cosmetics

MDPI

Article

Skin Anti-Aging Potentials of Phytochemicals from *Peperomia pellucida* against Selected Metalloproteinase Targets: An In Silico Approach

Babatunji Emmanuel Oyinloye [1,2,3,*], Emmanuel Ayodeji Agbebi [3,4], Oluwaseun Emmanuel Agboola [3], Chukwudi Sunday Ubah [5], Olutunmise Victoria Owolabi [6], Raphael Taiwo Aruleba [7], Sunday Amos Onikanni [8], Jerius Nkwuda Ejeje [9], Basiru Olaitan Ajiboye [10] and Olaposi Idowu Omotuyi [3,11]

1 Phytomedicine, Biochemical Toxicology and Biotechnology Research Laboratories, Department of Biochemistry, College of Sciences, Afe Babalola University, PMB 5454, Ado-Ekiti 360001, Nigeria
2 Biotechnology and Structural Biology (BSB) Group, Department of Biochemistry and Microbiology, University of Zululand, KwaDlangezwa 3886, South Africa
3 Institute of Drug Research and Development, S.E Bogoro Center, Afe Babalola University, PMB 5454, Ado-Ekiti 360001, Nigeria
4 Department of Pharmacognosy and Natural Products, College of Pharmacy, Afe Babalola University, Ado-Ekiti 360001, Nigeria
5 Department of Epidemiology and Biostatistics, College of Public Health, Temple University, Philadelphia, PA 19121, USA
6 Medical Biochemistry Unit, College of Medicine and Health Sciences, Afe Babalola University, PMB 5454, Ado-Ekiti 360001, Nigeria
7 Department of Physiology, East Carolina University, Greenville, NC 27834, USA
8 Graduate Institute of Biomedical Sciences, College of Medicine, China Medical University, Taichung 40402, Taiwan
9 Department of Chemistry/Biochemistry/Molecular Biology, Alex Ekwueme Federal University Ndufu-Alike, P.O. Box 1010, Abakaliki 482131, Nigeria
10 Phytomedicine and Molecular Toxicology Research Laboratory, Department of Biochemistry, Federal University Oye-Ekiti, Oye-Ekiti 371104, Ekiti State, Nigeria
11 Department of Pharmacology and Toxicology, College of Pharmacy, Afe Babalola University, PMB 5454, Ado-Ekiti 360001, Nigeria
* Correspondence: babatunjioe@abuad.edu.ng

Abstract: Skin aging and wrinkle formation are processes that are largely influenced by the overexpression of enzymes like tyrosinase, elastase, and collagenase. This study aimed to validate the skin anti-aging properties of phytochemicals from *Peperomia pellucida* (PP) as well as its attendant mechanism of action. Compounds previously characterized from PP were retrieved from the PubChem database and docked to the active sites of tyrosinase, elastase, and collagenase using Schrödinger's Maestro 11.5 and AutoDock tools to predict compounds with the best inhibitory potential to block these enzymes in preventing skin aging. It was observed that our hit compounds had favorable affinity and displayed key interactions at the active sites of these enzymes similar to those of the standards. With elastase, we observed key interactions with the amino acids in the S1 sub-pocket (especially ALA-181), Zn chelation, and histidine residues, which are key for inhibitory activity and ligand stability. The hit compounds showed H-bonds with the key amino acids of collagenase, including LEU-185 and ALA-186; phlobaphene and patuloside B were found to have better docking scores and inhibition constants (Ki) (−12.36 Kcal/mol, 0.87 nM and −12.06 Kcal/mol, 1.45 nM, respectively) when compared with those of the synthetic reference compound (−12.00 Kcal/mol, 1.67 nM). For tyrosinase, our hit compounds had both better docking scores and Ki values than kojic acid, with patuloside B and procyanidin having the best values of −9.43 Kcal/mol, 121.40 nM and −9.32 Kcal/mol, 193.48 nM, respectively (kojic acid = −8.19 Kcal/mol, 898.03 nM). Based on this study, we propose that acacetin, procyanidin, phlobaphene, patulosides A and B, palmitic acid, and hexahydroxydiphenic acid are responsible for the anti-aging effects of PP on the skin, and that they work synergistically through a multi-target inhibition of these enzymes.

Citation: Oyinloye, B.E.; Agbebi, E.A.; Agboola, O.E.; Ubah, C.S.; Owolabi, O.V.; Aruleba, R.T.; Onikanni, S.A.; Ejeje, J.N.; Ajiboye, B.O.; Omotuyi, O.I. Skin Anti-Aging Potentials of Phytochemicals from *Peperomia pellucida* against Selected Metalloproteinase Targets: An In Silico Approach. *Cosmetics* **2023**, *10*, 151. https://doi.org/10.3390/cosmetics10060151

Academic Editor: Perry Xiao

Received: 14 August 2023
Revised: 14 September 2023
Accepted: 19 September 2023
Published: 2 November 2023

Keywords: skin aging; metalloproteinases; kojic acid; *Peperomia pellucida*; procyanidin

1. Introduction

Life expectancy is steadily increasing, but the mystery of aging remains partially unsolved. Such mysteries are equally prevalent in mental and physical disability as well as in diseases associated with aging. One extrinsically notable aspect of aging is the changes observed on the skin, thus making the skin an excellent model organ for studying the aging process [1]. Skin is a multi-layered structure made up of an underlying matrix called the dermis and a functional epidermis, which has four closely adherent layers at the outer surface [2,3]. Aging has similar effects on both the skin and internal organs, causing irreversible degeneration [2,4]. With visible changes in the structure and function of the integument, the skin is the most visible indicator of the aging process. Human organs, including the skin (our body's outermost protective cover), undergo age-related changes. Because the skin is in direct contact with the external environment, it is one of the organs most vulnerable to environmental damage, which contributes significantly to the aging process [5,6].

The epidermis is densely packed with keratinocytes, which produce structural keratin protein and stratify toward the outer surface, forming an external barrier. The dermis, on the other hand, is densely packed with fibroblasts that secrete a variety of structural components with significant reparative and wound-healing properties. Furthermore, this layer contributes to the regulation of extracellular matrices as well as interstitial fluid volume [7].

Extrinsic and intrinsic skin aging (photoaging) also occurs. The former is a natural process that is brought on mainly by the metabolic by-products of skin cells, among which reactive oxygen species (ROS) are the most harmful. However, a wide range of external environmental elements play important role in mediating extrinsic aging. By facilitating the breakage and aberrant chain crossing of the fibrous proteins elastin, collagen, and glycosaminoglycan hyaluronic acid in the skin's extracellular matrix, excessive ROS generation contributes to the development of wrinkles [1,8,9]. Thus, aging generally occurs as a result of a disturbance in the balance between the regenerative and degenerative potential of the skin, leading to wrinkle formation and epidermis thinning [10,11].

Many metalloproteinases, a superfamily of protease enzymes whose catalytic action requires a metal, are responsible for degrading fibrous proteins. Collagenases are proteins which catalyze chemical processes and break the peptide bonds in collagen, while elastase is responsible for the degradation of the extracellular matrix component, elastin. Both excessive melanin formation and hyperpigmentation, which is regulated by the enzyme tyrosinase, also contribute significantly to skin aging [12]. Chemical inhibitors that target these enzymes offer a viable method of delaying and combating skin aging, particularly if they have significant antioxidant activity [11]. Thus, inhibiting these enzymes may provide promisingly youthful skin that defies aging [13].

Many efficient anti-aging products are derived from natural sources; these products have proven effective in restoring the imbalance between the regenerative/degenerative power of the skin via diverse mechanisms, including moisturizing effects and the potentiation of elastin and collagen production [10]. Owing to their abundant reservoir of phytochemicals, the use of medicinal plants in disease management as well as the source of major compounds of drugs has been a widespread practice. Specifically, a good number of drugs in the market today are obtained from natural sources, either directly (e.g., morphine, digoxin) or indirectly, as a base for the development of semisynthetic chemical compounds (e.g., aspirin from salicin, etc.) [14,15]. In this age of globalization and technology, consumer preferences are unexpectedly changing from synthetic chemicals to natural/herbal products. This trend is being driven by increased concerns about the undesirable side effects of chemical products. Several natural-based compounds have been approved for use by local

and international medical authorities in the management of various ailments, including skin disorders. For example, procyanidin B and kojic acid (a fungal metabolite commonly produced by many species of *Aspergillus*, *Acetobacter*, and *Penicillium*) have been used in many skin care products and soaps [16].

Peperomia pellucida (L.) Kunth (Scheme 1) is an annual herbaceous plant with terminal spike inflorescences opposite to its alternating oval, succulent leaves [17,18].

Scheme 1. *Peperomia pellucida* (L.) Kunth whole plant in its natural habitat (photographed by Emmanuel Agbebi).

It belongs to the family Piperaceae and is mainly distributed through the Neotropics, Africa, southeast Asia, and Australian regions. The plant thrives in areas with low sun exposure and moist, loose soils, and it grows better during rainy seasons [19,20]. It has short roots and fascicules that grow typically between 15 and 45 cm tall [21]. It has a glabrous, translucent green, erect, and succulent stem [22]. Many of its pharmacological activities are well documented. It is frequently used to treat a wide range of medical disorders, including skin sores, measles [23], and hypertension [24]. More specifically, locals utilize the plant's leaves to cure mental issues and topically to address skin problems, including acne, boils, and wounds [19]. It was also reported to possess high antioxidant activities, even at low concentrations, indicating that it could be effective for the treatment of diseases resulting from oxidative stress [25,26].

