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Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention

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Abstract

The search for effective therapeutics against COVID-19 remains imperative, and natural compounds have emerged as promising candidates. Our study explores the potential of bioactive phytochemicals from the traditional Siddha formulation MilagaiKudineer as inhibitors against key target proteins of the SARS-CoV-2 virus. Through in-silico docking analyses, the interactions of phytochemicals from Cuminum cyminum, Curcuma longa, and Capsicum annuum with the receptor-binding domain of the SARS-CoV-2 spike glycoprotein (PDB ID: 6VSB), the SARS-CoV2 RNA-dependent RNA polymerase (PDB ID: 6NUR), and the main protease, 3CL pro (PDB ID: 6LU7) were examined. Notable compounds such as Curcumin, Quercetin, Capsaicin, and Ascorbic acid demonstrated significant binding affinities towards these viral targets, suggesting mechanisms by which these phytochemicals may disrupt viral entry and replication. Our findings also highlight the potential of compounds like Carvacrol, Cuminaldehyde, Linalool, and Dihydrocapsaicin in mediating antiviral effects by interfacing with key amino acid residues of the spike glycoprotein. These interactions are indicative of their capacity to hinder the virus-host cellbinding process. Moreover, the interaction of select phytochemicals with the SARS-CoV2 RNA-dependent RNA polymerase and the 3CLpro enzyme suggests a possible inhibitory effect on viral replication. Given the promising interactions observed, these phytochemicals warrant further investigation through in vitro and in vivo studies to validate their antiviral efficacy against COVID-19. This research underscores the importance of exploring traditional medicinal formulations for potential therapeutic agents in the fight against emerging infectious diseases.

Keywords: COVID-19, Siddha formulation, MilagaiKudineer, bioactive compounds, molecular docking, spike glycoprotein, receptor-binding domain, main protease, SARS-CoV2 RNA-dependent RNA polymerase

Introduction

The Siddha system of medicine, deeply rooted in South India, particularly in Tamil Nadu, has garnered renown for its holistic approach to healthcare. During challenging times such as the outbreaks of diseases like Dengue and the COVID-19 pandemic, Siddha medicines have played a vital role in saving lives. Among these remedies, MilagaiKudineer, a formulation attributed to the Tiruvanamalai Guru Parambariyam, has emerged as particularly noteworthy during the COVID-19 crisis. This herbal concoction has demonstrated

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significant efficacy in combating the coronavirus, making it a crucial asset in the fight against the pandemic. The COVID-19 pandemic caused by the novel coronavirus SARS-CoV-2 has led to significant global health and economic challenges and underscores the need for effective antiviral interventions. The search for effective therapeutic agents to combat this virus is of paramount importance. The spike glycoprotein of SARS-CoV-2 plays a crucial role in viral entry by binding to host cell receptors. Targeting the receptor-binding domain of the spike protein presents a promising approach for therapeutic development. Siddha medicine offers a repertoire of natural compounds with potential antiviral properties, including those found in MilagaiKudineer. Traditional medicinal systems, such as Siddha medicine, offer a repertoire of natural compounds with potential pharmacological activities against viral infections.MilagaiKudineer is a traditional Siddha formulation comprising herbs like Cuminum cyminum, Curcuma longa, and Capsicum annuum, which are known for their bioactive constituents with diverse medicinalproperties. The main protease of SARS-CoV-2, 3-chymotrypsin-like protease (3CLpro), plays a crucial role in viral replication by cleaving the polyproteins into functional non-structural proteins.

Targeting 3CL pro presents a promising strategy for developing antiviral therapeutics. Siddha medicine, an ancient traditional medicinal system, offers potential sources of bioactive compounds for combating viral infections. MilagaiKudineer, a Siddha formulation, comprises various herbs known for their pharmacological properties. This study was envisaged with objectives such as i)to assess the inhibitory potential of bioactive compounds from MilagaiKudineer against the COVID-19 spike glycoprotein receptor-binding domain through computational docking analysis, ii)to investigate the potential of bioactive compounds derived from MilagaiKudineer against the SARS-CoV2 RNA-dependent RNA polymerase through computational methods and iii) to investigate the inhibitory potential of bioactive compounds from MilagaiKudineer against the SARS-CoV2 main protease, 3CL pro, through computational docking analysis.

Materials and Methods

Preparation of MilagaiKudineer:

Milagaikudineer, a kudineer formulation of Tiruvanamalai Guru Parambariyam is routinely prepared as described (Asan, 2023). All the ingredients in the proportion given in Table 1 are taken, and 200 ml of water is added and boiled for 2 minutes. After filtration 50 ml dose can be given onlyin the morning for 3 consecutive days. For children, 5-25 ml can be given as per their age.

Table-1: MilagaiKudineer Ingredients

S. No	Tamilname	Englishname	Scientific name	Ratio
1.	Pachai Milagai	Green chilly	CapsicumannuumLinn	1 no.
2.	Seeragam	Cumin	CuminumcyminumLinn	100 mg
3.	Manjalthool	Turmeric powder	CurcumalongaLinn	100 mg
4.	Uppu	Commonsalt	Sodium Chloride (NaCl)	100 mg

Phytocompounds Selected for docking:

Totally11 compounds, 4 phytocompounds from *Cuminum cyminum* viz. Carvacrol Cuminaldehyde, Linalool, Coumaric acid (Ali, 2016), 4 from *Curcuma longa* viz. Curcumin, Quercetin, Sabinene, Cineol (Hewlings*et al.,* 2017; Zhang and Kitts, 2021) and 3 compounds, Dihydrocapsaicin, Capsaicin, Ascorbic acid from *capsicum annuum* (Hamed *et al.,* 2019) were selected for this study.

