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Research Article

CHARACTERIZATION AND MOLECULAR DOCKING OF CINNAMIC ACID DERIVATIVES: POTENTIAL INHIBITORS OF CYCLOOXYGENASE ENZYMES

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ABSTRACT

Objective: The pathology of every disease passes through the inflammation stage; hence, the design and optimization of potential lead compounds as anti-inflammatory agents is still a significant part of medicinal chemistry globally.

Methods: In this study, we designed, synthesized, and characterized some cinnamic acid derivatives and performed molecular docking of the derivatives on the human cyclooxygenase-1 (COX-1) enzyme.

Results: The elemental analysis showed the presence of different functional groups. Molecular docking was performed on the active sites of COX-1 (PDB ID: 6Y3C). The derivatives as well as the standard compound, were observed to interact mainly with the arginine residue of the target protein. The dioxomethylene substituted derivative showed the highest binding affinity, compared with other derivatives, including the standard drug (−6.8 kcal/mol).

Conclusion: The binding affinity observed in the cinnamic derivatives, and biological activities correlations revealed that compounds with the dioxomethylene group would be good anti-inflammatory lead molecules, as they demonstrated high affinity to the target protein and biological activities. Thus, these compounds can serve as potential lead compounds for the design, and development of effective anti-inflammatory agents, targeted to inhibit the human COX-1 enzyme involved in biological inflammatory mechanisms.

Keywords: Anti-inflammatory, Inflammation, Cinnamic acid, Cyclooxygenase, Molecular docking.

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INTRODUCTION

Cinnamic acid (Fig. 1) is a natural aromatic carboxylic acid and a key chemical constituent found in *Cinnamomum cassia* (Chinese cinnamon) and *Panax ginseng*, as well as some fruits, vegetables, and honey [1]. Cinnamic acid derivatives possess antimicrobial, management of cancer, down-regulation of blood glucose levels in diabetes, and are useful in neurological disorders [2]. Some derivatives of cinnamic acid have been severally reported to be as effective as the conventional medications used in the management of different disease conditions *in vitro*, thereby making these compounds very significant potential therapeutic remedies [2]. The presence of an acrylic acid group substituted on the phenyl ring of cinnamic acid gives the compound either a *cis*- or *trans-configuration* [3,4]. Cinnamic acid derivatives activate the peroxisome proliferator-activated receptor-α, thereby stimulating lysosomal biogenesis and lowering amyloid protein aggregation (plaque) observed in Alzheimer's disease in mice [1]. Because of its natural origin, cinnamic acid and its derivatives are believed to have minimal side effects on biological tissues [3]; hence, the use of cinnamic acid derivatives' inflammatory properties was evaluated, to find alternative medicines that could be useful in the pharmacotherapy of inflammatory diseases [5].

The mechanism of action of most anti-inflammatory agents, especially, the non-steroidal anti-inflammatory drugs (NSAIDs) with analgesic and antipyretic activity, is the inhibition of the cyclooxygenase (COX-1 or COX-2) enzymes. These enzymes are responsible for the biosynthesis of prostaglandins and thromboxane [6]. Abnormalities in prostaglandin and thromboxane metabolism may contribute to the pathophysiology of several disease conditions including hypertension, pyrexia, pain regulation, and inflammation [7]. The inflammatory process is the initial phase of disease progression [8], making anti-inflammatory agents useful in almost every unusual clinical symptom. NSAIDs delay the progression of Alzheimer's disease, pointing to their potential in the inhibition of amyloid protein aggregation, which has also been shown by some cinnamic acid derivatives [1,9]. Some NSAIDs are known to cause severe neuroinflammation, gastritis, gastric ulcer, and even stomach cancer [10,11]. Structure-activity relationships (SAR) show the relationships between the physicochemical properties of chemical compounds and have been used severally in the design and development of medicinal agents and quantification of their biotransformation processes. A more elaborate quantitative-SAR (QSAR) helps to obtain a reliable statistical model for the prediction of the pharmacological activities of the samples under analysis [12], including hydrophobicity, hydrophilicity, and the nature of plasma binding of drug molecules [13,14]. Following these complications, the design and development of less toxic, effective, and stable analgesic and anti-inflammatory agents are imminent. This study utilized SAR and molecular docking of some synthesized cinnamic acid derivatives, previously evaluated pharmacologically and characterized using nuclear magnetic resonance (NMR) and Fourier transform infrared spectroscopy (FTIR), to quantify their COX-1 enzymes binding affinity and comparative anti-inflammatory activity with diclofenac (NSAIDs), as potential lead compounds for the design and discovery of therapeutically useful anti-inflammatory agents.

