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Synthesis And Biological Activity Of Some New Stilbene Derivatives And Their Analogues

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ABSTRACT

The current research was aimed at the synthesis of cis stilbene derivatives and testing their anti-cancer abilities. Various cis stilbenes substituted on the olefinic bridge were prepared and their characterizations were given using IR, NMR and MASS spectra. Sequences of the compounds that were tested regarding their cytotoxic effect on MCF-7 and HeLa cell lines. Of all the compounds, Compound 4a that was a methyl ester showed stronger activities against MCF-7 and HeLa cell lines with the IC50 value of 22.24 µm and 27.43 µm respectively.

1. INTRODUCTION

For centuries, nature has served as a rich reservoir of therapeutic agents, with a significant proportion of contemporary medicines derived from or inspired by natural products. This is particularly evident in the field of oncology, where many plant-based compounds have formed the foundation for novel anticancer agents. Compared to conventional chemotherapeutics, targeted molecules derived from natural sources often offer enhanced efficacy profiles and reduced toxicity.

Among these, stilbene-based compounds, including E-stilbene and Z-stilbene (Fig. 1), though not naturally occurring in their unsubstituted forms, are structurally represented through numerous hydroxylated derivatives found abundantly in medicinal plants. One such example is trans-3,5,4′-trihydroxystilbene, commonly known as resveratrol [1–7], which is present in grapes and has been associated with cardiovascular benefits, notably in populations consuming red wine [8–11].

Plants frequently produce stilbene derivatives as a defense mechanism against microbial invasion, and these compounds have demonstrated a broad spectrum of biological activities. Documented properties include antifungal [12], antibacterial [12], cytotoxic [13], anti-inflammatory [14], and anticonvulsant effects [15]. Resveratrol, in particular, exhibits potent antioxidant activity [16], modulates lipid metabolism, suppresses enzymes such as ribonucleotide reductase and DNA polymerase, and enhances MAP kinase activity — a pathway implicated in neurodegenerative diseases like Alzheimer's and Parkinson's disorders [17]. Additionally, some stilbene derivatives have shown the ability to inhibit platelet aggregation [18].

In the present study, conformational modifications of the resveratrol structure were explored by preserving its stilbene core while substituting hydroxyl groups with alternative functional moieties and introducing substituents at the olefinic linkage. This strategy aims to develop derivatives with improved pharmacological properties and better formulation potential

2. MATERIALS AND METHOD

All chemicals, reagents, and solvents used in this study were of analytical grade and procured from commercial suppliers, including Sigma-Aldrich, HiMedia, and SDFine. Spectral data for the synthesized compounds were recorded and compared with previously reported literature values for confirmation. The ^1H NMR spectra were obtained using a Bruker Avance 3400 spectrometer operating at 400 MHz, with either DMSO-d_6 or $CDCl_3$ as solvents. Chemical shifts (δ) were recorded in

parts per million (ppm), with coupling constants (J) expressed in Hertz (Hz). Signal multiplicities were denoted as s (singlet), d (doublet), t (triplet), and m (multiplet).

Thin Layer Chromatography (TLC) was performed on silica gel 60 F_{254} pre-coated plates (Merck) to monitor the progress of reactions. Melting points of the synthesized compounds were determined using a Stuart SMP3 melting point apparatus and are uncorrected. Elemental analyses (C, H, N, S) were conducted on a Vario MICRO CHNS analyzer. For the cytotoxicity evaluation, 3-(4,5-dimethylthiazolyl-2)-2,5-diphenyltetrazolium bromide (MTT), Dulbecco's Modified Eagle's Medium (DMEM), antibiotic-antimycotic solutions, trypsin-EDTA, and phosphate-buffered saline (PBS) were obtained from Sigma-Aldrich (St. Louis, MO, USA). Fetal Bovine Serum (FBS) was procured from GIBCO. Cell culture flasks (25 cm^2 and 75 cm^2) and 96-well plates were purchased from Eppendorf India. All other reagents and chemicals used were of analytical grade. Human cancer cell lines, including HeLa (cervical carcinoma), HT-29 (colorectal cancer), and MCF-7 (breast adenocarcinoma), were obtained from the National Centre for Cell Science (NCCS), Pune, India.

