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Employing an *in-silico* approach in the evaluation of *Aloe vera* metabolites as inhibitors of Omicron RBD and the ROS-dependent cellular signaling processes

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ABSTRACT

The pathogenesis of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection is related to increased reactive oxygen species (ROS) formation. This increasing ROS formation can mediate ROS-dependent cellular signaling processes inducing cytokines and inflammations that worsen the disease. The severity of coronavirus disease 2019 (COVID-19) can progress due to the self-sustaining cycle of ROS release, inflammatory mediators, and cellular damage. For the treatment, *Aloe vera* is a promising plant that has the potential to be used. In this study, therefore, we identified the metabolite composition of *A. vera* peel and gel using liquid chromatography-mass spectrometry (LC-MS). The metabolites were molecularly docked to Omicron receptor-binding domain (RBD) and ROS-producing enzymes to obtain medicinal compounds to inhibit these targets. The LC-MS analysis revealed the peel and gel compositions are distinct, in which 13 metabolites are identified in the peel and 12 in the gel. Furthermore, these metabolites might be promising inhibitors against Omicron variant SARS-CoV-2 RBD and ROS-producing enzymes based on the docking scores and the number of bonds formed. Thus, *A. vera* is one promising candidate for COVID-19 treatment due to its potential to alter the RBD function of forming a complex with ACE2 and inhibit the ROS-dependent cellular signaling processes related to COVID-19 pathogenesis and disease severity progression.

INTRODUCTION

The increased level of reactive oxygen species (ROS) appears to play a role in the pathogenesis of coronavirus disease 2019 (COVID-19) (Cecchini and Cecchini, 2020). ROS are small oxygen-derived molecules that can be in the form of oxidizing agents or also molecules that are easily converted into oxygen radicals (André-Lévigne *et al*., 2017). Increased production of ROS can lead to oxidative stress-induced molecular damage. Recent methodological advances have identified the enzymes responsible for producing and elevating ROS levels. Elevated ROS levels have been linked to a variety of diseases, including cardiovascular, immune, and nervous system disorders, as well as aging and cancer. According to a recent study, selectively targeting specific ROS levels enables the development of a futuristic refined redox medicine (Sammar *et al*., 2019; Sies and Jones, 2020). Production and elevation formation of ROS is correlated with almost every human disease. As a result, selectively targeting ROS-related proteins is an important step in the development of redox medicine. Network pharmacology and big data in drug discovery and development are modernized approaches in treating oxidative stress that has placed ROS as the basis of a number of diseases. However, studies on the regulation of ROS levels must be conducted in greater detail in order to elucidate the mechanism

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by which ROS causes disease and cellular damage (Casas *et al*., 2020).

Antioxidants have been extensively used as agents to lower ROS levels and treat oxidative stress. Antioxidant supplementation reduces COVID-19 severity due to its stress oxidative relieving effect (Delgado-Roche and Mesta, 2020; Pizzino *et al*., 2017).

The use of herbal medicine is becoming popular worldwide and well accepted and distributed as food composition and supplements that are safe as antioxidants (Ekor, 2014). *Aloe vera* is one of the plants known to exhibit great antioxidant activity. Hence, it has the potential to regulate ROS formation in the body. It contains a considerable number of metabolites, such as flavonoids, terpenoids, lectins, fatty acids, anthraquinones, pectins, hemicelluloses, glucomannan, tannins, campesterol, capreol, enzymes, salicylic acid, calcium, chromium, copper, iron, magnesium, manganese, potassium, phosphorus, sodium, zinc, retinol, ascorbic acid, alpha-tocopherol, β-carotene, B1–B3, B6, choline, B12, and folic acid (Hęś *et al*., 2019).

The variety of metabolite makeup of a plant depends on several aspects, such as the type and conditions of cultivation, climate condition, harvesting time, position of the harvested leaves, species, variety, and method used for harvesting leaves (Giannakoudakis *et al*., 2018). We hypothesized that *A. vera* grown in North Sulawesi, Indonesia, contains different metabolites than those grown in other regions, owing to the different cultivation conditions and climates. Information on metabolite classes has been published. Specific metabolites from the peel and gel of *A. vera* collected from the Indonesian island of North Sulawesi, on the other hand, have never been previously reported.

There is a dearth of information regarding the composition of *A. vera* metabolites from North Sulawesi and their effect on ROS levels and COVID-19 patients. Hence, we used liquid chromatography-mass spectrometry (LC-MS) to determine the metabolite composition of the peel and gel of *A. vera* collected in North Sulawesi, Indonesia. Further docking of the compounds to the ROS-producing enzymes was performed to identify compounds that have the potential to inhibit the proteins' function and stop their biological processes. In addition, to look at the direct inhibition potential against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), we also evaluated the inhibition potential of the metabolites against the receptor-binding domain (RBD) of the Omicron variant using the same method as against the ROS-producing enzymes. Since RBD plays an essential role in viral entry into human cells, targeting this domain can illustrate the role of the metabolites in halting the virus's life cycle.

METHODS

Metabolite identification

The identification of the metabolites was carried out using an LC-MS instrument in accordance with the protocol described by Nur *et al*. (2021), with slight modifications to the run time, specifically for 25 minutes.

Molecular docking protocol

The identified metabolites were used as ligands. The structures were obtained from PubChem, their energy was minimized, and then PyRx 8.0 was used to dock them to the ROS-producing enzymes. The targets used in this protocol are RBD of SARS-CoV-2 Omicron variant (PDB code: 7T9L), inducible nitric oxide synthase (NOS) (PDB code: 4UX6), endothelial nitric oxide synthase (eNOS) (PDB code: 6CIF), neuronal nitric oxide synthase (nNOS) (PDB code: 6PMY), cyclooxygenase-1 (PDB code: 5U6X), cyclooxygenase-2 (PDB code: 6V3R), and 5-lipoxygenase (5-LOX) (PDB code: 6N2W). Vitamin C was used as a benchmark because it is an extremely powerful antioxidant. The docking result was ranked based on the docking scores (kcal/mol) and the number of hydrogen bonds due to the evaluation of the two of these aspects is important and may enhance the linearity with experimental data (Cheshire *et al*., 2011; Cinelli *et al*., 2020; Cingolani *et al*., 2017; Dallakyan and Olson, 2015; Gilbert *et al*., 2020; Li *et al*, 2018; Mousavi *et al*., 2021; Uddin *et al*., 2020; Tallei *et al*., 2020). The top-ranked ligands were further visualized using the BIOVIA Discovery Studio Visualizer.

RESULTS AND DISCUSSION

Metabolite identification

The previous study lacked data on the metabolites found in *A. vera* peel. Only the peel metabolites classes had been extensively identified, such as steroids, tannins, terpenoids, catechin, carotenoids, and anthraquinones (Bista *et al*., 2020; Dharajiya *et al*., 2017; Muthukumaran *et al*., 2018; Nalimu *et al*., 2021). However, the majority of the available research data did not differentiate between the peel and gel metabolite classes, so the localized major metabolites in each component of the plant are poorly known (Raad *et al*., 2020; Sánchez *et al*., 2020). We found that there are differences in the metabolites in the peel and gel of *A. vera*. The only available data on the metabolites in *A. vera* peel was the result of the study conducted by López *et al*. (2013), using samples collected from the Canary Islands of Spain. They identified sinapic acid, quercitroside, kaempferol, chamomile, protocatechuic, catechin, vanillic acid, epicatechin, cedar acid, 3-O-caffeoylquinic acid, gentisic acid, 3,4-dihydroxycinnamic acid, coumaric acid, 4-hydroxy-3-methoxycinnamic acid, rutin, myricetin, and quercetin. Our finding showed that *A. vera* collected from North Sulawesi of Indonesia contained different types of metabolites. As a result, it can be justified that plants' metabolite content will vary according to their cultivation location (Kumar *et al*., 2017).

There are 13 metabolites identified in the peel and 12 metabolites in the gel of *A. vera*. Figures 1 and 2 depict the LC-MS chromatogram. The result showed that there are differences in metabolites in the peel and gel of *A. vera.* The metabolites detected in the peel include triacetyl-bromo-emodin, 6-O-(alpha-*L*-rhamnopyranosyl)-(1->6)-beta-*D*-glucopyranosyl)emodin, 8-demethyl-8-(alpha-*L*-rhamnosyl)tetracenomycinC,(10R)-1,8,10alpha-triacetoxy-3-acetoxymethyl-10-(2-O,3-O,4-O,6-O-tetraacetyl-beta-*D*-glucopyranosyl)anthrone, aloe-emodin dianthrone diglucoside, (9R)-9-[(9S)-2-carboxy-5-[(2S,3R,4S,5S,6R)-

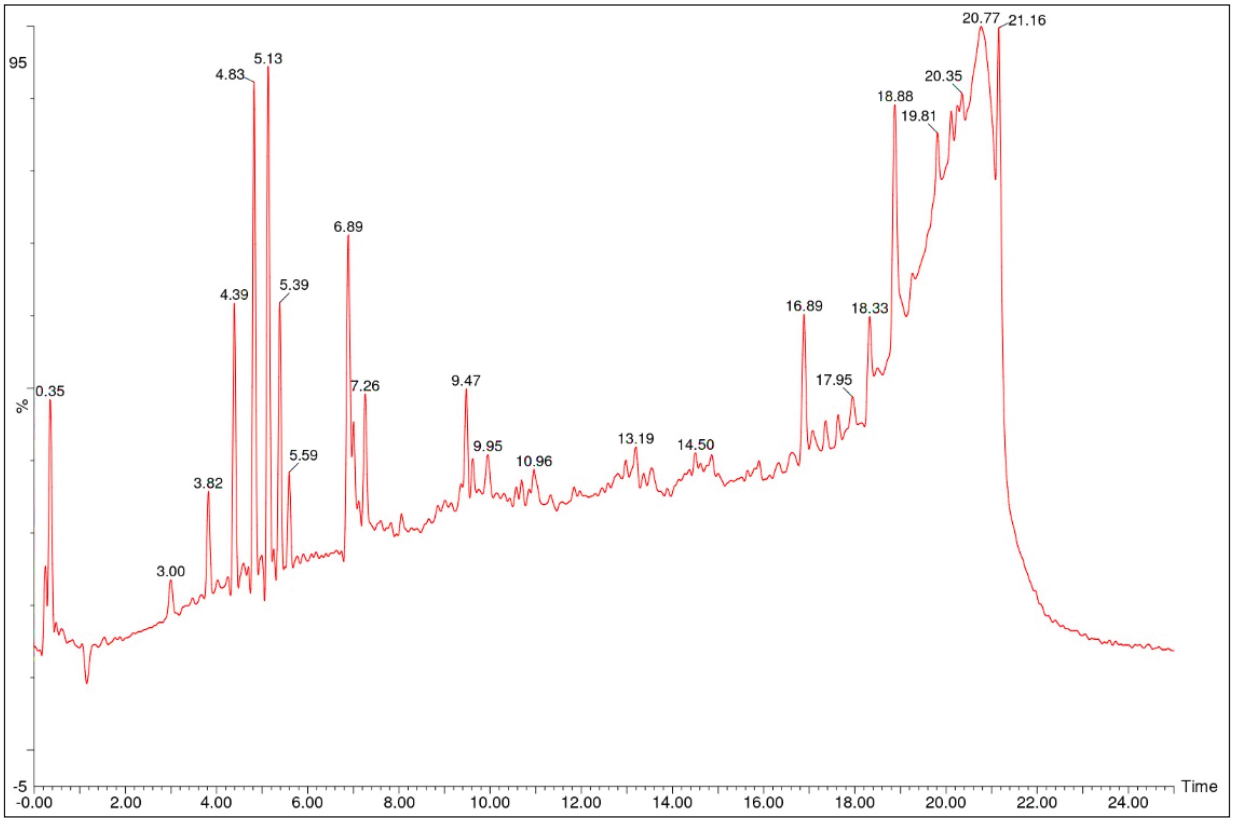


Figure 1. LC-MS result of *A. vera* peel.

