SYNTHESIS AND PRELIMINARY MOLECULAR DOCKING STUDIES OF NOVEL ETHYL-GLYCINATE AMIDE DERIVATIVES

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ABSTRACT

Ethyl glycinate was synthesized by the Fischer esterification protocol, and its amide derivatives; 2-amino-*N*-(nitrophenyl)acetamide **31**, 2-amino-*N*-(6-methylpyridin-2-yl) acetamide **33,** *N,N'*-(1,4-phenylene)bis-(2-aminoacetamide) **35,** *N,N'*-(6-chloropyrimidine-2,4-diyl)bis-(2-aminoacetamide) **37,** and 2,4-(diamino-*N'N*-6-hydroxypyrimidyl)acetamide **39** respectively were obtained by coupling reactions of 4-nitroaniline, 2-amino-6-methylpyridine, 1,4-diamino-N,N'-benzene, 2,6-diamino-4-chloropyrimidine and 2,4-diamino-6-hydroxypyrimidine respectively with ethyl glycinate. These compounds were characterized on the basis of their melting points, UV-Visible, IR, ^{1}HNMR and ^{13}CNMR spectroscopic analyses. The results obtained from the spectra were consistence with the assigned structures of the compounds. The synthesized compounds were subjected to molecular docking with a target protein, 1CVU to compare their binding energies with celecoxib and rofecoxib which are standard drugs that inhibit COX2 enzyme. From the docking results, the binding energies values of the above synthesized compounds are -5.8 kJmol^{-1}, -6.2 kJmol^{-1}, -7.2 kJmol^{-1}, -7.4 kJmol^{-1} and -7.6 kJmol^{-1} respectively. Compound **39** showed the highest binding energy of -7.6 kJmol^{-1}, close to celecoxib and rofecoxib with binding energy values of -8.0 kJmol^{-1} and -8.2 kJmol^{-1} respectively. This result indicates that compound **39** possess some level of inhibitory activity against COX2.

1.   INTRODUCTION

Amino acids are molecules containing both amino and carboxylic acid groups. There are basically twenty in number namely; glycine **1**, alanine **2**, serine**3**, threonine **4**, cysteine **5**, valine**6**, leucine**7**, isoleucine **8**, methionine **9**, proline**10**, phenylalanine **11**, tyrosine **12**, tryptophan **13**, aspartic acid **14**, glutamic acid **15**, asparagine **16**, glutamine **17**, histidine**18**, lysine **19**, arginine **20**, and of all these, glycine**1** is the simplest, *Young (1994)*. Out of these, nine of them are classified as essential amino acids, because they cannot be synthesized by the body and are therefore required to be taken in diets namely; histidine, leucine, isoleucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine according to Dietary Reference Intakes (2014). The non-essential amino acids can

be synthesized in the body and they include alanine, aspartic acid, asparagine, glutamic acid, serine, while arginine, cysteine, glycine, glutamine, proline, and tyrosine are classified as conditionally essential amino acids, meaning their synthesis can be limited under special pathophysiological conditions, such as prematurity in the infant or individuals in severe catabolic distress, *Young (1994) and* Dietary Reference Intakes (2014).

Amino acids are the building blocks for protein synthesis, through the formation of peptides linkages according to Wilson (2016). The structures of the twenty (20) alpha-amino acids are shown in fig 1 below.
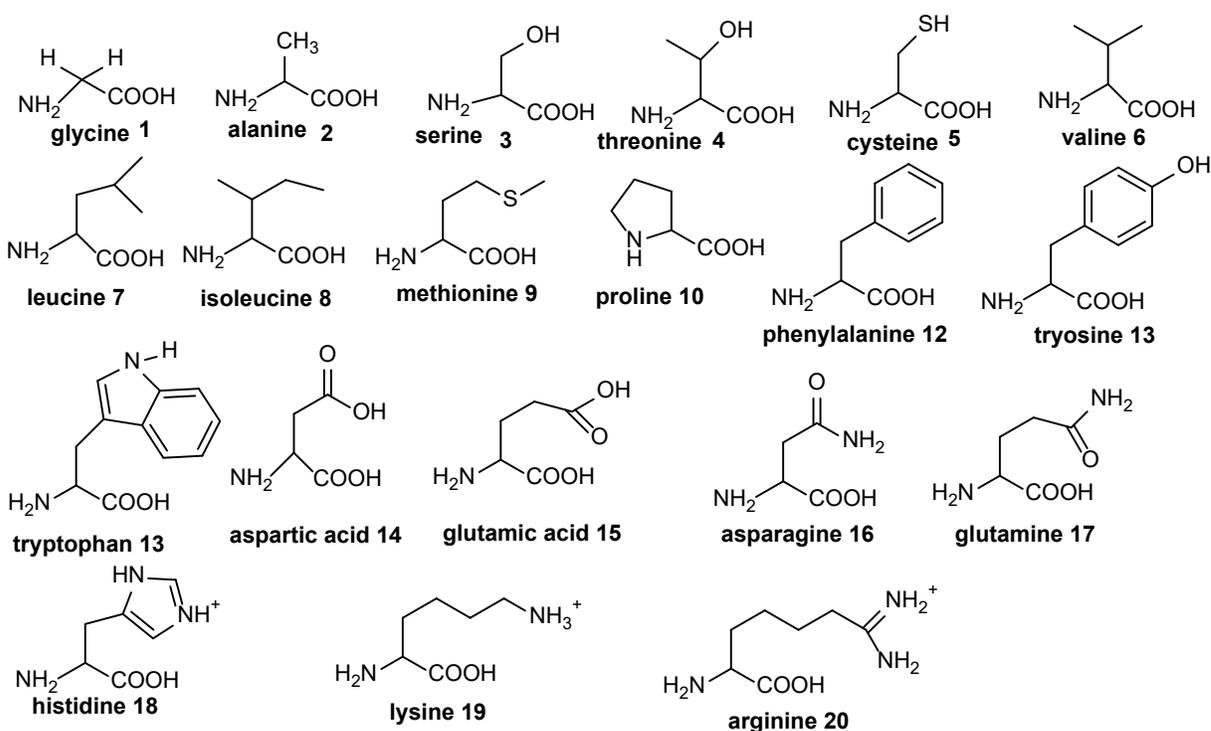

Figure 1: Structures and names of the 20 amino acids

Glycine **1** is the simplest and a conditionally essential amino acid; its chemical formula is $C_2H_5NO_2$. It is a white solid with density of 1.607g/mol. It is soluble in pyridine, sparingly soluble in ethanol and insoluble in ether. It was first produced by a French chemist, H. Braconnot from acid hydrolysis of protein in 1820, according to Wang *et al* (2013). It has a sweet taste like glucose and can also be produced by alkaline hydrolysis of meat and gelatin with potassium hydroxide. Because of its simplicity, it has only one form, unlike other amino acids that possess the L and D isomers.

Wu (2009) reported that glycine supports healthy kidney and liver function as well as the nervous system and serves as a major constituent in extracellular structural proteins (collagen and elastin) in animals. Although glycine has been traditionally classified as a "nutritionally conditionally essential amino acid" for mammals (including humans, pigs and rodents) due to the presence of its endogenous synthesis in the body according to Wu, (2010), and Darling *et al.*(1999), it has been reported that the amount of glycine synthesized in vivo is insufficient to meet metabolic demands in these species according to Jackson (1991), Melendez-Hevia*et al.* (2009), and Rezaei*et al.,* (2013).

