ORIGINAL RESEARCH ARTICLES

SYNTHESIS AND ANTICANCER ACTIVITY OF *N*-SUBSTITUTED INDOLE DERIVATIVES

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ABSTRACT

Thizolidine-2,4-dione (1), on reaction with *p*-fluorobenzaldehyde in the presence of piperidine and toluene, gives (*Z*)-5-(2-fluororobenzylidin)-thiazolidin-2,4-dione (2), which on reaction with indole and *o*-chlorobenzaldehyde in the presence of ethanol yielded final derivatives i.e (*Z*)-5-(4-fluororo-benzylidene)-3-[(1*H*-indol-1-yl) (substituted phenyl) methyl] thiazolidine-2,4-dione (3a-3e). All the synthesized compounds were characterized by UV, FTIR, ^{1}H NMR, MASS spectroscopy and elemental analysis. These compounds were screened for their anticancer activity against MCF-7 human breast cancer cell line by sulpho rhodamine B (SRB) assay method. Among the tested compounds, compound 3b with nitro group, which is a strong electron withdrawing group, was found to be most active for the inhibition of topoisomerase-I enzyme.

Keywords: Indole, anticancer activity, topoisomerase-I, human breast cancer cell line.

INTRODUCTION

Breast cancer is a global health problem and is the major cause of cancer-related deaths in women throughout the world. Many drugs are available for breast malignancies which are approved by US-FDA. Newer chemical moieties are required due to rise in the cases of drug resistance. Many drugs having indole moiety are used for the treatment of breast cancer and show multiple mechanisms of action[1-2]. Indole moiety is found in various natural products having different biological actions. Starting from the simple structural framework of serotonin, the well known neurotransmitter, to complex alkaloids which are used clinically as anticancer agent, mitomycin C, and the antihypertensive alkaloid reserpine, the indole moiety is considered as the active principle. Numerous indole derivatives with remarkable pharmacological activity and their utility as therapeutic agents have attracted chemist and researchers to find a potent lead against cancer disease. Sumatriptan, tadalafil, fluvastatin and rizatriptan are some examples of synthetic drugs containing indole nucleus and available in the market. Moreover, indole nucleus may be used as a lead compound for the discovery of new anticancer agents with less side effects and more potency.

Current research is based on the development of an indole-based potent anticancer agent with incorporation of thiazolidine-2,4-dione, which is an excellent strategy in drug design for the treatment of cancer due to its proper alkalinity, water solubility and the capacity to form hydrogen bonds. Thiazolidine-2,4-dione ring is a frequent moiety of numerous drugs, such as butamison (antihelmintic activity), clometocillin (antibiotic activity)[3], clospirazine (antipsychotic activity), dithiazanine (anthelmintic activity), etozoline (diuretic activity), letosteine (mucolytic activity), methicillin (antibiotic), mycobacidin (antimicrobial)[4-12], pidotimod (immunomodulator), pioglitazone (antidiabetic activity)[13-17], tiramide (anti-inflammatory)[17-19] and timonacic (hepatoprotective). The aim of this research, was to obtain a potent anticancer agent by the combination of thiazolidine ring with indole nucleus.

MATERIALS AND METHODS

Laboratory grade chemicals and reagents were used to synthesize all the reported compounds. The melting

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Table I: Physical data of the synthesized compounds (3a-3e)

Compound code	R	Melting Point (°C)	*R_f Value	% Yield
3a	4-Cl	189-190	0.71	85
3b	4-NO_2	161-162	0.70	75
3c	4-F	187-188	0.65	68
3d	3,4,5-OCH_3	201-202	0.76	65
3e	4-OH	151-152	0.72	73

***Solvent system:** Benzene: Ethyl Acetate: Glacial Acetic Acid (9.0:0.5:0.5)*

points were determined in open capillaries and are uncorrected. The IR spectra of compounds were recorded on Perkin-Elmer infrared–283 FTIR spectrometer by KBr pellet technique, ^{1}H NMR spectra were recorded on Bruker DRX-300 (300 MHZ, FT NMR) spectrophotometer using TMS as an internal standard and $CDCl_3$ as solvent. Mass spectra were obtained using LC-MS (Shimadzu-2010AT) under electron spray ionization (ESI) technique and elemental analysis was performed using Elemental Vario EL III, Carlo-Erba 1108. TLC was performed to monitor the reactions and to determine the purity of the products.

