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Design, synthesis, spectral analysis and molecular docking studies of some cyclic imide as potential anti breast and cervical cancer agents

Maha Abdul Razaq Essa
Department of Chemistry, College of Education for Pure Sciences, University of Basrah, Basrah 61001, Iraq

Adil Muala Dhumad
Department of Chemistry, College of Education for Pure Sciences, University of Basrah, Basrah 61001, Iraq
*Corresponding author email: adil.dhumad@uobasrah.edu.iq

Abstract---A number of cyclic imide derivatives were prepared in this study, using Diels- Alder reaction as a first step between maleic anhydride and anthracene. Use the reaction product as a reactant in the second step with different amines. The prepared cyclic imides were characterized by following the known spectroscopic methods to prove the proposed chemical composition of each of them using Mass, FT-IR, ^{1}HNMR and ^{13}CNMR. The biological activity of the prepared cyclic imides was tested theoretically using molecular docking to prove their efficacy as candidate inhibitors of breast and cervical cancer. The efficacy of prepared cyclic imides as inhibitors and antagonists of breast and cervical cancer, a practical candidate using the MTT assay to measure cellular metabolic activity as an indicator of cell viability, proliferation and cytotoxicity in MCF-7 and HeLa cell lines. Cytotoxic effects were in agreement with the molecular docking calculations, as the prepared cyclic imide derivatives showed good activity in inhibiting breast and cervical cancer cells. The results of MTT assay and molecular docking scores proved that the prepared (11R,15S)-13-(3-nitrophenyl)-9,10-dihydro-9,10-[3,4]epipyrrolo-anthracene-12,14-di one derivative a3 is considered as a potential candidate inhibitor for cervical cancer.

Keywords---cyclic imide, docking, breast cancer, cervical cancer, MCF-7, HeLa, DFT.

Introduction

Cyclic imides are commonly known for their structures in natural and laboratory prepared molecules with distinct pharmacological properties. It is also widely used for more functions through various chemical transformations [1-3]. Currently, worldwide renewed focus has been devoted to this important class of compounds for its potential new applications especially in medicinal and pharmaceutical chemistry as well as in drug discovery. Thus, the preparation of new cyclic imides or new methods of preparation has attracted great interest [4,5]. Several innovative methodologies have recently been developed in order to access this class of compounds and study their applications[6-8]. The cyclic imides are an important class of chemical compounds with wide applications in many fields if they are characterized by their widespread uses in therapeutic activities[9]. The building blocks are distinguished in the preparation of natural products, agricultural chemicals, pharmaceutical drugs and polymers [10-12].

Several studies demonstrated the biological efficacy of the prepared cyclic imides. The activity of cyclic imides was tested on many bacteria[13] and fungi[14], and it was also tested in inhibiting cancer cells on different lines[15-17]. In this study, a group of cyclic imides was prepared by Diels-Alder reaction [18]between maleic anhydride and anthracene, and the product was reacted with a five amines. Molecular docking calculations of the prepared compounds were performed to test their efficacy as reliable inhibitors of breast and cervical cancer. Cytotoxicity assay were carried out for the prepared cyclic imids, MTT assay with MCF-7 and HeLa [19,20]human breast and cervical, respectively cancer cells line were used to measure the activity of the prepared compounds to inhibit live breast and cervical human cancer cells.

Experimental

Apparatus

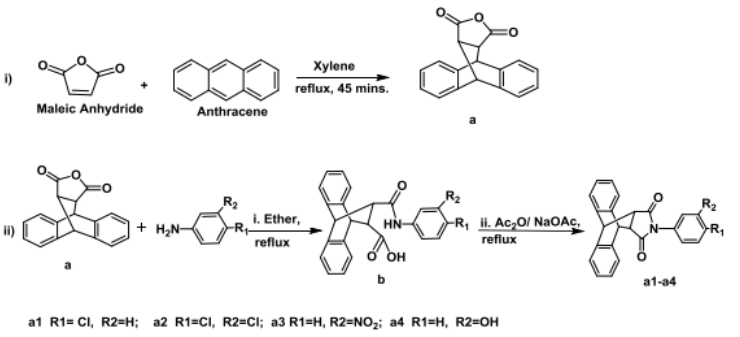
The FT-IR spectra was recorded on Shimadzu FT-IR 8400S spectrometer-Japan in the range 4000-500 cm^{-1} using KBr disc. ^{1}HNMR and ^{13}C NMR spectra were recorded on Bruker 400MHz spectrometer using Chloroform-d as a solvent and TMS as an internal reference. The mass spectrum was recorded on Agilent Technologies-5975C (EI, 70 eV).

