Applied Biochemistry and Biotechnology (2022) 194:37–53
https://doi.org/10.1007/s12010-021-03716-4

ORIGINAL ARTICLE

Study on the Interaction of Algal Peptides on Virulence Factors of *Helicobacter pylori*: In Silico Approach

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Received: 30 July 2021 / Accepted: 8 October 2021 /
Published online: 11 November 2021

Abstract
In the Asian region, *Helicobacter pylori* infects about 80% populations, which is most leading cause of peptic ulcers, and it is an asymptomatic infection. Studies reported that the particular bacteria carry specific virulence factors that leads to severe complications. These virulence factors can be used as a drug targets to inhibit their growth and pathogenicity. Chronic infection with *H. pylori* virulence factors are CagA, VacA and HtrA positive strains the risk factor of gastric cancer. In this study, we aimed to study the antagonistic interaction pattern between the potential eight algal peptides against the virulence factors of *H. pylori* through in silico analysis intended to treat peptic ulcer and prevent the further complications such as cancer. The proteins of virulent factors are docked using C-Docker algorithm and calculated the bind energy of the complexes. The results showed that the peptide derived from a green alga, *Tetradesmus* sp. are active against the three virulent factors such as cag-A, vac-A, and Htr-A with multiple hydrogen, vdW, electrostatic interactions, and mild π-hydrophobic bindings with the libdock energy score for CagA, VacA and HtrA are 175.625, 158.603 and 89.397 kcal/mol. These primes and the peptide lead to develop a better and potential inhibitors against *H. pylori* infection.

Keywords Antibacterial algal peptides; *H. pylori* · VacA · CagA · HtrA · Libdock

Introduction

H. pylori are more common in poorer nations because of the generally poor sanitation and hygiene conditions caused by poor socio-economic infrastructure. *H. pylori* are the main causative agent of peptic and gastric ulcers. It is a gram-negative, rod-shaped,

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microaerophilic, flagellated bacterium that causes stomach inflammation in more than half of the population; however, the affected individuals are asymptomatic. So this bacterium can enter our bodies and reside in the digestive tract. They cause sores called ulcers which may lead to cancer [1]. The World Health Organization (WHO) and the International Agency for Research on Cancer (IARC) have designated *H. pylori* as a class I carcinogen linked to the development of gastric cancer (GC) since 1994 [2].

Ulcers are the deep lesions penetrating through the entire thickness of the gastrointestinal tract that also lead to cancer because the stomach has a layer of mucus that is designed to protect from stomach acid. *H. pylori* attack this mucus lining and few part of the stomach exposed to acid together with the bacteria, and the acid can irritate the stomach causing ulcers or cancer There are different types of ulcers; most common are atrophy, metaplasia, and peptic and gastric ulcer, which appeared to damage the inner lining of the stomach. Furthermore, *H. pylori* infection is one of the most important risk factors for the development of stomach adenocarcinoma. Peptic ulcers develop due to an imbalance between aggressive factors and protective factors [3]. The virulence of the bacterial strains, environmental variables, and the genetic traits and lifestyles of the hosts all influence the clinical consequences of *Helicobacter pylori* infections. Certain foods or stress were considered to be the cause for peptic ulcer, however after the discovery of *H. pylori* [4].

Ulcers are not only the problems associated with *H. pylori*. Peptic ulcers can block the passage of food through the digestive tract, which causes vomiting and swelling from inflammation. Urease is the most important enzyme produced by the *H. pylori*, since it enables survival of the organism in a low pH environment and also aids colonization in the mucosal membrane of the stomach. The enzymes catalyze the degradation of urea to ammonia and carbon dioxide. Ammonia alkalizes the environment, leading to the neutralization of the acid fluid in the stomach, which allows bacterial survival. Urease was the first virulence factor of *H. pylori* used for diagnostic purposes in gastric pathology [5]. Medicinal plants and algae have achieved their therapeutic properties from their capability to produce renewable and various secondary metabolites which are known as phytochemical constituents. Algae used these phytochemicals as a protection mechanism against pathogen, and those algae are specific protein fragments, and this protein is an essential component required to repair cells, and also algal peptides have less toxicity and more antibacterial properties [6, 7]. Numerous species of algae are reported to be rich in proteins, carbohydrates, and bioactive compounds. Bioactive compounds isolated from algal peptides are showing anti-cancer activity and preliminary anti-COVID-19 [8, 9].