Chemistry

The scheme for the synthesis of (*Z*)-5-(4-fluororobenzylidene)-3-[(1*H*-indol-1-yl) (2-substitutedphenyl) methyl] thiazolidine-2,4-dione (3a-3e) is shown in Fig. 1. According to the scheme, the reaction is started with thizolidine-2,4-dione (1), which on reaction with *p*-fluorobenzaldehyde in the presence of piperidine and toluene gives (*Z*)-5-(2-fluororobenzylidin)-thiazolidin-2,4-dione (2). Compound (2) on reaction with indole and benzaldehyde derivative yields (*Z*)-5-(4-fluororobenzylidene)-3-[(1*H*-indol-1-yl) (substitutedphenyl) methyl] thiazolidine-2,4-dione (3a-3e). R_f value, melting point and percentage yield for the synthesized derivatives are given in Table I.

Procedure for the synthesis of thiazolidine-2, 4-dione (1)

Equimolar quantities of chloroacetic acid and thiourea were dissolved in 20 mL of water in a three necked flask. The mixture was stirred for 15 minutes to obtain a white precipitate. To the contents of the flask, 20 mL of concentrated hydrochloric acid was added slowly from the dropping funnel, flask was then connected with condenser

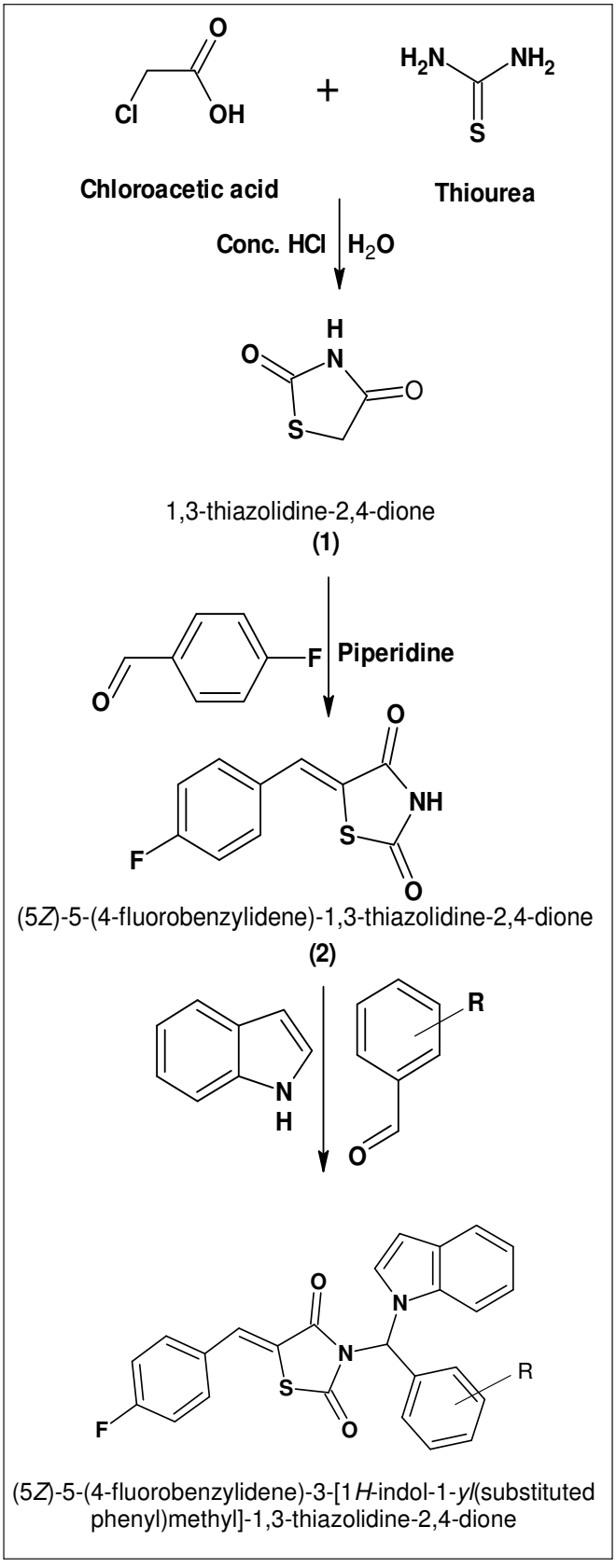
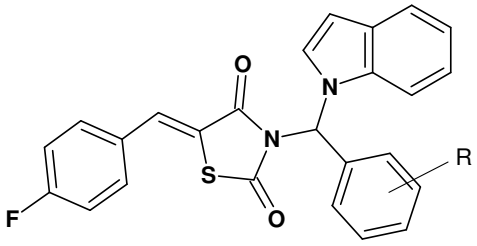