Materials and Methods

Starting materials were obtained from commercial suppliers and used without further purification. The open glass capillaries on a Fisher–Johns melting point apparatus was used to determine the melting point and it was uncorrected. The reaction was monitored by Thin-layer chromatography (TLC) on Silica Gel 60 F254 plates (VWR, Darmstadt); visualization by UV detection at 254 nm.

Synthesis

The reaction path shown in Scheme 1 was followed to synthesis cyclic imides derivatives a1-a4



Scheme 1. The reaction path of synthesis cyclic imides derivatives a1-a4

Synthesis of (11R,15S)-13-(4-chlorophenyl)-9,10-dihydro-9,10-[3,4]epipyrrol oanthracene-12,14-dione a1

A mixture of (0.5gm, 0.005mole) maleic anhydride and (1gm, 0.005mole) anthracene in 15ml of xylene was refluxed for 45 mins in a three nick round bottom flask on a sand bath. White crystalline product of adduct a was obtained on cooling. To a stirred solution of (0.018 mole, 0.552g) adduct a (which was prepared in the previous step) in 20 ml ether in round bottom flask, a solution of (0.018 mole, 0.2286 g) of p-chloroaniline was added portion wise with stirring up to 1 hours. The product was filtered, washed with ether and dried, giving pale yellow crystals of amic acid b. Mix the (0.001 mol, 0.385g) of b (which was prepared in the previous step) with 10 mL of acetic anhydride and (0.001 mol, 0.082 g) of sodium acetate in a round bottom flask, stirring the mixture with reflux for 1 hour, cooled to RT, then added the iced distilled water to it. The contents left for 2 hour, the precipitate was formed. collected by filtration, and re-crystallized from ethanol to give a yellow powder. yield of 68%, M.W. 385.85, m. p 207-209 ^{O}C.

FT-IR(KBr) υ(cm^{-1}): 3074, 3018 (υ Ar-H), 2972 (υ C-H), 1774, 1705 (υ C=O), 1580 (υ C=C), 1388 (υ C-N), 1227 (υ C-O). 1H NMR (400 MHz, Chloroform-d) 7.33 (dd, J = 5.4, 3.2 Hz, 1H, Ar-H), 7.30 – 7.21 (m, 1H, Ar-H), , 6.50 – 6.42 (m, 1H, Ar-H), 4.88 (t, J = 1.8 Hz, 1H, Ar- CH-CH), 3.37 (t, J = 1.7. ^{13}C NMR (101 MHz, Chloroform-d) δ 175.86 (C=O), 141(C-Cl), 138.71(Ar(C=C)), 134.68(C-N),129.82, 129.32, 127.77, 127.70, 127.21, 125.22, 125.14, 124.39, 124.06 Ar(C-C), 48.01, 47.05 (C-CO), 45.88, 45.44(Ar-CH-Ar).MS (EI, m/z (%)): M+[C24H16ClNO2], 385.85 (10.5%), 161 (100%).

Synthesis of (11R,15S)-13-(3,4-dichlorophenyl)-9,10-dihydro -9,10-[3,4]epi-pyrroloanthracene-12,14-dione a2

