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Synthesis in microwave, pharmacological evaluation, molecular docking and ADME studies of Schiff bases of Diclofenac targeting COX-2

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Abstract

Objective: To synthesize and preliminary pharmacological evaluation of new analogs of several new Schiff bases of diclofenac by investigating their interactions with COX-2 by docking and defining certain associations between their structures to improve selectivity towards COX-2 and to decrease side effects. **Methods**: Several new diclofenac Schiff bases were designed, prepared, and tested as potential COX-2 inhibitors. These new compounds were tested by molecular docking using genetic optimization for ligand docking suite for evaluated of their in vivo anti-inflammatory activity and COX-2 selectivity. **Results**: Because of their hydrogen bonding interaction with main amino acids in COX isozymes Arg121, Tyr356, and Ser120, all compounds evaluated in molecular docking exhibited important activities compared with diclofenac and 4PH9 as comparison drugs. The results of the ADME showed that all synthesized compounds absorbed from GIT while all compounds except (Yr 05 h-j) followed the Lipinski law. **Conclusion**: The production of the designed compounds has been managed successfully, the anti-inflammatory evaluation of the end products suggests that the new Schiff bases derivatives have strengthened their anti-inflammatory action, docking studies have shown that the preliminary analysis of anti-inflammatory activity has shown that all compounds (Yr 04, Yr 05 a-j) have strong anti-inflammatory properties, excluding Yr 03.

Key words: Diclofenac, Schiff base, docking, ADME, GOLD, Lipinski rule, microwave irradiations

Introduction

Diclofenac is a proven and widely used non-steroidal anti-inflammatory drug (NSAID) of the phenylacetic acid course. As with most NSAIDs, diclofenac exerts anti-inflammatory, analgesic, and antipyretic actions via inhibition of cyclooxygenases (COXs). The first created of Diclofenac was achieved by Alfred Sallmann and Rudolf Pfister in 1973 and since then the drug has been in clinical use for the treatment of rheumatoid arthritis, osteoarthritis, ankylosing spondylitis, gout attacks, postoperative pain, dysmenorrhea, and various ocular conditions (Davies and Anderson, 1997).

Besides its use in pathological processes related to inflammation, diclofenac and its chemical derivatives have been thoroughly examined for a varied range of pharmacological effects (Rossoni et al., 2008; Frantzias et al., 2012; Končič et al., 2009; Rojo et al., 2009; Johnsen et al., 2004; Wittine et al., 2009; Moody et al., 2010; Zhang et al., 2009). Several scientific communications report the broad-spectrum antibacterial activity of diclofenac in vitro and in vivo against isolates of various gram-positive and gram-negative bacteria that were either sensitive or resistant to antibiotics (Muñoz-Criado et al., 1996; Dastidar et al., 2000; Mazumdar et al., 2006; Mazumdar et al., 2009). The drug was reported to be highly bactericidal and exerts its action by inhibiting the DNA synthesis of bacteria (Dastidar et al., 2000; Mazumdar et al., 2009). Likewise, several small molecules bearing the 2-[(2,6-dichlorophenyl)amino]benzyl unit were shown to possess antibacterial potential (Sriram et al., 2006; Patel and Patel, 2011).

Antiviral activity of diclofenac was first studied by Gordon et al. in 1998. These researchers investigated topical NSAIDs direct pharmacological effects on adenoviral replication in vitro and only diclofenac appeared to have an inhibitory effect against different adenovirus serotypes (Gordon et al., 1998).

In a recent patent, the inventors claimed that diclofenac or its pharmaceutically acceptable salt inhibits the activity of herpes viruses, including herpes simplex virus type 1 (HSV-1), herpes simplex virus type 2 (HSV-2), and varicella-zoster virus. The drug was found to reduce the lesion size, lesion number, and viral titer when topically administered to herpesvirus lesions in animals (Lee and Shieh, 2011).

In a more recent study, some NSAIDs, antioxidants, and peroxisome proliferator-activated receptor-gamma agonists were tried for their capacity to interfere with rotavirus ECwt (wild type) infectivity in ICR mice. The work has to indicate that the treatment of rotavirus infected mice with diclofenac led to the reduced infection of villus cells (Guerrero et al., 2013).

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The function and morphology of the COX enzymes

The prostaglandins are lipid autacoids resulting from arachidonic acid and are synthesized via the cyclooxygenase pathway. Two linked isoforms of the cyclooxygenase enzyme have been defined. Cyclooxygenase-1 (COX-1) is accountable for the physiological creation of prostanoids while the cyclooxygenase-2 (COX-2) reasons the raised creation of prostanoids that happen at the site of disease and inflammation. COX-1 is defined as a "house-keeping enzyme" that controls ordinary cellular routes like gastric cytoprotection, vascular homeostasis, platelet aggregation, and kidney function. COX-2 is constitutively communicated in some tissues such as the brain, kidney, and bone, and its appearance at other sites is improved through states of inflammation. The two enzymes portion sixty-present homology in their amino acid sequence. However the complicated conformations for the substrate-binding places and catalytic areas are somewhat changed, for example, COX-2 has a greater and more elastic substrate station than COX-1 has, and also, COX-2 has a greater space at the site where inhibition bind and this structural change between COX-1 and COX-2 has allowed the development of COX-2 selective inhibiter (Picot et al., 1994).

Fig. 1 illustrates a diagram depiction of the amino acid structural variances between the substrate-binding channels of COX-1 and COX-2 that permitted the design of selective inhibitors. It is strong that the amino acid residues Val434, Arg513, and Val523 form a side compact in COX-2 that is absent in COX-1.

These findings resulted in that: (a) The non-selective inhibitors have access to the binding channels of both isoforms, while, (b) The more voluminous residues in COX-1, that are Ile434, His513, and Ile532, obstruct access of the bulky side chains of the COX-2 inhibitors (Grosser et al., 2006).