Other functions of glycine include: protection of the body against hyper toxicity by effectively and successively fighting against ethanol induced toxicity according to Senthilkumar *et al* (2004), Zeb and Rahman (2017), an effective therapy for shocks, Abello *et al* (1994), treatment of gastric ulcer by decreasing the acid secretions caused by pylorus ligation, prevention of organ transplanting failure (kidneys) when treated with a solution containing glycine and Carolina, Zeb and Rahman (2017). This mixture helps to protect the kidneys against storage injury as well as long survival after kidney transplantation Yin *et al* (2002). Glycine is a very successful immunomodulatory that suppresses inflammation. It also prevents aging in human system. Glycine could also help in the correction of erectile dysfunction, enables proper circulation of blood, helps in cholesterol reduction, prevention of diabetes, hair loss, insomnia and menopause, boosting of the immune system, quickens surgery recovery, improves fertility, it also helps

in weight loss and well-being. Shortage of glycine in small quantities is not harmful for health but severe shortage may lead to failure of immune response, low growth, abnormal nutrient metabolism as well as other undesirable effects on health, Lewis *et al* (2005). A typical example of a glycine derivative that can bring about reduction of cholesterol level in the body is dimethylglycine **21**.
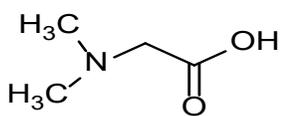
Dimethyl glycine 21

Figure 2: Structure of dimethylglycine

Esters are products obtained from the reaction of carboxylic acids and alcohols with the elimination of a water molecule by the process of esterification as shown in the reaction scheme below IUPAC, "in the Gold Book (1997).
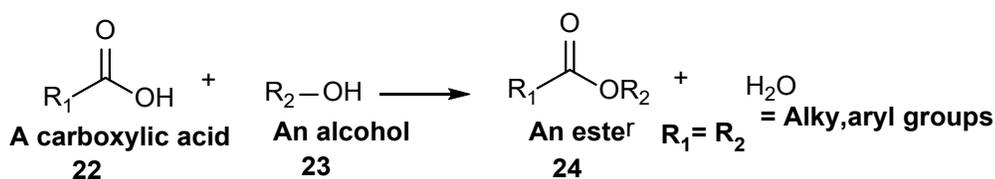

Figure 3: Reaction scheme for esters synthesis

Amides **26** on the other hand are compounds derived from the reaction of a carboxylic acid and an amino compound where a carboxylic acid group, and in an amide is replaced by the –NH of an NH_2 group as shown below, Montalbetti and Falque (2005), Smith and March (2007).
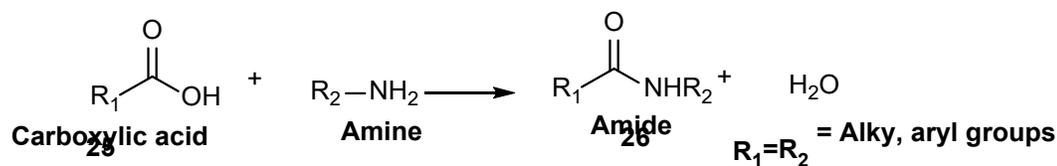

Figure 4: Reaction scheme for amides synthesis

Amide derivatives have been reported to possess broad spectrum of biological activities such as antituberculosis, Mohamed *et al* (2007), anticonvulsant, Nadeem*et al* (2008), analgesic, anti-inflammatory, Galewicz-Walesa *et al* (2003), insecticidal, Graybill*et al* (1992), antifungal, Miheala*et al* (2008), and antitumor properties, Andre *et al* (2007). Compounds containing amide functionalities have proven to be potentially active against various fungal strains and many of them have got wide acceptance in clinical trials according to Ledmicer and Mitschen (1980); Delegado and Remars (2004). They are considered as pro-drugs, biologically inactive compounds which can be metabolized in the body to produce drug activity, Surrender et al (2010).

Cyclooxygenase (COX) officially known as prostaglandin-endoperoxide synthase (PTGS) is an enzymethat is responsiblefor formation of prostanoids, including thromboxane and prostaglandins such as prostacyclin, from arachidonic acid, Kristina et al (2006). Various prostaglandin synthases then convert PGH_2 into several different prostaglandins and thromboxanes, Liu et al (2006). These prostaglandins and thromboxanes target specific G protein-coupled receptors and play major roles in regulation of renal function, platelet aggregation, protection of the stomach lining, and other numerous biological tasks, as well as mediation of the cellular inflammatory response, Kristina et al (2006) and Liu et al (2006). These functions are attributed mainly to the first of the two established COX isoforms, the COX-1, while the inflammatory response is largely associated with the inducible isoform, COX-2. Pharmaceutical inhibition of COX can provide relief from the symptoms of inflammation and pain. Those that are specific to the COX-2 isozyme are called COX-2 inhibitors. For example the active metabolite (AM404) of paracetamol believed to provide most or all of its analgesic effects is a COX inhibitor, and this is believed to provide part of its

effect, Liu et al (2006) and *Högestätt et al (2005)*. Inhibition of COX-2 produces the analgesic, antipyretic, and anti-inflammatory effects typical of non-steroidal anti-inflammatory drugs (NSAIDs), while inhibition of COX-1 is responsible for the antithrombotic effects of aspirin and other nonselective NSAIDs, as well as many of their side effects, such as gastric ulcer formation. The many therapeutically useful effects of COX inhibition have made the NSAIDs among the most widely used drugs of the past century according to *Högestätt et al (2005)*. Since selective COX-2 inhibition can provide analgesic and anti-inflammatory effects with reduced undesirable gastric side effects, COX-2 selective inhibitors such as celecoxib and rofecoxib have become some of the most widely used prescription medications in the developed world. However, recent reports that COX-2 selective inhibitors may increase the risk of heart attack in some patients has caused great concern, and stimulated increased interest in these enzymes, Masferrer et al (1994) and Solomon et al (2002).
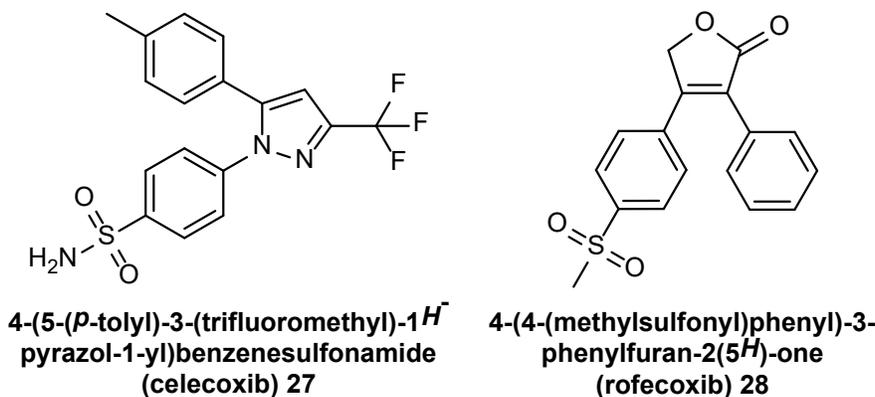

4-(5-(*P*-tolyl)-3-(trifluoromethyl)-1H^- pyrazol-1-yl)benzenesulfonamide (celecoxib) 27

4-(4-(methylsulfonyl)phenyl)-3-phenylfuran-2(5H)-one (rofecoxib) 28

Figure 5: Structures of Celecoxib and Rofecoxib

In this paper we have reported the successful synthesis as well the determination of the binding energies of novel ethyl-glycinate amide derivatives with the COX-2 construct which was used to obtain the 1CVU crystal structure for molecular docking in order compare their binding energy with the standard drugs, celecoxib and rofecoxib respectively used as COX-2 inhibitors. This is to determine if these derivatives could also serve as good drug molecules that can inhibit COX-2 enzyme or not.

2. EXPERIMENTAL

All the reagents were purchased from commercial supplier, Aldrich, and were used without further purification. Melting points were determined with electro thermal melting points apparatus in open capillaries and are uncorrected. UV and Visible spectra were recorded in DMF on a Jenway 6405 UV/Vis spectrophotometer, using matched 1cm quartz cell. IR spectra in (KBr) on a FTIR (NARICT, Zaria), ^{1}H–NMR and ^{13}C-NMR on a JEOL Associate E-400 instrument (chemical shift are reported on the δ scale relative to tetramelthylsilane (TMS) as an internal standard) and mass spectra on a Shimadzu QP2010 spectrophotometer. Analytical samples were obtained by column chromatography on aluminum oxide 90 (Merck, 70–230 Mesh ASTM) employing ethano-chloroform (9:1) as eluting solvent before recrystallization. The 3.0 Å resolution X-ray crystal structure of the ovine COX-1/AA complex, pdb entry 1CVU, was used to generate the initial model.