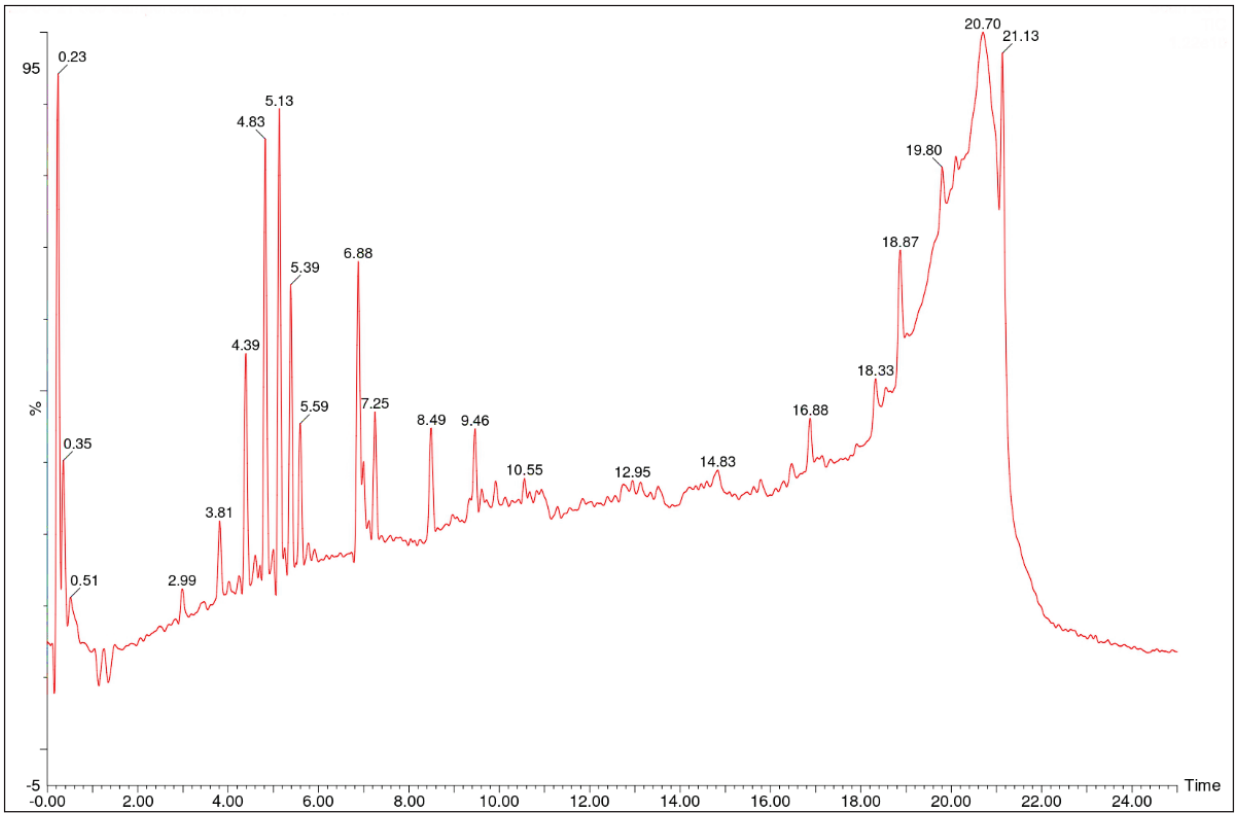


Figure 2. LC-MS result of *A. vera* gel.

4,5-dihydroxy-6-(hydroxymethyl)-3-oxalooxyoxan-2-yl] oxy-4-hydroxy-10-oxo-9H-anthracen-9-yl]-4-hydroxy-10-oxo-5-[(2S,3R,4S,5S,6R)-3,4,5-trihydroxy-6(hydroxymethyl) oxan-2-yl]oxy-9H-anthracene-2-carboxylic acid, landomycin X, emodin-8-*o*-beta-gentiobioside, *L*-rhodinose-2-deoxy-*L*-fucose-2-deoxy-*L*-fucose-10-decarbomethoxy-epsilon-rhodomycinone, adxanthromycin A, landomycin B, and landomycin W.

The metabolites found in gel include aloin 8-O-alpha-*D*-glucopyranoside, aloe-emodin-d5, 1-hydroxyanthrone, 6-O-(alpha-*L*-rhamnopyranosyl)-(1->6)-beta-*D*-glucopyranosyl) emodin, 8-demethyl-8-(alpha-*L*-rhamnosyl)tetracenomycin C, 12-demethyl-elloramycin, (10R)-1,8,10alpha-triacetoxy-3-acetoxymethyl-10-(2-O,3-O,4-O,6-O-tetraacetyl-beta-D-glucopyranosyl)anthrone, aloe-emodin dianthrone diglucoside, (9R)-9-[(9S)-2-carboxy-5-[(2S,3R,4S,5S,6R)-4,5-dihydroxy-6-(hydroxymethyl)-3-oxalooxyoxan-2-yl]oxy-4-hydroxy-10-oxo-9H-anthracen-9-yl]-4-hydroxy-10-oxo-5-[(2S,3R,4S,5S,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl] oxy-9H-anthracene-2-carboxylic acid, landomycin X, emodin-8-*o*-beta-gentiobioside, landomycin A, and adxanthromycin A (Table 1). The Pubchem CIDs of the identified metabolites are listed in Supplementary Tables S1-S2

Molecular docking

The docking scores of vitamin C and *A. vera* metabolites docked to target the enzymes are shown in Table 2. The docking score, measured in kcal/mol, which is an energy per molecule unit, can represent the binding energy of a ligand against a particular protein target. The lower this score is, the higher the affinity is or the better the binding of the ligand-target complex is (Tallei *et al*., 2020). Due to all of the metabolites found in *A. vera*'s peel and gel having lower docking scores than vitamin C, they would highly likely form a better binding complex with the enzymes.

Figure 3 shows vitamin C interactions with all the target proteins, while Figures 4–10 show the visualization of the binding interactions. Numerous bond formations occur in the complexes of the compounds and all of the targeted enzymes, such as conventional hydrogen bond (dark green), carbon-hydrogen bond (light green), alkyl/pi-alkyl bond (light pink), pi-pi stacked bond (dark pink), pi-cation/anion bond (dark orange), pi-sulfur bond (light orange), and pi-sigma bond (purple), with hydrogen and alkyl bonds dominating the interaction.

In addition, the ligands also interact with the enzymes' amino acids through van der Waals interactions. The interactions are also evaluated according to which amino acids in the metabolites bind to because each protein might have certain amino acids that are critical in the inhibition process (Kumar *et al*., 2021).

SARS-CoV-2 Omicron RBD inhibitor

There are 30 mutations that occur in the viral spike of the Omicron variant, among which eight mutations are situated in the spike RBD. These mutations invite the most interest from science and medical researchers due to their significant role as host recognition sites. D339, L371, P373, F375, N471, K440, S446, N477, K478, A484, R493, S496, R498, Y501, and H505 were the output residues of the Omicron variants. Landomycin A of *A. vera,* through hydrogen bond interactions, binds Ser484 and Ser493 in the mutation region of the Omicron RBD. It has been hypothesized that variants carrying multiple mutations in the RBD will probably exhibit an even higher stabilizing effect on the interaction between the viral spike and the host angiotensin-converting enzyme 2 (ACE2) receptor. Hence, landomycin A is predicted to alter this stabilizing effect of the Omicron RBD. Ortega *et al*. (2022) addressed several further analyses employing computational study which are needed to gain a novel perspective of the SARS-CoV-2 Omicron variant's detailed structure, residue formation, and changes that can be potentially inhibited as a druggable binding domain (Ahmad, 2021; Mengist *et al*., 2021; Ortega *et al*., 2021, 2022).

The previous study also found that residues 301 and 430 in the main receptor-binding motif have a critical mechanism due to their role as the antibody recognition region of the viral spike. The antibody recognition site is critical for efficiency. However, the Omicron variant's viral spike carries several mutations located in this region. Therefore, the current assumption is that the antibody will be less effective against the Omicron variant, and further study needs to be done to evaluate the immune response elicited by vaccines against recent emerging SARS-CoV-2 variants (Ewer *et al*., 2021; Harvey *et al*., 2021; Ravichandran *et al*., 2020).

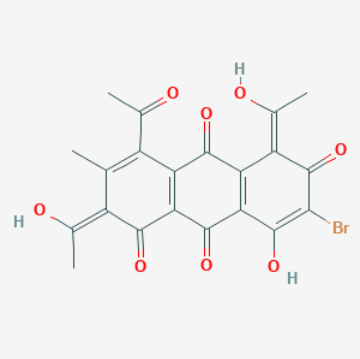
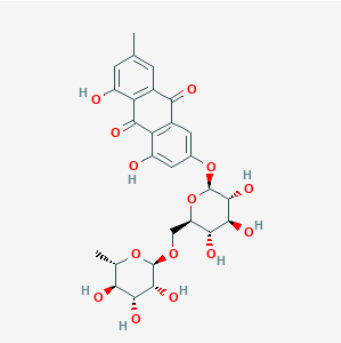
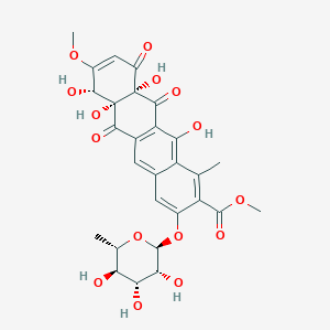
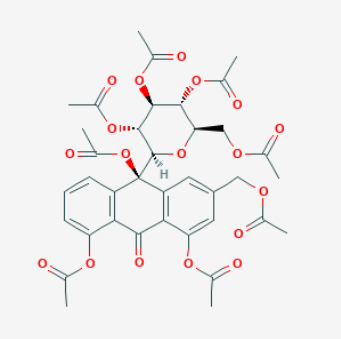
Aloe vera metabolites show great docking scores in the Omicron RBD binding domain, with the value ranging from −6 kcal/mol to −10 kcal/mol. In terms of the binding affinity with ACE2, a significant influence of the Omicron variant RBD was found. Residues 468–473 were reported to play a role in this significant change (Kumar *et al*., 2021). In comparison with the wild-type RBD, the Omicron variant has a better ACE2 binding capacity. Residues T478K and N501Y are associated with increased ACE2 binding capacity (Omotuyi *et al*., 2022). Neither vitamin C nor landomycin A interacts with these residues. However, the B501Y mutation did not enhance Omicron infectivity and epidermal growth factor receptor (EGFR) binding due to the mutations of the charged to the uncharged polar side chain on its interface of interaction. Therefore, N501Y failed to deliver immune disruption and did not lower vaccine efficiency (Baek *et al*., 2021; Kazybay *et al*., 2022; Liu *et al*., 2021).

