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Original Research Article

Synthesis and Molecular Docking Studies of New Tetrazole-acetamide Derivatives as Anti-cancer Agent

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ABSTRACT

The chemistry of condensed heterocyclic molecules in terms of their diverse biological properties and role in drug development has been the subject of numerous publications. Tetrazole is a naturally occurring chemical that has been used to create several commercially available drugs and as a result, plays an important role in pharmaceutical chemistry. The current study aimed to create and synthesize seven new 2,5-disubstituted-tetrazole-acetamide derivatives 3a–3g via an *N*-alkylation reaction of 5-(4-bromophenyl)-2H-tetrazole or 5-(4-chlorophenyl)-2H-tetrazole 1a,1b, and *N*-acetamide derivatives 2a–2f, and 2g in CH_3CN using potassium carbonate as a base in good yields. New molecules were assigned based on nuclear magnetic resonance results (^1H, ^{13}C NMR, and two-dimensional-NMR [heteronuclear single quantum coherence spectroscopy [HSQC]), along with mass spectrometry (EI-MS) techniques. The products' biological activities were confirmed using the tetrazolium (MTT) assay against MCF-7 (breast cancer) and PC3 (prostate cancer) cells and their effects on the normal hepatic cell line, WRL68. Results showed that compounds 3e-3g inhibited PC3 cells with average IC_{50} values of 32.59, 54.99, and 55.53 µM, respectively. Compounds 3a and 3b demonstrated cytotoxicity against the MCF-7 cell line, with average IC_{50} values of 94.25 and 68.16 µM, respectively. Compounds 3a, 3c, and 3e-3g on the 3ERT and 3ZK6 receptors demonstrated strong binding capabilities and improved protein interactions according to molecular docking experiments.

Keywords: Acetamide, Anti-cancer, Cytotoxic, Molecular docking, Tetrazole.

Introduction

The importance of heterocyclic molecules in pharmacology has sparked an abundance of interest in the discipline in recent years.[1,2] Nitrogen-containing heterocyclic molecules seem to be extremely promising vectors in the fields of industrial chemistry, synthetic organic chemistry, and medicine.[3,4] Furthermore, society expects chemists to develop more sustainable and green chemical processes. Researchers in the fields of chemistry, pharmacology, and science have long been interested in tetrazole derivatives.[5,6] They also form an important category of N-heterocyclic molecules due to their distinctive molecular construction, which is a chemical counterpart of carboxy or cis-amido groups with considerable lipophilicity and prolonged, refractory metabolism.[7,8] These compounds have a wide range of pharmacological actions, including antihypertensive, antifungal, antituberculosis, antimalaria, antileishmanial, antidiabetic, and anticancer properties.[9–15] Additionally, substances from this class of heterocycles have shown promise in a variety of areas, such as materials science, drug development, coordination, organometallic, and organocatalytic chemistry.[16–18] Tetrazole products are widely used in synthetic applications as they serve as the starting materials for the synthesis of potent heterocycles in industries that produce propellants, medicines, and explosives. Tetrazole analogs were the first successful treatment involving the dopamine D2 receptor.[19]

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Tetrazole compounds are highly effective antibacterial and anticancer agents.[20, 21] Kondo et al.[22] combined phenyl sulfonyl hydrazones of aryl aldehydes with arene diazonium salts to produce 2,5-diaryl substituted tetrazoles. Our research aimed to study the formation of new 2,5-disubstituted-tetrazole-acetamide derivatives 3a–3g via an *N*-alkylation reaction of 5-substituted-2*H*-tetrazole with *N*-acetamide derivatives under conditions of basic catalysis, Furthermore, these derivatives were evaluated for their anticancer activities against MCF-7 and PC3 cells. In addition, a molecular docking study of the newly synthesized compounds was also performed using the Auto Dock 4.2 software.[23,24]

Materials and Methods

Cell culture

Iran's National Cell Bank obtained the cell lines, MCF-7, PC3, and WRL68. The cell culture was supplemented with antibiotics (100 U/mL penicillin and 100 µg/mL streptomycin) using Roswell Park Memorial Institute (RPMI)-1640 medium (Gibco). The cells were passaged using trypsin/ethylenediaminetetraacetic acid (EDTA; from Gibco) and phosphate-buffered saline (PBS) solutions. They were housed at 37 °C in humidified air containing 5% CO_2. The conditions and media for growing cells into three-dimensional (3D) colonies were identical to those for monolayer culture.

General procedure for the synthesis of tetrazole-acetamide derivatives (3a-3g)

The synthesis of tetrazole acetamides is outlined in a multistep synthesis pathway by subsequent well-known procedures.[25,26] First, 4-bromobenzonitrile or 4-chlorobenzonitrile (19.8 mmol) and NaN_3 (24 mmol) were reacted in 10 mL dimethyl fluoride (DMF) with ammonium chloride (23.6 mmol). After 5 hours of agitation at a temperature of 120 ºC, the reaction mixture was allowed to cool to the ambient temperature. The precipitate underwent filtration, followed by three washes with cold water, and subsequent drying. This process