2.1. ETHYL GLYCINATE 29

The compound glycine ethyl ester **98** was prepared by using the esterification reaction of glycine **1** with ethanol in the presence of hydrochloric acid as a catalyst, Jiabo and Yaowu (2008) and Fischer and Speier (1895). A mixture of glycine (15.0g, 0.15mol), ethanol (200ml) and conc. hydrochloric acid (7ml) was refluxed for 15h, at 78°C. At the end of the reaction, the mixture was placed in a water bath and evaporated. The evaporated product was kept in an airtight desiccator for a week and a crystalline whitish product was obtained. This was later recrystallized from ethanol mixed with a little quantity of diethyl ether to precipitate the final product (14.50g, 96.5%). This was further dried for some days and brilliant white crystals were obtained, melting at 188. 2°C.The UV-Vis: λmax, 334nm (ε = 8.4). IR (KBr): 3119cm-1 (N-H stretching), 2838cm-1 (C-H stretching), 1718 cm-1 (C=O stretching), 1490cm-1(C-N

stretching), 1244cm-1 (C-O stretching).1H-NMR (DMSOd6) δ ppm: 8.70 (singlet, 2H, 1° amine), 4.16 (triplet, 2H, methylene protons), 4.04(singlet, 2H, methylene protons), 1.2 (doublet, 3H, methyl protons). 13C-NMR (DMSOd6) δ ppm: 167.6 (1C, carbonyl), 61.0 (1C, methylene), 41,8 (1C, methylene), 14.6 (1C, methyl).

2.2. 2-AMINO-*N*- (4-NITROPHENYL) ACETAMIDE (31)

The compound, 2-amino-*N*-(4-nitrophenyl) acetamide **31** was prepared by the reaction of the synthesized glycine ethyl ester **29** (2.0g, 0.02mol) and 4-nitroaniline **30** (2.0g, 0.01mol) with the stoichiometric ratio of 2:2. Both compounds were dissolved in (50ml) ethanol and boiled under reflux for 4h. The crude product was obtained with the help of a rotary evaporator under reduced pressure. This was dried and subjected to column chromatography using a mixture of ethanol and chloroform (9:1) as eluting solvent followed by recrystallization, to give a brown-yellowish compound **31**, yield 3.76g (94.0%), melting point, 186.2°C. UV-Vis -$\lambda_{max}(\varepsilon)$ 344nm (18.0) 364nm (19.5) 425nm (22.2) 445nm (21.6) 465nm (21.3).IR (KBr): 3481cm-1(N-H stretching), 2838cm-1(C-H stretching), 1751cm-1(C=O stretching) 1625cm-1(C=C stretching), 1584cm-1(C-N stretching), 913cm-1(C-H bending out of plane of aromatic ring).1H-NMR (DMSOd6) δ ppm: 10.23 (singlet, 1H, amide proton), 8.70 (triplet, 2H, 1°amine protons), 8.17 (doublet, 2H, aromatic protons), 7.82 (doublet, 2H, aromatic protons), 3.85 (triplets 2H, methylene protons). 13C-NMR (DMSOd6) δ ppm: 168.5 (1C, carbonyl), 144.6 (1C, Aromatic-C-NO2), 143.5 (1C, Aromatic-C-N), 124.1 (2C, aromatic), 119.9 (2C, aromatic), 43.2 (1C, methylene).

2.3. 2-AMINO-*N*-(6-METHYLPYRIDIN-2-YL) ACETAMIDE 33

The compound 2-Amino-*N*-(6-methylpyridyl) acetamide **33** was prepared by the reaction of ethyl glycinate 2**9** (2.0g, 0.02mol) and 2-amino-6-methylpyridine **32** (2.0g, 0.02mol) in the ratio of 2:2 respectively. These compounds were dissolved in 50mlethanol and boiled under reflux for 4h. The crude product was obtained with the help of a rotary evaporator under reduced pressure. This was dried and subjected to column chromatography using a mixture of ethanol and chloroform (9:1) as eluting solvent followed by recrystallization, to give a whitish crystalline compound **33**, yield 3.56g (89.0%), melting at 202.1°C. UV-Vis -$\lambda_{max}(\varepsilon)$; 354nm (15.09); IR (KBr): 3090cm-1(N-H stretching), 2806cm-1 (C-H stretching), 1662cm-1(C=O stretching), 1572cm-1(C=N stretching), 924cm-1 (C-Hbending of aromatics).1H-NMR (DMSOd6) δ ppm: 11.14 (singlet, 1H, amide proton), 8.70 (triplet, 2H, 1°amine protons), 8.17 (doublet, 2H, aromatic protons), 7.96 (singlet, 1H, aromatic proton),7.89 (singlet, 1H, aromatic proton), 6.88 (singlet, 1H, aromatic proton), 3.85 (triplets 2H, methylene protons);13C-NMR (DMSOd6) δ ppm: 168.5 (1C, carbonyl), 151.5 (2C, aromatic-C-N), 149.5 (1C, aromatic-C-N), 143.5 (1C, C-NO2), 125.5 (1C, aromatic), 112.8 (2C, aromatic), 43.2 (1C, methylene), 23.9 (1C, methyl).

2.4. *N, N'*-(1,4-PHENYLENE)-*BIS*-(2-AMINOACETAMIDE) 35

The compound 1,4-(2,2-diamino-*N, N'*-phenyl) diacetamide **35** was prepared by the reaction of glycine ester 2**9** (2.0g, 0.02mol) and 1,4-diamino-*N, N'*-benzene **34** (1.0g, 0.01mol) with the ratio of 2:1 respectively. Both compounds were dissolved in 50ml ethanol and left to boil under reflux for 4h, thereafter the crude product was obtained with the help of a rotary evaporator under reduced pressure. This was dried and subjected to column chromatography using a mixture of ethanol and chloroform (9:1) as eluting solvent followed by recrystallization, to give a black compound **35**, yield 2.84g (94.7%), melting at 235.4°C. UV-Vis -$\lambda_{max}(\varepsilon)$; 354nm (20.3) 420nm (24.8) 547nm (28.7) 597nm (29.6); IR (KBr): 3451cm-1(N-H stretching), 2944cm-1(C-H stretching), 1668cm-1(C=O stretching), 1386cm-1 (C-N stretching), 682cm-1 (C-H bending of aromatics).1H-NMR (DMSOd6) δ ppm: 10.23 (singlet, 2H, amide protons), 8.70 (triplet, 4H, 1°amine protons), 7.57 (singlet, 4H, aromatic protons), 3.85 (triplets 4H, methylene protons); 13C-NMR (DMSOd6) δ ppm: 168.5 (2Cs, carbonyl), 134.1 (2C, aromatic-C-N), 121.8 (4C, aromatic-C-N), 43.2 (1C, methylene).

2.5. *N, N'*-(6-CHLOROPYRIMIDINE-2,4-DIYL)-*BIS*-(2-AMINOACETAMIDE) 37

The compound 2,6-(2,2-diamino-*N, N'*-4-chloropyrimidyl) diacetamide **37**was prepared by the reaction of ethyl glycinante 2**9** (2.0g, 0.02mol) and 2,6-diamino-4-chloro-pyrimidine **36** (1.0g, 0.01mol) with the ratio of 2:1. Both

compounds were dissolved in 50ml ethanol and left to boil under reflux for 4h, thereafter the crude product was obtained with the help of a rotary evaporator under reduced pressure. This was dried and subjected to column chromatography using a mixture of ethanol and chloroform (9:1) as eluting solvent followed by recrystallization, to give a whitish compound **37**, yield 1.48g (49.6%), melting at 168.6°C. UV-Vis -$\lambda_{max}(\varepsilon)$; 344nm (3.6). IR (KBr): 3451cm^{-1}(N-H stretching), 3011cm^{-1}(C-H stretching), 1684cm^{-1}(C=O stretching) 1628cm^{-1} C=N stretching,874cm^{-1} (C-H bending of aromatics). ^{1}H-NMR (DMSOd$_6$) δ ppm: 11.14 (singlet, 1H, amide protons), 10.30 (singlet, 1H, amide protons) 8.70 (triplet, 4H, 1°amine protons), 8.14 (singlet, 4H, aromatic protons), 3.80 (singlet 4H, methylene protons); ^{13}C-NMR (DMSOd$_6$) δ ppm: 168.5 (2Cs, carbonyl), 161.4 (1C, aromatic-Cl), 147.6 (1C, aromatic-C-N), 105.5 (2C, aromatic), 43.2 (2C, methylene).