Nitric oxide synthase inhibitor

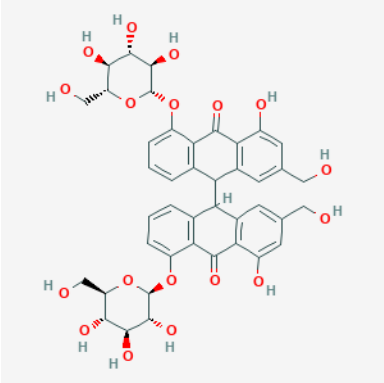
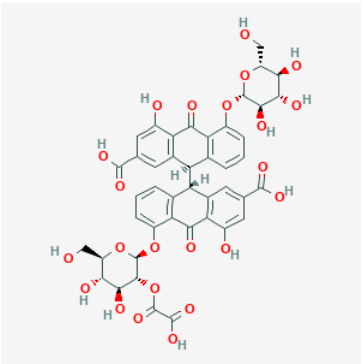
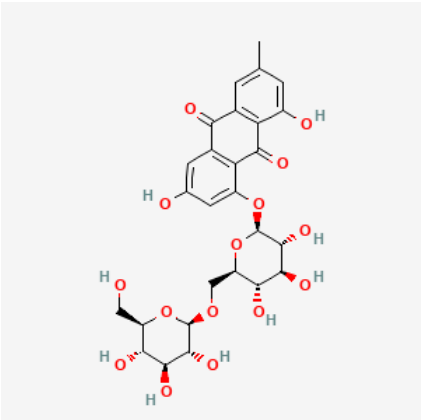
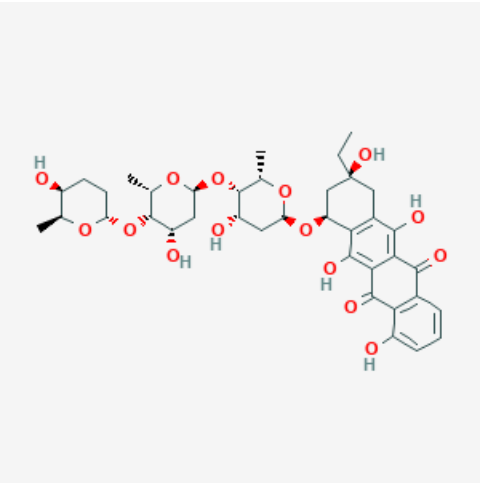
The release of ROS induces oxidative stress. In terms of improving the health conditions of COVID-19 patients, several clinical approaches have been used in order to lower ROS levels and production, as well as to repair the oxidative stress condition (Derouiche, 2020).

There are a number of studies showing that ROS plays a role in SARS-CoV-2 infections. The infections will generate a sustaining cycle of hematological damage, cytopathic hypoxia, and ROS production (Cecchini and Cecchini, 2020). During disease progression, patients with COVID-19 were shown to undergo hematological damage (Chen *et al*., 2020; Huang *et al*., 2020; Lippi and Mattiuzzi, 2020) and cytopathic hypoxia. Disseminated intravascular coagulopathy, sepsis, and decreased oxygen transport to the tissues are some of the main elements presented by COVID-19 patients. In the state of hypoxia, the body generates superoxide, such as hydrogen peroxide, that triggers inducible nitric oxide synthase (iNOS) activation resulting in an increased level of NO and peroxynitrite, which are toxic to mitochondria. Mitochondria impairment leads to cytopathic hypoxia. This creates a self-sustaining cycle and can progress to a

Table 1. Chemical structure of *A. vera* peel and gel metabolites.

	Metabolite	Structure
1	Triacetyl-bromo-emodin	
2	6-O-(Alpha-*L*-rhamnopyranosyl)-(1->6)-beta-*D*-glucopyranosyl)emodin	
3	8-Demethyl-8-(alpha-*L*-rhamnosyl)tetracenomycin C	
4	(10R)-1,8,10Alpha-triacetoxy-3-acetoxymethyl-10-(2-O,3-O,4-O,6-O-tetraacetyl-beta-*D*-glucopyranosyl)anthrone	

Continued

	Metabolite	Structure
5	Aloe-emodin dianthrone diglucoside	
6	(9R)-9-[(9S)-2-Carboxy-5-[(2S,3R,4S,5S,6R)-4,5-dihydroxy-6-(hydroxymethyl)-3-oxalooxyoxan-2-yl]oxy-4-hydroxy-10-oxo-9H-anthracen-9-yl]-4-hydroxy-10-oxo-5-[(2S,3R,4S,5S,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxy-9H-anthracene-2-carboxylic acid	
7	Emodin-8-*o*-beta-gentiobioside	
8	*L*-Rhodinose-2-deoxy-*L*-fucose-2-deoxy-*L*-fucose-10-decarbomethoxy-epsilon-rhodomycinone	

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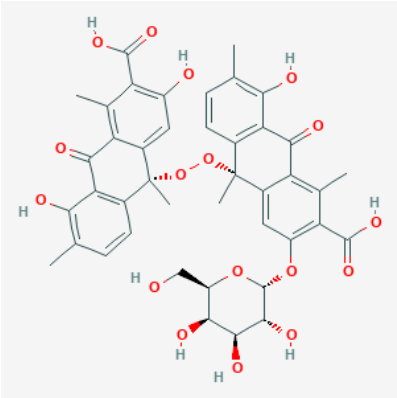
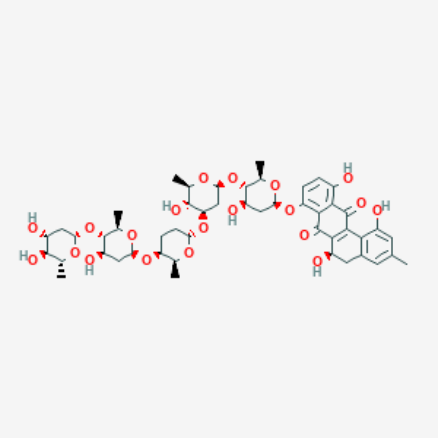
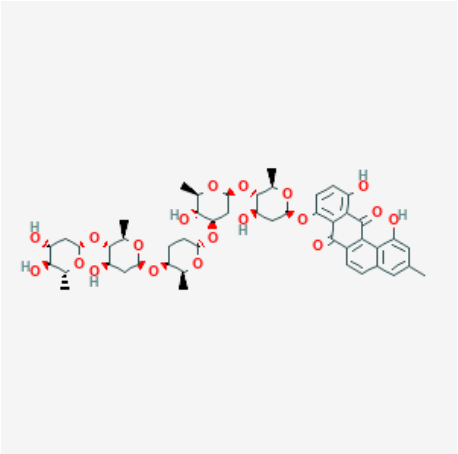
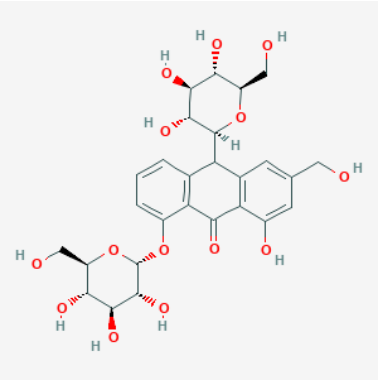
	Metabolite	Structure
9	Adxanthromycin A	
10	Landomycin B	
11	Landomycin W	
12	Aloin 8-O-alpha-*D*-glucopyranoside	

Continued

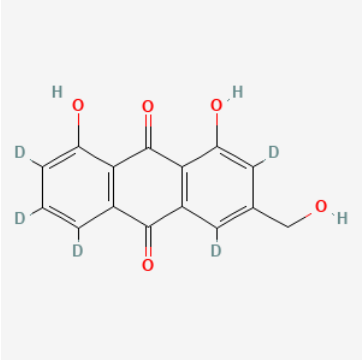
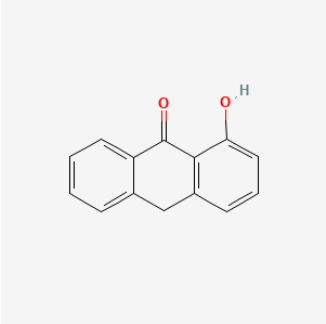
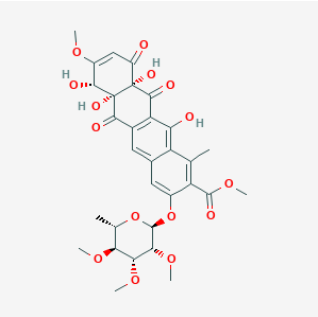
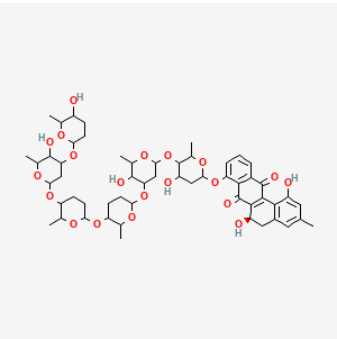
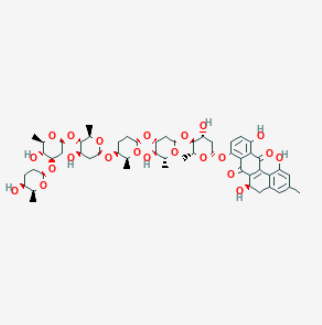
	Metabolite	Structure
13	Aloe-emodin-d5	
14	1-Hydroxyanthrone	
15	12-Demethyl-elloramycin	
16	Landomycin X	
17	Landomycin A	

Table 2. Molecular docking result of vitamin C and *A. vera* peel and gel metabolites against omicron RBD and ROS-producing enzymes. The lowest docking score values are bolded.

Metabolite	Docking score (kcal/mol)						
	RBD (7T9L)	iNOS (4UX6)	eNOS (6CIF)	nNOS (6PMY)	COX-1 (5U6X)	COX-2 (6V3R)	5-LOX (6N2W)
Vitamin C	−5.1	−6	−6.2	−5.8	−6.2	−6	−6
Triacetyl-bromo-emodin	−7.3	−9.9	−9.2	−9.4	−8.6	−8.1	−7.4
6-O-(Alpha-*L*-rhamnopyranosyl)-(1->6)-beta-*D*-glucopyranosyl) emodin	−8	−10.7	−11.6	−12.1	−10.7	−11.4	−9.4
8-Demethyl-8-(alpha-*L*-rhamnosyl)tetracenomycin C	−7.4	−9.9	−9.5	−9.5	−9.4	−9.1	−8.6
(10R)-1,8,10Alpha-triacetoxy-3-acetoxymethyl-10-(2-O,3-O,4-O,6-O-tetraacetyl-beta-*D*-glucopyranosyl)anthrone	−6	−8.5	−7.1	−7.8	−8.1	−7.3	−7.9
Aloe-emodin dianthrone diglucoside	−7.6	−10.8	−9.3	−11.1	−9.6	−9.5	−9.7
(9R)-9-[(9S)-2-Carboxy-5-[(2S,3R,4S,5S,6R)-4,5-dihydroxy-6-(hydroxymethyl)-3-oxalooxyoxan-2-yl]oxy-4-hydroxy-10-oxo-9H-anthracen-9-yl]-4-hydroxy-10-oxo-5-[(2S,3R,4S,5S,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl] oxy-9H-anthracene-2-carboxylic acid	−8.3	−10.6	−10.6	−11.5	−9.9	−10.4	−9.5
Emodin-8-*o*-beta-gentiobioside	−8	−10.5	−11.6	−10.9	−9.6	−11.1	−9.8
L-Rhodinose-2-deoxy-*L*-fucose-2-deoxy-*L*-fucose-10-decarbomethoxy-epsilon-rhodomycinone	−8.4	−11.7	−11.4	−11.3	−10.4	−11.1	−10.1
Adxanthromycin A	−7.5	−11.6	−11	−11.4	−9.4	−10.5	−9.8
Landomycin B	−9.3	−12.1	−13.2	−12.8	−12.1	−10.8	−10.7
Landomycin W	−9	−11.7	−13.5	−12.4	−11.7	−12.2	−11
Aloin 8-O-alpha-*D*-glucopyranoside	−6.9	−7.9	−9.4	−9	−8.8	−8.1	−9.6
Aloe-emodin-d5	−6.4	−8.8	−9.4	−9.1	−8.8	−9.1	−8.3
1-Hydroxyanthrone	−6.7	−9.4	−10.1	−9.7	−8.6	−8.7	−8.4
12-Demethyl-elloramycin	−7	−9.5	−8.4	−8.2	−8.7	−8.4	−8
Landomycin X	−9.5	−12.5	−13.4	−11.7	−11.9	−11.7	−11.1
Landomycin A	−10	−12.2	−13.6	−12	−12	−12	−11

more severe condition (Connors and Levy, 2020; Mantzarlis *et al*., 2017; Ottolenghi *et al*., 2020). In contrast with the NO-induced mitochondria impairment, the production of NO by the eNOS at the proper dose might intriguingly elevate the COVID-19 patient condition. Therefore, selectively targeting NOS is critical (Guan *et al*., 2020; Fang *et al*., 2021; Pieretti *et al*., 2021).