METHODS

Synthesis

Five derivatives of cinnamic acid were successfully synthesized. Malonic acid and derivatives of benzaldehydes were used in the

synthesis of cinnamic acid and derivatives following the Knoevenagel condensation reaction [15]. An equivalent weight of 13.6 g of *para*-methoxybenzaldehyde and 10.4 g of malonic acid was transferred into a 50 mL volumetric flask with the addition of 25 mL pyridine. The content was oiled under reflux for about 3 h, and then poured into a beaker with 40 mL of dilute HCl, acidified with 15 mL of concentrated 12M HCl, a precipitate was observed and allowed to settle then filtered. The filtered product was recrystallized from hot ethanol. The resultant crystals gave *para*-methoxycinnamic acid [(*E*)-3-(4-methoxyphenyl) acrylic acid] (A1). The procedure was repeated with 15.0 g of 3,4-dioxomethylenebenzaldehyde, resulting to 3,4-dioxomethylenecinnamic acid [(*E*)-3-(benzo[d][1,3]dioxol-5-yl) acrylic acid] (A2).

An equivalent weight of 1.70 g of *para*-methoxycinnamic powder was weighed and dissolved in 30 mL of methanol with gentle heating and constant stirring. An aliquot of 30 mL potassium hydroxide-KOH (10%) and 5 g Raney nickel were added and kept for 30 min with constant stirring. The clear solution was decanted, 10 mL of water was added to the residue and acidified with 20 mL of concentrated 12 M HCl, the precipitates observed were allowed to settle, filtered, and dried resulting in *para*-methoxydihydrocinnamic acid [3-(4-methoxyphenyl) propanoic acid] (A3). This was 4.5 g cinnamic acid repeated with acid resulting in dihydrocinnamic acid [3-phenylpropanoic acid] (A4).

Finally, cinnamic acid epoxide was formed. This was achieved by dissolving an equivalent weight of 1.48 g cinnamic acid powder in 30 mL of methanol, with gentle heat and constant stirring. An aliquot of 30 mL potassium hydroxide-KOH (10%) and 15 mL of hydrogen peroxide H_2O_2 was added and kept for about 30 min with constant stirring, and acidified with 40 mL of 10% acetic acid; a precipitate was formed, allowed to settle, filtered, and dried. This gave cinnamic acid epoxide [3-phenyloxirane-2-carboxylic acid] (A5).

Characterization and SAR

The synthesized molecules were subjected to various physicochemical analyses, including NMR (^{1}HNMR, ^{13}CNMR), infrared spectroscopy (FTIR), and QSAR. The compounds were characterized using ^{1}HNMR and ^{13}CNMR – NMR spectra were recorded on a Varian Gemini 200 (250 MHz), USA, the chemical shifts are reported in part per million (ppm) relative to tetramethylsilane. Melting points were determined with an electrothermal melting point apparatus, and infrared (IR) spectra were measured on a Buck scientific IR M500 instrument. Mass spectra were recorded on a Varian MAT 44S mass spectrometer operating at 70eV. The electron spray ionization mode of ionization was used for the mass spectrometry quantification.

The inhibitory concentration (IC50) of the derivatives was determined from the pharmacological profile (supporting file) with the following formula [16]:

$$Y=\text{Min}+\frac{\text{Max-Min}}{1+\left(\frac{X}{IC_{50}}\right)\text{HillCoefficient}}$$

Molecular docking

Molecular modeling and docking of the binding protein and synthesized ligands were done using the Maestro software of OPLS3, 2018 force field [17], and Pymol software [18]. The docking parameters and affinity were compared with the previously reported anti-inflammatory properties [5] of the synthesized compounds. The human COX-1 crystal structure protein (6Y3C) was obtained from the PDB website [19], and modeled with D3Pocket webserver [20,21], to obtain all possible binding pockets and apply the best fit.

RESULTS

The derivatives of cinnamic acid were successfully synthesized using the Knoevenagel condensation reaction pathway. All the chemical reagents and solvents used in the study were of analytical grades, and the synthetic reactions were monitored by thin layer chromatography, and spots were visualized with a UV lamp, and an iodine tank.

Binding affinity and SAR

The protein-ligand binding affinities from the *in-silico* molecular docking studies and relative structural features of all the cinnamic acid derivatives samples used in the study are shown in Table 4.