In the previous research Cbf-K16, a cathelicidin-like antimicrobial peptide, demonstrated wide antibacterial action which is followed Cbf-K16 was found to have effective antibacterial and anti-inflammatory properties, as well as down regulating the expression of adhesion- and cytotoxin-related genes in drug-resistant H. pylori, making it a promising anti-infective therapy option [10]. Sun et al. have recently reported that anti-adhesive peptide which is derived from wheat germ protein is effectively inhibited *H. pylori* adhesion to gastric epithelial cells [11]. The aim of this study was to investigate the antagonistic interaction pattern between the potential 8 algal peptides against the virulence factors of *H. pylori* through in silico analysis intended to treat peptic ulcer and prevent the further complication such as cancer.

Materials and Methods

Collections of Virulence Genes *H. pylori*

Literature studies reported about the *H. pylori* and also its virulence genes. Commonly there are several genes that have been implicated in disease related to *H. pylori*. Studies show that several virulence factors are thought to be important once contact with the host cell epithelium. The study reported showed that 21 virulence genes of *H. pylori* are the reason behind the ulcerogenesis, and specifically three gene markers CagA, VacA, and HtrA significantly play the role (Table 1).

Collections of Antimicrobial Algal Peptides

The potent antibacterial, antitumor, and antimicrobial algal peptide sequences (Table 2) were collected. Among the 12 peptides, 8 peptides were selected for the docking studies. Sequences of peptides are (a) peptide 1, VECYGPNRPQF (*Chlorella vulgaris*); (b) peptide 2, NIPP-1(PGWNQWFL) (*Navicula incerta*); (c) peptide 3, NIPP-2 (VEVLPAEL) (*Navicula incerta*); (d) peptide 4, VPGTPKNLDSPR (*Porphyra haitanensis*); (e) peptide 5, GPDRPKFLGPF (*Tetradesmus obliquus*); (f) peptide 6, WYGPDRPKFL (*Tetradesmus obliquus*); (g) peptide 7, SDWDRF (*Tetradesmus obliquus*); and (h) peptide 8, WPRGYFL (*Tetradesmus obliquus*). The aspect of peptide structure is an important factor influencing the conformations adapted by protein and large peptides, and the structures of all 8 algal peptides were fabricated using Discovery Studio software (2017 version) (Fig. 1).

Generation of Stable Confirmation of Peptide Through Minimization

In molecular mechanics simulations, an essential step is first to assign a force field to calculate the potential energy of input molecule. The Discovery Studio client uses a CHARMm-style residue topology file (RTF) to implement force field calculation. Additionally, each of the supported force fields may include some residue patches to allow variations of the residues. To make stable molecules, energy minimization is performed on structures before docking analysis; it relaxes the conformation and removes the steric overlap that produces bad contacts [24]. In the minimization algorithm, max steps are 2000, RMS gradient is 0.01, and the energy change is 0.0. In advanced, partial charge estimation is Momany-Rone fixed in the parameter window. Minimized molecules were subjected to the libdock protocol.

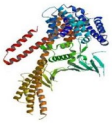
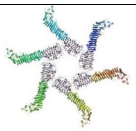
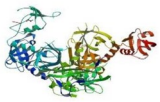
LIB Docking

Libdock is an algorithm for docking small molecules into an active receptor site. Initially, a hotspot map is calculated for the receptor active site which contains polar and a polar group. This hotspot map is subsequently used to rigidly align the ligand conformation to form favorable interactions [25]. In the protocol window, virulence factors are submitted in the input receptor column, and in the ligand column, all 8 minimized peptides are uploaded. The binding spot is chosen based on the receptor cavity in the coordinates of − 5.05467 (X), 60.4518 (Y), and − 24.6123 (Z) with the radius of 10.9 Å. Other docking preferences, conformation method, minimization algorithm, and advanced options are mentioned in Table 3.

