







# Molecular Docking Analysis of Chlorogenic Acid Against Matrix Metalloproteinases (MMPs)

Dhivyadharshini Govindharaj <sup>1</sup>, Saraswathy Nachimuthu <sup>1\*</sup>, Dymphan F Gonsalves <sup>1</sup>, Ram Kothandan <sup>1</sup>, Bhaarathi Dhurai <sup>2</sup>, Lakshminarayanan Rajamani <sup>3</sup>, Seeram Ramakrishana <sup>4</sup>

<sup>1</sup> Department of Biotechnology, Kumaraguru College of Technology, Coimbatore, 641049; dhivyadharshini.18mbt@kct.ac.in; saraswathy.n.bt@kct.ac.in; dymphan.17phd@kct.ac.in; ram.k.bt@kct.ac.in;

<sup>2</sup> Department of Fashion Technology, Kumaraguru College of Technology, Coimbatore -641049; bhaarathidhurai.txt@kct.ac.in;

<sup>3</sup> Anti-infective Research Group, Singapore Eye Research Institute (SERI), Singapore 169856; lakshminarayanan.rajamani@seri.com.sg;

<sup>4</sup> Centre for Nanofibre and Technology, Faculty of Engineering, National University of Singapore (NUS), Singapore; seeram@nus.edu.sg;

\* Correspondence: saraswathy.n.bt@kct.ac.in;

Scopus Author ID 6506474744

Received: 10.05.2020; Revised: 28.05.2020; Accepted: 30.05.2020; Published: 31.05.2020

**Abstract:** Wound healing is one of the most critical and complex processes in the human system, which involves many enzymes. Overexpression of Matrix metalloproteinases (MMPs) at the wound site delay the wound healing process. These overexpressed MMPs can be down-regulated or inhibited using small bioactive molecules derived from natural sources. Chlorogenic acid is a polyphenol derivative found in coffee and a well-known antioxidant. The main objective of the study is to unveil the molecular mechanism by which chlorogenic acid binds to the MMPs through molecular docking studies. The result of docking studies showed that chlorogenic acid showed an excellent binding affinity towards all four selected MMPs. The free binding energy of MMPs 2, 3, 8, and 12 were about -9.32, -8.17, -8.85, and -7.431kcal/mol, respectively. Thus, chlorogenic acid can be used to regulate the activity of metalloproteinases and help to promote wound healing activity.

**Keywords:** Chlorogenic acid, Matrix metalloproteinases, Antioxidant, Molecular Docking.

© 2020 by the authors. This article is an open-access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

## 1. Introduction

Skin is the largest and essential part of the body that protects the body from invasion of microorganisms [1, 2]. The normal healthy skin is remarkably intact and can control the entry and growth of microbes [3, 4]. Any damage in the skin that causes the loss of integrity of the skin is termed as wound. Generally, a wound is a regular event in every individual's life [5]. There are a series of events that help in wound healing [6]. The four events of wound healing are hemostasis, inflammation, proliferation, and tissue remodeling [7, 8]

The wound site consists of various enzymes that modulate the wound healing process [9]. Enzymes that are present in the extracellular matrix (ECM) are called matrix metalloproteinases (MMPs), also known as matrixins [10, 11]. MMPs belong to endoproteases that depend on zinc for their activity [12, 13]. There are about 23 MMPs in human beings, which are divided into six classes [14]. Gelatinase (MMP2), stromelysins (MMP3), collagenase (MMP8), and metalloelastase (MMP12) are few MMPs that are mainly involved in tissue repairing process [15, 16]. Under normal condition, MMPs are secreted in a balanced manner,

but due to oxidative stress in the wound site, there is an improper regulation in MMPs and endogenous regulatory process, which leads to degradation of newly formed ECM by MMPs, thus delaying the wound healing process and leading to chronic wound infections [17].

Many medicinal plants have been reported to possess compounds that are showing significant wound healing activity [18], which possess less or no side effects compared to synthetic compounds [19]. Chlorogenic acid (CGA) is a secondary metabolite belongs to a group of phenolic compounds [20], and it is majorly present in plants such as coffee, vegetables such as potatoes and fruits including apples, pears, berries, etc., [21, 22]. It is known to show free radical and metal scavenging activities [23, 24]. IUPAC chemical name of CGA is called 1,3,4,5-tetrahydroxy cyclohexane carboxylic acid 3-(3,4-dihydroxycinnamate) [25, 26]. It is known to show that CGA possesses antioxidant, anticarcinogenesis, hepato-protectant [27, 28] and also plays a significant role in the wound healing process [29]. CGA is helpful in the wound healing process by increasing hydroxyproline content, diminishing nitric oxide levels, and also promotes reduced-glutathione levels in wound bed [30].

Molecular docking is the *in silico* approach where the protein and ligand are used to find the best interactions between them. Hence, the results from *in silico* studies could be used to find the relevant information before *in vitro* and *in vivo* studies. This study is to understand of the interaction of CGA with and MMPs that are involved in the wound healing process.