resulted in the production of 5-(4-bromophenyl)-2H-tetrazole or 5-(4-chlorophenyl)-2H-tetrazole, denoted as 1a and 1b, respectively. In the second stage, p-substituted anilines or p-amino antipyrine (20 mmol) and triethyl amine (24 mmol) were reacted in $CHCl_3$ (20 mL). Afterward, a solution containing 24 millimoles of chloroacetyl chloride was slowly added to the reaction mixture at a temperature of 0 degrees Celsius. The reaction was then allowed to cool at room temperature for 20 h. The mixture was extracted, and the organic phase was washed with water and brine before being dried over anhydrous sodium sulfate, reduced under vacuum, and purified with an ethyl alcohol/water mixture to yield N-(p-substituted phenyl)-2-chloroacetamide or 2-chloro-N-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl) acetamide 2a–2g. The tetrazole-acetamide 3a–3g was prepared by substitution with 5-(4-bromophenyl)-2H-tetrazole or by addition of 5-(4-chlorophenyl)-2H-tetrazole 1a,1b (5 mmol) and *N*-acetamide derivatives 2a-2g (5 mmol) in CH_3CN (25 mL) using potassium carbonate (10 mmol) as a base. The reaction was mixed and refluxed for 24 h after which it was then cooled to room temperature. The precipitate was then filtered, followed by washing with ethyl alcohol, drying, and crystallizing from ethyl alcohol to produce 2-(5-(p-substituted phenyl)-2H-tetrazol-2-yl)-N-(p-substituted) acetamide and 2-(5-(4-substituted-phenyl)-1H-tetrazol-1-yl)-N-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl) acetamide.

Chemicals/Reagents

All chemicals were reagent grade. 4-bromobenzonitrile (99%), 4-chlorobenzonitrile (99%), sodium azide (99.5%), ammonium chloride (99.5%), 4-aminoantipyrine (97%), 4-methyl aniline (99.6%), 4-methoxy aniline (99%), 4-aminoacetophenone (99%), triethyl amine (99.5%), chloroacetyl chloride (98%), and potassium carbonate (99%), they were acquired from Sigma-Aldrich (Germany) and applied as is. Further reagents involving dimethyl fluoride (DMF) (99.90%), ethanol (98%), chloroform ($CHCl_3$) (98%), and acetonitrile (CH_3CN) (99%) were of analytical grade and were gotten from Fluka.

Instruments/Equipment

The melting point was determined using a thermo-scientific device at the University of Basrah, Iraq. Thin layer chromatography (TLC) plate silica gel is frequently used to track the progress of reactions. The nuclear magnetic resonance (NMR) spectra of ^{1}H-NMR at 400 MHz and ^{13}C-NMR at 100 MHz were acquired using a Bruker ARX-400 spectrometer at Basrah University in Iraq. Deuterated dimethyl sulfoxide (DMSO) was utilized as the internal solvent (^{1}H NMR: chemical shift of dimethyl sulfoxide-d_6: 2.5 ppm and water at 3.35 ppm; ^{13}C NMR: chemical shift of dimethyl sulfoxide-d_6: 39.52 ppm), and the standard used within was trimethylsilane (TMS). Agilent technology at 70 eV was used to generate mass spectra at Tehran University, Iran.

Cell feasibility/fast testing

The cytotoxic effect was measured using a tetrazolium (MTT) assay that was performed in 96-well plates.[27,28] The cell line was seeded at a density of 1 x 10^4 cells/well. MCF-7 cells (derived from breast cancer), PC3 cells (derived from prostate cancer), and WRL-68 cells (derived from normal human cells) were treated with trypsin to separate them, then gathered and adjusted to a density of 1.4 x 104 cells per well to create monolayer cultures. Uniform cells were placed into 96-well plates with 200 µl of fresh medium apiece and kept in a controlled environment for 24 hours. Following the formation of a monolayer, the cells were exposed to substances with concentrations varying from 25 to 400 µg/mL for a duration of 24 hours at a temperature of 37 °C in an environment containing 5% CO_2. Following a 24-hour treatment period, the liquid was extracted, and 200 µl/well of MTT solution (0.5 mg/mL in PBS) was introduced into each well. The monolayer culture survived undamaged in its original container. Subsequently, the dish was incubated at a temperature of 37 °C for a duration of 4 hours. After the cell supernatant was removed, 100 µL of DMSO was added to the MTT solution in each well. Cells were grown on a shaking apparatus at 37 °C until crystals dissolved completely. Cell viability was assessed using an enzyme-linked immunosorbent assay (ELISA) reader (Model Wave XS2, BioTek, USA) at 620 nm. The percentage cell growth inhibition (PCTI) was calculated using the equation shown below (1):

$$(PR) = \frac{B}{A} \times 100 \quad \text{..... (Eqn. 1)}$$

Where B is the optical density of treated wells, and A is the average optical density of untreated wells; IR = 100 − PR.

Statistical analysis

To depict the experimental values, three different studies' averages and standard deviations (SD) were used. Statistical significance was determined by $p < 0.0001$ based on the analysis of variance (ANOVA) performed with the GraphPad Prism 7 (2016).

Docking parameters and software

The docking parameters of compounds were calculated using Auto Dock4.2 (AD4.2), and the Biovia Discovery Studio visualizer was used to analyze the results.

Results and Discussion

Derivatives of tetrazole-acetamide are becoming increasingly significant for use in modern medicine, especially in cancer treatment. Syntheses of the intermediate and target compounds were performed according to Fig 1. Some physicochemical parameters for the tetrazole-acetamide products 3a–3g are given in Table 1. The starting 5-substituted tetrazoles 1a,1b, and N-(p-substituted phenyl)-2-chloroacetamide 2a–2g were obtained according to the described literature methods.[25,26] Good yields of substrates 3a–3g were obtained via the *N*-alkylation reaction of 5-(4-bromophenyl)-2H-tetrazole or 5-(4-chlorophenyl)-2H-tetrazole 1a,1b, and N-acetamide derivatives 2a–2g in CH_3CN using potassium carbonate as a base and refluxed for 24 h to yield 2-(5-(p-substituted phenyl)-2H-tetrazol-2-yl)-N-(p-substituted) acetamide and 2-(5-(4-substituted phenyl)-1H-tetrazol-1-yl)-N-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl) acetamide in good yield. The times, yields, and molecular formulas are summarized in Table 1. The chemical structures of all the resulting tetrazole-acetamide analogs were validated using NMR and mass spectrometry.

