Pharmacogn. Mag.
A multifaceted peer reviewed journal in the field of Pharmacognosy and Natural Products
www.phcog.com | www.phcog.net

ORIGINAL ARTICLE

In Silico Screening of Antibacterial Compounds from Herbal Sources Against *Vibrio cholerae*

Sabah Perveen, Hotam Singh Chaudhary

Department of Biotechnology, Madhav Institute of Technology and Science, Gwalior, Madhya Pradesh, India

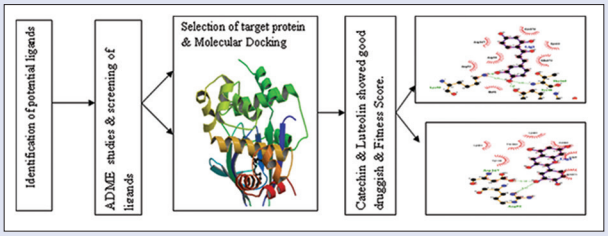
ABSTRACT

Background: The prolonged use of antibiotic viz., tetracycline, quinolones, ampicillin, etc., to reduce the infection of cholera, may failed due to the emergence of new *Vibrio cholerae* antibiotics resistant strains. Moreover, these antibiotics even restricted for patient suffering from severe dehydration. Hence, there is a call to find an alternative therapeutics against *V. cholerae*. The natures serve different herbs in its lap which might contain several natural therapeutic compounds almost all diseases. Computer-aided designing is the initial steps for screening the novel inhibitors. **Objective:** To identify and evaluate natural compounds with low side effects with high efficacy against *V. cholerae* has been done. **Materials and Methods:** *In silico* screening, absorption, digestion, metabolism, and excretion (ADME), and docking of herbal compounds have been performed on to the target ToxT (transcriptional activator of *V. cholerae*). The compound with good ADME properties and drug-likeness property were subjected to docking. **Results:** From 70 herbal compounds, some compounds such as aloin, campesterol, lupeol, and ursolic acid showed a violation of the rule of five and compounds such as lupeol and beta carotene showed negative binding energy. Luteolin, catechin, brevifolin, etc., compounds were selected based on ADME, drug-likeness property, and docking studies. **Conclusion:** Two compounds named catechin and luteolin showed better inhibition properties against ToxT and good ADME and drug-likeness property were selected as a better lead molecule for drug development in future. The Genetic Optimization for Ligand Docking fitness score for catechin is 48.74 kcal/mol and luteolin 38.12 kcal/mol.

Key words: Absorption, digestion, metabolism, and excretion, catechin, docking, luteolin, transcriptional activator ToxT

SUMMARY

- *Vibrio cholerae* became antibiotic resistance and associated with several cholera epidemic and pandemic. Hence, there is a need to find an alternative therapeutics against *V. cholerae*. Many herbal compounds present in nature having high medicinal value. From *in-silico* study,found two compound Luteolin from *Tulsi* and Catechin from *Green Tea* which showed good binding energy and druggish property.



Abbreviations used: *V. cholerae: Vibrio cholera,* ADME: Absorption, digestion, metabolism and excretion, CT: Cholera toxin, TCP: Toxin co-regulated Pilus, GOLD: Genetic Optimization for Ligand Docking, Asp: Aspartic acid, Arg: Arginine, Lys: Lysine, Thr: Threonine, Tyr: Tyrosine, KEGG: Kyoto encyclopedia of Genes and Genomes.

Correspondence:
Prof. Hotam Singh Chaudhary,
Department of Biotechnology, MITS, Gwalior,
Madhya Pradesh, India.
E-mail: hotamsingh@gmail.com

DOI : 10.4103/0973-1296.172960

Access this article online
Website: www.phcog.com
Quick Response Code:

INTRODUCTION

Vibrio cholerae, the causative organism of cholera is a comma shaped, Gram-negative bacterium responsible for severe morbidity and mortality in developing countries of the world including South Asian countries.[1-3] It has been the culprit of seven pandemics of the world.[4] *V. cholerae* encompasses more than 200 serogroups, two of which are associated with cholera epidemic and pendemic. The studies of these *V. cholerae* serogroup O1 and O139 have led to the identification of several critical virulence factors such as cholera toxin (CT) and toxin co-regulated pilus (TCP). In addition, *V. cholerae* produces a major zinc-dependent metalloprotease known as hemagglutinin/protease. All the serogroups of *V. cholerae* except O1 and O139 are considered as non-O1 and non-O139, and most are nonpathogenic. However, some members of serogroup are capable of causing sporadic cases of moderate to severe gastroenteritis and extraintestinal infections in humans, despite the fact that the genes encoding TCP and CT are absent, thus raising increasing concern in endemic area.[5]