DISCUSSION

The elemental analysis of the synthesized cinnamic derivative using infrared, ^{1}HNMR, and ^{13}CNMR, and mass spectroscopy showed the presence of different functional groups and splitting patterns, confirming the expected skeleton of the various cinnamic derivatives. All the derivatives showed bands at around 2900–3500 and 1600–1700, in the infrared spectrum, depicting the presence of carbonyl (-C=O) and hydroxy (-OH) functional groups (Table 1). The mass spectrometry showed different splitting patterns of the cinnamic acid derivatives, validating the different natures of the molecules obtained (Table 1). Other functional groups such as C=C, C-O, and chemical environment of proton bonding and carbon bonds were also observed in the NMR (^{1}HNMR and ^{13}CNMR) analysis shown in Tables 2 and 3, respectively

Inflammation is one of the common events in the majority of disease conditions that can lead to the development of arthritis, diabetes, cancer, Alzheimer's disease, and atherosclerosis along with pulmonary, autoimmune, and cardiovascular diseases. Either reduces or promotes inflammatory processes in the biological system [22]. At present, inflammatory diseases are being managed with the steroidal and non-steroidal anti-inflammatory agents. Furthermore, the development of drugs from plants and animals has complex difficulties with a very small yield that is insufficient for subsequent lead development [23]. These compounds have been previously evaluated for some biological properties, including antimicrobial and anti-inflammatory evaluation with promising activities on mice models, compared to the standard diclofenac anti-inflammatory compound [5].

Understanding the cellular and protein binding ability of these molecules is essential for the design and development of alternative medications for the management of inflammatory conditions. Prostaglandins (PGs) are the mediators of inflammation in the biological system, which help to sustain the homeostatic functions and exert a variety of pathophysiological roles to regulate the development of various diseases, including inflammation [22,24]. The inhibition of these endogenous chemicals either reduces or promotes inflammatory processes in the biological system [22].

For a ligand or drug to considerably inhibit an enzyme (protein), it must have some degree of binding affinity to the receptor proteins. Hence, high affinity could result in increased inhibition of the ligand on the target protein. The cinnamic derivatives were observed to interact mainly with the arginine residue of the target protein as well as the standard diclofenac compound (Fig. 2). Several pockets were detected on the COX-1 enzyme protein, using the D3Pocket webserver [21]. Using the pocket with the highest molecular docking scores, different interactions were observed. Based on the level of affinity, A2 (−7.0 kcal/mol) had the highest affinity even more than the standard compound, followed by A1 (−6.3 kcal/mol), A3

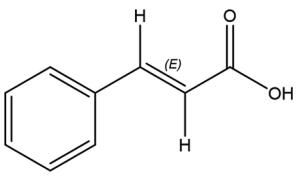


Fig. 1: Chemical structure of transcinnamic acid

Table 1: Infrared and mass spectrometry elemental characterization

Infrared spectroscopic analysis						
	Chemical component	**A1**	**A2**	**A3**	**A4**	**A5**
FTIR (KBr)	OH	2941.54	2939.61	3443.05	3437.26	2989.76
	C = O	1629.90	1653.05	1635.69	1653.05	1651.12
	C-O	1242.20	1249.91	1236.41	1255.70	1296.21
	C = C-H	979.87	1024.24	-	-	939.36
	C-C	-	1460.16	-	1454.38	-
Mass spectrometry						
Sample	**Molecular formula**	**Splitting pattern (m/z)**				
A1	$C_{10}H_{10}O_3$	m/z: 178.06 (100.0%), 179.07 (11.0%), 180.07 (1.2%)				
A2	$C_{10}H_8O_4$	m/z: 192.04 (100.0%), 193.05 (11.1%), 194.05 (1.4%)				
A3	$C_{10}H_{12}O_3$	m/z: 180.08 (100.0%), 181.08 (11.1%)				
A4	$C_9H_{10}O_2$	m/z: 150.07 (100.0%), 151.07 (9.9%)				
A5	$C_9H_8O_3$	m/z: 164.05 (100.0%), 165.05 (9.9%), 166.05 (1.0%)				