Generally, *Peperomia* has been used locally for topical application to address skin problems, including eczema, acne, and other skin diseases [18]. Thus, this study aims to

validate its skin anti-aging properties and the attendant mechanism of action as a viable topical agent.

2. Methods

2.1. Virtual Screening and Docking Platform

Compounds that have been previously characterized from *Peperomia pellucida* were collected from an online database and docked to the active sites of the selected targets to predict compounds with the best inhibitory potential to block these enzymes in preventing skin aging. The Schrödinger Suite software, Maestro 11.5, and AutoDock tools were used for the docking study, using the standard molecular docking principle [27,28].

2.2. Ligand Library Generation and Preparation

The two-dimensional (2D) structures of phytochemicals from the *Peperomia pellucida* plant in SDF format were retrieved from the PubChem online database https://pubchem.ncbi.nlm.nih.gov/ (accessed on 13 February 2023). The ligands were retrieved from research reviews of the plant [19]. The 2D structures were transformed into 3D structures using the ligprep tool by adding hydrogen atoms, ionizing at pH (7.2 $\pm$ 0.2), and removing salt using Ep2i/UNEP/-Zk. The OPLS3e force field was utilized for ionization and tautomeric state formation.

2.3. Target Retrieval and Preparation

The X-ray crystal structure of the selected targets, collagenase (PDB ID: 2D1N) [29], elastase (PDB ID: 1RMZ) [30], and tyrosinase (PDB ID: 5M8M) [31], with their corresponding bound ligands, were retrieved from the Protein Data Bank https://www.rcsb.org (accessed on 13 February 2023). The PyMOL Molecular Graphics System, Version 2.5 Schrödinger, LLC, was used for the visualization of the proteins. The proteins were prepared using the protein preparation wizard tool in Maestro's Schrödinger Suite. Following standard protocols, bond orders were assigned, hydrogens were added, zero-order metal bonds were made, disulfide bonds were created, water molecules were removed, and HET states were generated using Epik at pH 7.0 $\pm$ 0.2 during the protein preparation process. The protein refinement was completed by optimizing the H-bond assignment, and then, the protein was reduced using the OPLS3e (optimized potentials for liquid simulation) force field.

2.4. Receptor Grid Generation

The Receptor Grid Generation tool was used to create the prepared protein grid on the binding site (Glide Grid). The receptor grid depicts the area where the ligand and protein interact. The coordinates of the co-crystallized ligand were used to specify and generate the receptor grid/active site for docking. By selecting the co-crystallized ligand at the active site of the receptor, the binding location was automatically mapped (by a cubic grid box) covering all of the amino acid residues at the active site.

2.5. Molecular Docking

Docking was performed on Maestro 11.5 with the Glide tool using the Extra Precision (XP) docking techniques. The AutoDock tool was then used for molecular docking of the hit compounds to determine the predicted inhibition constant (Ki). The co-crystallized ligands were extracted and re-docked into the active site to validate the molecular docking study.

2.6. The Molecular Mechanics/Generalized Born Surface Area (MM/GBSA)

The Molecular Mechanics/Generalized Born Surface Area (MM/GBSA) continuum solvent model in the Schrödinger Suite's prime module was used to determine the binding free energy of the docked protein–ligand complex [32].

3. Results

As shown below, Figure 1 represents the Glide scores of all the phytochemicals recovered from the literature that have been isolated from *Peperomia* against elastase, collagenase, and tyrosinase.

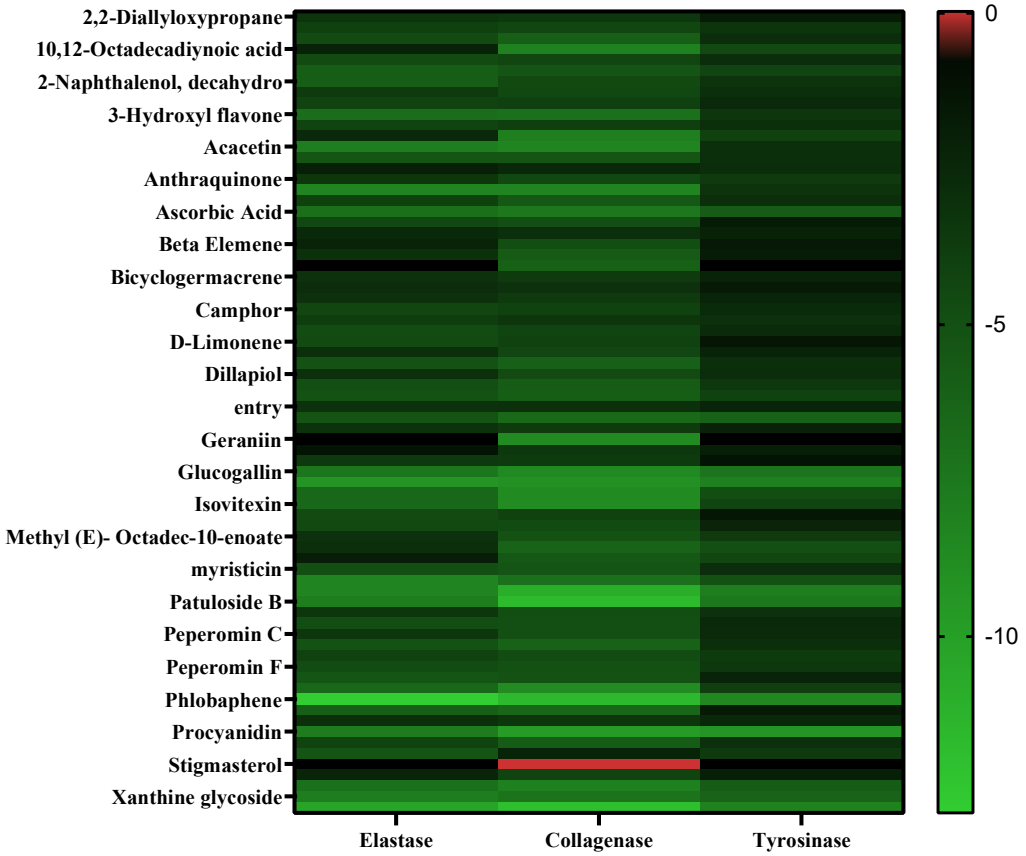

Figure 1. A heatmap, showing the Glide score of phytochemicals from *Peperomia pellucida* against Elastase, Collagenase, and Tyrosinase.

From this result, it was observed that acacetin, procyanidin, phlobaphene, patuloside A and B, palmitic acid, and hexahydroxydiphenic acid have good Glide scores and were able to inhibit at least two of the three receptors used at a comparable level to the synthetic inhibitors. Therefore, they were selected as our hit compounds, for further study using AutoDock, to determine their inhibition constants against these targets (as shown in Table 1).

The molecular mechanics generalized born surface area (MM-GBSA) is a computational thermodynamics method of determining the binding affinity of compounds. Schrödinger suite's Prime module MM-GBSA has previously been found to provide an accurate statistical post-docking analysis of docked complexes. The lower the score, the higher/better the binding and stability of the complex. The relative free binding energies of the compounds and that of the co-crystallized ligands are shown in Figure 2. The MM-GBSA results show that some of the hit compounds have similar binding energy with reference molecules. For the elastase, HHDP has a similar binding energy with the co-crystalized ligand/synthetic inhibitor (−41.83 vs. −41.75, respectively). Acacetin, procyanidin, and patuloside B also have scores of −38.59, −35.68, and −34.71, respectively. For collagenase, the synthetic inhibitor has a better binding energy (−54.66) than all the hit compounds. However, most of the compounds have a good score (≤ -30) except patuloside B (−15.96), with patuloside A and phlobaphene having scores of −43.97 and −48.41, respectively.

Table 1. Docking scores (Kcal/mol), inhibition constant, Ki, (nM), and interacting residues of each target with their co-crystallized ligands and the top 6 hit compounds.