3. EXPERIMENTAL

3.1 General Procedure for the Preparation of Compounds 3a-c. A mixture of phenylacetic acid derivatives (2a–b, 2 mmol), heteroaryl benzaldehydes (1a–d, 2 mmol), and triethylamine (0.5 mL) was refluxed in acetic anhydride (5 mL) for 12 hours. Upon completion, the reaction mixture was transferred into a flask containing 50 mL of hot saturated sodium carbonate solution and allowed to stand undisturbed overnight at room temperature. The reaction mixture was subsequently extracted twice with diethyl ether (2 × 50 mL), and the combined organic layers were discarded. The remaining aqueous phase was acidified with dilute hydrochloric acid to precipitate the crude product. The solid obtained was collected by filtration, washed with water, and recrystallized from a mixture of ethyl acetate and hexane to afford the pure target compounds [19].

3.2 General Procedure for the Preparation of Compounds 4 a-f. To a stirred solution of carboxylic acid derivative 3 (172 mg, 0.5 mmol) in absolute methanol (20 mL), concentrated sulfuric acid (0.5 mL) was added dropwise. The reaction mixture was refluxed for 6 hours under continuous stirring. After completion, approximately 90% of the excess methanol was removed by evaporation under reduced pressure. The residue was then poured into ice-cold water (300 mL), and the resulting mixture was extracted with diethyl ether (2 × 40 mL). The combined organic layers were sequentially washed with 2% aqueous sodium hydroxide solution (2 × 50 mL) followed by distilled water (200 mL). The ether layer was dried over anhydrous sodium sulfate, and solvent removal under reduced pressure afforded the corresponding methyl ester derivatives as solid products [19].

3.3 General Procedure for the Preparation of Compounds 5a-e.

Carboxylic acid derivative 3 (172 mg, 0.5 mmol) was dissolved in benzene (10 mL), and thionyl chloride (1 mL) was added to the solution. The mixture was refluxed for 6 hours. Upon completion, excess thionyl chloride and benzene were removed under reduced pressure, and the resulting residue was maintained under vacuum for 30 minutes to eliminate residual solvents. The crude acid chloride thus obtained was then added to an aqueous solution of methylamine (40%, 5 mL) and stirred at room temperature for 2 hours. The precipitated solid was collected by filtration, washed thoroughly with 2% aqueous sodium hydroxide and distilled water, and dried. The crude product was purified by recrystallization from a mixture of ethyl acetate and hexane to yield the analytically pure amide derivatives [19].

3.3. Biological assays:

3.3.1 Cell culture:

MCF-7 (breast adenocarcinoma), HT-29 (colorectal carcinoma), and HeLa (cervical carcinoma) cell lines were maintained in Minimum Essential Medium (MEM) supplemented with 10% fetal bovine serum (FBS) and 0.5 mL^{-1} of penicillin-streptomycin solution. The cultures were incubated at 37 °C in a humidified atmosphere containing 5% CO_2 and 95% air. The synthesized chalcone derivatives (3a–h) and flavonol derivatives (4a–g) were initially dissolved in dimethyl sulfoxide (DMSO) to prepare stock solutions, which were stored in aliquots at –20 °C until use. For the MTT assay, accurately weighed quantities of each test compound were dissolved in DMSO, and the resulting solutions were further diluted with culture medium to a final concentration of 1 mg/mL. Cells were treated with serial dilutions of these test compounds, ranging from 10 µM to 100 µM, to evaluate their cytotoxic effects.

3.3.2. MTT assay:

The antiproliferative activity of the synthesized chalcone (3a–h) and flavonol (4a–g) derivatives was evaluated against MCF-7, HT-29, and HeLa cell lines using the methyl thiazolyl tetrazolium (MTT) colorimetric assay. Experiments were performed in triplicate at six different concentrations for each compound, across three independent trials. The cultured cells were harvested by trypsinization and viability was assessed using the trypan blue exclusion method. The viable cell count was determined using a haemocytometer, and cells were seeded into 96-well plates at a density of 5.0×10^3 cells per well in MEM supplemented with 10% FBS. The plates were incubated overnight at 37 °C in a 5% CO_2 atmosphere to allow cell attachment.

Following incubation, the medium was aspirated and replaced with 100 µL of fresh medium containing serial dilutions of the test compounds. Control wells received an equivalent concentration of DMSO. After a 48-hour treatment period, the media containing test compounds were removed and replaced with 100 µL of medium containing MTT solution (0.5 mg/mL). Plates were incubated for an additional 3 hours at 37 °C to allow the reduction of MTT by metabolically active cells into insoluble formazan crystals. Subsequently, the medium was carefully removed, and the resulting formazan crystals were dissolved in DMSO. The absorbance of each well was measured at 570 nm using a microplate reader.