Protein targets selected for docking studies:

The three-dimensional structures of the SARS-CoV-2 viral spike glycoprotein receptor-binding domain (PDBID:6VSB),SARS-CoV2RNA-dependent RNApolymerase (PDB ID:6NUR)and SARS-CoV-2 viral main protease (3-chymotrypsin-like protease (3CL pro) (PDB ID: 6LU7) were retrieved from RCSB-PDB (Goodsell *et al.,*2020).

Preparation of the receptors:

The crystalline structure of the SARS-CoV-2 viral spike glycoprotein receptor-binding domain (PDB ID: 6VSB), SARS-CoV2 RNA-dependent RNA polymerase (PDB ID: 6NUR) and SARS- CoV-2 viral main protease (3-chymotrypsin-like protease (3CL pro) (PDB ID: 6LU7) were retrieved from RCSB-PDB and protein clean-up process was done and essential missing hydrogen atom were added. Watermolecules and cocrystallized ligands wereremoved. Different orientation of the lead molecules with respect to the target protein was evaluated by theAutodock program and the best dock pose was selected based on the interaction study analysis.

Ligand preparations:

All the 11 compounds, 4 phytocompounds from *Cuminum cyminum* viz. Carvacrol Cumin aldehyde, Linalool, Coumaric acid, 4 from *Curcuma longa* viz. Curcumin, Quercetin, Sabinene, Cineol and 3 compounds, Dihydrocapsaicin, Capsaicin, and Ascorbic acid from *capsicum annuum* were built using Chem Draw prof online tool version 12.0. Ligands prepared through geometry optimization method (MMFF94).

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Docking procedure:

Docking calculations were carried out for retrieved phytocomponents against all three target proteins. Essential hydrogen atoms, Kollman united atom type charges, and solvation parameters were added with the aid of AutoDock tools (Morris, Goodsell et al., 1998). Affinity (grid) maps of 25 Å grid points and 0.375 Å spacing were generated using the Autogrid program (Morris, Goodsell et al., 1998). AutoDock parameter set- and distance-dependent dielectric functionswere used in the calculation of the van der Waals and the electrostatic terms, respectively. Docking simulations were performed using the Lamarckian genetic algorithm (LGA) and the Solis & Wets local search method (Solis and Wets, 1981). The initial position, orientation, and torsions of the ligand molecules were set randomly. All rotatable torsions were released during docking. Each docking experiment was derived from 2 different runs that were set to terminate after a maximum of 250000 energy evaluations. The population size was set to 150. During the search, a translational step of 0.2 Å, and quaternion and torsion steps of 5 were applied.

Results and Discussion

Preparation of phyto chemicals from MilagaiKudineer:

Totally11 compounds were selected for this study and their structures are shown in Figure-1. These were four phytocompounds from *Cuminum cyminum* viz. Carvacrol Cumin aldehyde, Linalool,Coumaric acid, 4from *Curcumalonga* viz.Curcumin,Quercetin,Sabinene,Cineoland 3 compounds, Dihydrocapsaicin, Capsaicin, and Ascorbic acid from *capsicum annuum*.

Docking studies of phytochemicals from MilagaiKudineer against SARS-CoV-2spike glycoprotein receptor-binding domain:

The Summary of the molecular docking studies of compounds against SARS-CoV-2 spike glycoprotein receptor-binding – PDB 6VSB is shown in Table-2. The docked pose of the top molecule Carvacrol with SARS-CoV-2 spike glycoprotein receptor is shown in Figure-2.

Outof11compounds, the lead molecules such as Carvacrol,Cuminald eyde,Linalool, Curcumin, Quercetin, Cineol, Dihydrocapsaicin, Capsaicin and ascorbic acid reveals a maximum of 3-5 interactions with the core active amino acid residues present on the target SARS-CoV-2 spike glycoprotein receptor-binding domain. the interaction between lead molecules and with target protein are shown in Table-3

Based on the results of the computational analysis it was concluded that the bio-active compounds such as Carvacrol, Cuminaldeyde, Linalool, Curcumin, Quercetin, Cineol,