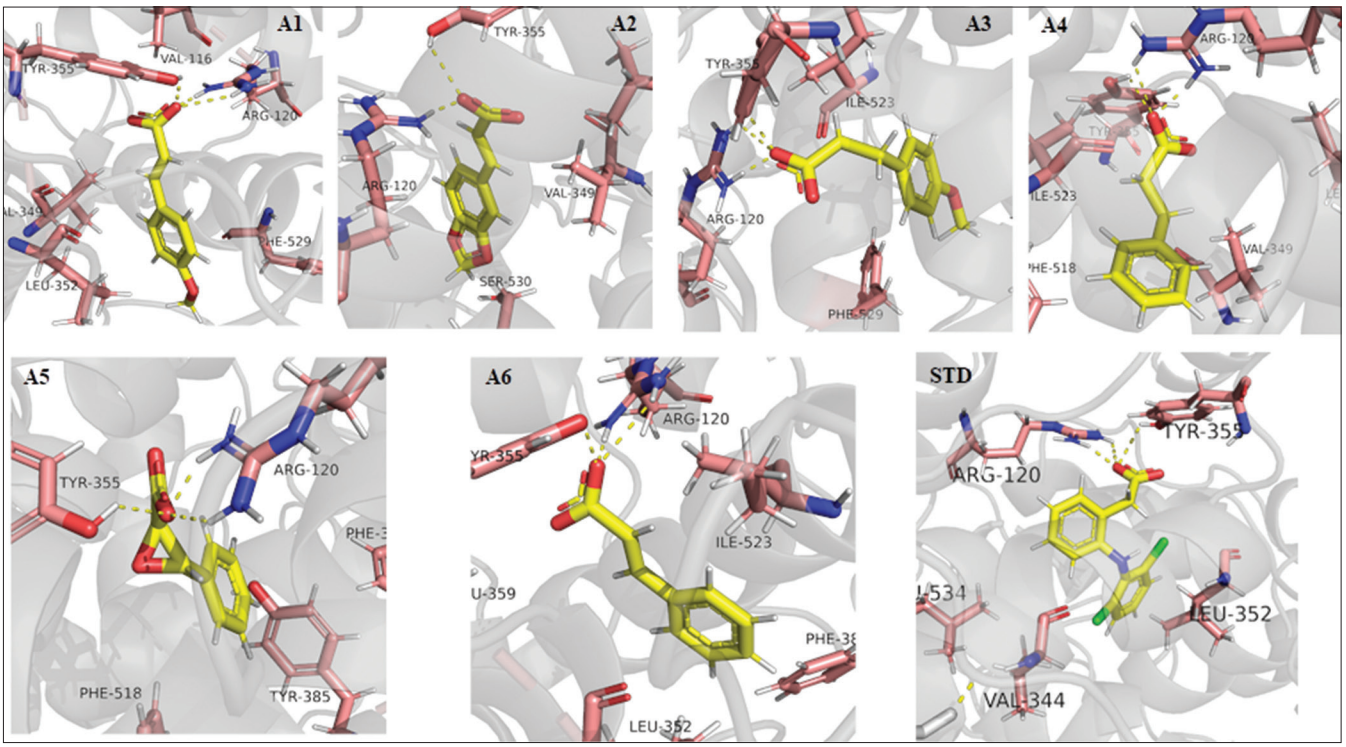


Fig. 2: COX Protein interactions with cinnamic acid derivatives: A1 – A6, STD – Standard – interaction at Arg-120, and Tyr-355 amino acid residues

(−6.3 kcal/mol), A6 (−6.4 kcal/mol), A4 (−6.3 kcal/mol), and the standard compound with −6.8 kcal/mol, respectively.

These results were in consonance with the biological activities of the molecules [5]. The standard compound had a $LogIC_{50}$ of 0.515 µg/L. Inhibitory concentration (IC_{50}) is a tool used to access the biological activities of therapeutic or potential agents with respect to their pharmacological actions [25]. The $logIC_{50}$ of the standard molecule was comparable with the synthesized compounds including A1 ($logIC_{50}$ of 0.521 µg/L), A5 ($logIC_{50}$ of 0.519 µg/L), A3 and A4 ($logIC_{50}$ of 0.522 µg/L). Higher $logIC_{50}$ was obtained in samples A2 and A6 (0.526 µg/L). The regression coefficient correlation of the synthesized cinnamic acid biological activities and molecular docking was 0.6023 (R_2), indicating a moderate correlation. This implies that the compounds inhibitory actions on the COX-1 enzyme may not be sufficient enough to alter its activities but can impair its functions.

Further structural modifications of the cinnamic acid derivatives may potentially lead to new anti-inflammatory agents. A1 and A2 have a methoxy and dioxomethylene side chains on the phenyl-base and showed good interaction with arginine and tyrosine bases of the protein residue with the highest binding affinities compared to other derivatives. Compared with the lead cinnamic acid compound, all the derivatives showed improved interactions with the protein molecules, starting with 2,3-dioxomethylene, methoxy (-OCH_3), oxirane (epoxide), and the dehydrated derivatives. This shows that with proper molecular optimization, these molecules would give many active chemical agents that can be employed as anti-inflammatory agents, especially the 2,3-dioxomethylene derivative. The previous anti-inflammatory also showed that the compound at about 2 h after administration showed more ant-inflammatory activities compared to the standard diclofenac molecule, while after 5 h, gave a comparable activity but was still less than the 2,3-dioxomethylene (A2), and methoxy (A1) derivatives. This is comparable to the report on the role of hydroxylated phenols' activity on inflammatory downregulation, in which the hydroxy group is a common moiety in all the derivatives, including the standard compound [26]. It is interesting to note that these