NOS, a homodimer nicotinamide adenine dinucleotide phosphate hydrogen (NADPH) binding enzyme, is regulated by several modulatory interactions with the function of converting *L*-arginine to nitric oxide, one of the ROS. According to the localization or the regulation, NOS is available in three isoforms, namely, nNOS, iNOS, and eNOS. In the state of excessive NO production, this enzyme regulation may lead to cellular or tissue damage (Casas *et al*., 2020; Stuehr and Vasquez-Vivar, 2017).

The peel and gel of *A. vera* were shown to have metabolites that bind to NOS with a stable interaction due to the great docking scores and number of bond formations in the binding complex with landomycin X, landomycin A, and landomycin B. This indicates that these compounds can act as the most potent inhibitors. Our finding on the inhibition of NOS, the enzyme that can induce cellular damage in the state of excessive NO production, may explain the result of a study conducted by Moriyama *et al*. (2016), where they found that *A. vera* treatment increases cell number and wound healing.

Cyclooxygenase (COX) inhibitor

Viral infections have been associated with molecular processes that promote inflammation and oxidative damage during the infection (Komaravelli and Casola, 2014). COVID-19 pathogenesis is closely related to the development of oxidative stress (Chernyak *et al*., 2020). COxs are a part of ROS-producing enzymes (Liu *et al*., 2018). COX produces ROS in the intestinal mucosa. In the presence of free arachidonic acid, COX is activated in the pathway which will result in the production of IL-8 as an end-product and ROS as a byproduct (Alzoghaibi, 2013). The production of ROS leads to oxidative stress, one of the characteristics found in severe COVID-19 patients, that contributes to a complex cellular signaling process in COVID-19 disease progression.

In a severe condition, COVID-19 patients were also shown to have increased fatty acid levels along with inflammatory lipid storms. COX produced inflammatory lipids that were involved in these characteristics (Archambault *et al*., 2020). Activation of the COX pathway plays a pivotal role in the development of inflammation. It leads to the formation of prostaglandins like PGE2

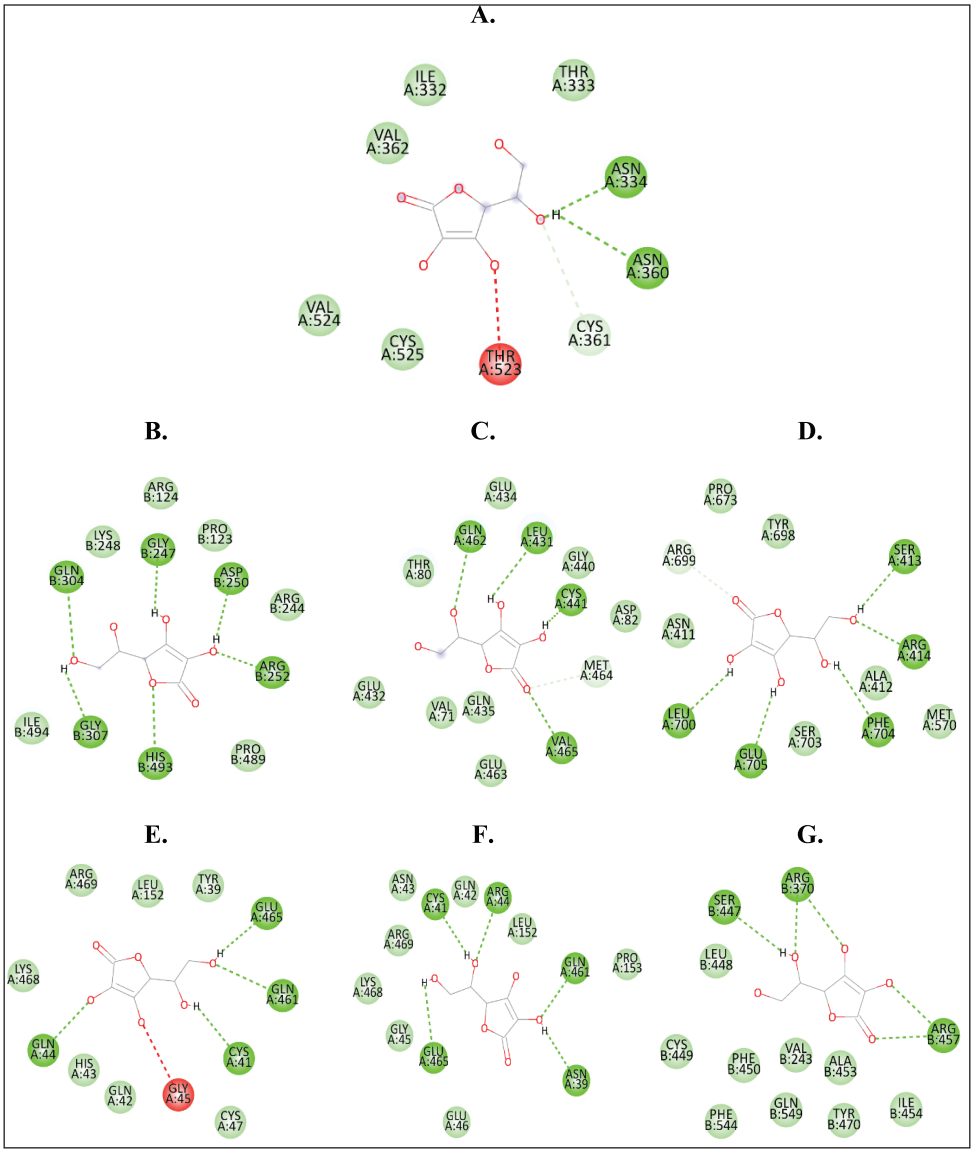


Figure 3. Binding interaction of vitamin C with (A) Omicron RBD, (B) iNOS, (C) eNOS, (D) nNOS, (E) COX-1, (F) COX-2, and (G) 5-LOX.

and PGI2 which are involved in various aspects of inflammation, including the increase of IL-6 production, migration of leukocytes, and fever (McCarthy and Weinberg, 2012).

Aloe vera metabolites can inhibit COX (COX-1 and COX-2) according to the molecular interactions formed in the complex in our finding. The most potential ligands as COX inhibitors are landomycin B and landomycin W. However, there is no clear line on whether COX-1 or COX-2 is more important in lowering ROS levels or in the progression of the viral infection. Therefore, the selectivity of the inhibition needs to be taken into consideration for further analysis.

5-Lipoxygenase inhibitor

The COVID-19 disease can also be characterized by alveolar and endothelial damage, thrombosis, pulmonary embolism, and inflammatory cell infiltration (Barton *et al*., 2020; Damiani *et al*., 2021; Huang *et al*., 2020; Zhang *et al*., 2020). The infiltrating inflammatory cells release ROS in order to clear the infection, leading to the production and accumulation of oxidized phospholipids (OxPLs), oxidized phospholipids, locally in the lungs. OxPLs, which were also detected in the lungs of SARS-CoV-2 patients, signal the activation and release of macrophages, TNF-α, and IL-1β (Merad and Martin, 2020; Poeckel and Funk, 2010). Respiratory symptoms are the hallmark manifestations of SARS-COV-2 infection (Terpos *et al*., 2020).

ROS, along with proinflammatory leukotrienes, are released via the lipoxygenase (LOX) pathway. The release can be derived from arachidonic acid or eicosapentaenoic acid or docosahexaenoic acid (Häfner *et al*., 2019; O'Flaherty *et al*., 2012). 5-LOX is one of the enzymes that mediate the LOX pathway. Other enzymes mediating the LOX pathway are 12-LOX, 15-LOX, and the epidermal LOX (Rådmark *et al*., 2015).

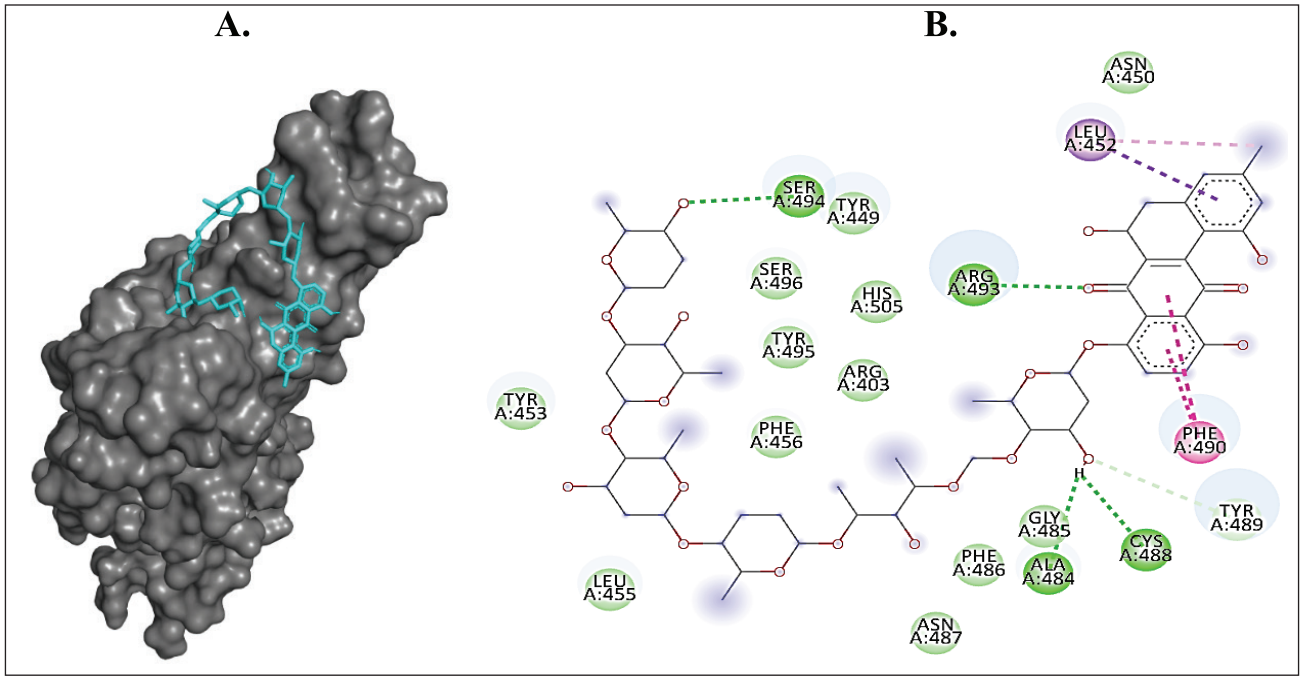


Figure 4. Binding interaction of ligands with the lowest binding energy (landomycin A, PubChem CID: 9988748) against SARS-CoV-2 Omicron RBD. (A) The pose of interactions of the ligand (cyan) and the RBD (gray) in a 3D representation. (B) 2D representation of the pose shows the amino acids of the RBD that interact with the ligand.

