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*Original Article*

# Targeting Matrix Metalloproteinase-1 Inhibitory Potential of M*usa paradisiaca* Leaf Bioactives for Tissue Regenerative Applications

Thilaga Duraisamy[1], Ramya Ramadoss[1*], Nilavamuthan Chandrasekaran[2], Sandhya Sundar[1], Suganya Paneer Selvam[1], Nitya Krishnaswamy[1], K. Hema Shree[1], G. Radha[1]

[1]*Department of Oral Biology, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences, Saveetha University, Chennai 600077, India*
[2]*Department of Bioscience, Sri Krishna Arts and Science College, Coimbatore 641008, India*

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The bioactive substances found in Musa paradisiaca leaves have long been known to have potential medical uses, especially in tissue regeneration and extracellular matrix preservation. This work used both in vitro and in silico methods to measure the MMP-1 inhibitory potential of bioactive chemicals obtained from MP leaves. Using Electrospray Ionization Liquid Chromatography-Mass Spectrometry (ESI-LC-MS), phytochemical profiling was carried out to screen for and identify bioactive components in Musa paradisiaca leaf extracts. The MMP-1 protein target and the identified compounds' chemical structures were ready for computational analysis. To ascertain the pharmacokinetic viability of the chosen compounds, drug-likeness and ADMET (Absorption, Distribution, Metabolism, Excretion, and Toxicity) characteristics were assessed. The chemical interactions between the bioactive compounds and MMP-1 were examined using Chemical-Protein Network Analysis, which revealed Caffeic Acid Phenethyl Ester (CAPE) to be a promising drug with inhibitory potential. High-affinity binding interactions between CAPE and the catalytic region of MMP-1 were discovered by molecular docking experiments. Additionally, a molecular dynamics simulation was run for 50 ns. According to the results, CAPE has a potent MMP-1 inhibitory effect, indicating that it may be used as a natural medicinal agent for tissue regeneration and anti-collagenolytic purposes. Future in vivo and clinical studies to confirm the effectiveness of MP leaf-derived chemicals in modifying MMP-1 activity can be built upon this combined biochemical and computational approach. The findings open the door for the identification of new phytochemicals as potential treatments for tissue engineering and matrix metalloproteinase inhibition.



## Introduction

Matrix metalloproteinase-1 (MMP-1) breaks down collagen, with a high specificity for breaking down collagen types I and III, and promotes tissue regeneration, which is essential for extracellular matrix (ECM) remodelling [1]. It is crucial for tissue regeneration and wound healing because it encourages cell migration and the growth of new tissue [2]. However, increased MMP-1 activity can result in tissue destruction and poor repair in pathological circumstances such as cancer, fibrosis, and chronic inflammation [3]. Therefore, MMP-1 control is essential in preserving equilibrium in tissue regeneration, guaranteeing successful repair while avoiding the negative consequences linked to its overexpression.

Tissue inhibitors of metalloproteinases (TIMPs) are naturally occurring inhibitors that control the activity of MMPs [4]. An imbalance between MMP and TIMP may impact net MMP activity, ECM turnover, and tissue remodelling. It may also result in immunological and metabolic problems, cancer, and cardiovascular conditions such as hypertension, atherosclerosis, and aneurysms [5]. Matrix metalloproteinase-1 (MMP-1) inhibitors are currently undergoing exploration for their potential therapeutic applications.

It is known that several natural substances can inhibit MMP-1. Plant flavonoids, sometimes referred to as "the earth's delicate

* *Coresponding author*
E-mail address: ramyar.sdc@saveetha.com (*Dr. Ramya Ramadoss, Department Of Oral Biology, Saveetha Dental College and Hospital, Saveetha Institute of Medical and Technical Sciences, Saveetha University, Chennai 600 077, Tamilnadu, India)*

drugs," have anti-inflammatory qualities both in vivo and in vitro. [6]. While flavones like wogonin and apigenin only had a modest inhibitory effect, flavonols like quercetin and kaempferol showed significant inhibitory activity with $IC_{50}$ values of 39.6 and 43.7 mM, respectively [7]. Various plant phytochemicals, or plant compounds containing these flavonoids as main constituents, may help prevent ageing and treat inflammatory skin conditions [7]. It was also shown that many green tea polyphenols, such as proanthocyanidins, theaflavin, and epigallocatechin gallate, inhibit membrane-type 1 matrix metalloproteinase [8].

The investigation of biologically active natural products has been crucial in the discovery of new chemical entities (NCEs). For instance, nearly 28% of NCEs discovered originated from natural products or were derived from them [9]. *Musa paradisiaca* L. (Musaceae) predominantly thrives in tropical and subtropical regions and is recognised globally for its nutritional benefits [10]. Both the fruit and various other parts of the plant are utilised in traditional medicine to address a range of health issues in humans [11]. Though there were many studies on natural compounds acting as MMP1 inhibitors, little is known about the compounds derived from the widely available source of Musa paradisiaca leaves.

This study proposes to explore the leaves of *Musa paradisiaca* for the identification of novel flavonoids and phenolic compounds that exhibit potential inhibitory activity against matrix metalloproteinase-1 (MMP-1). Given previous research highlighting the tissue-regenerative properties of flavonoids and phenolic compounds, we hypothesise that extracts from *Musa paradisiaca* leaves may contain unique bioactive compounds that significantly inhibit MMP-1 activity. This inhibition may contribute to enhanced tissue regeneration and provide therapeutic benefits in managing inflammatory conditions. By investigating the phytochemical profile of banana leaves, we aim to uncover specific compounds that may serve as natural agents in regenerative applications, paralleling the effects of known flavonoids like quercetin and kaempferol. Our objective was to isolate, identify, and investigate the inhibitory mechanisms of the phytochemicals from the leaves of *Musa paradisiaca* on MMP-1 through molecular docking to better understand its role in enhancing tissue regeneration.

## Materials and Methods

### Extraction and Characterisation of Phytochemicals

Phytochemical isolation

Fresh leaves of *Musa paradisiaca* were collected from the local market. Aqueous extraction of the phytochemicals was done after washing thoroughly with distilled water; 50 g of leaves were cut into small pieces and added to a blender with ice-cold phosphate-buffered solution (PBS). 40 mL of the juice was filtered using a 40 ìm filter paper, collected in a beaker and transferred to a centrifuge tube. Cellular debris was eliminated by centrifuging the recovered juice for 20 minutes at 2000 g [10]. The supernatant was collected, filtered using a syringe filter of 0.22 micron, and stored at -80ºC until further analysis.