To a stirred solution of (0.018 mole, 0.55g) adduct a in 20 ml ether in round

bottom flask, a solution of (0.002 mole, 0.29 g) of dichloroaniline was added portion wise with stirring up to 2 hours. The product was filtered, washed with ether and dried, giving pale yellow crystals of amic acid b. Then Mix the (0.001 mole, 0.385g) of b with 10 mL of acetic anhydride and (0.001 mole, 0.082 g) of sodium acetate, reflux for 1 hour, cooled to RT, yellow powder. yield of 65%, M.W. 420.29, m. p. 208-210 oC. FT-IR(KBr) υ(cm^{-1}): 3109, 3049 (υ Ar-H), 2972 (υ C-H), 1774, 1703 (υ C=O), 1589 (υ C=C), 1388 (υ C-N), 1255 (υ C-O). 1H NMR (400 MHz, Chloroform-d) 7.33 (dd, J = 5.4, 3.2 Hz, 1H, Ar-H), 7.30 – 7.21 (m, 1H, Ar-H), 6.50– 6.42 (m, 1H, Ar-H), 4.88 (t, J = 1.8 Hz, 1H, Ar- CH-CH), 3.37 (t, J = 1.7. 13C NMR (101 MHz, Chloroform-d) δ δ 175.86 (C=O), 141(C-Cl), 138.71(Ar(C=C)), 134.68(C- N),129.82, 129.32, 127.77, 127.70, 127.21, 125.22, 125.14, 124.39, 124.06 Ar(C-C), 48.01, 47.05 (C-CO), 45.88, 45.44(Ar-CH-Ar). (CH-CH). MS (EI, m/z (%)): M+[C24H15Cl2NO2], 419.2 (5.4%), 178.3 (100%).

Synthesis of (11R,15S)-13-(3-nitrophenyl)-9,10-dihydro-9,10-[3,4]epipyrroloanthracene-12,14-dione a3

To a stirred solution of (0.005 mole, 0.1381g) adduct a with (0.005 mole, 0.0693 g) of 3-nitroaniline was added portion wise with stirring up to 2 hours. The product was filtered, washed with ether and dried, giving pale yellow crystals of amic acid b. Then (0.013801 mol, 0.385g) of b with 10 mL of acetic anhydride and (0.001 mol, 0.082 g) of sodium acetate mixture with reflux for 1 hour, to give a yellow powder. yield of 66%, M.W. 396.40, m. p. 201-203 ^{o}C. FT-IR(KBr) υ(cm^{-1}): 3072, 3043 (υ Ar- H), 2978, 2960 (υ C-H), 1741, 1710 (υ C=O), 1620 (υ C=C), 1388 (υ C-N), 1238 (υ C-O). ^{1}H NMR (400 MHz, Chloroform-d) δ 8.14 (m, *J* = 8.4, 1H, Ar-H), 7.51 – 6.99 (m, 2H, Ar-H), 6.92 (m, 1H, Ar-H), 4.82 (t, *J* = 1.8 Hz, 2H, Ar-CH CH), 3.56 – 3.46 (m, 2H, CH-CO),. ^{13}C NMR (101 MHz, Chloroform-d) δ 170.46 (C=O), 140.65(C- NO2), 138.12(Ar(C=C)), 134.68(C-N), 127.76, 127.16, 127.21, 125.22, 125.41, 124.41, Ar(C-C), 48.24, 48.01 (C-CO), 45.44(Ar-CH-Ar).. MS (EI, m/z (%)): M+[$C_{24}H_{16}N_2O_4$], 396.40 (1%), 80.1 (100%), 178.2(73%).

Synthesis of (11R,15S)-13-(4-hydroxyphenyl)-9,10-dihydro-9,10-[3,4]epipyrroloanthracene-12,14-dione a4

(0.018 mole, 0.2762g) adduct a with (0.018 mole, 0.109 g) of p-hydroxyaniline was added portion wise with stirring up to 2 hours. to giving pale yellow crystals of amic acid b. The (0.001 mol, 0.385g) of b with 10 mL of acetic anhydride and (0.001 mol, 0.082 g) of sodium acetate mixture with reflux for 1 hour, to give a yellow powder. yield of 66%, M.W. 367.40, m. p. 201-202 ^{o}C. FT-IR(KBr) υ(cm^{-1}): 3450(υ O-H),3068, 3024 (υ Ar-H), 2954(υ C-H), 1764, 1712 (υ C=O), 1600 (υ C=C), 1384 (υ C- N), 1250 (υ C-O). ^{1}H NMR (400 MHz, Chloroform-d) δ 7.46 – 7.36 (m, 2H, Ar-H), 7.33 (dd, *J* = 5.4, 3.2 Hz, 2H, Ar-H), 7.08 – 6.98 (m, 2H, Ar-H), 6.57 – 6.49 (m, 2H, Ar-H), 4.88 (t, *J* = 1.7 Hz, 2H, Ar- CH-CH), 3.36 (t, *J* = 1.9 Hz, 2H, CH-CO). ^{13}C NMR (101 MHz, Chloroform-d) δ 175.69 (C=O), 168.60 (C=O), 150.77(C-O), 141.28 (C-N), 138.68 (C-N), 129.51 (Ar-C), 127.24

(Ar-C), 126.90 (Ar-C), 125.14 (Ar-C), 124.37 (Ar-C), 123.52 (Ar-C), 121.86 (Ar-C), 119.72 (Ar-C), 47.02 (CH- CH), 45.91 (CH-CH). MS (EI, m/z (%)): M+[$C_{24}H_{17}NO_3$], 367.4 (70%), 178.2 (100%).