Studies (Liou, 2010), have shown that glucocorticoids can suppress the creation of proteins involved in inflammation (resulting in their role as anti-inflammatory compounds). Aside from that, glucocorticoids further suppress inflammation by activating a group of enzymes known as lipocortins. Lipocortins have been found to inhibit or decrease the activity of phospholipase A2, a key enzyme that participates in the release of arachidonic acid from the cell membrane, where it is usually incorporated into. When the cell attacked by foreign substances, arachidonic acid is released from the cell membrane and is converted into substances such as prostaglandins which mediate inflammation. Free arachidonic acid is being used in the production of inflammatory prostaglandins by the COX-2 isozyme. The release of arachidonic acids requires the activation of the enzyme phospholipase A2. As mentioned earlier the lipocortins inhibit the phospholipase A2 activity. By activating lipocortins, glucocorticoids cause the inhibition of phospholipase A2, thereby inhibiting the release of arachidonic acid and consequently prostaglandin synthesis in the cell and therefore participate in their effect as significant anti-inflammatory agents. Because lower amounts of inflammatory prostaglandins are synthesized, inflammation is suppressed and damage caused by chronic inflammation is decreased.

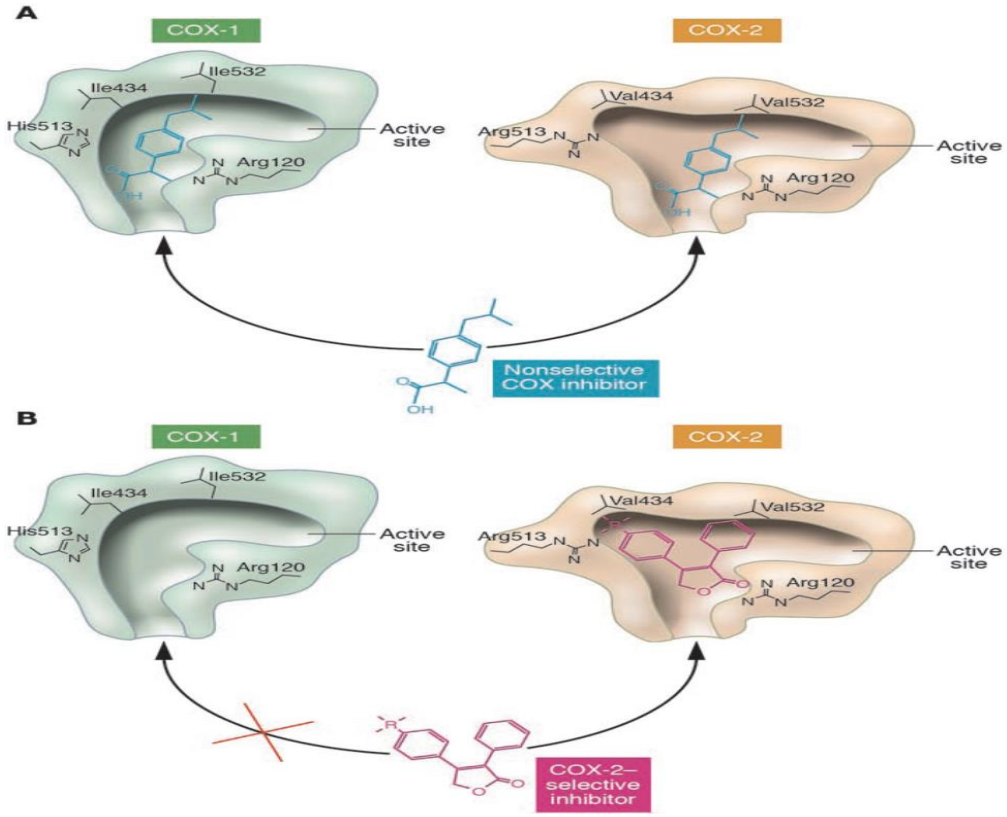

Fig. (1): The schematic depiction of the amino acid structural differences between the substrate-binding channels of COX-1 and COX-2 that allowed the design of selective inhibitors (Grosser et al., 2006).

Heterocyclic Compounds containing an azomethine group (-CH=N-), called as Schiff bases. The first synthesis of imine was done by Hugo Schiff in 1864 (Hussain et al., 2014). The Schiff base is formed by reacting compounds having active carbonyl groups with primary amines in presence acid (Saxena, 2013). Schiff bases possess a wide variety of biological activities such as antimicrobial activity (Al-Shemary et al., 2016), antileishmanial (Al Zoubi, 2013), anti-inflammatory (Tantaru et al., 2013), anti-HIV (Patel et al., 2012), Anticonvulsant (Singh and Kumar, 2016), anticancer (Arulmurugan et al., 2010), antifungal (Kailas et al., 2016) and anti-proliferative (Su et al., 2015). Schiff bases derivatives are important in the medical field.

Microwave chemistry is the science of applying microwave radiation to chemical reactions (Microwaves in Organic Synthesis, 2018; de la Hoz et al., 2005; Strauss and Trainor, 1995; Kidwai, 2001; Kappe et al., 2012). Microwaves act as high-frequency electric fields and will generally heat any material containing mobile electric charges, such as polar molecules in a solvent or conducting ions in a solid. Polar solvents are heated as their component molecules are forced to rotate with the field and lose energy in collisions. Semiconducting and conducting samples heat when ions or electrons within them form an electric current and energy is lost due to the electrical resistance of the material. Microwave heating in the laboratory began to gain wide acceptance following papers in 1986 (Gedye et al., 1986), although the use of microwave heating in chemical modification can be traced back to the 1950s. Although occasionally known by such acronyms as *MAOS* (Microwave-Assisted Organic Synthesis) (Pizzetti et al., 2012), *MEC* (Microwave-Enhanced Chemistry), or *MORE synthesis* (Microwave-organic Reaction Enhancement), these acronyms have had little acceptance outside a small number of groups.

Materials and Methods

General

All reagents and anhydrous solvents were of an annular type and generally used as received from the commercial suppliers (Merck, Germany, Reidel-De Haen, Germany, Sigma-Aldrich, Germany and BDH, England).

