DOI: https://doi.org/10.31351/vol34iss4pp66-75

Molecular Docking, Synthesis and Preliminary Anti-microbial Evaluation of Some New Sulfonohydrazide and 1,3,4-Oxadiaxzole Derivatives of Flurbiprofen

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Received 1/6/2024, Accepted 18 /8/2024, Published 20/12/2025

Abstract

A new series of compounds were derived from 2-(2-fluoro-[1,1'-biphenyl]-4-yl) propanoic acid by reacting flurbiprofen ethyl ester with hydrazine hydrate to get flurbiprofen hydrazid(**III**) which then reacted with three different sulfonyl chloride compounds (Benzenesulfonyl Chloride, 4-toleunesulfonyl Chloride, 4-chlorosulfonyl Chloride) in the existence of triethylamine. Additionally, compound (**III**) and carbon disulfide were reacted to yield oxadiazole 2-thione (**compound V**), which was subsequently reacted with 4-chlorophenacyl bromide and 4-bromophenacyl bromide to yield the final products **(VIa** and **VIb)** respectively. For characterization of the final compounds, FT-IR and ^{1}H-NMR spectroscopy were used. Each final compound (**IVa-c,VIa-b**) were examined for their Anti-Microbial activity against G(+)ve bacteria (*Streptococcus pyogenes*, *Staphylococcus aureus*), G(-)ve bacteria (*Escherichia coli, Klebsiella pneumoniae*) and strain of fungi (*Candida albicans*). Compound **(IVc)** showed highest antimicrobial activity against G(-)ve and G(+)ve bacteria and *Candida albicans* fungi. The ADMET study of the resulting compounds exhibited desirable pharmacokinetic properties along with acceptable estimated drug-like properties. molecular docking revealed good molecular docking results with target protein using ciprofloxacin as the reference.

Keywords: Anti-Microbial Activity ,Flurbiprofen, NSAID, Oxadiazole, Sulfonamide, Sulfonyl Chloride.

Introduction

Antimicrobial drug resistance is one of the causes of death and morbidity, which has caused scientists to focus on the rise in multidrug-resistant microbes as a major concern in recent years[(1)]. Numerous non-antibiotic medications with differing degrees of broad-spectrum antimicrobial property have been identified, including: anthelmintics, herbal antimicrobials, anticancer medications, antipsychotics, and antidepressants, antiplatelet drugs, and non-steroidal anti-inflammatory drugs (NSAIDs)[(2)]. NSAIDs are a group of medications that widely used as anti-inflammatory and pain killer[(3)]. Flurbiprofen, a NSAID with anti-inflammatory, antipyretic, and analgesic activity, is commonly used to treat pain, inflammation, rheumatoid arthritis, osteoarthritis, migraines, and acute gout[(4-8)]. Sulfonamides (SNs), with their general formula -SO_2NHR-, are a well-known class in medicinal chemistry with diverse biological activities, including: antifungal, antibacterial, hypoglycemic, diuretic, antithyroid, anti-inflammatory, antiglaucoma, and antineoplastic properties [(9,10)]. Bacterial strains have developed resistance against them, which has somehow hampered their uses [(11)]. Because SNs are not easily biodegradable, they have a number of undesirable side effects, such as respiratory and digestive tract illnesses [(12)].Oxadiazole is an aromatic heterocyclic nucleus with two nitrogen and one oxygen atom[(13-15)]. Oxadiazoles exist in four different isomers, depending on where the nitrogen atoms are located: 1,2,5-oxadiazole, 1,2,4-oxadiazole, 1,2,3-oxadiazole and 1,3,4-oxadiazole[(16)]. 1,3,4-Oxadiazole is the sole isomer of oxadiazole lacking an oxygen-nitrogen link, exhibiting a diverse range of a biological activity [(17)] such as anti-diabetic, anti-malarial, antibacterial, anti-cancer, analgesic, and anti-inflammatory properties, that have attracted a lot of researchers [(18-20)].

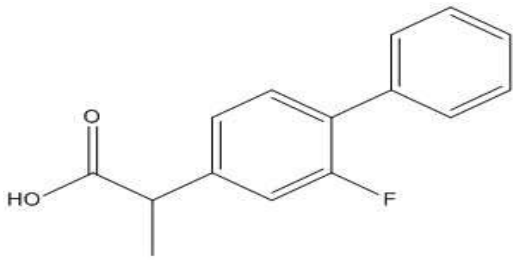


Figure 1. Chemical structure of flurbiprofen

Materials and Methods

Docking Study

The use of the maestro program from licensed Schrodinger in conjunction with the Glide application. The Staphylococcus aureus gyrase enzyme in combination with DNA and ciprofloxacin was downloaded from Protein Data Bank (PDB ID: 2XCT) [21]. To get the amino acid residues into the proper tautomeric state and ionization, remove all water molecules and add hydrogen atoms [22]. The receptor grid was made from the cocrystallized ligand that interacted with the protein. The collection of ligands to be docked was identified and prepared using ligprep. Docking of the prepared ligands to the gyrase enzyme (PDB ID: 2XCT) was done with the default XP docking option, which had a 10-pose limit [23]. To validate the docking, the co-crystallized ligand was re-docked into the binding site using the identical settings as described earlier[24].

ADMET studies

All final compounds are put via ADMET prediction utilizing Qikprobe software in Schrodinger Maestro in order to evaluate drug likeness properties.

Chemical synthesis Materials

Flurbiprofen is gotten from Picasso-e Company, ethanol from Honeywell Company, ,hydrazine hydrate from Thomas Baker Company, dichloromethane from Alpha Chemika company, 4-chlorobenzenesulfonyl chloride and p-toluenesulfonyl chloride are from Himidia Company, Benzene sulfonyl chloride from Nevend New Marerials Company, acetone from ACS Chemicals Company, potassium carbonat from GPR Company, 4-bromophenacyl bromide and 4-chlorophenacyl bromide from Fluka AG Company. The melting points were uncorrected and measured using the Stuart SMP30 apparatus. With the use of Thin-Layer Chromatography (TLC) method with an Aluminum-Precoated silica sheet material (Germany, Merck), the reaction completion was monitored and the purity of the products was assessed. Each target compound dot was observed by a UV 254 nm lamp. The infrared spectra were done at the College of Pharmacy, Bagdad University, utilizing the FT-IR (Shimadzu, Japan) with a measuring unit of ύ, cm^{-1}. ^{1}H-NMR was done using a Brucker model 400 MHz at Basra, Iraq, and DMSO was used as a solvent. The target compounds were tested for antibacterial properties ,minimum inhibitory concentration(MIC) and minimum bactericidal concentration (MBC) at the office located in Baghdad, Iraq named chemistry analysis center(CAC) .