Table 1 Comprehensive reports on peptide derived from algal species and its biomedical applications chronologically

S. no	Peptide	Algae	Mode of action	References
1	VECYGPNRPQF	*Chlorella Vulgaris*	Anticancer and antioxidant activities	[12]
2	NIPP-1(pro-gly-trp-asn-gln-trp-phe-leu) NIPP-2(val-glu-val-leu-pro-pro-ala-glu-leu)	*Navicula incerta*	Inhibited ethanol-induced cytotoxicity in HepG2/CYP2E1 cells	[13]
3	Gly-Met-Asn-Asn-Leu-Thr-Pro-Leu-Glu-Gln	*Nannochloropsis oculata*	Fractions on human umbilical vein endothelial cells (HUVECs)	[14]
4	LDAVNR, MMLDF	*Spirulina maxima*	Anti-inflammatory activity	[15]
5	Ile-Ala-Glu, Phe-Ala-Leu, Ala-Glu-Leu, Ile-Ala-Pro-Gly, and Val-Ala-Phe	*Spirulina platensis*	Anti-oxidant activity, anti-hypertensive activity, anti-microbial activity, anti-diabetics activity, and anti-obesity activity	[16]
6	ELWKTF	*Gracilariopsis lamaneiformis*	Antioxidant peptides	[17]
7	WPRGYFL, GPDRPKFLGPF, WYGPDRPKFL, SDWDRF	*Tetradesmus obliquus*	ACE inhibitory activity	[18]
8	PHA, PHP, PHS	*Arthrospira maxima*	Anti-oxidant, anti-hyaluronidase, anti-collagenase, anti-inflammatory activity	[19]
9	FGMPLDR, MELVLR	*Ulva intestinalis*	ACE inhibitory peptides	[20]
10	GVPMPNK, RNPFVFAPTLLTVAAR, LRSELAAWSR	*Spirulina platensis*	Anti-diabetic peptides	[21]
11	AIVFQAQH	*Dunaliella salina*	Anti-osteopenic activity	[22]
12	PIZ, FEIHCC	*Isochrysis zhanjiangensis*	ACE inhibitory peptides	[23]

Table 2 Summary of virulence factor available in *H. pylori* and its pathogenicity

Virulence factor	Molecular structure	Mechanism in pathogenicity	Effects and causes	References
CagA		• Exogenous cancer-promoting gene • Type 4 secretion system • Phosphorylation of tyrosine. • Cytotoxicity-associated immunodominant antigen	• Causes cell proliferation and cell elongation	[24]
VacA		• Multifunctional toxin • Ability to cause vacuolation of cell culture • Type 5 auto transport secretion • Vacuolating cytotoxin auto transporter	• Causes cell vacuolation and cell necrosis. • Enhances the activation of autophagy and increased cell death in ER.	
HtrA		• *H. pylori* can secrete proteases. • All clinical *H. pylori* separate possess htrA gene factor and suppression of HtrA proteolytic activity is adequate to kill *H. pylori.* • Disruption of junction proteins is especially vital for *H. pylori* to take advantage of the host receptors. • Periplasmic serine endoprotease DegP	• Induce the expression of host proteases to cleave extracellular matrix and intracellular junction proteins. • Cleaves the E-cadherin and degrades misfolded proteins	[25]

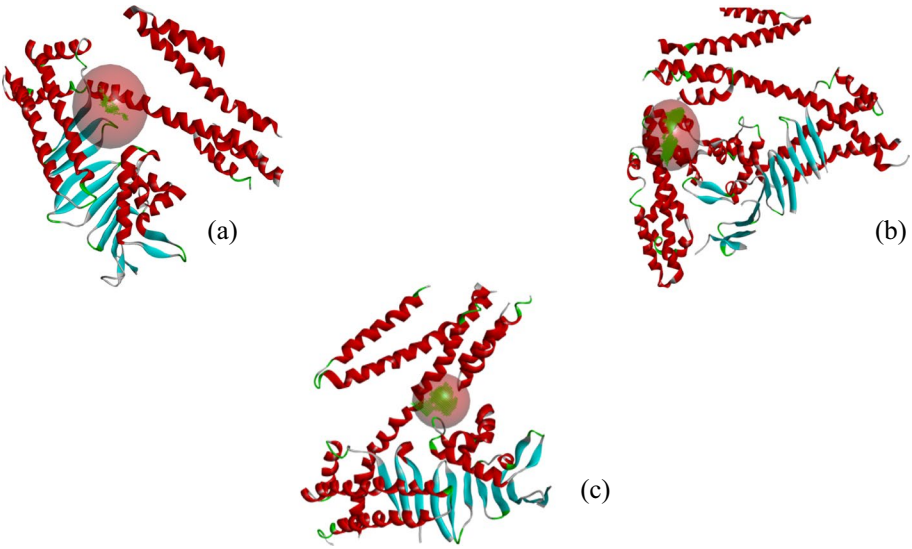


Fig. 1 Secondary structure of the virulence protein CagA (**a**), VacA (**b**), and HtrA (**c**) with the binding site