## 2. Materials and Methods

### 2.1. Protein preparation.

The three-dimensional structure of the selected matrix metalloproteinases (MMPs) were retrieved from RCSB database (<https://www.rcsb.org/>) in Protein Data Bank (PDB) format. The four selected MMPs are of *Homo sapiens* origin, and PDB code of MMP 2, MMP 3, MMP 8 and MMP 12 is 1QIB, 2DIO, 1MNC, and 1HNE, respectively [16].

### 2.2. Ligand preparation.

The three-dimensional structure of the CGA was retrieved from the PubChem database (<https://pubchem.ncbi.nlm.nih.gov/>). The compound ID of CGA is 1794427 with the molecular formula of C<sub>16</sub>H<sub>18</sub>O<sub>9</sub>, and the molecular weight is 354.31 g/mol. The CGA was screened by using an online server <http://www.scfbio-iitd.res.in/software/drugdesign/lipinski.jsp> in order to find its physicochemical properties such as molecular weight, hydrogen bond donors, hydrogen bond acceptors, lipophilicity and molar refractivity [31, 32] and its properties were compared with Lipinski's rule of five [33, 34].

### 2.3. Active sites of MMPs.

The active site is one of the essential criteria in docking studies where the ligand interacts with the protein's active site amino acid residues to give effective binding interactions. In the present study, the experimentally verified active site residues of the selected MMPs, MMP2, MMP 3, MMP8, and MMP12 were considered based on literature reports [35–38].

### 2.4. Dock preparation.

The retrieved MMPs were processed using USFC Chimera 1.31.1. The additional chains present in the protein structure were removed, and only one chain was used. During dock preparation, ions, ligands, and water molecules present in each protein crystal structure

were removed. Then, the charges were added additionally to other residues (Gasteiger) and standard residues (AMBERff14SB). These dock prepared proteins were used for further docking process. Similarly, the CGA structure was also dock prepared using USFC Chimera 1.31.1 [39, 40].

### *2.5. Molecular docking process.*

AutoDock tool 1.5.6 was used for analyzing the docking studies between MMPs and CGA. The dock prepared MMPs and CGA were reconstructed into PDBQT format. For CGA, a torsion tree was applied to rotate all the rotatable bonds. The original grid spacing of 0.375 Å was remodeled according to the active site residues and fitted into the active site cavity of the protein molecules. The chlorogenic acid was docked against all the four selected MMPs; MMP2, MMP 3, MMP8, and MMP12 separately. Here, Lamarckian genetic algorithm (LGA) was used, and the population size was about 150, the maximum number of generations was about 27000. The rate of gene mutation and crossover was set about 0.02 and 0.8, respectively [41]. Based on the RMSD and affinity score, out of 10 conformations, one of the best docking pose for each MMP was taken for further analysis. The resulting best conformation of CGA and MMP was visualized using USFC Chimera 1.31.1. The hydrogen bonds between active site residues of MMP and CGA were also noted for interaction analysis [42, 43].

## **3. Results and Discussion**

### *3.1. Protein and ligand preparation.*

MMPs are a group of enzymes involved in the wound healing process, and four MMPs such as MMP2, MMP3, MMP8, and MMP12 were selected for the docking study. Under natural conditions, these MMPs are regulated in a balanced manner, but under certain conditions like a chronic wound, these MMPs activity is upregulated, and it leads to delay in wound healing [44, 45]. CGA is a natural bioactive compound found in coffee was selected as a ligand to study the interaction with the selected MMPs [46]. The 3D structure of selected MMPs was retrieved from RCSB in PDB format, and CGA structure was obtained from PubChem database are shown in Figures 1 and 2. Selected MMPs and CGA were docks prepared using chimera software before subjecting to the docking process. The dock preparation includes removal of ions, ligands, the addition of hydrogen bonds, and also the addition of Gasteiger charges.

### *3.2. Ligand properties.*

The ligand properties of CGA were predicted using the Lipinski rule of five, and the detail is shown in table 1. The results showed that CGA follows all the five rules except one property. The number of hydrogen donors for CGA was found to be six whereas it is expected less than five. However, Lipinski's rule of five does not apply to natural compounds [47].

### *3.3. Active site prediction of MMPs.*

The amino acid residues present in the active site of selected MMPs, MMP2, MMP3, MMP8, and MMP12, are listed in table 2. Phe, Asn, Tyr, Asp, His, Ser, Val, Glu, etc., were some of the active site residues. These residues show stronger binding interaction with the ligand molecule.

### 3.4. Docking interaction and analysis.

The docking studies were done using AutoDock Tool 1.5.6. The dock prepared CGA was docked with all four selected dock prepared MMPs. The best interactions between MMPs and CGA were predicted using the Root Mean Square Deviation (RMSD) score and binding affinity values. The selected best poses were analyzed for hydrogen bond donor residues and also the relative bond distances between the MMPs active sites and CGA.