Chemical synthesis

Synthesis of flurbiprofen ester[ethyl 2-(2-fluoro-[1,1'- biphenyl] – 4 – yl)propanoate] Compound II[25,26].

After dissolving flurbiprofen (5g, 0.02mole) in 50 ml of absolute ethanol, 15 drops of H_2SO_4 were added, mixed, and refluxed for 6 hours. Reaction completion is checked by utilizing TLC. The solution was then neutralized by 10% $NaHCO_3$. Diethyl ether was used to separate the oily crude product. To get compound II, the organic layer was evaporated. A yellowish oil product was obtained. Yield: 88%, R_f = 0.8 (ethanol: n-hexane 4:6), Infra-Red (v cm^{-1}): 1732.08:(carbonyl) strch. Ester func. group, 1180:(C-O bond) strch. ether.

Synthesis of flurbiprofen hydrazide [2-(2-fluoro-[1,1'-biphenyl]-4-yl)propanehydrazide]Compound III[27,28]:

Hydrazine hydrate 99% (5 mL, 0.11 mole) was mixed with a solution of compound II (2g, 0.007 mole) in 30 mL of absolute ethanol, and then the combination was refluxed for 20 hours of time. The solution was then cooled to(25°C) temperature ,then decanted over squashed ice with continuous mixing. The solid after that filtered and recrystallized with ethanol. Yield: 85%, MP= (114-116)ºC, R_f = 0.3 (ethanol:n-hexane 4:6), Infra-Red{IR} (v cm^{-1}): 1631 (Carbonyl) strch. of amide functionality, 3309.85, 3282 ((N-H)) strch. of (amine)NH, 3032: (C-H bond) strch. of Ar. C-H.

Synthesis of N'-(2-(2-fluoro-[1,1'-biphenyl]-4-yl) propanoyl) argonsulfinohydrazide (IVa- c) [29]: A mixture of compound III (1g , 0.0038mole), 25 ml of dry dichloromethane, [Benzenesulfonyl chloride (0.67g, 0.0038 mole) ,4-toleunesulfonyl chloride (0.72g ,0.0038 mole), 4-chlorosulfonyl chloride (0.8g, 0.0038mole) respectively] and triethylamine (0.948g, 0.009 mole) was stirred continuously for 30 hours at room temperature.100 ml of distilled water was used to separate the organic layer, which was then evaporated. The powder then recrystallizes by ethanol solvent.

N'-(2-(2-fluoro-[1,1'-biphenyl]-4-yl) propanoyl) benzenesulfonohydrazide(IVa):beige powder, Yield 66%, MP= (180-181)ºC, R_f =0.75 (ethanol, n-hexane 5:5), IR (v cm^{-1}):3302:(N-H) strch. of amideNH,3155 :(N-H) strch. of sulf.NH, 3035: (C-H) strch. of Ar- ring,2974: (C-H) asymm. strch. of CH_3 CH, 2819:(C-H) Symm.strch. of CH_3, CH, 1685:(C=O) strch. of amide,1581: (C=C) strch. of Ar ring,1419:(SO_2) asymm. strch. of S=O,1165:(SO_2) symm. strch. of S=O, 694:(C-S) strch. ^{1}HNMR: 1.39 (3H,dub.- CH_3) , 3.71 (1H, qur. - CH),7.25-7.54(13 H, mul. _Ar-H), 9.16 (H, sig.-amide NH),10.19(H , sig.-sulfonamide NH).

N'-(2-(2-fluoro-[1,1'-biphenyl]-4-yl)propanoyl)-4-methylbenzene sulfonohydrazide(IVb): beige-white powder, Yield = 88% MP= (194-195)ºC, R_f = 0.66 (ethanol, n-hexane 5:5), IR (v cm^{-1}):3348:(N-

H) strch. of amide NH, 3205:(N-H) strch. of Sulf.NH,3059: (C-H) strch. of Ar-H, 2951:(C-H) Asymm. strch. of CH_3, CH , 2870:(C-H) Symm. strch. of CH_3, CH ,1674(C=O) strch. of amide, 1624:(C=C) strch. of Ar ring,1342:(SO_2) Asymm. strch. of S=O,1165:(SO_2) symm. strch. of S=O. ^{1}HNMR,1.22(3H , dub. – CH_3),2.32(3H , sig. – CH_3),3.61 (H , qur. – CH),7.20-7.55(12H , mul. – Ar_H), 9.85(H, sig, -CO-NH),10.21 (H ,sig.-sulf. NH).

4-chloro-N'-(2-(2-fluoro-[1,1'-biphenyl]-4-yl)propanoyl)benzenesulfonohydrazide (IVc): off white powder, Yield =62% MP= (185-186)ºC, R_f=0.8 (ethanol, n-hexane 5:5), IR (v cm^{-1}): IR (v cm^{-1}):3305:(N-H) strch. of amide NH, 3205:(N-H) strch. of Sulf.NH,3059,3035: (C-H) strch. of Ar-H, 2981,2943:(C-H) Asymm. strch. of CH_3, CH , 2823,2758:(C-H) Symm. strch. of CH_3, CH ,1670(C=O) strch. of amide, 1624:(C=C) strch. of Ar. ring,1342:(SO_2) Asymm. strch. of S=O,1157:(SO_2) symm. strch. of S=O. ^{1}HNMR, 1.40(3H, dub., CH_3),3.72(H, qur.-CH) , 7.26-7.55(12H , mul.- Ar_H) , 9.20(H , sig.-amide NH),10.20(H , sig.-sulf. NH).