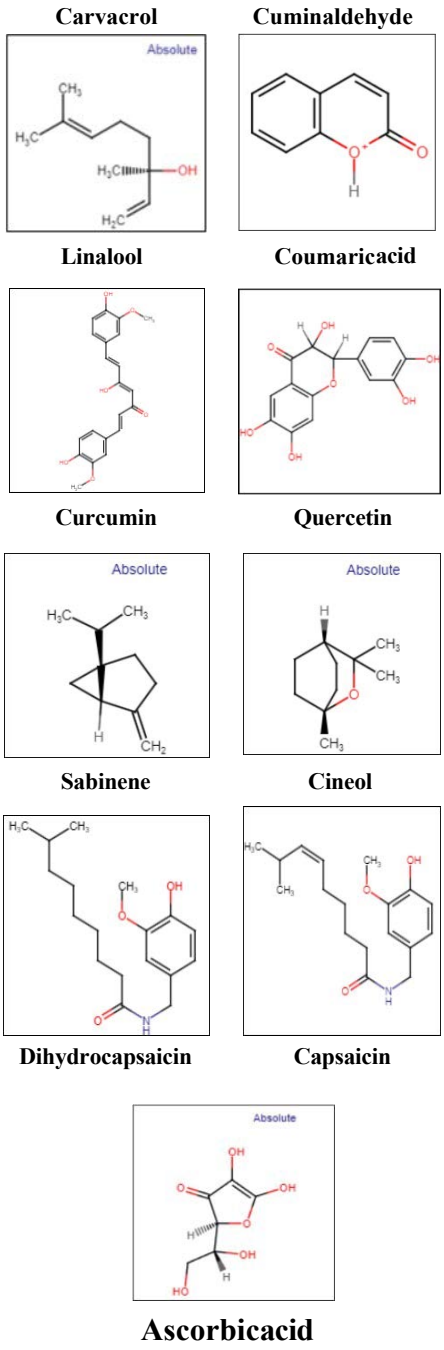

Figure1: 2 D Structure of Phytocomponents from MilagaiKudineer

Dihydrocapsaicin, Capsaicin and ascorbic acid present in the Siddha formulation MilagaiKudineer reveals significant binding against the target proteinSARS-CoV-2 spike glycoprotein receptor-binding domain.

Docking studies of phytochemicals from MilagaiKudineer against SARS-CoV-2 RNA-dependent RNA polymerase:

The Summary of the molecular docking studies of compounds against SARS-CoV-2 RNA- dependent RNA

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27

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Table 2: Summary of the molecular docking studies of compounds against SARS-CoV-2 spike glycoprotein receptor-binding – PDB 6VSB

Compounds	Est.FreeEnergy ofBinding	Est.Inhibition Constant,Ki	Electrostatic Energy	TotalIntermole c.Energy	Interact.Surface
Carvacrol	-4.51kcal/mol	495.77uM	-0.05kcal/mol	-5.44kcal/mol	533.228
Cuminaldehyde	-4.15kcal/mol	908.13uM	-0.04kcal/mol	-4.75kcal/mol	435.425
Linalool	-3.96kcal/mol	1.25mM	-0.16kcal/mol	-5.37kcal/mol	486.187
Coumaricacid	-4.31kcal/mol	697.38uM	-0.02kcal/mol	-4.31kcal/mol	400.076
Curcumin	-6.06kcal/mol	36.34uM	-0.15kcal/mol	-7.43kcal/mol	818.251
Quercetin	-6.41kcal/mol	19.91uM	-0.80kcal/mol	-5.79kcal/mol	546.743
Sabinene	-4.39kcal/mol	609.66uM	-0.01kcal/mol	-4.68kcal/mol	435.91
Cineol	-4.34kcal/mol	653.49uM	-0.02kcal/mol	-4.34kcal/mol	431.703
Dihydrocapsaicin	-5.43kcal/mol	104.67uM	-0.28kcal/mol	-7.07kcal/mol	662.568
Capsaicin	-5.94kcal/mol	44.13uM	-0.23kcal/mol	-7.18kcal/mol	661.061
Ascorbicacid	-5.02kcal/mol	210.36uM	-0.35kcal/mol	-4.15kcal/mol	368.451

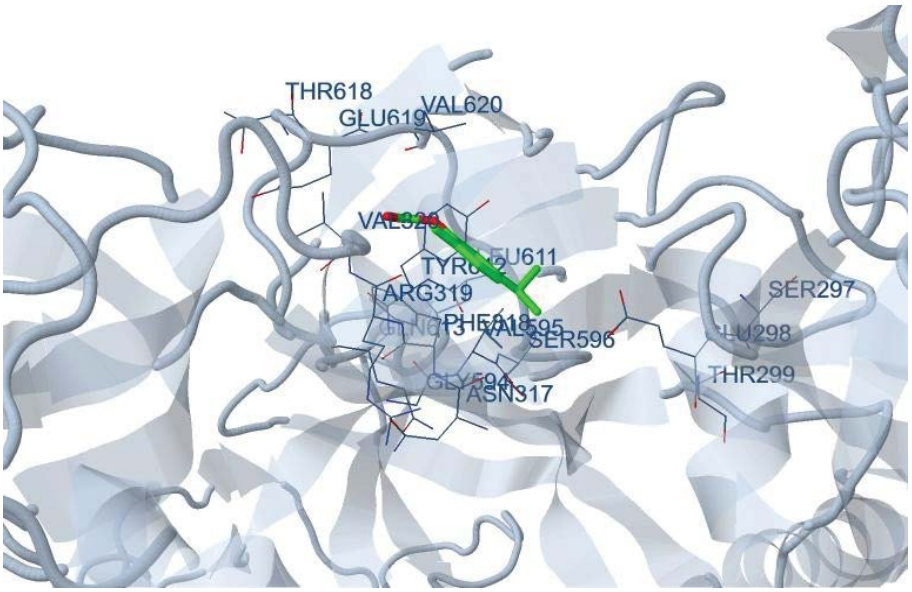

Figure 2: DockingPoseofCarvacrolwithSARS-Co 2spikeglycoproteinreceptor binding Domain

polymerase (PDB)-6NUR is shown in Table-4. The docked pose of the top molecule Carvacrol with SARS-CoV-2 RNA-dependent RNA polymerase is shown in Figure-3.

A total of 11 bioactive lead compounds were retrieved from the Siddha formulation Milagaikudineer and were subject edtoin-silicoinvestigationofwhichleadcompoundssuc hasLinalool, Coumaric acid, Curcumin, Quercetin, Carvacrol, Cineol, Dihydrocapsaicin and Capsaicin reveals maximum interactions (2-3) with the binding sites on the core active amino acid residues present on the target receptor RdRp. The interaction between lead molecules and with target protein is shown in Table-5.