Results and Discussions

The three main reasons for the selection of these particular virulence markers are as follows: first, CagA (cytotoxin associated gene) is a cancer-promoting gene. It causes cell proliferation and cell elongation. Second, VacA (Vacuolating cytotoxin gene) is a multifunctional toxin, and it causes cell vacuolation. The last is HtrA—suppression of HtrA proteolytic activity is sufficient to kill *H. pylori*. These three proteins were collected from their genes CagA, VacA, and HtrA, respectively, and downloaded from protein databases based on their residues length, classification, and resolution values.

Table 3 Libdock protocol parameter setup

Docking preferences		Minimization algorithm	
Max hits to save		RMSD cutoff	1.0
Max number of hits	100	Minimization max steps	1000
Minimum libdock score	100	Minimization RMS gradient	0.001
Final score cutoff	0.5	Minimization energy change	0.0
Max BFGS steps	50	Minimization force field	CHARMm
Max conformation hits	30	Implicit solvent model	Distance-dielectrics
Max start conformations	1000	Dielectric constant	1
Steric fraction	0.10	Solvent dielectric constant	80
Final cluster radius	0.5	Minimum hydrogen radius	0.8
A polar SASA cutoff	15.0	Non-polar surface constant	0.92
Polar SASA cutoff	5.0	Non-polar surface coefficient	0.00542
Surface grid steps	18	Salt concentration	0.0
Conformation method	FAST	Input atomic radii	Van der Waals radii
Maximum conformations	50	Nonbond list radius	13.0
Discard conformations	True	Nonbond higher cutoff	12.0
Energy threshold	20.0	Nonbond lower cutoff	9.0

Minimization

Energy minimization step for ligands and proteins before docking. Some proteins have less energy value with only in its minimized. Molecular mechanics mainly depend upon three-parameter force field, parameter sets, and minimizing algorithms. The potential energy of the molecules in molecular mechanics is calculated by using force field concepts. A force field is a set of functions and constants used to describe the potential energy of a molecule.

The parameter set includes atomic mass, Vander Waal's radii, bond length, bond angle, the dihedral angle which defines a reference point, and force constants allowing for the calculation of potential energy caused due to the inclusion of attractive or repulsive interactions between atoms. Algorithms to calculate new geometrical positions are so-called minimizers or optimizers [26]. Force field applied three virulence factors, and the 8 peptides were minimized by the smart minimizer (steepest descent 1000 steps + conjugated gradient 1000 steps). It resulted the stable conformers of VacA, HtrA, and CagA with the local energy level of − 46,328.26, 33,085.043, and − 27,465.8 kcal/mol, respectively (Tables 4, 5 and 6).

Similarly the peptide energy was minimized from the range − 295.572 to 16,358.4 kcal/mol to -295.572 to -992.423 kcal/mol. The binding pocked for peptide was selected based on the eraser algorithm, which find the cavities. For vacA XYZ coordinates fixed as a -5.0546, 60.4518, and -10.9000 with the radius of 9.1000 Å. Similarly, the coordinates for HtrA and cagA were found to be − 39.7488, 45.4658, and 4.9687 and − 21.5038, − 25.3823, and − 37.6440, respectively (Fig. 2).