Tetrazole-acetamide compounds 3a–3g were used for obtaining ^{1}H NMR spectra. The singlet signals at 10.67, 10.58, 10.53, 10.52, 11.0, and 9.87 ppm belong to the NH group with the remaining protons observed at expected points. Likewise, the ^{13}C NMR analysis of compounds 3a–3g exhibited signals at 163.1, 163.0, 163.8, 163.9, and 161.8 ppm, according to the C=N group, and signals at 163.8, 163.9, 164.0, 164.0, 164.0, 164.0, and 164.2 ppm refer to the carbonyl group (C=O). Two-dimensional NMR analysis reflected by the heteronuclear single quantum coherence spectroscopy (^{1}H-^{13}C HSQC) spectrum of chemical 3a revealed a correlation among the signals at δ 2.26 (3H, s) ppm and the CH signal at δ 20.9 ppm in addition to the signal at 5.77 (2H, s) ppm and the CH signal at 55.9 ppm.

Tetrazole–acetamide derivatives 3a–3g showed mass spectra with a molecular ion (m/z): 325 [M-2H+], 370 [M-H+], 346 [M+3H+], 388 [M+H+], 354 [M-H+], 399 [M+], and 422 [M-H+], which matched the likely values of m/z 327, 371, 343, 387, 355, 399, and 423.

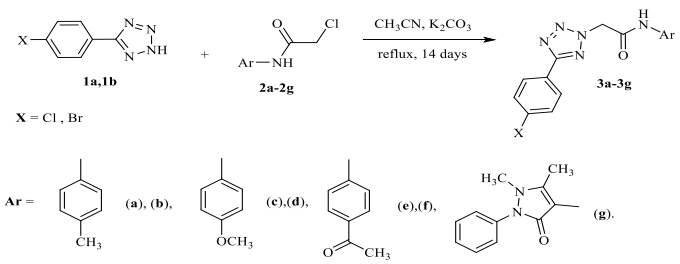

Figure 1: Synthetic route of target tetrazole-acetamide compounds 3a-3g.

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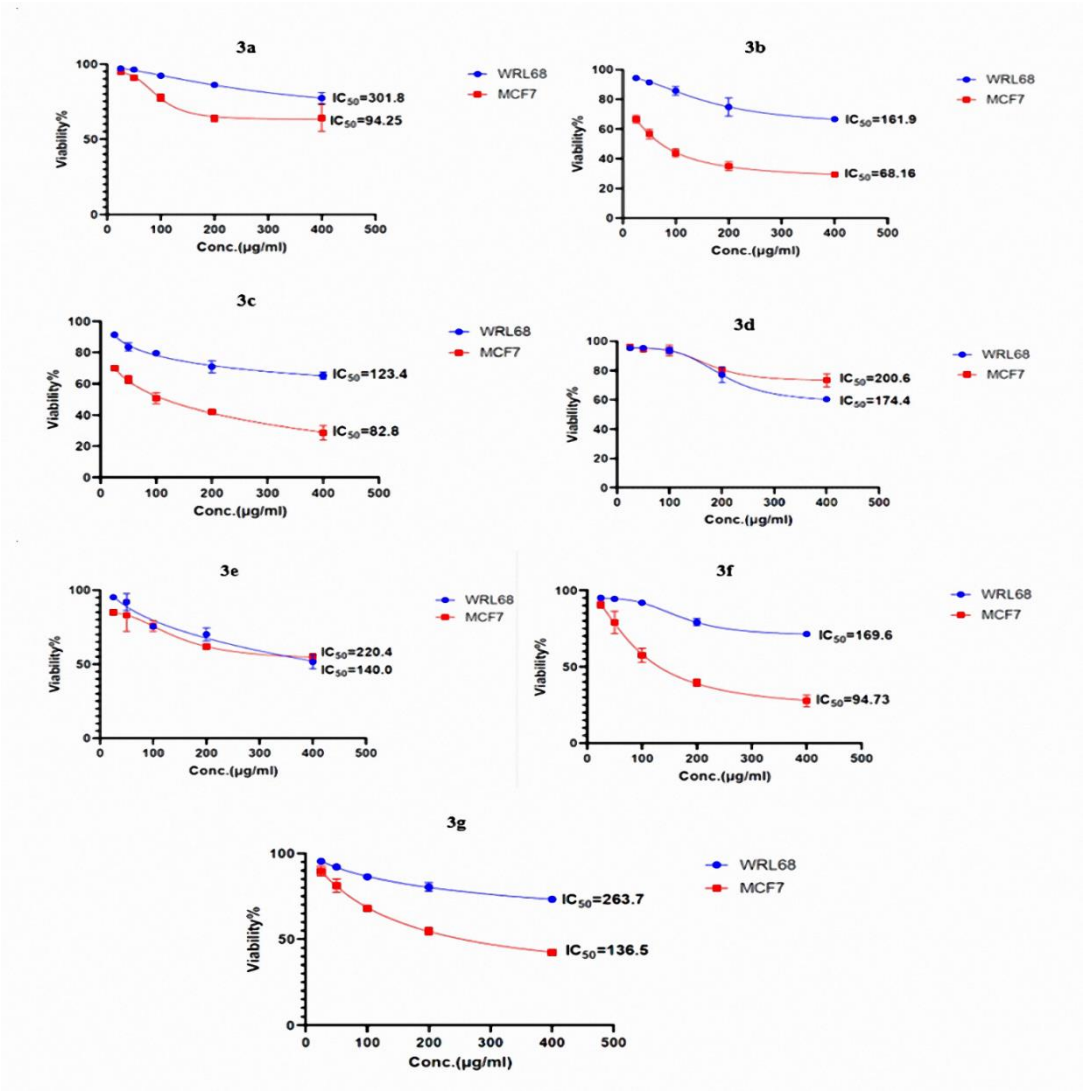

Figure 2: The activity of compounds against MCF-7 and WRL-68 normal cells.

