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Original Research Article

In Vitro Anti-Inflammatory and Anti-Ulcerogenic Potential of Ethanol Extract of *Carica papaya* Leaves: Phytochemical and Molecular Docking Studies

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Pharmaceutical treatments for inflammation and ulcers often have side effects, prompting interest in plant-based alternatives. This study investigates the anti-inflammatory and anti-ulcerogenic potential of ethanol extract of *Carica papaya* leaves using *in vitro* assays and molecular docking. The leaves were subjected to ethanol extraction, after which the resulting extract underwent GC-MS analysis and *in vitro* assays. The phytochemicals identified from the GC-MS profile were subsequently analyzed through molecular docking studies for further evaluation. The extract demonstrated significant anti-inflammatory activity by inhibiting protein denaturation (IC_{50}: 180.70±2.25 µg/ml), membrane stabilization (IC_{50}: 192.60±2.29 µg/ml), and proteinase activity (IC_{50}: 1289.00±3.11 µg/ml). Its anti-ulcerogenic potential was evident in its inhibitory effects on H^+/K^+-ATPase (IC_{50}: 196.10±2.29 µg/ml) and urease (IC_{50}: 700.90±2.85 µg/ml), suggesting a role in gastric protection. Gas Chromatography-Mass Spectrometry (GC-MS) analysis identified biologically active compounds, such as phenolic acids and long-chain fatty acid esters, known for their anti-inflammatory and gastroprotective properties. Molecular docking analyses additionally confirmed the therapeutic potential of 2,3-Benzofurandione 2-monooxime, 3-(4-Nitrophenyl) propiolic acid, and Phthalic acid, di-(1-hexen-5-yl) by demonstrating strong interactions with cyclooxygenase-2 (COX-2) and H^+/K^+-ATPase enzymes, with binding affinities comparable to standard drugs like celecoxib and omeprazole respectively. These results offer scientific validation supporting the traditional use of *C. papaya* leaves in managing inflammatory and ulcerative conditions. Further in vivo studies and clinical trials are necessary to confirm their efficacy and safety for therapeutic applications.

Keywords: *Carica papaya*, Anti-inflammatory, Anti-ulcerogenic, Phytochemicals, Molecular docking, Cyclooxygenase-2 inhibition, Gastric protection

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Introduction

Inflammation and ulcerative disorders are significant health concerns globally, contributing to morbidity and reduced quality of life.[1,2] Chronic inflammation is implicated in the development of various diseases, such as cardiovascular diseases, diabetes, and gastrointestinal conditions such as peptic ulcers.[3,4] Conventional anti-inflammatory and anti-ulcer drugs, while effective are frequently linked to negative effects, prompting the search for safer, natural alternatives.[5,6] Medicinal plants have been traditionally employed for their healing properties, and recent scientific investigations have validated their potential as sources of bioactive compounds with anti-inflammatory and anti-ulcerogenic activities.[7-9] *Carica papaya* (papaya), a tropical fruit-bearing plant, is broadly acknowledged for its nutritional and medicinal value.[10,11] The leaves of *Carica papaya* has traditionally been employed to manage various ailments, such as inflammation, gastrointestinal disorders, and wound healing.[12-15] Phytochemical studies have identified a diverse array of bioactive compounds in papaya leaves, including flavonoids, alkaloids, tannins, and phenolic acids, which are recognized for their ability to exhibit antioxidant, anti-inflammatory, and gastroprotective properties.[16,17]

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These compounds are thought to exert their effects via mechanisms such as scavenging free radicals, inhibiting pro-inflammatory cytokines, and enhancing mucosal defense mechanisms.[18,19] Despite the widespread traditional use of *Carica papaya* leaves, there is a scarcity of scientific evidence regarding their anti-inflammatory and anti-ulcerogenic potential. Preliminary studies have demonstrated the antioxidant and anti-inflammatory properties of papaya leaf extracts in vitro, but further research is needed to elucidate their mechanisms of action and therapeutic efficacy.[14,20] Additionally, the anti-ulcerogenic potential of papaya leaf extracts remains underexplored, particularly about their ability to safeguard against gastric mucosal injury caused by factors such as stress, alcohol, or nonsteroidal anti-inflammatory drugs (NSAIDs). This research seeks to assess the *in vitro* anti-inflammatory and anti-ulcerogenic potential of the ethanol extract obtained from *Carica papaya* leaves. By investigating its effects on key inflammatory markers and their ability to mitigate ulcer formation, this research seeks to provide scientific validation for the ethnomedicinal applications of papaya leaves and support the advancement of natural therapeutic agents for inflammation and ulcer-related disorders. The outcomes of this research could carry substantial significance for the management of these conditions, offering a safer and more accessible alternative to conventional treatments.

Materials and Methods

Chemicals and Reagents

Carica papaya leaf extract, Ethanol, Methanol, butylated hydroxytoluene (BHT), ferrous sulfate, hydrogen peroxide, salicylic acid, distilled water, sodium nitroprusside, Griess reagent, glacial acetic acid, naphthyl ethylenediamine, dichloride, Hydrogen peroxide (H_2O_2), phosphate buffer, antioxidant ascorbic acid, phenazine methosulfate,

sodium acetate, glacial acetic acid, TPTZ (2, 4, 6-tripyridyl-s-triazine), HCl, $FeCl_3$, $FeSO_4$, ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid), potassium persulfate, potassium persulfate, diclofenac sodium, bovine serum albumin (BSA), hypo saline, sulfuric acid and sodium phosphate ammonium molybdate.

Carica papaya Extraction

The *Carica papaya* leaves were collected on the 10th of October 2024, identified, and authenticated by Mr. Bolu Ajayi, a taxonomist at the Department of Plant Biology, University of Ilorin, Kwara State, Nigeria, and a voucher number (UILH/001/945/2025) was obtained. Ilorin GPS coordinates are latitude: 8.49664° N and longitude: 4.54214° E. The leaves were washed to remove dirt and dried in the shade at low temperature to prevent degradation of bioactive compounds.

The dried leaves were subsequently pulverized in a blender to obtain powder before extraction. After grinding, the powder was macerated in ethanol at room temperature for 48-72 hours with constant shaking. It was then filtered, and the filtrate was concentrated in a water bath at 50 °C. The resultant extract was then obtained and kept in a refrigerator at 4 ℃ for further analysis.

GC-MS Analysis

Gas chromatography–mass spectrometry (GC-MS) analysis of the extract was conducted employing an Agilent 7890A Gas Chromatograph in conjunction with an Agilent 7000 Triple Quadrupole Mass Spectrometer (Agilent Technologies, USA). Separation of compounds was achieved on an HP-5MS capillary column (30 m × 0.25 mm i.d., 0.25 µm film thickness). Helium served as carrier gas at a constant flow rate of 1 mL/min. The oven temperature was programmed to increase from 100 to 260 °C at a rate of 4 °C/min. The injector temperature was maintained at 250 °C, while the detector was set at 230 °C. Identification of compounds was based on comparison of the obtained mass spectra with entries in the NIST 2017 mass spectral library.[21]

In vitro Anti-inflammatory Assay

Protein Denaturation Assay: The inhibition of protein denaturation can be assessed using a modified method.[22] This method involves preparing a reaction mixture containing 1% bovine serum albumin (BSA) in phosphate-buffered saline (PBS) at pH 6.4 and concentrations of 7.8125, 15.625, 31.25, 62.5, 125, 250, 500, and 1000 µg/ml of the extract. The mixture undergoes incubation at 37 °C for 20 minutes and is subsequently heated at 57 °C for 30 minutes. After cooling, the turbidity was determined spectrophotometrically at a wavelength of 660 nm using a Shimadzu UV-1800 Spectrophotometer (Shimadzu Corporation, Kyoto, Japan). The percentage inhibition of protein denaturation was computed using the following formula (equation 1):

$$\text{Percentage inhibition} = \frac{(\text{Absorbance of control} - \text{Absorbance of sample})}{\text{Absorbance of control}} \times 100 \ldots$$

Equation 1

Membrane Stabilization Assay: The membrane stabilization assay relies on the prevention of hypotonicity-induced hemolysis of red blood cells (RBCs) as previously described.[23] Blood was collected from a healthy volunteer and washed three times with isotonic PBS. The RBC suspension was subsequently combined with 7.8125, 15.625, 31.25, 62.5, 125, 250, 500, and 1000 µg/ml concentrations of the extract and hypotonic saline. The mixture was maintained at room temperature for 10 minutes, followed by centrifugation at 3000 rpm for 10 minutes. The absorbance of the supernatant was measured at 560 nm. The percentage inhibition of hemolysis was determined using the formula (equation 1).