Table 4 Minimized energy results of 8 peptides

S. no	Peptide derived from algae	Force field	Minimization criteria	Initial RMS gradient	Initial potential energy	Potential energy	Van der Waals energy	RMS gradient
1	*Chlorella_vulgaris*	CHARMm	CONJUG > minimization exiting with gradient tolerance (0.0100000) satisfied	0.04593	−572.359	−572.423	−24.024	0.0095
2	NIPP-1	CHARMm	CONJUG > minimization exiting with gradient tolerance (0.0100000) satisfied	0.00991	−301.886	−301.886	−24.835	0.00779
3	NIPP-2	CHARMm	CONJUG > minimization exiting with gradient tolerance (0.0100000) satisfied	0.00951	−341.699	−341.699	−29.439	0.00734
4	*Porphyra*_peptide	CHARMm	CONJUG > Minimization exiting with gradient tolerance (0.0100000) satisfied	0.02003	−425.009	−425.049	−40.527	0.00991
5	*Tetradesmus* (2)	CHARMm	CONJUG > minimization exiting with gradient tolerance (0.0100000) satisfied	0.00933	−387.059	−387.059	−16.767	0.00734
6	*Tetradesmus* (3)	CHARMm	CONJUG > minimization exiting with gradient tolerance (0.0100000) satisfied	0.01	−295.572	−295.572	−5.215	0.00738
7	*Tetradesmus*(4)	CHARMm	CONJUG > minimization exiting with gradient tolerance (0.0100000) satisfied	0.01	−295.572	−295.572	−5.215	0.00738
8	*Tetradesmus* (1)	CHARMm	CONJUG > minimization exiting with gradient tolerance (0.0100000) satisfied	13,956.7	16,358.4	−295.572	−24.62	0.03148

Table 5 Minimized energy results of proteins

Virulence protein	Force field	Initial potential energy (kcal/mol)	Potential energy (kcal/mol)	Van der Waals energy (kcal/mol)	Electrostatic energy (kcal/mol)	Initial RMS gradient (kcal/mol)	Final RMS gradient (kcal/mol)	Minimization criteria
CagA	CHARMm	−13,919.6	−27,465.8	−2,481.29	27,992.736	43.83504	1.12101	CONJUG > minimization exiting with number of steps limit (200) exceeded
VacA	CHARMm	−10,460.11	−46,328.26	−4,102.72	−47,362.04	994.22647	0.40513	CONJUG > minimization exiting with the number of steps limit (500) exceeded
HtrA	CHARMm	−13,956.746	33,085.04390	−2,845.14	33,794.920	111.21234	0.48696	CONJUG > minimization exiting with the number of steps limit (500) exceeded

Table 6 Libdock score for the peptide and virulence factor complex

Peptide name	Virulence factor		
	VacA	**HtrA**	**CagA**
	Libdock score		
Chlorella_vulgaris	N	N	N
NIPP-1	134.878	N	N
NIPP-2	125.310	N	N
Porphyra_peptide	N	N	N
Tetradesmus (2)	140.739	N	166.954
Tetradesmus (3)	158.603	89.397	175.625
Tetradesmus (4)	N	N	N
Tetradesmus (1)	140.112	N	N

**N* no conformation dock, *D* dock, energy indicated in kcal/mol

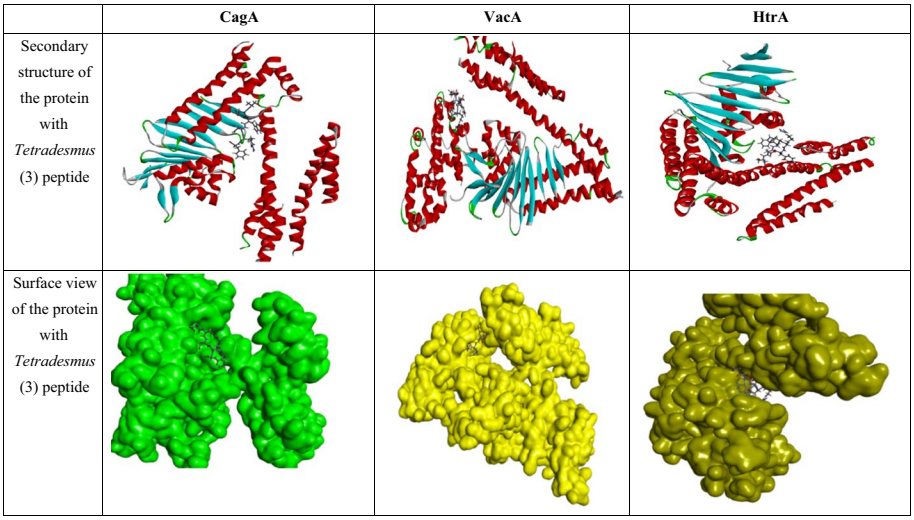


Fig. 2 Secondary structure of the protein CagA, VacA, and HtrA with *Tetradesmus* (3) peptide using LIB-Docking

Docking Pattern Examination

vacA

The docking algorithm run between the vacA virulence factor and 8 peptides results illustrated that, totally 222 conformers generated, 81 poses docked and 2 molecules failed to dock. The failing of the legends due to the clashes of active site amino acids with the amino acids of peptide. Figure 3 shows that the highest binding molecule *Tetradesmus* (3) formed 17 hydrogen bonds and 3 attractive charges that made the molecule bind with highest energy of 158.603 k cal/mol compared to other molecules.