Based on the results of the computational analysis it was concluded that compounds such as Linalool, Coumaric acid, Curcumin, Quercetin, Carvacrol, Cineol, Dihydrocapsaicin and Capsaicin present in the Siddha formulation Milagaikudineer revels significant binding efficacy against active aminoacid present on the target enzyme and it was concluded that these compounds exert promising inhibiting against RdRp enzyme and thereby halt the viral replication.

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27

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Table 3: Amino acid Residue Interaction of Lead against SARS-CoV-2spikeglycoprotein receptor-binding – PDB 6VSB

Compounds	Interactions	AminoacidResidues								
Carvacrol	4	298	318	595	612	619				
		GLU	PHE	VAL	TYR	GLU				
Cuminaldehyde	3	318	320	612	619	620				
		PHE	VAL	TYR	GLU	VAL				
Linalool	5	295	298	318	595	610	612	620		
		PRO	GLU	PHE	VAL	VAL	TYR	VAL		
Coumaricacid	2	318	320	612	619					
		PHE	VAL	TYR	GLU					
Curcumin	5	274	291	292	298	316	318	595	612	619
		THR	CYS	ALA	GLU	SER	PHE	VAL	TYR	GLU
Quercetin	3	318	319	321	595	612				
		PHE	ARG	GLN	VAL	TYR				
Sabinene	2	318	320	612	619					
		PHE	VAL	TYR	GLU					
Cineol	3	318	612	619	620					
		PHE	TYR	GLU	VAL					
Dihydrocapsaicin	5	295	298	316	318	595	612	620		
		PRO	GLU	SER	PHE	VAL	TYR	VAL		
Capsaicin	5	298	316	318	319	595	612	619	620	
		GLU	SER	PHE	ARG	VAL	TYR	GLU	VAL	
Ascorbicacid	4	298	316	595	612	620				
		GLU	SER	VAL	TYR	VAL				

Table 4: Summary of the molecular docking studies of compounds against SARS-CoV-2- RNA dependent RNA polymerase PDB-6NUR

Compounds	Est.FreeEnergy ofBinding	Est.Inhibition Constant,Ki	Electrostatic Energy	TotalIntermole c.Energy	Interact.Surface
Carvacrol	-5.05kcal/mol	198.16uM	-0.16kcal/mol	-5.94kcal/mol	556.056
Cuminaldehyde	-4.70kcal/mol	357.69uM	-0.07kcal/mol	-5.30kcal/mol	417.791
Linalool	-4.91kcal/mol	251.80uM	-0.04kcal/mol	-6.35kcal/mol	473.4
Curcumin	-4.56kcal/mol	457.94uM	-0.01kcal/mol	-4.85kcal/mol	455.07
Quercetin	-7.13kcal/mol	5.93uM	-1.49kcal/mol	-6.76kcal/mol	556.705
Sabinene	-4.66kcal/mol	384.15uM	-0.01kcal/mol	-4.96kcal/mol	421.055
Cineol	-5.27kcal/mol	136.00uM	-0.09kcal/mol	-5.27kcal/mol	448.845
Dihydrocapsaicin	-5.89kcal/mol	48.44uM	-0.24kcal/mol	-8.09kcal/mol	753.146
Capsaicin	-6.18kcal/mol	29.56uM	-0.30kcal/mol	-7.69kcal/mol	718.291
Ascorbicacid	-5.63kcal/mol	74.94uM	-0.95kcal/mol	-4.64kcal/mol	387.778

Figure 3: Docking Pose of Carvacrol with SARS-CoV-2RNA-dependent RNA polymerase

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27

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Table 5: Amino acid Residue Interaction of Lead against SARS-CoV-2 RNA-dependent RNA Polymerase

Compounds	Interaction s	AminoacidResidues						
Carvacrol	3	618	619	760	761	800	811	
		ASP	TYR	ASP	ASP	TRP	GLU	
Cuminaldehyde	1	761	800	811				
		ASP	TRP	GLU				
Linalool	2	617	619	695	760	761	800	811
		TRP	TYR	ASN	ASP	ASP	TRP	GLU
Coumaricacid	2	617	618	619	695	760		
		TRP	ASP	TYR	ASN	ASP		
Curcumin	2	618	761	800	811			
		ASP	ASP	TRP	GLU			
Quercetin	3	617	618	619	621	623	760	761
		TRP	ASP	TYR	LYS	ASP	ASP	ASP
Sabinene	1	761	800					
		ASP	TRP					
Cineol	2	618	761	800	811			
		ASP	ASP	TRP	GLU			
Dihydrocapsaici n	3	618	619	622	623	760	761	800
		ASP	TYR	CYS	ASP	ASP	ASP	TRP
Capsaicin	3	618	760	761	800	814		
		ASP	ASP	ASP	TRP	SER		
Ascorbicacid	1	618	621	622	623	798		
		ASP	LYS	CYS	ASP	LYS		

Docking studies of phytochemicals from MilagaiKudineer against SARS-CoV-2Main protease PDB 6LU7:

The Summary of the molecular docking studies of compounds against SARS-CoV-2 Main protease PDB 6LU7 is shown in Table-6. The docked pose of the top molecule Carvacrol with SARS-CoV-2 spike glycoprotein receptor is shown in Figure-4.

Out of eleven compounds, the lead compounds such as Curcumin, Quercetin, Sabinene, Cineol, Dihydrocapsaicin and Capsaicin reveal a maximum of 3 to 5 interactions with the core active amino acid residues present on the target 3CLpro.The interaction between the lead molecule and with target protein is shown in Table-7.