Many antibiotics use against *V. cholerae* such as tetracycline and fluoroquinolones, ciprofloxacin, ampicillin, nalidixic acid, and erythromycin. With time, the *V. cholerae* strains became resistant to these antibiotics, and this is evident from some recent cholera outbreaks.[6-8] In this current scenario, there is a demand to find a better therapeutics against *V. cholerae*. The best alternative is the bioactive natural compounds present in the lab of herbal plants. Importantly many herbal plants contain natural compounds of high medicinal value. Computer-aided methods are preliminary approach to screening novel therapeutic candidates and an emerging strategy to reduce many complexities of drug discovery process. Absorption, digestion,

Cite this article as: Perveen S, Chaudhary HS. *In silico* screening of antibacterial compounds from herbal sources against *vibrio cholerae*. Phcog Mag 2015;11:550-5.

metabolism, and excretion (ADME) and drug-likeness properties of all biological active compounds present in nature can be easily found by the computer-aided method.[9] The study of receptor-ligand interaction is a fundamental concept of rational drug design and the prediction of such interactions by computational methods has highlighted the importance of structure-based drug discovery.[10]

There are many herbal compounds reported in the literature with medicinal value and may be used as therapeutics agent against *V. cholerae*.[1,9,11] The study is mainly focused on the screening of potential herbal compound which might inhibit the infection of targeted *V. cholerae* by computer-based studies.

MATERIALS AND METHODS

Identification of potential ligands

Many herbal compounds having high medicinal value reported against *V. cholerae*.[12] Hence, an initial survey of 70 herbal compounds from 15 different plants was carried out from extensive review studies. The three-dimensional (3D) structure of these compounds is present in drug database such as PubChem, ChemSpider, and Kyoto Encyclopedia of Genes and Genomes, and the structures were retrieved from the PubChem[13] and ChemSpider.[14]

Absorption, digestion, metabolism, and excretion studies and screening of ligands

The ADME are the most important part of pharmacological studies of lead molecules, and these can be predicted by computational tools such as MedChem designer and OSIRIS property explorer. Hence, all the 70 ligands were tested for their ADME profile, drug-likeliness, drug score, and toxicity risks. MedChem designer was used for ADME profiling. The SDF file of ligands was uploaded in MedChem designer software and then calculated the ADME property. Computer programmer OSIRIS used for drug-likeliness, toxicity risks, and drug score.

MedChem designer is an advanced molecule design software application that combines an intuitive sketch interface with fast and accurate ADME property predictions from ADME Predictor software package.[15] It also predicts rule of five. Rule of five evaluate drug-likeliness property that would make it an orally active drug. Updated OSIRIS properties explorer help to search chemical structures by name, Simplified Molecular Input Line Entry System, CAS No., and calculates on-the-fly various drug-relevant properties whenever a structure is valid. Prediction results are valued and color coded. Properties with high risks of undesired effects like mutagenicity or a poor intestinal absorption are shown in red. Whereas a green color indicates drug-conform behavior.[16]

Selection of target protein and molecular docking

Transcriptional activator ToxT activates transcription for structural genes of CT and TCP virulence factors. Hence *V. cholerae* strain deficient for ToxT production lack expression of CT and TCP and thus could not cause disease. Thus, ToxT is now a novel and potent target in *V. cholerae* for drug designing.[17] The crystal structure of transcriptional activator ToxT (PDB ID: 3GBG) was retrieved from Protein Data Bank (PDB). The active site of this target protein is identified by Active Site Prediction (http://www.scfbio-iitd.res.in/dock/ActiveSite.jsp). The selected herbal compounds docked onto ToxT using Genetic Optimization for Ligand Docking (GOLD). GOLD uses a genetic algorithm for searching binding ligand conformational space and provides a score for binding residues. GOLD scores are used to rank poses.[18]