#### 3.4.1. Interaction between CGA and MMP 2.

The molecular interaction between MMP2 and CGA was visualized using chimera software is shown in figure 3a. From figure 3a, it was found that CGA fits into the active site pocket, and it interacts with amino acid residues of MMP 2 that is likely to show more excellent inhibition activity in under in vivo condition. CGA interaction with MMP 2 (Gelatinase) showed the second-highest affinity score of about - 9.32 kcal/mol out of 10 poses and RMSD score 72.55. The bond length and hydrogen bonding residues are listed in table 3. Thus, CGA could help in regulating the enzyme gelatinase and thereby enhancing the amount of gelatin, collagen, elastin, and fibronectin in the wound site and decreasing the aggregation of platelets, pro-inflammatory factors and also reducing proteolytic affect at the wound site thereby faster ECM development and helps in the wound healing process [16, 48].

#### 3.4.2. Interaction between CGA and MMP 3.

MMP3 belongs to stromelysins subfamily, and it enhances the production of laminin, aggregan, gelatin, and fibronectin in wound site, which leads to a faster wound healing process. The hydrogen bonding that is formed between active site residues of MMP 3 and CGA was visualized using chimera and shown in figure 3b. The bond length and hydrogen bonding residues are given in table 3. It showed the interactions score between MMP3 and CGA of about - -8.17 kcal/mol out of 10 poses and RMSD scores 93. Figure 3b indicates that CGA fits into the active site pocket of MMP 3 and forms hydrogen bonds with interacting amino acid residues. Hence CGA can be considered as a potential inhibitor molecule. It can down-regulate the MMP 3 (stromelysins) activity and which in turn reduces the activity of TGF- $\beta$  and anti-inflammatory factors at the wound site [27].

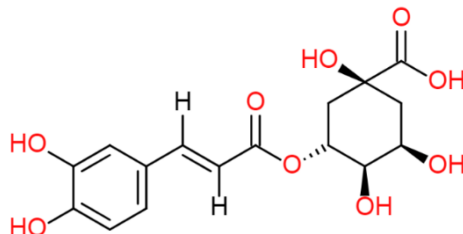
#### 3.4.3. Interaction between CGA and MMP 8

MMP 8 belongs to the collagenase 2 subfamily, which is involved in the degradation of collagen deposition in the extracellular matrix [49]. Excess degradation of collagen type I results in a decrease in wound healing. Therefore, inhibiting the activity of MMP 8 at the wound site is essential to enhance the collagen deposition [50]. Interaction between MMP8 and CGA through hydrogen bonding was visualized using chimera is shown in figure 3c. From figure 3c, it was found that CGA fits into the active site pocket, and out of four MMPs selected, MMP 8 showed maximum affinity score of about - 8.85kcal/mol out of 10 poses and RMSD score was found to be 34.03. Hence, CGA may provide more excellent inhibitor activity towards the MMP 8. The bond length and hydrogen bonding residues were listed in table 3. Regulating the collagenase at the wound site can help to decrease the degradation of collagen at the wound site, and increase the accumulation of gelatin, aggregan, fibronectin synthesis. Inactivation of MMP 8 leads to lowering the activity of chemokines and cell migration towards the wound site, which leads to reducing the inflammatory activity and promoting the wound healing process [51].

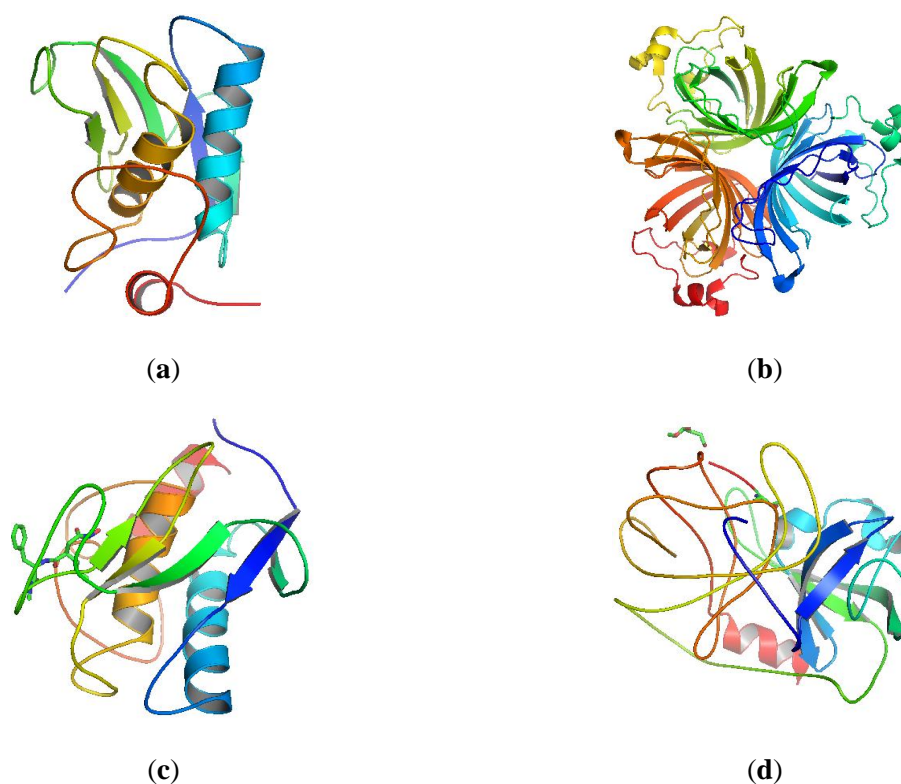
#### 3.4.4 Interaction between CGA and MMP 12