Based on the results of the computational analysis it was concluded that the bio-active compounds such as Curcumin, Quercetin, Sabinene, Cineol, Dihydrocapsaicin and Capsaicin present in the Siddha formulation reveal significant binding against the target protein3CLpro thereby it was concluded that these compounds may exert promising inhibiting against 3CLpro enzyme and hereby halt the formation of 16 non-structural proteins (nsp1-nsp16) that are highly essential for viral replication and thereby prevents the viral survival in the host environment.

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27

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Table 6: Summary of the molecular docking studies of compounds against SARS-CoV-2 Main protease PDB 6LU7

Compounds	Est.FreeEnergy ofBinding	Est.Inhibition Constant,Ki	Electrostatic Energy	TotalIntermole c.Energy	Interact.Surface
Carvacrol	-5.74kcal/mol	61.54uM	-0.01kcal/mol	-6.66kcal/mol	542.922
Cuminaldehyde	-4.69kcal/mol	365.37uM	-0.11kcal/mol	-5.28kcal/mol	469.209
Linalool	-4.87kcal/mol	268.82uM	-0.03kcal/mol	-6.15kcal/mol	467.784
Coumaricacid	-4.62kcal/mol	412.16uM	-0.06kcal/mol	-4.62kcal/mol	396.462
Curcumin	-7.68kcal/mol	2.36uM	-0.05kcal/mol	-9.01kcal/mol	858.356
Quercetin	-6.92kcal/mol	8.45uM	-0.15kcal/mol	-6.19kcal/mol	610.849
Sabinene	-5.35kcal/mol	119.11uM	-0.24kcal/mol	-5.65kcal/mol	420.699
Cineol	-5.82kcal/mol	54.07uM	-0.02kcal/mol	-5.82kcal/mol	421.586
Dihydrocapsaicin	-7.02kcal/mol	7.17uM	-0.14kcal/mol	-9.00kcal/mol	779.094
Capsaicin	-7.43kcal/mol	3.58uM	-0.08kcal/mol	-8.86kcal/mol	728.808
Ascorbicacid	-5.30kcal/mol	130.13uM	-0.18kcal/mol	-4.36kcal/mol	430.556

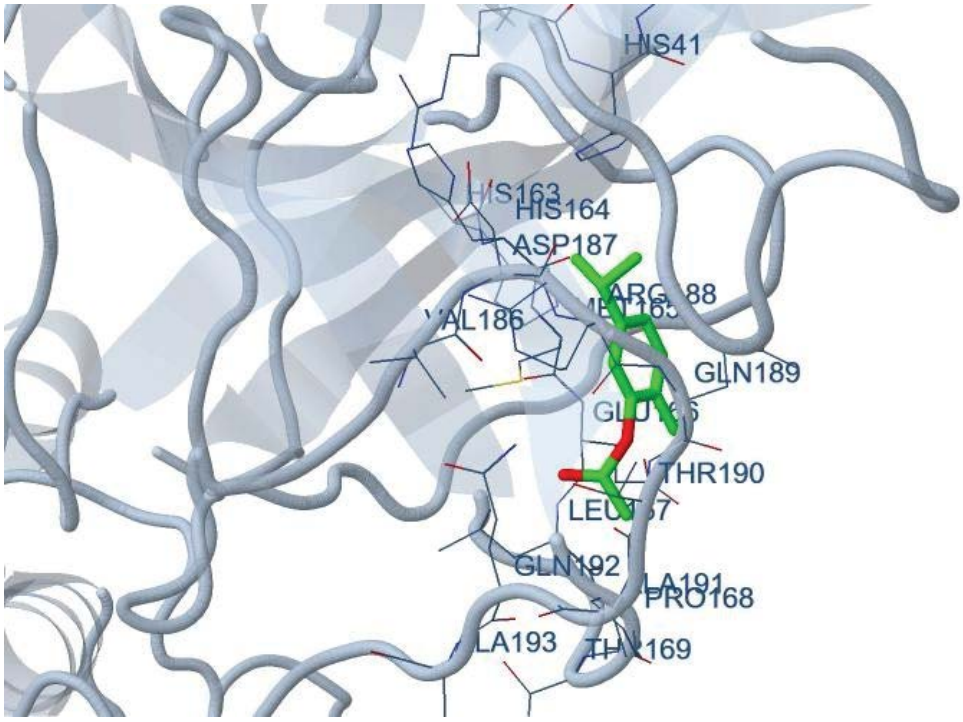

Figure 4: DockingPoseofCarvacrolwithSARS-CoV-2MainproteasePDB6LU7

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27

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Table-7: Amino acid ResidueInteraction of Lead againstSARS-CoV-2 Main proteasePDB 6LU7

