Research Article

SYNTHESIS, CHARACTERISATION AND ANTICANCER ACTIVITY OF SCHIFF BASE DERIVATIVES OF 5-(2-PHENOXYPYRIDIN-3-YL)-1, 3, 4-THIADIAZOL-2-AMINEAdimule Vinayak¹, Medapa Sudha², Rao Prakash Kumar^{2*}, Kumar Lalita.S³¹Mount Carmel Centre for Scientific and Advanced Research, Mount Carmel College, Vasanth Nagar, Bengaluru, Karnataka, India²Department of Chemistry, Mount Carmel College (Autonomous), Vasanth Nagar, Bengaluru, Karnataka, India³Department of Chemistry, School of Sciences, IGNOU, New-Delhi, India

*Corresponding Author Email: pkras1960@gmail.com

Article Received on: 20/10/13 Revised on: 21/11/13 Approved for publication: 20/12/13

DOI: 10.7897/2230-8407.041214**ABSTRACT**

Schiff Bases of 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-aminoderivatives (a-h) have been synthesized with different aromatic aldehyde and tested for *in vitro* anticancer activity; the 1, 3, 4-thiadiazole derivatives were prepared by the reaction of thiosemicarbazide, phosphorous oxychloride and aromatic acids. The synthesized final Schiff base compounds were purified by column-chromatography and the structures of the titled Schiff bases were elucidated by IR, ¹H NMR and LCMS spectral measurements. Three different cell lines namely Hela, Hep-G2 and MCF7 are used for the present study. The compounds tested were, in most of cases shows cytotoxic effect but, only one compound showed good activity on breast carcinoma cell line having IC₅₀ = 2.28 µg/ mL.

Keywords: Thiosemicarbazide, 1, 3, 4-Thiadiazoles, Schiff bases, Anticancer, Phosphorous-oxy chloride, Hela**INTRODUCTION**

Many five member aromatic systems having three hetero atoms (N, O, S) at symmetrical position have been studied for their interesting physiological properties. The derivatives of 1, 3, 4-thiadiazole were known to possess various pharmacological activities like anti-inflammatory^{1,2}, anticancer³, anti-microbial⁴, anti-tubercular³, anti-convulsant⁵ activities. Other activities that are reported in thiadiazole containing drug include anthelmintic⁶, and analgesic⁷ activities. Their synthesis is outlined in the synthetic schemes 1 and 2. The aim of this study was to synthesise some novel Schiff base derivatives of 1, 3, 4-thiadiazole and testing these compounds on anticancer cell lines. The structures of the compounds were established on the basis of their IR, LCMS and ¹H NMR spectral data. Cancer is known medically as a malignant neoplasm is a broad group of diseases involving unregulated cells growth that are affecting millions of people worldwide. This importance of thiadiazole moiety and continuing demand for new anticancer agents, prompted us to synthesise biologically important eight different novel Schiff base derivatives of 1, 3, 4-thiadiazole ring.

Experimental

The chemicals required for the study were obtained from spectrochem and s-d fine chemicals. The melting points of these synthesized compounds were determined by open capillary tube. The IR spectra were recorded by preparing KBr pellets containing 1 % compounds using FTIR-8400 spectrometer. Liquid Crystal Mass Spectra (LCMS) of the samples were recorded using Agilent and the ¹H NMR spectra were recorded using Varian (400MHz) NMR spectrometer. The purity of the compounds were analysed by Shimadzu HPLC 2010. The structures of synthesized compounds are in good agreement with the proposed ones.