MMP 12 belongs to metalloelastase group of enzymes, which are essential for tissue repairing process. But overexpression of MMP 12 results in infiltration of inflammatory cells and delays wound healing [52]. Docking between CGA and MMP12 showed that CGA fit into the active site of the MMP

12, and the interaction was visualized using chimera and represented in figure 3d. Hydrogen bond length and hydrogen bonding residues in the active site of MMP12 are listed in table 3. From the figure 3d, it was found that CGA fits to the active site pocket of MMP12 with affinity score of binding was estimated to -7.43 kcal/mol out of 10 poses and RMSD score of about 64.84. The inhibitory nature of CGA under in vivo condition will help in reducing the activity of MMP12 activity at the wound site and enhance the accumulation of elastin, gelatin, collagen, fibronectin, laminin, vitronectin, proteoglycan, and angiogenesis help in faster wound healing process [53].



**Figure 1.** Structure of Chlorogenic acid.



**Figure 2.** Structure of Matrix Metalloproteinases (MMPs) (a)MMP2, (b)MMP3, (c)MMP8 and (d)MMP12.

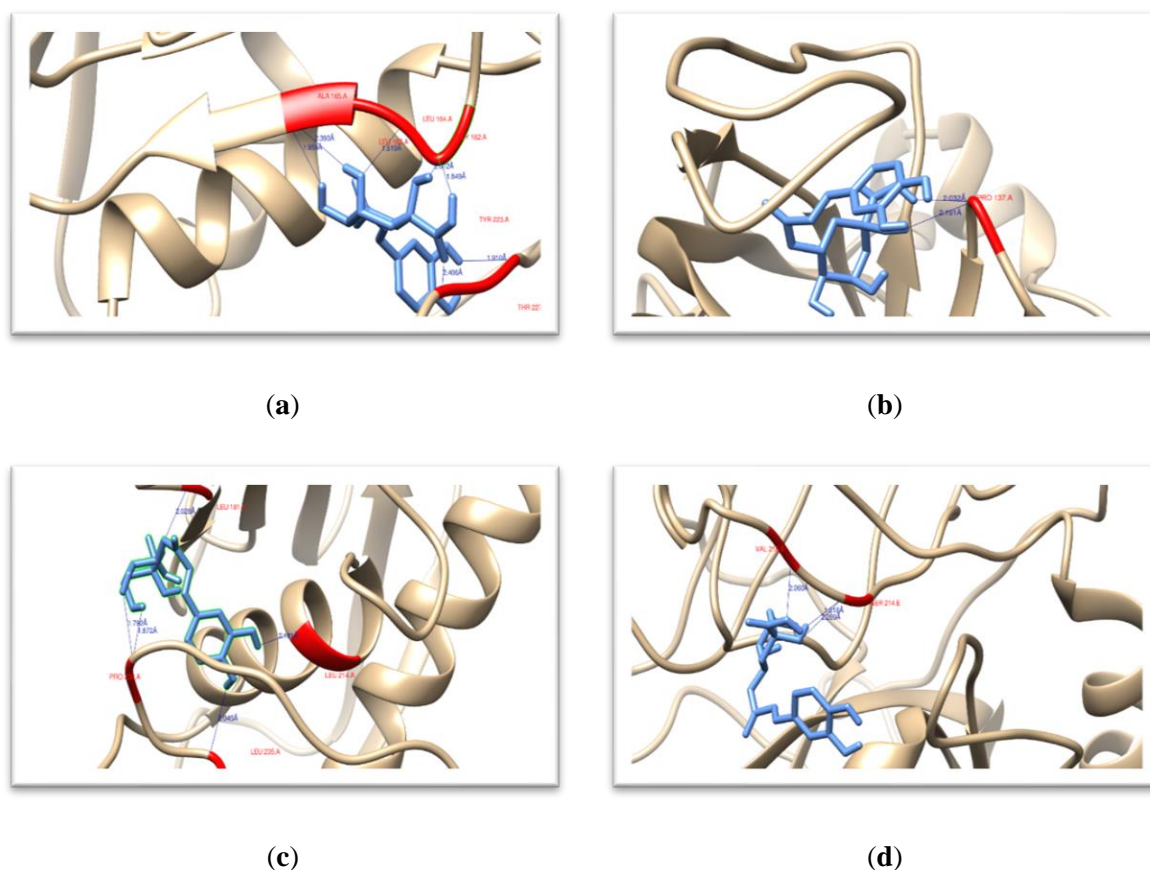
**Table 1.** Lipinski rule of Five.