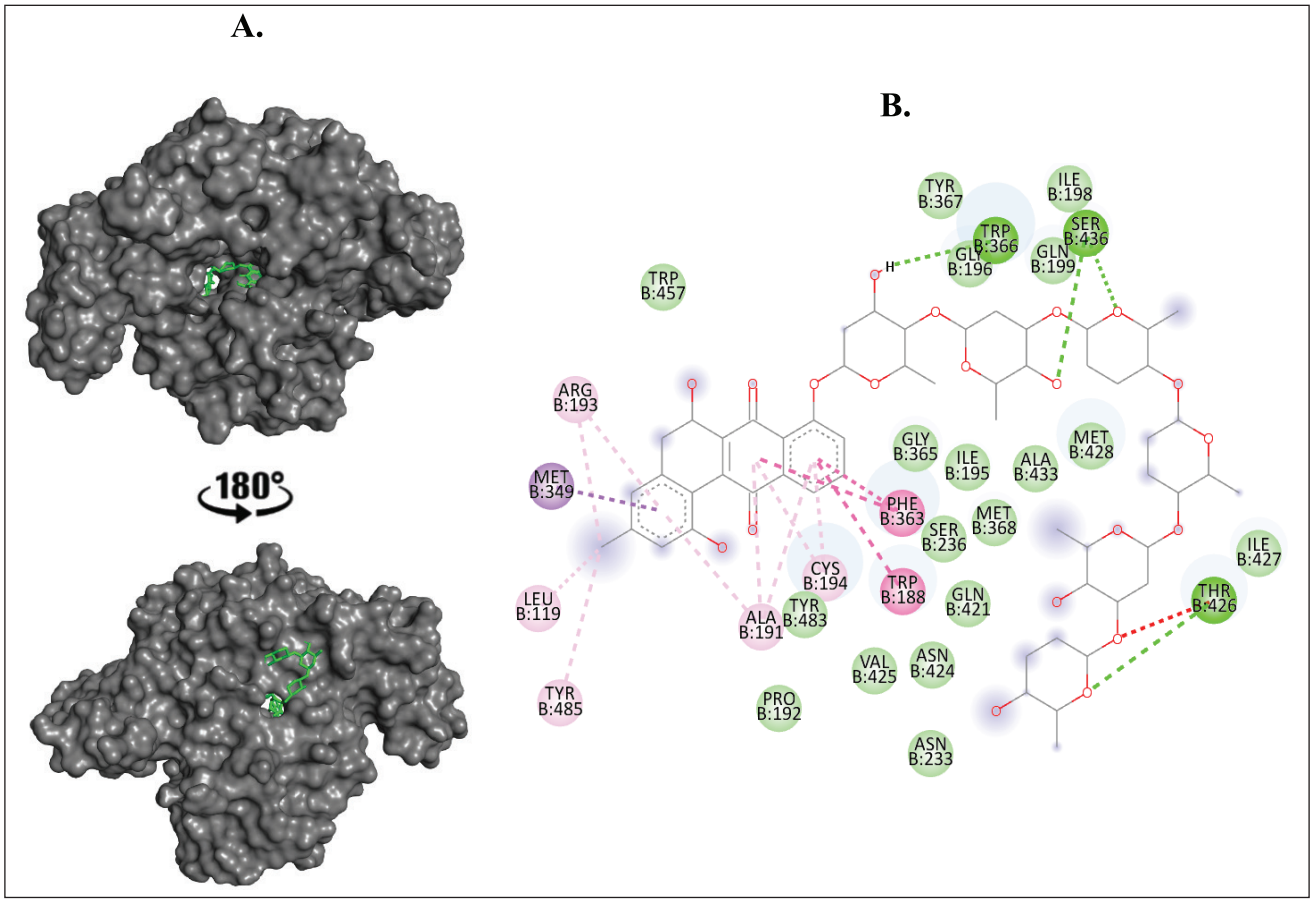


Figure 5. Binding interaction of ligands with the lowest binding energy (landomycin X, PubChem CID: 156580396) against iNOS. (A) The pose of interactions of the ligand (green) and the iNOS (gray) in a 3D representation with 180° rotation views. (B) 2D representation of the pose shows the amino acids of the iNOS that interact with the ligand.

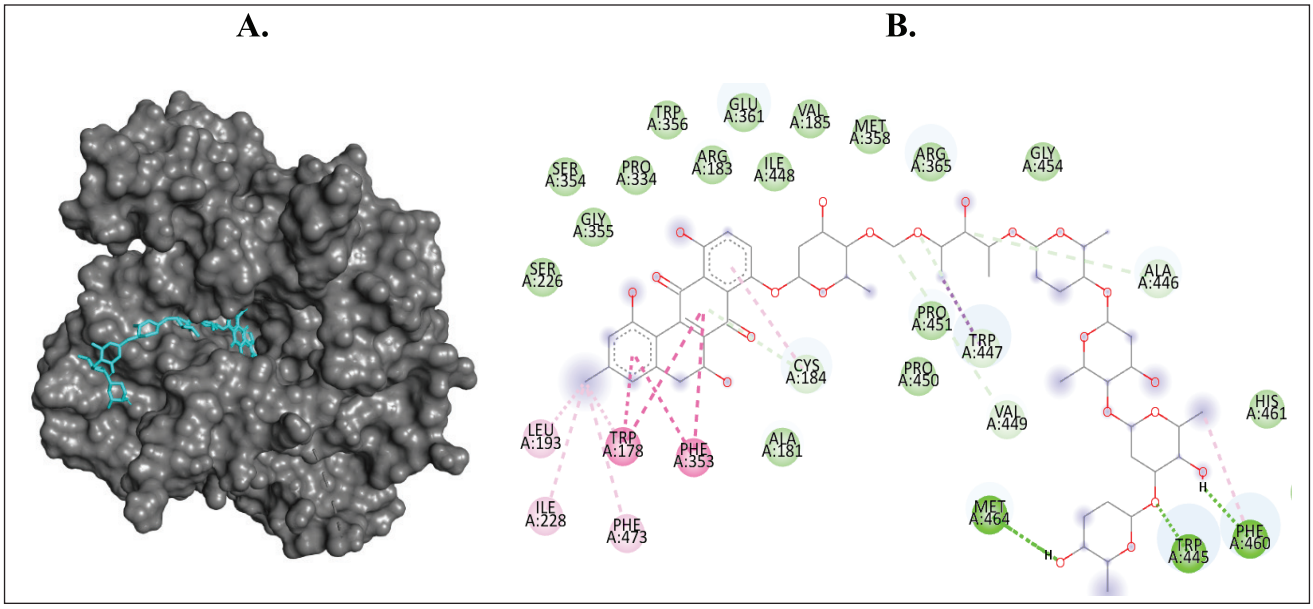


Figure 6. Binding interaction of ligands with the lowest binding energy (landomycin A, PubChem CID: 9988748) against eNOS. (A) The pose of interactions of the ligand (cyan) and the eNOS (gray) in a 3D representation. (B) 2D representation of the pose shows the amino acids of the eNOS that interact with the ligand.

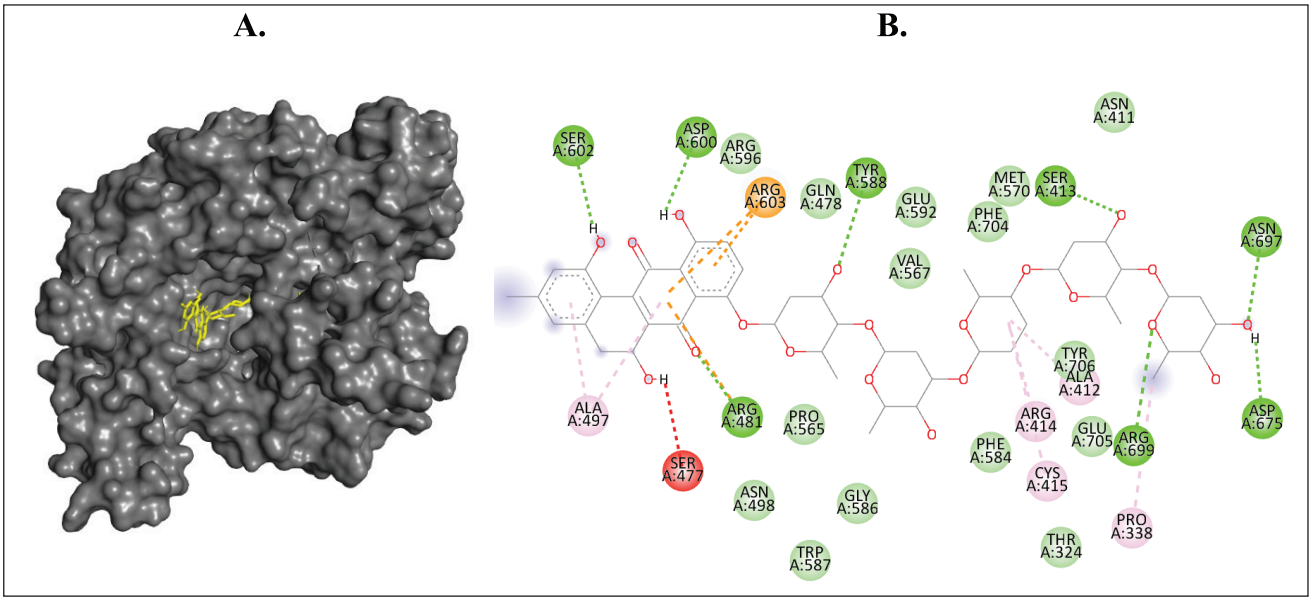


Figure 7. Binding interaction of ligands with the lowest binding energy (landomycin B, PubChem CID: 53297405) against nNOS. (A) The pose of interactions of the ligand (yellow) and the nNOS (gray) in a 3D representation. (B) 2D representation of the pose shows the amino acids of the nNOS that interact with the ligand.

5-LOX, along with COX, can generate ROS as byproducts of its activation. In the presence of free arachidonic acid or NADH or NADPH, 5-LOX produces superoxide, one class of ROS. The activation of 5-LOX mediates the accumulation of intracellular superoxide (Alzoghaibi, 2013; Zhang *et al.*, 2014). Landomycin W, a metabolite localized only in the peel of *A. vera*, is the most potential metabolite to be developed as a 5-LOX inhibitor due to the high number of bond formations and great docking scores. Landomycin W can possibly act as an inhibitor to stop 5-LOX activation. This inhibition is critical due to the activation of 5-LOX, and the release of leukotrienes may contribute to the severity of the disease (Ayola-Serrano *et al.*, 2021). 5-LOX inhibitors can also stop the disease progression by preventing 5-LOX from inducing TNF-a activation and the translocation of NF-κB into the nucleus (Lin *et al.*, 2014).