MATERIALS AND METHODS**Scheme 1: Synthesis of 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine (Figure 1)****Step 1: Synthesis of ethyl 2-chloropyridine-3-carboxylate**

2-Chloropyridine-3-carboxylic acid (10 g, 0.0534 mole) was taken in a 500 ml single necked round bottom flask fitted with a reflux condenser. To this reaction mixture absolute ethanol (100 ml) was added while stirring. Concentrated H₂SO₄ was added drop wise at room temperature under stirring. Reaction mixture was refluxed for 4 h. The formation of ethyl 2-chloropyridine-3-carboxylate was monitored by TLC. After completion of the reaction solvent was removed under reduced pressure; Ice cold water was added to the residue and neutralised with saturated solution of NaHCO₃. The product was extracted with ethyl acetate (50 ml x 3) and organic layer was washed with brine (20 ml), dried over Na₂SO₄ and evaporated under reduced pressure. The crude product was used for the next step without any further purification. Yield (88 %), LCMS (M + 1) = 187, HPLC purity = 96 %, ¹H NMR- 1.15 (t, 3H), 3.85 (q, 2H), 7.4 (t, 1H), 8.45 (dd, 1H), 8.85 (d, 1H) TLC-ethyl acetate: hexane (1:9). IR -2845 cm⁻¹ (C-H stretching, s), 3006 cm⁻¹ (C-H stretching, w), 1089 cm⁻¹ (C-Cl stretching, w), 980 cm⁻¹ (C-O stretching esters, w)

Step 2: Synthesis of ethyl 2-phenoxy pyridine-3-carboxylate

Ethyl 2-chloropyridine-3-carboxylate (8.8 g, 0.0474 mole), phenol (8.913 g, 0.09482 mole), anhydrous K₂CO₃ (19.6236 g, 0.1422 mole) and solvent DMF were taken in a single necked round bottom flask fitted with a reflux condenser and heated to 100°C for 10 h. The formation of ethyl 2-phenoxy pyridine-3-carboxylate was monitored by TLC. After the completion of the reaction the reaction mixture was cooled to room temperature added with ice cold water and the product is extracted with ethyl acetate (50 ml x 3) the ethyl

acetate layer was washed with 10 % of NaOH solution (10 ml x 3), and washed with brine (20 ml), dried over Na_2SO_4 and evaporated under reduced pressure. The crude product was purified by column-Chromatography packed with silica gel 100-200 mesh. To begin the eluent is 100 % n-hexane and finally the product is eluted completely using 25 % of ethyl acetate. Yield (68 %), LCMS- $M + 1 = 243.2$, HPLC purity = 92.5 %, $^1\text{H NMR}$ -0.85 (t, 3H), 3.55 (q, 2H), 7.05 (m, 2H), 7.35 (dd, 2H), 8.48 (d, 1H), 8.52 (d, 1H), 8.9 (d, 1H), 8.93 (d, 1H) IR-2865 cm^{-1} (C-H stretching, s) 1485 cm^{-1} (C-H bending), 3060 cm^{-1} (C-H stretching, w), 1080 cm^{-1} (C-stretching esters, w), 1170 cm^{-1} (C-O stretching, ethers, m). TLC ethyl acetate: hexane (2:8).

Step 3: Synthesis of 2-phenoxy pyridine-3-carboxylic acid

The ethyl 2-phenoxy pyridine-3-carboxylate (5.98 g, 0.0278 mol) was taken in a 100 ml single necked round bottom flask. LiOH (2.00 g, 0.838), THF (50 ml) and water (20 ml) were added under stirring. The reaction mixture was stirred at room temperature overnight. The reaction was monitored by TLC. After the completion of the reaction the solvent was removed under reduced pressure. The residue was cooled to 5°C by adding ice cold water and neutralised using concentrated HCl. The solid thus obtained was filtered, washed with cold water and dried under vacuum. Yield (45 %), LCMS = 214 (-ve), HPLC purity = 94.5 %, TLC Ethyl acetate: hexane (4:6). $^1\text{H NMR}$ - 7.05 (m, 2H), 7.35 (dd, 2H) 7.45 (dd, 2H), 9.2 (m, 2H), 10.9 (bs, 1H). IR-2765 cm^{-1} (C-H stretching), (s) 1477 cm^{-1} (C-H bending), 1785 cm^{-1} (C-O stretching), 3060 cm^{-1} (C-H stretching, w), 1160 cm^{-1} (C-O stretching, ethers, m), 2650 cm^{-1} (O-H stretching, bs)