Experimental Part

1. Melting points were recorded using the digital STUART scientific SMP30 melting point apparatus and are uncorrected.
2. FTIR spectra were recorded on SHIMADZU FTIR-8400S using ZnSe PRISM in the (4000-600) cm^{-1} spectral range.
3. 1HNMR was recorded on BRUKER 300MHzistrument using DMSO as solvent and TMS as an internal reference.

Synthesis of [2-(2,6-dichloroanilino)phenyl]acetic acid (Yr 02)

Diclofenac sodium (2g, 6.28mmol), was dissolved in 50ml of warm water with stirring for 10 minutes, then 2N HCl (3.1ml, 6.28mmol) was added, followed by addition of excess cold water (100ml), the acid was precipitated then filtered, dried and used in the following step without further purification (Banerjee and Amidon, 1981).

IR spectra (KBr, υ_{max}, cm^{-1}): 3323 (N-H), 3066 (O-H), 1689 (C=O), 1303 (C-O), 740 (C-Cl).

^{1}H–NMR (DMSO, δ, ppm): 3.72 (s, 2H, CH_2), 6.29 (s, 1H, NH), 6.85–7.53 (m, 7H, ArH), 12.73 (s, 1H, OH).

Synthesis of methyl [2-(2,6-dichloroanilino)phenyl]acetate (Yr 03)

The diclofenac methyl ester was prepared as per the procedure described previously in the literature (Ballini and Carott, 1983). These steroidal carboxylic acids were methylated using Potassium Carbonate as a base in refluxing acetone.

Yield: 98% , mp: 97.5°C.

IR spectra (KBr, υ_{max}, cm^{-1}): 3352 (N-H), 3024 (C-H), 1737 (C=O), 1294 (C-O), 750 (C-Cl).

^{1}H-NMR (DMSO, δ, ppm): 3.66 (s, 3H, O-CH_3), 3.82 (s, 2H, CH_2), 6.26 (s, 1H, NH), 6.84–7.55 (m, 7H, ArH).

Synthesis of 2-[2-(2,6-dichloroanilino) phenyl]acetohydrazide (Yr 04)

Compound (Yr 03, 0.01 mol) and hydrazine hydrate (0.02 mmol) were refluxed in absolute ethanol (50 ml) for 24 h (examined by thin-layer chromatography [TLC]).

Compound synthesis (Yr 04) was performed as stated in previous literature (Palkar et al., 2014). The mixture was concentrated, cooled, and placed in ice water. So isolated white amorphous solid was washed, dried, and recrystallized from ethanol and water to provide compound (Yr 04).

Yield: 99 % , mp 158°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3348(NH_2), 3327 (N-H), 3030 (C-H), 1635 (C=O), 742 (C-Cl).

^{1}H-NMR (DMSO, δ, ppm): 3.36 (s, 2H, CH_2), 4.35 (s, 2H, NH_2), 6.30 (s, 1H, NH), 6.85–7.53 (m, 7H, ArH), 9.51 (s, 1H, CO-NH).

General method for the production of 2-(2-(2,6-dichloroanilino)phenyl)-N'- ethylideneaceto hydrazide derivatives (Yr 05a-j)

A finely ground mixture of Compound (Yr 04, 0.01 mmol) was added in the irradiation tube, the appropriately substituted

aromatic aldehyde (0.01 mmol) was added with 2 drops from glacial acetic acid and the reaction mix was irradiated in microwave synthesis reactor (450w) for a suitable time. Then, the reaction combination was cool to the room temperature, filtered, and the Schiff base is collected. The product was recrystallized using absolute ethanol as per the earlier report.

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(E)-(pyridin-4-yl)methylidene]acetohydrazide (Yr 05a)

Yield: 71 % , mp 228°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3317 (N-H), 3028 (C-H), 1647 (C=O), 744 (C-Cl).

^{1}H-NMR (DMSO, δ, ppm): 4.17 (s, 2H, CH_2), 6.29 (s, 1H, NH), 6.84–8.65 (m, 11H, ArH), 11.90 (s, 1H, N=CH), 12.10 (s, 1H, CO-NH).

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(1E)-1-(pyridin-4-yl)ethylidene]acetohydrazide (Yr 05b)

Yield: 75 % , mp 241-243°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3275 (N-H), 3024 (C-H), 1660 (C=O), 759 (C-Cl).

^{1}H‑NMR (DMSO, δ, ppm): 2.30 (s, 3H, CH_3), 4.21 (s, 2H, CH_2), 6.30 (s, 1H, NH), 6.84–8.62 (m, 11H, ArH), 10.99 (s, 1H, CO-NH).

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(1E)-1-(pyridin-3-yl)ethylidene]acetohydrazide (Yr 05c)

Yield: 73 % , mp 250°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3288 (N-H), 3057 (C-H), 1668 (C=O), 752 (C-Cl).

^{1}H‑NMR (DMSO, δ, ppm): 2.34 (s, 3H, CH_3), 4.19 (s, 2H, CH_2), 6.28 (s, 1H, NH), 6.84–9.03 (m, 11H, ArH), 11.00 (s, 1H, CO-NH).

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(E)-(furan-2-yl)methylidene]acetohydrazide (Yr 05d)

Yield: 89 % , mp 267°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3288 (N-H), 3022 (C-H), 1643 (C=O), 746 (C-Cl).

^{1}H‑NMR (DMSO, δ, ppm): 4.06 (s, 2H, CH_2), 6.26 (s, 1H, NH), 6.32–8.13 (m, 10H, ArH), 11.55 (s, 1H, N=CH), 11.74 (s, 1H, CO-NH).

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(1E)-1-(furan-2-yl)ethylidene]acetohydrazide (Yr 05e)

Yield: 88 % , mp 281°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3248 (N-H), 3068 (C-H), 1660 (C=O), 746 (C-Cl).

^{1}H‑NMR (DMSO, δ, ppm): 2.20 (s, 3H, CH_3), 4.09 (s, 2H, CH_2), 6.21 (s, 1H, NH), 6.31–7.81 (m, 10H, ArH), 10.82 (s, 1H, CO-NH).