3.5 Structure characterization:

3.5.1 (E)-3-(1,4-diphenyl-1H-pyrazol-3-yl)-2-phenylacrylic acid (3a):

IR (cm^{-1}): The structure of the synthesized compound was confirmed by IR, ^1H NMR, elemental analysis, and mass spectrometry. The IR spectrum (KBr, cm^{-1}) showed characteristic absorption bands at 3290 (O–H stretching), 1631 (carbonyl C=O stretching), and 1547 (olefinic C=C stretching).

The ^1H NMR spectrum (400 MHz, $CDCl_3$, δ ppm) exhibited a singlet at 6.84 (1H, olefinic proton), a doublet at 7.66–7.64 (J = 6.8 Hz, 6H, aromatic protons), a multiplet at 7.83–7.79 (7H, aromatic protons), and a triplet at 7.90–7.87 (J = 7.4 Hz, aromatic protons). Elemental analysis calculated for $C_{24}H_{18}N_2O_2$ (Molecular weight: 366) was: C, 78.67%; H, 4.95%; N, 7.65%. The experimental values obtained were: C, 78.65%; H, 4.93%; N, 7.67%, closely matching the theoretical values. The ESI-MS spectrum revealed a molecular ion peak at m/z 367 $[M+H]^+$, supporting the proposed molecular formula.

3.5.2 (E)-3-(1-(4-methoxyphenyl)-4-phenyl-1H-pyrazol-3-yl)-2-phenylacrylic acid (3b):

IR (cm^{-1}): The synthesized compound was characterized using ^1H NMR, elemental analysis, and mass spectrometry. The ^1H NMR spectrum (400 MHz, $CDCl_3$, δ ppm) displayed a singlet at 6.63 (1H, olefinic proton), a doublet at 7.04–7.02 (J = 8.4 Hz, 1H, aromatic), a multiplet at 7.40–7.34 (2H, aromatic), a multiplet at 7.52–7.45 (1H, aromatic), a doublet at 7.67–7.65 (J = 8.4 Hz, 2H, aromatic), and a doublet at 7.80–7.78 (aromatic protons).

Elemental analysis calculated for $C_{25}H_{20}N_2O_3$ (Molecular weight: 396) gave theoretical values: C, 75.74%; H, 5.08%; N, 7.07%. The experimentally determined values were: C, 75.76%; H, 5.10%; N, 7.05%, closely consistent with the calculated composition.The ESI-MS analysis showed a molecular ion peak at m/z 397 $[M+H]^+$, confirming the expected molecular formula.

3.5.3 (E)-3-(1,4-diphenyl-1H-pyrazol-3-yl)-2-(3-fluorophenyl)acrylic acid (3c):

IR (cm^{-1}): The synthesized compound was characterized by IR, ^1H NMR, elemental analysis, and mass spectrometry. The IR spectrum (KBr, cm^{-1}) showed prominent absorption bands at 3350 (O–H stretching), 1650 (C=O stretching), and 1560 (olefinic C=C stretching). The ^1H NMR spectrum (400 MHz, $CDCl_3$, δ ppm) revealed a singlet at 6.80 (1H, olefinic proton), a doublet at 7.11–7.09 (J = 4.8 Hz, 1H, aromatic), a multiplet at 7.33–7.21 (2H, aromatic), and a multiplet at 7.57–7.40 (8H, aromatic protons). Elemental analysis calculated for $C_{24}H_{17}FN_2O_2$ (Molecular weight: 384) gave theoretical values: C, 74.99%; H, 4.46%; F, 4.94%; N, 7.29%. The experimentally determined values were: C, 74.97%; H, 4.48%; F, 4.92%; N, 7.27%, in excellent agreement with the calculated composition. The ESI-MS spectrum displayed a molecular ion peak at m/z 385 $[M+H]^+$, consistent with the proposed molecular formula.

3.5.4 (E)-methyl 3-(1,4-diphenyl-1H-pyrazol-3-yl)-2-phenylacrylate (4a):

The structure of the synthesized compound was confirmed through IR, ^1H NMR, and mass spectrometric analysis. The IR spectrum (KBr, cm^{-1}) exhibited characteristic absorption bands at 3010 (aromatic C–H stretching), 1648 (carbonyl C=O stretching), and 1562 (olefinic C=C stretching).

The ^1H NMR spectrum (400 MHz, $CDCl_3$, δ ppm) displayed a singlet at 3.60 (3H, $–OCH_3$), a singlet at 6.93 (1H, olefinic proton), a multiplet at 7.40–7.34 (4H, aromatic), and a multiplet at 7.57–7.47 (6H, aromatic).