Fig. 1: Scheme for the synthesis of compound 3a-3e



(5*Z*)-5-(4-fluorobenzylidene)-3-[1*H*-indol-1-yl(substituted phenyl)methyl]-1,3-thiazolidine-2,4-dione

Fig. 2: General structure of compound 3a-3e

and gentle heat applied to complete the reaction. The mixture was stirred and refluxed for 12 h. On cooling, the contents of the flask solidified to a cluster of white needles. The product was filtered, washed and recrystallized from ethanol to give compound (1).

Procedure for the synthesis of (*Z*)-5-(2-fluororobenzylidin)-thiazolidin-2, 4-dione (2)

Equimolar quantities of thiazolidin-2, 4-dione and *p*-fluorobenzaldehyde were taken and suspended in dry toluene. To this, catalytic amount of piperidine was added. The reaction mixture was stirred and refluxed for 3 h at 110 °C. The compound was filtered and washed with cold dry toluene. The compound was dried and recrystallized with ethyl alcohol to obtained intermediate product (2).

General procedure for the synthesis of (*Z*)-5-(4-fluororobenzylidene)-3-[(1*H*-indol-1-yl)(substitutedphenyl) methyl] thiazolidine-2,4-dione (3a-3e)

Equimolar quantities of (*Z*)-5-(4-fluororobenzylidin)-thiazolidin-2,4-dione and indole were dissolved in 5 mL of ethanol and benzaldehyde derivative was added to this reaction mixture. The reaction mixture was refluxed for 11 h with continuous stirring. The reaction mixture was filtered and cold water was added to the filtrate. The product precipitated out, which was filtered, dried and recrystallized from ethanol to obtain the compound (3a-3e).

Spectral data for the compounds 3a-3e

The analytical and spectral data of final compounds 3a-3e (Fig. 2) are given below:

(*Z*)-5-(4-fluororobenzylidene)-3-[(1*H*-indol-1-yl)(4-chlorophenyl) methyl] thiazolidine-2,4-dione (3a)

UV λ_{max}(DMSO): 287.0 nm; FTIR (KBr) (ν): 3052.11 (Aromatic C-H str.), 1706.03 (C=O str., amide (cyclic lactums)), 1608.59 (Aromatic C=C str.), 1324.65 (C-F str.), 1143.35 (C-N str.), 904.12 (C-S str.), 810.63 (C-H def*p*-disubstituted bezene), 692.47 cm^{-1} (C-Cl str.); ^{1}H NMR (DMSO-d_6) δ 1.620 (s, 1H, C-H), 5.391 (s, 1H, vinylic C-H), 7.104-7.132 (d, 1H, Ar-H), 7.134-7.160 (d, 1H, Ar-H), 7.161-7.182 (t, 1H, Ar-H), 7.185-7.245 (t, 1H, Ar-H), 7.247-7.273 (d, 1H, Ar-H), 7.274-7.387 (d, 1H, Ar-H) 7.390-7.507 (t, 1H, Ar-H), 7.510-7.518 (t, 1H, Ar-H) 7.521-7.556 (d, 1H, Ar-H), 7.560-7.623 (d, 1H, Ar-H), 7.625-7.643 (d, 1H, Ar-H), 7.646-7.918 (d, 1H,Ar-H), 7.921-8.021 (d, 1H, Ar-H), 8.025-8.059 ppm (d, 1H, Ar-H); EIMS (m/z) (% relative abundance): $[M]^+$ 462.98 (100), [M+1] 463.04 (30), [M+2] 464.10 (26); Fragments: 443.07 (58), 427.51 (28), 333.08 (19), 243.05 (30), 167.02 (40), 77.98 (13), 633.96 (30); Elemental analysis: Calculated for $C_{25}H_{16}ClFN_2O_2S$: C, 64.90; H, 3.44; N, 6.06; Found: C, 64.92; H, 3.45; N, 6.07%.