Compound	Elastase (1RMZ)			Collagenase (2D1N)			Tyrosinase (5M8M)		
	Score	**Ki (nM)**	**Key Interactions**	**Score**	**Ki (nM)**	**Key Interactions**	**Score**	**Ki (nM)**	**Key Interactions**
Co-crystallized ligands (CCL)/synthetic inhibitors	−10.32	24.48	Zn chelation–Zn264 H-bond–LEU181, ALA 182	−12.00	1.67	Zn chelation–Zn270 H-bond–ALA186, PRO236, ALA238, ILE243. π-cat–TYR176, HIS226	−8.19	898.03	Zn chelation–Zn512 H-bond–SER394 π–π–HIS381
Hexahydroxydiphenic acid	−9.33	137.99	Zn chelation–Zn264 H-bond–GLY179, ALA 182, TYR240	−8.07	1210	H-bond–GLY183, LEU185, ALA 186, TYR244. π-π–HIS232	−8.03	1258.0	H-bond–TYR362, ARG374, ASN378, THR391, SER394
Patuloside A	−12.91	0.35	Zn chelation–Zn264 H-bond–THR215, GLU219, VAL235. π–π–HIS228	−11.03	8.29	H-bond–LEU185, GLU223, PHE241, ILE243, TYR244.	−8.47	614.69	H-bond–TYR362, ASN378, THR391, SER394
Patuloside B	−12.01	1.56	Zn chelation–Zn264 H-bond–ALA 182, GLU219, TYR240. π–π–HIS228	−12.06	1.45	H-bond–LEU185, ALA 186, GLU223, ALA238, TYR244. π–π–HIS222	−9.43	121.40	H-bond–GLU216, ASN318, ARG321, ASN378, GLY389.
Procyanidin	−9.57	96.11	Zn chelation–Zn264 H-bond–LEU181, ALA 182, GLU219, TYR240. π–π–HIS228	−10.96	9.33	Zn chelation–Zn270 H-bond–GLU223, PRO242. π–π–HIS232	−9.32	193.48	H-bond–VAL196, ASP212, GLU216, ARG321, ARG374, GLY389. π–π–TYR362
Phlobaphene	−12.83	0.39	Zn chelation–Zn264 H-bond–LEU214. π–π–HIS228	−12.36	0.87	H-bond–GLY183, LEU185, ALA 186, ALA188, ASP231, PRO242, TYR244. π–π–TYR176	−8.51	582.93	H-bond–GLU216, GLY389, THR391, SER394 π–π–HIS215
Acacetin	−9.60	91.86	Zn chelation–Zn264 H-bond–LEU181, ALA 182. π–π–HIS218	−9.40	129.05	H-bond–ALA238. π–π–HIS222	−7.34	4180	H-bond–ARG321, 374

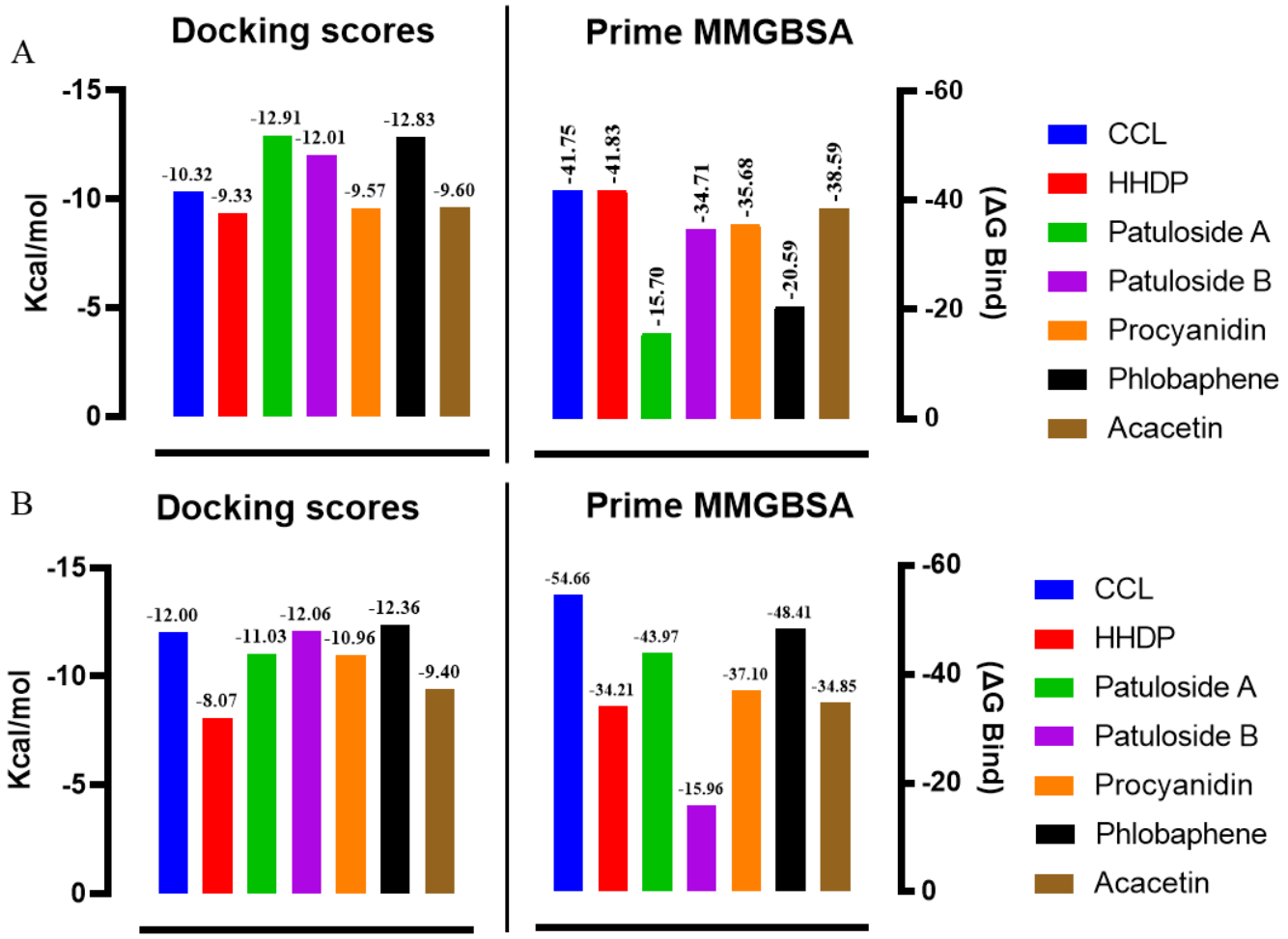

Figure 2. Docking and Prime MM-GBSA scores of our hit compounds and CCL with (**A**) Elastase and (**B**) Collagenase.

Figures 3–5 show the 2D interaction of the amino acids at the active sites of elastase, collagenase, and tyrosinase with the hit compounds and the co-crystallized ligands/synthetic inhibitors. Our hit compounds were observed to display key interactions, including metal chelation, and interaction with key amino acids at the catalytic site of the enzymes, for example, a hydrogen bond with Ala-181 and Ala-182 for elastase, and Leu-185 and Ala-186 for collagenase. They also have inhibition constants in the nanomolar range (similar to the synthetic inhibitors), as shown in Table 1.

Figures 6–8 show the 3D surface interaction of our hit compounds and the co-crystallized ligands with these enzymes. Similar to the co-crystallized ligands/synthetic inhibitors, our hit compounds also displayed their ability to penetrate deep into active site pockets and interact with residues. For example, for collagenase, one of the remarkable features of the co-crystallized synthetic inhibitor, FA4, was the deep penetration of its long aliphatic chain into the S1 pocket. This was also demonstrated by some of our compounds (acacetin, and patulosides A and B), as shown in Figure 7.