Table 1: The physical properties of tetrazole-acetamide compounds 3a-3g.

Compounds	Molecular formula	M.wt g/mol	Color	Time, day	mp, °C	Yield, %
3a	$C_{16}H_{14}ClN_5O$	327.77	White	14	240-242	85
3b	$C_{16}H_{14}BrN_5O$	372.23	White	14	243-245	75
3c	$C_{16}H_{14}ClN_5O_2$	343.77	White	14	236-238	75
3d	$C_{16}H_{14}BrN_5O_2$	388.23	White	14	242-244	65
3e	$C_{17}H_{14}ClN_5O_2$	355.78	White	14	239-240	75
3f	$C_{17}H_{14}BrN_5O_2$	400.24	White	14	241-243	77
3g	$C_{20}H_{18}ClN_7O_2$	423.86	White	14	262-264	66

Characteristics of the prepared compounds

2-(5-(4-chlorophenyl)-2H-tetrazol-2-yl)-N-(p-tolyl) acetamide (3a)

The compound is a white color with a yield of 85% at mp 240-242°C. ^{1}H NMR (DMSO-d_6): H_{23} (3H, δ = 2.26 ppm), H_{13} (2H, δ = 5.77 ppm), $H_{19,21}$ (2H, δ = 7.15 ppm, *J* = 8.2 Hz), $H_{18,22}$ (2H, δ =7.49 ppm, *J* = 8.2 Hz), $H_{1,5}$ (2H, δ =7.66 ppm, *J* = 8.4 Hz), $H_{2,4}$ (2H, δ = 8.10 ppm, *J* = 8.4 Hz), H_{15} (1H, δ =10.67 ppm). ^{13}C -NMR (DMSO-d_6): C_{23} (δ= 20.9 ppm), C_{13} (δ=55.9 ppm), $C_{18,22}$ (δ=119.8 ppm), C_6 (δ=126.1 ppm), $C_{1,5}$ (δ=128.6 ppm), $C_{2,4}$ (δ=129.7 ppm), $C_{19,21}$ (δ =129.9 ppm), C_{20} (δ =133.5 ppm), C_3 (δ =135.8 ppm), C_{17} (δ =136.2 ppm), C_8 (δ =163.1 ppm), C_{14} (δ =163.8 ppm). MS (ESI), m/z = 325 [M-2H$^+$].

2-(5-(4-bromophenyl)-2H-tetrazol-2-yl)-N-(p-tolyl) acetamide (3b)

The compound is a white color with a yield of 75% at mp 243-245 °C. ^{1}H NMR (DMSO-d_6): H_{23} (3H, δ= 2.26 ppm), H_{13} (2H, δ= 5.76 ppm), $H_{19,21}$ (2H, δ = 7.15 ppm, *J* = 8.2 Hz), $C_{18,22}$ (2H, δ =7.47 ppm), $H_{2,4}$ (2H, δ =7.80 ppm), $H_{1,5}$ (2H, δ =8.03 ppm), H_{15} (1H, δ =10.58 ppm). ^{13}C NMR (DMSO-d_6): C_{23} (δ= 20.9 ppm), C_{13} (δ=55.9 ppm), $C_{18,22}$ (δ=119.8 ppm), C_3 (δ=124.6 ppm), C_6 (δ=126.4 ppm), $C_{19,21}$ (δ =128.8 ppm), C_{20} (δ=129.8 ppm), $C_{1,5}$ (δ=132.9 ppm), $C_{2,4}$ (δ=133.5 ppm), C_{17}

(δ=136.1 ppm), C_8 (δ=163.0 ppm), C_{14} (δ=163.9 ppm). MS (ESI), m/z = 370 [M-H$^+$].

2-(5-(4-chlorophenyl)-2H-tetrazol-2-yl)-N-(4-methoxyphenyl) acetamide (3c)

The compound is a white color with a yield of 75% at mp 236-238 °C. ^{1}H NMR (DMSO-*d*$_6$): H_{24} (3H, δ= 3.73 ppm), H_{13} (2H, δ= 5.75 ppm), $H_{19,21}$ (2H, δ = 6.92 ppm, *J* = 9.0 Hz), $H_{18,22}$ (2H, δ =7.51 ppm, *J* = 9.0 Hz), $H_{2,4}$ (2H, δ = 7.66 ppm, *J* = 8.6 Hz), $H_{1,5}$ (2H, δ =8.10 ppm, *J*= 8.6 Hz), H_{15} (1H, δ =10.53 ppm). ^{13}C NMR (DMSO-*d*$_6$): C_{24} (δ= 55.7 ppm), $C_{19,21}$ (δ =114.3 ppm), $C_{18,22}$ (δ =121.3 ppm), C_6 (δ= 126.1 ppm), $C_{1,5}$ (δ =128.6 ppm), $C_{2,4}$ (δ =129.9 ppm), C_{17} (δ =131.7 ppm), C3 (δ =135.8 ppm), C_{20} (δ =156.2 ppm), C_8 (δ =162.8 ppm), C_{14} (δ =163.8 ppm). MS (ESI), m/z = 346 [M+3H$^+$].