In severe disease progression, COVID-19 patients are characterized by robust levels of lipids produced by the COX and LOX pathways. Leukotrienes such as LTC4, LTB4, 20-COOH-LTB4, LTE4, and eoxin E4 are also increased (Archambault *et al.*, 2020). A treatment targeting leukotrienes production has been

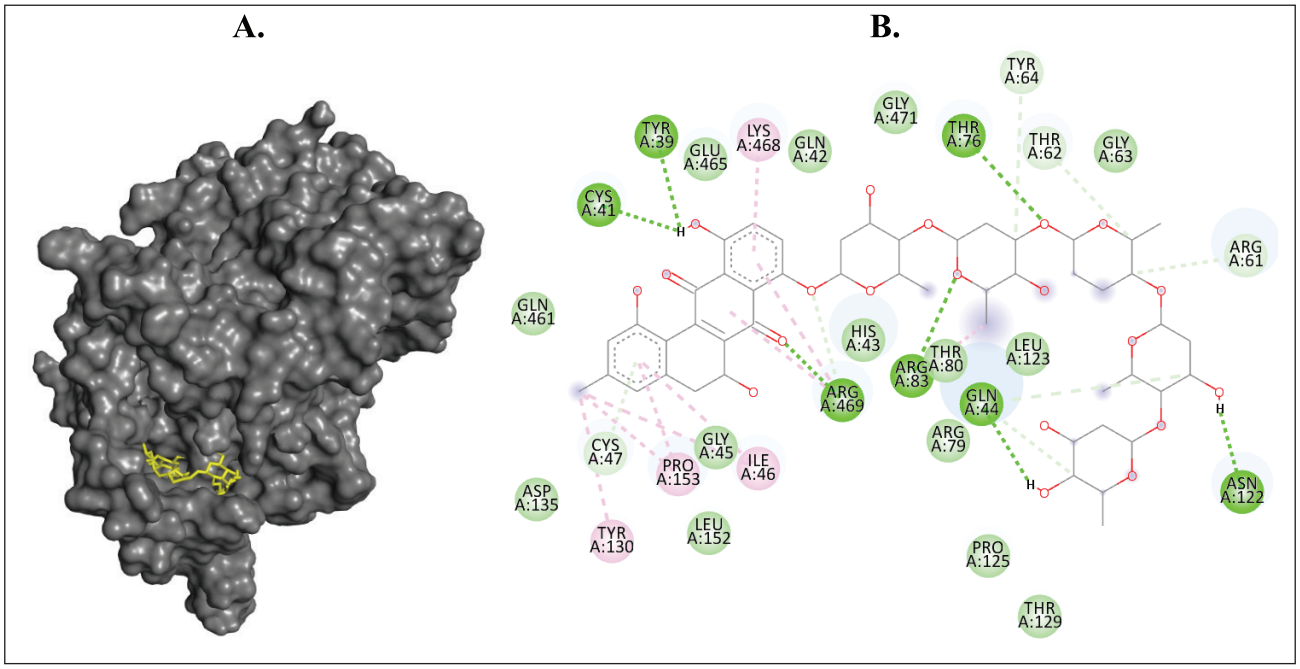


Figure 8. Binding interaction of ligands with the lowest binding energy (landomycin B, PubChem CID: 53297405) against COX-1. (A) The pose of interactions of the ligand (yellow) and the COX-1 (gray) in a 3D representation. (B) 2D representation of the pose shows the amino acids of COX-1 that interact with the ligand.

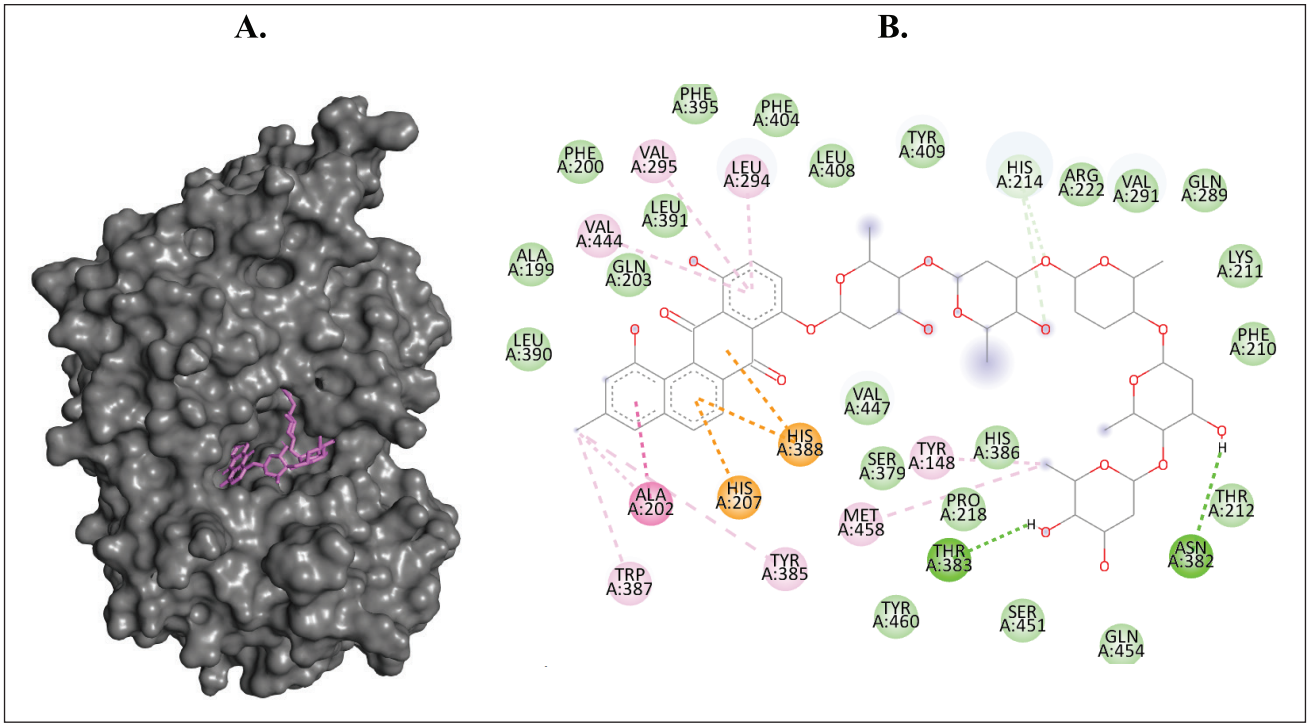


Figure 9. Binding interaction of ligands with the lowest binding energy (landomycin W, PubChem CID: 50993663) against COX-2. (A) The pose of interactions of the ligand (violet) and the COX-2 (gray) in a 3D representation. (B) 2D representation of the pose shows the amino acids of COX-2 that interact with the ligand.

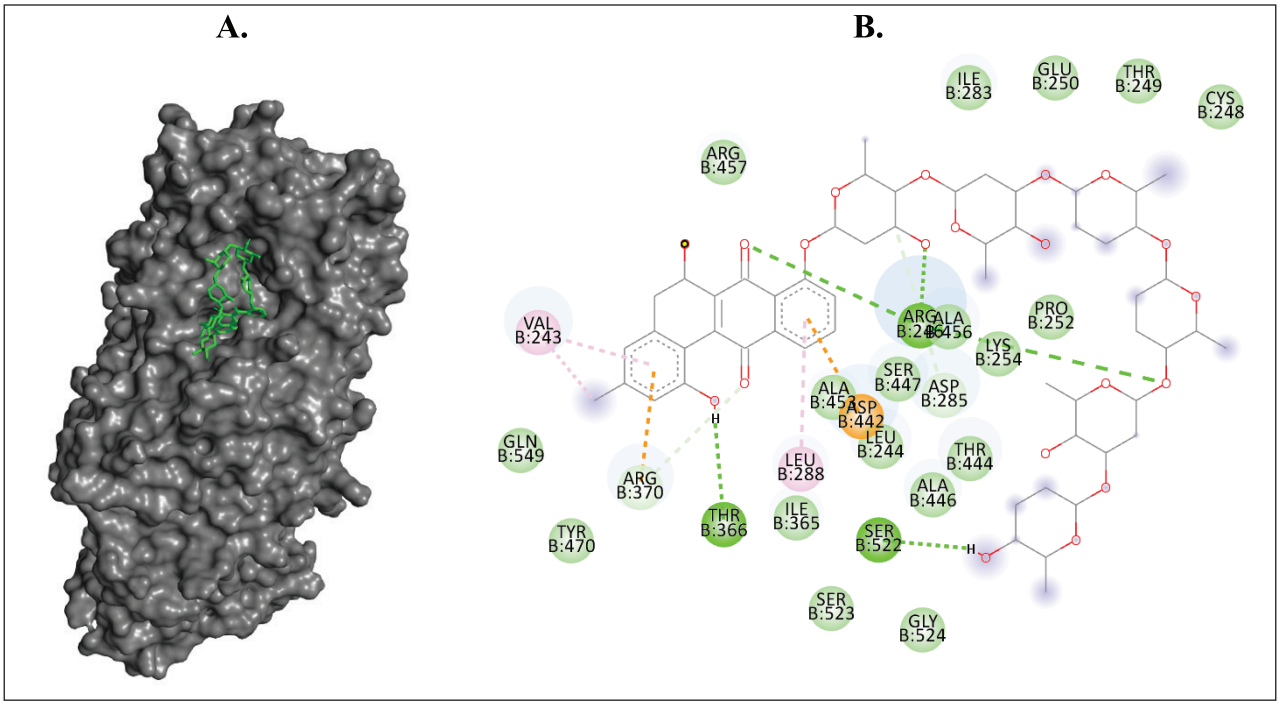


Figure 10. Binding interaction of ligands with the lowest binding energy (landomycin X, PubChem CID: 53297405) against 5-LOX. (A) The pose of interactions of the ligand (green) and the 5-LOX (gray) in a 3D representation. (B) 2D representation of the pose shows the amino acids of 5-LOX that interact with the ligand.

previously offered to treat mild to severe COVID-19 infection (Funk and Ardakani, 2020; Halpin *et al*., 2020).

CONCLUSION

In conclusion, there are differences in the metabolite composition of the peel and gel of *A. vera*. These metabolites of *A. vera* collected from North Sulawesi of Indonesia are found to be different from those collected from different cultivation areas. The docking score and bond formation evaluation of the metabolites in complex with Omicron RBD and ROS-producing enzymes suggest that *A. vera* can inactivate their function and alter the Omicron RBD-ACE2 complex formation and inhibit ROS production, which contributes to the pathogenesis and severity progression of COVID-19.

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AUTHORS' CONTRIBUTIONS

BJK and F designed the study. BJK and F carried out the laboratory work. BJK, MJK, and TET did the *in silico* work. BJK, MJK, and TET analyzed the data. BJK wrote the draft of the manuscript. BJK, MJK, TET, and F read and enriched the manuscript discussion. F and TET reviewed the final manuscript. All authors approved the final version of the manuscript.

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CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

ETHICAL APPROVALS

This study does not involve experiments on animals or human subjects.

DATA AVAILABILITY

All data generated and analyzed are included in this research article.