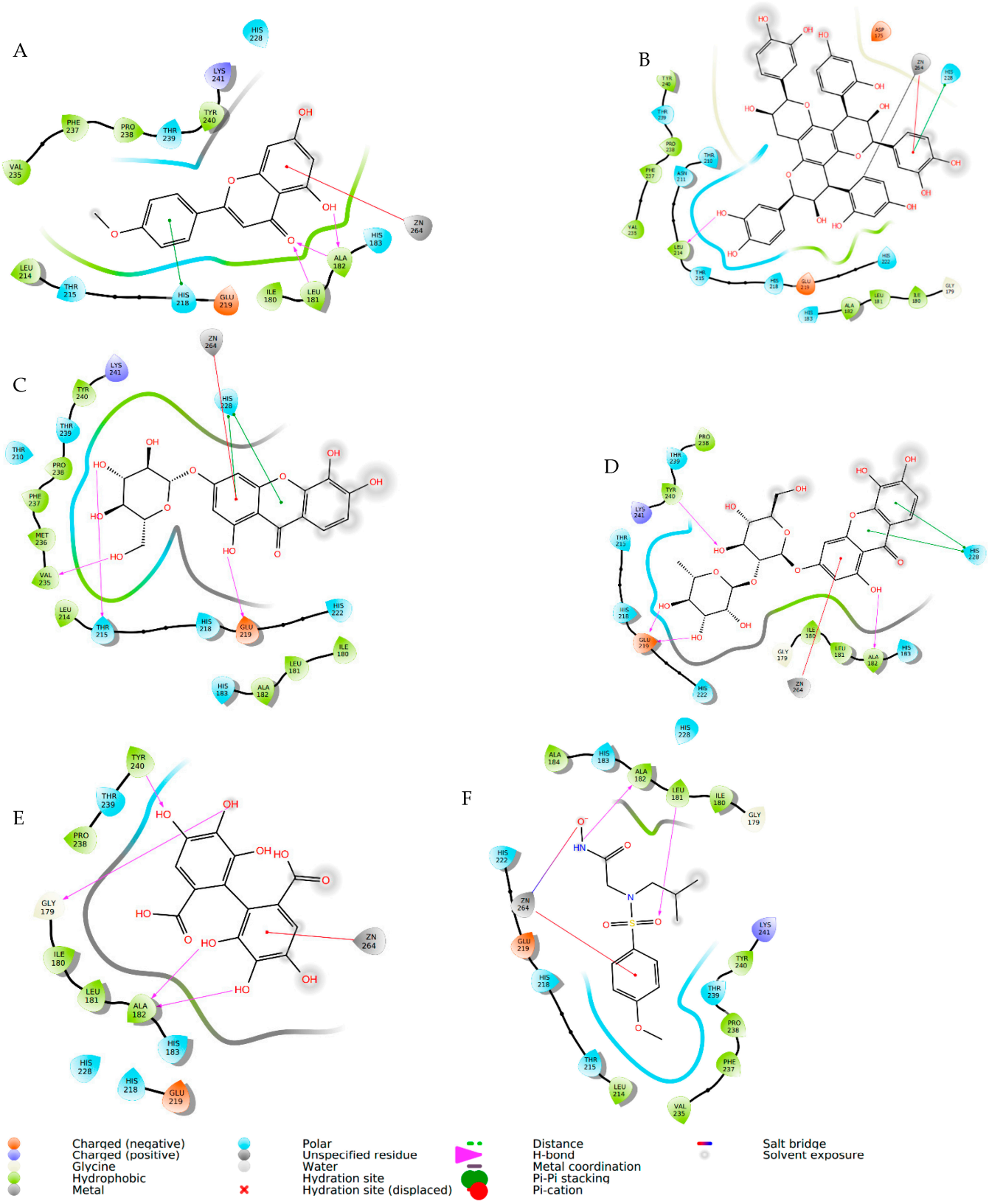

Figure 3. Two-dimensional (2D) molecular interactions of amino-acid residues of human elastase (1RMZ) with hit compounds from *Peperomia pellucida*. (**A**) Acacetin, (**B**) Phlobaphene, (**C**) Patuloside A, (**D**) Patuloside B, (**E**) Hexahydroxydiphenic acid, and (**F**) Standard (NGH).

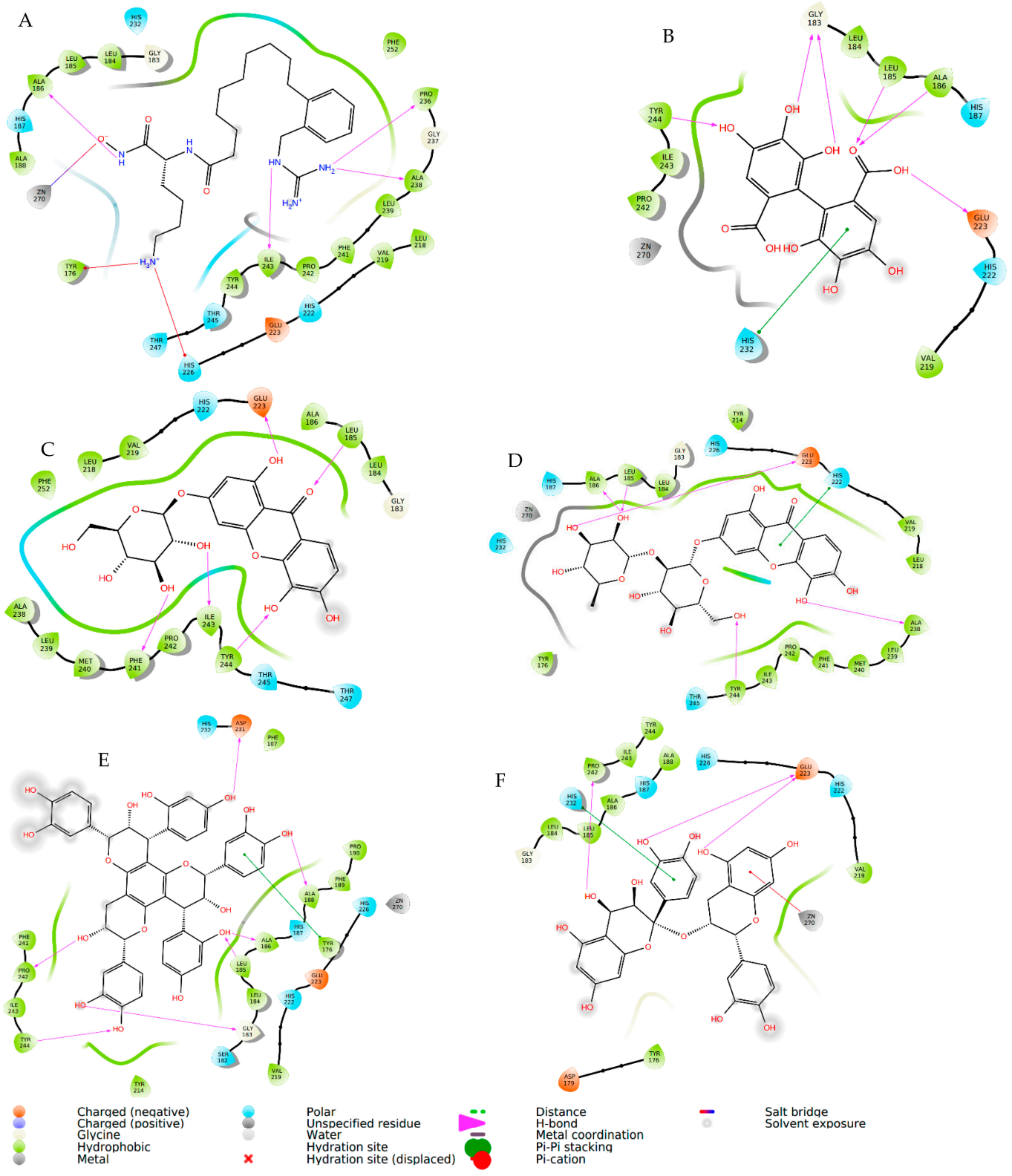

Figure 4. Two-dimensional (2D) molecular interactions of amino acid residues of human collagenase (2D1N) with hit compounds from *Peperomia pellucida*. (**A**) Standard (FA4), (**B**) Hexahydroxydiphenic acid, (**C**) Patuloside A, (**D**) Patuloside B, (**E**) Phlobaphene, and (**F**) Procyanidin.

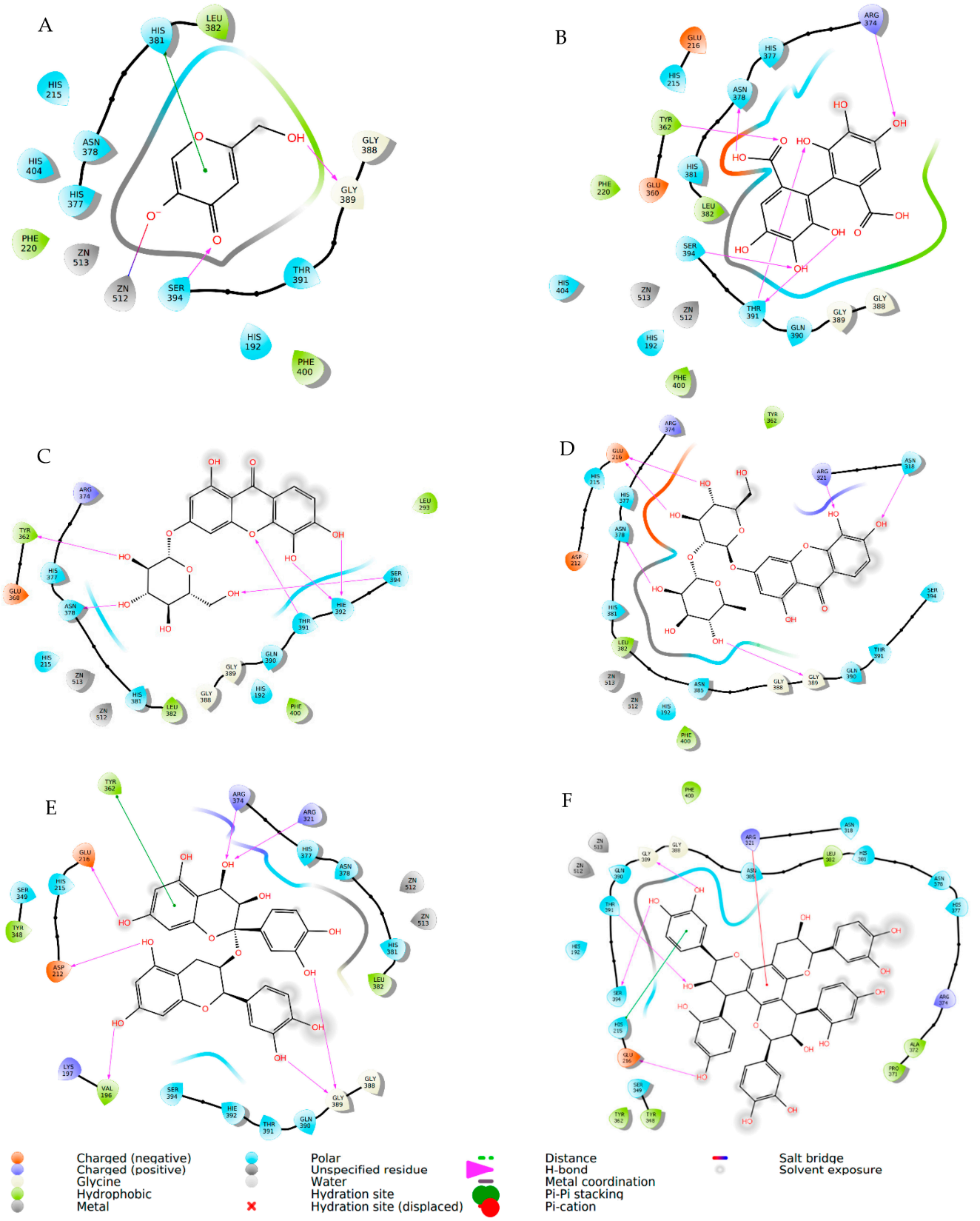

Figure 5. Two-dimensional (2D) molecular interactions of amino-acid residues of human collagenase (2D1N) with hit compounds from *Peperomia pellucida*. (**A**) Kojic Acid, (**B**) Hexahydroxydiphenic acid, (**C**) Patuloside A, (**D**) Patuloside B, (**E**) Procyanidin, and (**F**) Phlobaphene.