Step 4: Synthesis of 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine

The 2-phenoxy pyridine-3-carboxylic acid (2.691 g, 0.00997 mol) was taken in a 100 ml beaker containing thiosemicarbazide (1.824 g, 0.0199 mole). To this reaction mixture was added 10 ml of phosphorous oxy chloride and mixed well to get the homogeneous mixture. The reaction mixture was irradiated with microwave (30 seconds/interval) for 3 minutes. The reaction was monitored by TLC. After completion of the reaction the residue was added with few ice pieces and neutralised using saturated solution of NaHCO_3 . The solid thus obtained was filtered, washed with cold water and dried over vacuum. Yield (45 %), LCMS = 270, HPLC purity = 94 %, $^1\text{H NMR}$ - 5.15 (bs, 2H), 7.21 (m, 2H), 7.35 (dd, 2H), 8.15 (dd, 2H), 8.8 (m, 2H). IR-2815 cm^{-1} (C-H stretching), (s) 1487 cm^{-1} (C-H bending), 3060 cm^{-1} (C-H stretching, w), 1160 cm^{-1} (C-O stretching, ethers, m), 2890 cm^{-1} (C-H stretching), 1585 cm^{-1} (C-H bending), 2630 cm^{-1} (S-H stretching, w), 1500 cm^{-1} (C-N stretching), 3490 cm^{-1} (NH- stretching, s), and 1630 cm^{-1} (NH bend), TLC ethyl acetate: hexane (5:5).

Scheme-2: General procedure for the synthesis N-[(4-aryl) methylidene]-5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine (Figure 2)

The 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine (250 mg, 1 equivalent, 0.9248 mmole) was dissolved in absolute ethanol (10 ml) containing catalytic amount of acetic acid (2-5 drops). To this reaction mixture various substituted aromatic aldehydes (1.1 equivalent) was added while stirring. The reaction mixture was heated to 80°C for 2-5 h. Reaction was monitored by TLC. After the completion of the reaction, solvent was removed under reduced pressure, ice cold water

was added and the product was extracted using ethyl acetate (10 ml x 2). The organic layer was washed with brine (10 ml), dried over sodium sulphate and evaporated to dryness (Synthetic scheme-2). The crude product thus obtained was purified by column chromatography.

Purification

All the final compounds were purified by column chromatography using silica gel 100-200 mesh, 100 % n-hexane has been used as eluent for column and later using ethyl acetate it was decreased to 75 %. All the purified compounds are 94-98 % pure as reported by HPLC. Table 2 summarizes the physical and analytical data of compounds.

Anticancer activity

The novel 1, 3, 4-Thiadiazole Schiff base compounds (6a-6h) have been selected for the *in vitro* anticancer screening. The compounds were subjected for four different concentrations 1, 2.5, 5 and 10 mg/ mL; (100 $\mu\text{g}/\text{ml}$ to 1000 $\mu\text{g}/\text{ml}$). Three human cell lines were used in this experiment namely: a) human cervical carcinoma cell lines b) human liver carcinoma cell line (HepG2) and c) human breast carcinoma cell line (MCF7).

Preparation of Cell culture

The cell lines were grown in RPMI 1640 medium added with 10 % of fetal bovine serum. To this antibiotic were added (100 units/mL penicillin, 100 $\mu\text{g}/\text{mL}$ streptomycin, L-glutamine 0.03 %, w/v and sodium bicarbonate 2.2 %, w/v). All the cell lines were cultured in flasks kept in a 100 % humidified atmosphere incubated with 10 % of CO_2 for 24 h prior to the addition of the experimental molecules. After 24 h, the Aliquots of 10 μl the test compounds (100 $\mu\text{g}/\text{ml}$, 250 $\mu\text{g}/\text{ml}$, 500 $\mu\text{g}/\text{ml}$ and maximum of 1000 $\mu\text{g}/\text{ml}$) of different dilutions were added to the appropriate micro-titre plate are containing the 90 μl of cells. Cells from one plate of each cell line were added with TCA, to represent a measurement of the cell population for each cell line at the time of drug addition (Ta). After compound addition, plates were incubated at standard conditions for 48 hours and assay was terminated by the addition of cold TCA.