Synthesis of 5-(1-(2-fluoro-[1,1'-biphenyl]-4-yl)ethyl)-1,3,4-oxadiazole-2-thiol(V)[30]:

(0.22g, 0.004 mole) potassium hydroxide was dissolved in absolute ethanol, and then (1g, 0.004mole) of compound III was gradually added to the solution. At 0 °C, 25 drops of carbon disulfide were added dropwise to the mixture with continuous stirring for 2 hours. reflux for about 17 hours until the hydrogen sulfide's evolution stopped. Concentrated HCl was added dropwise until the solution's pH was reduced to 2-3.. The resulting precipitate was filtered.

Off white powder, Yield =84% MP= (141-142)ºC, R_f=0.8 (ethanol, n-hexane 5:5), IR (v cm^{-1}): IR (v cm^{-1}):3122:(N-H) strch. of thioamide (oxadiazole), 3062: (C-H) strch. of Ar-H, 2924:(C-H) Asymm. strch. of CH_3, CH , 2854:(C-H) Symm. strch. of CH_3, CH ,1612(C=N) strch. of imine, 1581,1562:(C=C) strch. of Ar ring,1269(C=S) strch. of thion,1180:(C-O-C) strch. of cyclic ether group of the ring. ^{1}HNMR, 1.61(3H, dub., CH_3),4.44(H, qur.-CH) , 7.20-7.56(8H , mul.- Ar_H),13.84(1H,sig.-NH of thioamide).

synthesis of S-alkylated derivatives of 5-(1-(2-fluoro-[1,1'-biphenyl]-4-yl)ethyl)-1,3,4-oxadiazole -2-thiol derivatives compounds (VIa,b)[31,32]:After dissolving (0.55 g, 0.004 mole) of potassium carbonate in (30 ml) of dry acetone, compound V (0.9 g, 0.003 mole) was added to the mixture while it was being heated and stirred continuously. After that, (0.003 mole) of aryl halides (4-chlorophenacyl bromide or 4-bromophenacyl bromide) was added, and the mixture refluxed for 25 hours. Chloroform and distilled water were used to separate the organic layer which was then evaporated and the precipitate washed with diethyl ether.

1-(4-chlorophenyl)-2-((5-(1-(2-fluoro-[1,1'-biphenyl]-4-yl)ethyl)-1,3,4-oxadiazol-2-yl)thio)ethan-1-one(VIa):off white powder, Yield =60% MP= (103-104)ºC, R_f = 0.7 (ethanol, n-hexane 5:5), IR (v cm^{-1}): 3059,3032: (C-H) strch. of Ar-H, 2924:(C-H) Asymm. strch. of CH_3, CH , 2854:(C-H) Symm. strch. of CH_3, CH ,1654(C=O) strch. of carbonyl, 1624:(C=N) strch. of imin,1581:(C=C) str. of Ar ring, 1118:(C-O-C) strch. of oxadiazole ring. ^{1}HNMR, 1.62(3H, dub., CH_3),4.53(1H, qur.-CH) ,5.04(2H,sig.-CH) 7.17-8.03(12H , mul.- Ar_H) ***1-(4-bromophenyl)-2-((5-(1-(2-fluoro-[1,1'-biphenyl]-4-yl)ethyl)-1,3,4-oxadiazol-2-yl)thio)ethan-1-one(VIb):*** beige powder, Yield =67% MP= (99-100)ºC, R_f=0.74 (ethanol, n-hexane 5:5), IR (v cm^{-1}): IR (v cm^{-1}): 3052: (C-H) strch. of Ar-H, 2954,2920:(C-H) Asymm. strch. of CH_3, CH , 2850:(C-H) Symm. strch. of CH_3, CH ,1678(C=O) strch. of carbonyl, 1626:(C=N) strch. of imin,1581:(C=C) strch. of Ar ring, 1149:(C-O-C) strch. of oxadiazole ring. ^{1}HNMR, 1.63(3H, dub., CH_3),4.55(H, qur.-CH) ,5.06(2H,sig.,CH), 7.19-7.97(12H , mul.- Ar_H).

Antimicrobial Activity

Antibacteial and antifungal activity of all final compounds (IVa-c,VIa,b) were assessed against two G(-)ve bacteria (*Klebsiella pneumonia* and *E. coli*), two G(+)ve bacteria (*Streptococcus pyogenes* and *Staphylococcus aureus*), and one fungus (*Candida albicans*) with use the disc diffusion method. All of the final compounds were dissolved at a concentration of 1000µg/ML in DMSO. The activity was then verified by measuring the zone of inhibition in millimeters and comparing it to the reference antibacterial (amoxicillin, ciprofloxacin) medications and antifungal (fluconazole) .

Results and Discussion

Docking study

Molecular docking was used to investigate the potential interactions between compounds IVa-c,VIa,b and the gyrase enzyme's active site (PDB: 2XCT). Protein residues, DNA bases, and the Mn^{+2} ion comprise the binding pocket of the benchmark drug, ciprofloxacin. Mn^{+2} ion interaction with the ligand is necessary for the binding process between the ligand and protein.In this work, all compounds have a good docking score but less than that of ciprofloxacin “Table 1”. Compound IVc has highest docking score amongst other compounds which is equal to -7.057 in contrast to ciprofloxacin docking score, -8.164.c which has hydrogen bond interaction between NH of amide and DNA nucleotide DG8. Another hydrogen bond interaction is between oxygen of sulfonyl group and DNA nucleotide DC13. Mn2000 interact with nitrogen of sulfonamide and carbonyl group through metal

coordination. pi-pi stacking interaction between the DNA nucleotide DC13 and the aromatic ring of compound IVc .there is also salt bridge between nitrogen of sulfonamide and Mn2000 "Figure. 2A" .

Ciprofloxacin, on the other hand, exhibits metal coordination interactions between Mn^{2+} and two C=O groups in addition to pi-pi stacking interactions between its aromatic rings and the DNA nucleotides DG8 and DG9 "Figure. 2B".