Heat-Induced Hemolysis Assay: The heat-induced hemolysis assay involves incubating the RBC suspension with multiple concentrations (7.8125, 15.625, 31.25, 62.5, 125, 250, 500, and 1000 µg/ml) of the extract and PBS as previously described.[24] The mixtures were incubated at 54 °C for 30 minutes. After incubation, the samples were cooled under running tap water, centrifuged at 2500 rpm for 5 minutes, and the absorbance of the supernatant was measured at 560 nm. The inhibition percentage of hemolysis was calculated with the formula (equation 1).

Proteinase Inhibition Assay: The proteinase inhibition assay was conducted in accordance with the method previously described.[23] A reaction mixture containing 0.06 mg of trypsin, 20 mM Tris-HCl buffer (pH 7.4), and at 7.8125, 15.625, 31.25, 62.5, 125, 250, 500, and 1000 µg/ml concentrations of the extract was incubated for 5 minutes at 37 °C. After incubation, 0.8 % casein was added, then the resulting mixture was incubated for 20 minutes. The reaction was stopped by adding 70 % trichloroacetic acid, followed by centrifugation. The absorbance of the supernatant is measured at 210 nm. The percentage inhibition of proteinase activity was determined using the following formula (equation 1).

In vitro Anti-ulcerogenic Assay

H^+/K^+-ATPase Assay: The H^+/K^+-ATPase assay was conducted to assess the inhibitory potential of the extract on gastric proton pump activity, a key mechanism in gastric acid secretion. The assay was carried out in accordance with an earlier reported method, with slight modifications.[25] Briefly, gastric mucosal tissues were homogenized using ice-cold sucrose buffer (0.25 M sucrose, 0.02 M Tris-HCl, pH 7.4) and centrifuged at 10,000 × g for 15 minutes at 4 °C. The obtained supernatant was subsequently centrifuged at 100,000 × g for 60 minutes to isolate the microsomal fraction containing H^+/K^+-ATPase. The enzyme activity was determined spectrophotometrically by quantifying the release of inorganic phosphate (Pi) from ATP hydrolysis. The reaction mixture consisted of 50 µg of microsomal protein, 2 mM ATP, 10 mM $MgCl_2$, and 10 mM KCl in 50 mM Tris-HCl buffer (pH 7.4). The extract was added at varying concentrations (10–100 µg/mL), and the reaction was initiated by adding ATP. After incubation at 37℃ for 30 minutes, the reaction was terminated by adding 10% trichloroacetic acid (TCA). The released Pi was calculated using the method of Fiske and Subbarow,[26] with absorbance measured at 660 nm. Omeprazole (20 µM) served as a positive control. The percentage inhibition of H^+/K^+-ATPase activity was determined using the formula (equation 2):

$$\text{Percentage inhibition} = (1 - \frac{\text{Activity with extract}}{\text{Activity without extract}} \times 100) \ldots$$

Equation 2

Urease Assay: The urease inhibitory activity of the extract was assessed using a previously reported method with slight modification.[27] Urease, a significant enzyme in the pathogenesis of gastric ulcers, particularly those associated with *Helicobacter pylori* infection. The assay was conducted using jack bean urease (Sigma-Aldrich, USA) as the enzyme source. The reaction mixture consisted of 25 µL of urease solution (0.1 U/ml), 55 µL of phosphate buffer (100 mM, pH 6.8), and 5 µL of the extract at varying concentrations (10–100 µg/ml). The mixture was pre-incubated at 37 °C for 15 minutes, after which 65 µL of urea solution (100 mM) was included to initiate the reaction. The reaction proceeded for 30 minutes at 37 °C and was terminated by adding 45 µL of phenol-nitroprusside solution (1% phenol, 0.005 % sodium nitroprusside) followed by 70 µL of alkaline hypochlorite solution (0.5 % NaOH, 0.1 % sodium hypochlorite). The absorbance of the resulting indophenol complex was measured at 625 nm using a microplate reader. Thiourea (1 mM) served as a positive control. The percentage inhibition of urease activity was determined using the formula (equation 2).

Ligand Docking with Schrödinger Maestro 12.8

Top-binding phytochemicals from the extract were docked alongside the standard drugs pioglitazone and oxaprazin[17,28] using Schrödinger Maestro 12.8 (Schrödinger LLC, NY, USA) with HTVS, SP, and XP docking protocols. Ligands were prepared via the LigPrep module, using structures from PubChem,[29,30] with hydrogen addition, ionization at pH 7.0 ± 0.2, and geometry optimization under the OPLS4 force field. Final energy minimization ensured low-energy, stable conformers.

The 3D structures of human COX-2 and gastric H^+/K^+-ATPase (PDB IDs: 5F1A and 8D3U) were retrieved from the Protein Data Bank. Proteins were prepared in Maestro with the aid of the Protein Preparation Wizard, which introduced missing hydrogens, adjusted

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bond orders and charges, and removed water molecules beyond 5 Å from the binding site. Protonation states were set at pH 7.0 ± 0.2 using Epik, and energy minimization was carried out using the OPLS4 force field until the RMSD of heavy atoms reached 0.3 Å. Receptor grids were created surrounding the co-crystallized ligand sites using default settings for van der Waals scaling (1.0) and partial charge cutoff (0.25) to support accurate docking.[31]

Molecular docking was performed using the Glide Standard Precision (SP) protocol,[31,32] treating receptors as rigid and ligands as flexible. Top-ranked phytochemicals were further refined using Extra Precision (XP) docking for more accurate interaction assessment.[33,34] Binding free energies (ΔG_bind) were estimated using the MM-GBSA method in the Prime module, incorporating solvation, van der Waals forces, electrostatics, and hydrogen bonding.[35,36] The best candidates for COX-2 and H^+/K^+-ATPase were identified based on XP scores and MM-GBSA results. Ligand-receptor interactions were visualized in BIOVIA Discovery Studio Visualizer v21.1.0.20298 (2020), highlighting key binding features underlying their anti-inflammatory and anti-ulcerogenic potential.

Ethical Clearance

This research was conducted in line with the ethical principles stated in the Declaration of Helsinki. Ethical clearance for the study was secured from the Centre for Research and Development of Kwara State University, Malete, under approval number KWASU/CR&D/REA/2024/0093, dated 6th February 2025. No human or animal subjects were harmed during this research. All procedures involving plant materials were handled in compliance with institutional, national, and international guidelines.

Statistical Analysis

Statistical analyses were conducted using SPSS software (version 25.0), with all data expressed as mean ± standard deviation. One-way analysis of variance (ANOVA) was employed to evaluate differences between groups, followed by Duncan's post-hoc test for multiple comparisons. Statistical significance was defined at $p < 0.05$. The half-maximal inhibitory concentration (IC_{50}) values were calculated using GraphPad Prism 10 (version 10.0.2, GraphPad Software, Inc., La Jolla, CA, USA).

Results and Discussion

The *in vitro* anti-inflammatory and anti-ulcerogenic potentials of the extract demonstrated significant activity across multiple biochemical assays. The extract effectively inhibited protein denaturation (IC_{50}: 180.70±2.25 µg/ml), proteinase activity (IC_{50}: 1289.00±3.11 µg/ml), membrane stabilization (IC_{50}: 192.60±2.29 µg/ml), and heat-induced hemolysis (IC_{50}: 201.40±2.30 µg/ml), though its potency was lower than that of diclofenac, which served as the reference anti-inflammatory drug (Figure 1). The extract's ability to stabilize cell membranes and prevent protein denaturation suggests its potential to mitigate inflammatory damage by inhibiting key pathways associated with inflammation-related cellular injury.