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REFERENCES

Ahmad L. Implication of SARS-CoV-2 immune escape spike variants on secondary and vaccine breakthrough infections. Front Immunol, 2021; 12:742167; https://doi.org/10.3389/fimmu.2021.742167

Alzoghaibi MA. Concepts of oxidative stress and antioxidant defense in Crohn's disease. World J Gastroenterol, 2013; 19(39):6540–7; https://doi.org/10.3748/wjg.v19.i39.6540

André-Lévigne D, Modarressi A, Pepper MS, Pittet-Cuénod B. Reactive oxygen species and NOX enzymes are emerging as key players in cutaneous wound repair. Int J Mol Sci, 2017; 18(10):2149; https://doi.org/10.3390/ijms18102149

Archambault A-S, Zaid Y, Rakotoarivelo V, Doré E, Dubuc I, Martin C, Amar Y, Cheikh A, Fares H, El Hassan A, Tijani Y, Laviolette M, Boilard E, Flamand L, Flamand N. Lipid storm within the lungs of severe COVID-19 patients: extensive levels of cyclooxygenase and lipoxygenase-derived inflammatory metabolites. MedRxiv; https://doi.org/10.1101/2020.12.04.20242115

Ayola-Serrano NC, Roy N, Fathah Z, Anwar MM, Singh B, Ammar N, Sah R, Elba A, Utt RS, Pecho-Silva S, Rodriguez-Morales AJ, Dhama K, Quraishi S. The role of 5-lipoxygenase in the pathophysiology of COVID-19 and its therapeutic implications. Inflamm Res, 2021; 70(8):877–89; https://doi.org/10.1007/s00011-021-01473-y

Baek M, DiMaio F, Anishchenko I, Dauparas J, Ovchinnikov S, Lee GR, Wang J, Cong Q, Kinch LN, Schaeffer RD, Millán C, Park H, Adams C, Glassman CR, DeGiovanni A, Pereira JH, Rodrigues AV, van Dijk AA, Ebrecht AC, Opperman DJ, Sagmeister T, Buhlheller C, Pavkov-Keller T, Rathinaswamy MK, Dalwadi U, Yip CK, Burke JE, Garcia KC, Grishin NV, Adams PD, Read RJ, Baker D. Accurate prediction of protein structures and interactions using a three-track neural network. Science, 2021; 373(6557):871–6; https://doi.org/10.1126/science.abj8754

Barton LM, Duval EJ, Stroberg E, Ghosh S, Mukhopadhyay S. COVID-19 autopsies, Oklahoma, USA. Am J Clin Pathol, 2020; 153(6):725–33; https://doi.org/10.1093/ajcp/aqaa062

Bista R, Ghimire A, Subedi S. Phytochemicals and antioxidant activities of *Aloe Vera* (Aloe Barbadensis). J Nut Sci Heal Diet, 2020; 1(1):25–36; https://doi.org/10.47890/JNSHD/2020/RBista/10243803

Casas AI, Nogales C, Mucke HAM, Petraina A, Cuadrado A, Rojo AI, Ghezzi P, Jaquet V, Augsburger F, Dufrasne F, Soubhye J, Deshwal S, Di Sante M, Kaludercic N, Di Lisa F, Schmidt HHHW. On the clinical pharmacology of reactive oxygen species. Pharmacol Rev, 2020; 72(4):801–28; https://doi.org/10.1124/pr.120.019422

Cecchini R, Cecchini AL. SARS-CoV-2 infection pathogenesis is related to oxidative stress as a response to aggression. Med Hypotheses, 2020; 143:110102; https://doi.org/10.1016/j.mehy.2020.110102

Chen N, Zhou M, Dong X, Qu J, Gong F, Han Y, Qiu Y, Wang J, Liu Y, Wei Y, Xia J, Yu T, Zhang X, Zhang L. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. Lancet, 2020; 395(10223):507–13; https://doi.org/10.1016/S0140-6736(20)30211-7

Chernyak BV, Popova EN, Prikhodko AS, Grebenchikov OA, Zinovkina LA, Zinovkin RA. COVID-19 and oxidative stress. Biochem, 2020; 85(12):1543–53; https://doi.org/10.1134/S0006297920120068

Cheshire DR, Åberg A, Andersson GMK, Andrews G, Beaton HG, Birkinshaw TN, Boughton-Smith N, Connolly S, Cook TR, Cooper A, Cooper SL, Cox D, Dixon J, Gensmantel N, Hamley PJ, Harrison R, Hartopp P, Käck H, Leeson PD, Luker T, Mete A, Millichip I, Nicholls DJ, Pimm AD, St-Gallay SA, Wallace AV. The discovery of novel, potent and highly selective inhibitors of inducible nitric oxide synthase (iNOS). Bioorg Med Chem Lett, 2011; 21(8):2468–71; https://doi.org/10.1016/j.bmcl.2011.02.061

Cinelli MA, Reidl CT, Li H, Chreifi G, Poulos TL, Silverman RB. First contact: 7-phenyl-2-aminoquinolines, potent and selective neuronal nitric oxide synthase inhibitors that target an isoform-specific aspartate. J Med Chem, 2020; 63(9):4528–54; https://doi.org/10.1021/acs.jmedchem.9b01573

Cingolani G, Panella A, Perrone MG, Vitale P, Di Mauro G, Fortuna CG, Armen RS, Ferorelli S, Smith WL, Scilimati A. Structural basis for selective inhibition of cyclooxygenase-1 (COX-1) by diarylisoxazoles mofezolac and 3-(5-chlorofuran-2-yl)-5-methyl-4-phenylisoxazole (P6). Eur J Med Chem, 2017; 138:661–8; https://doi.org/10.1016/j.ejmech.2017.06.045

Connors JM, Levy JH. COVID-19 and its implications for thrombosis and anticoagulation. Blood, 2020; 135(23):2033–40; https://doi.org/10.1182/blood.2020006000

Dallakyan S, Olson AJ. Small-molecule library screening by docking with PyRx. Methods Mol Biol, 2015; 1263:243–50; https://doi.org/10.1007/978-1-4939-2269-7_19

Damiani S, Fiorentino M, De Palma A, Foschini MP, Lazzarotto T, Gabrielli L, Viale PL, Attard L, Riefolo M, D'Errico A. Pathological post-mortem findings in lungs infected with SARS-CoV-2. J Pathol, 2021; 253(1):31–40; https://doi.org/10.1002/path.5549

Delgado-Roche L, Mesta F. Oxidative stress as key player in severe acute respiratory syndrome coronavirus (SARS-CoV) infection. Arch Med Res, 2020; 51(5):384–7; https://doi.org/10.1016/j.arcmed.2020.04.019

Derouiche S. Oxidative stress associated with SARS-Cov-2 (COVID-19) increases the severity of the lung disease - a systematic review. J Infect Dis Epidemiol, 2020; 6:121; https://doi.org/10.23937/2474-3658/1510121

Dharajiya D, Pagi N, Jasani H, Patel P. Antimicrobial activity and phytochemical screening of *Aloe vera* (Aloe barbadensis Miller). Int J Curr Microbiol Appl Sci, 2017; 6(3):2152–62; https://doi.org/10.20546/ijcmas.2017.603.246

Ekor M. The growing use of herbal medicines: issues relating to adverse reactions and challenges in monitoring safety. Front Pharmacol, 2014; 4:177; https://doi.org/10.3389/fphar.2013.00177

Ewer KJ, Barrett JR, Belij-Rammerstorfer S, Sharpe H, Makinson R, Morter R, Flaxman A, Wright D, Bellamy D, Bittaye M, Dold C, Provine NM, Aboagye J, Fowler J, Silk SE, Alderson J, Aley PK, Angus B, Berrie E, Bibi S, Cicconi P, Clutterbuck EA, Chelysheva I, Folegatti PM, Fuskova M, Green CM, Jenkin D, Kerridge S, Lawrie A, Minassian AM, Moore M, Mujadidi Y, Plested E, Poulton I, Ramasamy NM, Robinson H, Song R, Snape MD, Tarrant R, Voysey M, Watson MEE, Douglas AD, Hill AVS, Gilbert SC, Pollard AJ, Lambe T, Oxford COVID Vaccine Trial Group. T cell and antibody responses induced by a single dose of ChAdOx1 nCoV-19 (AZD1222) vaccine in a phase 1/2 clinical trial. Nat Med, 2021; 27(2):270–8; https://doi.org/10.1038/s41591-020-01194-5

Fang W, Jiang J, Su L, Shu T, Liu H, Lai S, Ghiladi RA, Wang J. The role of NO in COVID-19 and potential therapeutic strategies. Free Radic Biol Med, 2021; 163:153–62; https://doi.org/10.1016/j.freeradbiomed.2020.12.008

Funk CD, Ardakani A. A novel strategy to mitigate the hyperinflammatory response to COVID-19 by targeting leukotrienes. Front Pharmacol, 2020; 11:1214; https://doi.org/10.3389/fphar.2020.01214

Giannakoudakis DA, Hosseini-Bandegharaei A, Tsafrakidou P, Triantafyllidis KS, Kornaros M, Anastopoulos I. *Aloe vera* waste biomass-based adsorbents for the removal of aquatic pollutants: a review. J Environ Manage, 2018; 227:354–64; https://doi.org/10.1016/j.jenvman.2018.08.064

Gilbert NC, Gerstmeier J, Schexnaydre EE, Börner F, Garscha U, Neau DB, Werz O, Newcomer ME. Structural and mechanistic insights into 5-lipoxygenase inhibition by natural products. Nat Chem Biol, 2020; 16(7):783–90; https://doi.org/10.1038/s41589-020-0544-7

Guan SP, Seet RCS, Kennedy BK. Does eNOS derived nitric oxide protect the young from severe COVID-19 complications? Ageing Res Rev, 2020; 64:101201; https://doi.org/10.1016/j.arr.2020.101201

Häfner A-K, Kahnt AS, Steinhilber D. Beyond leukotriene formation—the noncanonical functions of 5-lipoxygenase. Prostaglandins Other Lipid Mediat, 2019; 142:24–32; https://doi.org/10.1016/j.prostaglandins.2019.03.003

Halpin DMG, Criner GJ, Papi A, Singh D, Anzueto A, Martinez FJ, Agusti AA, Vogelmeier CF. Global initiative for the diagnosis, management, and prevention of chronic obstructive lung disease. The 2020 GOLD science committee report on COVID-19 and chronic obstructive pulmonary disease. Am J Respir Crit Care Med, 2020; 203(1):24–36; https://doi.org/10.1164/rccm.202009-3533SO

Harvey WT, Carabelli AM, Jackson B, Gupta RK, Thomson EC, Harrison EM, Ludden C, Reeve R, Rambaut A, COVID-19 Genomics UK (COG-UK) Consortium, Peacock SJ, Robertson DL. SARS-CoV-2 variants, spike mutations and immune escape. Nat Rev Microbiol, 2021; 19(7):409–24; https://doi.org/10.1038/s41579-021-00573-0

Hęś M, Dziedzic K, Górecka D, Jędrusek-Golińska A, Gujska E. *Aloe vera* (L.) Webb.: natural sources of antioxidants - a review. Plant Foods Hum Nutr, 2019; 74(3):255–65; https://doi.org/10.1007/s11130-019-00747-5

Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, Zhang L, Fan G, Xu J, Gu X, Cheng Z, Yu T, Xia J, Wei Y, Wu W, Xie X, Yin W, Li H, Liu M, Xiao Y, Gao H, Guo L, Xie J, Wang G, Jiang R, Gao Z, Jin Q, Wang J, Cao B. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet, 2020; 395(10223):497–506; https://doi.org/10.1016/S0140-6736(20)30183-5

Kazybay B, Ahmad A, Mu C, Mengdesh D, Xie Y. Omicron N501Y mutation among SARS-CoV-2 lineages: insilico analysis of potent binding to tyrosine kinase and hypothetical repurposed medicine. Travel Med Infect Dis, 2022; 45:102242; https://doi.org/10.1016/j.tmaid.2021.102242

Komaravelli N, Casola A. Respiratory viral infections and subversion of cellular antioxidant defenses. J Pharmacogenomics Pharmacoproteomics, 2014; 5(4):1000141; https://doi.org/10.4172/2153-0645.1000141