Table 1. Docking score of final compounds (IVa-c, VIa,b)

Compounds	Docking Score	Interactions					
		metal coordination		H.B		*Pi*- interaction	
		metal	gp.	nucleotide	gp.	nucleotide	gp.
IVa	-6.833	Mn^{+2}	C=O N	DG8	NH	DG8 DG9 Mn^{+2}	ring
IVb	-6.796	Mn^{+2}	C=O S=O	DG8	NH	DG8 DG9	ring
IVc	-7.057	Mn^{+2}	C=O N	DG8 DC13	NH S=O	DC13	ring
VIa	-6.325	Mn^{+2}	C=O	-	-	DA7 DG8 DG8 DG9	Oxadiazole ring ring
VIb	-6.232	Mn^{+2}	C=O	-	-	DA7 DG8 DG8 DG9	Oxadiazole ring ring
Ciprofloxacin	-8.164	Mn^{+2}	C=O	-	-	DG8 DG9	ring

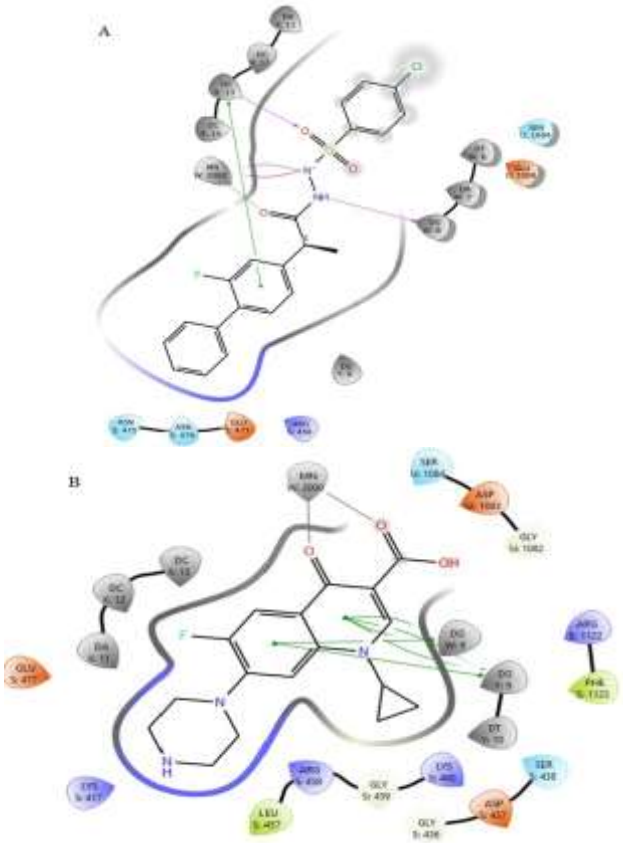


Figure 2. Two-Dimensional docking pose of A: IVc, B: ciprofloxacin into the gyrase active site (PDB:2XCT)

ADMET studies

All of the final compounds possessed the appropriate expected pharmacokinetic parameters (oral absorption, rule of five, rule of three, and metabolism) as shown in "Table 2". Compounds IVa-c have fewer CNS adverse effects than other compounds since they had the lowest CNS activity. There are probably a number of metabolic reactions for every compound. All compounds demonstrated high percentages of human oral absorption, with compounds IVa–c having the highest oral absorption. Compounds VIa and VIb only showed one rule of three and rule of five deviation.

Table 2. ADMET study of final compounds(IVa-c, VIa,b)

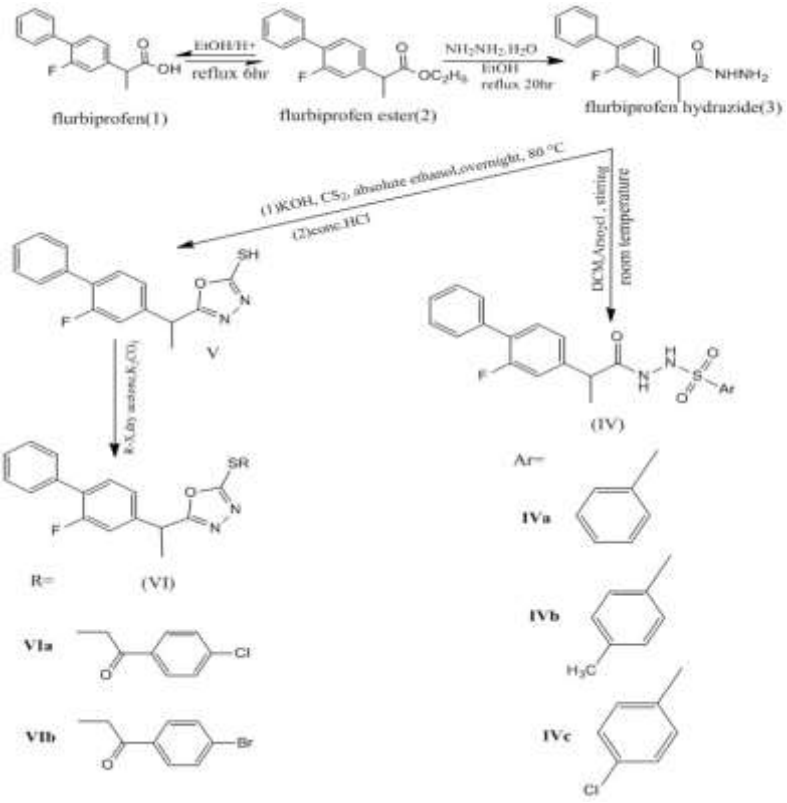
Compound	CNS	#metab	Human Oral Absorption	Percent Human Oral Absorption	Rule Of Five	Rule Of Three
IVa	-1	1	3	100	0	0
IVb	-1	2	3	100	0	0
IVc	-1	1	3	100	0	0
VIa	0	3	1	100	1	1
VIb	0	3	1	100	1	1

CNS; suggested a scale for central nervous system activity ranging from –2 (inactive) to +2 (active). **#metab**; Probable number of metabolic reactions: 1–8. **Human Oral Absorption;** 1, 2, or 3 for low, medium, or high, respectively.**Percent Human Oral Absorption**; >80% is high, <25% is poor. **Rule Of Five**; There are a maximum of four instances of breaking Lipinski's rule of five. **Rule Of Three**; There are a maximum of three violations of Jorgensen's rule of three[23].