Cell line fixation and SRB assay

Sulphorhodamine B (SRB) is a bright pink aminoxanthine dye with two sulfonic groups. Under mild acidic conditions, SRB binds dye to basic amino acid residues in TCA (trichloro acetic acid) fixed cells to provide a sensitive index of cellular protein content that is linear over a cell density range of visible at least two order of magnitude. After compound addition, plates were incubated for 48 hours and assay was terminated by the addition of cold TCA. Cells were fixed by the gentle addition of 50 μl of cold 30 % TCA and incubated for 60 minutes at 4°C. The supernatant layer was removed and the plates were washed four times with tap water and the air dried. Sulphorhodamine B (SRB) solution (50 μl) in 1 % acetic acid was added to each of the wells, and plates were incubated for 20 minutes at room temperature. After staining the residual dye was removed by washing five times with 1 % acetic acid. The plates were then air dried. Bound stain was subsequently eluted with 10 M Mtrizma base, and the absorbance was read on a micro plate reader at a wavelength of 540 nm. Percent growth was calculated on each plate on the basis for test wells relative to control wells. Percent Growth was expressed as the ratio of average absorbance of the test well to the average absorbance of the

control wells multiplied by 100. Time zero (Ta), control concentration levels (Tb). Percentage growth inhibition was calculated as:

$$[(Tb-Ta)/(C-Ta)] \times 100$$

Table 1: Obtained percentage yield and melting point of the various synthesised novel 1, 3, 4-thiadiazole Schiff base derivatives

Compounds No	L = Linking to R	Molecular Formula	M.P (°C)	Obtained Yield (%)
6a		C ₂₆ H ₁₇ FN ₄ OS	131	81
6b		C ₂₂ H ₁₈ N ₄ O ₃ S	143	56
6c		C ₂₀ H ₁₄ N ₄ O ₂ S	96	55
6d		C ₁₉ H ₁₄ N ₄ OS ₂	148(D)	46
6e		C ₁₈ H ₁₁ BrN ₄ OS ₂	155(D)	78
6f		C ₂₀ H ₁₃ ClN ₄ OS	129	72
6g		C ₂₀ H ₁₃ BrN ₄ OS	88	52
6h		C ₂₀ H ₁₂ BrFN ₄ OS	103	68

Where, D = Decomposes

Table 2: IR, 1H-NMR data of the Synthesised novel 1, 3, 4-Oxadiazoles 6(a-h)