Biological test
Cell lines and culture

MCF7 (a human Breast cancer cell line) and HeLa (a human cervical cancer cell line) were purchased from National Cell Bank of Iran (Pasteur Institute, Iran) and Biotech Cell Bank Unit-Iraq, respectively. Cells were grown in RPMI-1640 medium (Gibco) with 10% FBS (Gibco) supplemented with antibiotics (100 U/ml penicillin and 100 µg/ml streptomycin). Cells were maintained at 37 °C under humidified air containing 5% CO2 and were passaged using trypsin/EDTA (Gibco) and phosphate- buffered saline (PBS) solution[21].

MTT cell viability assay in MCF-7 and HeLa Cells.

Cell growth and cell viability were quantified using the MTT [3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium Bromide] (Sigma-Aldrich) assay. In brief, for monolayer culture, cells were digested with trypsin, harvested, adjusted to a density of 1.4×10^4 cells/well and seeded to 96-well plates filled with 200 µl fresh medium per well for 24 h. When cells formed a monolayer, they were treated with 100-6.25 µg/ml of the compound in five serial dilution series and in triplicates for 48 h at 37 °C and 5% CO2. At the end of the treatment (24h), while the monolayer culture was left untouched in the original plate, the supernatant was removed and 200 µl/well of MTT solution (0.5 mg/ml in phosphate-buffered saline [PBS]) was added and the plate was incubated at 37 °C for an additional 4 h. MTT solution (the supernatant of cells was removed and dimethyl sulfoxide was added (100 µl per well). Cells were incubated on a shaker at 37 °C until crystals were completely dissolved. Cell viability were quantified by measuring absorbance at 570 nm using an ELISA reader (Model wave xs2, BioTek, USA). The concentration of the compounds that resulted in 50% of cell death (IC_{50}) was determined from respective dose-response curves[22].

Computational details

All computations were performed using the Gaussian09 package [23] software package. Full geometry optimizations were performed using the Density Function Theory (DFT) at the B3LYP exchange–correlation functional using the 6-311+G(d,p) basis set level[24,25]. The optimized structures of prepared cyclic imides derivatives a1-a4 are shown in Figure1.

Retrieval of protein from database

The ligands were docked into the crystallographic structures of Human androgen receptors (AR) (PDB ID: 3pp0) and aromatase (PDB ID: 3EQM), obtained from the Protein Data Bank (PDB) [26].

Protein and Ligand preparation

Raw proteins from protein data bank with PDB (3pp0) and (3EQM) named Human estrogen receptor is further prepared for docking studies by removing all water molecules and the H atoms and followed by subsequent energy minimization using the AutoDock Vina and Discovery Studio 2017 R2 Client [27-30].

Molecular docking

Molecular docking procedure was carried out using the AutoDock Tools 4.0 (version: 1.5.6) program docking between Human estrogen receptors (PDB ID:3PP0) and (PDB ID: 3PP0) as a targets and optimize prepared cyclic imide derivatives (a1-a4) as a ligands. Automated dockings were carried out to calculated binding energy and locate the convenient binding orientations and conformations of prepared cyclic imide derivatives as a ligands in the Human estrogen receptor binding pocket by AutoDock Vina [27].

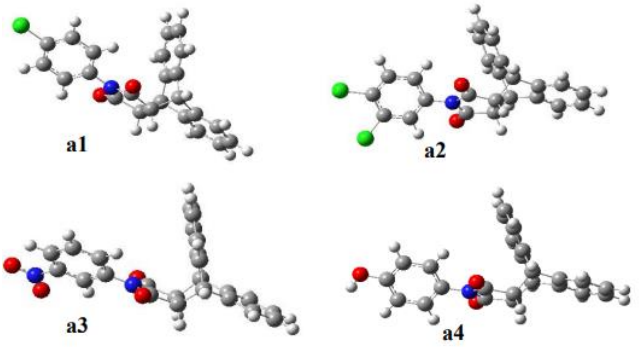


Figure 1. Optimized structures of prepared cyclic imides derivatives a1-a4

Results and Discussion

Synthesis

A number of cyclic imide derivatives were prepared in this study, using Diels-Alder reaction as a first step between maleic anhydride and anthracene. Use the reaction product as a reactant in the second step with four amines to produced analogies amic acids. Cyclization reaction was last step to prepared suggested cyclic imide derivatives (a1-a4).