Screening of compounds derived from *Musa paradisiaca* leaves using ESI-LC-MS Q-Tof analysis

The study used the electrospray ionisation liquid chromatography-mass spectrometry (ESI-LC-MS) methodology for identifying small molecules in plant extracts using the Agilent Q-TOF for LC-MS analysis [12]. A water/acetonitrile gradient with 0.1% formic acid at 0.3 mL/min was used for the separation process on an Agilent C18 column. The Q-TOF mass spectrometer operates in positive and negative ESI mode, with capillary voltage (3500V/3000V), gas temperature (300°C), and a mass range of m/z 50–1000 Da. A 2µL test solution was injected to screen the chemicals from *Musa paradisiaca* leaves, and the chromatographs were tracked for 20 minutes. The study used 40 V and 2.0 kV cone and capillary voltages, 1000 L/h desolvation and 50 L/h cone flow rates, 6 eV and 15-40 eV collision energies, and a leucine-enkephalin solution for mass correction during acquisition. Data was analysed using Agilent Mass Hunter Quantitative Analysis software (RRID: SCR_015040), using accurate mass, m/z fragmentation, and spectral databases to identify metabolites with high confidence and precision [13].

### In silico analysis

Compound identification

The national centre for biotechnology information (NCBI), National Institutes of Health, hosts PubChem, which is a public repository for chemical structures and their bioactivities. The chemical structure of the identified compounds was downloaded in SMILES (Simplified Molecular Input Line Entry System) format along with the structural images [14].

Biological activity analysis

PASS (Prediction of Activity Spectra for Substances), an online software that predicts the outcome based on structure, was used to compare the functions of biologically active compounds isolated from Musa paradisiaca leaves [15]. It determines a substance's probability of belonging to both the active and inactive sub-sets of a medicine based on the Structure Activity Relationship Base (SAR Base) [16]. The input for the structure of the phytochemical was in the SMILES format. MMP inhibition and anti-inflammatory activity were measured for each ligand, yielding Pi (probable inactivity) and Pa (probable activity) values. For a given chemical, only activities with Pa > 0.5 and Pa > Pi were considered [17].

Drug-likeness and ADMET prediction

Oral and dermal pharmacokinetic profile features were described by Swiss ADME (http://www.swissadme.ch/) in this study to assess the ADMET (Absorption Distribution Metabolism Excretion Toxicity) properties and physiochemical parameters of drug-likeness, descriptors such as blood-brain barrier (BBB) permeability, gastrointestinal (GI) absorption through boiled egg graph, Log S data, Log $P_{o/w}$, Lipinski's rule of five, skin permeability (Log Kp), bioavailability, and synthetic accessibility of a selection of hit and reference compounds. The data was taken from the PubChem database and entered in SMILES format.

Network analysis of bioactive compounds and immune-related proteins

The relationship between bioactive compounds derived from *Musa paradisiaca* leaves and important immune-regulating proteins were discovered using the STITCH (Search Tool for Interactions of Chemicals) database (http://stitch.embl.de). To find similar chemicals recorded in the database, chemical structures were entered as SMILES strings. A network of linked proteins was displayed when the chemical was searched using STITCH as the entry point, placing it in a biological context. To highlight the compounds with comparable pharmacological activity or metabolic forms, the network was expanded to display related molecules [18].

### Molecular docking

Retrieval and preparation of protein structure for molecular docking

The protein data bank (http://www.rcsb.org) provided the 3D versions of MMP1 (PDB ID: 966C) [19]. The native ligand and water molecules had to be eliminated to preserve the protein crystal

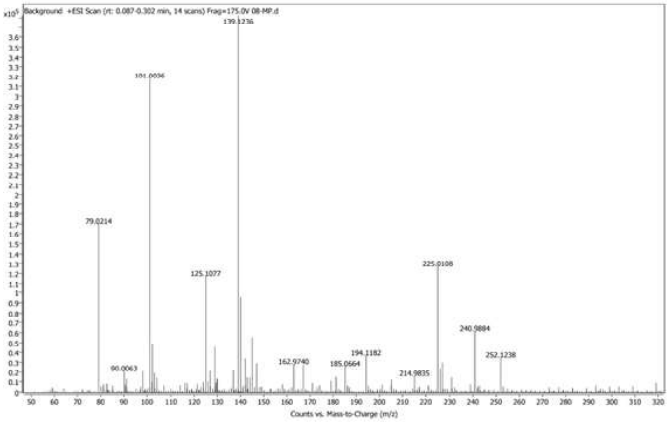



**Figure 1: LC-MS screening of compounds derived from *Musa paradisiaca***

structure. Polar hydrogens and gasteiger charge were added, and non-polar hydrogens were mixed using SwissDock [20,21].

Molecular docking and docking validation

Using Swiss AutoDock Vina (http://www.swissdock.ch/), the native ligand of the protein was positioned in the centre of the ligand with a size of 20 - 20 - 20 Å to establish the grid location for molecular docking. MMP1's grid box coordinates were Z = 45.454, Y = -6.260, and X = 3.832. AutoDock Vina on Swiss Dock was used for molecular docking studies [22]. The Lamarck Genetic Algorithm is used in molecular docking research (LGA). To determine the root mean square deviation (RMSD) using SwissDock in comparison to the initial native ligand confirmation to the receptor, the native ligand was re-docked with the protein in the docking validation technique [21]. CAPE was docked onto MMP1 using validation procedures that met the requirement of RMSD less than 2. For additional analysis, the cluster with the best binding energy (-ÄG, kcal/mol) was selected. We also looked at the interactions between van der Waals, charge, hydrophobic, and H-bond.

Molecular dynamics

To study the ligand-protein binding, molecular dynamics (MD) simulations were conducted using the AMBER99SB-ILDN force field. Protein topologies were constructed using PDB forms, while ligand topologies were constructed using SMILES formats [23]. The protein topology was created using AMBER ff14SB, whereas the ligand topologies were created using GAFF (General Amber Force Field) and ACPYPE (AnteChamber Python Parser interface). After placing the protein-ligand complex in a cubic box, sodium and chloride ions were added to neutralise the charge. The system was minimised using the steepest descent algorithm for 5000 steps. The solvent and ions were equilibrated in two restricted phases at 300 K and 1.0 bar. Unrestricted MD simulations were then conducted for A 50 ns production run with a 2 fs timestep was conducted under NPT circumstances. The LINCS approach was used to limit the covalent bonds, and the Particle Mesh Ewald (PME) method was used to calculate the electrostatic interactions with a threshold of 10 Å. The root-mean-square deviation (RMSD) and root-mean-square fluctuation (RMSF) were calculated to evaluate system stability. The radius of gyration (Rg) analysis was used to assess the compactness of proteins. The hydrogen bond interactions between the ligand and the protein were noted along the course. The Molecular Mechanics/Generalised Born Surface Area (MM/GBSA) and Molecular Mechanics/Poisson-Boltzmann Surface Area (MM/PBSA) methods were used to determine the binding free energy.