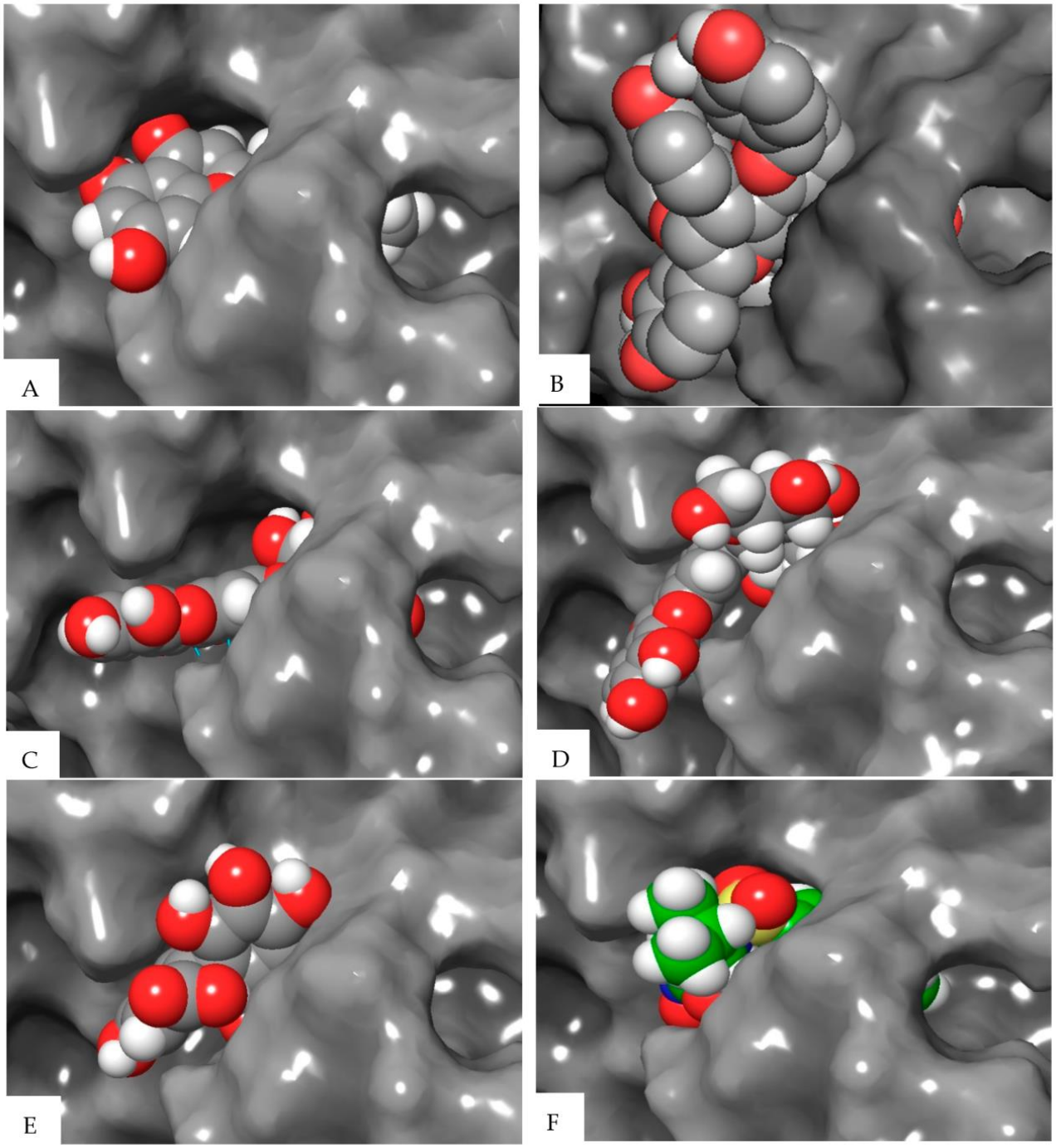

Figure 6. Three-dimensional (3D) surface interactions of human elastase (1RMZ) with hit compounds from *Peperomia pellucida*. (**A**) Acacetin, (**B**) Phlobaphene, (**C**) Patuloside A, (**D**) Patuloside B, (**E**) Hexahydroxydiphenic acid, and (**F**) Standard (NGH).

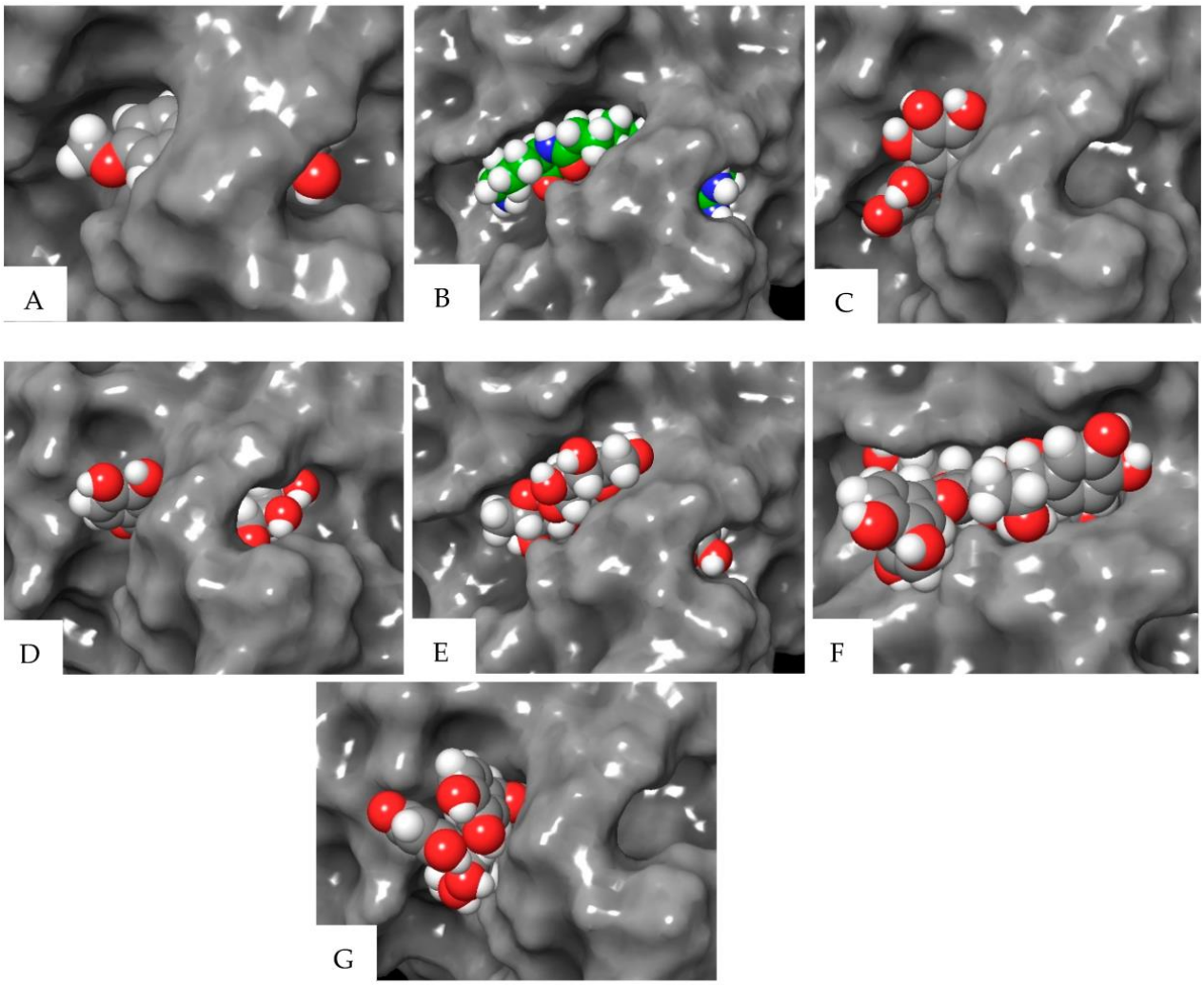

Figure 7. Two-dimensional (3D) surface interactions of human collagenase (2D1N) with hit compounds from *Peperomia pellucida*. (**A**) Standard (FA4), (**B**) Acacetin, (**C**) Hexahydroxydiphenic acid, (**D**) Patuloside A, (**E**) Patuloside B, (**F**) Phlobaphene, (**G**) Procyanidin.