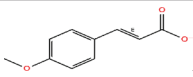
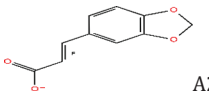
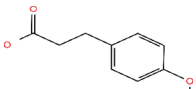
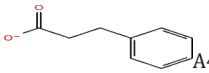
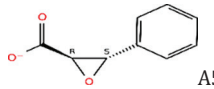
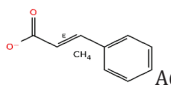
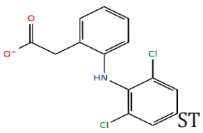
Table 2: ^{1}HNMR elemental analysis of cinnamic acid derivatives

A1 ^{1}HNMR analysis (DMSO) δ ppm					A2 ^{1}HNMR analysis (DMSO) δ ppm				
Node	**Coupling atoms**	**Chemical Shift**	**Atom index**	**Probable group (ring)**	**Node**	**Coupling atoms**	**Chemical Shift**	**Atom index**	**Probable group (ring)**
OH	H-C*C-H	12.05	14	COOH	OH	H-C*C*C-H	12.05	10	C = O
CH	H-C*C*C-H	7.06	1	Ar/-O-C/-C = C	CH_2	H-C*C-H	6.06	15	1,3-dioxole
CH	H-C*C-H	7.68	6	Ar/-O-C/-C = C	CH	H-C*C-H	7.12	6	Ar, -O-C, -C = C
CH	H-C*C*C-H	7.06	3	Ar/-O-C/-C = C	CH	H-C*C*C-H	6.94	3	Ar, -O-C, -C = C
CH	H-C*C-H	7.68	4	Ar/-O-C/-C = C	CH	H-C*C-H	7.06	4	Ar, -O-C, -C = C
CH_3	H-C*C*C-H	3.81	8	-CH_3/-O-/	H	H > C = C > H	7.45	11	Ethylene (-CH_2-CH_2-), C*C, -C(=O) O (*cis*)
H	H > C = C > C	7.45	12	-CH_2H_2/-C*C/-C(=O) O	H	H > C = C > H	6.27	12	-CH_2-CH_2-, C*C, -C(=O) O
H	H > C = C > H	6.27	13	-CH_2H_2/-C*C/-C(=O) O					
A3 ^{1}HNMR analysis (DMSO) δ ppm					**A4 ^{1}HNMR analysis (DMSO) δ ppm**				
OH	H-C*C-H	12.03	13	-COOH, -C-R	OH	H-C*C-H	12.03	11	-COOH, -C-C
CH	H-C*C*C-H	6.86	1	Ar-, -O-C, -C-C-	CH	H-C*C-H/H-C*CH*C-H	7.23	6	Ar-C, -C-C
CH	H-C*C-H/H-C*C*C-H	7.13	6	Ar-, -O-C, -C-C-	CH	H-C*C-H	7.23	4	Ar-C, -C-C
CH	H-C*C-H/H-C*C*C-H	6.86	3	Ar-, -O-C, -C-C-	CH	H-C*C-H/H-C*CH*C-H	7.28	3	Ar-C, -C-C
CH	H-C*C-H/H-C*C*C-H	7.13	4	Ar-, -O-C, -C-C-	CH	H-C*C-H	7.28	1	Ar-C, -C-C
CH_3	H-CH-CH-H	3.81	9	-CH_3, α-O-, -C*C-	CH	H-C*C-H/H-C*CH*C-H	7.19	2	Ar-C, -C-C
CH_2	H-CH-CH-H	2.73	7	-CH_2, α-C-, β-C(=O) O-	CH_2	H-C-CH-CH-H	2.73	7	-CH_2, α-C*C, β-C(=O) O
CH_2	H-CH-CH-H	2.51	11	-CH_2, α-C(=O) O, β-C*C-	CH_2	H-C-CH-CH-H	2.51	9	-CH_2, α-C(=O) O, β-C*C
H	H-CH-CH-H	2.73	8	CH_2, α-C-, β-C(=O) O-	H	H-C-CH-CH-H	2.73	8	-CH_2, α-C*C, β-C(=O) O
H	H-CH-CH-H	2.51	15	CH_2, α-C-, β-C(=O) O-	H	H-C-CH-CH-H	2.51	13	-CH_2, α-C(=O) O, β-C*C
A5 ^{1}HNMR Analysis (DMSO) δ ppm									
OH	H-C*C-H	13.36	11	-COOH, -C-C					
CH	H-C*CH*C-H	7.32	6	Ar-, -C-O					
CH	H-C*CH*C-H	7.32	4	Ar-, -C-O					
CH	H-C*C*C-H/H-C*C-H	7.25	3	Ar-, -C-O					
CH	H-C*C-H/H-C*CH*C-H	7.25	1	Ar-, -C-O					
CH	H-C*CH*C-H	7.25	2	Ar-, -C-O					
H	H-C-C-H	4.14	8	Oxiran-, α-C*C, β-C(=O) O					
H	H-C-C-H	3.79	13	Oxiran-, α-C(=O) O, β-C*C					