3.5.5 (E)-methyl 3-(1-(4-methoxyphenyl)-4-phenyl-1H-pyrazol-3-yl)-2-phenylacrylate (4b):

The synthesized compound was characterized by IR, ^1H NMR, elemental analysis, and mass spectrometry. The IR spectrum (KBr, cm^{-1}) exhibited absorption bands at 3040 (aromatic C–H stretching), 1632 (C=O stretching), and 1566 (olefinic C=C stretching), confirming the presence of key functional groups.

The ^1H NMR spectrum (400 MHz, $CDCl_3$, δ ppm) showed singlets at 3.76 (3H, $–OCH_3$) and 3.87 (3H, $–OCH_3$), a singlet at 6.74 (1H, olefinic proton), a doublet at 7.03–7.02 (J = 2 Hz, 1H, aromatic), multiplets at 7.37–7.31 (4H, aromatic) and 7.52–7.49 (5H, aromatic), doublets at 7.64–7.62 (1H, aromatic) and 7.80–7.78 (1H, aromatic), a singlet at 8.17–8.21 (1H, aromatic), a singlet at 8.52 (1H, aromatic), and a singlet at 8.56 (1H, –NH). Elemental analysis calculated for $C_{26}H_{22}N_2O_3$ (Molecular weight: 410) gave theoretical values: C, 76.08%; H, 5.40%; N, 6.82%. The experimentally obtained values were C, 76.10%; H, 5.38%; N, 6.80%, in excellent agreement with the theoretical values. The ESI-MS spectrum showed a molecular ion peak at m/z 411 $[M+H]^+$, consistent with the proposed molecular structure..

3.5.6 (E)-methyl 2-(3-fluorophenyl)-3-(1-(4-methoxyphenyl)-4-phenyl-1H-pyrazol-3-yl)acrylate (4c):

The synthesized compound was characterized by IR, ^1H NMR, elemental analysis, and mass spectrometry. The IR spectrum (KBr, cm^{-1}) exhibited characteristic absorption bands at 3078 (aromatic C–H stretching), 1574 (C=O stretching), and 1575 (olefinic C=C stretching), confirming the presence of key functional groups.

The ^1H NMR spectrum (400 MHz, $CDCl_3$, δ ppm) displayed singlets at 3.41 (3H, $–OCH_3$) and 3.73 (3H, $–OCH_3$), a singlet at 6.80 (1H, olefinic proton), a doublet at 7.11–7.09 (J = 4.8 Hz, 1H, aromatic), a multiplet at 7.33–7.29 (1H, aromatic), a multiplet at 7.60–7.38 (9H, aromatic), a doublet at 7.66–7.64 (J = 7.2 Hz, 2H, aromatic), a doublet at 7.84–7.72 (2H, aromatic), and a singlet at 8.43 (1H, aromatic). Elemental analysis calculated for $C_{26}H_{21}FN_2O_3$ (Molecular weight: 428) gave theoretical values: C, 72.88%; H, 4.94%; N, 6.54%. The experimentally determined values were: C, 72.90%; H, 4.92%; N, 6.56%, closely aligning with the calculated composition.

3.5.7 (E)-methyl 2-phenyl-3-(pyridine-3-yl)acrylate (4d):

The structure of the synthesized compound was confirmed by IR, ^1H NMR, elemental analysis, and mass spectrometry. The IR spectrum (KBr, cm^{-1}) displayed characteristic absorption bands at 3040 (aromatic C–H stretching), 1632 (C=O stretching), and 1570 (olefinic C=C stretching).

The ^1H NMR spectrum (400 MHz, $CDCl_3$, δ ppm) exhibited a singlet at 3.64 (3H, $–OCH_3$), a singlet at 6.84 (1H, olefinic proton), a multiplet at 7.05–6.92 (3H, aromatic), a triplet at 7.24–7.23 (J = 3.54 Hz, 1H, aromatic), a doublet at 7.48–7.46 (J = 8.8 Hz, 1H, aromatic), a doublet at 7.64–7.62 (J = 4.64 Hz, 1H, aromatic), a doublet at 7.76–7.74 (J = 8.7 Hz, 1H, aromatic), a multiplet at 7.66–7.71 (1H, aromatic), a doublet at 8.02–8.01 (J = 2.56 Hz, 1H, aromatic), and a singlet at 8.34 (1H, aromatic).