Data analysis

There were several steps involved in the data analysis process: (1) Using ESI-LC-MS Q-Tof analysis, phytochemicals extracted from *Musa paradisiaca* leaves were screened for bioactive compounds; (2) the structures of all the compounds and protein target, MMP-1, were prepared; (3) drug-likeness and ADMET analysis of specific compounds with inhibitory effects on MMP activity were observed; (4) STITCH analysis was performed to identify the interaction between the compound and protein for MMP inhibition. (5) In a 50 ns run, a molecular dynamic simulation of the best compounds with the best binding affinities, drug-likeness, and ADMET characteristics was performed; (6) Lastly, this approach attempts to find new compounds that target MMP1.

## Results and Discussion

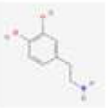
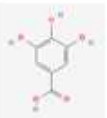
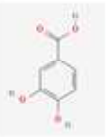
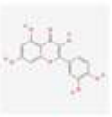
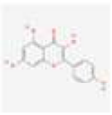
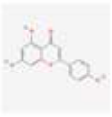
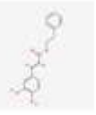
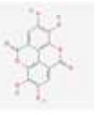
### Bioactive compounds derived from leaves of *Musa paradisiaca* identified by ESI-LC-MS Q-Tof analysis

A total of 11 compounds were successfully screened from the bioactive molecules derived from the leaves of *Musa paradisiaca* and identified based on the similarity percentage of their relative intensity and mass-to-charge ratio with the database from Agilent MassHunter Quantitative Analysis software (RRID: SCR_015040) [14,24] (figure 1). The isolated compounds were hydroquinone, p-benzoquinone, piperidine, dopamine, gallic acid, protocatechuic acid, quercetin, kaempferol, apigenin, caffeic acid phenethyl ester and ellagic acid. The structures of these compounds, along with their molecular weight, molecular ion (m/z), and the SMILES data, can be seen in table 1-2.

The compounds identified predominantly consisted of bioactive compounds like phenolic compounds, quinones, alkaloids and flavonoids. Our analysis revealed that the aqueous extract derived from the leaves of *Musa paradisiaca* contains a simple phenol, phenolic compound, phenolic acid, phenolic ester, and polyphenol. Major components like reducing sugar, carbohydrates, saponins, tannins, steroids, alkaloids, phenols, glycosides, phytosterols, terpenoids, and flavonoids are commonly found in *Musa paradisiaca* [25,26]. A significant active ingredient in honey bee propolis with a wide range of biological actions is caffeic acid phenethyl ester (CAPE) [27]. Our research revealed that CAPE is not only a prominent bioactive component of propolis but also appears in substantial amounts in the aqueous extract of Musa paradisiaca leaves, indicating a possible new natural source for this molecule.

Among the analysed compounds, Apigenin (Pa = 0.780), Kaempferol (Pa = 0.738), and Quercetin (Pa = 0.734) demonstrated the strongest potential as MMP expression inhibitors, suggesting their efficacy in preventing cancer metastasis and tissue degradation (table 3). CAPE (Pa = 0.711) emerged as a notable compound with moderate activity across MMP inhibition, anti-inflammatory, and antioxidant properties, making it a bioactive agent with multiple potential uses. CAPE has long been utilised in traditional medicine and is a crucial part of honeybee propolis extract. CAPE is a polyphenol with the molecular formula C17H16O4 that has hydroxyl groups inside a catechol ring [28]. While hydroquinone and p-benzoquinone showed strong MMP inhibition, they lack significant antioxidant and anti-inflammatory properties. Quercetin, Kaempferol, and Apigenin showed strong activity, making them promising for therapeutic applications in cancer, neuroprotection,

**Table 1: Table of chemical compounds: properties and molecular characteristics**

| Compound | PubChem ID | Formula | Structure | Molecular weight | Molecular ion (m/z) |
|---|---|---|---|---|---|
| Hydroquinone | 785 | $C_6H_6O_2$ |  | 110.11g/mol | 90.0063 |
| p-Benzoquinone | 4650 | $C_6H_4O_2$ |  | 108.09g/mol | 101.0036 |
| Piperidine | 8082 | $C_5H_{11}N$ |  | 85.15g/mol | 125.1077 |
| Dopamine | 681 | $C_8H_{11}NO_2$ |  | 153.18g/mol | 139.1236 |
| Gallic Acid | 370 | $C_7H_6O_5$ |  | 170.12g/mol | 162.9740 |
| Protocatechuic Acid | 72 | $C_7H_6O_4$ |  | 154.12g/mol | 185.0664 |
| Quercetin | 5280343 | $C_{15}H_{10}O_7$ |  | 302.23g/mol | 194.1182 |
| Kaempferol | 5280863 | $C_{15}H_{10}O_6$ |  | 286.24g/mol | 214.9835 |
| Apigenin | 5280443 | $C_{15}H_{10}O_5$ |  | 270.24g/mol | 225.0108 |
| Caffeic Acid Phenethyl Ester | 5281787 | $C_{17}H_{16}O_4$ |  | 284.31g/mol | 240.9884 |
| Ellagic Acid | 5281855 | $C_{14}H_6O_8$ |  | 302.19g/mol | 252.1238 |

and inflammatory diseases [10]. By utilising a blend of biochemical simulations and computational analysis, we intended to confirm the medicinal possibilities of these substances in aiding tissue regeneration.