2.6. 2-AMINO-*N*-(4-HYDROXY-6-UREIDOPYRIMIDIN-2-*YL*) ACETAMIDE 39

The compound 2,4-(diamino- *N, N'*-6-hydroxypyrimidyl) diacetamide **39** was prepared from the reaction of ethyl glycinate **29** (2.0g, 0.02mol) and 2,4-diamino-6-hydroxypyrimidine **38** (1.0g, 0.01mol) with the ratio of 2:1. Both compounds were dissolved in 50ml ethanol and left to boil under reflux for 4h, thereafter the crude product was obtained with the help of a rotary evaporator under reduced pressure. This was dried and subjected to column chromatography using a mixture of ethanol and chloroform (9:1) as eluting solvent followed by recrystallization, to give a whitish crystalline compound **39**, yield 2.77g (94.7%), melting at 227.3°C.UV-Vis -$\lambda_{max}(\varepsilon)$; 359nm (19.9); IR(KBr): 3324cm^{-1} O-H stretching, 3281cm^{-1} (N-H stretching), 2993cm^{-1}(C-H stretching), 1673cm^{-1}(C=O stretching), 1506cm^{-1} (C=N stretching) 1133cm^{-1}(C-O stretching), 883cm^{-1} (C-H bending).^{1}H-NMR (DMSOd$_6$) δ ppm: 11.62 (s, 1H, OH), 11.14 (singlet, 1H, amide protons), 10.30 (singlet, 1H, amide protons) 8.70 (triplet, 4H, 1°amine protons), 7.37 (singlet, 4H, aromatic protons), 3.80 (singlet 4H, methylene protons); ^{13}C-NMR (DMSOd$_6$) δ ppm: 168.5 (2Cs, carbonyl), 167.5 (1C, aromatic-OH), 151.3 (1C, aromatic-C-N), 138.8 (1C, aromatic-C-N), (101.3 (1C, aromatic), 43.2 (2C, methylene).

2.7. MOLECULAR DOCKING EXPERIMENTS

2.7.1. PREPARATION OF LIGANDS

The docking studies were carried out on a Zinox Laptop, Model T5101. ACD/ Chemdraw 2015 (Ref: ACD/Structure Elucidator, version 15.01, Advanced Chemistry Development, Inc., Toronto, ON, Canada, www.acdlabs.com, 2015.) was used to draw the structures of compounds 31-39 (Figs. 6 ,7,10, 13, 16, 19 and 22) and also convert them to 3D formats.

2.7.2. PREPARATION OF PROTEIN TARGETS

The 3D structure of the cyclooxygenase active site of COX-2(PDB: 1CVU) was retrieved from the RCSB Protein Data Bank (PDB) (www.rcsb.org/pdb/home/home.do), Picot et al (1994). All bound ligands, cofactors, andwater molecules were removed from the proteins using Discovery Studio Visualizer v16. 1.0. 15,350. All file conversions required for the docking study were performed using the open source chemical toolbox. Open Babel version 2.3.2 (www.openbabel.org). Finally Auto Dock was used to calculate the binding free energy of a given inhibitor conformation in the macromolecular structure.

Figure 6: Structure of Cyclooxygenase-2 (+-PDB: 1CVU)

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3. RESULT AND DISCUSSIONS

The synthesis of ethyl glycinate **29** was achieved through Fischer esterification protocol, Jiabo and Yaowu (2008) and Fischer and Speier (1895) whereas its amide derivatives; 2-amino-*N*-(nitrophenyl)acetamide **31**, 2-amino-*N*-(6-methylpyridin-2-yl) acetamide **33**, *N,N'*-(1,4-phenylene)bis-(2-aminoacetamide) **35**, *N,N'*-(6-chloropyrimidine-2,4-diyl)bis-(2-aminoacetamide) **37**, and 2,4-(diamino-*N,N'*-6-hydroxypyrimidyl)acetamide **39** respectively were synthesized by coupling reactions of the following starting reagents namely; 4-nitroaniline, 2-amino-6-methylpyridine, 1,4-diamino-N,N'-benzene, 2,6-diamino-4-chloropyrimidine and 2,4-diamino-6-hydroxypyrimidine respectively with the key intermediate, ethyl glycinate **29**, FitzGerald (200). These compounds were characterized on the basis of their melting points, UV-Visible, IR, ¹HNMR and ¹³CNMR spectroscopic analyses. The results obtained from the spectra were consistent with the assigned structures of the compounds. The ¹HNMR and ¹³CNMR spectra as well as the reaction schemes for the synthesis of the above compounds are outline in the Figs (6-22) below;
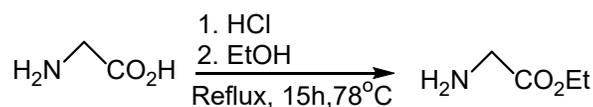
Figure 7: Reaction scheme for the synthesis of ethyl glycinate
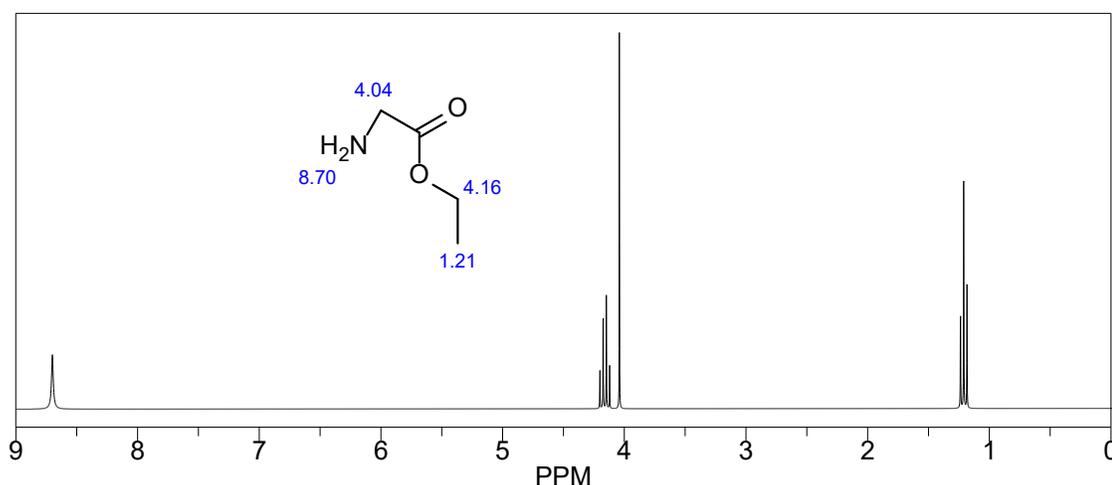
Figure 8:¹HNMR spectrum of ethyl glycinate **(29)**
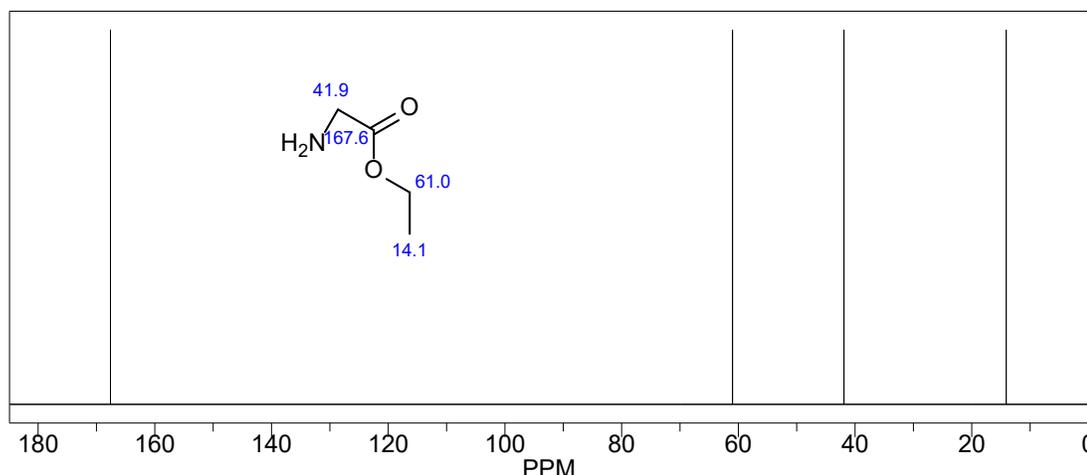
Figure 9: ¹³CNMR spectrum of ethyl glycinate **(29)**
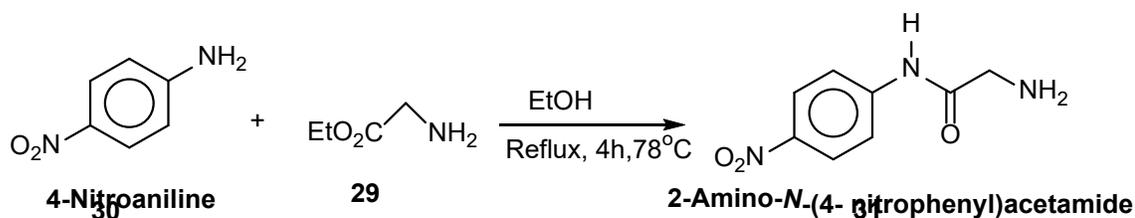
Figure 10: Reaction scheme for the synthesis of 2-amino-*N*-(4-nitrophenyl) acetamide **(31)**
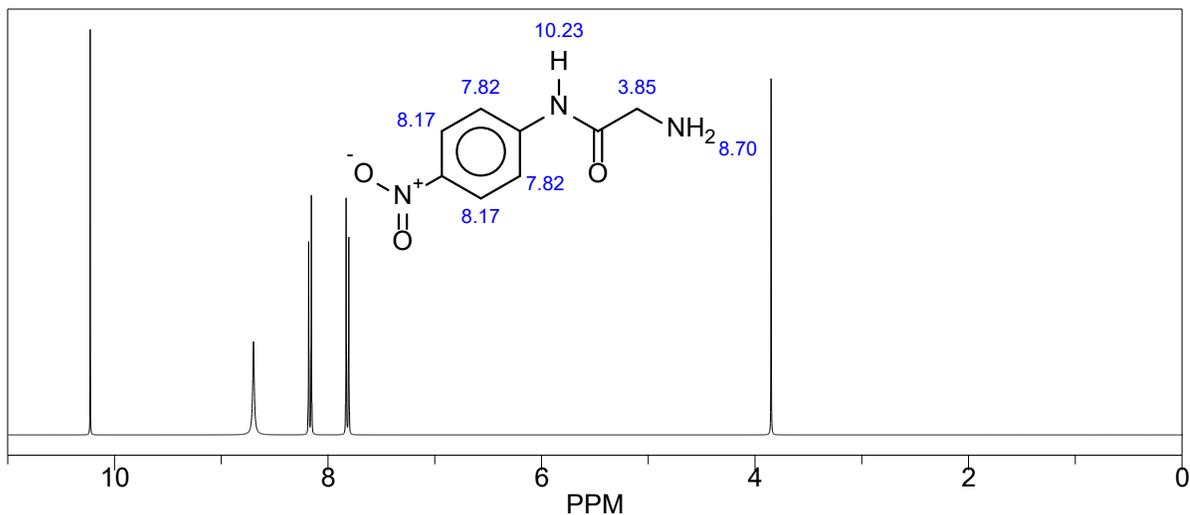