Lipinski rule	Accepted values	Value for Chlorogenic acid
Molecular mass(Da)	< 500	354.0
Hydrogen bond donor	< 5	6
Hydrogen bond acceptors	< 10	9
High lipophilicity (LOGP)	< 5	-0.6459
Molar refractivity	40-130	82.518768

**Table 2.** Active site residues present in selected MMPs.

Enzymes	Residues in the active site	Amino acids	Reference
MMP2	162, 163, 164, 165, 201, 202, 221, 222, 223	Asn, Val, Leu, Ala, His, Glu, Pro, Leu, Tyr	[35]
MMP3	23, 25, 31, 43, 49, 51, 53, 71, 85, 89, 105, 136	Glu, Asn, Ser, Phe, Val, Phe, Cys, Phe, Tyr, Tyr, Tyr	[36]
MMP8	179, 181, 182, 219, 238, 240	Gly, Leu, Ala, Glu, Pro, Tyr	[37]
MMP12	57, 102, 192, 193, 194, 195, 213, 214, 215, 216, 226	His, Asp, Phe, Gly, Asp, Ser, Ala, Ser, Phe, Val, Asp	[38]





**Figure 3.** Hydrogen bonding interaction of CGA in the active site of MMPs (a) MMP2 (b) MMP3 (c) MMP8 (d) MMP12

**Table 3.** Molecular interactions observed between CGA and selected MMPs.

Receptor protein	No. of H bonds present	Amino acid residues that forms H bond	Length of H-bond (Å)	Binding energy (kcal/mol)	Inhibitor constant $k_i$ (micromolar) at temperature 298.15K	Final intermolecular energy (kcal/mol)	Reference RMSD	Cluster RMSD
MMP2	6	Gly 162	1.849	-9.32	1.50	-11.61	72.55	0
			2.072					
		Leu 164	1.819					
		Ala 165	1.959					
		Try 223	2.939					
	2.406							
MMP3	2	Pro 137	2.032	-8.17	5.56	- 10.45	93	0
			2.191					
MMP8	2	Leu 181 Pro 238	2.028	-8.85	51.82	-9.13	34.03	0
			1.792					
			1.872					
MMP12	2	Ser 214 Val 216	1.818	-7.43	710.69	-7.58	64.84	0
			2.269					
			2.060					

#### 4. Conclusions

The molecular docking studies between CGA and MMPs revealed that CGA is a potential inhibitory molecule. From the interaction results, it is observed that CGA fits into the active site of MMPs and also interacts with active site amino acid residues present in the MMPs through hydrogen bonding. It is also shown that CGA showed an affinity towards all four MMPs. Further, *in vivo* studies to be carried out to evaluate the inhibitory potential of CGA to

down-regulate excess activity of selected the MMPs in the chronic wound site to promote the faster wound healing process.

## Funding

This research was funded by MHRD, Government of India, under the SPARC scheme (SPARC/2018-2019/141/SL(IN) dated 18<sup>th</sup> March 2019).

## Acknowledgments

This research has no acknowledgment.

## Conflicts of Interest

The authors declare no conflict of interest.

## References

1. Yousef, H.; Alhadj, M.; Sharma, S. Anatomy, skin (integument). *Epidermis* **2019**. Treasure Island (FL): StatPearls Publishing, <https://www.ncbi.nlm.nih.gov/books/NBK470464/>
2. Polańska, A.; Cieplewicz, P.; Adamski, Z.; Żaba, R.; Dańczak-Pazdrowska, A. The influence of ultraviolet radiation on the aging process of the skin. *Journal of Face Aesthetics* **2019**, *2*, 28-37, <https://doi.org/10.20883/jofa.8>.
3. Mir, M.; Ali, M.N.; Barakullah, A.; Gulzar, A.; Arshad, M.; Fatima, S.; Asad, M. Synthetic polymeric biomaterials for wound healing: a review. *Progress in Biomaterials* **2018**, *7*, 1-21, <https://doi.org/10.1007/s40204-018-0083-4>.