Table 1: ADME prediction of selected herbal compound by MedChem designer

Ligand	Plant source	MlogP	S + logP	S + logD	Rule of five	Rule of five code	Mwt	M_No	T_PSA	HBDH
Quercetin	Neem	−0.235	1.958	1.529	0.00		302.24	7	131.36	5.00
Emodin	*A. vera*	1.372	3.064	2.0189	0.00		270.24	5	94.830	3.00
Aloin	*A. vera*	−0.156	0.017	−0.040	1.00	Hb	418.40	9	167.91	7.00
Campesterol	*A. vera*	6.591	9.182	9.182	1.00	LP	400.69	1	20.203	1.00
Lupeol	*A. vera*	6.786	9.518	9.518	1.00	LP	414.72	1	20.230	1.00
Beta-sitosterol	*A. vera*	6.979	9.281	9.281	1.00	LP	426.73	1	20.230	1.00
Oleanolic acid	Tulsi	5.561	7.261	5.099	1.00	LP	456.71	3	57.530	2.00
Ursolic acid	Tulsi	5.561	7.017	4.892	1.00	LP	456.71	3	57.530	2.00
Eugenol	Tulsi	2.621	2.230	2.229	0.00		164.20	2	29.460	1.00
Carvacrol	Tulsi	2.813	3.128	3.127	0.00		150.22	1.00	20.230	1.00
Methyl cinnamate	Tulsi	1.894	2.538	2.538	0.00		162.18	2.00	26.300	0.00
Linalool	Tulsi	2.642	2.894	2.894	0.00		154.25	1.00	20.230	1.00
Beta-caryophllene	Tulsi	4.631	5.761	5.761	1.00	LP	204.35	0.00	0.00	0.00
Luteotin	Tulsi	0.525	2.428	2.007	0.00		286.24	6.00	111.13	4.00
Apigenin	Tulsi	1.296	2.858	2.530	0.00		270.24	5.00	90.900	3.00
Curcumin	Turmeric	2.256	2.994	2.943	0.00		368.38	6.00	93.060	2.00
Allicin	Garlic	1.238	1.959	1.959	0.00		162.27	1.00	17.070	0.00
Alpha-atlantone	Turmeric	3.427	4.368	4.368	0.00		218.34	1.00	17.070	0.00
Allyl sulfide	Garlic	2.352	2.203	2.203	0.00		114.21	0.00	0.00	0.00
Ellagic acid	Pomegranate	−0.57	1.914	1.382	0.00		302.19	8.00	141.34	4.00
Diallyl disulphide	Garlic	2.352	2.420	2.420	0.00		146.27	0.00	0.00	0.00
(−) epigallocatechin gallate	Tea	−1.21	2.170	1.574	2.00	Hb, NO	458.38	11.00	197.37	8.00
Corilagin	Tea	−4.01	0.853	−0.267	3.00	Hb, Mw, NO	634.46	18.00	310.66	11.00
Alpha-pinene	Eucalyptus	4.286	4.362	4.362	1.00	LP	136.23	0.00	0.00	0.00
Benzyl benzoate	Eucalyptus	3.108	3.457	3.457	0.00		212.25	2.00	26.300	0.00
(+) limonene	Eucalyptus	3.267	4.274	4.274	0.00		136.29	0.00	0.00	0.00
Ethyl cinnamate	Eucalyptus	2.186	3.003	3.003	0.00		176.21	2.00	26.300	0.00
Cinnamaldehyde	Cinnamon	2.066	1.964	1.964	0.00		132.16	1.00	17.070	0.00
Cinnamyl alcohol	Cinnamon	2.152	1.902	1.902	0.00		134.17	1.00	20.230	1.00
Cinnamyl acetate	Cinnamon	2.186	2.707	2.707	0.00		176.21	1.00	26.300	0.00
Ferulic acid	Guava	1.298	1.542	−1.225	0.00		194.18	4.00	66.760	2.00

Contd...

Table 1: Contd...

