and G. Kirsch, Eur. J. Med. Chem., **40**, 757-763 (2005).

28. K. Starcevic, G. Karminski-Zamola, I. Piantanida, M. Zinic, L. Sŭman and M. Kralj, J. Am. Chem. Soc., **127**, 1074-1075 (2005).

29. R. Romagnoli, P. G. Baraldi, M. D. Carrion, C. L. Cara, O. Cruz-Lopez, D. Preti, M. Tolomeo, S. Grimaudo, A. D. Cristina, N. Zonta, J. Balzarini, A. Brancale, T. Sarkar and E. Hamel, Design, Bioorg. Med. Chem., **16**, 5367-5376 (2008).

30. I. C. F. R. Ferreira, M. R. P. Queiroz, M. Vilas-Boas, L. M. Estevinho, A. Begouin and G. Kirsch, Bioorg. Med. Chem. Lett., **16**, 1384-1387 (2006).

31. M. G. Mamolo, V. Falagiani, D. Zampieri, L. Vio and E. Banfo, IIFARMACO, **56**, 587-592 (2001).

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