(*Z*)-5-(4-fluororobenzylidene)-3-[(1*H*-indol-1-yl)(4-nitrophenyl) methyl] thiazolidine-2,4-dione (3b)

UV λ_{max}(DMSO): 370.0 nm; FTIR (KBr) (ν): 3043.11 (Aromatic C-H str.), 1744.98 (C=O str., amide (cyclic lactums)), 1614.66 (Aromatic C=C str.), 1515.16 (Asymmetric N=O str.), 1346.58 (C-F str.), 1319.47 (Symmetric N=O str.), 1166.85 (C-N str.), 918.12 (C-S str.), 815.83 cm^{-1} (C-H def*p*-disubstituted bezene); ^{1}H NMR (DMSO-d_6): δ 1.625 (s, 1H, C-H), 5.397 (s, 1H, vinylic C-H), 7.104-7.132 (d, 1H, Ar-H), 7.134-7.158 (d, 1H, Ar-H), 7.161-7.183 (t, 1H, Ar-H), 7.185-7.246 (t, 1H, Ar-H), 7.247-7.272 (d, 1H, Ar-H), 7.274-7.388 (d, 1H, Ar-H) 7.390-7.508 (t, 1H, Ar-H), 7.510-7.518 (t, 1H, Ar-H) 7.521-7.558 (d, 1H, Ar-H), 7.560-7.624 (d, 1H, Ar-H), 7.625-7.642 (d, 1H, Ar-H), 7.646-7.919 (d, 1H,Ar-H), 7.921-8.021 (d, 1H, Ar-H), 8.203-8.352 ppm (d, 1H, Ar-H); EIMS (m/z) (% relative abundance): $[M]^+$ 473.53(100), [M+1] 474.02 (43); Fragments: 454.54 (38), 427.53 (29), 333.08 (23), 243.05 (22), 167.02 (36), 77.98 (22), 63.96 (34); Elemental analysis: Calculated for $C_{25}H_{16}FN_3O_4S$: C, 63.43; H, 3.37; N, 8.86; Found: C, 63.44; H, 3.38; N, 8.88 %.

(*Z*)-5-(4-fluororobenzylidene)-3-[(1*H*-indol-1-yl)(4-fluorophenyl) methyl] thiazolidine-2,4-dione (3c)

UV λ_{max}(DMSO): 285.0 nm; FTIR (KBr) (ν): 3035.47 (Aromatic C-H str.), 1710.06 (C=O str., amide (cyclic lactums)), 1595.89 (Aromatic C=C str.), 1338.58 (C-F str.), 1157.35 (C-N str.), 915.12 (C-S str.), 816.83 cm^{-1} (C-H def *p*-disubstituted bezene); ^{1}H NMR (DMSO-d_6) δ 1.636 (s, 1H, C-H), 5.376 (s, 1H, vinylic C-H), 7.173-7.210 (d, 1H, Ar-H), 7.213-7.234 (d, 1H, Ar-H), 7.236-7.292 (t, 1H, Ar-H), 7.295-7.331 (t, 1H, Ar-H), 7.332-7.374

(d, 1H, Ar-H), 7.376-7.413 (d, 1H, Ar-H) 7.415-7.442 (t, 1H, Ar-H), 7.445-7.482 (t, 1H, Ar-H) 7.485-7.543 (d, 1H, Ar-H), 7.545-7.639 (d, 1H, Ar-H), 7.641-7.661 (d, 1H, Ar-H), 7.764-7.785 (d, 1H, Ar-H), 7.787-7.891 (d, 1H, Ar-H), 8.108-8.121 ppm (d, 1H, Ar-H); EIMS (m/z) (% relative abundance): $[M]^+$ 446.53 (100), [M+1] 447.54 (44); Fragments: 427.10 (22), 333.08 (19), 243.05 (25), 167.02 (26), 77.98 (17), 63.96 (24); Elemental analysis: Calculated for $C_{25}H_{16}FN_2O_2S$: C, 67.18; H, 3.58; N, 6.27; Found: C, 67.19; H, 3.59; N, 6.25 %.