Chemistry

The overall synthetic procedure that produced the final compounds (IVa-c,VIa,b) were depicted in Scheme 1. The first step is the production of flurbiprofen ethyl ester II, which is synthesized by reacting flurbiprofen in ethanol in the presence of H_2SO_4. The FT-IR spectra of Compound II showed an absorption band at 1732 cm^{-1} because of the C=O group of the ester and at 1180 cm^{-1} because of the C-O stretching vibration of the ether. Compound II was refluxed with 99% hydrazine hydrate in absolute ethanol to create flurbiprofen hydrazide. Compound III's FT-IR spectra revealed an absorption band at 1631 cm^{-1} of C=O for the amide's stretching vibration and 3309.85, 3282 cm^{-1} for the primary amine's stretching vibration. Compound III was reacted with (benzenesulfonyl chloride, 4-toleunesulfonyl chloride and 4-chlorosulfonyl chloride), respectively in dry dichloromethane in the presence of the triethyamine as a base at room temperature. Derivatives (IVa-c) are derived from sulfonyl chloride. The synthesis was verified by the formation of SO_2 FT-IR absorption bands at 1419,1165 cm^{-1} for compound IVa, peaks at 1342, 1165 cm^{-1} for compound IVb and peaks at 1342, 1157 cm^{-1} for compound IVc . Moreover, absorption bands of the amide's carbonyl C=O are at 1670-1685 cm^{-1} . Additionally, the disappearance of the primary amine's absorption bands at 3309 cm^{-1} and 3282 cm^{-1} and the appearance of the N-C=O absorption bands at 3302 $cm^{-1}$and N-S=O group band of absorption at 3155 cm^{-1} for IVa ,N-C=O group moiety at 3348 $cm^{-1}$and N-S=O at 3205 cm^{-1} for IVb and N-C=O at 3305 cm^{-1} and N-S=O at 3205 cm^{-1} for IVc confirmed the formation of sulfonyl derivatives. ^{1}HNMR spectroscopy revealed a singlet signal of amide's NH at 8.29-9.85 PPM and a singlet signal at 10.19-10.21 PPM representing the proton of the sulfonamide. Compound IVb had two signals for Ar-CH-C=O due to the presence of chiral center that explain R and S configurations.

Compound V was prepared by reacting flurbiprofen hydrazide with carbone disulfide in the presence of potassium hydroxide in absolute ethanol, which was characterized by FT-IR absorption band at 3062 cm^{-1} for thioamide of the oxadiazole ring, 3062 cm^{-1} absorption band of aromatic C-H, 2924 cm^{-1} asymmetric stretching of CH_3, CH. 2854 cm^{-1} for symmetric stretching of CH_3,CH. 1612 cm^{-1} is the imine absorption band of the oxadiazole ring, 1581,1562 cm^{-1} is the absorption band of the C=C of the aromatic ring, and 1269 cm^{-1}C=S stretching vibration of the oxadiazole. However, oxadiazole's ^{1}H-NMR spectra revealed a singlet signal at 13.84 SH. Finally compounds VIa, VIb were prepared by reacting suitable aryl halides with oxadiazole ring in the presence of potassium carbonate in dry acetone. The synthesis was characterized by FT-IR absorption band at 1654,1678 cm^{-1} for C=O moiety stretching vibration of carbonyl group , 1624-1626 cm^{-1} for C=N group stretching of the ring , 1118,1149 cm^{-1} as a result of the ring's cyclic ether stretching. A singlet signal at (5.04-5.06) ppm can be seen in ^{1}H-NMR spectra of VIa and VIb respectively because of two CH_2 protons (-S-CH_2-COC_6H_5-) and the disappearance of the NH signal.



Scheme 1 .Synthesis of target compounds (V_{a-c} , VI_{a-b})

Anti-microbial activity

The synthetic derivatives (IVa-c,VIa,b) were evaluated for their anti-microbial activity against fungi, G(-)ve, and G(+)ve bacteria using the disc well diffusion method, with amoxicillin, ciprofloxacin and fluconazole serving as standards. DMSO was used as a solvent and negative control and, as shown in the accompanying "Table3". All the final compounds have good antifungal activity but amongst them compounds IVc and VIb have more antifungal activity . Compound IVc has the highest activity against G(-)ve and G(+)ve bacterial species in "Table3".

Table 3. The anti-microbial evaluation of target compounds (IVa-c,VIa,b)

Compound	Inhibition zone (mm)					
	Conc. µg/ml	*S. aureus*	*S. pyogenes*	*K. Pneumonia*	*E.Coli*	*C.albicans*
IVa	10^3	24	26	25	25	25
IVb	10^3	22	25	25	25	25
IVc	10^3	25	28	26	38	29
VIa	10^3	22	25	12	20	20
VIb	10^3	23	26	-	-	28
Amoxicillin	10^3	30	35	25	36	-
Ciprofloxacin	10^3	41	43	35	40	-
Fluconazole	10^3					20
DMSO	Control and solvent					

(zone of inhibition more than 15 mm): highly active, (zone of inhibition between 10-15mm) : moderately active, (zone of inhibition between 5–10 mm): slightly active, (-):no activity.

Statistical analysis

IBM SPSS Statistics 25 software and Two-factor without replication ANOVA TEST was used for statistical analysis of antibacterial activity "Table 4" , "Figure. 3" and one-way ANOVA TEST was used for antifungal activity "Table 5" , "Figure. 4" . The values are shown as the mean ± SEM of triple measurements of the zone of inhibition, with a significance level of 0.05.

Table 4. Antibacterial statistical analysis of final compounds(IVa-c, VIa,b)

Mean ± SEM				
Compounds	***S. aureus***	***S. pyogene***	***K. pneumonia***	***E. coli***
IVa	23.66±0.321*#	25.66±0.321*#	24.66±0.321#	24.66±0.321*#
IVb	21.66±0.321*#	24.66±0.321*#	24.66±0.321#	24.66±0.321*#
IVc	24.66±0.321*#	27.66±0.321*#	25.66±0.321*#	37.66±0.321*#
VIa	21.66±0.321*#	24.66±0.321*#	11.66±0.321*#	19.66±0.321*#
VIb	22.66±0.321*#	25.66±0.321*#	0.00±0.321*#	0.00±0.321*#

*Significant difference with respect to Amoxicillin P<0.05). # Significant difference with respect to Ciprofloxacin P<0.05).