Compounds	Interactions	AminoacidResidues											
Carvacrol	2	41	165	167	168	187	189	192					
		HIS	MET	LEU	PRO	ASP	GLN	GLN					
Cuminaldehyde	2	41	54	165	187	189							
		HIS	TYR	MET	ASP	GLN							
Linalool	2	41	44	49	54	165	187	189					
		HIS	CYS	MET	TYR	MET	ASP	GLN					
Coumaricacid	2	41	44	49	52	54	165	189					
		HIS	CYS	MET	PRO	TYR	MET	GLN					
Curcumin	5	25	27	140	142	144	145	163	165	167	168	189	192
		THR	LEU	PHE	ASN	SER	CYS	HIS	MET	LEU	PRO	GLN	GLN
Quercetin	4	140	144	145	163	165	166	189	192				
		PHE	SER	CYS	HIS	MET	GLU	GLN	GLN				
Sabinene	3	41	49	54	164	165	187	189					
		HIS	MET	TYR	HIS	MET	ASP	GLN					
Cineol	3	41	49	54	164	165	187	189					
		HIS	MET	TYR	HIS	MET	ASP	GLN					
Dihydrocapsaicin	5	41	49	54	144	145	163	165	166	187	189		
		HIS	MET	TYR	SER	CYS	HIS	MET	GLU	ASP	GLN		
Capsaicin	6	41	49	140	142	144	145	163	164	165	166	172	189
		HIS	MET	PHE	ASN	SER	CYS	HIS	HIS	MET	GLU	HIS	GLN
Ascorbicacid	2	49	165	166	189								
		MET	MET	GLU	GLN								

Conclusion

MilagaiKudineer exhibits promising antiviral properties attributed to its constituents. *Capsicumannuum* Linn (Green chilly) contains capsaicin, which possesses antiviral effects. *Cuminum cyminum* Linn (Cumin) and *Curcuma longa* Linn (Turmeric) are known for theirimmunomodulatory and anti-inflammatory properties, which can help combat viral infections. Computational analysis revealed the potential of bioactive compounds from MilagaiKudineer to bind effectively to the receptor-binding domain of the SARS-CoV-2 spike glycoprotein, the SARS-CoV-2 RNA-dependent RNA polymerase and the SARS-CoV-2 Main protease. Compounds such as Carvacrol, Cuminaldehyde, Linalool, Curcumin, Quercetin, Cineol, Dihydrocapsaicin, Capsaicin, and Ascorbic acid hold promise as inhibitors of viral entry and replication, highlighting their potential as therapeutic agents against COVID-19. Furtherexperimental studies are warranted to validate these findings and explore their clinical applications.

References

1. Ali A.M, Mackeen M.M, Ei-Sharkawy S.H, Hamid J.A, Ismail N.H, Ahmad F.B.H, et al. Antiviral and Cytotoxic Activities of Som. e Plants Used in Malaysian Indigenous Medicine Pertanika. J Trop Agric Sci. 1996;19(2/3):129–136.

2. Ali Esmail Al-Snafi. The pharmacological activities of Cuminum cyminum - A review. IOSR Journal of Pharmacy. 2016;6(6): 46-65.

3. Halgren T. A A. Merck molecular force field. I. Basis, form, scope, parameterization, and performance of MMFF94. Journal of Computational Chemistry (1998). 17 (5-6), 490-519.

4. Morris G. M, D. S. Goodsell, et al. Automated docking using a Lamarckian genetic algorithm and an empirical binding free energy function. Journal of Computational Chemistry (1998). 19 (14), 1639-1662.

5. Solis, F.J. and Wets, R.J.B. (1981) Minimization by

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27

Rajamanickam B, et al., J Pharm Pharmacol Res 2025
DOI:10.26502/fjppr.0105

Random Search Techniques. Mathematics of Operations Research, 6, 19-30.