Ligand	Plant source	MlogP	S + logP	S + logD	Rule of five	Rule of five code	Mwt	M_No	T_PSA	HBDH
Ascorbic acid	Guava	−2.22	−1.638	−1.633	0.00		176.12	6.00	107.22	4.00
Cyanidin	Guava	0.600	−0.195	−1.299	0.00		287.25	6.00	114.29	5.00
Geraniol	LemonGrass	2.642	3.190	3.190	0.00		154.25	1.00	20.230	1.00
Citral	Lemon Grass	2.545	3.012	3.012	0.00		152.23	1.00	17.070	0.00
Geranic acid	Lemon Grass	2.070	3.131	0.369	0.00		168.23	2.00	37.300	1.00
Chlorogenic acid	Lemon Grass	−0.96	−0.398	−2.353	1.00	Hb	354.31	9.00	164.75	6.00
Orientin	Lemon Grass	−1.76	−0.349	−0.542	2.00	Hb, NO	448.38	11.00	201.28	8.00
Myrcene	Lemon Grass	3.562	4.466	4.66	0.00		136.23	0.00	0.00	0.00
Beta pinene	Lemon Grass	4.286	4.152	4.152	1.00	LP	136.23	0.00	0.00	0.00
Barneol	Lemon Grass	2.502	2.765	2.765	0.00		154.25	1.00	20.230	1.00
Caffeic acid	Lemon Grass	0.994	1.262	−1.395	0.00		180.16	4.00	77.760	3.00
Alpha Tocopherols	Lemon Grass	6.237	11.481	11.481	1.00	LP	430.71	2.00	29.460	1.00
Beta carotene	Grape	8.957	11.618	11.618	2.00	Mw, LP	536.89	0.00	0.00	0.00
Benzethonium	Grape	1.189	1.391	1.391	0.00		421.46	3.00	18.460	0.00
Bergamottin	Grape	2.782	5.399	5.399	0.00		338.40	4.00	52.580	0.00
Bergapten	Grape	0.727	2.092	2.092	0.00		216.19	4.00	52.580	0.00
Citric acid	Grape	−2.39	−1.669	−4.709	0.00		192.12	7.00	132.13	4.00
Limonin	Grape	1.031	1.913	1.913	0.00		470.52	8.00	104.57	0.00
Narangenin	Grape	1.450	2.420	2.104	0.00		272.25	5.00	86.990	3.00
Nootkatone	Grape	3.517	3.639	3.639	0.00		218.34	1.00	17.070	0.00
Putrescine	Grape	−0.08	−0.857	−3.968	0.00		88.153	2.00	52.040	4.00
Resveratron	Grape	2.402	3.094	3.080	0.00		228.24	3.00	60.690	3.00
Farnesol	Grape	3.909	4.998	4.998	0.00		222.37	1.00	20.230	1.00
Triclosan	Grape	4.353	5.580	5.441	1.00	LP	289.24	2.00	29.460	1.00
Nimbin	wasabi	1.246	2.948	2.948	1.00	Mw	540.61	9.00	118.34	0.00
Gallate	Piper betel	0.178	0.682	−2.652	0.00		170.12	5.00	97.990	4.00
Procyanidin	Piper betel	−1.16	1.742	1.674	3.00	Hb, Mw, NO	594.53	13.00	229.99	10.00
Allyl isothiocyanate	Piper betel	2.033	2.104	2.102	0.00		99.155	1.00	12.360	0.00
Capsaicin	Piper betel	3.392	3.721	3.720	0.00		305.42	4.00	58.560	2.00
Catechin	Piper betel	0.757	0.775	0.746	0.00		290.27	6.00	110.38	5.00
Guineesine	Piper betel	4.060	5.959	5.959	0.00		383.53	4.00	47.560	1.00
Pinorensinol	Daio	1.984	2.644	2.634	0.00		358.39	6.00	77.380	2.00
Dehydropipernonaline	Guazuma	3.415	3.982	3.982	0.00		339.43	4.00	38.770	0.00
Piperrolein-B	Green tea	3.578	4.822	4.822	0.00		343.46	4.00	38.770	0.00
chlorogenic acid	Red chili	−0.96	-0.398	−2.353	1.00	Hb	354.31	9.00	164.75	6.00
Eugenyl acetate	Grape	2.167	2.611	2.611	0.00		206.24	3.00	35.530	0.00
Thymol	Pomegranate	1.237	2.148	−1.280	0.00		138.12	3.00	57.530	2.00
Azadirachtin	Neem	−1.44	0.400	0.400	2.00	Mw, NO	720.73	16.00	215.34	3.00
Brevifolin	Clove	1.513	1.905	1.884	0.00		196.20	4.00	55.760	1.00

The result gave the value of S + logP, S + logD, MlogP, HBDH, M_NO, T_PSA, Lipinski's rule of five and rule of five_code. Many Compounds have satisfied the different parameters of ADME study; *A. vera: Aloe vera*

RESULTS

The 70 herbal ligands are firstly screened on the basis of ADME studies. ADME prediction is done by MedChem designer. The result of ADME is tabulated in Table 1. The toxicity risk, drug-likeness, and drug score is calculated by computer programmer OSIRIS property explorer and the results are given in Table 2. After ADME studies of ligands we have found that many herbal compounds were suitable for drug development against *V. cholerae*. β-sitosterol from *Aloe vera*, lupeol from *Ocimum tenuiflorum*, brevifolin and ellagic acid from *Punica granatum*, catechin from green tea, luteolin from Tulsi, and many more showed better pharmacokinetic and drug-likeness properties. Using GOLD software, the docking of ligands to the target (PDB ID: 3GBG) has been done, and the GOLD fitness scores of all the ligands have been given in Table 3.

On the basis of drug score, ADME and docking studies we have found luteolin and catechin as a best ligands which shows good drug-score, no toxicity risk, and best GOLD score.