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(E)-(thiophen-2-yl)methylidene]acetohydrazide (Yr 05f)

Yield: 70 % , mp 278°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3273 (N-H), 3021 (C-H), 1643 (C=O), 746 (C-Cl).

^{1}H‑NMR (DMSO, δ, ppm): 4.06 (s, 2H, CH_2), 6.28 (s, 1H, NH), 6.83–8.24 (m, 10H, ArH), 11.59 (s, 1H, N=CH), 11.74 (s, 1H, CO-NH).

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(1E)-1-(thiophen-2-yl)ethylidene]acetohydrazide (Yr 05g)

Yield: 79 % , mp 287°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3269 (N-H), 3026 (C-H), 1662 (C=O), 746 (C-Cl).

^{1}H‑NMR (DMSO, δ, ppm): 2.31 (s, 3H, CH_3), 4.10 (s, 2H, CH_2), 6.28 (s, 1H, NH), 6.84–7.85 (m, 10H, ArH), 10.90 (s, 1H, CO-NH).

N'-[(E)-(4-chlorophenyl)methylidene]-2-[2-(2,6-dichloroanilino)phenyl]acetohydrazide (Yr 05h)

Yield: 75 % , mp 227°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3340 (N-H), 3068 (C-H), 1656 (C=O), 750 (C-Cl).

^{1}H‑NMR (DMSO, δ, ppm): 4.12 (s, 2H, CH_2), 6.28 (s, 1H, NH), 6.32–8.17 (m, 11H, ArH), 11.59 (s, 1H, N=CH), 11.78 (s, 1H, CO-NH).

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(E)-(4-dimethylanilino)methylidene]acetohydrazide (Yr 05i)

Yield: 76 % , mp 250°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3317 (N-H), 3028 (C-H), 1647 (C=O), 744 (C-Cl).

^{1}H-NMR (DMSO, δ, ppm): 3.32 (s, 6H, CH_3), 4.10 (s, 2H, CH_2), 6.30 (s, 1H, NH), 6.86–8.26 (m, 11H, ArH), 11.32 (s, 1H, N=CH), 11.49 (s, 1H, CO-NH).

2-[2-(2,6-dichloroanilino)phenyl]-N'-[(E)-(4-nitrophenyl)methylidene]acetohydrazide (Yr 05j)

Yield: 78 % , mp 242°C.

IR spectra (KBr,υ_{max}, cm^{-1}): 3362 (N-H), 3080 (C-H), 1658 (C=O), 750 (C-Cl).

Computational Method

The computational method approved in this work is outlined in Fig. 2. A full licensed CCDC genetic optimization for ligand docking (GOLD) Suite (v. 5.7.3) was used to achieve the molecular docking studies for the compounds. CCDC Hermes visualizer software (v. 1.10.3) was used to envisage: the protein, ligands, interactions of hydrogen bonding, short contacts, and length of bonds calculation. The chemical structures of our ligands were drawn using ChemDraw Professional software (v. 16.0) Fig. 3.

The pharmacokinetic profile, i.e., adsorption, distribution, metabolism, excretion (ADME) of the synthesized compounds was expected with the assistance of the Swiss ADME server. (Daina et al., 2017)

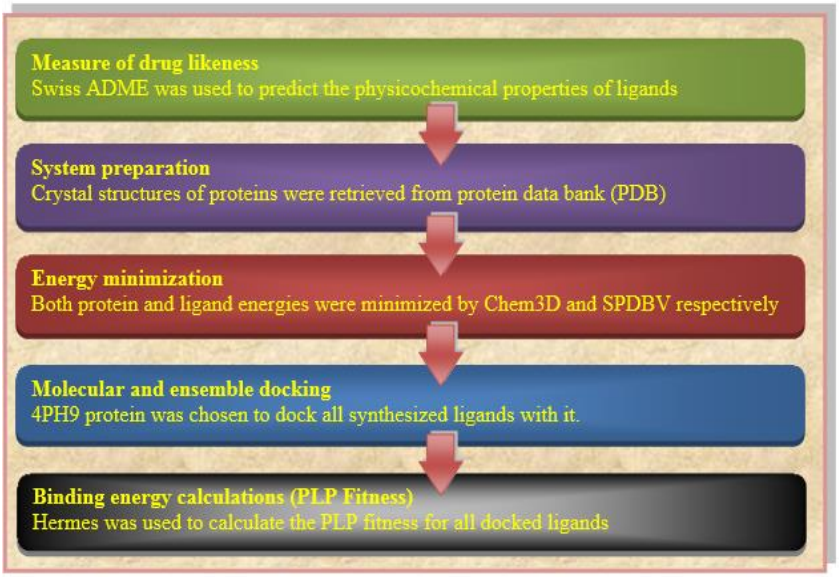

Fig. (2) Outline of computational procedure.

Fig. (3) A full licensed CCDC genetic optimization for ligand docking (GOLD)

ADME procedures

All ligands were drawn by ChemSketch (v. 14), converted to SMILE name by Swiss ADME tool which predicts the physicochemical descriptors and pharmacokinetic properties. BOILED-EGG was used to compute the lipophilicity and polarity of the small molecule. (Friedman, 1951)

Preparation of ligands and protein receptor

The crystal structures of the enzyme COX 2 [PDB ID: 4PH9] were downloaded from the Protein Data Bank (PDB) and their missing atoms were added with the assistance of Swiss PDB Viewer (v. 3.7). The crystal structures of our downloaded protein were prepared by eliminating all water molecules and by the addition of hydrogen atoms to get a right ionization and tautomeric states of amino acid residues. Chem3D (v. 16.0) was used to minimize the energy for our synthesized ligands by applying the MM2 force field.

Docking procedures

The full license version of GOLD (v. 5.7.3) was used for molecular docking. (Jones et al., 1997; Jones et al., 1995) The Hermes visualizer software in the GOLD Suite was used to set up the receptors for the docking process additionally. The binding site used to dock GOLD has been identified as all the protein residues inside the 10 Å of the source ligands that occur in the complexes of the downloaded protein structure. Two COX-2 proteins were downloaded from the PDB website (1PXX and 4PH9) to dock the whole process. (Huang and Zou, 2007) As a result, 4PH9 was selected to dock the compounds.