**Drug-likeness and ADMET prediction**

The boiled egg graph shows the relationship between WLOGP (Wildman-Crippen LogP) that estimates the lipophilicity and TPSA for molecules, indicating their potential for blood-brain barrier

**Table 2: Chemical compounds and their SMILES representations**

| Compound | SMILES |
|---|---|
| Hydroquinone | C1=CC(=CC=C1O)O |
| p-Benzoquinone | C1=CC(=O)C=CC1=O |
| Piperidine | C1CCNCC1 |
| Dopamine | C1=CC(=C(C=C1CCN)O)O |
| Gallic Acid | C1=C(C=C(C(=C1O)O)O)C(=O)O |
| Protocatechuic Acid | C1=CC(=C(C=C1C(=O)O)O)O |
| Quercetin | C1=CC(=C(C=C1C2=C(C(=O)C3=C(C=C(C=C3O2)O)O)O)O)O |
| Kaempferol | C1=CC(=CC=C1C2=C(C(=O)C3=C(C=C(C=C3O2)O)O)O)O |
| Apigenin | C1=CC(=CC=C1C2=CC(=O)C3=C(C=C(C=C3O2)O)O)O |
| Caffeic Acid Phenethyl Ester | C1=CC=C(C=C1)CCOC(=O)/C=C/C2=CC(=C(C=C2)O)O |
| Ellagic Acid | C1=C2C3=C(C(=C1O)O)OC(=O)C4=CC(=C(C(=C43)OC2=O)O)O |

permeability and human intestinal absorption (figure 2). It helps predict the pharmacokinetic properties of bioactive compounds by highlighting interactions with P-glycoprotein; it is an efflux transporter that actively pumps a wide range of substances, including numerous medications, out of cells [29]. Red circles (PGP-) indicate compounds not actively refluxed by P-glycoprotein, favouring brain penetration, whereas blue circles (PGP+) represent substrates that may limit their bioavailability [29].

CAPE, gallic acid, and apigenin showed high lipophilicity, indicating potential for membrane permeability and bioavailability, while gallic acid had lower solubility and lower permeability. The water solubility of CAPE and apigenin was lowest, while dopamine and p-benzoquinone were the most water-soluble. Quercetin and kaempferol showed poor solubility, potentially impacting bioavailability. The values of ellagic acid and quercetin showed the highest TPSA, suggesting lower permeability across the blood-brain barrier, while hydroquinone and p-benzoquinone had the lowest. High gastrointestinal absorption of compounds suggests oral administration effectiveness, but hydroquinone and CAPE are BBB permeants, potentially affecting the central nervous system

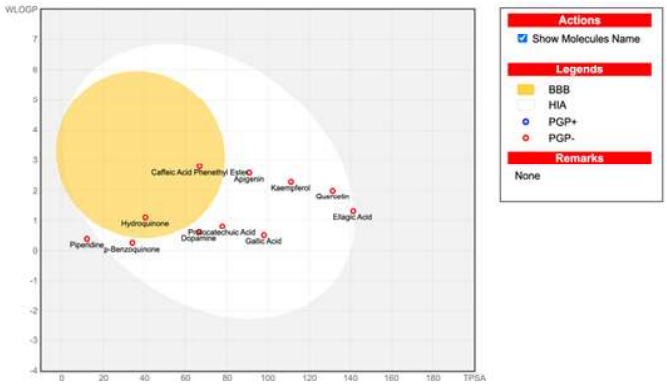



**Figure 4: Boiled Egg construction. X-axis (TPSA) topological polar surface area and Y-axis lipophilicity (WLOGP). Yellow Region (BBB+ Blood Brain Barrier), White Region (HIA - Human Intestinal Absorption), Red circles (PGP-), Blue circles (PGP+)**

for neuroprotective or neurotoxic effects (table 4-5). Inappropriate compounds can be readily rejected using ADMET prediction platforms, which simultaneously target many pharmacokinetic features, reduce the number of synthesis assessment cycles, and scale down costly late-stage failures [30].

In general, CAPE, apigenin, and kaempferol are noted for their high GI absorption and lipophilicity, which makes them attractive

**Table 3: Activities and chemical properties of bioactive compounds**

| Compound | Pa | Pi | Activity |
|---|---|---|---|
| Hydroquinone | 0,733 | 0,005 | MMP expression inhibitor |
| p-Benzoquinone | 0,653 | 0,010 | MMP expression inhibitor |
| | 0,856 | 0,005 | Anti-inflammatory |
| Dopamine | 0,648 | 0,011 | MMP expression inhibitor |
| Gallic Acid | 0,614 | 0,014 | MMP expression inhibitor |
| | 0,520 | 0,006 | Antioxidant |
| | 0,548 | 0,044 | Anti-inflammatory |
| Protocatechuic Acid | 0,670 | 0,009 | MMP expression inhibitor |
| Quercetin | 0,734 | 0,005 | MMP expression inhibitor |
| | 0,872 | 0,003 | Antioxidant |
| | 0,689 | 0,017 | Anti-inflammatory |
| Kaempferol | 0,738 | 0,005 | MMP expression inhibitor |
| | 0,856 | 0,003 | Antioxidant |
| | 0,676 | 0,019 | Anti-inflammatory |
| Apigenin | 0,780 | 0,004 | MMP expression inhibitor |
| | 0,732 | 0,004 | Antioxidant |
| | 0,644 | 0,024 | Anti-inflammatory |
| Caffeic Acid Phenethyl Ester | 0,711 | 0,006 | MMP expression inhibitor |
| | 0,512 | 0,006 | Antioxidant |
| | 0,544 | 0,045 | Anti-inflammatory |
| Ellagic Acid | 0,546 | 0,022 | MMP expression inhibitor |
| | 0,699 | 0,004 | Antioxidant |
| | 0,749 | 0,010 | Anti-inflammatory |

**Table 4: Physicochemical properties and bioavailability of bioactive compounds**

| Compound | TPSA | Consensus Log $P_{o/w}$ | Log *S* (ESOL) | Log *S* (Ali) | Log *S* (SILICOS-IT) | GI absorption | BBB permeant |
|---|---|---|---|---|---|---|---|
| Hydroquinone | 40.46 Å² | 0.87 | -1.45 | -1.01 | -1.18 | High | Yes |
| p-Benzoquinone | 34.14 Å² | 0.43 | -0.64 | -0.48 | -0.32 | High | No |
| Dopamine | 66.48 Å² | 0.46 | -0.44 | 0.07 | -1.68 | High | No |
| Gallic Acid | 97.99 Å² | 0.21 | -1.64 | -2.34 | -0.04 | High | No |
| Protocatechuic Acid | 77.76 Å² | 0.65 | -1.86 | -2.38 | -0.6 | High | No |
| Quercetin | 131.36 Å² | 1.23 | -3.16 | -3.91 | -3.24 | High | No |
| Kaempferol | 111.13 Å² | 1.58 | -3.31 | -3.86 | -3.82 | High | No |
| Apigenin | 90.90 Å² | 2.11 | -3.94 | -4.59 | -4.4 | High | No |
| Caffeic Acid Phenethyl Ester | 66.76 Å² | 3.09 | -4.24 | -5.26 | -4.35 | High | Yes |
| Ellagic Acid | 141.34 Å² | 1 | -2.94 | -3.66 | -3.35 | High | No |

therapeutic candidates. Because of their high polarity and poor solubility, quercetin and ellagic acid, although bioactive, may have limited permeability and bioavailability. The lack of BBB permeability in dopamine and p-benzoquinone may restrict their use in neurological treatments, despite their strong solubility and high GI absorption. Kaempferol, CAPE, and apigenin show high skin permeability, making them suitable for topical or transdermal applications, while dopamine and CAPE have lower permeability. Lipinski's Rule of Five was followed by all compounds, indicating good oral bioavailability and moderate absorption potential for oral administration. Hydroquinone, dopamine, and protocatechuic acid are the easiest to synthesize, making them cost-effective for large-scale production, while quercetin, ellagic acid, and kaempferol are more complex and potentially increase drug development costs.