(*Z*)-5-(4-fluororobenzylidene)-3-[(1*H*-indol-1-yl) (3,4,5-tri-methoxyphenyl) methyl] thiazolidine-2,4-dione (3d)

UV λ_{max}(DMSO): 342.0 nm; FTIR (KBr) (ν): 3043.11 (Aromatic C-H str.), 2823.88 (Aliphatic C-H str.), 1714.08 (C=O str., amide (cyclic lactums)), 1587.09 (Aromatic C=C str.), 1339.8 (C-F str.), 1176.85 (C-N str.), 915.14 (C-S str.), 815.83 cm^{-1} (C-H def *p*-disubstituted bezene); ^{1}H NMR (DMSO-d_6) δ 1.636 (s, 1H, C-H), 4.012 (s, 3H, OCH_3), 4.015 (s, 3H, OCH_3), 4.016 (s, 3H, OCH_3,) 5.376 (s, 1H, vinylic C-H), 7.213-7.234 (d, 1H, Ar-H), 7.236-7.293 (d, 1H, Ar-H), 7.376-7.412 (d, 1H, Ar-H), 7.415-7.443 (d, 1H, Ar-H) 7.445-7.484 (t, 1H, Ar-H), 7.485-7.542 (t, 1H, Ar-H) 7.545-7.639 (d, 1H, Ar-H), 7.641-7.659 (d, 1H, Ar-H), 7.662 (s, 1H, Ar-H), 7.764 (s, 1H,Ar-H), 7.787-7.891 (d, 1H, Ar-H), 7.893-7.910 ppm (d, 1H, Ar-H); EIMS (m/z) (% relative abundance): $[M]^+$ 518.61(100), [M+1] 519.62 (29); Fragments: 499.61 (51), 427.51 (39), 333.08 (23), 243.05(49), 167.02 (29), 77.98 (28), 63.96 (30); Elemental analysis: Calculated for $C_{28}H_{23}FN_2O_5S$: C, 64.86; H, 4.44; N, 5.40; Found: C, 64.84; H, 4.42; N, 5.41 %.

(*Z*)-5-(4-fluororobenzylidene)-3-[(1*H*-indol-1-yl) (4-hydroxyphenyl) methyl] thiazolidine-2,4-dione (3e)

UV λ_{max}(DMSO): 310.0 nm; FTIR (KBr) (ν): 3437.63 (Phenolic O-H str.) 3057.32 (Aromatic C-H str.), 1714.92 (C=O str., amide (cyclic lactums)), 1583.09 (Aromatic C=C str.), 1346.88 (C-F str.), 1141.42 (C-N str.), 919.98 (C-S str.), 815.83 (C-H def*p*-disubstituted bezene); ^{1}H NMR (DMSO-d_6): δ 1.625 (s, 1H, C-H), 5.390 (s, 1H, Vinylic C-H), 7.100 (s, 3H, Phenolic O-H), 7.104-7.132 (d, 1H, Ar-H), 7.134-7.160 (d, 1H, Ar-H), 7.161-7.183 (d, 1H, Ar-H), 7.185-7.245 (d, 1H, Ar-H), 7.247-7.388 (d, 1H, Ar-H), 7.390-7.508 (d, 1H, Ar-H), 7.560-7.623 (d, 1H, Ar-H), 7.625-7.644 (d, 1H, Ar-H), 7.646-7.918 (d, 1H, Ar-H), 7.921-8.023 (d, 1H,Ar-H), 8.025-7.060 (d, 1H, Ar-H), 8.061-8.223 (d, 1H, Ar-H), 8.225-8.274 (d, 1H, Ar-H), 8.276-8.289 ppm (d, 1H, Ar-H); EIMS (m/z) (% relative abundance): $[M]^+$ 454.07 (100), [M+1] 455.07 (32); Fragments: 427.10 (49), 425.54 (38), 333.08 (21), 244.05 (29), 167.02 (42), 77.98 (14), 63.96 (24);Elemental analysis: Calculated for $C_{25}H_{16}FN_2O_3S$: C, 66.06; H, 3.52; N, 6.16; Found: C, 66.05; H, 3.50; N, 6.17 %.