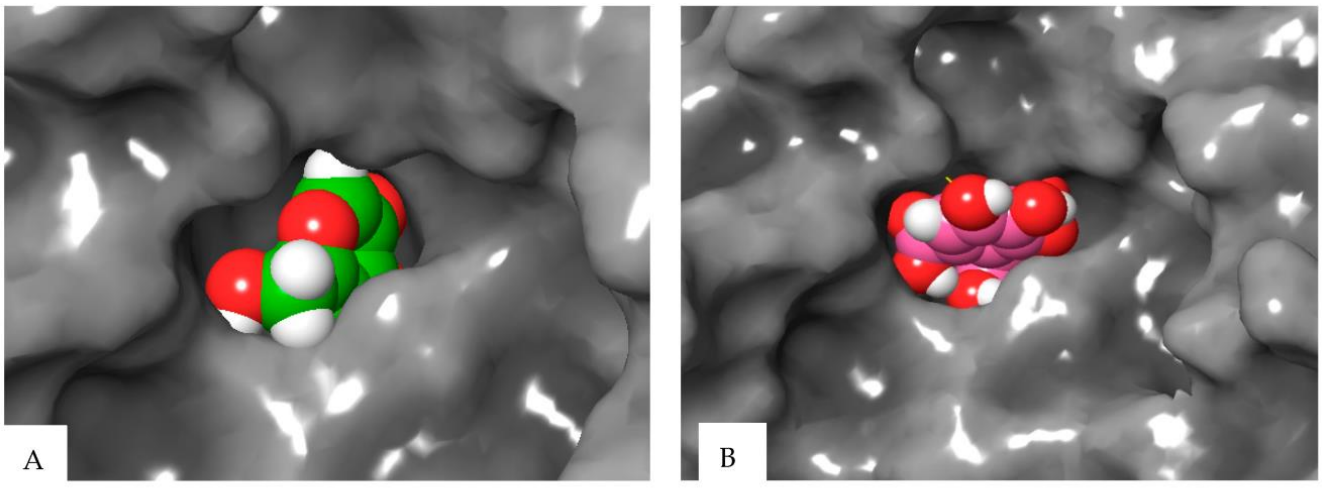

Figure 8. *Cont*.

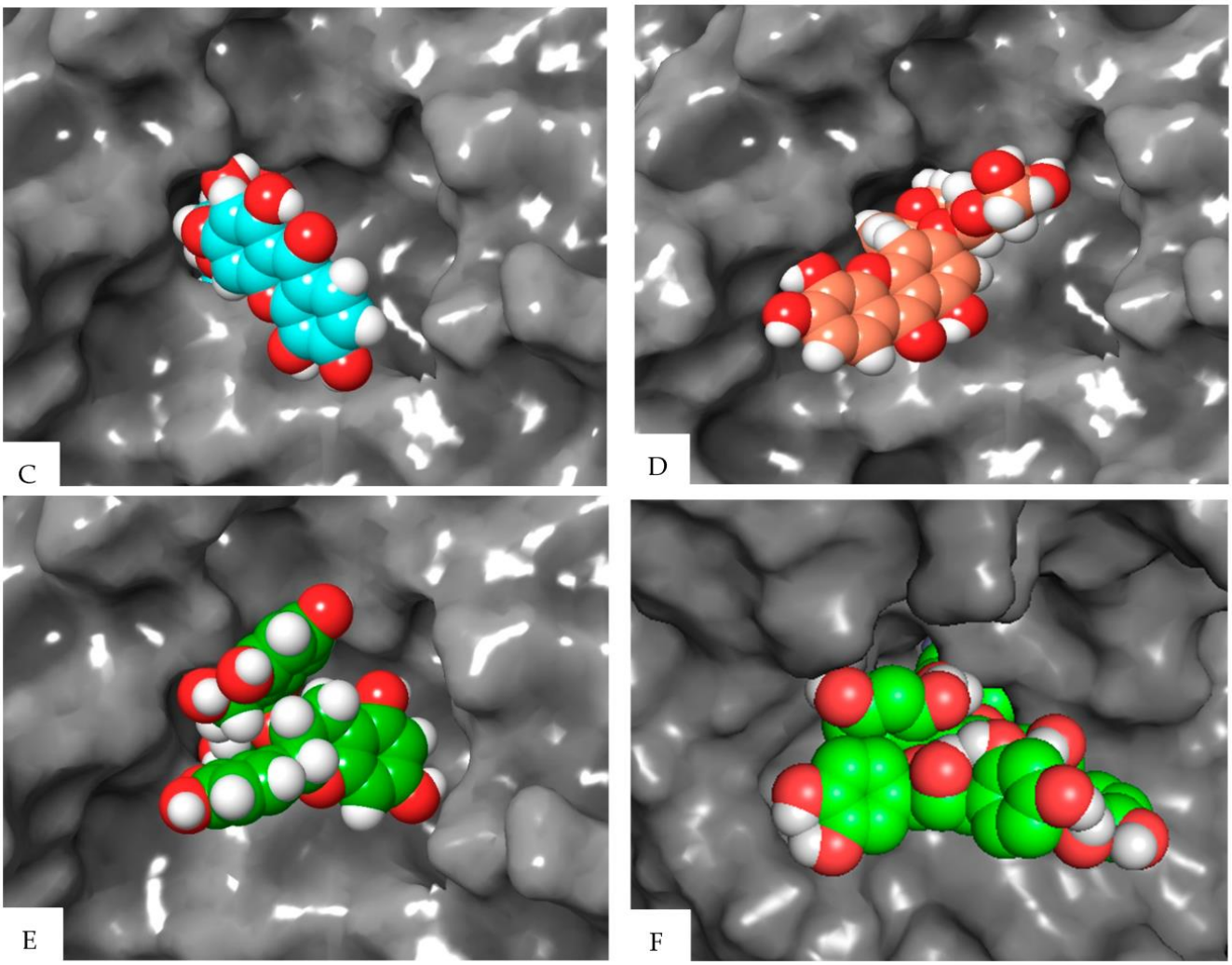

Figure 8. Three-dimensional (3D) surface interactions of human tyrosinase (5M8M) with hit compounds from *Peperomia pellucida*. (**A**) Standard (Kojic acid), (**B**) Hexahydroxydiphenic acid, (**C**) Patuloside A, (**D**) Patuloside B, (**E**) Procyanidin, and (**F**) Phlobaphene.

4. Discussion

Skin aging and wrinkle formation are processes that are largely influenced by enzymes like elastase and collagenase. These enzymes are responsible for the degradation of the extracellular matrix components, elastin and collagen, which are responsible for skin elasticity, flexibility, and strength. The overexpression of these enzymes will lead to matrix proteolytic degradation, skin aging, and wrinkle formation [29,33,34]. Therefore, blocking these enzymes will help to preserve the skin's integrity, elasticity, and strength, preventing skin aging and wrinkle formation.

The surface interaction, docking score, and inhibition constants revealed that our compounds, especially the selected hit compounds, fit properly into the active sites and have lower binding energies compared with the co-crystallized synthetic inhibitors (Table 1, Figures 2 and 6–8). For elastase, it has been reported that the interaction of the inhibitor with Zn ion and the S1 subsite of the active site is key for activity, as observed with the synthetic inhibitor [30,35]. Our hit compounds show similar interaction at the active site, including the metal chelation and Ala-182 H-bond, which is important for its inhibitory activity. Additionally, the histidine moieties (His-218, His-222, and His-228) have been reported to contribute to the stability of ligands at the receptor site [30]. Acacetin, phlobaphene, and patulosides A and B were also observed to exhibit a pie–pie interaction with His-218 and His-228 (Table 1 and Figures 3–5).

In addition to these, our study also shows that palmitic acid, a known emollient used in the cosmetic industry as a cleansing agent and moisturizer, was observed to bind with good affinity and fitting in the S1 subsite, affording it maximum interaction with key amino acids, including Ala-181 and Ala-182.

For the collagenase, one of the remarkable features of the co-crystallized synthetic inhibitor, FA4, was the deep penetration of its long aliphatic chain into the S1 pocket. This was also demonstrated by some of our compounds (acacetin, and patulosides A and B) as shown in Figure 7. Most of our compounds (except procyanidin) did not exhibit metal chelation; however, they all show interactions with key amino acids at the active site, including an H-bond with Leu-185 and Ala-186, pie interaction with His-222 and His-232, and hydrophobic interaction with Leu-239 and Phe-241. Phlobaphene and patuloside B were found to have the best binding score and inhibition constant (Ki) values of −12.36 Kcal/mol, 0.87 nM, and −12.06 Kcal/mol, 1.45 nM, respectively, in comparison with the synthetic reference compound value of −12.00 Kcal/mol, 1.67 nM.

Hyperpigmentation, which occurs as a result of the overproduction of melanin, contributes to skin aging. It is caused by various internal and external factors, including hormonal imbalances and exposure to UV radiation and chemicals. Skin pigmentation is controlled by melanin, and melanin synthesis is under the control of tyrosinase. Thus, inhibiting the tyrosinase enzyme can improve overall skin appearance, prevent hyperpigmentation, and retard the aging process [35,36]. Kojic acid, a known compound for the management of hyperpigmentation, acts by inhibiting the tyrosinase enzyme [37].