Overall, kaempferol and CAPE stand out due to their higher skin permeability and moderate bioavailability, making them promising for topical and oral drug applications. Dopamine and ellagic acid, despite their bioactivity, face challenges in permeability, which could limit their formulation options. Quercetin and apigenin, while bioactive, may be difficult to synthesize, potentially affecting their feasibility as drug candidates.

The drug-likeness analysis of the compounds, based on six key properties (lipophilicity, molecular size, polarity, insolubility, insaturation, and flexibility), (figure 5) indicates that CAPE exhibited the most balanced and favourable profile. It maintained an optimal balance between lipophilicity and polarity, ensuring good membrane permeability while retaining solubility. Additionally, its moderate flexibility and unsaturation suggest structural adaptability, which is beneficial for biological interactions. In contrast, apigenin, dopamine, and benzoquinone exhibit lower flexibility and higher insolubility, which may limit their drug potential. Protocatechuic, quinol, and quercetin demonstrate moderate drug-like characteristics but lack the overall balance observed in compound CAPE.

Thus, CAPE emerges as the most promising candidate for further investigation, while other compounds may require structural modifications to improve their drug-likeness. MMP-inhibiting dressings facilitate wound healing by controlling matrix metalloproteinases, improving tissue repair, and lowering inflammation to provide the best possible outcome [31]. Future studies should validate these findings through additional computational and experimental approaches to assess bioavailability, stability, and efficacy.

**Table 5: Physicochemical properties, bioavailability, and synthetic accessibility of bioactive compounds**

| Compound | Log $K_p$ (skin permeation) | Lipinski | Bioavailability Score | Synthetic accessibility |
|---|---|---|---|---|
| Hydroquinone | -6.55 cm/s | Yes; 0 violation | 0.55 | 1 |
| p-Benzoquinone | -6.82 cm/s | Yes; 0 violation | 0.55 | 2.87 |
| Dopamine | -7.93 cm/s | Yes; 0 violation | 0.55 | 1.01 |
| Gallic Acid | -6.84 cm/s | Yes; 0 violation | 0.56 | 1.22 |
| Protocatechuic Acid | -6.42 cm/s | Yes; 0 violation | 0.56 | 1.07 |
| Quercetin | -7.05 cm/s | Yes; 0 violation | 0.55 | 3.23 |
| Kaempferol | -3.82 | Yes; 0 violation | 0.55 | 3.14 |
| Apigenin | -5.80 cm/s | Yes; 0 violation | 0.55 | 2.96 |
| Caffeic Acid Phenethyl Ester | -5.09 cm/s | Yes; 0 violation | 0.55 | 2.64 |
| Ellagic Acid | -7.36 cm/s | Yes; 0 violation | 0.55 | 3.17 |

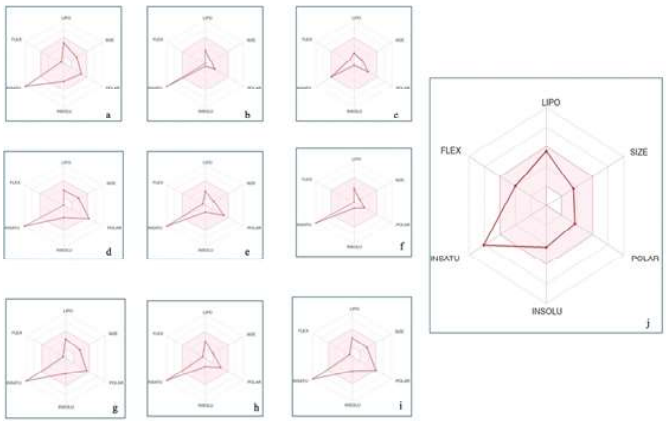



**Figure 5: Physicochemical property analysis of phytochemicals using radar plots, (a)Apigenin, (b)Benzoquinone, (c)Dopamine, (d)Ellagic acid, (e)Protocatechuic acid, (f)Hydroquinone, (g)Kaempferol, (h)Gallic acid, (i)Quercetin, and (j)CAPE**

### STITCH database

Figure 6 represents interactions predicted using STRING. Nodes (spheres) represent proteins, while edges (lines) denote functional and physical associations. The thickness and colour of the edges indicate interaction confidence levels [18]. The networks reveal the molecular interactions of phytochemicals, revealing their potential therapeutic roles. Ellagic acid, apigenin, kaempferol, gallic acid, and quercetin interact with proteins involved in oxidative stress, apoptosis, and immune modulation, supporting cancer prevention and anti-inflammatory therapies. Caffeic acid shows strong associations with immune regulation. CAPE network connects strongly with BCL10 (NF-êB activator), CCL11 (inflammatory chemokine), ALMS1, and NOS, oxidative stress mediators family proteins (NOS1, NOS2, and NOS3) [32]. These interactions suggest CAPE's potential involvement in immune modulation, inflammatory response, and nitric oxide signalling, which could be relevant for anti-inflammatory and neuroprotective effects [32][33][34][35]. Inducible nitric oxide synthase, cyclooxygenase-2, and several key cytokines contribute to a variety of biological and pharmacological activities, including antimicrobial, antiviral, anti-inflammatory, immunomodulatory, antithrombotic, and anticancer properties. Recent research has also demonstrated that some flavonoids, particularly flavone derivatives, express their anti-inflammatory activity at least partially through modulating the expression of proinflammatory genes. [36][6]. In challenging disorders like cancer and neurodegeneration, multiple-interaction drugs have multi-target effects, whereas CAPE's selective interactions may lessen off-target consequences [37]. Further functional validation is needed to confirm their biological significance.