Anticancer activity

All the newly synthesized indole derivatives were screened for anticancer activity against MCF 7 (Human Breast Cancer) cell line by SRB (Sulpho rodamine B) assay. The SRB assay possesses a colorimetric end point and is non- destructive and indefinitely stable. These practical advances make the SRB assay an appropriate and sensitive assay to measure percent growth inhibition. The results are shown in Table II.

Determination of Cell viability by SRB assay

Principle

SRB is a dark pink aminoxanthine dye with sulphonic group. Under mild conditions, SRB binds to protein basic amino acid residue in tricholoro acetic acid (TCA) fixed cells to provide sensitive index of cellular protein content that is linear over a cell density range of at least two orders of magnitude. Colour development in SRB assay is rapid, stable and visible. The developed colour can be measured over a broad range of visible wavelength in either a spectrophotometer or a 96 well plate reader. When TCA fixed, SRB stained samples are dried they can be stored indefinitely without deterioration.

Cell culture used

The cell lines were growth in RPMII640 medium containing 10 % fetal bovine serum and 2 m MoL L-glutamine. For present screening experiment, cells were inoculated into 96 well micro titer plates in 100 µL at plating densities as shown in the study details, depending on the doubling time of individual cell lines. After cell inoculation, the microtiter plates were inoculated at 37 ^oC, 5 % CO_2, 95 % air and 100 % relative humidity for 24 h prior to addition of experimental drugs.

Procedure

- After 24 h one 96 plate containing $5X10^{3n}$ cells $well^{-1}$ was fixed *in situ* with TCA, to represent a measurement of the cell population at the time of drug addiction (Tz).
- Experimental drugs were initially solubilized in dimethyl sulfoxide at 100 µg mL^{-1} and diluted to 1 mg mL^{-1} using water and stored frozen prior to use.
- At the time of drug addition, an aliquote of frozen concentrate (1 mg mL^{-1}) was thawed and diluted to

100 µg mL^{-1}, 200 µg mL^{-1}, 400 µg mL^{-1} and 800 µg mL^{-1} with complete medium containing test article.

- Aliquots of 10 µL of these different drug dilutions were added to the appropriate micro titer wells already containing 90 µL of medium, resulting in the required final drug concentrations i.e. 10, 20, 40 and 80 µg mL^{-1}.
- After compound addition, plates were incubated at standard conditions for 48 h and assay was terminated by the addition of cold TCA.
- Cells were fixed *in situ* by the gentle addition of 50 µL of cold 30 % (w/w) TCA (final concentration, 10 %TCA) and incubated for 60 minutes at 4 °C.
- The supernatant was discarded; the plates were washed five minutes with tap water and air dried.
- Sulfo rhodamine B (SRB) solution (50 µL) at 0.4 % (w/V) in acetic acid was added to each of the wells and plates were incubated for 20 minutes at room temperature.
- After staining, unbound dye was recovered and the residual dye was removed by washing five times with 1 % acetic acid, the plates were air dried.
- Bound stain was subsequently eluted with 10 mmoL trizma base and the absorbance was read on a plate reader at a wavelength of a wavelength of 540 nm with 690 nm reference wavelength.
- Percent growth was expressed as the ratio of average absorbance of the test well to the absorbance of the control wells X 100.