The anti-ulcerogenic assessment revealed the extract's inhibitory effects on gastric proton pump activity and urease enzymes, with IC_{50} values of 196.10±2.29 and 700.90±2.85 µg/ml, respectively (Figure 2). While these values indicate moderate activity, they are significantly higher than those of omeprazole (standard drug) (IC_{50}: 14.43±1.16 µg/ml for H^+-K^+ ATPase and 32.26±1.51 µg/ml for urease). These findings suggest that the extract may possess gastroprotective effects through the modulation of gastric acid secretion and the inhibition of urease activity, which is often implicated in ulcer formation. The GC-MS investigation of *Carica papaya* leaf extract indicated the presence of a complex phytochemical profile, highlighting the presence of bioactive compounds with potential anti-inflammatory and anti-ulcerogenic properties. Notably, the chromatogram identified several key constituents, including benzoic acid methyl ester, 2,4-di-tert-butylphenol, and hexadecanoic acid derivatives, which have been reported for their antioxidant and anti-inflammatory activities previously (Figure 3, Table 1). The presence of dibutyl phthalate and 1,2-benzenedicarboxylic acid derivatives suggests additional pharmacological relevance, as these compounds exhibit cytoprotective and gastroprotective effects. Furthermore, long-chain fatty acid esters such as hexadecanoic acid ethyl ester and docosanoic acid ethyl ester may contribute to membrane stabilization and ulcer protection by modulating lipid peroxidation.

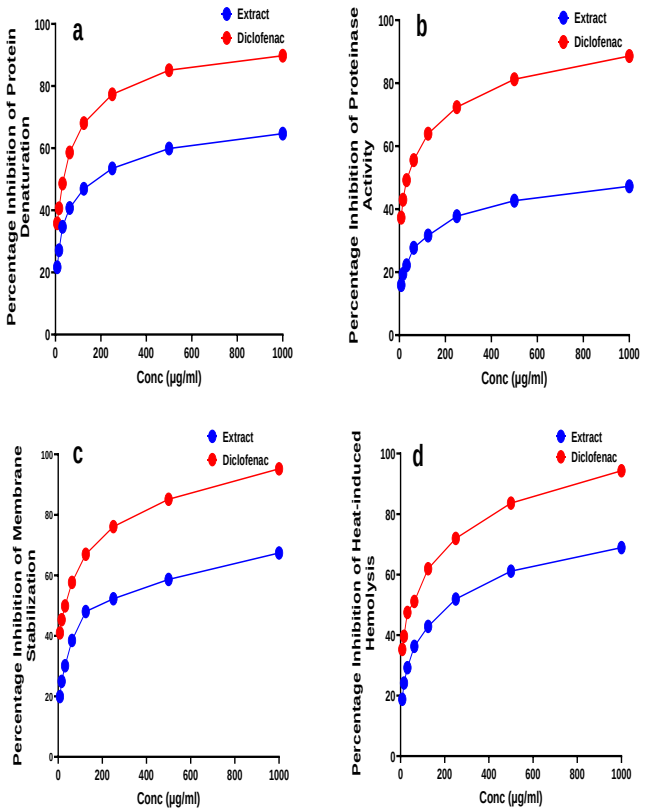

Figure 1: Percentage inhibition of (a) protein denaturation (b) proteinase (c) membrane stabilization and (d) heat-induced hemolysis by the extract. IC_{50} PD: Extract – 180.70±2.25 µg/ml, Diclofenac – 29.45±1.47 µg/ml, PI: Extract – 1289.00±3.11 µg/ml, Diclofenac – 30.33±1.48 µg/ml, MS: Extract – 192.60±2.29 µg/ml, Diclofenac – 24.37±1.39 µg/ml, HIH: Extract – 201.40±2.30 µg/ml, Diclofenac – 36.26±1.51 µg/ml

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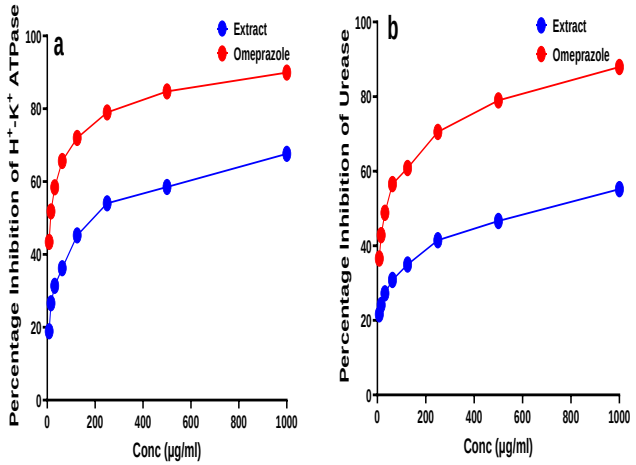

Figure 2: Percentage inhibition of (a) H+-K+ ATPase and (b) urease by the extract. IC_{50} H+-K+ ATPase: Extract –

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196.10±2.29 µg/ml, Omeprazole – 14.43±1.16 µg/ml, Urease: Extract – 700.90±2.85 µg/ml, Omeprazole – 32.26±1.51 µg/ml

The identification of phenolic and benzoic acid derivatives within the extract further supports its therapeutic potential in inflammatory and ulcerative conditions. Compounds such as 3-(4-nitrophenyl) propiolic acid and phenol, 2-(6,7-dimethylbenzo[e]dithiepan-2-yl) are known for their radical-scavenging properties, potentially capable of mitigating oxidative stress, a major contributor to inflammation and ulcer formation. The significant peak areas corresponding to these bioactive molecules indicate their dominant presence in the extract, suggesting a synergistic role in its pharmacological effects.The molecular docking study of COX-2 inhibitors using Glide molecular docking simulation yielded the identification of several lead compounds exhibiting potential anti-inflammatory activity. Only 36 compounds were dockable against COX-2 (Table 2). Celecoxib, a well-established COX-2 inhibitor, was used as the reference molecule, with a Glide GScore of -6.43 kcal/mol. Among the screened compounds, 2,3-Benzofurandione 2-monooxime demonstrated the best binding affinity with a Glide GScore of -6.23 kcal/mol, followed by 9-Azabicyclo[3.3.1]nona-2,6-diene-9-carboxaldehyde (-6.03 kcal/mol) and 1,2-Benzisoxazol-5-ol, 4,6-dimethyl- (-5.87 kcal/mol) (Table 2).

Further refinement through Standard Precision (SP) and Extra Precision (XP) docking was conducted to evaluate ligand binding energies more accurately. The Molecular Mechanics Generalized Born Surface Area (MMGBSA) computational analyses provided a more comprehensive understanding of the binding stability. Notably, Phthalic acid, di-(1-hexen-5-yl) ester displayed an XP GScore of -7.13 kcal/mol and MMGBSA of -51.06 kcal/mol, demonstrating strong interactions with the COX-2 active site (Table 3). Additionally, Docosanoic acid, ethyl ester and Z,Z-4,16-Octadecadien-1-ol acetate showed favorable docking scores (XP GScore: -6.54 and -5.94 kcal/mol, respectively) and significant binding free energy (MMGBSA: -55.72 and -56.05 kcal/mol, respectively). Figure 4 illustrates the binding pocket interactions between the selected lead compounds and COX-2, revealing hydrogen bonding, hydrophobic interactions, and π-π stacking with key active site residues. The interaction patterns suggest that these phytochemicals from Carica papaya exhibit competitive binding with Celecoxib, reinforcing their potential as COX-2 inhibitors.