6. Aartjan JW. The RNA polymerase activity of SARS-coronavirus nsp12 is primer-dependent. Nucleic Acids Research, 2010, Vol. 38, No. 1 203–214.
7. Sivaraman D, P.S. Pradeep. AI Vital Finding New Targets for Existing Drugs. Artificial intelligence is harnessed to accelerate drug repurposing in the search for COVID-19 solutions. COVID-19 Engineering Solutions Special Issue. Nature India.2020: 6-7.
8. Dhanasekaran Sivaraman, Puspharajselvadoss Pradeep. Scope of phytotherapeutics in targeting ACE2 mediated Host-Viral Interface of SARS-CoV2 that causes COVID-19. ChemRxi 2020. Doi.org/10.26434/chemrxiv.12089730.v1
9. Sivaraman D, P.S. Pradeep, S. Sundar Manoharan, C. Ramachandra Bhat, K.V. Leela, V. Venugopal. Revealing potential binding affinity of FDA approved therapeutics targeting Main protease (3CLpro) in impairing novel coronavirus (SARS-CoV-2) replication that causes COVID-19. Coronaviruses. Bentham publisher.2020;1:98-107. DOI: 10.2174/2666796701999200701122817
10. Dhanasekaran Sivaraman, Puspharajselvadoss Pradeep. Revealing Anti-viral Potential of Bio-active Therapeutics Targeting SARS-CoV2- polymerase (RdRp) in Combating COVID-19: Molecular Investigation on Indian Traditional Medicines. Preprints. March 2020. DOI: 10.20944/preprints202003.0450.v1.
11. Arumugam G, Swamy M.K,Sinniah U.R. Plectranthusamboinicus (Lour.) Spreng: Botanical, phytochemical, pharmacological, and nutritional significance. Molecules. 2016;21(4):369. doi: 10.3390/molecules21040369.
12. Asan R.PALPANDIAN, Nature Mimics, Earth India Siddha Private Limited, Tiruvannamalai, Tamilnadu,2023. ISBN:978-81-966690-1-0.
13. Bikadi, Z, Hazai, E. Application of the PM6 semi-empirical method to modeling proteins enhances docking accuracy of AutoDock. J. Cheminf. (2009). 1, 15.
14. Banegas-Luna A.J, Ceron-Carrasco J.P, Perez-Sanchez H. A review of ligand-based virtual screening web tools and screening algorithms in large molecular databases in the age of big data. Future Med Chem. 2018;10(22):2641–2658.
15. Bauer M.R, Mackey M.D. Electrostatic complementarity as a fast and effective tool to optimizebindingand selectivityof protein–ligand complexes. J Med
16. Borgio J, Alsuwat H, Al Otaibi W, Ibrahim A, Almandil N, Al Asoom L, et al. State-of-the-art tools unveil potent drug targets amongst clinically approved drugs to inhibit helicase in SARS-CoV-2. Arch Med Sci. 2020;16(3):508–518. doi: 10.5114/aoms.2020.94567.
17. Coronavirus disease 2019. World Health Organization; 15 March 2020.
18. Daina A,Michielin O, Zoete V. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Sci Rep. 2017;7:42717.
19. Denyer C.V, Jackson P,Loakes D.M, Ellis M.R, Young D.A. Isolation ofantirhinoviralsesquiterpenes fromginger(Zingiberofficinale) J Nat Prod. 1994;57(5): 658–662.
20. Eweas A.F,Maghrabi I.A,Namarneh A.I. Advances in molecular modeling and docking as a tool for modern drug discovery. Sch Res Lib Der Pharma Chem. 2014;6:211–228.
21. Flare, version, Cresset®, Litlington, Cambridgeshire, UK. Cheeseright T, Mackey M, Rose S, Vinter A. Molecular field extrema as descriptors of biological activity: definition and validation. J Chem Inf Model. 2006;46(2):665–676. http://www.cresset-group.com/flare
22. Goodsell, D. S,Zardecki, C, di Costanzo, L, Duarte, J. M, Hudson, B. P,Persikova, I, Segura, J, Shao, C, Voigt, M, Westbrook, J. D, Young, J. Y,& Burley, S. K. RCSB Protein Data Bank: Enabling biomedical research and drug discovery. Protein Science, 2020. 29(1), 52–65. https://doi.org/https://doi.org/10.1002/pro.3730.
23. Guo Z.Y.L, Zheng X, Hu L, Yang Y, Wang J.A. A comparison of various optimization algorithms of protein-ligand docking programs by fitness accuracy. J Mol Model. 2014;20(7):2251–2261.
24. Hamed, M, Kalita, D, Bartolo, M. E,& Jayanty, S. S. (2019). Capsaicinoids, Polyphenols and Antioxidant Activities of Capsicum annuum: Comparative Study of the Effect of Ripening Stage and Cooking Methods. Antioxidants (Basel, Switzerland), 8(9), 364. https://doi.org/10.3390/antiox8090364.
25. Hewlings SJ, Kalman DS. Curcumin: A Review of Its Effects on Human Health. Foods. 2017;6(10):92.
26. https://www.nhp.gov.in/pitha-suram-dengue-fever-_mtl
27. https://www.nhp.gov.in/swine-flu_mtl
28. Jain J, Kumar A, Narayanan V, Ramaswamy R.S,Sathiyarajeswaran P, Devi M.S, et al. Antiviral activity of ethanolic extract of NilavembuKudineer against dengue and chikungunya virus through in vitro evaluation. J Ayurveda Integr Med. 2019;S0975-9476(18):30073-1. doi: 10.1016/j.jaim.2018.05.006.

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27

Rajamanickam B, et al., J Pharm Pharmacol Res 2025
DOI:10.26502/fjppr.0105

29. Jain J, Narayanan V, Chaturvedi S, Pai S, Sunil S. In vivo evaluation of withaniasomnifera–based Indian traditional formulation (Amukkarachoornam), against chikungunya virus–induced morbidity and arthralgia. J Evid Based Integr Med. 2018 Feb 26;23 2156587218757661.

30. Jain J, Pai S, Sunil S. Standardization of in vitro assays to evaluate the activity of polyherbal siddha formulations against Chikungunya virus infection. Virus Dis. 2018;29(1):32–39.

31. Jiang Z.Y, Liu W.F, Zhang X.M, Luo J, Ma Y.B, Chen J.J. Anti-HBV active constituents from Piper longum. Bioorg Med Chem Lett. 2013;23(7):2123–2127.

32. Kalikar M.V,Thawani V.R,Varadpande U.K, Sontakke S.D, Singh R.P,Khiyani R.K. Immunomodulatory effect of Tinospora cordifolia extract in human immuno-deficiency virus positive patients. Indian J Pharmacol. 2008;40(3):107–110.

33. Koch C, Reichling J,Schneele J, Schnitzler P. Inhibitory effect of essential oils against Herpes Simplex virus type 2. Phytomedicine. 2008;15(1–2):71–78.

34. Murugesamudaliyar K.S. 8th ed. Directorate of Indian Medicine & Homeopathy; Chennai: 2006. Siddha materia media (medicinal plants division) pp. 7–713.

35. Pereira F, Aires-de-Sousa J. Computational methodologies in the exploration of the marine natural product leads. Mar Drugs. 2018;16(7):E236.

36. Ramalingam M, Annamalai S, Sekar A, et al. In-silico Identification of Potential Phytochemicals from Siddha Formulation "KabasuraKudineer" against SARS-CoV-2 Spike Protein. Evidence-based Complementary and Alternative Medicine. 2021; 2021: 8892507.

37. Ramaswamy RS, Sathiyarajeswaran P, Shree Devi MS, Mohana Srinivasan V, Subathra Devi C, et al. Simple and low-cost process for the preparation of synergistic bio active compound JACOM for the management of H1N1 influenza virus infection, (Patent no: 201741016901 A, 18.05.2018).