Percentage growth inhibition was calculated by using the formula:

% Growth Inhibition (GI_{50})= [(Ti-Tz)/(C-Tz)]X100

Concentrations for which Ti>Tz, (Ti-Tz)) is positive or zero

% Growth Inhibition (GI_{50})= [(Ti-Tz)/Tz]X100

Concentrations for which Ti<Tz, (Ti-Tz) is negative

where,

GI_{50}= drug concentration resulting in 50 % reduction in the net protein increase (as measured by SRB staining) in control cells during the drug incubation

C =Control growth

Ti= test growth in the presence of drug at the four concentration levels

Tz= Cell population at the time of drug addiction

Table II: Anticancer data of synthesized 3a-3e derivatives

Human Breast Cancer Cell Line MCF7 % Control Growth Drug Concentrations (µg mL^{-1})					
Compound no.	**10**	**20**	**40**	**80**	**IC_{50} Value (µM)**
3a	55.4	34.6	10.6	-15.4	13.8
3b	40.4	45.3	38.2	30.8	9.9
3c	52.4	40.9	34.7	29.2	30.3
3d	58.6	25.6	10.0	2.8	22.3
3e	59.6	41.5	30.2	22.6	37.9
ADR (Std.)	0.3	-10.7	-33.6	-59.7	8.2

RESULTS AND DISCUSSION

Newer indole derivatives in combination with thiazolidine-2,4-dione were synthesized by the scheme shown in Fig. 1. The resultant products (3a-3e) have consistent values of C, H and N contents with predicted structure. The structures of newly synthesized thiazolidine-2,4-dione derivatives were elucidate through IR, ^{1}HNMR and mass spectral data. In IR spectra of series, significant bands appeared at 3045.39 (aromatic C-H str.), 1704.96 (C=O str., amide (cyclic lactums), 1593.09 (aromatic C=C str.), 1336.58 (C-F str.), 1153.35 (C-N str.), 916.12 (C-S str.). In ^{1}HNMR spectra of these compounds, a broad multiplet of aromatic proton was appeared between 6.90-8.23 ppm. All derivatives showed M+1 peak in their mass spectra. All the newly synthesized benzimidazole derivatives were screened for anticancer activity against MCF 7 (human breast cancer) cell line by SRB (sulpho rodamine B) assay. The SRB assay possesses a colorimetric end point and is nondestructive and indefinitely stable. These practical advantages make the SRB assay an appropriate and sensitive assay to measure percent growth inhibition. Results of anticancer activity states that compound 3b was found to be most potent with 35.2 % control growth. Compound 3b was also to be active with 40.5 % control growth <10 µg mL^{-1} GI_{50} value.

CONCLUSION

All the synthesized compounds were found to possess significant anticancer activity. Compound 3b was found most potent with minimum IC_{50} value**.** Present research work illustrates that indole moiety has lot of opportunity to find out a potent anticancer agent. Compound 3b can be

used as a lead molecule for further research. It may give a direction to our researcher to find out a potent anticancer agent with promising activity and less side effects.