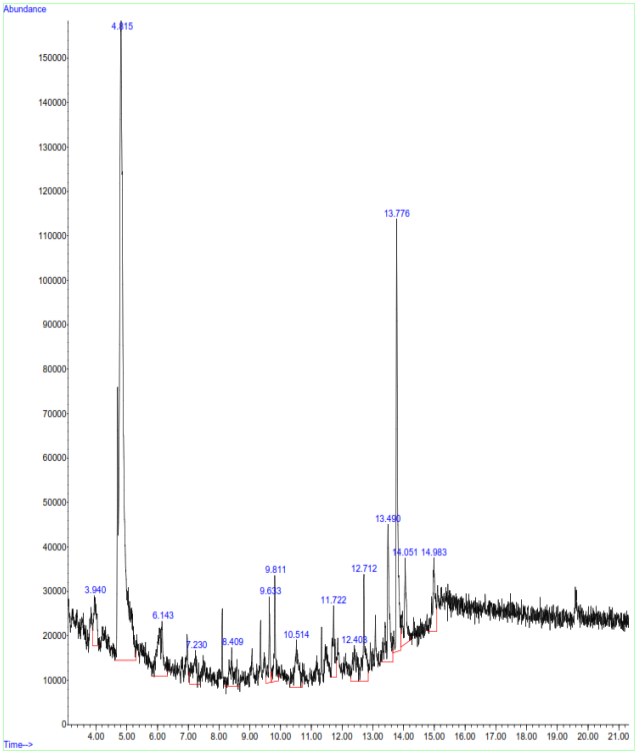

Figure 3: GC-MS Chromatogram of *Carica papaya* leaf

Table 1: GC-MS Analysis Results of *Carica papaya* leaf

Peak No	S No	Ligand	Retention time	Area	Quality	Mol. Formula	Mol. Weight (g/mol)	CID
1	1	1,1-Cyclopropanedicarbonitrile, 2, 3-dimethyl-, trans-	3.940	2.42	46	$C_{14}H_{22}O$	206	22212542
1	2	1,1-Cyclopropanedicarbonitrile, 2, 3-dimethyl-, cis-	3.940	2.42	41	$C_{15}H_{16}S_2O$	276	22212541
1	3	2-Amino-6-methoxy-4-(2H-1,2,3,4-tetrazol-5-yl) phenol	3.940	2.42	37	$C_{10}H_{12}O_3$	180	135629805
2	4	Benzoic acid, methyl ester	4.815	47.75	55	$C_9H_9NO_2$	163	7150
3	5	Silane, methylenebis [methyl-	6.143	5.24	43	$C_9H_{11}NO$	149	6329178
3	6	3-(4-Nitrophenyl) propiolic acid	6.143	5.24	43	$C_{20}H_{26}O_4$	330	247394
3	7	Dimethyl 3-hydroxypentanedioate, TMS derivative	6.143	5.24	43	$C_{17}H_{15}BrO_4$	363	232491
4	8	Cedran-diol, (8S,14)-	7.230	2.25	10	$C_9H_9NO_2$	163	51924376
4	9	Benzaldehyde, 2-nitro-, diaminomethylidenhydrazone	7.230	2.25	10	$C_8H_5NO_4$	179	9562955
4	10	2-Pyridinemethanamine, N-(diethylboryl)-	7.230	2.25	9	$C_{11}H_{12}O_6$	240	534975
5	11	1-Decanol, 2-methyl-	8.409	2.38	14	$C_9H_{16}O$	140	86776
5	12	7-Methyl-Z-tetradecen-1-ol acetate	8.409	2.38	14	$C_{20}H_{40}O$	296	5363222
5	13	Dodecanoic acid, 3-hydroxy-	8.409	2.38	14	$C_9H_{14}O_2$	154	94216
6	14	2,4-Di-tert-butylphenol	9.633	1.94	74	$C_9H_{12}O_2$	152	7311
6	15	Phenol, 2,5-bis(1,1-dimethylethyl)	9.633	1.94	64	$C_{10}H_{18}$	138	79983
7	16	Phenol, 2-(6,7-dimethylbenzo[e]dithiepan-2-yl)-	9.811	2.59	14	$C_{20}H_{36}O_2$	308	596134
7	17	Ethyl (2-hydroxyphenyl) acetate, S derivative	9.811	2.59	10	$C_{17}H_{34}O_2$	270	521075

7	18	1,2-benzisoxazol-5-ol, 4,6-dimethyl-	9.811	2.59	10	$C_{17}H_{34}O_2$	270	91723934
8	19	9-Azabicyclo [3.3.1] nona-2,6-diene-9-carboxaldehyde	10.514	3.15	38	$C_{16}H_{22}O_4$	278	590848
8	20	Phthalic acid, di-(1-hexen-5-yl) ester	10.514	3.15	38	$C_{16}H_{22}O_4$	278	590929
8	21	Phthalic acid, 3-bromobenzyl ethyl ester	10.514	3.15	27	$C_{20}H_{30}O_4$	334	91720576
9	22	benzoxazole, 7-methoxy-2-methyl-	11.722	2.54	18	$C_{11}H_{20}O_2$	184	12157234
9	23	2,3-Benzofurandione 2-monooxime	11.722	2.54	14	$C_{18}H_{36}O_2$	284	595970
9	24	1,4-benzenedicarboxylic acid, 2-hydroxyethyl methyl ester	11.722	2.54	14	$C_{24}H_{48}O_2$	368	77207
10	25	2-Nonyn-1-ol	12.403	2.17	16	$C_{16}H_{32}O$	240	80017
10	26	1-Hexadecen-3-ol, 3,5,11,15-tetramethyl	12.403	2.17	14	$C_{16}H_{30}O_2$	254	551282
10	27	3-Nonynoic acid	12.403	2.17	14	$C_{20}H_{40}O_2$	312	534236
11	28	1,2-Dimethyl-4-oxocyclohex-2-enecarboxaldehyde	12.712	3.88	30	$C_{14}H_{22}O$	206	565417
11	29	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl	12.712	3.88	27	$C_{15}H_{16}S_2O$	276	10129
11	30	Z, Z-4,16-Octadecadien-1-ol acetate	12.712	3.88	22	$C_{10}H_{12}O_3$	180	5364627
12	31	Hexadecanoic acid, methyl ester	13.490	5.75	93	$C_9H_9NO_2$	163	8181
12	32	Pentadecanoic acid, 14-methyl-, methyl ester	13.490	5.75	70	$C_9H_{11}NO$	149	21205
13	33	Dibutyl phthalate	13.776	11.11	90	$C_{20}H_{26}O_4$	330	3026
13	34	1,2-Benzenedicarboxylic acid, bis 2-methylpropyl) ester	13.776	11.11	78	$C_{17}H_{15}BrO_4$	363	6782
13	35	Phthalic acid, isobutyl octyl ester	13.776	11.11	78	$C_9H_9NO_2$	163	6423815
14	36	Ethyl cyclohexanepropionate	14.051	3.54	58	$C_8H_5NO_4$	179	61466
14	37	Hexadecanoic acid, ethyl ester	14.051	3.54	52	$C_{11}H_{12}O_6$	240	12366
14	38	Docosanoic acid, ethyl ester	14.051	3.54	52	$C_9H_{16}O$	140	22199
15	39	Oxirane, tetradecyl-	14.983	3.29	49	$C_{20}H_{40}O$	296	23741
15	40	1,2-15,16-Diepoxyhexadecane	14.983	3.29	43	$C_9H_{14}O_2$	154	543423
15	41	2-Octyldecyl acetate	14.983	3.29	25	$C_9H_{12}O_2$	152	91691480

Table 2: Summary of the hit molecules against COX-2 identified by High-Throughput Virtual Screening (HTVS) using the Glide molecular docking simulation protocol.

S. No	Ligand Name	Glide GScore (kcal/mol) COX-2
1	Celecoxib (**Reference**)	-6.43
2	2,3-Benzofurandione 2-monooxime	-6.23
3	9-Azabicyclo [3.3.1] nona-2,6-diene-9-carboxaldehyde	-6.03
4	1,2-Benzisoxazol-5-ol, 4,6-dimethyl-	-5.87
5	Phenol, 2-(6,7-dimethylbenzo[e]dithiepan-2-yl)-	-5.85
6	2-Pyridinemethanamine, N-(diethylboryl)-	-5.79
7	Phthalic acid, di-(1-hexen-5-yl) ester	-5.70
8	1,2-Dimethyl-4-oxocyclohex-2-enecarboxaldehyde	-5.68
9	2-Amino-6-methoxy-4-(2H-1,2,3,4-tetrazol-5-yl) phenol	-5.68
10	3-(4-Nitrophenyl) propiolic acid	-5.42
11	Phenol, 2,5-bis(1,1-dimethylethyl)	-5.29
12	1,1-Cyclopropanedicarbonitrile, 2,3-dimethyl-, trans-	-5.22
13	Benzoic acid, methyl ester	-5.14
14	Ethyl cyclohexanepropionate	-4.89
15	Ethyl (2-hydroxyphenyl) acetate, S derivative	-4.87
16	Benzoxazole, 7-methoxy-2-methyl-	-4.79
17	Benzaldehyde, 2-nitro-, diaminomethylidenhydrazone	-4.56
18	Silane, methylenebis [methyl-	-4.51
19	1,4-Benzenedicarboxylic acid, 2-hydroxyethyl methyl ester	-4.51
20	3-Nonynoic acid	-4.41
21	1,1-Cyclopropanedicarbonitrile, 2,3-dimethyl-, cis-	-4.13
22	2-Pyridinemethanamine, N-(diethylboryl)-	-5.48
23	2,3-Benzofurandione 2-monooxime	-5.87
24	1,2-Benzenedicarboxylic acid, bis 2-methylpropyl) ester	-3.45
25	Dodecanoic acid, 3-hydroxy-	-3.14
26	2-Nonyn-1-ol	-2.73