Figure 11:¹HNMR spectrum of 2-amino-*N*- (4-nitrophenyl) acetamide **(31)**
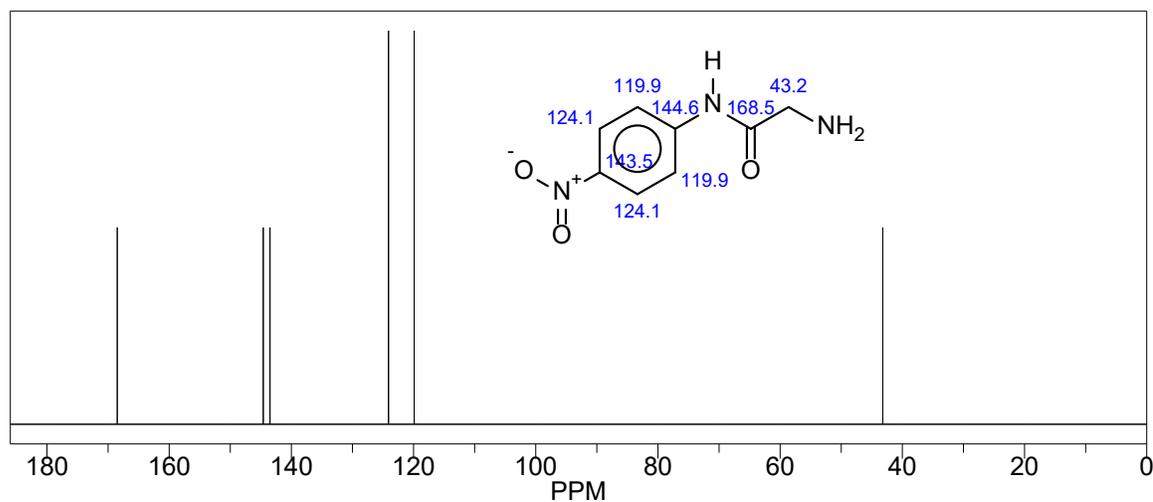

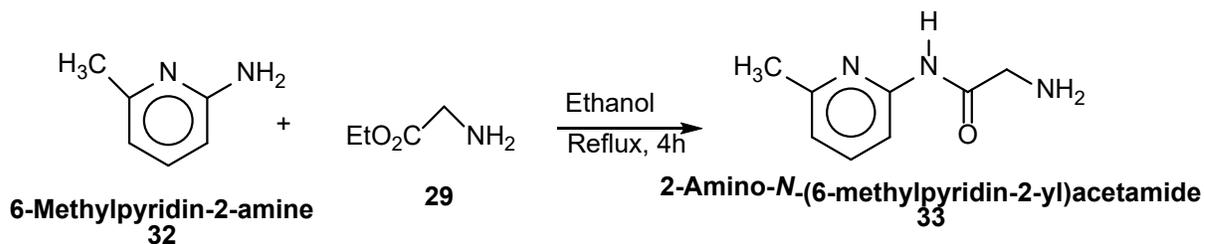**Figure 12:**¹³CNMR spectrum of 2-amino-*N*- (4-nitrophenyl) acetamide **(31)**