38. Saravanan J, Devasia N,Gopalasatheeskumar K, Sanish Devan V, Thanga Kokila K, Sanjay M. Anti-inflammatory, antipyretic and antibacterial study of Kabasurakudineerchoornam. Int J Curr Adv Res. 2018;7(2):9992–9997.

39. Selvaraj J, Jayaprakash R, Arunachalam C, et al. Molecular docking analysis of Siddha drug ThirikaduguChooranam on SARS-CoV-2 spike protein. Journal of Traditional and Complementary Medicine. 2021; 11(3): 225-229.

40. Senthil Kumar N, Kumari BP, Meenakumari R, et al. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against COVID-19 Spike Glycoprotein with Receptor-Binding Domain: An In-Silico Investigation for Antiviral Intervention. Frontiers in Pharmacology. [Online ahead of print]. DOI: 10.3389/fphar.2021.757823.

41. Shahid M, Dar F.K, Ismaeel A.Y, Al-Mahmeed A, Al Sindi K, Malik A, et al. Recent trends in biotechnology and therapeutic applications of medicinal plants. Springer; Dordrecht: 2013. Plant natural products as a potential source of antimicrobial agents: an overview and a glimpse on recent developments; pp. 93–107.

42. Shaikh S.A, Jain T, Sandhu G, Latha N, Jayaram B. From drug target to leads-sketching a physico-chemical pathway for lead molecule design in silico. Curr Pharmaceut Des. 2007;13(34):3454–3470.

43. Shanmugavelu M. 1st ed. Directorate of Indian Medicine & Homeopathy; Chennai: 2014. Noi nadalnoimudhalnadalthirattu part-I (in Tamil) p. 370.

44. Shanmugvelu N. Noi Nadal Noi Mudal Nadal. Chennai: Department of Indian Medicine and Homoeopathy; 1967. 164 p.

45. Siddha pharmacopoeia of India. Part 1, vol 2, First edition, Govt. of India, Ministry of Health and Family Welfare, Department of AYUSH, NewDelhi,2011.p43,119

46. The Siddha Formulary of India. Part 1, vol 2 , First edition, Govt. of India, Ministry of Health and Family Welfare, Department of Health, NewDelhi,1992.p27,185

47. Siddha formulary of India Part II. The controller of Publications; Delhi: 2011. p.160,173,178, 179,181, 182,183,186,187,191,192.

48. Singh S, Awasthi M, Tiwari S, Pandey V.P, Dwivedi U.N. Computational approaches for therapeutic application of natural products in Alzheimer's disease. Neuromethods. 2018;132:483–511.

49. Sood R, Raut R, Tyagi P, Pareek P.K, Barman T.K, Singhal S, et al. Cissampelos pareira Linn: natural source of potent antiviral activity against all four dengue virus serotypes. PLoS Neglected Trop Dis. 2015;9(12).

50. Srinivasan D, Perumal K, Nadimuthu K. Siddha treatment for COVID-19: A review of recent advances and future prospects. Journal of Traditional and Complementary Medicine. 2021; 11(1): 26-34.

51. Venkatasubramanian P, Priya A, Gnanamani A. Identification of Potential Anti-SARS-CoV-2 Molecules from Siddha Formulations – An in Silico Approach. European Journal of Molecular & Clinical Medicine. 2021; 8(5): 1895-1909.

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27

Rajamanickam B, et al., J Pharm Pharmacol Res 2025
DOI:10.26502/fjppr.0105

52. Wadhwa B, Mahajan P, Barik M.R, Malik F,Nargotra A. Combining ligand- and structure-based in silico methods for the identification of natural product-based inhibitors of Akt1. Mol Divers. 2019;24(1):45–60.

53. Wang W, Wang J, Dong S.F, Liu C.H, Italiani P, Sun S.H, et al. Immunomodulatory activity of andrographolide on macrophage activation and specific antibody response. Acta Pharmacol Sin. 2010;31(2):191–201.

54. Worachartcheewan A,Prachayasittikul V,Shoombuatong W,Songtawee N, Simeon S,Prachayasittikul V, et al. Computer aided drug design of bioactive natural products. Curr Top Med Chem. 2015;15(18):1780–1800.

55. World Health Organization . 2020-01-30. Statement on the second meeting of the International Health Regulations (2005) Emergency Committee regarding the outbreak of novel coronavirus (2019-nCoV) [2020].

56. Wrapp D, Wang N, Corbett K.S, Goldsmith J.A, Hsieh C.L, Abiona O, et al. Cryo- EM structure of the 2019-nCoV spike in the prefusion conformation. Science. 2020;367(6483):1260–1263.

57. Zhang, H. A, & Kitts, D. D. (2021). Turmeric and its bioactive constituents trigger cell signaling mechanisms that protect against diabetes and cardiovascular diseases. Molecular and cellular biochemistry, 476(10), 3785–3814. https://doi.org/10.1007/s11010-021-04201-6

Citation: Baskar Rajamanickam, Ramesh Balasubramanian, Manekshah Yovas Rajammal, Bharathkumar Govindaraju, Selvapriya Sikkal Selvaraaju, Hemadevi Thangarasu, Kanakavalli Kadarkarai. Exploring the Potential of Siddha Formulation MilagaiKudineer-Derived Phytotherapeutics Against SARS-CoV-2: An In-Silico Investigation for Antiviral Intervention. Journal of Pharmacy and Pharmacology Research. 8 (2025): 17-27