Table 3: ^{13}CNMR elemental analysis of cinnamic acid derivatives

A1 ^{13}CNMR analysis (DMSO) δ ppm			**A2 ^{13}CNMR analysis (DMSO) δ ppm**		
Node	**C-bonding shift**	**Overall Shift**	**Node**	**C-bonding shift**	**Overall Shift**
C	Ar-C: 128.5, -O-C: 33.5, -C = C: -0.8	159.8	CH_2	Aliphatic-C-C:-2.3, α-O: 98.0, β-C*C: 9.3	101.2
C	Ar-C: 128.5, -O-C: -7.7, -C = C: 6.4	127.5	C	Ar-C: 128.5, -O-C: 33.5,-14.4, -C = C:-0.1	148.7
CH	Ar-C: 128.5, -O-C: -14.4, -C = C: -0.1	114.2	C	Ar-C: 128.5, -O-C:-14.4, 33.5, -C = C:-0.8	148.0
CH	Ar-C: 128.5, -O-C: 1.0, -C = C: -2.3	130.2	CH	Ar-C: 128.5, -O-C:-14.4, 1.0, -C = C:-2.3	106.7
CH	Ar-C: 128.5, -O-C: -14.4, -C = C: -0.1	114.2	CH	Ar-C: 128.5-O-C: 1.0, -14.4, -C = C:-0.1	108.4
CH	Ar-C: 128.5, -O-C: 1.0, -C = C: -2.3	130.2	C	Ar-C: 128.5, -O-C: 1.0, -7.7, -C = C: 6.4	127.3
C	C = O: 166.0, -C = C: 4.0	171.5	CH	Ar-C: 128.5, -O-C:-7.7, 1.0, -C = C:-2.3	122.5
CH_3	C-C: -2.3, α-O: 49.0, β-O: 9.3	55.8	C	C = O: 166.0, -C = C: 4.0	171.5
CH	$-CH_2-CH_2$: 123.3, -C*C*C: 12.5, -C(=O)-O: 9.8	144.2	CH	$-CH_2-CH_2$:123.3, C*C*C: 12.5, -C(=O)-O: 9.8	144.9
CH	$-CH_2-CH_2$: 123.3, -C*C*C: -11.0, -C(=O)-O: 4.6	116.5	CH	$-CH_2-CH_2$:123.3, C*C*:12.5, -C(=O)-O: 4.6	116.5
A3 ^{13}CNMR analysis (DMSO) δ ppm			**A4 ^{13}CNMR analysis (DMSO) δ ppm**		
C	Ar-C: 128.5, -O-C: 33.5, -C-C:-2.8	157.8	C	Ar-C: 128.5, -C-C: 11.7	141.3
C	Ar-C: 128.5, -O-C:-7.7, -C-C: 11.7	133.6	CH	Ar-C: 128.5, -C-C:-0.6	127.7
CH	Ar-C: 128.5, -O-C:-14.4, -C-C:-0.1	114.2	CH	Ar-C: 128.5, -C-C:-0.6	127.7
CH	Ar-C: 128.5, -O-C: 1.0, -C-C:-0.6	129.8	CH	Ar-C: 128.5, -C-C:-0.1	128.6
CH	Ar-C: 128.5, -O-C:-14.4, -C-C:-0.1	114.2	CH	Ar-C: 128.5, -C-C:-0.1	128.6
CH	Ar-C: 128.5, -O-C: 1.0, -C-C:-0.6	129.8	CH	Ar-C: 128.5, -C-C:-2.8	125.9
C	-C = O: 166.0, -C-C: 11.0	174.4	C	-C = O: 166.0, -C-C: 11.0	174.4
CH_3	Aliphatic-C:-2.3, α-O-:49.0, β-C*C: 9.3	55.8	CH_2	Aliphatic-C:-2.3, α-C* C: 24.3, α-C: 9.1, β-C(=O)-O: 2.0	30.2
CH_2	Aliphatic-C:-2.3, α-C*C: 24.3, α-C: 9.1, β-C(=O)-O: 2.0	30.2	CH_2	Aliphatic-C:-2.3, α-C (=O)-O: 21.0, α-C: 9.1, β-C*C: 9.3	34.2
CH_2	Aliphatic-C:-2.3, α-C(=O)-O; 21.8, α-C: 9.1, β-C*C: 9.3	32.2			
A5 ^{13}CNMR analysis (DMSO) δ ppm					
CH	-Oxiran:-18.6, aliphatic -α-C*C: 24.3, α-C: 9.1, β-C(=O)-O: 2.0	60.1			
CH	-Oxiran:-18.6, aliphatic-α- C(=O)-O: 21.8, α-C: 9.1, α-O: 49.0, β-C*C: 9.3	60.5			
C	Ar-C: 128.5, -C-O-C: 9.2	135.5			
CH	Ar-C: 128.5, -C-O-C:-3.1	125.5			
CH	Ar-C: 128.5, -C-O-C:-3.1	125.2			
CH	Ar-C: 128.5, -C-O-C:-0.1	125.2			
CH	Ar-C: 128.5, -C-O-C:-0.1	128.6			
CH	Ar-C: 128.5, -C-O-C:-0.5	128.2			
C	-C = O: 166.0, -C-C: 11.0	173.2			