Compound d. No		MS [M+ and M+1], 1H NMR [DMSO-d ₆ , δ ppm], IR [KBr,vcm-1]
6a	MS	452.5 [M+], 453 and 454 [M+1]
	IR	2825 cm ⁻¹ (C-H stretching, s), 1487 cm ⁻¹ (C-H bending), 3030 cm ⁻¹ (C-H stretching, w), 1426 cm ⁻¹ (C-F stretching), 1130 cm ⁻¹ (C-O stretching, ethers, m), 2830 cm ⁻¹ (C-H stretching), 1525 cm ⁻¹ (C-H bending), 2610 cm ⁻¹ (S-H stretching, w), 1509 cm ⁻¹ (C-N stretching), 3390 cm ⁻¹ (NH- stretching, s), and 1630 cm ⁻¹ (NH bend).
	1H-NMR	7.3 (m, 3H), 7.45 (dd, 2H), 7.5 (dd, 2H), 7.9 (dd, 2H) 7.96 (dd, 2H), 8.25 (d, 2H), 8.35 (d, 2H), 9.05 (d, 1H), 9.1 (d, 1H), 10.2 (s, 1H)
6b	MS	418.4 [M+], 419 and 420 [M+1]
	IR	2855 cm ⁻¹ (C-H stretching, s), 1497 cm ⁻¹ (C-H bending), 3021 cm ⁻¹ (C-H stretching, w), 1260 cm ⁻¹ (C-O stretching, ethers, m), 2880 cm ⁻¹ (C-H stretching), 1525 cm ⁻¹ (C-H bending), 2638 cm ⁻¹ (S-H stretching, w), 1520 cm ⁻¹ (C-N stretching), 3470 cm ⁻¹ (NH- stretching, s), and 1630 cm ⁻¹ (NH bend).
	1H-NMR	2.3 (s, 6H), 7.1 (m, 2H), 7.25 (m, 2H), 8.2 (d, 2H), 8.25 (d, 2H), 8.95 (dd, 2H), 9.1 (d, 1H), 10.11 (s, 1H)
6c	MS	374.4 [M+], 373 [M-1]
	IR	2814 cm ⁻¹ (C-H stretching, s), 1489 cm ⁻¹ (C-H bending), 1320 cm ⁻¹ (O-H bending), 3545 cm ⁻¹ (O-H stretching), 3160 cm ⁻¹ (C-H stretching, w), 1160 cm ⁻¹ (C-O stretching, ethers, m), 2790 cm ⁻¹ (C-H stretching), 1565 cm ⁻¹ (C-H bending), 2635 cm ⁻¹ (S-H stretching) (w), 1504 cm ⁻¹ (C-N stretching), 3490 cm ⁻¹ (NH- stretching, s), and 1633 cm ⁻¹ (NH bend)
	1H-NMR	4.85 (bs, 1H), 7.1 (m, 1H), 7.3 (m, 2H), 8.15 (dd, 2H), 8.25 (dd, 2H), 8.9 (d, 2H), 9.15 (d, 2H), 10.08 (s, 1H).
6d	MS	378.4 [M+], 380 [M+1]
	IR	2844 cm ⁻¹ (C-H stretching, s), 1499 cm ⁻¹ (C-H bending), 3040 cm ⁻¹ (C-Stretching, w), 1160 cm ⁻¹ (C-O stretching, ethers, m), 2870 cm ⁻¹ (C-H stretching), 1595 cm ⁻¹ (C-H bending), 2638 cm ⁻¹ (S-H stretching) (w), 1512 cm ⁻¹ (C-N stretching), 3495 cm ⁻¹ (NH- stretching, s), and 1640 cm ⁻¹ (NH bend), 3035 cm ⁻¹ (thiophene C-H stretching)
	1H-NMR	1.2 (s, 3H), 7.2 (m, 2H), 7.3 (m, 2H), 8.2 (dd, 2H), 8.35 (dd, 2H), 9.05 (dd, 2H), 10.15 (s, 1H)
6e	MS	443 [M+], 445 and 446 [M+1]
	IR	2845 cm ⁻¹ (C-H stretching, s), 1477 cm ⁻¹ (C-H bending), 3080 cm ⁻¹ (C-H stretching, w), 720 cm ⁻¹ (C-Br stretching), 1260 cm ⁻¹