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27	Docosanoic acid, ethyl ester	-2.66
28	2-Pyridinemethanamine, N-(diethylboryl)-	-4.99
29	Dimethyl 3-hydroxypentanedioate, T MS derivative	-2.59
30	7-Methyl-Z-tetradecen-1-ol acetate	-2.51
31	1-Hexadecen-3-ol, 3,5,11,15-tetramethyl	-1.95
32	Hexadecanoic acid, ethyl ester	-1.25
33	1,2-15,16-Diepoxyhexadecane	-1.02
34	Z,Z-4,16-Octadecadien-1-ol acetate	-0.89
35	2-Octyldecyl acetate	0.04
36	1-Decanol, 2-methyl-	1.02
37	Pentadecanoic acid, 14-methyl-, methyl ester	1.69

Table 3: Standard Precision and Quantum Polarized Ligand Molecular Docking of Hit Compounds against the COX-2 Protein Receptor

S. No	Ligand Name	Glide GScore (kcal/mol) COX-2	MMGBSA (kcal/mol) COX-2	XP GScore (kcal/mol) COX-2	MMGBSA (kcal/mol) COX-2
1	Celecoxib (**Reference**)	-6.43	-54.61	-7.29	-48.33
2	Phenol, 2,5-bis(1,1-dimethylethyl)	-6.23	-27.17	-5.77	-27.15
3	Phenol, 2-(6,7-dimethylbenzo[e]dithiepan-2-yl)-	-6.03	-53.79	-6.02	-48.59
4	3-(4-Nitrophenyl) propiolic acid	-5.87	-22.2	-4.01	-28.82
5	2,4-Di-tert-butylphenol	-5.85	-35.36	-5.09	-34.95
6	Phthalic acid, di-(1-hexen-5-yl) ester	-5.79	-53.83	-7.13	-51.06
7	2,3-Benzofurandione 2-monooxime	-5.70	-26.6	-4.69	-27.42
8	1,2-Dimethyl-4-oxocyclohex-2-enecarboxaldehyde	-5.68	-25.16	-3.86	-25.15
9	2-Amino-6-methoxy-4-(2H-1,2,3,4-tetrazol-5-yl) phenol	-5.68	-7.92	-4.91	-5.55
10	2-Octyldecyl acetate	-5.42	-43.57	-6.13	-46.54
11	Phthalic acid, isobutyl octyl ester	-5.29	-51.78	-5.39	-55.61
12	1,1-Cyclopropanedicarbonitrile, 2,3-dimethyl-, trans-	-5.22	-21.25	-4.07	-21.27
13	Benzoxazole, 7-methoxy-2-methyl-	-5.14	-29.89	-4.18	-29.79
14	1,1-Cyclopropanedicarbonitrile, 2,3-dimethyl-, cis-	-4.89	-22.05	-3.95	-15.25
15	2-Pyridinemethanamine, N-(diethylboryl)-	-4.87	-36.08	-4.36	-41.62
16	9-Azabicyclo [3.3.1] nona-2,6-diene-9-carboxaldehyde	-4.79	-20.66	-3.40	-20.73
17	Benzoic acid, methyl ester	-4.56	-28.17	-3.78	-28.05
18	1,2-Benzisoxazol-5-ol, 4,6-dimethyl-	-4.51	-31.73	-4.31	-31.77
19	Docosanoic acid, ethyl ester	-4.51	-61.86	-6.54	-55.72
20	Z,Z-4,16-Octadecadien-1-ol acetate	-4.41	-59.82	-5.94	-56.05
21	Silane, methylenebis [methyl-	-4.13	-10.63	-2.84	-6.84
22	1,4-Benzenedicarboxylic acid, 2-hydroxyethyl methyl ester	-5.48	-44.38	-4.80	-39.93
23	Ethyl cyclohexanepropionate	-5.87	-37.09	-4.31	-38.97
24	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl	-3.45	-19.03	-3.56	-12.62
25	Ethyl (2-hydroxyphenyl) acetate, S derivative	-3.14	-39.96	-4.32	-36.32
26	Benzaldehyde, 2-nitro-, diaminomethylidenhydrazone	-2.73	-31.05	-3.49	-32.74
27	1,2-Benzenedicarboxylic acid, bis 2-methylpropyl) ester	-2.66	-43.58	-5.52	-56.23
28	3-Nonynoic acid	-4.99	-9.47	-4.58	-11.55
29	2-Nonyn-1-ol	-2.59	-32.92	-3.21	-32.28
30	2,3-Benzofurandione 2-monooxime	-2.51	-3.74	-4.69	-27.42
31	Dimethyl 3-hydroxypentanedioate, T MS derivative	-1.95	-30.94	-3.55	-29.16
32	Oxirane, tetradecyl-	-1.25	-52.84	-5.54	-31.58
33	2-Pyridinemethanamine, N-(diethylboryl)-	-1.02	-32.16	-2.59	-32.84
34	2-Pyridinemethanamine, N-(diethylboryl)-	-0.89	-36.04	-3.87	-36.66
35	7-Methyl-Z-tetradecen-1-ol acetate	0.04	-44.29	-4.81	-42.27
36	1-Hexadecen-3-ol, 3,5,11,15-tetramethyl	1.02	-47.17	-4.93	-24.09
37	Pentadecanoic acid, 14-methyl-, methyl ester	1.69	-39.49	-4.85	-49.93

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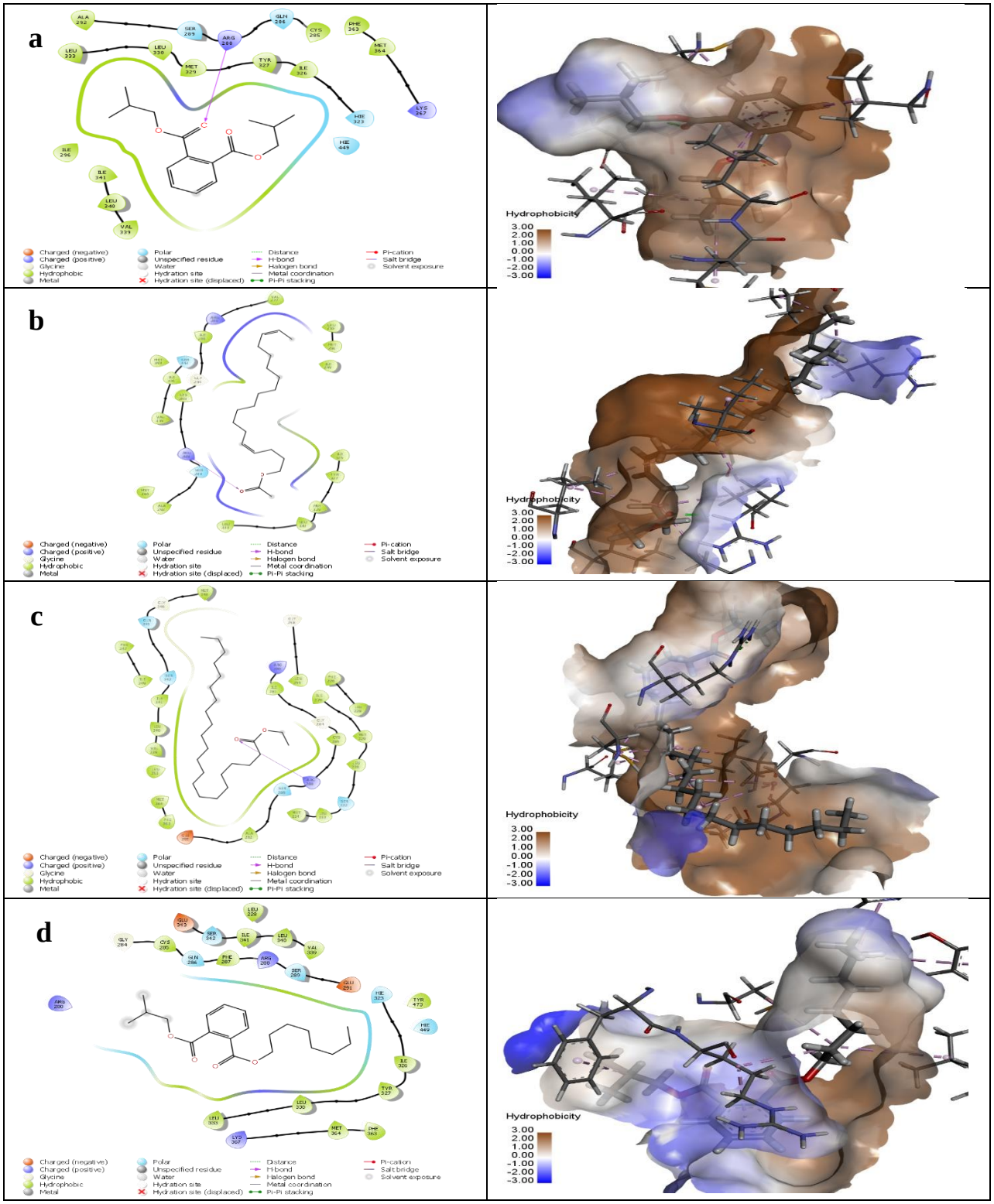