4. Grada, A.; Obagi, Z.; Phillips, T. Management of chronic wounds in patients with pemphigus. *Chronic Wound Care Management and Research* **2019**, *6*, 89, <https://doi.org/10.2147/CWCMS.S141948>.
5. Price, A.; Naik, G.; Harding, K. Skin repair technology. In: *Biomaterials for Skin Repair and Regeneration*. Woodhead Publishing. 2019; 27-57, <https://doi.org/10.1016/B978-0-08-102546-8.00002-9>.
6. Perchyonok, V.T. Copazan herbal gel and wound healing in vitro: Assessment of the functional biomaterial for veterinary application. *Biointerface Research in Applied Chemistry* **2018**, *8*, 3084-3088.
7. Bhatt, T.; Kansagar, G.; Pincha, N.; Jamora, C. How does the skin heal wounds? *Iwonder* **2019**, *3*, 13-18. <http://publications.azimpremjifoundation.org/id/eprint/2092>
8. Gonzalez, A.C.d.O.; Costa, T.F.; Andrade, Z.d.A.; Medrado, A.R.A.P. Wound healing - A literature review. *Anais Brasileiros de Dermatologia* **2016**, *91*, 614-620, <http://dx.doi.org/10.1590/abd1806-4841.20164741>.
9. De Sousa, F.; Rogênio, F.; Mendes, F.; Bermudez, J.; Brandão da Silva, A.; Da, M.; Vasconcelos, M.; Lourenzoni, M.; Oliveira Filho, R.; Campos, A.; De, R.; Moreira, A.; Cristina, A.; Monteiro-Moreira, O. Plant Macromolecules as Biomaterials for Wound Healing. 2019; In: *Wound Healing*. IntechOpen 2019; 21, <https://doi.org/10.5772/intechopen.89105>.
10. Nguyen, T.T.; Mobashery, S.; Chang, M. Roles of matrix metalloproteinases in cutaneous wound healing. *Wound Healing - New insights into Ancient Challenges* **2016**, 37-71, <http://dx.doi.org/10.5772/64611>.
11. Rousselle, P.; Montmasson, M.; Garnier, C. Extracellular matrix contribution to skin wound re-epithelialization. *Matrix Biology* **2019**, 75-76, 12-26, <https://doi.org/10.1016/j.matbio.2018.01.002>.
12. Krejner, A.; Litwiniuk, M.; Grzela, T. Matrix metalloproteinases in the wound microenvironment: therapeutic perspectives. *Chronic Wound Care Management and Research* **2016**, *23*, 29-39, <https://doi.org/10.2147/CWCMS.S73819>.
13. Ibrahim, W.; Syaiful, M.; Doolaanea, A.A. Matrix Metalloproteinases In Cancer Biology: A Review. *International Medical Journal Malaysia* **2019**, *18*, 147-152. <https://journals.iium.edu.my/kom/index.php/imjm/article/view/75>
14. Jabłońska-Trypuć, A.; Matejczyk, M.; Rosochacki, S. Matrix metalloproteinases (MMPs), the main extracellular matrix (ECM) enzymes in collagen degradation, as a target for anticancer drugs. *Journal of Enzyme Inhibition and Medicinal Chemistry* **2016**, *31*, 177-183, <https://doi.org/10.3109/14756366.2016.1161620>.
15. Li, K.; Tay, F.R.; Yiu, C.K.Y. The past, present and future perspectives of matrix metalloproteinase inhibitors. *Pharmacology & Therapeutics* **2020**, 207, <https://doi.org/10.1016/j.pharmthera.2019.107465>.
16. Selvaraj, G.; Kaliyamurthi, S.; Thiruganasambandam, R. Molecular docking studies of rutin on matrix metalloproteinase. *Insights Biomed* **2016**, 1(1).