		¹ (C-O stretching, ethers, m), 2790 cm ⁻¹ (C-H stretching), 1565 cm ⁻¹ (C-H bending), 2650 cm ⁻¹ (S-H stretching), 1520 cm ⁻¹ (C-N stretching), 3490 cm ⁻¹ (NH- stretching, s), and 1630 cm ⁻¹ (NH bend),
	¹ H-NMR	7.25 (m, 2H), 7.5 (d, 2H), 7.7 (d, 2H), 8.2 (2d, 2H), 9.25 (dd, 2H), 10.2 (s, 1H).
6f	MS	392.8 [M+], 394 and 395 [M+1]
	IR	2915 cm ⁻¹ (C-H stretching, s) 1434 cm ⁻¹ (C-H bending), 1100 cm ⁻¹ (C-Cl stretching, s), 3050 cm ⁻¹ (C-H stretching, w), 1150 cm ⁻¹ (C-O stretching, ethers, m), 2790 cm ⁻¹ (C-H stretching), 1565 cm ⁻¹ (C-H bending), 2640 cm ⁻¹ (S-H stretching, w), 1556 cm ⁻¹ (C-N stretching), 3490 cm ⁻¹ (NH- stretching, s), and 1630 cm ⁻¹ (NH bend).
	¹ H-NMR	7.15 (m, 2H), 7.3 (m, 2H), 7.6 (m, 2H), 7.65 (m, 2H), 8.9 (d, 2H), 9.2 (d, 2H), 10.03 (s, 1H).
6g	MS	437.3 [M+], 438 and 439 [M+1]
	IR	2888 cm ⁻¹ (C-H stretching, s), 680 cm ⁻¹ (C-Br stretching, s) 1480 cm ⁻¹ (C-H bending), 3160 cm ⁻¹ (C-H stretching, w), 1160 cm ⁻¹ (C-O stretching, ethers, m), 2890 cm ⁻¹ (C-H stretching), 1685 cm ⁻¹ (C-H bending), 2640 cm ⁻¹ (S-H stretching, w), 1578 cm ⁻¹ (C-N stretching), 3490 cm ⁻¹ (NH- stretching, s), and 1650 cm ⁻¹ (NH bend),
	¹ H-NMR	7.19 (m, 2H), 7.29 (m, 2H), 7.6 (m, 2H), 8.2 (dd, 2H), 8.95 (d, 2H), 9.25 (d, 2H), 10.1 (s, 1H)
6h	MS	455.3 [M+], 456 and 458 [M+1]
	IR	2835 cm ⁻¹ (C-H stretching, s), 1497 cm ⁻¹ (C-H bending), 3068 cm ⁻¹ (C-H stretching, w), 1260 cm ⁻¹ (C-O stretching, ethers, m), 2890 cm ⁻¹ (C-H stretching), 1585 cm ⁻¹ (C-H bending), 2630 cm ⁻¹ (S-H stretching, w), 1500 cm ⁻¹ (C-N stretching), 3490 cm ⁻¹ (NH- stretching, s), and 1630 cm ⁻¹ (NH bend), 1425 cm ⁻¹ (C-F stretching, s), 690 cm ⁻¹ (C-Br stretching).
	¹ H-NMR	7.25 (m, 2H), 7.56 (m, 2H), 7.85 (dd, 2H), 8.19 (d, 2H), 8.23 (d, 2H), 9.15 (dd, 1H), 10.02 (s, 1H)

d = doublet, dd = doublet of doublet, m = multiple, s = singlet, bs = broad singlet, s = strong (IR), m = medium, q = quintet, w = weak

Table 3: Effect of the synthesised 1, 3, 4-thiadiazole compounds on Hela, Hep-G2, MCF7 cell lines

Compounds	IC 50 (µg/ml)		
	Hela	Hep-G2	MCF7
6a	-	-	117.89
6b	-	116.78	-
6c	106.78	44.34	55.45
6d	33.54	19.56	18.98
6e	-	19.32	14.56
6f	-	44.10	2.28
6g	22.04	23.62	-
6h	-	-	-

Reaction scheme-1: Synthesis of 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine:

Figure 1

Scheme 2:- Synthesis of schiff bases of 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine:

Figure 2

RESULTS AND DISCUSSIONS

Chemistry (Table 1)