Elemental analysis calculated for $C_{15}H_{13}NO_2$ (Molecular weight: 239) gave theoretical values: C, 75.30%; H, 5.48%; N, 5.85%. The experimentally obtained values were: C, 75.32%; H, 5.46%; N, 5.87%, closely consistent with the calculated composition.

3.5.8 (E)-methyl 2-phenyl-3-(thiophen-2-yl)acrylate (4e):

IR (cm^{-1}): 3021 –CH aromatic, 1628 –C=O, 1551 (olefinic C=C): ^{1}HNMR at δ ppm: 3.68 (s, 3H, $-OCH_3$), 7.01 (s, 1H, olefinic -CH), 7.44-7.40 (t, j = 7.5, 2H), 7.73-7.58 (m, 3H), 7.08-7.04 (t, j = 5.2Hz, 1H), 8.04-8.06 (d, j = 7.94Hz, 2H); Elemental analysis: calculated for $C_{14}H_{12}O_2S$ (244); calculated; C, 68.83; H, 4.95; found C, 68.81; H, 4.92; ESI MS (m/z): 245[M+1].

3.5.9 (E)-3-(1,4-diphenyl-1H-pyrazol-3-yl)-N-methyl-2-phenylacrylamide (5a):

IR (cm^{-1}): 3259 –NH, 3059 –CH aromatic, 1643 –C=O, 1571 (olefinic C=C): ^{1}H NMR at δ ppm: 2.62-.261 (d, j = 5.2, 3H, $-CH_3$), 5.44 ,(bs, 1H, -NH), 6.695 (s, 1H), 7.41-7.38 (m, 3H), 7.53-7.46 (m, 6H), 7.79-7.83 (m, 6H), 8.556 (s, 1H);); Elemental analysis: calculated for $C_{25}H_{21}N_3O$ (379); calculated; C, 79.13; H, 5.58; N, 11.07 found C, 79.11; H, 5.60; N, 11.05; ESI MS (m/z): 380[M+1].

3.5.10 (E)-3-(1-(4-methoxyphenyl-4-phenyl-1H-pyrazol-3-yl)-N-methyl-2-phenylacrylamide (5b):

IR (cm^{-1}): 3260 –NH, 3078 –CH aromatic, 1919 –CH aliphatic, 1534 –C=O, 1568 (olefinic C=C). ^{1}HNMR at δ ppm:. (d, 3H, $-CH_3$, J= Hz), 3.87 (s, 3H, $-OCH_3$), 5.5 (NH), 6.80 (s, 1H, olefinic CH), 7.05-7.02 (m, 2H, aromatic), 7.40-7.33(m, 5H, aromatic), 7.52-7.44(m, 3H, aromatic), 7.62-7.60 (d, j = 8.4, 2H, aromatic), 7.80-7-78 (d, j = 8.8, 2H, aromatic), 8.54(s, 1H, aromatic);); Elemental analysis: calculated for $C_{26}H_{23}N_3O_2$ (409); calculated; C, 79.13; H, 5.58; N, 11.07 found C, 79.11; H, 5.60; N, 11.05; ESI MS (m/z): 410[M+1].

3.5.11 (E)-2-(3-fluorophenyl)-3-(1-(4-methoxyphenyl)-4-phenyl-1H-pyrazol-3-yl)-N-methylacrylamide (5c):

IR (cm^{-1}): 3259 –NH, 3059 –CH aromatic, 2950 –CH aliphatic, 1643 –C=O, 1552 (olefinic C=C). ^{1}H NMR at δ ppm: 1.59-1.57 (d, j = 5.6Hz, 3H, $-CH_3$), 4.51 (bs, 1H, -NH), 6.90 (s, 1H, aromatic), 7.03-7.01 (m, 2H, aromatic), 7.20-7.18 (m, 8H, aromatic), 7.60-7.79 (m, 3H, aromatic), 8.41 (s, 1H, aromatic); Elemental analysis: calculated for $C_{26}H_{22}FN_3O_2$ (427); calculated; C, 73.05; H, 5.19; F, 4.44; N, 9.83 found C, 73.02; H, 5.21; F, 4.40; N, 9.83; ESI MS (m/z): 428[M+1].

3.5.12 (E)-N-methyl-2-phenyl-3-(pyridine-3-yl)acrylamide (5d):

IR (cm^{-1}): 3289 –NH, 3021 –CH aromatic, 1628 –C=O, 1556 (olefinic C=C). ^{1}HNMR at δ ppm: 2.78 (d, j = 4.3Hz, 3H, $-CH_3$), 5.23 (bs, 1H, -NH), 6.98(s, 1H, olefinic -CH),7.45- 7.42(m, 2H, aromatic), 7.59-7.63(m, 3H, aromatic), 8.00-8.05 (m, 4H, aromatic); Elemental analysis: calculated for $C_{15}H_{14}N_2O$ (238); calculated; C, 75.61; H, 5.92; N, 11.76 found C, 75.59; H, 5.94; N, 11.72; ESI MS (m/z): 239[M+1].