Figure 13: Reaction scheme for the synthesis of 2-Amino-*N*-(6-methylpyridin-2-yl) acetamide **(33)**
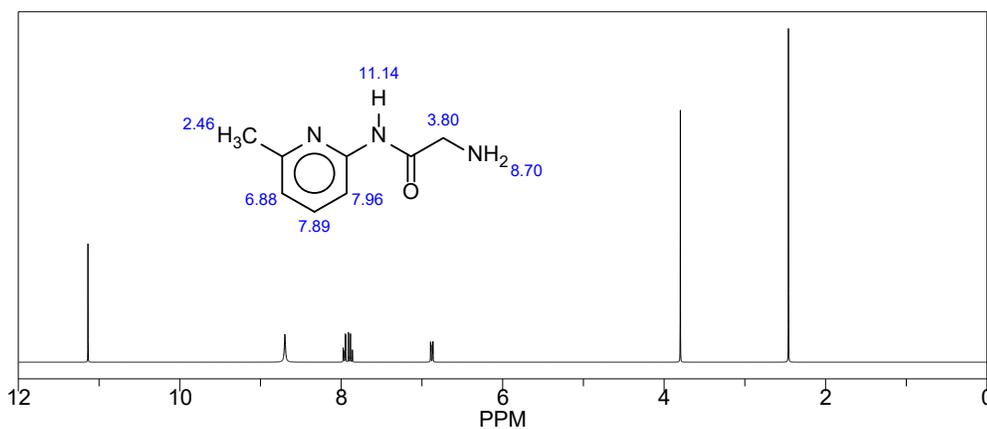
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Figure 14: ¹HNMR spectrum of 2-Amino-*N*-(6-methylpyridin-2-yl) acetamide **(33)**
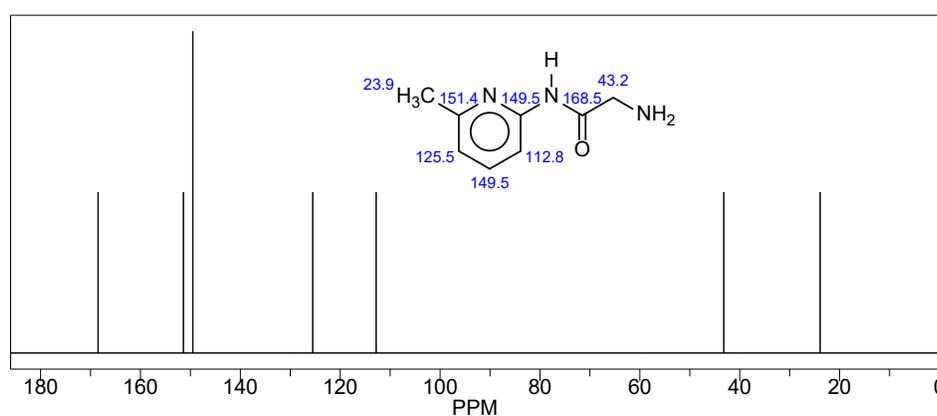
Figure 15: ¹³CNMR spectrum of 2-Amino-*N*-(6-methylpyridin-2-yl) acetamide **(33)**
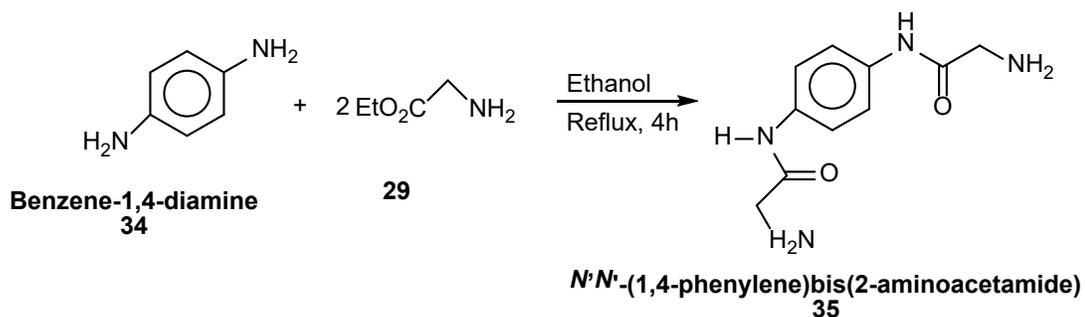
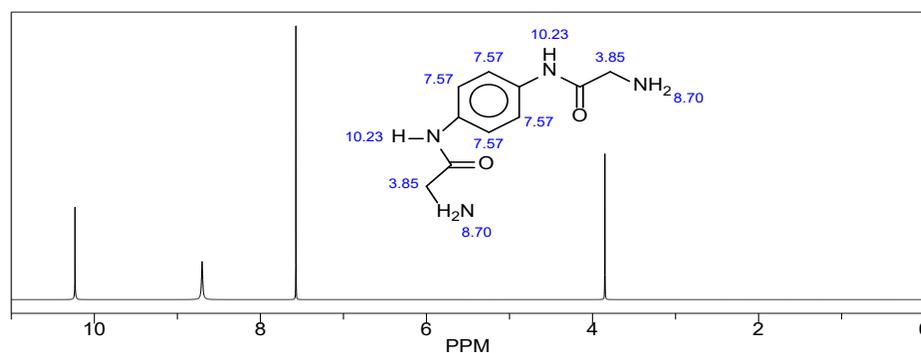**Figure 16:** Reaction scheme for the synthesis of *N, N'*-(1,4-phenylene)-*bis*-(2-aminoacetamide) **(35)**

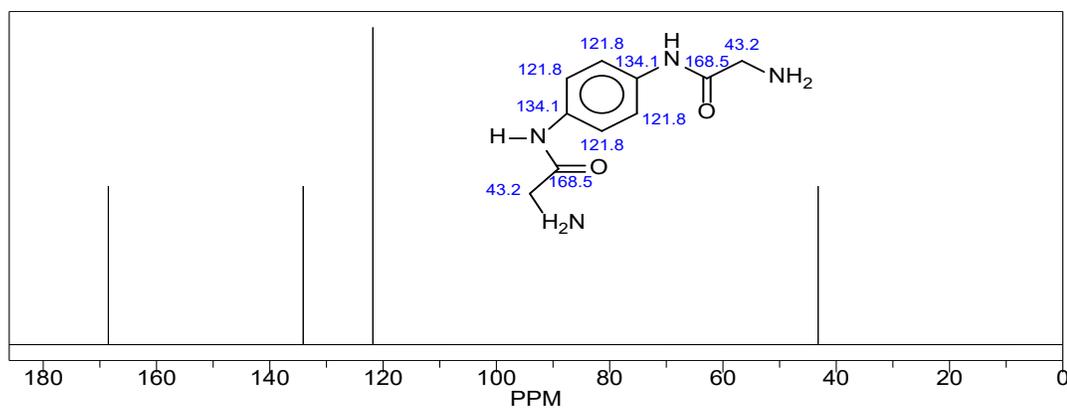**Figure 17:** ¹HNMR spectrum of *N, N'*-(1,4-phenylene)-*bis*-(2-aminoacetamide) **(35)**

Figure 18: ^{13}CNMR spectrum of *N, N'*-(1,4-phenylene)-*bis*-(2-aminoacetamide) **(35)**
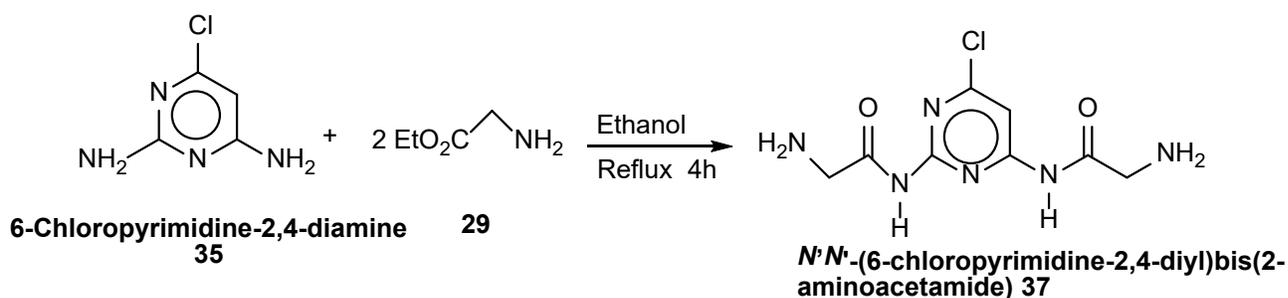

Figure 19: Reaction scheme for the synthesis of*N, N'*-(6-Chloropyrimidine-2,4-diyl)-*bis*-(2-aminoacetamide**)
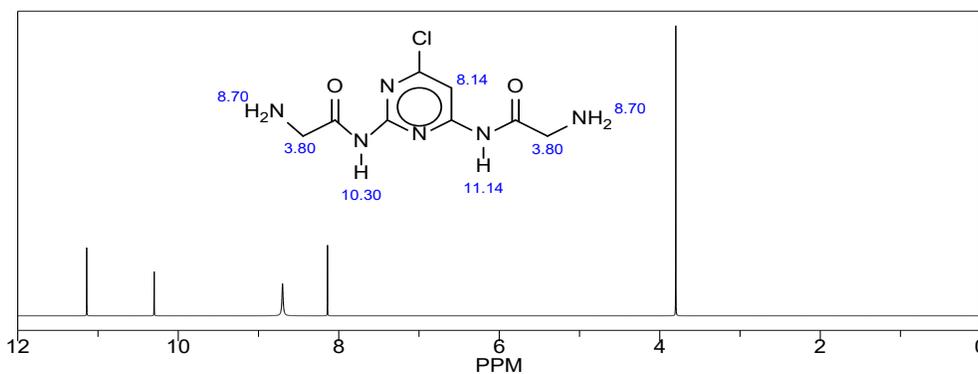(37)**