2-(5-(4-bromophenyl)-2H-tetrazol-2-yl)-N-(4-methoxyphenyl) acetamide (3d)

The compound is a white color with a yield of 65% at mp 242-244 °C. ^{1}H NMR (DMSO-*d*$_6$): H_{24} (3H, δ= 3.73 ppm), H_{13} (2H, δ= 5.74 ppm), $H_{19,21}$ (2H, δ = 6.92 ppm, *J* = 9.0 Hz), $H_{18,22}$ (2H, δ =7.51 ppm, *J* = 9.0 Hz), $H_{2,4}$ (2H, δ = 7.80 ppm, *J* = 8.6 Hz), $H_{1,5}$ (2H, δ =8.03 ppm, *J*= 8.6 Hz), H_{15} (1H, δ =10.52 ppm). ^{13}C NMR (DMSO-*d*$_6$): C_{13} (δ= 55.7 ppm), C_{24} (δ= 55.8 ppm), $C_{19,21}$ (δ =114.5 ppm), $C_{18,22}$ (δ =121.4 ppm), C3 (δ =124.6 ppm), C_6 (δ= 126.5 ppm), $C_{1,5}$ (δ =128.8 ppm), $C_{2,4}$ (δ =131.7 ppm), C_{17} (δ =132.9 ppm), C_{20} (δ =156.2 ppm), C_8 (δ =162.8 ppm), C_{14} (δ =163.9 ppm). MS (ESI), m/z = 388 [M+H$^+$].

N-(4-acetylphenyl)-2-(5-(4-chlorophenyl)-2H-tetrazol-2-yl) acetamide (3e)

The compound is a white color with a yield of 75% at mp 239-240 °C. ^{1}H NMR (DMSO-*d*$_6$): H_{25} (3H, δ= 2.54 ppm), H_{13} (2H, δ= 5.85 ppm), $H_{18,22,2,4}$ (4H, δ =7.70 ppm), $H_{19,21,1,5}$ (4H, δ =8.04), H_{15} (1H, δ =11.0). ^{13}C NMR (DMSO-*d*$_6$): C_{25} (δ= 26.9 ppm), C_{13} (δ= 55.9 ppm), $C_{18,22}$ (δ =118.9 ppm), C_6 (δ= 126.0 ppm), $C_{1,5}$ (δ =128.6 ppm), $C_{2,4}$ (δ =130.0 ppm), $C_{19,21}$ (δ =130.1 ppm), C_{20} (δ=130.8 ppm), C_3 (δ=135.8 ppm), C_{17} (δ =142.8 ppm), C_8 (δ =163.8 ppm), C_{14} (δ =164.0 ppm), C_{23} (δ =197.1 ppm). MS (ESI), m/z = 354 [M-H$^+$].

N-(4-acetylphenyl)-2-(5-(4-bromophenyl)-2H-tetrazol-2-yl) acetamide (3f)

The compound is a white color with a yield of 77% at mp 241-243 °C. ^{1}H NMR (DMSO-*d*$_6$): H_{25} (3H, δ= 2.54 ppm), H_{13} (2H, δ= 5.85 ppm), $H_{2,4,18,22}$ (4H, δ =7.77 ppm), $H_{1,5,19,21}$ (4H, δ =8.01), H_{15} (1H, δ =11.0). ^{13}C NMR (DMSO-*d*$_6$): C_{25} (δ= 26.9 ppm), C_{13} (δ= 55.9 ppm), $C_{18,22}$ (δ =119.2 ppm), C_3 (δ=124.6 ppm), C_6 (δ= 126.4 ppm), $C_{19,21}$ (δ =128.8 ppm), $C_{1,5}$ (δ =130.1 ppm), C_{20} (δ=132.8 ppm), $C_{2,4}$ (δ =132.9 ppm), C_{17} (δ =142.8 ppm), C_8 (δ =163.9 ppm), C_{14} (δ =164.0 ppm), C_{23} (δ =197.1 ppm). MS (ESI), m/z = 399 [M$^+$].

2-(5-(4-chlorophenyl)-2H-tetrazol-2-yl)-N-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl) acetamide (3g)

The compound is a white color with a yield of 66% at mp 262-264 °C. ^{1}H NMR (DMSO-*d*$_6$): H_6 (3H, δ= 2.15 ppm), H_7 (3H, δ= 3.07 ppm), H_{17} (2H, δ= 5.77 ppm), $H_{10,12,14}$ (3H, δ =7.35 ppm), $H_{11,13}$ (2H, δ =7.51 ppm), $H_{26,28}$ (2H, δ =7.66 ppm, *J* = 8.6 Hz), $H_{25,29}$ (2H, δ =8.01 ppm, *J* = 8.6 Hz), H_{15} (1H, δ =9.87). ^{13}C NMR (DMSO-*d*$_6$): C_6 (δ= 11.7 ppm), C_7 (δ= 36.3 ppm), C_{17} (δ= 55.2 ppm), C_2 (δ= 106.7 ppm), $C_{10,14}$ (δ =124.2 ppm), C_{12} (δ= 125.1 ppm), C_{24} (δ= 126.9 ppm), $C_{25,29}$ (δ =128.6 ppm), $C_{11,13}$ (δ =129.6 ppm), $C_{26,28}$ (δ =129.7 ppm), C_8 (δ= 135.3 ppm), C_{27} (δ= 135.8 ppm), C_3 (δ= 152.5 ppm), C_1 (δ= 161.8 ppm), C_{22} (δ= 163.8 ppm), C_{16} (δ= 164.2 ppm). MS (ESI), m/z = 422 [M-H$^+$].