3.5.13 (E)-N-methyl-2-phenyl-3-(thiophen-2-yl)acrylamide (5e):

IR (cm^{-1}): 3244 -NH, 3046 -CH aromatic, 1626-C=O ; ^{1}HNMR at δ ppm: 2.72-2.71 (d, j = 5.2Hz, 3H, $-CH_3$), 5,42 (bs, 1H, NH), 6.87 (S, 1H), 6.97-6.94 (t, j = 7.2Hz, 1H), 7.05-7.03 (d, j = 8.4Hz, 1H, aromatic), 7.14-7.11 (m, 1H, aromatic), 7.52-

7.47 (m, 3H, aromatic), 7.43-7.42 (m, 1H, aromatic), 7.85-7.84 (d, j = 7.6Hz, 1H, aromatic); Elemental analysis: calculated for $C_{14}H_{13}NOS$ (243); calculated; C, 69.11; H, 5.39; N, 5.76 found C, 69.09; H, 5.42; N, 5.78 ESI MS (m/z): 244[M+1].

3.5.14 (E)-3-(1,4-diphenyl-1H-pyrazol-3-yl)-N-ethyl-2-phenylacrylamide (5f):

The structural confirmation of the synthesized compound was established through IR, ^{1}H NMR, elemental analysis, and mass spectrometric techniques. The IR spectrum (KBr, cm^{-1}) displayed absorption bands at 3280 (N–H stretching), 3045 (aromatic C–H stretching), 1642 (carbonyl C=O), and 1559 (olefinic C=C stretching), indicating the presence of characteristic functional groups.

^{1}H NMR (400 MHz, $CDCl_3$, δ ppm) exhibited signals at 1.24–1.20 (triplet, 3H, $–CH_3$, J = 3.8 Hz), 2.46–2.40 (quartet, 2H, $–CH_2$), 5.44 (broad singlet, 1H, –NH), 6.64 (singlet, 1H, olefinic proton), 7.43–7.39 (multiplet, 3H, aromatic), 7.45–7.57 (multiplet, 7H, aromatic), 7.79–7.83 (multiplet, 5H, aromatic), and 8.52 (singlet, 1H, aromatic).

Elemental analysis calculated for $C_{26}H_{23}N_3O$ (Molecular weight: 393) yielded C, 79.36%; H, 5.89%; N, 10.68%. The experimentally obtained values were C, 79.34%; H, 5.91%; N, 10.70%, consistent with the theoretical calculations.

3.5.15 (E)-N-ethyl-3-(1-(4-methoxyphenyl-4-phenyl-1H-pyrazol-3-yl)-N-methyl-2-phenylacrylamide (5g):

The synthesized compound was characterized by IR, ^{1}H NMR, elemental analysis, and mass spectrometry. The IR spectrum (KBr, cm^{-1}) exhibited characteristic absorption bands at 3280 (N–H stretching), 3079 (aromatic C–H stretching), 2959 (aliphatic C–H stretching), 1654 (carbonyl C=O), and 1564 (olefinic C=C). ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm) displayed signals at 1.10–1.06 (triplet, 3H, $–CH_3$, J = 7.1 Hz), 3.37–3.30 (quartet, 2H, $–CH_2$), 3.87 (singlet, 3H, $–OCH_3$), 5.45 (broad singlet, 1H, –NH), 6.61 (singlet, 1H, olefinic proton), 7.03–6.98 (multiplet, 3H, aromatic protons), 7.32–7.39 (multiplet, 4H, aromatic), 7.43–7.58 (multiplet, 4H, aromatic), 7.62–7.65 (doublet, 1H, aromatic), 7.78–7.80 (doublet, 2H, aromatic), and 8.52 (singlet, 1H, aromatic). Elemental analysis calculated for $C_{27}H_{25}N_3O_2$ (Molecular weight: 423) was C, 76.57%; H, 5.95%; N, 9.92%. The experimentally found values were C, 76.58%; H, 5.92%; N, 9.90%, which were in good agreement with the theoretical values. The ESI-MS spectrum showed a molecular ion peak at m/z 424 $[M+H]^+$, confirming the proposed molecular structure.