Starting from 2-Chloro nicotinic acid which is converted into 2-Chloro pyridine ethyl benzoate according to the method of Wang and Pengetal^{9,11}, this on reaction with phenol and K_2CO_3 yields ethyl 2-phenoxy pyridine-3-carboxylate. The ethyl 2-phenoxy pyridine-3-carboxylate was reacted with aqueous KOH in the presence of THF solvent yields the appropriate 2-phenoxy pyridine-3-carboxylic acid. This compound on reaction with thiosemicarbazide in presence of phosphorous oxy chloride under microwave irradiation gave the corresponding 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine¹². The 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine which is a key intermediate for the synthesis N-[(4-Aryl) methylidene]-5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine.^{12,13} Different substituted aryl aldehydes were reacted in the presence of ethanol and few drops of acetic acid with the intermediate No. (5) To get the final Schiff bases. Eight different Schiff base derivatives have been synthesized. In order to know the potency, these molecules have been tested with leukemic cell lines. The AR grade aromatic aldehydes were purchased from the commercial sources for the above synthesis. On the other hand, according to the known chemotherapeutic activities of 1, 3, 4-Thiadiazoles as antimicrobial⁴ and anticancer agents³, author has synthesized various Schiff base derivatives of 1, 3, 4-thiadiazole into the parent 2-chloro nicotinic acid backbone to obtain more active and less toxic anticancer agents. Thus, fusion of phenol into 2-chloro position of pyridine enhances the solubility. Reaction with 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine and various aromatic aldehydes yields N-[(4-Aryl) methylidene]-5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine¹³ 6(a-h) respectively. The presence of phenoxy pyridine moiety might overcome the water insolubility problem of 1, 3, 4-thiadiazole Schiff base compounds, thus increasing their Bio availability. On the other hand, since many N-[(4-Aryl) methylidene]-5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine¹³ derivatives possess anticancer activity³ it is prompted us to synthesize some novel 5-(2-phenoxy pyridin-3-yl) 1, 3, 4-thiadiazole Schiff base derivatives.

Anticancer activity: (Table 3): Discussion Based on Results

Above eight 6(a-h) compounds were screened for anti-cancer activity at Centre for Cellular and Molecular Platforms (C-CAMP), Tata institute of Fundamental Research (TIFR), Bangalore, India. Three cell lines were used for the evaluation (Human cervix carcinoma cell line, human liver carcinoma cell line and human breast carcinoma cell line). The results are expressed in the form of the concentration of compound that causes 50 % inhibition of cells growth (IC-50). The *in vitro* evaluation revealed that the activity of the tested compounds showed cytotoxicity on all the three cell lines except one compound (6f) which showed good activity on MCF7 cell line. The compound (6a) has showed no effect on cervical cancer and liver cancer cell lines but cytotoxic to breast cancer cell lines whereas the compound (6b) was cytotoxic to human liver carcinoma cell lines, Two compounds (6c, 6d) cytotoxic to all the 3-cell lines. The compounds (6e) and (6g) showed moderate activity. The compound (6e) is moderate active on Hep-G2 and MCF7 cell lines whereas compound (6g) is moderate active on Hela and Hep-G2 cell lines. Compound (6f) has showed good activity on MCF7 cell line having IC-50 of 2.28 $\mu\text{g/ml}$ and

moderately active on Hep-G2 cell line. The compound (6h) has not showed any activity on all the 3-cell lines. The results of the anticancer screening of the tested compounds are illustrated in Table 3.

CONCLUSIONS

From the data it is found that the compounds 6(e) and 6(f) were the most effective against breast carcinoma cell line having IC₅₀ value 14.56 $\mu\text{g/ml}$ and 2.28 $\mu\text{g/ml}$ respectively; whereas compounds (6a), (6c), (6d) were cytotoxic against the breast cancer cell line. Compounds (6e) and (6f) are the only two compounds showed very good IC-50 values and they were selective towards breast cancer cell lines showing the IC₅₀ of 14.56 $\mu\text{g/ml}$ and 2.28 $\mu\text{g/ml}$ respectively. The activity of the tested compounds could be correlated with structure variation and modification.

ACKNOWLEDGEMENT

The authors are thankful to Sr. Juanita, Principal Mount Carmel College, (Autonomous) Bengaluru, India Mrs Sudha Medapa, and HOD of Chemistry for providing facilities to carrying out research work and constant encouragement. I thank Dr. Prakash Kumar Rao and Dr. Lalita. S. Kumar for guiding and for providing needful materials in time to carryout this research work. Thanks are also due to Director, IISc Bengaluru, India for providing spectral data.