CCDC Superstar was used to assessing the cavity and the active site. The protein reference ligand has been used for determining the active site radius (10 Å). ChemScore kinase was used as a template for the configuration. For the scoring feature ChemPiecewise linear potential (CHEMPLP) was utilized. The scores of all parameters used during the docking process remained the default, and all solutions are scored according to the fitness function of CHEMPLP. According to CHEMPLP, the steric adjunct between protein and ligand is determined while the distance and angle-dependent hydrogen are considered. The results of docking, i.e., the binding mode, docked pose, and binding free energy was studied to evaluate the interaction between the amino acid residues of the proteins COX-2 and our synthesized ligands.

Molecular Modeling

Genetic optimization for ligand docking is a “genetic algorithm for docking flexible ligands into protein binding sites” (Webb and Griswold, 1984). GOLD has been broadly verified and has shown superb rendering for pose prediction and excellent results for virtual screening (Palm et al., 1997). It is provided as a part of the GOLD Suite, which contains additional software components, Hermes, Mercury, Isostar and Conquest, and GoldMine, etc.

Ligands and protein-energy minimization will repair distorted geometries by transferring atoms to release internal restrictions. After the minimization of the energy, the geometry is fixed which means a minimum of energy has been reached.

To predict the selectivity and binding energies of the synthesized compounds for COX-1 and COX-2, docking studies were performed with the help of GOLD Suite software to study the molecular interactions involved in between active binding sites of the protein target and the synthesized compounds (Table 1).

Table 1: The binding energies for derivatives and reference NSAIDs docked with COX-2

Compounds	COX-2 binding energy (PLP Fitness)	No. of Amino acids included in H-bonding	Amino acids included in H-bonding	no. of bonding	Length of bonding	
4PH9	66.32	3	ARG 121	1	3.047	
			ARG 121	1	3.007	
			TYR 356	1	2.761	
YR 05 i	93.22	3	TYR 356	2	3.036	2.597
			ARG 121	1	3.025	
YR 05 a	84.93	2	ARG 121	1	3.034	
			TYR 356	1	2.771	
YR 05 f	84.06	3	ARG 121	1	2.950	
			ARG 121	1	2.877	
			TYR 356	1	2.542	
YR 05 h	83.39	3	ARG 121	1	3.059	
			ARG 121	1	2.952	
			TYR 356	1	2.498	
YR 05 d	79.39	3	ARG 121	1	2.916	
			ARG 121	1	2.854	
			TYR 356	1	2.456	
YR 05 j	77.92	1	SER 531	1	2.979	
YR 05 g	77.47	2	ARG 121	1	2.984	
			TYR 356	1	2.892	
YR 05 c	75.93	5	SER 120	1	3.006	
			ARG 121	2	3.019	2.738
			ARG 121	1	3.012	
			TYR 356	1	2.967	
YR 05 e	72.42	3	ARG 121	1	2.976	
			ARG 121	1	2.870	
			TYR 356	1	2.747	
YR 05 b	70.61	2	ARG 121	1	2.923	
			TYR 356	1	2.681	
YR 04	68.34	4	HIS 90	1	3.080	
			LEU 353	1	2.770	
			SER 354	2	2.738	2.462
YR 03	62.66	1	TYR 356	1	2.733	

The distance of short contacts and hydrogen bonding between a specific protein atom and our synthesized ligands is calculated by GOLD and all bond length below 3 Å. (Verdonk et al., 2003) The short contacts are described as other interacting forces like van der Waals, electrostatic, steric, p – p stacking, dipole-dipole, and others.

All the synthesized compounds having promising docking results with COXs, fitted in the COX-2 active site as shown in Fig. 4–16

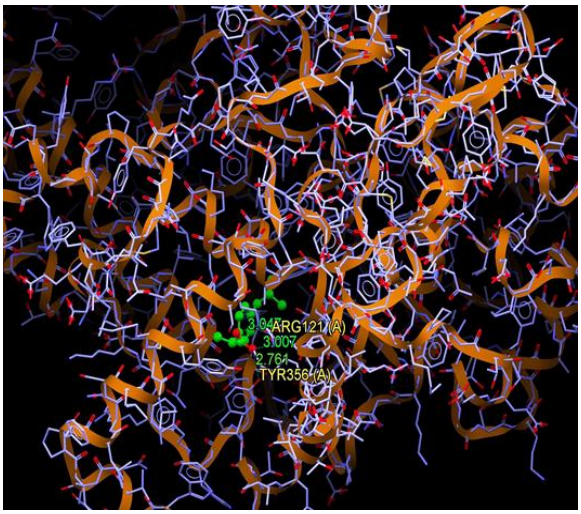

Fig. (4) : Short contacts interaction profile for the compound reference 4PH9.

The interaction between compound (4PH9) and amino acid residues [4Ph9: Ball-and-stick style, residues of amino acid in the capped form].

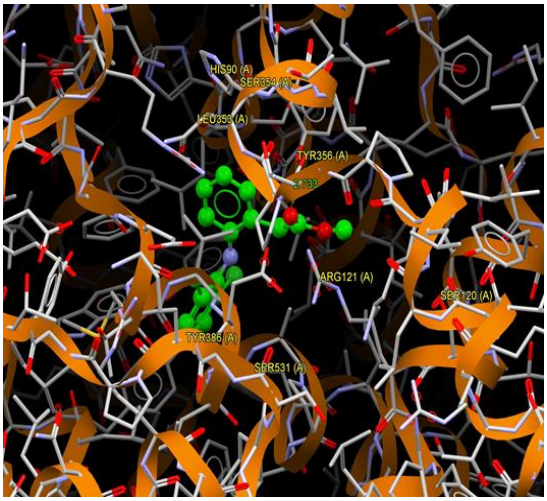

Fig. (5) : Short contacts interaction profile for the compound Yr 03.