Biological study

A new series of tetrazole–acetamide 3a–3g was tested for *in vitro* anticancer activities against MCF-7 and PC3 cells within 24 h and compared with normal WRL-68 hepatic cells. Compounds 3b, 3c, and 3g were found to be active and nontoxic in normal cell lines. On the other hand, compounds 3a and 3e–3g demonstrated rapid critical activity on the PC3 cell line, while the WRL-68 cell line exhibited only moderate susceptibility (Table 2). According to the MTT assay results, the synthesized compounds exhibited cytotoxic action against cancer cells at different doses. These results show that these substances can be experts in the fight against cancer because of their antitumor effects.[29–31] These compounds significantly inhibited the proliferation of MCF-7 and PC3 cells, whereas their effects on WRL-68 were mildly cytotoxic (Figures 1, 2), which summarises the appropriate IC_{50} values.

Molecular docking study

Our investigation showed considerable anticancer activities when docked with the PC3 (Pdb: 3ZK6) and MCF-7 (Pdb: 3ERT) [32, 33] proteins. The docking investigations were carried out with the Auto Dock 4.2 program [34], and the data were analyzed using the Biovia Discovery Studio visualizer. Compound 3a docked well with the 3ERT receptor site, creating a hydrogen bond with the amino acid ARG A:394 at 2.04 Å. Other p-p interactions with amino acid residues include HIE A:524, MET A:421, ILE A:424, LEU A:525, LEU A:346, LEU A:349, PHE A:404, LEU A:408, ALA A:350, GLU A:353, LEU A:391, LEU A:384, MET A:388, LEU A:387, LEU A:428, and PHE A:425. Compound 3c formed strong hydrogen bonds with three distinct residues: (1) THR A:347 (2.18 Å), (2) LEU A:346 (2.11 Å), and (3) ARG A:394 (2.09 Å). Other interactions with amino acids include ASP A:351, LEU A:525, CYS A:530, TRP A:383, ALA A:350, LEU A:387, GLU A:353, MET A:343, PHE A:404, LEU A:349, and LEU A:391. 3g also demonstrated very good binding energy by creating two hydrogen bonds with two distinct residues. LEU A:130 (distance: 2.19 Å) and SER A:145 (distance: 2.08 Å) interact with PHE A:146, THR A:109, LEU A:108, ASP A:107, GLU A:129, ARG A:139, PHE A:131, ASP A:133, ARG A:132, ARG A:102, PHE A:97, ALA A:149, ALA A:142, and PHE A:105 amino acid residues (Table 3, Figure 3). On the other hand, compounds 3a, 3c, and 3g had moderate to very good binding energy. Compound 3g showed a favorable interaction with LEU A:536, LEU A:354, TRP A:383, ASP A:351, LEU A:391, ALA A:350, LEU A:346, MET A:421, LEU A:346, LEU A:525, and MET A:343 amino acid residues. On the other hand, compound 3f interacted well with amino acid residues GLY A: 138, ALA A: 149, PHE A: 146, SER A: 145, GLU A: 98, ALA A: 142, ARG A: 102, ARG A: 139, LEU A: 108, PHE A: 105, PHE A:97, LEU A: 130, GLU A: 129, and SER A:106. In contrast, compound 3e formed one hydrogen bond with amino residue ARG A:102 (2.34 Å), and had contact with amino acid residues GLY A:138, PHE A105, ARG A:139, ALA A:142, PHE A:146, ALA A:149, SER A:145, PHE A:97, LEU A:130, and LEU A:108 (Table 4, Figure 4). Overall, the compounds evaluated with the target receptors demonstrated good docking scores due to hydrogen and p–p interactions with various residues. These findings could lead to beneficial effects on the apoptotic cell death process through activation of the apoptotic pathway, perhaps leading to the identification of new medications. [35,36]

Table 2: Cytotoxic effects of tested compounds against PC3, MCF-7, and WRL-68 cell lines.

Compounds	$IC_{50\ MCF-7}$	$IC_{50\ WRL68}$	$IC_{50\ PC3}$	$IC_{50\ WRL68}$
3a	94.25	301.8	112.0	301.8
3b	68.16	161.9	203.0	161.9
3c	82.8	123.4	74.8	123.4
3d	174.4	200.6	164.3	200.6
3e	140.0	220.4	32.59	220.4
3f	94.73	169.6	54.99	169.6
3g	136.5	263.7	55.53	263.7

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Table 3: Parameters of the compounds with the best conformer with the 3ERT receptor.

Compounds	B.E Kcal/mol	Ki uM	RMSD (A)
3a	-8.83	336.11	3.034
3c	-7.55	2910	4.797
3g	-10.06	42.34	3.266

Table 4: Parameters of the compounds with the best conformer with the 3ZK6 receptor.

Compounds	B.E Kcal/mol	Ki uM	RMSD (A)
3e	-9.12	208.10	2.143
3f	-9.19	183.78	2.299
3g	-9.71	75.70	2.176