DISCUSSIONS

The outbreaks of cholera are critical because the organism is recently emerged as antibiotic resistant.[19] Hence, the discovery of an alternative treatment has profound scope and significance. Herbal compounds are the best alternative because of their high medicinal value. There are many herbal compounds present in the nature which has good druggish property and have some medicinal value. The traditional method of drug designing is time-consuming and complicated. There are many clinical and preclinical trials. One of the major reasons for drug failures is the poor drug-likeness and pharmacokinetic properties of lead compounds. Computer- aided drug designing method is a good and rapid method. It gives significant screening approach because it selects the lead molecules with good pharmacological and druggish properties. We have selected 70 herbal compounds from 15 different plants. The 3D structure of the ligands was retrieve from the drug database, PubChem, and ChemSpider. However, many ligands were found suitable as per Lipinski's rule of five [Table 1, Supplementary Data].

Another important concern in drug designing is the pharmacokinetics properties. The ADME study gives the property of drug such as ADME. The ADME profiling of selected ligand was done by MedChem designer, and the values are provided in Table 1. *In silico* toxicity prediction is the final step in any drug designing process. In this study, we have found that the predicted toxicity risks, drug-likeness, and drug score of some herbal compounds were suitable. β-sitosterol from *A. vera,* brevifolin from

Table 2: Toxicity risk, drug score, and drug-likeness are predicted by OSIRIS property explorer

Ligands	Plant source	Mutagenic	Tumorigenic	Irritant	Reproductive effective	Drug- likeness	Drug-score
Quercetin	Neem	Red	Red	Green	Green	1.6	0.3
Emodin	*A. vera*	Red	Red	Red	Red	-	-
Aloin	*A. vera*	Green	Green	Green	Green	−2.33	0.44
Campesterol	*A. vera*	Green	Green	Green	Green	−8.19	0.14
Lupeol	*A. vera*	Green	Green	Green	Green	−22.13	0.13
Beta-sitosterol	*A. vera*	Green	Green	Green	Green	−4.48	0.13
Eugenol	Tulsi	Red	Red	Red	Green	−2.78	0.11
Carvacrol	Tulsi	Green	Green	Red	Green	−2.59	0.29
Methyl cinnamate	Tulsi	Green	Green	Red	Green	−5.55	0.28
Linalool	Tulsi	Red	Green	Red	Green	−6.68	0.16
Beta-caryophyllene	Tulsi	Green	Green	Green	Green	−6.48	0.31
Luteolin	Tulsi	Green	Green	Green	Green	1.9	0.84
Apigenin	Tulsi	Red	Green	Green	Green	1.21	0.47
Curcumin	Turmeric	Green	Green	Green	Green	−3.95	0.4
Allicin	Garlic	Green	Green	Green	Green	−6.13	0.48
Allyl-sulfide	Garlic	Red	Green	Green	Green	−3.93	0.29
Ellagic acid	Pomegranate	Green	Green	Green	Green	−1.6	0.51
Diallyl disulfide	Garlic	Green	Green	Green	Green	−4.7	0.45
(−)-epigallacatechin gallate	Tea	Green	Green	Green	Green	1.58	0.7
Alpha-pinene	Eucalyptus	Green	Green	Red	Green	−1.8	0.31
Benzyl-benzoate	Eucalyptus	Green	Green	Red	Green	−11.8	0.26
(+)-limonene	Eucalyptus	Green	Green	Yellow	Green	−21.8	0.35
Ethyl cinnamate	Eucalyptus	Green	Green	Red	Green	−9.49	0.28
Cinnamaldehyde	Cinnamon	Red	Red	Red	Red	−6.47	0.06
Cinnamyl alcohol	Cinnamon	Red	Green	Red	Red	−2.81	0.11
Cinnamyl Acetate	Cinnamon	Red	Green	Red	Green	−2.29	0.18
Ferulic acid	Guava	Red	Red	Green	Red	1.12	0.18
Ascorbic acid	Guava	Green	Green	Green	Green	0.02	0.74
Geranoil	Lemon Grass	Green	Green	Red	Green	−3.67	0.27
Citral	Lemon Grass	Yellow	Red	Yellow	Red	−7.31	0.1
Geranic acid	Lemon Grass	Green	Green	Red	Green	−4.49	0.27
Chlorogenic acid	Lemon Grass	Green	Green	Green	Green	0.17	0.7
Orientin	Lemon Grass	Red	Green	Green	Green	−0.71	0.32
Myrcene	Lemon Grass	Green	Red	Red	Red	−7.82	0.09
Beta-pinene	Lemon Grass	Green	Green	Red	Green	−7.56	0.27
Farnesol	Lemon Grass	Green	Green	Green	Green	−3.38	0.32
Borneol	Lemon Grass	Red	Green	Red	Green	−3.53	0.17
Caffeic acid	Lemon Grass	Red	Red	Green	Red	1.62	0.19
Alpha-tocopherol	Grape	Green	Green	Green	Green	−4.78	0.12
Beta-carotene	Grape	Green	Green	Green	Green	−3.35	0.1
Benzethonium	Grape	Green	Green	Red	Green	−13.3	0.21
Bergamottin	Grape	Green	Green	Red	Green	−4.97	0.13
Bargapten	Grape	Red	Red	Green	Red	−3.38	0.1
Citric acid	Grape	Green	Green	Red	Green	3.56	0.38
Limonin	Grape	Green	Green	Green	Green	−3.0	0.35
Naringenin	Grape	Red	Green	Green	Green	1.9	0.51
Nootkatone	Grape	Green	Green	Green	Green	−20.9	0.4
Putrescine	Grape	Red	Green	Green	Red	−3.53	0.18
Resveratrol	Grape	Red	Green	Green	Red	−3.25	0.16
Triclosan	Grape	Red	Red	Red	Red	1.41	0.06
Guineesine	Piper betel	Green	Green	Green	Red	−9.8	0.1
Piperrolein-B	Piper betel	Green	Green	Green	Red	−7.85	0.14
Chlorogenic acid	Piper betel	Green	Green	Green	Green	0.17	0.7
Eugenyl acetate	Piper betel	Green	Red	Red	Green	-0.98	0.21
Procyanidin	Guazuma	Green	Green	Green	Green	1.77	0.53
Catechin	Green tea	Green	Green	Green	Green	1.92	0.87
Capsaicin	Red chili	Green	Green	Green	Green	−9.65	0.39
Brevifolin	Pomegranate	Green	Green	Green	Green	−0.77	0.63
Nimbin	Neem	Green	Green	Red	Green	−4.87	0.16
Thymol	Clove	Red	Green	Green	Red	−3.02	0.17