Table 4: The binding affinity of cinnamic acid derivatives on COX-1 enzyme protein

S/N	**Molecule**	**Binding affinity (Docking Score with 6Y3C: PDB)**	**Experiment Results (% inhibition) – IC_{50}**	**LogIC_{50}**
1.	 A1	−6.3	3.32	0.521
2.	 A2	−7.0	3.36	0.526
3.	 A3	−6.3	3.33	0.522
4.	 A4	−6.3	3.33	0.522
5.	 A5	−6.3	3.30	0.519
6.	 A6	−6.4	3.36	0.526
7.	 ST	−6.8	3.27	0.515

ST: Standard molecule

polyphenolic compounds, which are mostly from plant origin, have also been reported to have various pharmacological uses, such as anti-oxidant, and anticancer, among others [27]. With appropriate QSAR, molecular modifications, and clinical studies, more valuable molecules with diverse pharmacotherapeutic applications can be obtained from cinnamic acid derivatives.

CONCLUSION

The compounds showed more inhibitory affinity on the target protein compared to the standard drug sample used in the molecular docking and the previously reported clinical studies. The dioxomethylene (-O-CH2-O-) substituted derivative (A2) showed the highest binding affinity (−7.0 kcal/mol), compared with other derivatives, including the standard drug (−6.8 kcal/mol). The binding affinity observed on the derivatives with a specific docking score was −6.3 kcal/mol (A1, A3, A4, and A5), and −6.4 kcal/mol, for A6, respectively. The biological anti-inflammatory correlation studies revealed that compounds with the dioxomethylene group would be good anti-inflammatory lead molecules, as they exhibited good biological activity and high affinity to the target protein in the *in-silico* molecular docking analysis. This means, the higher the level of inhibition on the target protein receptor, the more the molecule would exert its potential pharmacological effects. All the cinnamic acid derivatives can be adequately optimized and further modified to reduce potential toxicities, and increase therapeutic efficacy.

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AUTHORS CONTRIBUTION

SJB –conceptualization, data curation; project administration; formal analysis design, experimental works, original draft; software; OM – writing – review and editing; DA – data curation, validation, manuscript review; HB – supervision, validation, manuscript review, and approval.

CONFLICTS OF INTERESTS

The author declares no conflicts of interest.