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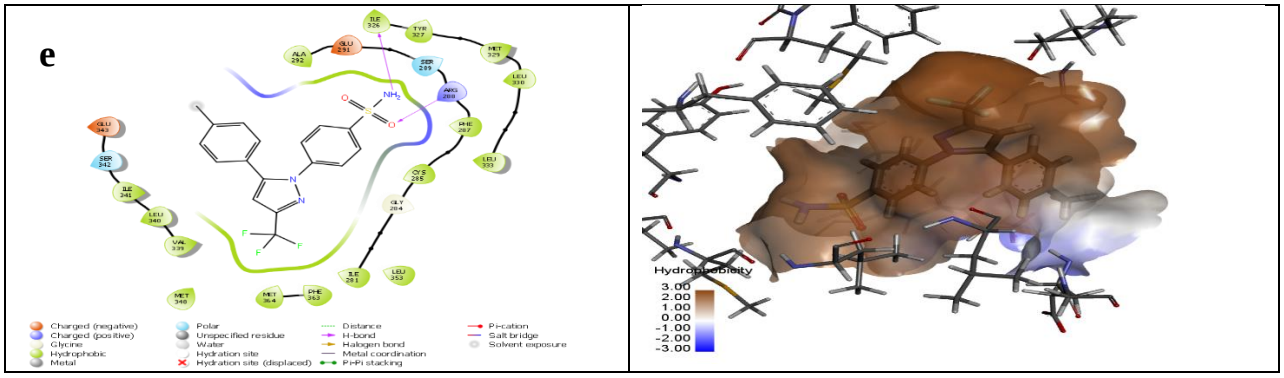

Figure 4: Binding pocket and two-dimensional interactions between COX-2 and *C. papaya* compounds. This figure presents a detailed view of the binding pocket and two-dimensional interactions between COX-2 and four *C. papaya* lead compounds: (a) 1,2-Benzenedicarboxylic acid, bis 2-methylpropyl) ester; (b) Z, Z-4,16-Octadecadien-1-ol acetate; (c) Docosanoic acid, ethyl ester; (d) Phthalic acid, isobutyl octyl ester; plus, reference drug: (d) Celecoxib. These interactions are elucidated following glide quantum polarized ligand docking, offering insights into the molecular dialogue between these phytochemicals and the active site amino acids of COX-2.

The molecular docking study aimed to identify potent inhibitors against H^+/K^+-ATPase through high-throughput virtual screening (HTVS) and quantum polarized ligand molecular docking simulations. The Glide docking scores (GScore) and Molecular Mechanics Generalized Born Surface Area (MMGBSA) binding energies were used as primary evaluation parameters for binding affinity.

In HTVS docking results (Table 4), the top-scoring ligand, Phenol, 2,5-bis(1,1-dimethylethyl), exhibited the highest binding affinity with a GScore of -6.66 kcal/mol. Other promising compounds included 2,4-Di-tert-butylphenol (-6.55 kcal/mol) and Phenol, 2-(6,7-dimethylbenzo[e]dithiepan-2-yl)- (-6.31 kcal/mol). The reference drug, omeprazole, yielded a significantly weaker GScore (-4.97 kcal/mol), suggesting that several test compounds may exhibit stronger inhibitory potential.

Table 4: Summary of the Hit Molecules Against H^+/K^+-ATPase Identified by High-Throughput Virtual Screening (HTVS) Using Glide Molecular Docking Simulation Protocol

S. No	Ligand Name	Glide GScore (kcal/mol) H^+/K^+-ATPase
1	Phenol, 2,5-bis(1,1-dimethylethyl)	-6.66
2	2,4-Di-tert-butylphenol	-6.55
3	Phenol, 2-(6,7-dimethylbenzo[e]dithiepan-2-yl)-	-6.31
4	Benzoic acid, methyl ester	-5.74
5	1,2-Dimethyl-4-oxocyclohex-2-enecarboxaldehyde	-5.71
6	1,2-Benzisoxazol-5-ol, 4,6-dimethyl-	-5.37
7	9-Azabicyclo [3.3.1] nona-2,6-diene-9-carboxaldehyde	-5.04
8	Omeprazole (**Reference**)	-4.97
9	3-(4-Nitrophenyl) propiolic acid	-4.88
10	Ethyl (2-hydroxyphenyl) acetate, S derivative	-4.78
11	1,1-Cyclopropanedicarbonitrile, 2,3-dimethyl-, cis-	-4.74
12	Silane, methylenebis [methyl-	-4.64
13	Benzoxazole, 7-methoxy-2-methyl-	-4.59
14	Docosanoic acid, ethyl ester	-4.41
15	2-Pyridinemethanamine, N-(diethylboryl)-	-4.41
16	2,3-Benzofurandione 2-monooxime	-4.36
17	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl	-4.33
18	2-Amino-6-methoxy-4-(2H-1,2,3,4-tetrazol-5-yl) phenol	-4.28
19	Z,Z-4,16-Octadecadien-1-ol acetate	-4.19
20	1,4-Benzenedicarboxylic acid, 2-hydroxyethyl methyl ester	-4.02
21	Ethyl cyclohexanepropionate	-3.96
22	1,1-Cyclopropanedicarbonitrile, 2,3-dimethyl-, trans-	-3.92
23	Benzaldehyde, 2-nitro-, diaminomethylidenhydrazone	-3.77
24	2-Pyridinemethanamine, N-(diethylboryl)-	-5.21
25	3-Nonynoic acid	-3.46
26	Dimethyl 3-hydroxypentanedioate, T MS derivative	-3.30
27	2-Pyridinemethanamine, N-(diethylboryl)-	-5.65
28	2,3-Benzofurandione 2-monooxime	-5.20
29	Phthalic acid, di-(1-hexen-5-yl) ester	-2.68
30	2-Octyldecyl acetate	-2.59
31	2-Nonyn-1-ol	-2.57
32	Pentadecanoic acid, 14-methyl-, methyl ester	-1.38
33	1,2-Benzenedicarboxylic acid, bis 2-methylpropyl) ester	-1.25
34	7-Methyl-Z-tetradecen-1-ol acetate	-0.77
35	Hexadecanoic acid, ethyl ester	-0.56
36	1-Hexadecen-3-ol, 3,5,11,15-tetramethyl	-0.46
37	Dibutyl phthalate	-0.38

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38	Dodecanoic acid, 3-hydroxy-	-0.34
39	1,2-15,16-Diepoxyhexadecane	-0.15
40	Oxirane, tetradecyl-	0.11
41	1-Decanol, 2-methyl-	0.81
42	Hexadecanoic acid, methyl ester	3.72

Subsequent Standard Precision (SP) and Quantum Polarized Ligand Docking (XP) analyses (Table 5) further refined the candidate selection. The highest XP GScore was observed in 1-Hexadecen-3-ol, 3,5,11,15-tetramethyl (-6.11 kcal/mol), followed by Phthalic acid, di-(1-hexen-5-yl) ester (-6.16 kcal/mol) and Z,Z-4,16-Octadecadien-1-ol acetate (-6.21 kcal/mol). MMGBSA binding energies also supported the strong binding interactions, with Z,Z-4,16-Octadecadien-1-ol acetate showing the most favorable energy (-63.03 kcal/mol), significantly outperforming omeprazole (-47.52 kcal/mol).