The result is given in color and value. Property with a high risk of undesirable effect shows red color and green color show drug confirmation behavior. *A. vera: Aloe vera*

P. granatum, luteolin from Tulsi, catechin from green tea, and many more showed better pharmacokinetics property and drug score [Tables 1 and 2, Supplementary Material]. After this screening process, these ligands are found suitable for docking studies. All the herbal compounds were reported to have high medicinal value against different pathogens, and we have tested the efficiency of the same against *V. cholerae* by computer-aided approach.

Table 3: The GOLD score (kcal/mol) of various plant-derived compounds with ToxT of *Vibrio cholerae*

Compounds ID	Herbal active compounds	GOLD score (kcal/mol)
CID:5280343	Quercetin	36.59
CID:3220	Emodin	40.33
CID:12305761	Aloin	34.80
CID:173183	Campesterol	21.48
CID:259846	Lupeol	−9.03
CID:222284	Beta-sitosterol	23.05
CID:3314	Eugenol	36.76
CID:10364	Carvacrol	35.88
CID:637520	Methyl cinnamate	33.20
CID:6549	Linalool	36.03
CID:5281515	Beta-caryophyllene	23.90
CID:5280445	Luteolin	38.12
CID:5280443	Apigenin	36.70
CID:969516	Curcumin	50.24
CID:65036	Allicin	37.25
CID:558173	Alpha-atlantone	44.91
CID:11617	Allyl sulfide	28.35
CID:5281855	Ellagic acid	25.95
CID:16590	Diallyl disulphide	41.42
CID:65064	(−) epigallocatechin gallate	36.34
CID:6654	Alpha-pinene	28.26
CID:2345	Benzyl benzoate	42.01
CID:440917	(+) limonene	25.72
CID:637758	Ethyl cinnamate	45.89
CID:637511	Cinnamaldehyde	33.03
CID:5315892	Cinnamyl alcohol	28.43
CID:5282110	Cinnamyl acetate	35.96
CID:445858	Ferulic acid	34.25
CID:54670067	Ascorbic acid	39.66
CID:637566	Geraniol	39.13
CID:638011	Citral	38.57
CID:5275520	Geranic acid	31.61
CID:1794427	Chlorogenic acid	47.77
CID:5281675	Orientin	40.98
CID:31253	Myrcene	31.18
CID:14896	Beta pinene	25.89
CID:64685	Barneol	24.08
CID:689043	Caffeic acid	38.29
CID:14985	Alpha Tocopherols	38.92
CID:5280489	Beta carotene	-21.71
CID:2335	Benzethonium	30.49
CID:5471349	Bergamottin	48.39
CID:311	Citric acid	47.99
CID:179651	Limonin	26.65
CID:932	Narangenin	42.42
CID:1045	Putrescine	16.55
CID:445154	Resveratron	35.69
CID:445070	Farnesol	47.20
CID:5564	Triclosan	48.76
CID:108058	Nimbin	29.21
CID:107876	Procyanidin	26.64
CID:6442405	Guineesin	50.74
CID:21580213	Piperrolein-B	55.39
CID:1794427	Chlorogenic acid	40.26
CID:7136	Eugenyl acetate	32.01
CID:98389	Brevifolin	40.46
CID:5281303	Azadirachtin	39.48
CID:73160	Catechin	48.74
CID:1548943	Capsaicin	55.47
CID:6989	Thymol	32.75