17. Lallemand, L.A.; Zubieta, C.; Lee, S.G.; Wang, Y.; Acajjaoui, S.; Timmins, J.; McSweeney, S.; Jez, J.M.; McCarthy, J.G.; McCarthy, A.A. A Structural Basis for the Biosynthesis of the Major Chlorogenic Acids Found in Coffee. *Plant Physiology* **2012**, *160*, 249–260, <https://doi.org/10.1104/pp.112.202051>.
18. Ibrahim, N.; Wong, K.S.; Mohamed, N.I.; Mohamed, N.; Chin, K.-Y.; Ima-Nirwana, S.; Shuid, N.A. Wound Healing Properties of Selected Natural Products. *International Journal of Environmental Research and Public Health* **2018**, *15*, <https://doi.org/10.3390/ijerph15112360>.
19. Hassan, A., Ullah, H. IsrarMatejczyk, M., The antioxidant activity and phytochemical analysis of medicinal plant *Veronica biloba*. *Letter in applied nanaoscience*. **2019**. 8 (4), 732 – 738. <https://doi.org/10.33263/LIANBS84.732738>
20. Matejczyk, M.; Ofman, P.; Dąbrowska, K.; Świsłocka, R.; Lewandowski, W. The study of biological activity of transformation products of diclofenac and its interaction with chlorogenic acid. *Journal of Environmental Sciences* **2020**, *91*, 128-141., <https://doi.org/10.1016/j.jes.2020.01.022>.
21. Bagdas, D.; Gul, N.Y.; Topal, A.; Tas, S.; Ozyigit, M.O.; Cinkilic, N.; Gul, Z.; Etoz, B.C.; Ziyank, S.; Inan, S.; Turacozen, O.; Gurun, M.S. Pharmacologic overview of systemic chlorogenic acid therapy on experimental wound healing. *Naunyn-Schmiedeberg's Archives of Pharmacology* **2014**, *387*, 1101-1116, <https://doi.org/10.1007/s00210-014-1034-9>.
22. Park, J.J.; Hwang, S.J.; Park, J.-H.; Lee, H.-J. Chlorogenic acid inhibits hypoxia-induced angiogenesis via down-regulation of the HIF-1 $\alpha$ /AKT pathway. *Cellular Oncology* **2015**, *38*, 111-118, <https://doi.org/10.1007/s13402-014-0216-2>.
23. Kim, J.; Jeong, I.-H.; Kim, C.-S.; Lee, Y.M.; Kim, J.M.; Kim, J.S. Chlorogenic acid inhibits the formation of advanced glycation end products and associated protein cross-linking. *Archives of Pharmacol Research* **2011**, *34*, 495-500, <https://doi.org/10.1007/s12272-011-0319-5>.
24. Holopainen, J.M.; Moilanen, J.A.O.; Sorsa, T.; Kivelä-Rajamäki, M.; Tervahartiala, T.; Vesaluoma, M.H.; Tervo, T.M.T. Activation of Matrix Metalloproteinase-8 by Membrane Type 1-MMP and Their Expression in Human Tears after Photorefractive Keratectomy. *Investigative Ophthalmology & Visual Science* **2003**, *44*, 2550-2556, <https://doi.org/10.1167/iovs.02-1190>.
25. Kim, J.K.; Park, S.U. Chlorogenic acid and its role in biological functions: an up to date. *EXCLI journal* **2019**, *18*, 310-316, <http://dx.doi.org/10.17179/excli2019-1404>.
26. Cho, H J.; Kang, H.J.; Kim, Y.J.; Lee, D.H.; Kwon, H.W.; Kim, Y.Y.; Park, H.J. Inhibition of platelet aggregation by chlorogenic acid via cAMP and cGMP-dependent manner. *Blood Coagul. Fibrinolysis* **2012**, *23*, 629–635. <https://doi.org/10.1097/MBC.0b013e3283570846>.
27. Chen, W.C.; Liou, S.S.; Tzeng, T.F.; Lee, S.L.; Liu, I.M. Effect of Topical Application of Chlorogenic Acid on Excision Wound Healing in Rats. *Planta Med* **2013**, *79*, 616-621.
28. De Camargo, A.C.; Lima, R.S. A perspective on phenolic compounds, their potential health benefits, and international regulations: The revised Brazilian normative on food supplements. *Journal of Food Bioactives* **2019**, *7*, <https://doi.org/10.31665/JFB.2019.7193>.
29. Ave, O.R. Exploring the Potential of Green Coffee Extract for Wound Healing Treatment. *IOP Conference Series: Earth and Environmental Science* **2019**, *391*, 012057. IOP Publishing.
30. Bagdas, D.; Etoz, B.C.; Gul, Z.; Ziyank, S.; Inan, S.; Turacozen, O.; Gul, N.Y.; Topal, A.; Cinkilic, N.; Tas, S.; Ozyigit, M.O.; Gurun, M.S. In vivo systemic chlorogenic acid therapy under diabetic conditions: Wound healing effects and cytotoxicity/genotoxicity profile. *Food and Chemical Toxicology* **2015**, *81*, 54-61, <https://doi.org/10.1016/j.fct.2015.04.001>
31. Lipinski, C.A. Lead- and drug-like compounds: the rule-of-five revolution. *Drug Discovery Today: Technologies* **2004**, *1*, 337-341, <https://doi.org/10.1016/j.ddtec.2004.11.007>.
32. Kataria, R.; Khatkar, A. In-silico design, synthesis, ADMET studies and biological evaluation of novel derivatives of Chlorogenic acid against Urease protein and H. Pylori bacterium. *BMC Chemistry* **2019**, *13*, <https://doi.org/10.1186/s13065-019-0556-0>.
33. Bare, Y.; Sari, D R.T.; Rachmad, Y.T.; Krisnamurti, G.C.; Elizabeth, A.; Maulidi, A. In Silico Insight the Prediction of Chlorogenic Acid in Coffee through Cyclooxygenase-2 (COX2) Interaction. *Biogenesis: Jurnal Ilmiah Biologi* **2019**, *7*, 100-105, <https://doi.org/10.24252/bio.v7i2.9847>.
34. Mohammadhassan, R., Fallahi, S., & Mohammadalipour, Z. ADMET and pharmaceutical activity analysis of caffeic acid diversities by in silico tools. *Letters in Applied NanoBioScience*. **2020**, *9*, 840 – 848. <https://doi.org/10.33263/LIANBS91.840848>
35. Dhanaraj, V.; Williams, M.G.; Ye, Q.; Pavlovsky, A.; Rubin, J.R.; Skeeane, R.W. X-ray Structure of Gelatinase A Catalytic Domain Complexed with a Hydroxamate Inhibitor. *Croatica chemica acta* **1999**, *72*, 575–591.
36. Hofmann, E.; Zerbe, P.; Schaller, F. The Crystal Structure of <math>\gamma</math>-Arabidopsis thaliana<math>\gamma</math>-Allene Oxide Cyclase: Insights into the Oxylipin Cyclization Reaction. *The Plant Cell* **2006**, *18*, 3201-3217, <https://doi.org/10.1105/tpc.106.043984>.
37. Koteswara Reddy, G.; Nagamalleswara Rao, K.; Yarrakula, K. Insights into structure and function of 30S Ribosomal Protein S2 (30S2) in Chlamydomonas reinhardtii: A potent target of pneumonia. *Computational Biology and Chemistry* **2017**, *66*, 11-20, <https://doi.org/10.1016/j.compbiolchem.2016.10.014>.