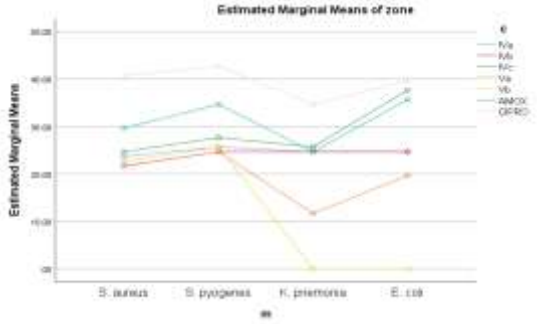


Figure 3. Histogram of the Antibacterial inhibition zone of final compounds (IVa-c, VIa,b)

Table 5. Antifungal statistical analysis of final compounds(IVa-c, VIa,b)

Mean ± SEM	
Compounds	***C. albicans***
IVa	24.66±0.333*
IVb	24.66±0.333*
IVc	28.66±0.333*
VIa	19.66±0.333
VIb	27.66±0.333*

*Significant difference with respect to Fluconazole P<0.05).

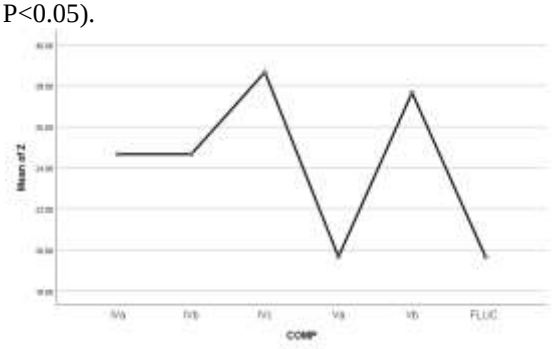


Figure 4. Histogram of the Antifungal inhibition zone of final compounds (IVa-c, VIa,b)

Minimum bactericidal concentration (MBC)

Compound (IVc) was more effective than amoxicillin and had the same MBC activity of ciprofloxacin against both G(+)ve and G(-)ve bacteria as well as fungi "Table 6".

Table 6. Minimum bactericidal concentration of final compounds (IVa-c, VIa,b)

Compound	**Minimum bactericidal concentration µg/ml**				
	S. aureus	***S. pyogenes***	***K. Pneumonia***	***E.Coli***	***C.albicans***
IVa	500	1000	1000	1000	500
IVb	500	1000	1000	1000	500
IVc	250	250	500	500	250
VIa	1000	1000	-	-	1000
VIb	1000	1000	-	-	1000
Amoxicillin	500	500	500	1000	-
Ciprofloxacin	250	250	250	500	-
Fluconazole					250

The minimum inhibitory concentration(MIC)

Antibiotic sensitivity test was used to assess MIC of final compounds. Compound IVc was more potent than amoxicillin and had the same potency of ciprofloxacin against gram +ve bacteria, somewhat less potent than ciprofloxacin and slightly more potent than amoxicillin against gram -ve bacteria and had the same antifungal potency as fluconazole "Table 7" , "Figure. 5".

Table 7. The minimum and sub-minimum inhibitory concentration of the final compounds(IVa-c, VIa,b)

Isolates	**IVa**		**IVb**		**IVc**		**VIa**		**VIb**		**AMOX.**		**CIP.**		**FLC.**	
	MIC	**SUB**	**MIC**	**SUB**	**MIC**	**SUB**	**MIC**	**SUB**	**MIC**	**SUB**	**MIC**	**SUB**	**MIC**	**SUB**	**MIC**	**SUB**
S. aureus	250	125	250	125	125	62.5	500	250	500	250	250	125	125	62.5	125	62.5
S. pyogen	500	250	500	250	125	62.5	500	250	500	250	250	125	125	62.5	125	62.5

K. Pneumonia	500 250	500 250	250 125	1000 500	1000 500	500 250	250 125	250 125
E. coli	500 250	500 250	250 125	1000 500	1000 500	250 125	125 62.5	125 62.5
C.albicans	250 125	250 125	125 62.5	500 250	500 250	250 125	125 62.5	125 62.5

AMOX: amoxicillin, CIP: ciprofloxacin, FLC: fluconazole

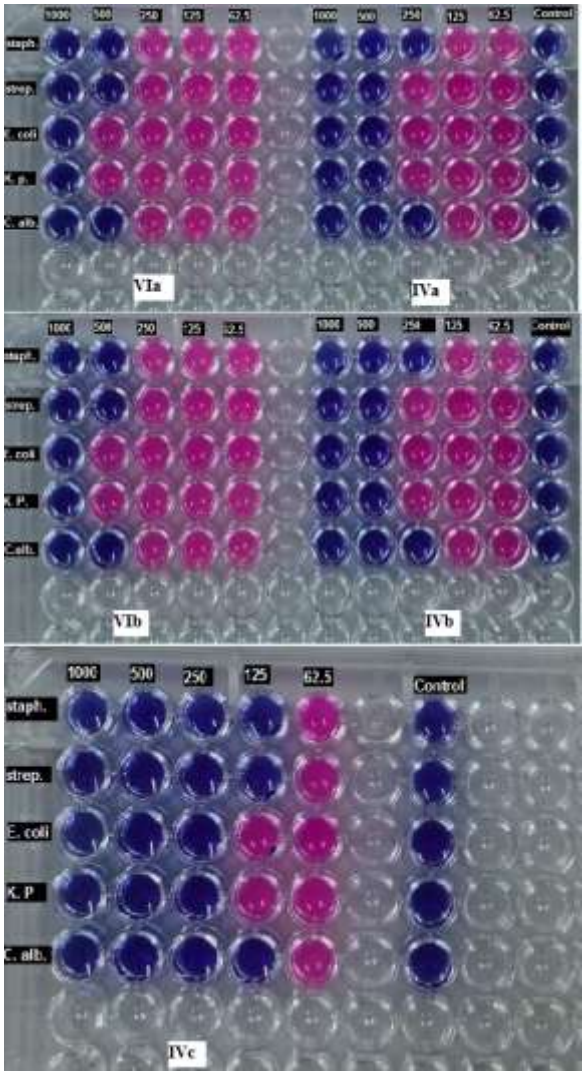


Figure 5. The minimum inhibitory concentration of final compounds

Conclusion

The disc diffusion method was used to assess antimicrobial efficacy of new derivatives of flurbiprofen.. The compound with the strongest action against G(-)ve and G(+)ve bacteria and fungus was IVc which was due to the presence of halogen chlorine. Compound IVc revealed highest MBC and MIC that may be due to the presence of halogen atom . All compounds showed good predicted pharmacokinetic properties and docking score .FT-IR and ^{1}HNMR were used to describe all of the final compounds.