Spectroscopic studies

The prepared cyclic imide derivatives (a1-a4) were characterized by FTIR, Mass and (^{1}H&^{13}C) NMR are mentioned in the experimental section. The mass spectra of the prepared cyclic imides were characterized by the appearance of a peak of the molecular ion [M+] at m/z in agreement with the proposed formula for these compounds. It is also observed in all mass spectra that the ion peak appears at m/z =178, which represents the spallation product of the Retro-Diel – Alder's

reaction. This is as a result of the breaking of the ring bond formed by the cyclic addition of the Diels-Alder reaction. This proves the cycloaddition reaction that we performed in the first step to prepare the cyclic imides (a1-a4) using the Diels-Alder reaction. The ^{1}H NMR spectra recorded on Chloroform-d and the signals attributable to aliphatic protons (from cycloaddition) and to aromatic phenyls protons. The ^{13}C NMR spectra recorded in Chloroform-d provides distinguished peaks confirming the presence of nonequivalent magnetic carbons. The C=O groups peaks indicates around175-168 ppm. The peaks around 130-119 ppm with a weak intensity representing the present aromatic carbons. In the aliphatic region, the peaks appeared at 48-45 ppm indicating the presence of CH_2 carbons.

Molecular docking

Molecular docking were performed to elevated of the prepare the cyclic imides (a1-a4) as conceder anticancer agents breast and cervical cancer. The binding energy (affinity) and interactions of synthesized compounds with Human androgen receptor (AR) as breast cancer androgen receptor (PDB ID:3pp0) and aromatase with the (PDB ID:3EQM) codes were calculated. Molecular docking studies were carried out for prepared cyclic imides (a1-a4) as a ligands against breast and cervical cancer androgen receptor (AR) structure (PDB ID: 3pp0) and aromatase the (PDB ID:3EQM) by AutoDock vina. Visualization ligands and protein interactions (complexes) using Discovery Studio 2017 R2 Client software. The results shows that good interaction of studies compounds with the target receptors. The binding energy of studied compounds with 3pp0 and 3EQM protein calculation shown in Tables 1 and 2. The values of the binding energy (affinity) between the protein 3pp0 and the prepared cyclic imides (a1-a4) calculated in molecular docking, were (-8.3, -8.8, -8.9 and -8.2) kcal/mol respectively, while their values calculated for protein 3EQM as a target were (-10.1, -10.3, -11.4 and -10.4) kcal/mol respectively.It is noticed that the values of binding energy between cyclic imides and the receptor 3EQM are less than that of the receptor 3pp0, and this is a good indication that the interaction with the receptor 3EQM was greater, and therefore the possibility of inhibition is greater. At the same time, it is noted that the number of hydrogen bonds formed was more with the 3EQM receptor, and this is an additional indicator of activity.

The cyclic imide a3 gave the lowest value of the bonding energy with the 3EQM receptor and the most number of hydrogen bonds, where the value of the bonding energy was -11.4 kcal/mol with the formation of five hydrogen bonds with the residues Arg115, Arg 145, Trp141 and Arg 438 were labelled as critical residues for exerting inhibitory activities[26]. On the other hand, there are a number of hydrophobic, hydrophilic, electrostatic and Van der Waals forces interactions [31] with a number of amino acids in the active sites of the protein, Ile 132, Gly 439, Ser 478, Phe 221, Trp 224, Thr 310, Asp 309, Ile 133, Ala 438, Leu 477, Cys 437 and Val 370 which gives strength in addition to the interaction and bonding, indicating that the prepared cyclic imide a4 is consider as a pioneering candidate for cervical cancer. The best docking position in the 2D and 3D structure molecules of prepared cyclic imides (a1-a4) with receptors 3pp0 and 3EQM can be seen in Figures 2-5 were plot by Discovery Studio 2017 R2 Client software.