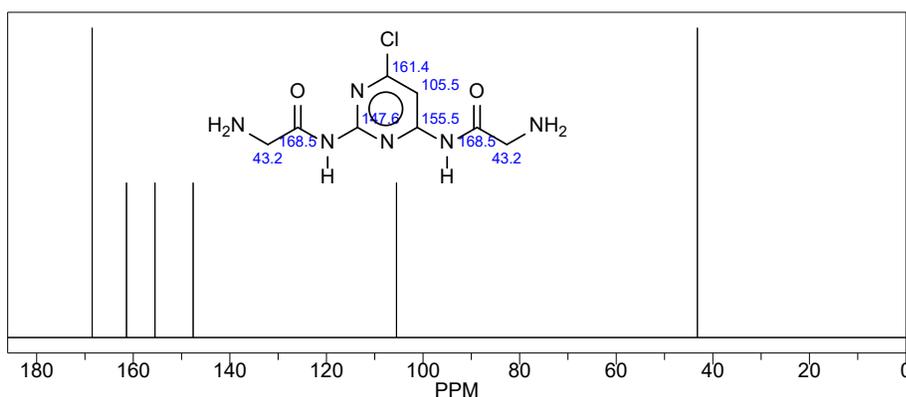**Figure 20:**^{1}HNMR of *N, N'*-(6-Chloropyrimidine-2,4-diyl)-*bis*-(2-aminoacetamide) **(37)**

Figure 21: ^{13}CNMR of *N, N'*-(6-Chloropyrimidine-2,4-diyl)-*bis*-(2-aminoacetamide) **(37)**
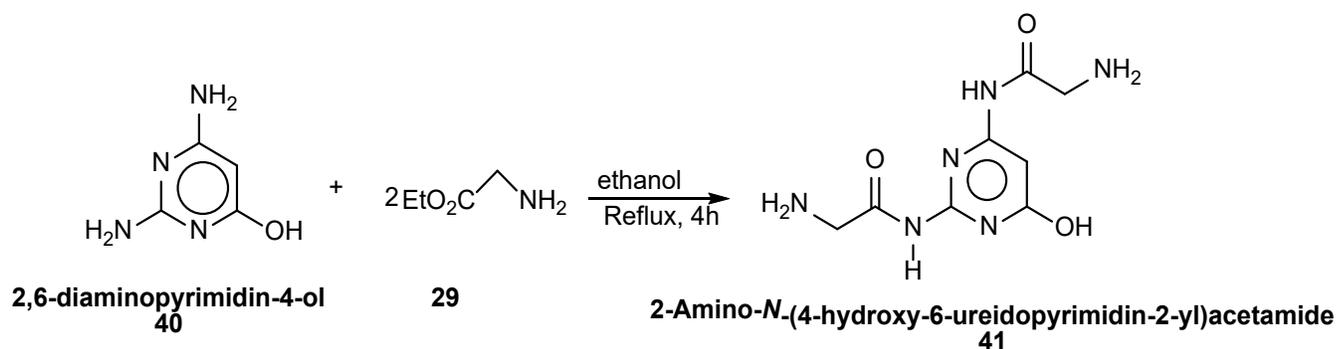
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Figure 22: Reaction scheme for the synthesis of 2-amino-*N*-(4-hydroxy-6-ureidopyrimidin-2-yl)acetamide **(39)**
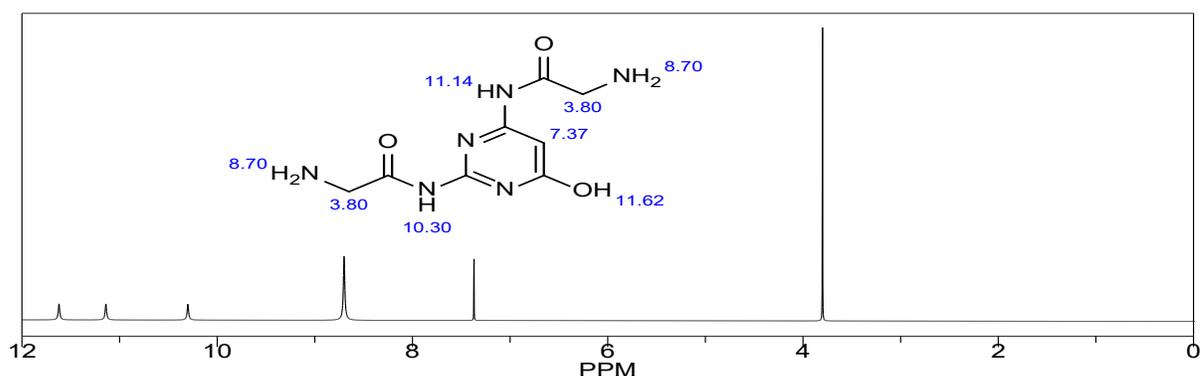

Figure 23: ^{1}HNMR of 2-amino-*N*-(4-hydroxy-6-ureidopyrimidin-2-yl) acetamide **(39)**

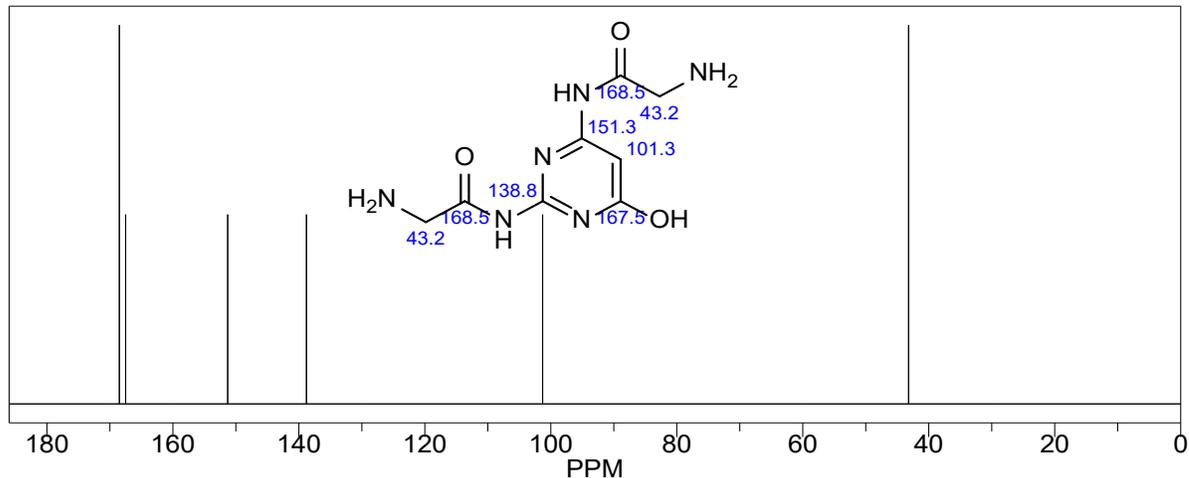**Figure 24**: ^{13}CNMR of 2-amino-*N*-(4-hydroxy-6-ureidopyrimidin-2-yl) acetamide **(39)**

The synthesized compounds were subjected to molecular docking with a target protein, 1CVU to compare their binding energies with celecoxib and rofecoxib which are used as standard drugs for the inhibition of COX2 enzyme, Kurumbail (1994). From the docking result, the binding energy values of the above synthesized compounds were found to be -5.8 kJmol^{-1}, -6.2 kJmol^{-1}, -7.2 kJmol^{-1}, -7.4 kJmol^{-1} and -7.6 kJmol^{-1} respectively as shown in table 1 and the chart in Fig 23 below. These values are actually below that of the standard drugs; however compound **(39)** showed the highest binding energy of -7.6 kJmol^{-1}, close to that for celecoxib and rofecoxib whose values are -8.0 kJmol^{-1} and -8.2 kJmol^{-1} respectively. This result indicates that compound **(39)** possess some level of inhibitory activity against COX2.