REFERENCES

1. Chandra S, Roy A, Jana M, Pahan K. Cinnamic acid activates PPARα to stimulate Lysosomal biogenesis and lower Amyloid plaque pathology in an Alzheimer's disease mouse model. Neurobiol Dis 2019;124:379-95.
2. Ruwizhi N, Aderibigbe BA. Cinnamic acid derivatives and their biological efficacy. Int J Mol Sci 2020;21:5712.
3. Yilmaz S, Sova M, Ergün S. Antimicrobial activity of trans-cinnamic acid and commonly used antibiotics against important fish pathogens and nonpathogenic isolates. J Appl Microbiol 2018;125:1714-27.
4. Rodrigues MP, Tomaz DC, de Souza LA, Onofre TS, de Menezes WA, Almeida-Silva J, *et al*. Synthesis of cinnamic acid derivatives and leishmanicidal activity against *Leishmania braziliensis*. Eur J Med Chem 2019;183:111688.
5. Jacob BB, Oluwadiya JO. Synthesis, Characterization and evaluation of Anti-inflammatory and antimicrobial properties of some cinnamic acid derivatives. Niger J Pharm Res 2020;16:1-8.
6. Bacchi S, Palumbo P, Sponta A, Coppolino MF. Clinical pharmacology of non-steroidal anti-inflammatory drugs: A review. Antiinflamm Antiallergy Agents Med Chem 2012;11:52-64.
7. Stoff JS. Prostaglandins and hypertension. Am J Med 1986;80:56-61.
8. Zappavigna S, Cossu AM, Grimaldi A, Bocchetti M, Ferraro GA, Nicoletti GF, *et al*. Anti-inflammatory drugs as anticancer agents. Int J Mol Sci 2020;21:2605.
9. Vane JR, Botting RM. Anti-inflammatory drugs and their mechanism of action. Inflamm Res 1998;47 Suppl 2:S78-87.
10. Bindu S, Mazumder S, Bandyopadhyay U. Non-steroidal anti-inflammatory drugs (NSAIDs) and organ damage: A current perspective. Biochem Pharmacol 2020;180:114147.
11. Hawkey CJ. Non-steroidal anti-inflammatory drug gastropathy: Causes and treatment. Scand J Gastroenterol Suppl 1996;220:124-7.
12. Umemori Y, Handa K, Sakamoto S, Kageyama M, Iijima T. QSAR model to predict $K_{p,uu,brain}$ with a small dataset, incorporating predicted values of related parameter. SAR QSAR Environ Res 2022;33:885-97.
13. Verma J, Khedkar VM, Coutinho EC. 3D-QSAR in drug design--a review. Curr Top Med Chem 2010;10:95-115.
14. Bunu JS, Ebeshi B, Ere D. Quantification of efavirenz plasma levels and associated adverse reactions using simple reversed-phase HPLC technique. Arab J Med Sci 2020;4:9-12.
15. Sawazaki T, Sohma Y, Kanai M. Knoevenagel condensation between 2-methyl-thiazolo[4,5-b]pyrazines and aldehydes. Chem Pharm Bull (Tokyo) 2022;70:82-4.
16. Quest Graph™ IC50 Calculator. AAT Bioquest, Inc. (2023). Retrieved from: https://www.aatbio.com/tools/ic50-calculator.
17. Salam NK, Adzhigirey M, Sherman W, Pearlman DA. Structure-based approach to the prediction of disulfide bonds in proteins. Protein Eng Des Sel 2014;27:365-74.
18. Seeliger D, de Groot BL. Ligand docking and binding site analysis with PyMOL and autodock/vina. J Comput Aided Mol Des 2010;24:417-22.
19. Miciaccia M, Belviso BD, Iaselli M, Cingolani G, Ferorelli S, Cappellari M, *et al.* Three-dimensional structure of human cyclooxygenase (hCOX)-1. Sci Rep 2021;11:4312.
20. Chen Z, Zhang X, Peng C, Wang J, Xu Z, Chen K, *et al*. D3Pockets: A method and web server for systematic analysis of protein pocket dynamics. J Chem Inf Model 2019;59:3353-8.
21. Shi Y, Zhang X, Mu K, Peng C, Zhu Z, Wang X, *et al.* D3Targets-2019-nCoV: A webserver for predicting drug targets and for multi-target and multi-site based virtual screening against COVID-19. Acta Pharm Sin B 2020;10:1239-48.
22. Ricciotti E, FitzGerald GA. Prostaglandins and inflammation. Arterioscler Thromb Vasc Biol 2011;31:986-1000.
23. Patil KR, Mahajan UB, Unger BS, Goyal SN, Belemkar S, Surana SJ, *et al*. Animal models of inflammation for screening of anti-inflammatory drugs: Implications for the discovery and development of phytopharmaceuticals. Int J Mol Sci 2019;20:4367.
24. Fujimori K. Prostaglandin D_2 and F_2^{α} as regulators of adipogenesis and obesity. Biol Pharm Bull 2022;45:985-91.
25. Aykul S, Martinez-Hackert E. Determination of half-maximal inhibitory concentration using biosensor-based protein interaction analysis. Anal Biochem 2016;508:97-103.
26. Al-Khayri JM, Sahana GR, Nagella P, Joseph BV, Alessa FM, Al-Mssallem MQ. Flavonoids as potential anti-inflammatory molecules: A review. Molecules 2022;27:2901.
27. Zhou Y, Zheng J, Li Y, Xu DP, Li S, Chen YM, *et al*. Natural polyphenols for prevention and treatment of cancer. Nutrients 2016;8:515.