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REFERENCES

1. Sakineh D. and Saeed E.: Indole in the target-based design of anticancer agents: A versatile scaffold with diverse mechanisms. **Eur. Med. Chem.,** 2018, 150, 9-29.
2. Sidhu J.S., Singla R. and Mayank J. V.: Indole Derivatives as Anticancer Agents for Breast Cancer Therapy: A Review. **Anti-Cancer Agents Med. Chem.,** 2015, 16(2), 160-173.
3. Albrecht U., Gordes D., Schmidt E., Thurow K., Lalk M. and Langer P.: Synthesis and structure-activity relationship of 2-alkylidenethiazolidine-4,5-diones as antibiotic agents. **Bioorg. Med. Chem.,** 2005, 13, 4402-4407.
4. Dundar O.B., Ozgen O., Mentese A., Altanlar N., Ath O., Kendi E., and Ertan R.: Synthesis and antimicrobial activity of some new thiazolyl thiazolidine-2,4-dione derivatives. **Bioorg. Med. Chem.** 2007, 15, 6012-6017.
5. Gouveia F.L., Oliveira R.M. and Oliveira T.B.: Synthesis, antimicrobial and cytotoxic activities of some 5-arylidene -4-thioxo-thiazolidine-2-ones. **Eur. J. Med. Chem.,** 2009, 44, 2038-2048.
6. Kallangouda R.A. and Shankar G.A.: Synthesis, characterization and antimicrobial activity evaluation of new 2,4-Thiazolidinones bearing imidazo[2,1-b][1,3,4] thiadiazole moiety. **Arabian J. Chem.**, 2010,1-8.
7. Karthikeyan M.S. and Holla B.S.: Synthesis, anti-inflammatory and antimicrobial activities of some 2,4-Dichloro-5-fluorophenyl substituted arylidene triazolothiazolidinones. **Monatsh. Chem.,** 2009,139, 691-696.
8. Patel N.B. and Shaikh F.M.: Synthesis and antimicrobial activity of new 4-thiazolidinone derivatives containing 2-amino-6-methoxybenzothiazole. **Saudi Pharm. J**., 2010, 1-8.
9. Prasad J.S., Rangappa K.S. and Kavitha C.V.: Synthesis of new bioactive venlafaxine analoge: novel thiazolidine-4-ones as antimicrobials. **Bioorg. Med. Chem**., 2006, 14, 2290-2299.
10. Rangappa K.S., Kavitha C.V., Swamy S.N. and Mantelingu D.: Synthesis of new bioactive venlafaxine analogs: Novel thiazolidin-4-ones as antimicrobials. **Bioorg. Med. Chem**., 2006, 14, 2290-2299.
11. Rani M., Kabilan S. and Ramachandrn R.: Design, synthesis and biological evaluation of novel 2-[(2, 4-diaryl-3-azabicyclo[3.3.1]nonan-9-ylidene)hydrazono]-1,3-thiazolidin-4-ones as a new class of antimicrobial agents. **Bioorg. Med. Chem**., 2009, 19, 2819-2823.
12. Srivastava S.D., Upashyay A. and Srivastava S.K.: Conventional and microwave assisted synthesis of some new *N*-[(4-oxo-2-substituted aryl-1, 3-thiazolidine)-acetamidyl]-5-nitroindazole and its antimicrobial activity. **Eur. J. Med. Chem**., 2010, 1-8.
13. Desai R.C., Han W., Metzger E.J., Bergman J.P., Gratale D.F., Macnaul K.L., Berger J.P., Doebber T.W., Leung K., Moller D.E., Heck J.V. and Sahoo S.P.: 5- Aryl Thiazolidine-2,4-diones: Discovery of PPAR Dual α/γ Agonist as Antidiabetic Agents. **Bioorg. Med. Chem. Lett**., 2003, 13, 2795-2798.
14. Ide T., Nakazawa T., Mochizuki T. and Murakami K.: Tissue-specific actions of antidiabetic thiazolidinediones on the reduced fatty acid oxidation in skeletal muscle and liver of zucker diabetic fatty rats. **Metabolism**, 2000, 49, 521-525.
15. Lohray B. B., Reddy K. A. and Bhushan V.: Novel Antidiabetic and hypolipidemic agents benzofuran–containing thiazolidinediones. **J. Med. Chem**., 1999, 42, 1927-1940.
16. Madhavan G.R., Chakrabarti R., Kumar S.K.B., Misra P., Mamidi R.N.V.S., Balraju V., Kasiram K., Babu R.K., Suresh J., Lohray B.B., Lohray V.B., Iqbal J. and Rajagopalan R.: Novel phthalazinone and benzoxazinone containing thiazolidinediones as antidiabetic and hypolipidemic agents. **Eur. J. Med. Chem**., 2001, 36, 627-637.
17. Pattan S.R., Suresh C., Pujar V.D., Reddy V.V.K., Rasal V.P. and Koti B.C.: Synthesis and antidiabetic activity of 2-Amino[5(4-sulphonylbenzylidine)-2,4-thiazolidinone]-7-chloro-6-fluorobenzo thiazole. **Indian J. Chem**., 2005, 44, 2404-2408.
18. Barros C.D., Amato A.A., Oliveira T.B., Iannini K.B.R., Silva A.L., Silva T.G., Leite E.S., Hernandes M.Z. and Pitta I.R.: Synthesis and anti-inflammatory activity of new arylidene-thiazolidine-2, 4-diones as PPARγ ligands. **Bioorg. Med. Chem**., 2010, 18, 3805-3811.
19. Karthikeyan M.S. and Holla B.S.: Synthesis, anti-inflammatory and antimicrobial activities of some 2,4-Dichloro-5-fluorophenyl substituted arylidene triazolothiazolidinones. **Monatsh. Chem**., 2008, 139, 691-696.