The interaction between compound (Yr 03) and amino acid residues [Yr 03: Ball-and-stick style, residues of amino acid in the capped form].

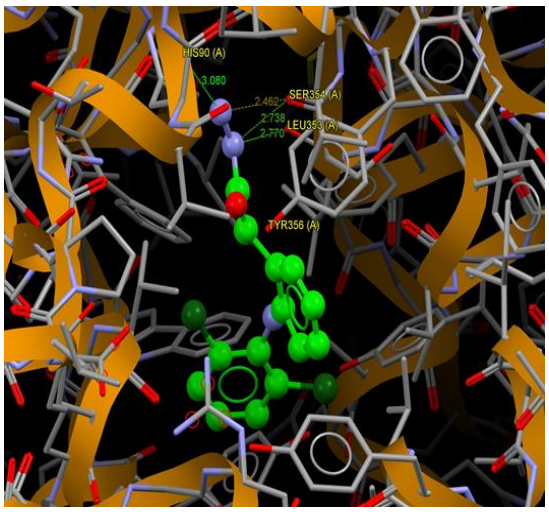

Fig. (6) : Short contacts interaction profile for the compound Yr 04.

The interaction between compound (Yr 04) and amino acid residues [Yr 04: Ball-and-stick style, residues of amino acid in the capped form].

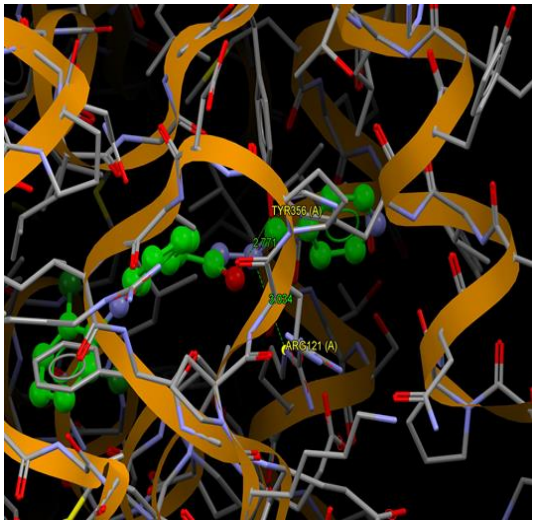

Fig. (7) : Short contacts interaction profile for the compound Yr 05 a.

The interaction between compound (Yr 05 a) and amino acid residues [Yr 05 a: Ball-and-stick style, residues of amino acid in the capped form].

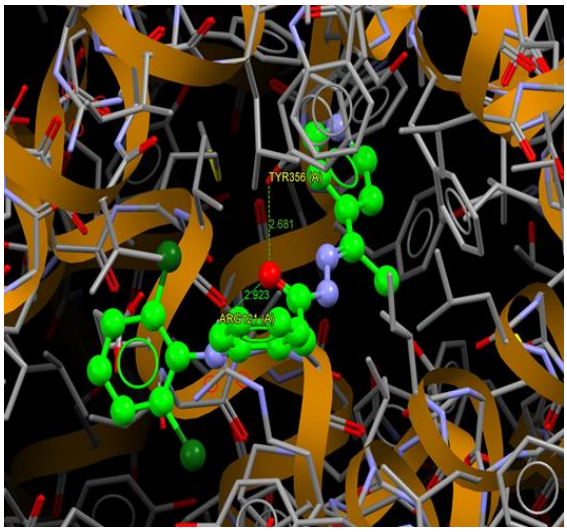

Fig. (8) : Short contacts interaction profile for the compound Yr 05 b.

The interaction between compound (Yr 05 b) and amino acid residues [Yr 05 b: Ball-and-stick style, residues of amino acid in the capped form].

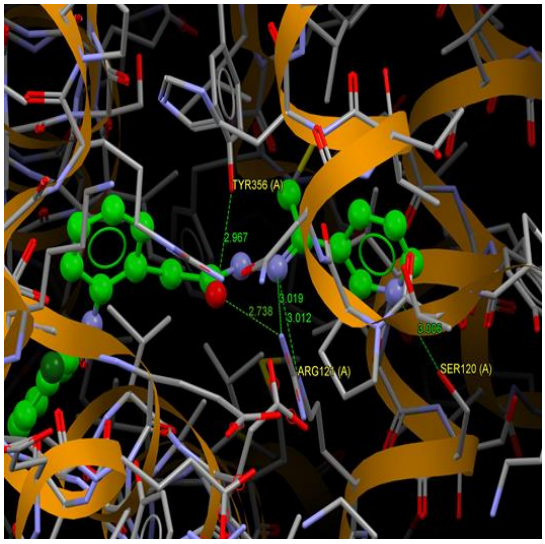

Fig. (9) : Short contacts interaction profile for the compound Yr 05 c.

The interaction between compound (Yr 05 c) and amino acid residues [Yr 05 c: Ball-and-stick style, residues of amino acid in the capped form].

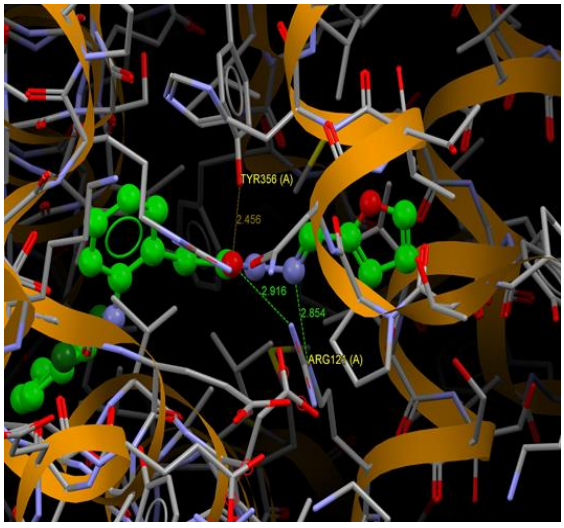

Fig. (10) : Short contacts interaction profile for the compound Yr 05 d.

The interaction between compound (Yr 05 d) and amino acid residues [Yr 05 d: Ball-and-stick style, residues of amino acid in the capped form].