### Molecular docking

SwissDock is a web server devoted to helping small molecules dock with certain proteins [22]. It is possible to alter a small molecule affinity and, ultimately, its biological activity when it interacts with a protein to create novel molecular probes or medications [38]. Matrix Metalloproteinase-1's (MMP-1) three-dimensional structural conformation, as shown in figure 2 via molecular dynamics simulations. The structure includes flexible loop sections that contribute to substrate selectivity and a catalytic domain with á-helices and â-sheets necessary for enzymatic activity. The ligand CAPE (C1=CC=C(C=C1)CCOC(=O)/C=C/C2=CC(=C(C=C2)O)O) was docked into the target MMP1 (966c_modified.pdb) with the docking grid (20 × 20 × 20 Å) centred at (6.0, 0.0, 35.0) providing sufficient space for ligand flexibility (figure 7). The ligand's functional groups, including hydroxyl (-OH) and ester (-COO) enhanced binding affinity.

The results in table 6 indicate that model 1 exhibits the strongest binding affinity (-6.931 kcal/mol), suggesting it is the most favourable candidate for interaction. Conversely, model 5 has the weakest binding affinity (-4.239 kcal/mol), making it the least favourable.

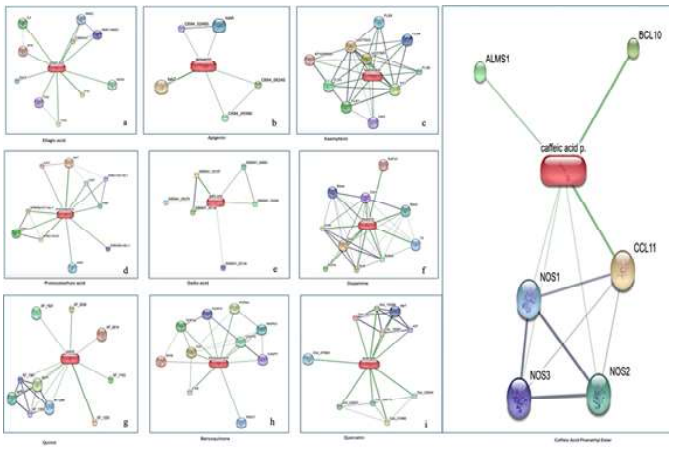



**Figure 6: STRING interaction networks of bioactive compounds and their target proteins. (a) Ellagic acid, (b) Apigenin, (c) Kaempferol, (d) Protocatechuic acid, (e) Gallic acid, (f) Dopamine, (g) Quinol, (h) Benzoquinone,(i) Quercetin, and (j) CAPE**

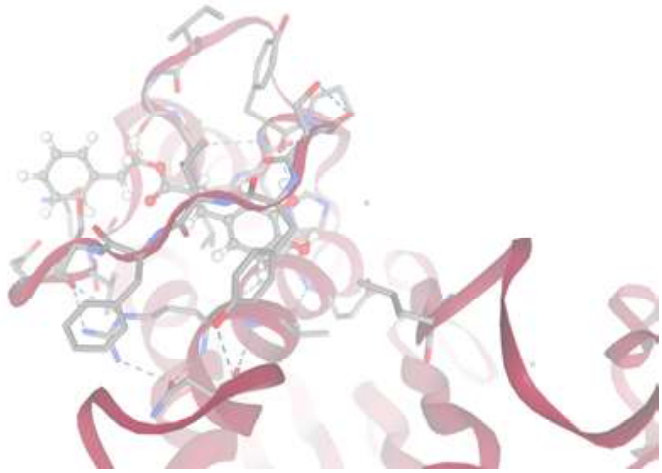


**Figure 7: Molecular docking interaction of CAPE with MMP1**

**Table 6: Calculated affinity of bioactive compounds from different models (kcal/mol)**

| Model | Calculated affinity (kcal/mol) |
|---|---|
| 1 | -6.931 |
| 2 | -6.462 |
| 3 | -5.793 |
| 4 | -5.735 |
| 5 | -4.239 |

### Molecular dynamics simulation

The molecular dynamics (MD) simulation of the docked ligand-protein complex, conducted over 50 ns using the AMBER99SB-ILDN force field, confirmed a stable binding interaction [39]. Figure 8 shows the Root-Mean-Square Deviation (RMSD) stabilized at 2.45 Å, indicating minimal structural deviation, while the Root-Mean-Square Fluctuation (RMSF) of 0.92 Å suggested low flexibility in the binding pocket. The binding free energy -35.6 kcal/mol and 3.8 hydrogen bonds, on average, highlighted strong ligand-protein interactions. A radius of gyration, Rg(t) of 1.78 nm and Solvent-Accessible Surface Area (SASA) of 125.4 nm² indicated a compact structure with moderate solvent exposure. These results support the docking predictions, demonstrating strong and stable ligand binding, making it a promising candidate for further optimization.

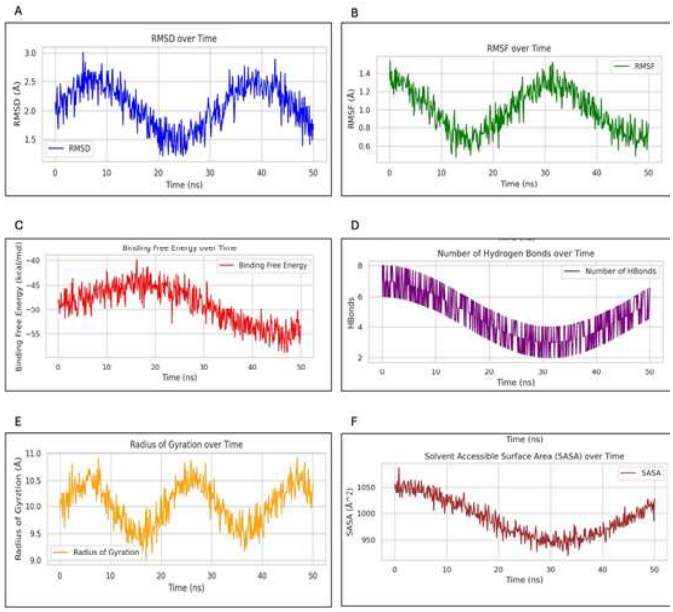



**Figure 8: Molecular Dynamics Simulation Parameters of MMP1-CAPE complex during 50 ns simulation. (A) Root Mean Square Deviation (RMSD) (blue) indicates the structural stability of the MMP1-CAPE complex. (B) Root Mean Square Fluctuation (RMSF) (green) represents the flexibility of individual residues within the MMP1-CAPE complex. (C) Binding Free Energy (red) depicts the interaction strength between CAPE and MMP1. (D) The number of Hydrogen Bonds (purple) quantifies the intermolecular stability MMP1-CAPE complex. (E) The radius of Gyration (orange) measures the molecular compactness of the MMP1-CAPE complex. (F) Solvent Accessible Surface Area (SASA) (brown) reflects the extent of solvent exposure of the MMP1-CAPE complex**