38. Navia, M.A.; McKeever, B.M.; Springer, J.P.; Lin, T.Y.; Williams, H.R.; Fluder, E.M.; Dorn, C.P.; Hoogsteen, K. Structure of human neutrophil elastase in complex with a peptide chloromethyl ketone inhibitor at 1.84-Å resolution. *Proceedings of the National Academy of Sciences* **1989**, *86*, 7–11, <https://doi.org/10.1073/pnas.86.1.7>.
39. Kumar, G.; Patnaik, R. Inhibition of Gelatinases (MMP-2 and MMP-9) by Withania somnifera Phytochemicals Confers Neuroprotection in Stroke: An In Silico Analysis. *Interdisciplinary Sciences: Computational Life Sciences* **2018**, *10*, 722-733, <https://doi.org/10.1007/s12539-017-0231-x>.
40. Liang, P.; Zhang, Y.Y.; Yang, P.; Grond, S.; Zhang, Y.; Qian, Z.-J. Viridicatol and viridicatin isolated from a shark-gill-derived fungus *Penicillium polonicum* AP2T1 as MMP-2 and MMP-9 inhibitors in HT1080 cells by MAPKs signaling pathway and docking studies. *Medicinal Chemistry Research* **2019**, *28*, 1039-1048, <https://doi.org/10.1007/s00044-019-02358-w>.
41. Yadav, E.; Singh, D.; Debnath, B.; Rathee, P.; Yadav, P.; Verma, A. Molecular Docking and Cognitive Impairment Attenuating Effect of Phenolic Compound Rich Fraction of *Trianthema portulacastrum* in Scopolamine Induced Alzheimer's Disease Like Condition. *Neurochemical Research* **2019**, *44*, 1665-1677, <https://doi.org/10.1007/s11064-019-02792-7>.
42. Naeem, S.; Hylands, P.; Barlow, D. Docking Studies of Chlorogenic Acid against Aldose Reductase by using Molgro Virtual Docker Software. *JAPS* **2013**, *3*, 13–20, <https://doi.org/10.7324/JAPS.2013.30104>.
43. Pathak, J.; Mondal, S.; Ahmed, H.; Rajneesh; Singh, S.P.; Sinha, R.P. In silico study on interaction between human polo-like kinase 1 and cyanobacterial sheath pigment scytonemin by molecular docking approach. *Biointerface Research in Applied Chemistry* **2019**, *9*, 4374-4378. <https://doi.org/10.33263/BRIAC95.374378>
44. Caley, M.P.; Martins, V.L.C.; O'Toole, E.A. Metalloproteinases and Wound Healing. *Adv Wound Care (New Rochelle)* **2015**, *4*, 225-234, <https://doi.org/10.1089/wound.2014.0581>.
45. Wlaschek, M.; Singh, K.; Sindrilaru, A.; Crisan, D.; Scharffetter-Kochanek, K. Iron and iron-dependent reactive oxygen species in the regulation of macrophages and fibroblasts in non-healing chronic wounds. *Free Radical Biology and Medicine* **2019**, *133*, 262-275, <https://doi.org/10.1016/j.freeradbiomed.2018.09.036>.
46. Zhou, L.; Ren, M.; Zeng, T.; Wang, W.; Wang, X.; Hu, M.; Su, S.; Sun, K.; Wang, C.; Liu, J.; Yang, C.; Yan, L. TET2-interacting long noncoding RNA promotes active DNA demethylation of the MMP-9 promoter in diabetic wound healing. *Cell Death & Disease* **2019**, *10*, <https://doi.org/10.1038/s41419-019-2047-6>.
47. Bickerton, G.R.; Paolini, G.V.; Besnard, J.; Muresan, S.; Hopkins, A.L. Quantifying the chemical beauty of drugs. *Nature Chemistry* **2012**, *4*, 90-98, <https://doi.org/10.1038/nchem.1243>.
48. Rodrigues, M.; Kosaric, N.; Bonham, C.A.; Gurtner, G.C. Wound Healing: A Cellular Perspective. *Physiological Reviews* **2018**, *99*, 665-706, <https://doi.org/10.1152/physrev.00067.2017>.
49. Kähäri, V.M.; Saarialho-Kere, U. Matrix metalloproteinases and their inhibitors in tumour growth and invasion. *Ann Med*. **1999**, *31*, 34-45, <https://doi.org/10.3109/07853899909019260>.
50. Danielsen, P.L.; Holst, A.V.; Maltesen, H.R.; Bassi, M.R.; Holst, P.J.; Heinemeier, K.M.; Olsen, J.; Danielsen, C.C.; Poulsen, S.S.; Jorgensen, L.N.; Ågren, M.S. Matrix metalloproteinase-8 overexpression prevents proper tissue repair. *Surgery* **2011**, *150*, 897-906, <https://doi.org/10.1016/j.surg.2011.06.016>.
51. Moghadam, E.S.; Ebrahimi, N.S.; Salehi, P.; Moridi Farimani, M.; Hamburger, M.; Jabbarzadeh, E. Wound Healing Potential of Chlorogenic Acid and Myricetin-3-O-β-Rhamnoside Isolated from *Parrotia persica*. *Molecules* **2017**, *22*, 1–15, <https://doi.org/10.3390/molecules22091501>.
52. Chan, M.F.; Li, J.; Bertrand, A.; Casbon, A.-J.; Lin, J.H.; Maltseva, I.; Werb, Z. Protective effects of matrix metalloproteinase-12 following corneal injury. *Journal of Cell Science* **2013**, *126*, 948–3960, <https://doi.org/10.1242/jcs.128033>.
53. Liang, N.; Kitts, D.D. Role of Chlorogenic Acids in Controlling Oxidative and Inflammatory Stress Conditions. *Nutrients* **2016**, *8*, 1–20, <https://doi.org/10.3390/nu8010016>.