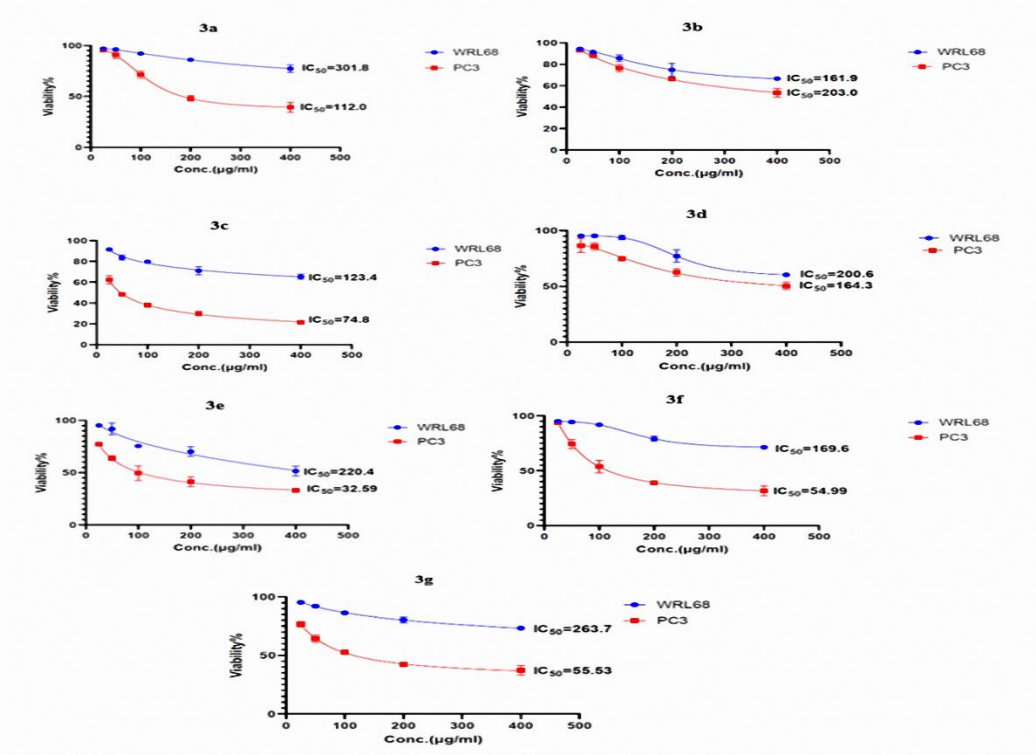

Figure 3: Cytotoxicity of compounds with PC3 and WRL-68 normal cell

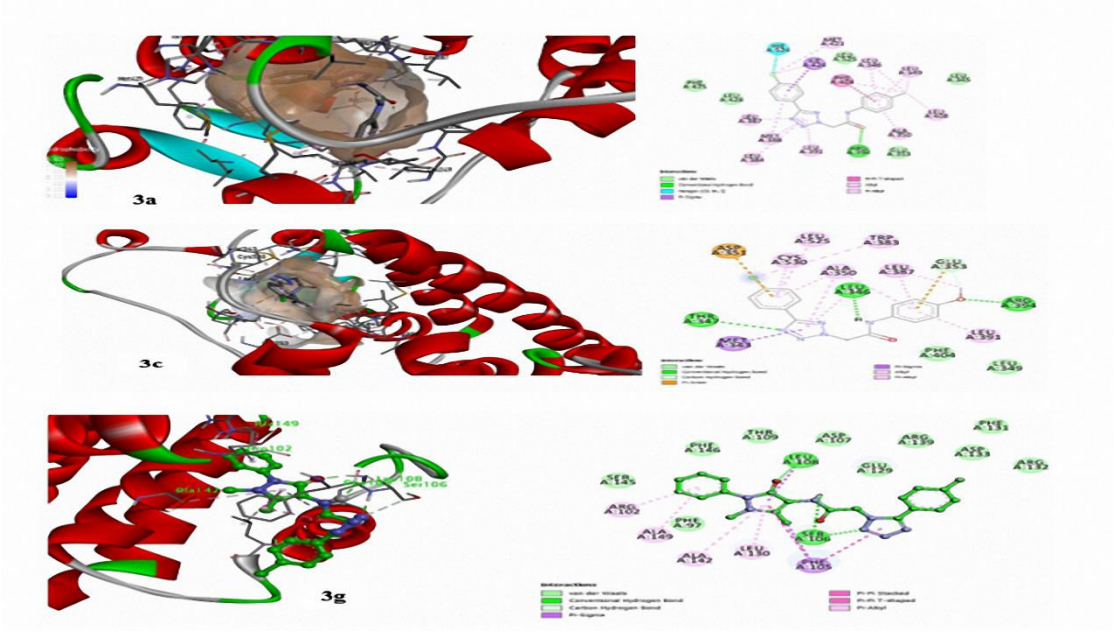

Figure 4: The protein-ligand interaction of the potent compounds 3a, 3c, and 3g against the 3ERT receptor

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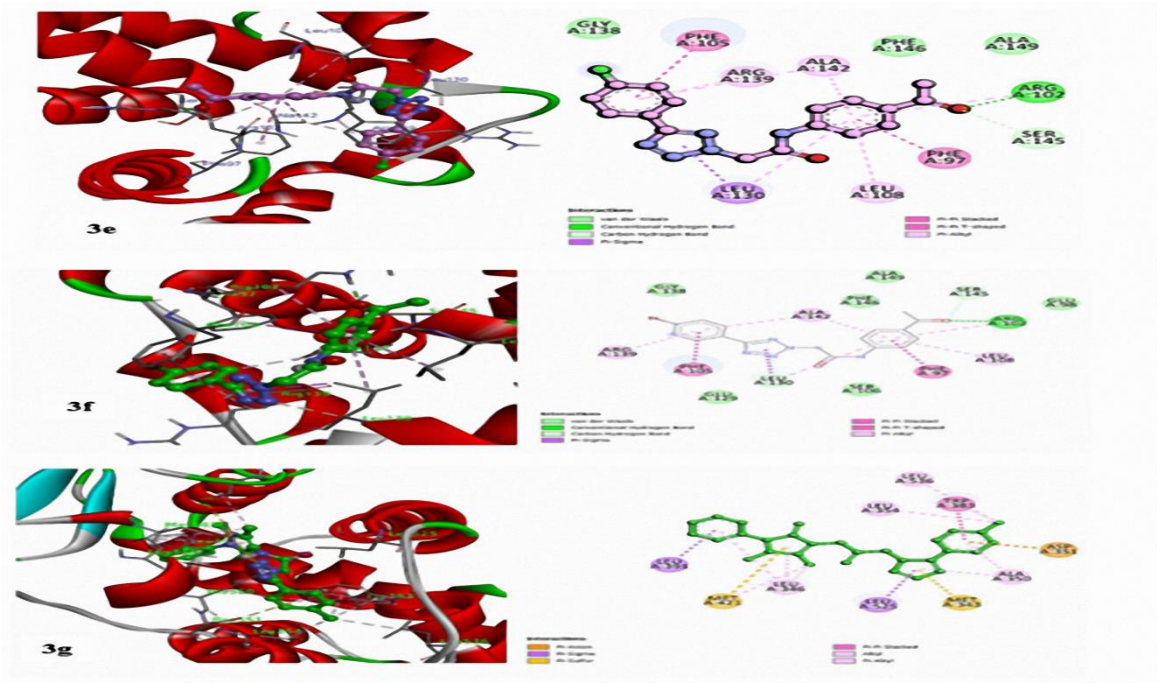