Table 1
Calculated binding energies and H-bond and other interaction of the prepared cyclic imides (a1-a4) with target receptors 3pp0.

Comps.	Affinity (kcal/mol)	H-bond count			Amino acid residues	
		No. H-bonds	Amino acid residues	Distances Å	Van der Waals forces	other forces
a1	-8.3	1	Arg978	2.74	Arg 840, Met 953, Asp 769, Met 953, Arg 985, Asp 950 Phe 986	Tyr 772, Leu 869, Pro 567, Ile 954, Glu 975, Lys 957, Asp 871
a2	-8.8	2	Tyr 772 Arg 985	3.12 2.90	Lys957, Asp950, Phe 986, Arg 840, Leu 870, Asp 871, Leu 869, Asp769	Ile 872, Met 953 Arg 978
a3	-8.9	2	Lys 957 Lys 765	2.09 2.14	Arg 840, Arg 985, Arg 978, Asp 950, Leu 869, Asp 871	Tyr 772, Glu 975
a4	-8.2	4	Arg 978 Arg 985	3.05 3.16	Gle 975, Asp 950, Lys 957, Met 953, Phe 986, Asp 871, Leu 869, Asp 769	Tyr 772, Ile 872

Table 2
Calculated binding energies and H-bond and other interaction of the prepared cyclic imides (a1-a4) with target receptors 3EQM.

Comps.	Affinity (kcal/mol)	H-bond count			Amino acid residues	
		No. H-bonds	Amino acid residues	Distances Å	Van der Waals forces	other forces

a1	-10.1	-	-	-	Leu 477, Phe 430, Gly 439, Phe 134, Trp 224, Ala 438, Thr 310, Met 311	Met 446, Ile 442, Val 370, Ala 443, Ala 306, Arg 115, Cys 437, Ile 133
a2	-10.3	-	-	-	Phe 430, Gly 439, Leu 477, Phe 134, Trp 224, Ala 438	Thr 310, Ile 442, Met 446 Ala 306, Leu 152, Ala 443, Val 370, Ile 133, Arg 115, Cys 437
a3	-11.4	5	Arg 115 Arg 115 Arg145 Trp 141 Arg 438	2.97 3.02 3.18 3.28 2.68	Ile 132, Gly 439, Ser 478, Phe 221, Trp 224, Thr 310, Asp 309	Ile 133, Ala 438, Leu 477, Cys 437, Val 370
a4	-10.4	-	-	-	Gly 439, Thr 310, Ile 442, Met 446, Met 311, Phe 134, Phe 430, Leu 477, Trp 224, Ala 438	Ala 306, Ala 306, Val 370, Ile133, Arg 115, Cys 437