Kumar S, Yadav M, Yadav A, Yadav JP. Impact of spatial and climatic conditions on phytochemical diversity and in vitro antioxidant activity of Indian *Aloe vera* (L.) Burm.f. South Afr J Bot, 2017; 111:50–9; https://doi.org/10.1016/j.sajb.2017.03.012

Kumar S, Thambiraja TS, Karuppanan K, Subramaniam G. Omicron and delta variant of SARS-CoV-2: a comparative computational study of spike protein. J Med Virol, 2021; 1–9; https://doi.org/10.1002/jmv.27526

Li H, Evenson RJ, Chreifi G, Silverman RB, Poulos TL. Structural basis for isoform selective nitric oxide synthase inhibition by thiophene-2-carboximidamides. Biochemistry, 2018; 57(44):6319–25; https://doi.org/10.1021/acs.biochem.8b00895

Lin H-C, Lin T-H, Wu M-Y, Chiu Y-C, Tang C-H, Hour M-J, Liou H-C, Tu H-J, Yang R-S, Fu W-M. 5-Lipoxygenase inhibitors attenuate TNF-α-induced inflammation in human synovial fibroblasts. PLoS One, 2014; 9(9):e107890; https://doi.org/10.1371/journal.pone.0107890

Lippi G, Mattiuzzi C. Hemoglobin value may be decreased in patients with severe coronavirus disease 2019. Hematol Transfus Cell Ther, 2020; 42(2):116–7; https://doi.org/10.1016/j.htct.2020.03.001

Liu Y, Liu J, Plante KS, Plante JA, Xie X, Zhang X, Ku Z, An Z, Scharton D, Schindewolf C, Widen SG, Menachery VD, Shi PY, Weaver SC. The N501Y spike substitution enhances SARS-CoV-2 infection and transmission. Nature, 2021; https://doi.org/10.1038/s41586-021-04245-0

Liu Z, Ren Z, Zhang J, Chuang CC, Kandaswamy E, Zhou T, Zuo L. Role of ROS and nutritional antioxidants in human diseases. Front Physiol, 2018; 9:477; https://doi.org/10.3389/fphys.2018.00477

López A, de Tangil MS, Vega-Orellana O, Ramírez AS, Rico M. Phenolic constituents, antioxidant and preliminary antimycoplasmic activities of leaf skin and flowers of *Aloe vera* (L.) Burm. f. (syn. A. barbadensis Mill.) from the Canary Islands (Spain). Molecules, 2013; 18(5):4942–54; https://doi.org/10.3390/molecules18054942.

Mantzarlis K, Tsolaki V, Zakynthinos E. Role of oxidative stress and mitochondrial dysfunction in sepsis and potential therapies. Oxid Med Cell Longev, 2017; 2017:5985209; https://doi.org/10.1155/2017/5985209

McCarthy MK, Weinberg JB. Eicosanoids and respiratory viral infection: coordinators of inflammation and potential therapeutic targets. Mediators Inflamm, 2012; 2012:236345; https://doi.org/10.1155/2012/236345

Mengist HM, Kombe Kombe AJ, Mekonnen D, Abebaw A, Getachew M, Jin T. Mutations of SARS-CoV-2 spike protein: implications on immune evasion and vaccine-induced immunity. Semin Immunol, 2021; 55:101533; https://doi.org/10.1016/j.smim.2021.101533

Merad M, Martin JC. Pathological inflammation in patients with COVID-19: a key role for monocytes and macrophages. Nat Rev Immunol, 2020; 20(6):355–62; https://doi.org/10.1038/s41577-020-0331-4

Moriyama M, Moriyama H, Uda J, Kubo H, Nakajima Y, Goto A, Akaki J, Yoshida I, Matsuoka N, Hayakawa T. Beneficial effects of the genus aloe on wound healing, cell proliferation, and differentiation of epidermal keratinocytes. PLoS One, 2016; 11(10):1–15; https://doi.org/10.1371/journal.pone.0164799

Mousavi SS, Karami A, Haghighi TM, Tumilaar SG, Fatimawali, Idroes R, Mahmud S, Celik I, Ağagündüz D, Tallei TE, Emran TB, Capasso R. In silico evaluation of Iranian medicinal plant phytoconstituents as inhibitors against main protease and the receptor-binding domain of SARS-CoV-2. Molecules, 2021; 26(18):5724; https://doi.org/10.3390/molecules26185724

Muthukumaran P, Divya R, Indhumathi E, Keerthika C. Total phenolic and flavonoid content of membrane processed *Aloe vera* extract: a comparative study. Int Food Res J, 2018; 25(4):1450–6; http://ifrj.upm.edu.my/25%20(04)%202018/(17).pdf

Nalimu F, Oloro J, Kahwa I, Ogwang PE. Review on the phytochemistry and toxicological profiles of *Aloe vera* and *Aloe ferox*. Futur J Pharm Sci, 2021; 7(1):145; https://doi.org/10.1186/s43094-021-00296-2

Nur S, Aisyah AN, Lukitaningsih E, Rumiyati, Juhardi RI, Andirah R, Hajar AS. Evaluation of antioxidant and cytotoxic effect against cancer cells line of *Angiopteris* ferox Copel tuber and its compounds by LC-MS analysis. J Appl Pharm Sci, 2021; 11(8):54–61; https://doi.org/10.7324/JAPS.2021.110808

O'Flaherty JT, Hu Y, Wooten RE, Horita DA, Samuel MP, Thomas MJ, Sun H, Edwards IJ. 15-Lipoxygenase metabolites of docosahexaenoic acid inhibit prostate cancer cell proliferation and survival. PLoS One, 2012; 7(9):e45480; https://doi.org/10.1371/journal.pone.0045480

Omotuyi O, Olubiyi O, Nash O, Afolabi E, Oyinloye B, Fatumo S, Femi-Oyewo M, Bogoro S. SARS-CoV-2 Omicron spike glycoprotein receptor binding domain exhibits super-binder ability with ACE2 but not convalescent monoclonal antibody. Comput Biol Med, 2022; 142:105226; https://doi.org/10.1016/j.compbiomed.2022.105226

Ortega JT, Jastrzebska B, Rangel HR. Omicron SARS-CoV-2 variant spike protein shows an increased affinity to the human ACE2 receptor: an in silico analysis. Pathog, 2022; 11(1):45; https://doi.org/10.3390/pathogens11010045

Ortega JT, Pujol FH, Jastrzebska B, Rangel HR. Mutations in the SARS-CoV-2 spike protein modulate the virus affinity to the human ACE2 receptor, an in silico analysis. EXCLI J, 2021; 20:585–600; https://doi.org/10.17179/excli2021-3471

Ottolenghi S, Zulueta A, Caretti A. Iron and sphingolipids as common players of (Mal) adaptation to hypoxia in pulmonary diseases. Int J Mol Sci, 2020;21(1):307; https://doi.org/10.3390/ijms21010307

Pieretti JC, Rubilar O, Weller RB, Tortella GR, Seabra AB. Nitric oxide (NO) and nanoparticles – potential small tools for the war against COVID-19 and other human coronavirus infections. Virus Res, 2021; 291:198202; https://doi.org/10.1016/j.virusres.2020.198202

Pizzino G, Irrera N, Cucinotta M, Pallio G, Mannino F, Arcoraci V, Squadrito F, Altavilla D, Bitto A. Oxidative stress: harms and benefits for human health. Oxid Med Cell Longev, 2017; 2017:8416763; https://doi.org/10.1155/2017/8416763

Poeckel D, Funk CD. The 5-lipoxygenase/leukotriene pathway in preclinical models of cardiovascular disease. Cardiovasc Res, 2010; 86(2):243–53; https://doi.org/10.1093/cvr/cvq016

Raad B, Ali S, Rehman K, Akhtar N, Ullah B, Wali S. Phytochemical screening and biological activities of *Aloe vera* (L.) Burm. F. Pure Appl Biol, 2020; 10(2):360–7; http://dx.doi.org/10.19045/bspab.2021.100039

Rådmark O, Werz O, Steinhilber D, Samuelsson B. 5-Lipoxygenase, a key enzyme for leukotriene biosynthesis in health and disease. Biochim Biophys Acta Mol Cell Biol Lipids, 2015; 1851(4):331–9; https://doi.org/10.1016/j.bbalip.2014.08.012

Ravichandran S, Coyle EM, Klenow L, Tang J, Grubbs G, Lie S, Wang T, Golding H, Khurana S. Antibody signature induced by SARS-CoV-2 spike protein immunogens in rabbits. Sci Transl Med, 2020; 12(550):eabc3539; https://doi.org/10.1126/scitranslmed.abc3539

Sammar M, Abu-Farich B, Rayan I, Falah M, Rayan A. Correlation between cytotoxicity in cancer cells and free radical-scavenging activity: in vitro evaluation of 57 medicinal and edible plant extracts. Oncol Lett, 2019; 18(6):6563–71; https://doi.org/10.3892/ol.2019.11054

Sánchez M, González-Burgos E, Iglesias I, Gómez-Serranillos MP. Pharmacological update properties of *Aloe Vera* and its major active constituents. Molecules, 2020; 25(6):1324; https://doi.org/10.3390/molecules25061324

Sies H, Jones DP. Reactive oxygen species (ROS) as pleiotropic physiological signalling agents. Nat Rev Mol Cell Biol, 2020; 21(7):363–83; https://doi.org/10.1038/s41580-020-0230-3

Stuehr DJ, Vasquez-Vivar J. Nitric oxide synthases-from genes to function. Nitric oxide Biol Chem, 2017; 63:29; https://doi.org/10.1016/j.niox.2017.01.005

Tallei TE, Tumilaar SG, Niode NJ, Fatimawali, Kepel BJ, Idroes R, Effendi Y, Sakib SA, Emran TB. Potential of plant bioactive compounds as SARS-CoV-2 main protease (Mpro) and spike (S) glycoprotein inhibitors: a molecular docking study. Scientifica (Cairo), 2020; 2020:6307457; https://doi.org/10.1155/2020/6307457

Terpos E, Ntanasis-Stathopoulos I, Elalamy I, Kastritis E, Sergentanis TN, Politou M, Psaltopoulou T, Gerotziafas G, Dimopoulos MA. Hematological findings and complications of COVID-19. Am J Hematol, 2020; 95(7):834–47; https://doi.org/10.1002/ajh.25829

Uddin MJ, Xu S, Crews BC, Aleem AM, Ghebreselasie K, Banerjee S, Marnett LJ. Harmaline analogs as substrate-selective cyclooxygenase-2 inhibitors. ACS Med Chem Lett, 2020; 11(10):1881–5; https://doi.org/10.1021/acsmedchemlett.9b00555

Zhang H, Lei Y, Yuan P, Li L, Luo C, Gao R, Tian J, Feng Z, Nice EC, Sun J. ROS-mediated autophagy induced by dysregulation of lipid metabolism plays a protective role in colorectal cancer cells treated with gambogic acid. PLoS One, 2014; 9(5):1–14; https://doi.org/10.1371/journal.pone.0096418

Zhang H, Zhou P, Wei Y, Yue H, Wang Y, Hu M, Zhang S, Cao T, Yang C, Li M, Guo G, Chen X, Chen Y, Lei M, Liu H, Zhao J, Peng P, Wang CY, Du R. Histopathologic changes and SARS-CoV-2 immunostaining in the lung of a patient with COVID-19. Ann Intern Med, 2020; 172(9):629–32; https://doi.org/10.7326/M20-0533

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SUPPLEMENTARY MATERIAL

Supplementary data can be downloaded from the journal's website

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