Figure 5: The protein-ligand interaction of the potent compounds 3e, 3f, and 3g against the 3ZK6 receptors.

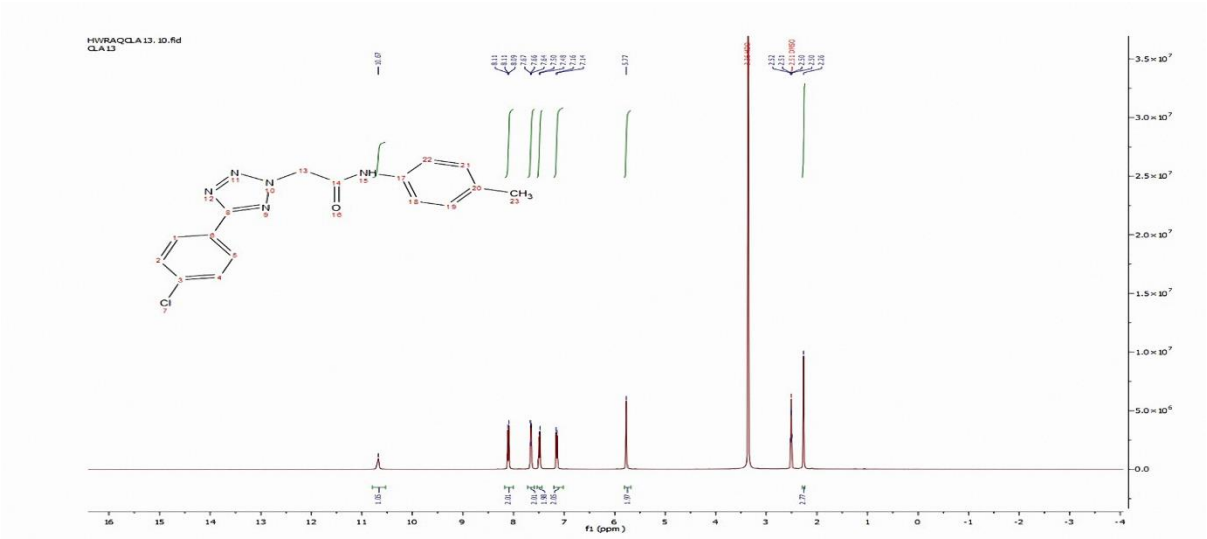

Figure 6: ^{1}H-NMR spectrum of compound 3a.

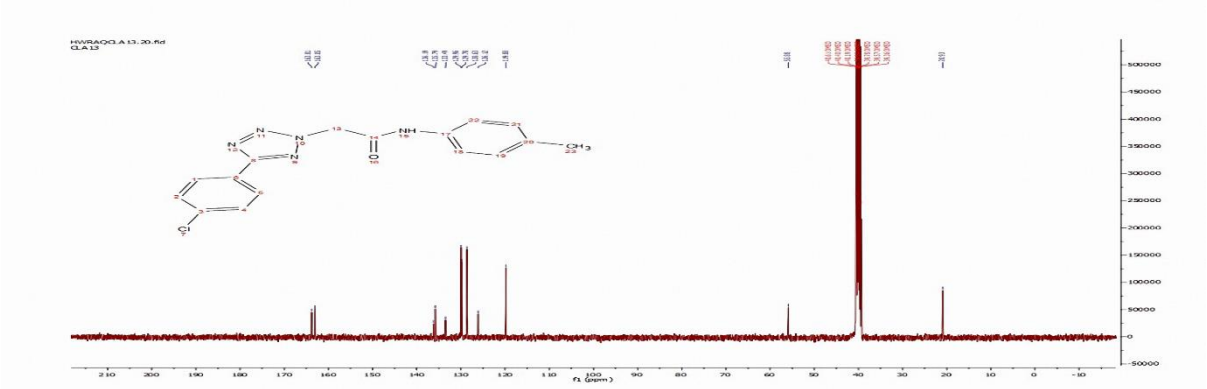

Figure 7: ^{13}C-NMR spectrum of compound 3a.

Conclusion

Dracaena spicata Roxb. is a potential source of polyphenol-like phytoconstituents and antioxidants of natural origin. The notable bioactivity of the plant also makes it a potential source of active metabolites that could be used as lead molecules for the drug development process. However, research should be conducted focusing on the isolation of more bioactive components of the plant responsible for its bioactivities.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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