Many herbal compounds like catechin, luteolin, brevifolin, etc. gave a good fitness score. GOLD: Genetic Optimization for Ligand Docking

There are two main virulence factors, CT and TCP in *V. cholerae*. The key protein involved in the virulence of *V. cholerae* is CT and TCP. The expression of both proteins is regulated by the transcriptional activator ToxT. Thus, the ToxT has been selected as a probable drug target. The crystal structure of ToxT (3GBG) was retrieved from PDB. The protein consists of a single chain. Chain A is the key domain consists of 260 amino acids residues, 4274 number of atoms, and 4313 number of bonds present. For interaction between ligand and target, the active site information is necessary. The active site was predicted by the online server Active Site Prediction. There were total 20 active cavities present in the target protein for docking. The docking study is carried out by GOLD. During docking studies, some compound did not give any fitness scores such as bergapten, gallate, and Nootkatone. The docking studies and the fitness score of all the selected ligands are given in Table 3. As the best catechin, active inhibitor present in green tea showed the fitness score 48.74. Similarly, the luteolin from Tulsi gave the fitness score 38.12. After docking the visualization of the result was done by LIGPLOT. The LIGPLOT program automatically generates schematic two-dimensional representations of protein-ligand complexes from standard PDB file input. The output is a color, or black-and-white, PostScript file giving a simple and informative representation of the intermolecular interactions and their strengths, including hydrogen bonds, hydrophobic interactions, and atom accessibilities. The program is completely general for any ligand and can also be used to show other types of interaction in proteins and nucleic acids. It gives the residues with which the ligand is binded with the target protein.[20] For ligand catechin, the main interacting residue is aspartic acid 167, arginine 92. Similarly for luteolin the main interacting residue is threonine 165, tyrosine 30, and lysine 95 in Figure 1.

CONCLUSION

The increase of multidrug resistant has led to the evolution of many notorious pathogens such as *V. cholerae*. This study concluded that the computer-based screening is an effective alternative for the remedies when all antibiotics seem to be failed. *In silico* ADME and toxicity

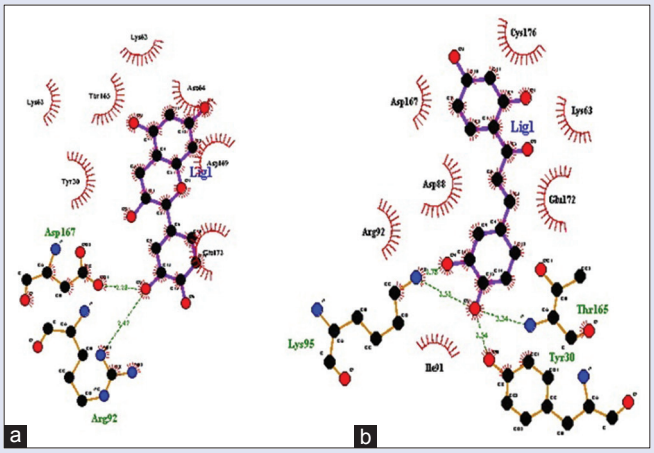


Figure 1: Binding of ligand to the target residues. The main interacting residues of catechin (a) and the main interacting residues of luteolin (b). The residues of catechin are aspartic acid 167 (bond length 2.23 Å) and arginine-92 (bond length 2.47 Å). The residue of luteolin are lysine 95 (bond length 3.75, 3.53 Å), tyrosine 30 (bond length 3.54 Å), and threonine 165 (bond length 3.34 Å)