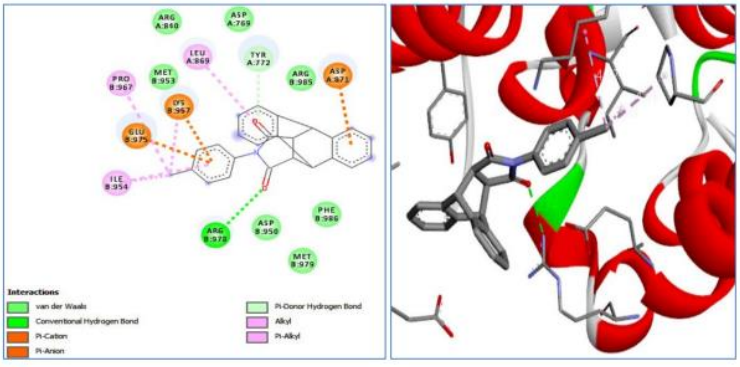


Figure 2. The binding mode (2D, 3D) of cyclic amide a1 with the receptor 3pp0

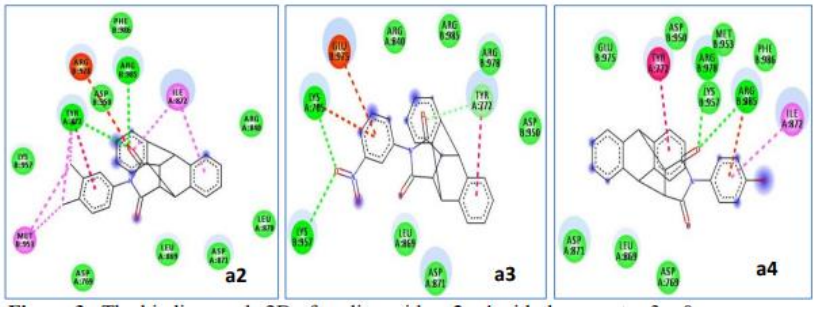


Figure 3. The binding mode 2D of cyclic amide a2-a4 with the receptor 3pp0

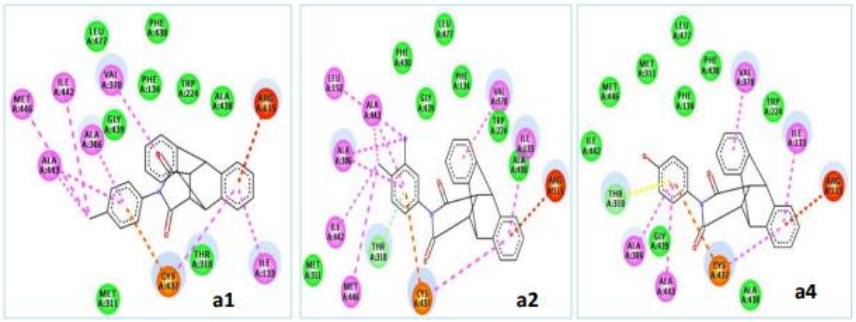


Figure 4. The binding mode 2D of cyclic amide a1, a2 and a4 with the receptor EQM

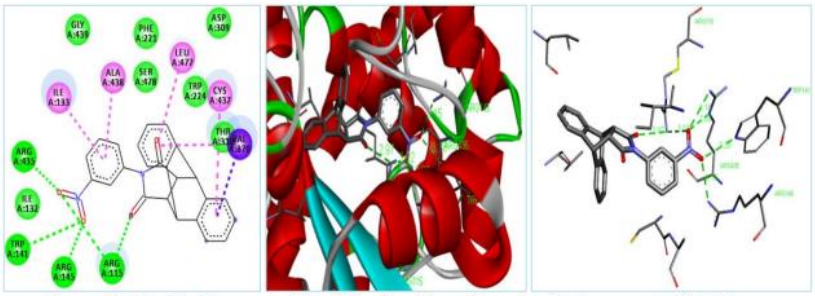


Figure 5. The binding mode (2D, 3D) of cyclic amide a3 with the receptor 3EQM

Evaluation of anticancer efficacy

Cytotoxicity was tested using the MMT assay for the prepared cyclic imide. Two cell lines were selected, the first representing live breast cancer cells type MCF-7 and the second line representing cervical cancer cells type HeLa.

Evaluation of breast anticancer efficacy

The MTT method was used to measure the activity of the prepared imides derivatives (a1-a4) in inhibiting live breast cancer cells of the MCF-7 cell line. Five different concentrations (100, 50, 25, 12.5, 6.25) µg/ml were prepared for each prepared imide, and the viability (%) were calculated at each concentration after treating the live cancer cells with these concentrations of the prepared imides. IC_{50}, were calculated for each prepared cyclic imide.

The percentages of residual viability of breast cancer cells (%) after being treated with prepared cyclic imide (a1-a4) at a concentration of 100 µg/ml. The rates of inhibition of the growth of live breast cancer cells for each compound are shown in Figure 6. Cyclic imide a3 gave the highest percentage of growth inhibition of MCF-7 breast cancer cells (the lowest percentage of live cancer cells at 40.83) by 59.17%. This was in agreement with the results of the molecular docking calculations, where the cyclic imide a3 gave the lowest binding energy with the receptor 3pp0 is -8.9 kcal/mol, with two hydrogen bonds. The IC_{50} values were (64.61, 71.11, 46.97, 75.12) µg/mL for the prepared cyclic imide derivatives (a1-

a4) respectively, the cyclic imide a3 gave the lowest value for IC_{50} at 46.97 µg/ml, while the imide a4 gave the highest value at 75.12 µg/ml.