4. RESULTS AND DISCUSSIONS

4.1 Chemistry

A series of stilbene derivatives were prepared through a base-promoted condensation reaction involving phenylacetic acids (compound 2) and various heteroaryl aldehydes (compound 1) in the presence of triethylamine, resulting in the formation of carboxylic acids (compound 3) as outlined in Table 1. These carboxylic acids were subsequently esterified using methanol and a catalytic quantity of sulfuric acid, producing methyl esters (compound 4), detailed in Table 2. Further, treatment of carboxylic acids 3 with thionyl chloride under reflux in benzene generated the corresponding acid chlorides, which were then reacted with selected amines to yield amide derivatives (compound 5), as presented in Table 3. Complete characterization data for these synthesized compounds is summarized in Tables 1–3. Infrared (IR) spectral analysis of the synthesized stilbenes displayed characteristic carbonyl stretching bands within 1600–1654 cm^{-1}, alongside olefinic C=C stretching absorptions between 1540–1575 cm^{-1}. Carboxylic acid functionalities showed hydroxyl bands in the 3290–3350 cm^{-1} region and C=O absorptions near 1609–1613 cm^{-1}, while amide derivatives exhibited N–H stretching frequencies in the range of 3244–3289 cm^{-1}. In the ^1H NMR spectra of hydroxylated chalcones, hydroxyl protons appeared as singlets between δ 12.40–12.60 ppm, and amide protons were observed as broad singlets around δ 5.0–6.0 ppm. The presence of the olefinic segment was confirmed by singlets corresponding to olefinic protons appearing in the δ 6.61–6.98 ppm range. Additionally, mass spectrometry analysis in positive ion mode revealed molecular ion peaks consistent with the expected molecular weights of the stilbene derivatives, confirming their structures.

4.2 Cytotoxicity activity

Cytotoxic potential of fifteen synthesized cis-stilbene derivatives, modified at the olefinic linkage between the aromatic rings (compounds 3a–c, 4a–e, and 5a–g), was assessed in vitro against two human cancer cell lines: MCF-7 (breast cancer) and HeLa (cervical cancer), employing the MTT assay method [20]. IC_{50} values were calculated by plotting semi-log graphs of compound concentration versus percentage inhibition using Microsoft Excel, as illustrated in Figure 2. Cisplatin served as the reference drug, demonstrating IC_{50} values of 4.15 µM for MCF-7 and 6.05 µM for HeLa cell lines. A comprehensive summary of the anticancer activities of all tested derivatives is presented in Tables 4, 5, and 6.

Introducing a carboxylic acid group at the olefinic position led to compound 3a, which exhibited notable activity against HeLa cells with an IC_{50} of 41.39 µM. Converting this COOH group into a methyl ester (compound 4a) significantly enhanced cytotoxicity, showing over a twofold increase in potency against both MCF-7 and HeLa lines compared to 3a. Further modification of the COOH group in compounds 3a and 3b into N-methyl amides resulted in increased cytotoxicity in both cell lines relative to 5a and 5b, though a decrease was noted for 5a specifically against HeLa cells.

Additionally, transforming the carboxylic acid group in compounds 3a and 3b into N-ethyl amides (compounds 5f and 5g)

improved cytotoxic effects against HeLa cells but showed a marginal reduction against MCF-7 cells. Overall, the majority of the derivatives (3a–b, 4a–d, and 5a–g) demonstrated greater cytotoxic activity toward HeLa cells compared to MCF-7.

Among the entire series, compound 4a, featuring a methyl ester group, emerged as the most potent agent against both MCF-7 and HeLa cell lines, recording IC_{50} values of 22.24 μM and 27.43 μM, respectively.

5. CONCLUSION

In this investigation, a series of cis-stilbene derivatives were synthesized by condensing various heteroaryl aldehydes with substituted phenylacetic acids in the presence of triethylamine and acetic anhydride. The purified compounds were subsequently evaluated for their cytotoxic potential against MCF-7 (breast cancer) and HeLa (cervical cancer) cell lines. Among the synthesized derivatives, compound 4a demonstrated the most promising anticancer activity, with IC_{50} values of 22.24 μM for MCF-7 and 27.43 μM for HeLa cell lines.

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REFERENCES

[1] Pervaiz S, Holme AL. Resveratrol: its biologic targets and functional activity. Antioxid Redox Signal. 2009; 11: 2851-2897.

[2] Burns J, Yokota T, Ashihara H. Plant foods and herbal sources of resveratrol. J Agric Food Chem. 2002; 50: 3337-3340.