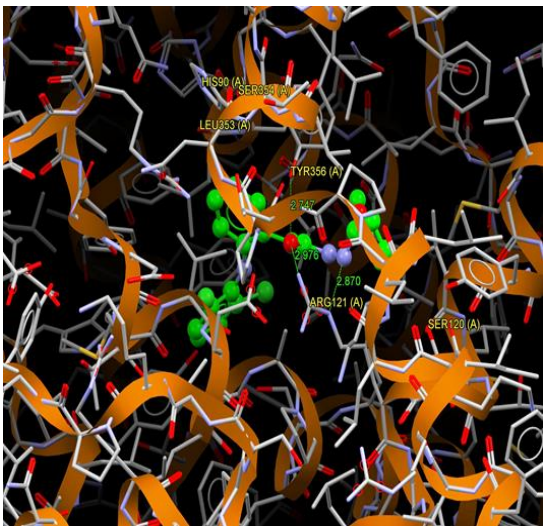

Fig. (11) : Short contacts interaction profile for the compound Yr 05 e.

The interaction between compound (Yr 05 e) and amino acid residues [Yr 05 e: Ball-and-stick style, residues of amino acid in the capped form].

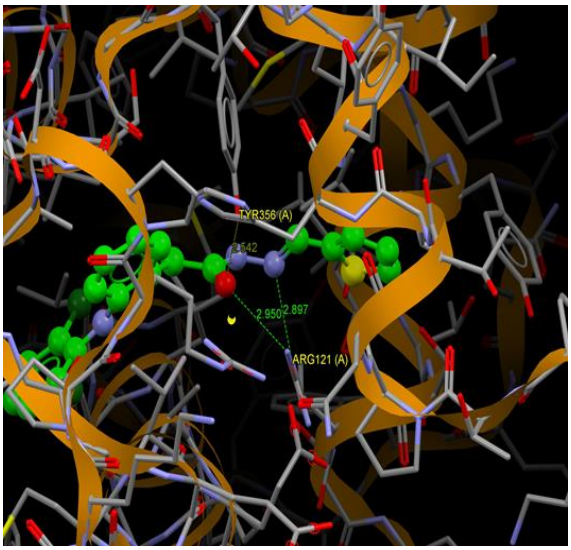

Fig. (12) : Short contacts interaction profile for the compound Yr 05 f.

The interaction between compound (Yr 05 f) and amino acid residues [Yr 05 f: Ball-and-stick style, residues of amino acid in the capped form].

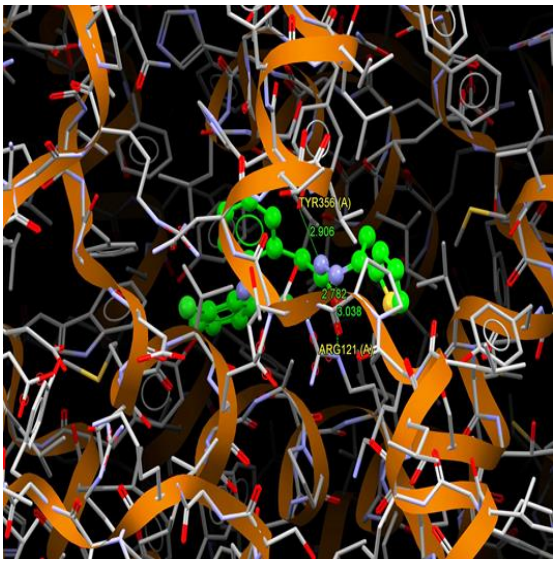

Fig. (13) : Short contacts interaction profile for the compound Yr 05 g.

The interaction between compound (Yr 05 g) and amino acid residues [Yr 05 g: Ball-and-stick style, residues of amino acid in the capped form].

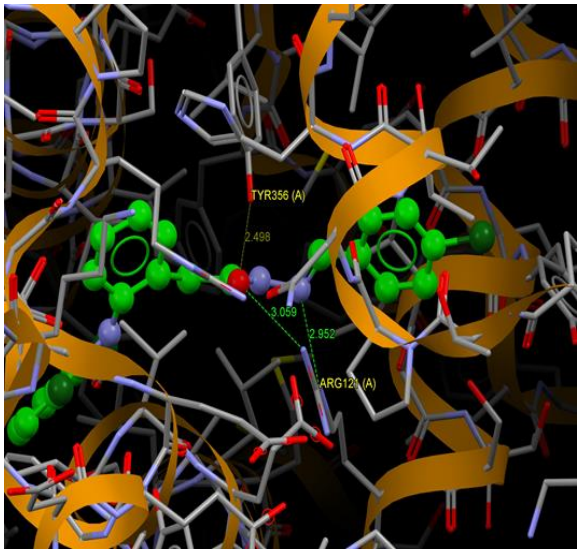

Fig. (14) : Short contacts interaction profile for the compound Yr 05 h.

The interaction between compound (Yr 05 h) and amino acid residues [Yr 05 h: Ball-and-stick style, residues of amino acid in the capped form].

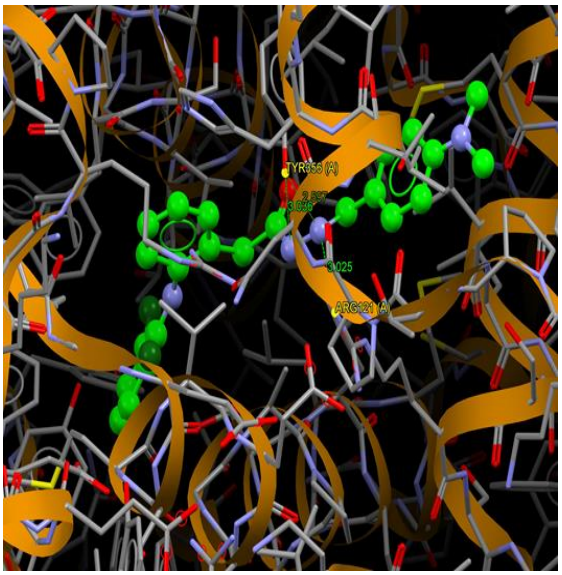

Fig. (15) : Short contacts interaction profile for the compound Yr 05 i.