Table 1: Binding energy (ΔG (kJ/mol) of the synthesized compounds with target protein, 1CVU

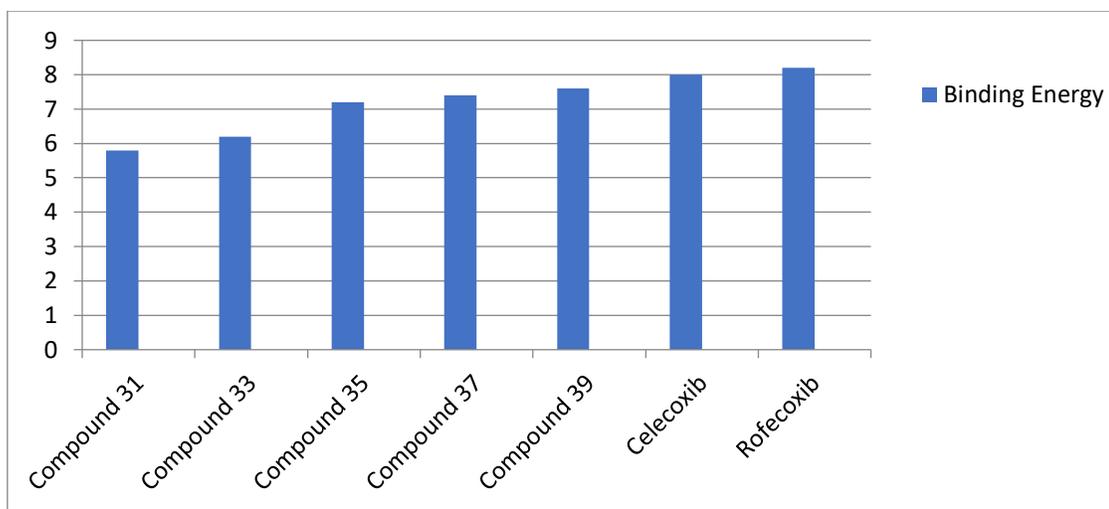| S/No	Compound	Binding Affinity ΔG (kJ/mol)
1	2-Amino-*N*- (4-nitrophenyl) acetamide31	-5.8
2	2-Amino-*N*-(6-methylpyridin-2-yl) acetamide33	-6.2
3	*N, N'*-(1,4-Phenylene)-*bis*-(2-aminoacetamide)35	-7.2
4	*N, N'*-(6-Chloropyrimidine-2,4-diyl)-*bis*-(2-aminoacetamide)37	-7.4
5	2-Amino-*N*-(4-hydroxy-6-ureidopyrimidin-2-yl) acetamide39	-7.6
	Celecoxib (standard COX2 Inhibitor)	-8.0
	Rofecoxib (standard COX2 Inhibitor)	-8.2

Figure 25: A chart comparing the binding energies of the synthesized compounds and the standard inhibitory drugs for COX2, celecoxib and rofecoxib respectively.
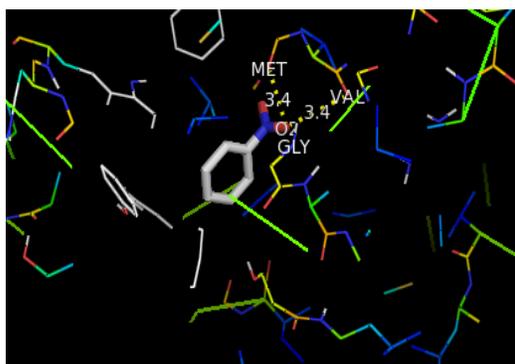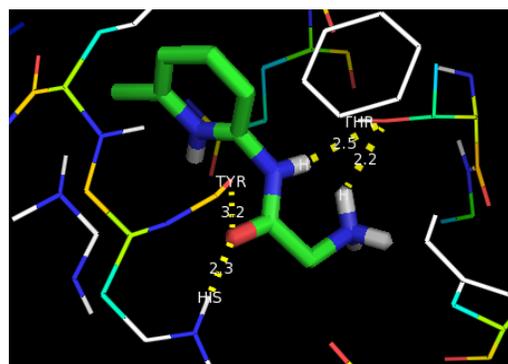

Figure 26: Binding mode of 1CVU–31

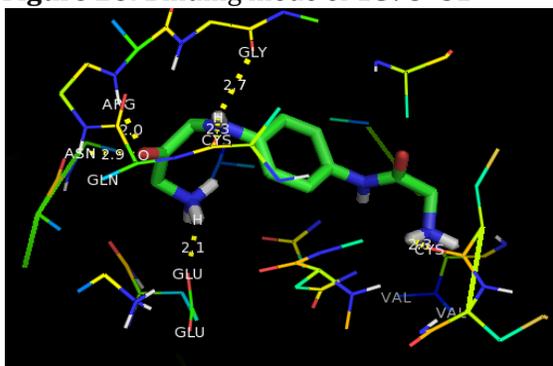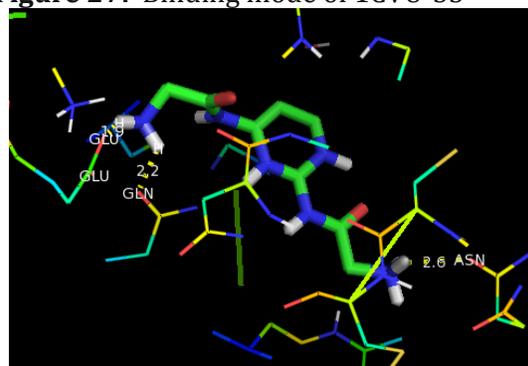
Figure 27: Binding mode of 1CVU-33

Figure 25: Binding mode of 1CVU – 35

Figure 26: Binding mode of 1CVU - 37

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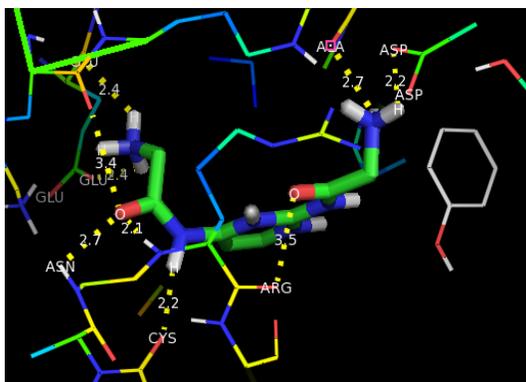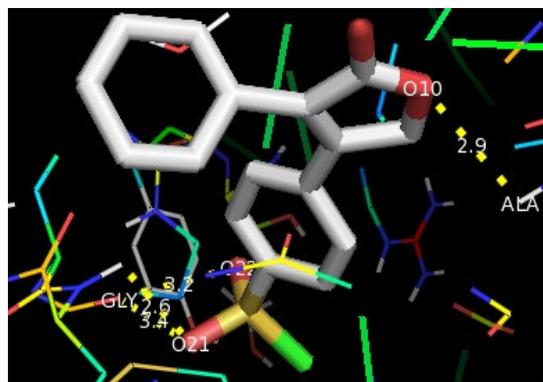
Figure 27: Binding mode of 1CVU–39

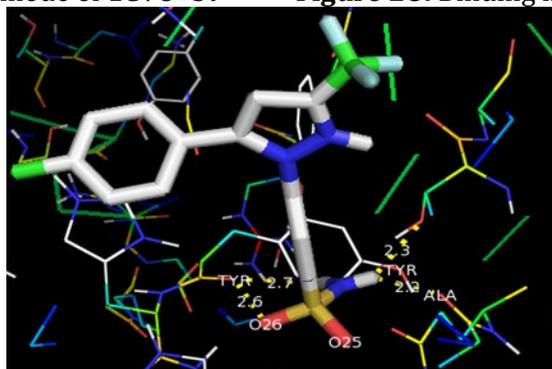**Figure 28:** Binding mode of 1CVU–Rofecoxib

Figure 29: Binding mode of 1CVU–Celecoxib

4. CONCLUSION

Five new derivatives of ethyl glycinate bearing carboxamide pharmacophores have been synthesized and characterized in this work. All the compounds showed appreciable binding energies ranging from − 5.8 to -7.6 kcal/mol with target protein, 1CVU. Compound showed the highest binding energy of 7.6 kcal/mol. Although the binding energies values were not as high as that of the standard drugs used, these novel compounds could be used as starting materials for the synthesis of drugs that can inhibit COX2 enzyme responsible for causing inflammation in the body.

SOURCES OF FUNDING

CONFLICT OF INTEREST

The author have declared that no competing interests exist.

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