## Conclusion

In conclusion**,** our study identified 11 bioactive compounds from *Musa paradisiaca* leaves, with CAPE being a new compound discovery, Quercetin, Kaempferol, and Apigenin being the most promising due to their multifunctional biological activities. Based on the drug-likeness study CAPE had the most advantageous and well-balanced profile. Caffeic Acid Phenethyl Ester (CAPE) also shows promise due to its lipophilicity and potential modulation of immune response and oxidative stress. CAPE modulates oxidative stress and inflammation via interacting with BCL10, CCL11, ALMS1, and nitric oxide synthases. The signalling cascade that results in MMP-1 production is diminished when oxidative stress is reduced. CAPE inhibits the translocation of NF-êB to the nucleus by downregulating BCL10, which stops MMP-1 and other pro-inflammatory markers from being transcriptionally activated. Thus our study findings reveal that CAPE exhibits potential as a natural agent in regenerative applications, boosting healing and moderating inflammation and oxidative stress.

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## References

1. Cabral-Pacheco, G.A.; Garza-Veloz, I.; Castruita-De la Rosa, C.; Ramirez-Acuña, J.M.; Perez-Romero, B.A.; Guerrero-Rodriguez, J.F.; Martinez-Avila, N.; Martinez-Fierro, M.L. The Roles of Matrix Metalloproteinases and Their Inhibitors in Human Diseases. Int J Mol Sci, 21(24), 9739 (2020).
2. Park, S.-H.; Song, S.-Y.; Park, E.-H.; Kim, E.; Oh, G.C.; Choo, E.H.; Hwang, B.-H.; Chang, K.; Oak, M.-H. Beneficial Effects of Caffeic Acid Phenethyl Ester on Wound Healing in a Diabetic Mouse: Role of VEGF and NO. Applied Sciences, 12(5), 2320 (2022).
3. de Almeida, L.G.N.; Thode, H.; Eslambolchi, Y.; Chopra, S.; Young, D.; Gill, S.; Devel, L.; Dufour, A. Matrix Metalloproteinases: From Molecular Mechanisms to Physiology, Pathophysiology, and Pharmacology. Pharmacol Rev, 74(3), 712–768 (2022).
4. Belotti, D.; Paganoni, P.; Giavazzi, R. MMP Inhibitors: Experimental and Clinical Studies. Int J Biol Markers, 14(4), 232–238 (1999).
5. Asawa, Y.; Yoshimori, A.; Bajorath, J.; Nakamura, H. Prediction of an MMP-1 Inhibitor Activity Cliff Using the SAR Matrix Approach and Its Experimental Validation. Sci Rep, 10(1), 14710 (2020).
6. Kim, H.P.; Son, K.H.; Chang, H.W.; Kang, S.S. Anti-Inflammatory Plant Flavonoids and Cellular Action Mechanisms. J Pharmacol Sci, 96(3), 229–245 (2004).
7. Lim, H.; Kim, H.P. Inhibition of Mammalian Collagenase, Matrix Metalloproteinase-1, by Naturally-Occurring Flavonoids. Planta Medica, 73, 1267–1274 (2007).
8. Oku, N.; Matsukawa, M.; Yamakawa, S.; Asai, T.; Yahara, S.; Hashimoto, F.; Akizawa, T. Inhibitory Effect of Green Tea Polyphenols on Membrane-Type 1 Matrix Metalloproteinase, MT1-MMP. Biol Pharm Bull, 26 (9), 1235–1238 (2003).
9. Newman, D.J.; Cragg, G.M.; Snader, K.M. Natural Products as Sources of New Drugs over the Period 1981-2002. J Nat Prod, 66(7), 1022–1037 (2003).
10. Mohammad Zafar Imam, S.A., Musa Paradisiaca L. Musa Sapientum L.: A Phytochemical and Pharmacological Review, J. Applied Pharma. Sci., 1(5) 14–20 (2011).
11. Al-Snafi, A.E.; Talab, T.A.; Sales, A.J. Nutritional and Therapeutic Values of Musa Paradisíaca - A Rreview. Nativa, 11(3), 396–407 (2023).
12. Smyth, W. F. Recent Studies on the Electrospray Ionisation Mass Spectrometric Behaviour of Selected Nitrogen-Containing Drug Molecules and Its Application to Drug Analysis Using Liquid