From our study, our hit compounds have better (lower) docking scores and Ki than Kojic acid, with patuloside B and procyanidin having the best binding score and Ki values of −9.43 Kcal/mol, 121.40 nM and −9.32 Kcal/mol, 193.48 nM, respectively, compared to Kojic acid with −8.19 Kcal/mol, 898.03 nM (Table 1).

The molecular mechanics generalized born surface area (MM-GBSA) is a computational thermodynamics method of determining the binding affinity of compounds. Schrödinger suite's Prime module MM-GBSA has previously been found to provide an accurate statistical post-docking analysis of docked complexes. It provides information on the binding affinity and stability of the ligand–receptor complex. The lower the score (i.e., the more negative it is), the higher/better the binding and stability of the complex [32]. The relative free binding energies of the compounds and that of the co-crystallized ligands, as shown in Figure 2, revealed that some of the hit compounds have a similar binding energy with reference molecules. For the elastase, HHDP has a similar binding energy with the co-crystalized ligand/synthetic inhibitor (−41.83 vs. −41.75, respectively). This shows that the stability of the HHDP–elastase complex is similar/comparable to that of the NGH–elastase complex.

Generally, our hit compounds, which were selected based on the ability to inhibit at least two of the three receptors used at a comparable level to the synthetic inhibitors, displayed a multi-target inhibition of these enzymes that play key roles in the skin aging process. Procyanidin, phlobaphene, and hexahydroxydiphenic acid belong to the flavonoid (tannin) class of compounds. They have been known to possess antioxidant activities, which contributes to their efficacy, considering that reactive oxygen species (ROS) play a key role in skin aging and wrinkle formation [8,38,39]. Procyanidin has been reported to possess antioxidant properties and promote skin moisturization and elasticity [40–42]. Patulosides A and B belong to the xanthone group of compounds. Xanthones (e.g., Mangostins) have been reported in nature to have good antioxidant properties, protect against free radicals and UV radiation, regulate melanin synthesis, and improve skin elasticity [39,43]. Acacetin has been reported/patented to reduce the biosynthesis of MMP-1 by ultraviolet rays and promote the biosynthesis of type 1 procollagen (Patent number: KR100851489B1). In addition, palmitic acid, a known emollient used in the cosmetic industry as a cleansing agent and moisturizer, is also present in the studied plant and they fit properly and bind with good affinity and low docking score to the target receptors. These reported activities are in agreement with our findings in this study. In addition to these, *Peperomia* has been reported to have good

anti-bacterial activity against acne-causing bacteria, *Propionibacterium acnes* [44]. Thus, the presence of these compounds in *Peperomia pellucida*, their reported use in the cosmetic industry, their antioxidant properties and antibacterial activity against *Propionibacterium acnes*, coupled with our report on their affinity, binding energy, Ki, and conformation at the active sites of these key enzymes implicated in skin aging and wrinkle formation, indicate that they will be an invaluable natural material in the cosmetics industry for the production of anti-aging skin care products and general maintenance of skin integrity.

5. Conclusions

It can be proposed from this study that phytochemicals from *Pepperomia pellucida*, especially acacetin, procyanidin, phlobaphene, patuloside A and B, palmitic acid, and hexahydroxydiphenic acid, are responsible for the anti-aging effects of the plant on the skin, and they work synergistically through a multi-target inhibition of elastase, collagenase, and tyrosinase enzyme activity. This medicinal plant can find application in the cosmetics industry as a natural raw material for the production of anti-aging skin care products and general maintenance of skin integrity. The identified phytochemicals can also serve as a lead compound for the development of anti-aging compounds for use in cosmetics industries. Further studies should be conducted in vitro and in vivo to provide more data for the commercialization of this plant for skincare products.

Author Contributions: Conceptualization, B.E.O., B.O.A. and O.I.O.; methodology, E.A.A., O.E.A., C.S.U. and O.V.O.; software and validation, E.A.A., O.E.A., C.S.U., S.A.O. and R.T.A.; writing original draft, E.A.A., O.E.A. and J.N.E.; supervision, B.E.O., B.O.A. and O.I.O.; project administration, B.E.O. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data presented in this study are available upon request from the corresponding author.

Acknowledgments: The authors acknowledge the Institute of Drug Research and Development, SE Bogoro Center, Afe Babalola University, for making their biocomputational facility available for this study.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Aburjai, T.; Natsheh, F.M. Plants used in cosmetics. *Phytother. Res. PTR* **2003**, *17*, 987–1000. [CrossRef]
2. Agbebi, E.A.; Alabi, O.S.; Nkrumah, A.O.; Ogbole, O.O. Evaluation of the antibacterial and antifungal potentials of peptide-rich extracts from selected Nigerian Plants. *Eur. J. Integr. Med.* **2022**, *54*, 102163. [CrossRef]
3. Altyar, A.E.; Ashour, M.L.; Youssef, F.S. Premna odorata: Seasonal Metabolic Variation in the Essential Oil Composition of Its Leaf and Verification of Its Anti-Ageing Potential via In Vitro Assays and Molecular Modelling. *Biomolecules* **2020**, *10*, 879. [CrossRef] [PubMed]
4. Alves, N.S.F.; Setzer, W.N.; da Silva, J.K.R. The chemistry and biological activities of *Peperomia pellucida* (Piperaceae): A critical review. *J. Ethnopharmacol.* **2019**, *232*, 90–102. [CrossRef]
5. Amirah, S.; Zain, H.H.M.; Husni, I.; Kassim, N.K.; Amin, I. In vitro Antioxidant Capacity of *Peperomia pellucida* (L.) Kunth Plant from two different locations in Malaysia using different Solvents Extraction. *Res. J. Pharm. Technol.* **2020**, *13*, 1767. [CrossRef]
6. Arrigoni-Blank, M.D.F.; Dmitrieva, E.G.; Franzotti, E.M.; Antoniolli, A.R.; Andrade, M.R.; Marchioro, M. Anti-inflammatory and analgesic activity of *Peperomia pellucida* (L.) HBK (Piperaceae). *J. Ethnopharmacol.* **2004**, *91*, 215–218. [CrossRef] [PubMed]
7. Bae-Harboe, Y.-S.C.; Park, H.-Y. Tyrosinase: A Central Regulatory Protein for Cutaneous Pigmentation. *J. Investig. Dermatol.* **2012**, *132*, 2678–2680. [CrossRef] [PubMed]
8. Bertini, I.; Calderone, V.; Cosenza, M.; Fragai, M.; Lee, Y.-M.; Luchinat, C.; Mangani, S.; Terni, B.; Turano, P. Conformational variability of matrix metalloproteinases: Beyond a single 3D structure. *Proc. Natl. Acad. Sci. USA* **2005**, *102*, 5334–5339. [CrossRef]
9. Buonocore, D.; Lazzeretti, A.; Tocabens, P.; Nobile, V.; Cestone, E.; Santin, G.; Bottone, M.G.; Marzatico, F. Resveratrol-procyanidin blend: Nutraceutical and antiageing efficacy evaluated in a placebo-controlled, double-blind study. *Clin. Cosmet. Investig. Dermatol.* **2012**, *159*, 159–165. [CrossRef]