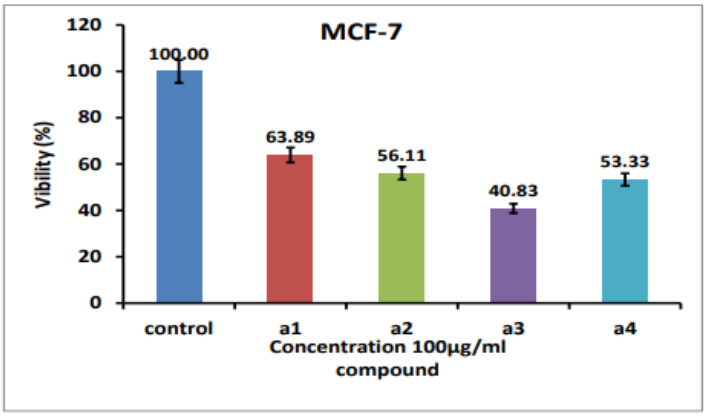


Figure 6. The percentages of residual viability of breast cancer cells (%) after being treated with prepared cyclic imide (a1-a4) at a concentration of 100 µg/ml.

Evaluation of the anti-cervical cancer efficacy

The MTT assay was used to estimate efficacy of the prepared cyclic imide (a1- a4) in inhibiting human cervical cancer cells of the HeLa cell line. Five different concentrations (750, 500, 250, 125, 62) µg/ml. The percentages of residual viability of cervical cancer cells (%) after being treated with prepared cyclic imide (a1-a4) at a concentration of 100 µg/ml are shown in Figure 8. The cyclic imide a3 had the highest percentage of inhibition of the growth of cervical HeLa cancer cells with a percentage of 99.05% (viability 0.95%) . The IC_{50} values were (45.10, 60.24, 10.57, 31.65) µg/mL for the prepared cyclic imide derivatives (a1-a4) respectively, the cyclic imide a3 gave the lowest value for IC_{50} at 10.57 µg/ml, while the imide a2 gave the highest value at 60.24 µg/ml.These results are in agreement with the results of the molecular docking, where the prepared cyclic imide a3 gave the lowest binding energy with the receptor 3EQM at -11.4 kcal/mol, and it was associated with it with five hydrogen bonds. The results of MTT assay and molecular docking scores proved that the prepared (11R,15S)-13-(3-nitrophenyl)-9,10-dihydro-9,10-[3,4]epipyrrolo-anthracene-12,14-di one derivative a3 is considered as a potential candidate inhibitor for cervical cancer.

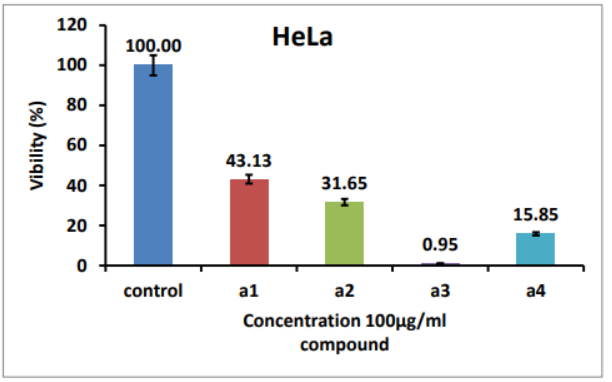


Figure 8. The percentages of residual viability of cervical cancer cells (%) after being treated with prepared cyclic imide (a1-a4) at a concentration of 100 µg/ml

Conclusions

In the present work, we are employed the Diels-Alder reaction to prepare new cyclic imide with high yield and by Click chemistry method. In an in vitro evaluation of anti-breast and anti-cervical cancer efficacy including cytotoxic test, the cyclic imide derivatives were screened by MTT assays with MCF-7 and Hela cell lines. All the prepared compounds showed good activity, but compound prepared (11R,15S)-13-(3-nitrophenyl)-9,10-dihydro-9,10-[3,4]epipyrrolo-anthracene-12,14-dione deriv- ative a3 was distinguished as having the highest activity in both assays and they have relatively low IC_{50} value. Theoretical studies using computational chemistry, especially molecular docking studies, are a useful and powerful tool to determine the activity of prepared compounds against cancer receptors. Thus, it will provide the researcher with important biological information that helps in the design, preparation and selection of candidate compounds as cancer inhibitors. The results of MTT assay and molecular docking scores proved that the prepared (11R,15S)-13-(3-nitrophenyl)-9,10-dihydro-9,10-[3,4]epipyrrolo-anthracene-12,14-dione derivative a3 is considered as a potential candidate inhibitor for cervical cancer.

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