studies help to identify the best ligand against the target. Several herbal compounds are screened against *V. cholerae*. It was found that herbal compounds such as catechin, luteolin shows good inhibitory action against ToxT (3GBG). However, the *in vitro* study is needed to perform to validate the computer-based result.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Hannan A, Humayun T, Hussain MB, Yasir M, Sikandar S. *In vitro* antibacterial activity of onion (*Allium cepa*) against clinical isolates of *Vibrio cholerae*. J Ayub Med Coll Abbottabad 2010;22:160-3.
2. Kumar P, Mishra DK, Deshmukh DG, Jain M, Zade AM, Ingole KV, *et al*. Haitian variant ctxB producing *Vibrio cholerae* O1 with reduced susceptibility to ciprofloxacin is persistent in Yavatmal, Maharashtra, India, after causing a cholera outbreak. Clin Microbiol Infect 2014;20:O292-3.
3. Kumar P, Jain M, Goel AK, Bhadauria S, Sharma SK, Kamboj DV, *et al*. A large cholera outbreak due to a new cholera toxin variant of the *Vibrio cholerae* O1 El Tor biotype in Orissa, Eastern India. J Med Microbiol 2009;58(Pt 2):234-8.
4. Hannan A, Raja M, Arshad MU, Absar M. 'Gastro' of Mirpur Khas (Sindh) is resolved. J Ayub Med Coll Abbottabad 2008;20:10-2.
5. Tripathi R, Singh Naorem S, Dureja C, Haldar S, Mondal AK, Raychaudhuri S. VopF, a type III effector protein from a non-O1, non-O139 *Vibrio cholerae* strain, demonstrates toxicity in a *Saccharomyces cerevisiae* model. J Med Microbiol 2010;59(Pt 1):17-24.
6. Rafi S, Qureshi AH, Saeed W, Ali A, Ahmadani M, Khawaji SA. Changing epidemiology and sensitivity pattern of *Vibrio cholera* at Rawalpindi. Pak J Med Sci 2004;20:357-60.
7. Jain M, Kumar P, Goel AK, Kamboj DV, Singh L. Class 1 integrons and SXT elements conferring multidrug resistance in *Vibrio cholerae* O1 strains associated with a recent large cholera outbreak in Orissa, Eastern India. Int J Antimicrob Agents 2008;32:459-60.
8. Kumar P, Jain M, Goel AK, Kamboj DV, Kumar O. Tetracycline resistant *V. cholerae* O1 biotype El Tor serotype Ogawa with classical ctxB from a recent cholera outbreak in Orissa, Eastern India. J Infect Public Health 2012;5:217-9.
9. Skariyachan S, Krishnan RS, Siddapa SB, Salian C, Bora P, Sebastian D. Computer aided screening and evaluation of herbal therapeutics against MRSA infections. Bioinformation 2011;7:222-33.
10. Lyskov S, Gray JJ. The RosettaDock server for local protein-protein docking. Nucleic Acids Res 2008;36:W233-8.
11. Praveen S, Saurabh M, Hemant S. To study the therapeutic role of Indian species in the treatment of gastrointestinal disease caused by *Vibrio cholerae*. IJIRSET 2013;2:2371-5.
12. Yamasaki S, Asakura M, Neogi SB, Hinenoya A, Iwaoka E, Aoki S. Inhibition of virulence potential of *Vibrio cholerae* by natural compounds. Indian J Med Res 2011;133:232-9.
13. Li Q, Cheng T, Wang Y, Bryant SH. PubChem as a public resource for drug discovery. Drug Discov Today 2010;15:1052-7.
14. Pence HE, Williams A. ChemSpider: An online chemical information resource. J Chem Educ 2010;87:1123-4.
15. van de Waterbeemd H, Gifford E. ADMET *in silico* modelling: Towards prediction paradise? Nat Rev Drug Discov 2003;2:192-204.
16. Ertl P. Molecular structure input on the web. J Cheminform 2010;2:1.
17. Mondal SI, Khadka B, Akter A, Roy PK, Sultana R. Computer based screening for novel inhibitors against *Vibrio cholerae* using NCI diversity set-II: An alternative approach by targeting transcriptional activator ToxT. Interdiscip Sci 2014;6:108-17.
18. Tjelvar O, Simon B. An Overview of Protein-ligand Docking Using GOLD. Cambridge Crystallographic Data Centre EMBO Workshop; 2014.
19. Kumar P, Mishra DK, Deshmukh DG, Jain M, Zade AM, Ingole KV, *et al*. *Vibrio cholerae* O1 Ogawa El Tor strains with the ctxB7 allele driving cholera outbreaks in south-western India in 2012. Infect Genet Evol 2014;25:93-6.
20. Wallace AC, Laskowski RA, Thornton JM. LIGPLOT: A program to generate schematic diagrams of protein-ligand interactions. Protein Eng 1995;8:127-34.


Sabah Perveen


Hotam Singh Chaudhary

ABOUT AUTHORS

Sabah Perveen is B.tech Biotechnology student.

Prof. Hotam Singh Chaudhary is M.tech biotechnology Asst. Prof. Madhav Institute of Technology and Science.