[3] Maite IG, Sonia RP, Rosa ML. Resveratrol and piceid levels in natural and blended peanut butters. J Agric Food Chem. 2000; 48(12): 6352-6354.

[4] Maria GC, Panagiotis A, Nikolaos K. Trans-Resveratrol in wines from the major greek red and white grape varieties. Food Control. 2006; 17(9): 700-706.

[5] Fremont L. Biological effects of resveratrol. Life Sci. 2000; 66(8): 663-673.

[6] Bavaresco L, Vezzulli S, Civardi S. Effect of lime-induced leaf chlorosis on ochratoxin a trans-resveratrol, and ε-viniferin production in grapevine (*Vitis vinifera* L.) berries infected by *Aspergillus carbonarius*. J Agric Food Chem. 2008; 56(6): 2085-2089.

[7] Sobolev VS. Localized production of phytoalexins by peanut (*Arachis hypogaea*) kernels in response to invasionby *Aspergillus* species. J Agric Food Chem. 2008; 56(6): 1949-1954.

[8] Burns J, Yokota T, Ashihara H, Lean ME, Crozier A. Plant foods and herbal sources of resveratrol. J Agric Food Chem. 2002; 50: 3337–3340.

[9] Soleas GJ, Diamandis EP, Goldberg DM. Resveratrol: a molecule whose time has come? and gone?. Clin Biochem. 1997; 30: 91–113.

[10] Zhou HB, Chen JJ, Wang WX, Cai JT, Du Q. Anticancer activity of resveratrol on implanted human primary. World Gastroenterol. 2005; 11(2):280–284.

[11] Pace-Asciak CR, Hahn S, Diamandis EP, Soleas G, Goldberg DM. The red wine phenolics trans-resveratrol and quercetin block human platelet aggregation and eicosanoid synthesis implications for protection against coronary heart disease. Clin Chim Acta. 1995; 235(2):207–219.

[12] Albert S, Horbach R, Holger B, Bianka Rene C. Synthesis and antimicrobial activity of (E) stibene derivatives. Bioorganic & Medicinal Chemistry. 2011; 19: 5155-5166.

[13] Belluti F, Fontana G, Dal L, Carenini N, Giommarelli C. Design, synthesis and anticancer activities stilbene-coumarin hybrid compounds: identification of novel proapoptotic agents. Bioorganic & Medicinal Chemistry. 2010; 18: 3543-3550.

[14] Marek M, Norbert H, Thomas E, Karin P. Resveratrol analogues as selective cyclo oxygenase-2 inhibitors: synthesis and structure activity relationship. Bio organic and Medicinal chemistry. 2004; 12: 5571-5578.

[15] Ryoji K, Akhiro T, Harukazu F, Kunihiro N. Substituted (w-Amino alkoxy) stilbene Derivatives as a New class of Anti convulsants. J.Med.chem. 1984; 27: 645-649.

[16] Genevieve X, Sean W, Cameron P, Michael G, Ralph I, Erica A. The functional group on (E)-4,40–disubstituted stilbenes influences toxicity and antioxidative activity in differentiated PC-12 cells. Bioorganic & Medicinal Chemistry Letters. 2013; 13: 6355–6359.

[17] Ferretta A, Gaballo A , Tanzarella P, Piccoli C, Capitanio N, Nico B, Annese T, Di Paola M, Dell'aquila C, De Mari M, Ferranini E, Bonifati V, Pacelli C, Cocco T. Effect of resveratrol on mitochondrial function:

implications in parkin-associated familiar Parkinson's disease. Biochim Biophys Act. 2014; 7: 902–905.

[18] Orsini F, Francesca P, Luisella V, Talal A. Isolation, synthesis, and antiplatelet aggregation activity of resveratrol 3-O-β-D glucopyranoside and related compounds. J.Nat.prod. 1997; 60: 1082-87.

[19] Mark C, D Nagarathnam, D Gopal, Cnii M. Synthesis and Evaluation of Analogues of (Z)-1-(4-Methoxy phenyl)-2-(3,4,5-Trimethoxy phenyl) ethane as potential cytotoxic and Antimitotic Agents. J.Med.chem.1992; 35: 2293-2306.

[20] Venkanna, A, Siva B, Poornima B, Rao P. R, Vadaparthi K., Rajendra P, Ashok Reddy K, BhanuPrakash Reddy G, Suresh K. Phytochemical investigation of sesquiterpenes from the fruits of Schisandrachinensis and their cytotoxic activity. Fitoterapia. 2014; 95: 102–108.