Table 5: Standard Precision and Quantum Polarized Ligand Molecular Docking of Hit Compounds Against the H^+/K^+-ATPase Protein Receptor

S. No.	Ligand Name	Glide GScore (kcal/mol) H^+/K^+-ATPase	MMGBSA (kcal/mol) H^+/K^+-ATPase	XP GScore (kcal/mol) H^+/K^+-ATPase	MMGBSA (kcal/mol) H^+/K^+-ATPase
1	Ethyl cyclohexanepropionate	-6.60	-37.58	-5.06	-29.24
2	2-Amino-6-methoxy-4-(2H-1,2,3,4-tetrazol-5-yl) phenol	-6.34	-48.73	-5.13	-20.88
3	Omeprazole (**Reference**)	-5.95	-32.06	-2.01	-47.52
4	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl	-5.74	-23.86	-4.31	-25.98
5	3-(4-Nitrophenyl) propiolic acid	-5.70	-26.30	-3.98	-23.06
6	2-Octyldecyl acetate	-5.69	-25.13	-5.30	-47.55
7	Phthalic acid, isobutyl octyl ester	-5.55	-37.53	-3.94	-47.21
8	Z,Z-4,16-Octadecadien-1-ol acetate	-5.43	-63.03	-6.21	-52.88
9	1,1-Cyclopropanedicarbonitrile, 2,3-dimethyl-, trans-	-5.36	-37.55	-3.15	-21.16
10	Benzoxazole, 7-methoxy-2-methyl-	-5.35	-18.98	-4.16	-26.10
11	1,1-Cyclopropanedicarbonitrile, 2,3-dimethyl-, cis-	-5.28	-36.50	-3.21	-17.61
12	Ethyl (2-hydroxyphenyl) acetate, S derivative	-5.15	-27.34	-5.07	-35.28
13	Silane, methylenebis [methyl-	-5.03	-23.20	-3.61	-12.10
14	9-Azabicyclo [3.3.1] nona-2,6-diene-9-carboxaldehyde	-4.97	-44.61	-3.35	-18.97
15	Benzaldehyde, 2-nitro-, diaminomethylidenhydrazone	-4.90	-48.07	-4.22	-30.43
16	2,3-Benzofurandione 2-monooxime	-4.89	-55.99	-0.87	-19.51
17	1,4-Benzenedicarboxylic acid, 2-hydroxyethyl methyl ester	-4.80	-21.22	-5.01	-30.17
18	Phthalic acid, di-(1-hexen-5-yl) ester	-4.79	-22.13	-6.16	-52.36
19	3-Nonynoic acid	-4.79	-17.23	-3.83	-26.42
20	Dimethyl 3-hydroxypentanedioate, T MS derivative	-4.69	-33.08	-3.45	-24.73
21	2-Nonyn-1-ol	-4.65	-13.39	-4.06	-33.84
22	1-Hexadecen-3-ol, 3,5,11,15-tetramethyl	-4.46	-14.83	-6.11	-54.16
23	1,2-Benzenedicarboxylic acid, bis 2-methylpropyl) ester	-4.36	-22.95	-4.75	-47.07
24	Dibutyl phthalate	-4.36	-19.54	-4.14	-45.65
25	7-Methyl-Z-tetradecen-1-ol acetate	-4.08	-28.64	-4.03	-42.21
26	Pentadecanoic acid, 14-methyl-, methyl ester	-3.98	-41.60	-3.37	-46.33
27	1,2-15,16-Diepoxyhexadecane	-3.56	-25.53	-3.58	-33.19
28	Hexadecanoic acid, ethyl ester	-3.52	-27.81	-3.43	-48.69
29	Dodecanoic acid, 3-hydroxy-	-3.17	-37.06	-3.64	-27.42
30	Hexadecanoic acid, methyl ester	-2.47	-55.56	-3.50	-47.76
31	Oxirane, tetradecyl-	-2.27	-35.95	-3.86	-41.86
32	1-Decanol, 2-methyl-	-2.26	-37.97	-3.34	-34.45

Figure 5 provides insight into the binding pocket and molecular interactions of selected lead compounds. The interactions highlight key hydrogen bonds and hydrophobic contacts stabilizing ligand binding at the active site of H^+/K^+-ATPase, suggesting these molecules' potential as competitive inhibitors.

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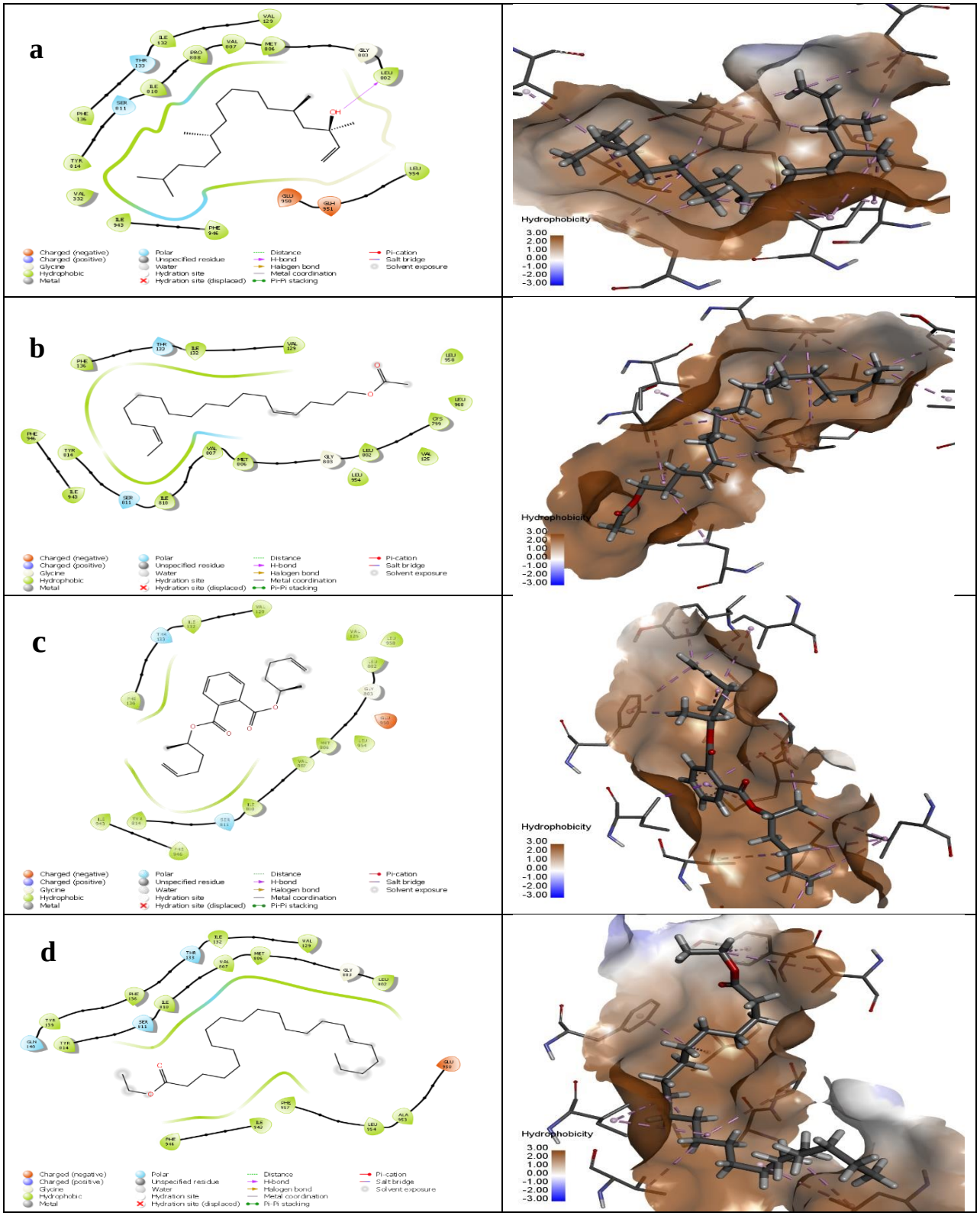