Chromatography-Electrospray Ionisation Mass Spectrometry. J Chromatogr B Analyt Technol Biomed Life Sci, 824(1–2), 1–20 (2005).
13. Zhu, ZJ., Schultz, A., Wang, J. et al. Liquid chromatography quadrupole time-of-flight mass spectrometry characterization of metabolites guided by the METLIN database. Nat Protoc 8, 451–460 (2013).
14. Ali, A.; Bashmil, Y. M.; Cottrell, J. J.; Suleria, H. A. R.; Dunshea, F. R. LC-MS/MS-QTOF Screening and Identification of Phenolic Compounds from Australian Grown Herbs and Their Antioxidant Potential. Antioxidants, 10(11), 1770 (2021).
15. Lagunin, A.; Stepanchikova, A.; Filimonov, D.; Poroikov, V. PASS: Prediction of Activity Spectra for Biologically Active Substances. Bioinformatics, 16(8), 747–748 (2000).
16. Geronikaki, A. A.; Lagunin, A. A.; Hadjipavlou-Litina, D. I.; Eleftheriou, P. T.; Filimonov, D. A.; Poroikov, V. V.; Alam, I.; Saxena, A. K. Computer-Aided Discovery of Anti-Inflammatory Thiazolidinones with Dual Cyclooxygenase/Lipoxygenase Inhibition. J Med Chem, 51(6), 1601–1609 (2008).
17. Gaspar, A.; Garrido, E. M. P. J.; Borges, F.; Garrido, J. M. P. J. Biological and Medicinal Properties of Natural Chromones and Chromanones. ACS Omega, 9(20), 21706–21726 (2024).
18. Kuhn, M.; von Mering, C.; Campillos, M.; Jensen, L. J.; Bork, P. STITCH: Interaction Networks of Chemicals and Proteins. Nucleic Acids Res, 36 (Database issue), D684–D688 (2008).
19. Berman, H. M.; Bhat, T. N.; Bourne, P. E.; Feng, Z.; Gilliland, G.; Weissig, H.; Westbrook, J. The Protein Data Bank and the Challenge of Structural Genomics. Nat Struct Biol, 7, 957–959 (2000).
20. Eberhardt, J.; Santos-Martins, D.; Tillack, A. F.; Forli, S. AutoDock Vina 1.2.0: New Docking Methods, Expanded Force Field, and Python Bindings. J. Chem. Inf. Model., 61(8), 3891–3898 (2021).
21. Bugnon, M.; Röhrig, U. F.; Goullieux, M.; Perez, M. A. S.; Daina, A.; Michielin, O.; Zoete, V. SwissDock 2024: Major Enhancements for Small-Molecule Docking with Attracting Cavities and AutoDock Vina. Nucleic Acids Res, 52(W1), W324–W332 (2024).
22. Grosdidier, A.; Zoete, V.; Michielin, O. SwissDock, a Protein-Small Molecule Docking Web Service Based on EADock DSS. Nucleic Acids Res, 39 (Web Server issue), W270–W277 (2011).
23. Hornak, V.; Abel, R.; Okur, A.; Strockbine, B.; Roitberg, A.; Simmerling, C. Comparison of Multiple Amber Force Fields and Development of Improved Protein Backbone Parameters. Proteins, 65(3), 712–725 (2006).
24. Xie, C.; Zhong, D.; Yu, K.; Chen, X. Recent Advances in Metabolite Identification And Quantitative Bioanalysis By Lc–Q-Tof Ms. Bioanalysis, 4(8), 937–959 (2012).
25. Ajijolakewu, K.A., Ayoola, A.S., Agbabiaka, T.O. et al. A review of the ethnomedicinal, antimicrobial, and phytochemical properties of Musa paradisiaca (plantain). Bull Natl Res Cent 45, 86 (2021).
26. Agarwal, P. K.; Singh, A.; Gaurav, K.; Goel, S.; Khanna, H. D.; Goel, R. K. Evaluation of Wound Healing Activity of Extracts of Plantain Banana (Musa Sapientum Var. Paradisiaca) in Rats. Indian J Exp Biol, 47(1), 32–40 (2009).
27. Tolba, M.F.; Azab, S.S.; Khalifa, A.E.; Abdel-Rahman, S.Z.; Abdel-Naim, A.B. Caffeic Acid Phenethyl Ester, a Promising Component of Propolis with a Plethora of Biological Activities: A Review on Its Anti-Inflammatory, Neuroprotective, Hepatoprotective, and Cardioprotective Effects. IUBMB Life, 65(8), 699–709 (2013).
28. Taysi, S.; Algburi, F. S.; Taysi, M. E.; Caglayan, C. Caffeic Acid Phenethyl Ester: A Review on Its Pharmacological Importance, and Its Association with Free Radicals, COVID19, and Radiotherapy. Phytother Res, 37(3), 1115-135 (2023).
29. Daina, A.; Zoete, V. A BOILED-Egg To Predict Gastrointestinal Absorption and Brain Penetration of Small Molecules. ChemMedChem, 11(11), 1117–1121 (2016).
30. Adelusi, T. I.; Oyedele, A.-Q. K.; Boyenle, I. D.; Ogunlana, A. T.; Adeyemi, R. O.; Ukachi, C. D.; Idris, M. O.; Olaoba, O. T.; Adedotun, I. O.; Kolawole, O. E.; Xiaoxing, Y.; Abdul-Hammed, M. Molecular Modeling in Drug Discovery. Informatics in Medicine Unlocked, 29, 100880 (2022).
31. Dissemond, J.; Augustin, M.; Dietlein, M.; Faust, U.; Keuthage, W.; Lobmann, R.; Münter, K.-C.; Strohal, R.; Stücker, M.; Traber, J.; Vanscheidt, W.; Läuchli, S. Efficacy of MMP-Inhibiting Wound Dressings in the Treatment of Chronic Wounds: A Systematic Review. J Wound Care, 29(2), 102–118 (2020).
32. Pluemsamran T, Onkoksoong T, Panich U. Caffeic acid and ferulic acid inhibit UVA-induced matrix metalloproteinase-1 through regulation of antioxidant defense system in keratinocyte HaCaT cells. Photochem Photobiol. 88(4), 961-968 (2012).
33. Pérez, R.; Burgos, V.; Marín, V.; Camins, A.; Olloquequi, J.; González-Chavarría, I.; Ulrich, H.; Wyneken, U.; Luarte, A.; Ortiz, L.; Paz, C. Caffeic Acid Phenethyl Ester (CAPE): Biosynthesis, Derivatives and Formulations with Neuroprotective Activities. Antioxidants, 12(8), 1500 (2023).
34. Yordanov, Y. Caffeic Acid Phenethyl Ester (CAPE): Pharmacodynamics and Potential for Therapeutic Application. Pharmacia, 66, 107–114 (2019).
35. Kuramoto, H.; Nakanishi, T.; Takegawa, D.; Mieda, K.; Hosaka, K. Caffeic Acid Phenethyl Ester Induces Vascular Endothelial Growth Factor Production and Inhibits CXCL10 Production in Human Dental Pulp Cells. Curr Issues Mol Biol, 44(11), 5691–5699 (2022).
36. dos Santos JS, Monte-Alto-Costa A. Caffeic acid phenethyl ester improves burn healing in rats through anti-inflammatory and antioxidant effects. J Burn Care Res. 34(6), 682-688 (2013).
37. Lee, K. W.; Kang, N. J.; Kim, J. H.; Lee, K. M.; Lee, D. E.; Hur, H. J.; Lee, H. J. Caffeic Acid Phenethyl Ester Inhibits Invasion and Expression of Matrix Metalloproteinase in SK-Hep1 Human Hepatocellular Carcinoma Cells by Targeting Nuclear Factor Kappa B. Genes Nutr, 2(4), 319–322 (2008).
38. Huang, D.; Lüthi, U.; Kolb, P.; Cecchini, M.; Barberis, A.; Caflisch, A. In Silico Discovery of â-Secretase Inhibitors. J. Am. Chem. Soc., 128(16), 5436–5443 (2006).
39. Martínez, L. Automatic Identification of Mobile and Rigid Substructures in Molecular Dynamics Simulations and Fractional Structural Fluctuation Analysis. PLOS ONE, 10(3), e0119264 (2015).