Acknowledgement

The College of Pharmacy at the University of Baghdad especially department of pharmaceutical chemistry provided all the facilities needed for the research, for which the authors are grateful.

Conflicts of Interest

There is no conflict.

Funding

There was no dedicated funding for this research.

Ethics Statements

The authors emphasized that no ethics committee permission was required for the synthesis of the target compounds.

Author Contribution

Both authors contributed to design, preparation of target compounds and interpretation of FT-IR and ^{1}HNMR and antimicrobial activity of final compounds.

References

1. Hadi MK, Rahim NA, Sulaiman AT, Ali RM. Synthesis, characterization and preliminary antimicrobial evaluation of new schiff bases and aminothiadiazole derivatives of N-substituted phthalimide. Research Journal of Pharmacy and Technology. 2022;15(9):3861-5.
2. Singh, Bhoj R., et al. Comparative antimicrobial activity of aspirin, paracetamol, flunixin meglumine, tolfenamic acid, diclofenac sodium and pheniramine maleate. Acta Scientific Veterinary Sciences. 2021,3(9):30-42
3. Abdulhamza, Hayder M., and Muthanna S. Farhan. "Synthesis, Characterization and Preliminary Anti-inflammatory Evaluation of New Fenoprofen Hydrazone Derivatives." Iraqi Journal of Pharmaceutical Sciences. 2020, 29(2):239-244
4. Ahmed w. Naser, ahmed j. al-musawi, ammar a. fadhil and nada n. al-shawi , synthesis, characterization and anti-inflammatory evaluation of some new 2-(3-fluorobiphenyl-4-yl) propanoic acid derivatives, international journal of pharmaceutical sciences and research, 2017.8(4).1598-05
5. Aftab Alam , Mumtaz Ali, Najeeb Ur Rehman, Saeed Ullah, Sobia Ahsan Halim, Abdul Latif, Zainab, Ajmal Khan, Obaid Ullah , Shujaat Ahmad , Ahmed Al-Harrasi, and Manzoor Ahmad, Bio-Oriented Synthesis of Novel (S)-Flurbiprofen Clubbed Hydrazone Schiff's Bases for Diabetic Management: In Vitro and In Silico Studies, Pharmaceuticals 2022,15(6):672.
6. Khan M, Alam A, Khan KM, Salar U, Chigurupati S, Wadood A, Ali F, Mohammad JI, Riaz M, Perveen S. Flurbiprofen derivatives as novel α-amylase inhibitors: Biology-oriented drug synthesis (BIODS), in vitro, and in silico evaluation. Bioorganic chemistry. 2018, 1;81:157-67.
7. Aydın S, Kaushik-Basu N, Arora P, Basu A, Nichols D, Talele TT, Akkurt M, Çelik İ, Büyükgüngör O, Küçükgüzel ŞG. Microwave assisted synthesis of some novel Flurbiprofen hydrazidehydrazones as anti-HCV NS5B and anticancer agents. Marmara Pharmaceutical Journal. 2013 1;17(1):26-34.
8. Bondì ML, Craparo EF, Picone P, Giammona G, Di Gesù R, Di Carlo M. Lipid nanocarriers containing ester prodrugs of flurbiprofen preparation, physical-chemical characterization and biological studies. Journal of Biomedical Nanotechnology. 2013 1;9(2):238-46.
9. Hadi MK, Abdulkadir MQ, Abdul-Wahab AH. Synthesis and Antimicrobial Evaluation of Sulfonylhydrazide Derivatives of Etodolac. International Journal of Drug Delivery Technology. 2021;11(3):1000-1003.
10. Halah A. Sahib, Mohammed K. Hadi, Maadh Qusay Abdulkadir. Synthesis, and Antimicrobial Evaluation of New hydrazone Derivatives of (2,4-dinitrophenyl) hydrazine. Research Journal of Pharmacy and Technology. 2022; 15(4):1743-8
11. Pervaiz, Muhammad, et al. Synthesis and characterization of sulfonamide metal complexes as antimicrobial agents. Journal of Molecular Structure, 2019,1202(10):127284
12. Ovung, Aben; bhattacharyya, Jhimli. Sulfonamide drugs: Structure, antibacterial property, toxicity, and biophysical interactions. Biophysical reviews, 2021,13(2): 259-272.
13. AL-wahaibi, Lamya H., et al. Investigation of potential anti-malarial lead candidate 2-(4-fluorobenzylthio)-5-(5-bromothiophen-2-yl)-1, 3, 4-oxadiazole: Insights from crystal structure, DFT, QTAIM and hybrid QM/MM binding energy analysis. Journal of Molecular Structure, 2019, 1175: 230-240.
14. AL-omary, Fatmah AM, et al. Crystal structure of 2-[(4-bromobenzyl) thio]-5-(5-bromothiophen-2-yl)-1, 3, 4-oxadiazole, C13H8Br2N2OS2. Zeitschrift für Kristallographie-New Crystal Structures, 2023, 238(5): 911-913.
15. ALmalki, Abdulraheem SA, et al. Synthesis and biological evaluation of 1, 2, 3-triazole tethered thymol-1, 3, 4-oxadiazole derivatives as anticancer and antimicrobial agents. Pharmaceuticals, 2021, 14(9): 866.
16. Biernacki, Karol, et al. Novel 1, 2, 4-oxadiazole derivatives in drug discovery. Pharmaceuticals, 2020, 13(6): 111.
17. MA, Jinchao, et al. 1, 3, 4-oxadiazole bridges: A strategy to improve energetics at the molecular level. Angewandte Chemie, 2021, 133(10): 5557-5564.
18. Glomb, Teresa; ŚWIĄTEK, Piotr. Antimicrobial activity of 1, 3, 4-oxadiazole derivatives. International journal of molecular sciences, 2021, 22(13): 6979.
19. Othman, Adil A.; Kihel, Mebrouk; Amara, Sarah. 1, 3, 4-Oxadiazole, 1, 3, 4-thiadiazole and 1, 2, 4-triazole derivatives as potential antibacterial agents. Arabian Journal of Chemistry, 2019, 12(7): 1660-1675.
20. Chortani, Sarra, et al. Novel 1, 3, 4-oxadiazole linked benzopyrimidinones conjugates: Synthesis, DFT study and antimicrobial evaluation. Journal of Molecular Structure, 2020, 1217(4): 128357.
21. Hasan, Md Rakhibul, et al. In silico analysis of ciprofloxacin analogs as inhibitors of DNA gyrase of Staphylococcus aureus. Informatics in Medicine Unlocked, 2021, 26: 100748.
22. Jwaid, Mazen Mohammed, Karima Fadhil Ali, and Mayada Hadi Abd-Alwahab. "Design, synthesis, molecular docking and antibacterial evaluation of novel isoniazid derivatives bearing 1, 3, 4-oxadiazole and 1, 2, 3-triazol moieties