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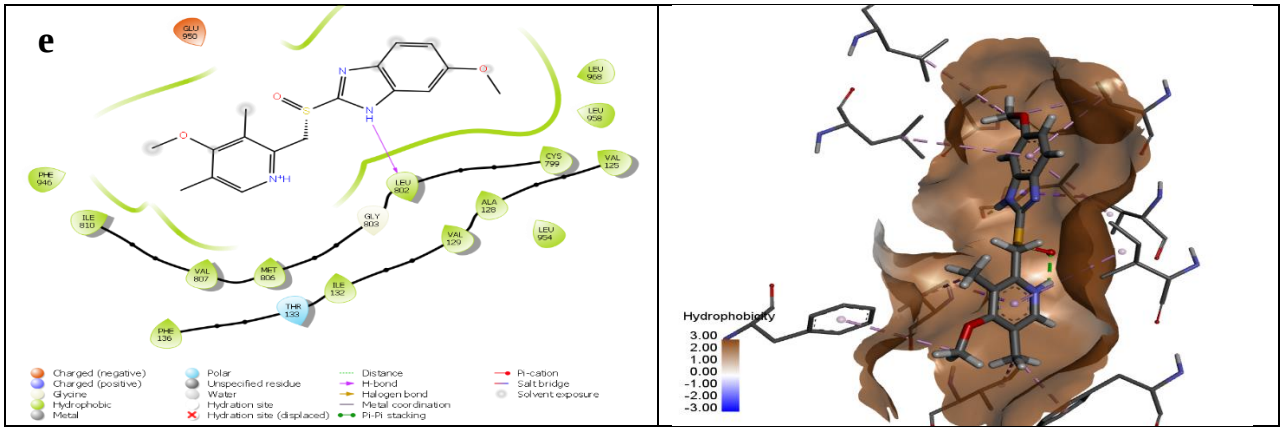

Figure 5. Binding pocket and two-dimensional interactions between H^+/K^+-ATPase and *C. papaya* compounds. This figure presents a detailed view of the binding pocket and two-dimensional interactions between H^+/K^+-ATPase and four *C. papaya* lead compounds: (a) 1-Hexadecen-3-ol, 3,5,11,15-tetramethyl; (b) Z, Z-4,16-Octadecadien-1-ol acetate; (c) Phthalic acid, di-(1-hexen-5-yl) ester; (d) Docosanoic acid, ethyl ester; plus, reference drug: (d) Omeprazole. These interactions are elucidated following glide quantum polarized ligand docking, offering insights into the molecular dialogue between these phytochemicals and the active site amino acids of H^+/K^+-ATPase.

The results highlighted the anti-inflammatory and anti-ulcerogenic potential of the ethanol extract of *Carica papaya* leaves, supporting their traditional use in treating inflammatory and gastrointestinal disorders. The study utilized various *in vitro* assays to evaluate the extract's efficacy, demonstrating significant inhibition of key inflammatory mediators and gastric ulcer-related enzymes.

Inflammation is a fundamental biological response to harmful stimuli, but chronic inflammation is implicated across multiple disease conditions, notably cardiovascular diseases, diabetes, and peptic ulcers.[4,37]. The extract exhibited substantial inhibition of protein denaturation, a well-established marker of anti-inflammatory activity. This result aligns with previous study suggesting that bioactive compounds in *Carica papaya*, such as flavonoids, tannins, and phenolic acids, contribute to anti-inflammatory effects by stabilizing proteins and preventing their denaturation.[38,39]

Additionally, the membrane stabilization assay demonstrated the ability of the extract to prevent hypotonicity-induced hemolysis of red blood cells (RBCs), indicating its potential to stabilize cell membranes and inhibit inflammation-related cell damage. This result is in agreement with previous report, which highlighted the significance of *Carica papaya* in membrane protection and oxidative stress reduction.[23,40]

The extract also inhibited proteinase activity, which plays a crucial role in inflammatory responses by degrading extracellular proteins and increasing tissue permeability. This result suggests that *Carica papaya* may modulate inflammation by targeting proteolytic enzymes involved in tissue damage.[20,41,42]

Gastric ulcers are commonly caused by NSAID use, stress, alcohol consumption, and *Helicobacter pylori* infection.[43] The study demonstrated that the extract significantly inhibited H^+/K^+-ATPase activity, plays a crucial role in gastric acid secretion. By targeting this enzyme, the extract may function similarly to proton pump inhibitors (PPIs), reducing acid secretion and minimizing mucosal damage.[44-46]

Furthermore, the extract exhibited urease inhibitory activity, suggesting a potential role in combating *H. pylori*-induced ulcers. Urease is critically involved in *H. pylori* survival by neutralizing gastric acid and contributing to ulcer formation.[45,46] The ability of *Carica papaya* extract to inhibit urease suggests a possible mechanism for reducing bacterial colonization and ulcer risk.

The molecular docking results of COX-2 are in agreement with prior molecular docking studies on natural anti-inflammatory compounds.[47-50] Several plant-derived polyphenols and fatty acid esters were reported to exhibit strong COX-2 inhibitory effects by binding within the enzyme's active site.[51,52] The observed docking scores suggest that *Carica papaya* phytochemicals could be identified as potential candidates for selective COX-2 inhibition, reducing inflammation demonstrating a lower incidence of gastrointestinal side effects relative to non-selective NSAIDs.

Earlier studies have provided evidence suggesting that selective COX-2 inhibitors, such as celecoxib, interact primarily with the hydrophobic pocket of COX-2, engaging residues such as Arg120, Tyr355, and Ser530.[53,54] The molecular interactions observed in this study suggest that the top-ranked compounds may similarly exploit these binding sites, potentially offering COX-2 selectivity with lower cardiovascular risks than synthetic coxibs.[55,56]

The molecular docking results for H^+/K^+-ATPase align with previous docking studies reporting the identification of natural polyphenols as potent inhibitors of gastric proton pumps.[57,58] Studies have demonstrated that polyphenolic compounds such as flavonoids and phenolic acids inhibit H^+/K^+-ATPase activity through hydrogen bonding and hydrophobic interactions.[39,59,60] The strong binding affinities observed in this study support the hypothesis that *Carica papaya* phytochemicals can serve as alternative ATPase inhibitors with improved pharmacokinetic properties.

Compared to synthetic inhibitors, natural compounds generally offer better biocompatibility and lower toxicity.[61,62] The presence of hydrogen bond donors and acceptors in the lead molecules suggests potential molecular interactions with essential amino acid residues, a characteristic observed in previous studies on ATPase inhibitors.[63,64]

An important finding emerging from this study is the dual inhibitory potential of certain phytochemicals. 2,3-Benzofurandione 2-monooxime, 3-(4-Nitrophenyl) propiolic acid, and Phthalic acid, di-(1-hexen-5-yl) ester showed promising binding affinities for both ATPase and COX-2. This combined activity provides evidence that these compounds may exert anti-inflammatory and gastroprotective effects simultaneously, making them ideal candidates for conditions such as NSAID-induced gastric ulcers.

Earlier investigations have demonstrated that dual-target inhibitors can offer superior therapeutic outcomes by addressing both inflammation and gastric mucosal damage.[65,66] For example, polyphenolic derivatives have been explored as both COX-2 inhibitors and proton pump inhibitors, providing an integrated approach to inflammatory disorders.[53,67,68] The findings from this investigation provide support for this paradigm, indicating that *Carica papaya* extracts may offer a natural, multifunctional therapeutic alternative.

Conclusion

This study validates the traditional use of *Carica papaya* leaves as a potential natural remedy for inflammatory and ulcerative conditions. The ethanol extract exhibited significant anti-inflammatory effects by inhibiting protein denaturation, membrane destabilization, and proteinase activity. Its anti-ulcerogenic properties were demonstrated through H^+/K^+-ATPase and urease inhibition, suggesting gastroprotective effects comparable to standard medications. GC-MS analysis revealed the presence of bioactive phytoconstituents, notably

phenolic acids and long-chain fatty acid esters, contributing to these pharmacological activities. Molecular docking further confirmed their strong interactions with COX-2 and H^+/K^+-ATPase, reinforcing their potential as alternative anti-inflammatory and anti-ulcer agents. These findings support the therapeutic application of *C. papaya* leaf extract in managing inflammation and gastric ulcers. However, further *in vivo* and clinical studies are essential to establish its efficacy, safety, and mechanism of action, thereby facilitating its potential integration into modern pharmaceutical formulations.

Conflict of interest
The authors declare no conflict of interest

Authors' Declaration
The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them

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