10. Burdock, G.A.; Soni, M.G.; Carabin, I.G. Evaluation of Health Aspects of Kojic Acid in Food. *Regul. Toxicol. Pharmacol.* **2001**, *33*, 80–101. [CrossRef]
11. Elgamal, A.M.; El Raey, M.A.; Gaara, A.; Abdelfattah, M.A.O.; Sobeh, M. Phytochemical profiling and anti-ageing activities of *Euphorbia retusa* extract: In silico and in vitro studies. *Arab. J. Chem.* **2021**, *14*, 103159. [CrossRef]
12. Eun Lee, K.; Bharadwaj, S.; Yadava, U.; Gu Kang, S. Evaluation of caffeine as inhibitor against collagenase, elastase and tyrosinase using in silico and in vitro approach. *J. Enzym. Inhib. Med. Chem.* **2019**, *34*, 927–936. [CrossRef] [PubMed]
13. Friesner, R.A.; Murphy, R.B.; Repasky, M.P.; Frye, L.L.; Greenwood, J.R.; Halgren, T.A.; Sanschagrin, P.C.; Mainz, D.T. Extra precision glide: Docking and scoring incorporating a model of hydrophobic enclosure for protein-ligand complexes. *J. Med. Chem.* **2006**, *49*, 6177–6196. [CrossRef] [PubMed]
14. Kamimura, A.; Takahashi, T.; Watanabe, Y. Investigation of topical application of procyanidin B-2 from apple to identify its potential use as a hair growing agent. *Phytomedicine* **2000**, *7*, 529–536. [CrossRef] [PubMed]
15. Karamanos, N.K.; Theocharis, A.D.; Piperigkou, Z.; Manou, D.; Passi, A.; Skandalis, S.S.; Vynios, D.H.; Orian-Rousseau, V.; Ricard-Blum, S.; Schmelzer, C.E.H.; et al. A guide to the composition and functions of the extracellular matrix. *FEBS J.* **2021**, *288*, 6850–6912. [CrossRef]
16. Kartika, I.G.A.A.; Insanu, M.; Safitri, D.; Putri, C.A.; Adnyana, I.K. New update: Traditional uses, phytochemical, pharmacological and toxicity review of *Peperomia pellucida* (L.) kunth. *Pharmacologyonline* **2016**, *2016*, 30–43.
17. Kohno, T.; Hochigai, H.; Yamashita, E.; Tsukihara, T.; Kanaoka, M. Crystal structures of the catalytic domain of human stromelysin-1 (MMP-3) and collagenase-3 (MMP-13) with a hydroxamic acid inhibitor SM-25453. *Biochem. Biophys. Res. Commun.* **2006**, *344*, 315–322. [CrossRef]
18. Amid Koparde, A.; Chandrashekar Doijad, R.; Shripal Magdum, C. Natural products in drug discovery. In *Pharmacognosy—Medicinal Plants*; IntechOpen: London, UK, 2019. Available online: http://dx.doi.org/10.5772/intechopen.82860 (accessed on 14 August 2023).
19. Kosasih, S.; Ginting, N.; Chiuman, L.; Nyoman, I.; Lister, E. The Effectiveness of *Peperomia Pellucida* Extract Against Acne Bacteria. *Am. Sci. Res. J. Eng. Technol. Sci.* **2019**, *59*, 149–153.
20. Lai, X.; Wichers, H.J.; Soler-Lopez, M.; Dijkstra, B.W. Structure of Human Tyrosinase Related Protein 1 Reveals a Binuclear Zinc Active Site Important for Melanogenesis. *Angew. Chem. Int. Ed.* **2017**, *56*, 9812–9815. [CrossRef]
21. Majumder, P. Phytochemical, pharmacognostical and physicochemical standardization of *Peperomia pellucida* (L.) HBK. STEM. *Pharm. Glob.* **2011**, *2*, 1–4.
22. Majumder, P.; Abraham, P.; Satya, V. Ethno-medicinal, phytochemical and pharmacological review of an amazing medicinal herb *Peperomia pellucida* (L.) HBK. *Res. J. Pharm. Biol. Chem. Sci.* **2011**, *2*, 358–364.
23. Matos, M.S.; Romero-Díez, R.; Álvarez, A.; Bronze, M.R.; Rodríguez-Rojo, S.; Mato, R.B.; Cocero, M.J.; Matias, A.A. Polyphenol-Rich Extracts Obtained from Winemaking Waste Streams as Natural Ingredients with Cosmeceutical Potential. *Antioxidants* **2019**, *8*, 355. [CrossRef] [PubMed]
24. Morris, G.M.; Huey, R.; Olson, A.J. Using AutoDock for Ligand-Receptor Docking. *Curr. Protoc. Bioinform.* **2008**, *24*, 8–14. [CrossRef] [PubMed]
25. Ohno, R.; Moroishi, N.; Sugawa, H.; Maejima, K.; Saigusa, M.; Yamanaka, M.; Nagai, M.; Yoshimura, M.; Amakura, Y.; Nagai, R. *Mangosteen pericarp* extract inhibits the formation of pentosidine and ameliorates skin elasticity. *J. Clin. Biochem. Nutr.* **2015**, *57*, 27–32. [CrossRef]
26. Oloyede, G.; Onocha, P. Phytochemical, toxicity, antimicrobial and antioxidant screening of leaf extracts of *Peperomia pellucida* from Nigeria. *Adv. Environ. Biol.* **2011**, *5*, 3700–3709.
27. Olugbogi, E.A.; Bodun, D.S.; Omoseeye, S.D.; Onoriode, A.O.; Oluwamoroti, F.O.; Adedara, J.F.; Oriyomi, I.A.; Bello, F.O.; Olowoyeye, F.O.; Laoye, O.G.; et al. *Quassia amara* bioactive compounds as a Novel DPP-IV inhibitor: An in-silico study. *Bull. Natl. Res. Cent.* **2022**, *46*, 217. [CrossRef]
28. Ooi, D.J.; Iqbal, S.; Ismail, M. Proximate Composition, Nutritional Attributes and Mineral Composition of *Peperomia pellucida* L. (Ketumpangan Air) Grown in Malaysia. *Molecules* **2012**, *17*, 11139–11145. [CrossRef] [PubMed]
29. Papaccio, F.; D'Arino, A.; Caputo, S.; Bellei, B. Focus on the Contribution of Oxidative Stress in Skin Ageing. *Antioxidants* **2022**, *11*, 1121. [CrossRef]
30. Parrado, C.; Mercado-Saenz, S.; Perez-Davo, A.; Gilaberte, Y.; Gonzalez, S.; Juarranz, A. Environmental Stressors on Skin Ageing. Mechanistic Insights. *Front. Pharmacol.* **2019**, *10*, 461144. [CrossRef]
31. Puizina-Ivić, N.; Mirić, L.; Čarija, A.; Karlica, D.; Marasović, D. Modern approach to topical treatment of ageing skin. *Coll. Antropol.* **2010**, *34*, 1145–1153.
32. Quan, T.; Wang, F.; Shao, Y.; Rittié, L.; Xia, W.; Orringer, J.S.; Voorhees, J.J.; Fisher, G.J. Enhancing Structural Support of the Dermal Microenvironment Activates Fibroblasts, Endothelial Cells, and Keratinocytes in Aged Human Skin In Vivo. *J. Investig. Dermatol.* **2013**, *133*, 658–667. [CrossRef]
33. Rue, E.A.; Rush, M.D.; van Breemen, R.B. Procyanidins: A comprehensive review encompassing structure elucidation via mass spectrometry. *Phytochem. Rev. Proc. Phytochem. Soc. Eur.* **2018**, *17*, 1–16. [CrossRef]
34. Saeedi, M.; Eslamifar, M.; Khezri, K. Kojic acid applications in cosmetic and pharmaceutical preparations. *Biomed. Pharmacother.* **2019**, *110*, 582–593. [CrossRef] [PubMed]

35. Saputri, F.C.; Hutahaean, I.; Mun'im, A. *Peperomia pellucida* (L.) Kunth as an angiotensin-converting enzyme inhibitor in two-kidney, one-clip Goldblatt hypertensive rats. *Saudi J. Biol. Sci.* **2021**, *28*, 6191–6197. [CrossRef] [PubMed]
36. Senol Deniz, F.S.; Orhan, I.E.; Duman, H. Profiling cosmeceutical effects of various herbal extracts through elastase, collagenase, tyrosinase inhibitory and antioxidant assays. *Phytochem. Lett.* **2021**, *45*, 171–183. [CrossRef]
37. Shoko, T.; Maharaj, V.J.; Naidoo, D.; Tselanyane, M.; Nthambeleni, R.; Khorombi, E.; Apostolides, Z. Anti-ageing potential of extracts from *Sclerocarya birrea*, (A. Rich.) Hochst and its chemical profiling by UPLC-Q-TOF-MS. *BMC Complement. Altern. Med.* **2018**, *18*, 54. [CrossRef] [PubMed]
38. Sonibare, M.A.; Moody, J.O.; Adesanya, E.O. Use of medicinal plants for the treatment of measles in Nigeria. *J. Ethnopharmacol.* **2009**, *122*, 268–272. [CrossRef] [PubMed]
39. Tanigawa, T.; Kanazawa, S.; Ichibori, R.; Fujiwara, T.; Magome, T.; Shingaki, K.; Miyata, S.; Hata, Y.; Tomita, K.; Matsuda, K.; et al. (+)-Catechin protects dermal fibroblasts against oxidative stress-induced apoptosis. *BMC Complement. Altern. Med.* **2014**, *14*, 133. [CrossRef] [PubMed]
40. Tobin, D.J. Introduction to skin ageing. *J. Tissue Viability* **2017**, *26*, 37–46. [CrossRef]
41. Venkatesh, S.; Maymone, M.B.C.; Vashi, N.A. Ageing in skin of color. *Clin. Dermatol.* **2019**, *37*, 351–357. [CrossRef] [PubMed]
42. Vierkötter, A.; Krutmann, J. Environmental influences on skin ageing and ethnic-specific manifestations. *Derm. Endocrinol.* **2012**, *4*, 227–231. [CrossRef] [PubMed]
43. Widowati, W.; Ginting, C.N.; Lister, I.N.E.; Girsang, E.; Amalia, A.; Wibowo, S.H.B.; Kusuma, H.; Rizal, R. Anti-ageing Effects of Mangosteen Peel Extract and Its Phytochemical Compounds: Antioxidant Activity, Enzyme Inhibition and Molecular Docking Simulation. *Trop. Life Sci. Res.* **2020**, *31*, 127–144. [CrossRef] [PubMed]
44. Zolghadri, S.; Bahrami, A.; Hassan Khan, M.T.; Munoz-Munoz, J.; Garcia-Molina, F.; Garcia-Canovas, F.; Saboury, A.A. A comprehensive review on tyrosinase inhibitors. *J. Enzym. Inhib. Med. Chem.* **2019**, *34*, 279–309. [CrossRef] [PubMed]