REFERENCES

- Pandey Alok, Dewangan Dhansay, Verma Shekhar, Mishra Achal, Dubey Ravindra Dhar. Synthesis of Schiff bases of 2-amino-5-aryl-1, 3, 4-thiadiazole and its Analgesic, Anti Inflammatory, Anti-Bacterial and Anti-Tubercular Activity; International Journal of Chem Tech Research 2011; 3(1): 178.
- Sainy Jitendra, Mishra Ganesh P, Sharma Rajesh and Chaturvedi Subhash C. 2-Amino-5-Sulfonyl-1, 3, 4-Thiadiazole a novel series of anti-inflammatory and analgesic agents; Pharmaceutical Chemistry Journal 2009; 43: 1-2.
- DA Ibrahim. Synthesis and biological evaluation of 3, 6-disubstituted 1, 2, 4 triazolo-thiadiazole derivatives as a novel class of potential antitumor agents; Eur. J. Med. Chem 2009; 44: 2776-2781. <http://dx.doi.org/10.1016/j.ejmech.2009.01.003> PMID:19203813
- Mullick Pooja *et al.* Synthesis, characterization and anti-microbial activity of new thiadiazole derivatives; Bulletin of the Korean Chemical Society 2010; 31(8): 2345-2350. <http://dx.doi.org/10.5012/bkcs.2010.31.8.2345>
- Christopher B Chapleo, Peter L Myers, Alan CB, Smith Ian F, Tulloch Donald S. Walter :Substituted 1, 3, 4-thiadiazoles with anticonvulsant activity; J. Med. Chem 1987; 30(5): 951-954. <http://dx.doi.org/10.1021/jm00388a038>
- SM Khawass, MA Khalil, AA Hazzaa, HA Bassiouny, NF Loutfy. Synthesis of some 1, 2, 4-triazolo [3, 4-b][1, 3, 4] thiadiazoles as potential anthelmintics; Farmaco 1989; 44: 703-709. PMID:2590368
- Dsouza Ullas P, Revanasiddappa BC and EVS Subrahmanyam. Analgesic and anti-inflammatory activity of 1, 3, 4- ox diazoles derivatives 2012; 2(3): 202-205.
- Luo, Ming Yang, Zhen. From PCT Int. Appl; 2009. WO 20090592 43 A1 20090507.
- Wan, Rongetal. Microwave assisted synthesis of propesticides 1, 3, 4-thiadiazole aminophosphonates Synthetic Communications 2011; 41(6): 864-870. <http://dx.doi.org/10.1080/00397911003707030>
- Han, Feng *et al.* Synthesis and biological activity of 1, 3, 4-thiadiazole aroyl urea derivatives; Youji Huaxue 2010; 30(1): 132-136.
- Wang, Peng *et al.* One-pot synthesis of 1, 3, 4-thiadiazolethiazolidinone derivatives; Journal of Chemical Research 2009; 11: 671-673. <http://dx.doi.org/10.3184/030823409X12559727242438>
- Chinnagiri T, Kumar Kirti, Jathi Keshavayya. Synthesis, Characterization, and Biological Activity of 5-Phenyl-1, 3, 4-thiadiazole-2-amine incorporated azo dye derivatives; Organic Chemistry International; 2013. p. 1-7.
- Sawa, YoshiroIrisawa, Junji Maeda, Ryozyo, Ger. Offen; 1997. DE 2030981 A 19710107

Cite this article as:

Adimule Vinayak, Medapa Sudha, Rao Prakash Kumar, Kumar Lalita.S. Synthesis, characterisation and anticancer activity of Schiff base derivatives of 5-(2-phenoxy pyridin-3-yl)-1, 3, 4-thiadiazol-2-amine. Int. Res. J. Pharm. 2013; 4(12):62-66 <http://dx.doi.org/10.7897/2230-8407.041214>