The interaction between compound (Yr 05 i) and amino acid residues [Yr 05 i: Ball-and-stick style, residues of amino acid in the capped form].

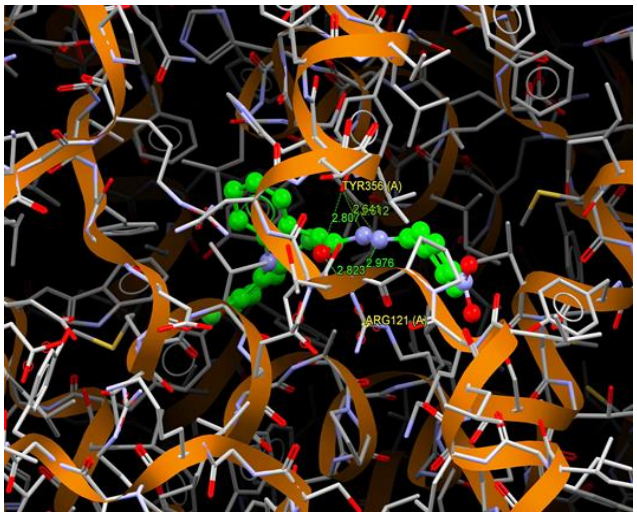

Fig. (16) : Short contacts interaction profile for the compound Yr 05 j.
The interaction between compound (Yr 05 j) and amino acid residues [Yr 05 j: Ball-and-stick style, residues of amino acid in the capped form].

Compounds Yr 05 (a-i) (Figs. 7–15) show H-bond interactions with Arg121 and Tyr356 and these two amino acids exist in the binding with five approved NSAIDs (Ibuprofen, Naproxen, Indomethacin, Flurbiprofen, and Des-methylflurbiprofen). Compound (Yr 05 j) (Fig. 16) has H-bond with Ser531 which is the binding site of diclofenac, lumiracoxib, tolfenamic acid. Compound (Yr 03) (Fig. 5) has one H-bond with Tyr355 like in aspirin.

ADME Studies

The ADME properties profile of our created compounds was studied by the Swiss ADME server to detect the safer and potential drug candidate(s) to filter out the compounds which are most likely to fail in the subsequent stages of drug development due to unfavorable ADME properties.

We assessed all synthesized compounds' ADME method.

Also, we measured the topological polar surface area (TPSA), as this is another important property related to the bioavailability of drugs. Thus, passively absorbed molecules with a TPSA > 140 Å are thought to have low oral bioavailability. (Suralkar et al., 2008) Our findings indicate that all produced compounds have TPSA below 140, which is within the range of 38–99 and that all ligands have a bioavailability of 0.55, indicating that all ligands enter the systemic circulation.

Compounds **Yr (02-04) and Yr 05 (a-g)** fulfilled Lipinski rule. Also, it fulfilled the topological descriptors and fingerprints of molecular drug-likeness structure keys as Log*P* and Log*S*.

The GI absorption score is a measure of the extent of absorption of a molecule from the intestine following oral administration. The absorption could be excellent if the result were high. In this study, the GI absorption of all compounds was high predicting them to be well absorbed from the intestine.

The ADME properties profiles for the created compounds are shown in (Table 2).

Table 2: ADME properties profile of the synthesized compounds

Comps.	Formula	M.Wt (g/mol)	H-bond acceptors	H-bond donors	MR	TPSA Å²	GI Abs.	BBB permeant	Lipinski violations
Yr 02	$C_{14}H_{11}Cl_2NO_2$	296.15	2	2	77.55	49.33	High	Yes	0
Yr 03	$C_{15}H_{13}Cl_2NO_2$	310.17	2	1	81.87	38.33	High	Yes	0
Yr 04	$C_{14}H_{13}Cl_2N_3O$	310.18	2	3	81.49	67.15	High	Yes	0
Yr 05 a	$C_{20}H_{16}Cl_2N_4O$	399.28	3	2	109.55	66.38	High	Yes	0
Yr 05 b	$C_{21}H_{18}Cl_2N_4O$	413.30	3	2	114.36	66.38	High	No	0
Yr 05 c	$C_{21}H_{18}Cl_2N_4O$	413.30	3	2	114.36	66.38	High	No	0

Yr 05 d	$C_{19}H_{15}Cl_2N_3O_2$	388.25	3	2	104.03	66.63	High	Yes	0
Yr 05 e	$C_{20}H_{17}Cl_2N_3O_2$	402.28	3	2	108.83	66.63	High	No	0
Yr 05 f	$C_{19}H_{15}Cl_2N_3OS$	404.31	2	2	109.64	81.73	High	No	0
Yr 05 g	$C_{20}H_{17}Cl_2N_3OS$	418.34	2	2	114.44	81.73	High	No	0
Yr 05 h	$C_{21}H_{16}Cl_3N_3O$	432.73	2	2	116.77	53.49	High	No	1
Yr 05 i	$C_{23}H_{22}Cl_2N_4O$	441.36	2	2	125.97	56.73	High	Yes	1
Yr 05 j	$C_{21}H_{16}Cl_2N_4O_3$	443.28	4	2	120.58	99.31	High	No	1

Conclusion

1. The production of the designed compounds has been magnificently achieved.
2. Characterization and identification of the created compounds were confirmed by the determination of physical properties (melting point and description), FT-IR spectroscopy, and 1H-NMR spectra.
3. The anti-inflammatory valuation of the final products shows that the incorporation of some aldehydes and ketones into sodium Diclofenac improved its anti-inflammatory action.
4. The ADME studies showed compounds **Yr (02-04) and Yr 05 (a-g)** fulfilled the Lipinski rule and all created compounds riveted from GIT.
5. The preliminary study of anti-inflammatory activity showed that compounds Yr 05 (a-j) have significantly more anti-inflammatory outcomes than sodium Diclofenac.

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