Jwaid." International Journal of Pharmaceutical Research 13(2) (2020): 2277-2286.

23. Saeed, Ali Mohammed, and Ayad Abed Ali Al-Hamashi. "Molecular Docking, ADMET Study, Synthesis, Characterization and Preliminary Antiproliferative Activity of Potential Histone Deacetylase Inhibitors with Isoxazole as New Zinc Binding Group." Iraqi Journal of Pharmaceutical Sciences. 2023,32(Suppl.): 188-203.

24. Mohammed, Hamada HH, et al. Synthesis and molecular docking of new N 4-piperazinyl ciprofloxacin hybrids as antimicrobial DNA gyrase inhibitors. Molecular Diversity, 2023, 27(4): 1751-1765.

25. Abbas S, Zaib S, Ali S, Iqbal J. 15-LOX Inhibitors: Biochemical Evaluation of Flurbiprofen and its Derivatives. Life and Science. 2020 ; 8;1(3):6-.

26. Kamms ZD, Hadi MK. Synthesis, Characterization and Preliminary Antimicrobial and Anti-inflammatory Evaluation of New Ibuprofen Hydrazide Derivatives. International Journal of Drug Delivery Technology. 2023;13(1):376-38.

27. Hmood KS, Kubba AA, Al-bayati RI, Saleh AM. Synthesis, and anti-tumor evaluation of some new Flurbiprofen derivatives against MCF-7 and WRL-68 cell lines. Indonesian Journal of Pharmacy. 2021;1;32(1):17-34.

28. Abdulraheem SS, Hadi MK. Synthesis and Characterization of New Coumarin Derivatives as Possible Antimicrobial Agents. International Journal of Drug Delivery Technology. 2021; 11(4):1484-1490.

29. Askar FW, Aldheif YA, Jinzeel NA, Nief OA. Synthesis and Biological Activity Evaluation of New Sulfonamid Derivatives. Iraqi Journal of Science. 58(.4B) : 2012-2021.

30. AL-wahaibi, Lamya H., et al. 1, 3, 4-Oxadiazole N-Mannich bases: synthesis, antimicrobial, and anti-proliferative activities. Molecules, 2021, 26(8): 2110.

31. Mamatha, S. V.; Belagali, S. L.; Bhat, Mahesh. Synthesis, characterisation and evaluation of oxadiazole as promising anticancer agent. SN Applied Sciences, 2020, 2(882): 1-12.

32. Noureldin NA, Richards J, Kothayer H, Baraka MM, Eladl SM, Wootton M, Simons C. Design, computational studies, synthesis and in vitro antimicrobial evaluation of benzimidazole based thio-oxadiazole and thio-thiadiazole analogues. BMC chemistry. 2021;15(1):1-9.

الرسو الجزيئي والتصنيع و التقييم ألاولي للمضادات الميكروبات لبعض مشتقات السلفونوهيدرازيد و ١و٣و٤-أوكساديازول الجديده لفلوربيبروفين

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الخلاصة

تم تحضير سلسلة جديدة من المركبات اشتقت من فلوربيبروفين عن طريق تفاعل إستر إيثيل فلوربيبروفين مع هيدرازين هيدرات لتحضير فلوربيبروفين هيدرازيد (III). بعد ذلك تفاعل مركب (٣) مع ثلاثة مركبات مختلفة من مركبات كلوريد السلفونيل (بنزين سولفونيل كلورايد، ٤-تولوين سولفونيل كلورايد،٤-كلوروسوفلونيل كلورايد). بالإضافة إلى ذلك تم تفاعل مركب (III) مع سلفيد الكاربون لإنتاج أكسادايزول ٢ – ثيون(V) الذي تم تفاعله مع ٤ - بروميد رابع كلوريد الفينيل و ٤ - بروميد رابع بروميد الفينيل لإنتاج المركبات النهائيه (VIa & VIb). تم استخدام الأشعة تحت الحمراء و طيف الرنين المغناطيسي النووي لتحديد هياكل المركبات النهائية. تم فحص كل مركب نهائي (IVa-c، VIa,b) من أجل نشاطه المضاد للميكروبات ضد البكتيريا الموجبة لصبغة الغرام (البكتيريا العقدية المقيحة، البكتيريا الكروية العنقودية)والبكتيريا السالبة لصبغة الغرام (الكلبسيلا الرئوي والإشريكية القولونية) وسلالة الفطريات (المبيضات البيضاء). ظهر المركب(IVc)أعلى نشاط مضاد للجراثيم ضد كلا السلالتين السالب والموجب لصبغة الغرام من البكتيريا والفطريات. الدراسة التنبؤيه للحركية الدوائية و السمية ظهرت خصائص تقديرية مقبولة شبيهة للأدوية وخصائص دوائية مرغوبة . ظهر برنامج التصميم الدوائي أن هناك نتائج جيدة مع البروتين المستهدف .

الكلمات المفتاحيه: الفعالية المضادة للميكروبات، فلوربيبروفين، مضادات الالتهاب غير الستيرويدية، أكسادايزول ،سولفونامايد ،سولفونيل كلوريد.