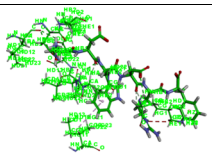
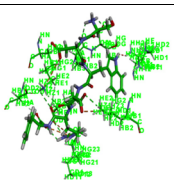
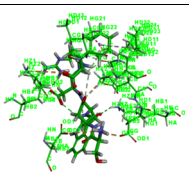
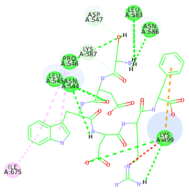
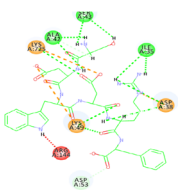
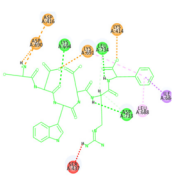
	CagA	VacA	HtrA
Interactions of active amino acids with *Tetradesmus* (3) peptide (3D view)			
Interactions of active amino acids with *Tetradesmus* (3) peptide (2D view)			
Inference	There is no unfavourable donor. ILE A: 675 is the pi- alkyl group and there are more than two conventional hydrogen bond they are marked as green in colour	ARG A:53 is the unfavourable donor donor and ALA A:42, SER A:43, ILE A:35 are the conventional hydrogen bond	LYS A:694 and LEU A: 734 are conventional hydrogen bond and SER A: 437 is the unfavourable donor. There are four attractive charges. They are: ASP A: 416, ASP A:690, LYS A:691, and LYS A:414.

Fig. 3 Interactions of active amino acids of CagA (a, d), Vac (b, e), and Htr (c, f) with *Tetradesmus* (3) peptide

HtrA

Libdock protocol run result of HtrA and 8 peptides showed that the only 1 peptide docked well and other possess are failed and totally 222 conformers generated. The interaction analysis shown in the figure explicates the interaction of docked Tetradesmus (3) peptide with HtrA. Five salt bridges, two hydrogen bonds, two π-alkyls, and one π–σ bond network were formed. Among the 8, *Tetradesmus* (3) only showed better inhibition with the energy of 89.397 kcal/mol.

CagA

Only two ligands docked and 35 conformers generated in the CagA libdock with peptides. *Tetradesmus* (3) and (2) only formed bonding with the CagA. Specifically, Tetradesmus (3) formed around 8 H-bonds, one π-alkyl, and π-cationic interaction that made the molecule fit proper orientation inside the binding pocket of CagA. The binding energy was found to be 175.625 kcal/mol (Table 7).

In silico molecular modeling studies will lead to generation of a potent molecule in less time with reduction in the usage of chemicals and animals and also reduce the expenses. Owing to its invasiveness and pathogenicity, numerous treatment and control methods were employed to target the *H. pylori* [27, 28]. Hence, to develop the vaccine against the pathogen is the significant task and at the same time the discovery and development of antibiotic among the most powerful and successful achievements of modern science and technology for the control of infectious diseases. Prolonged usage of broad spectrum antibiotics leads to the emergence of drug resistance. Apart from that, the algae have achieved their therapeutic properties from their capability to produce renewable and various secondary metabolites which are known as phytochemical constituents, and these phytochemicals were used as a protection mechanism against pathogen.

Table 7 Libdock score of docked peptides with CagA, VacA, and HtrA

Virulence protein vs. algae	Initial potential energy	Initial RMS gradient	Potential energy	RMS gradient	Van der Waals energy	Bond energy	Pose Number	Libdock score
1. CagA								
Tetradesmus(3)	169.227	81.0252	81.0252	0.00997	−5.21477	3.86543	1	175.625
Tetradesmus(3)	169.227	81.0252	81.0252	0.00997	−5.21477	3.86543	2	141.18
Tetradesmus(3)	169.227	81.0252	81.0252	0.00997	−5.21477	3.86543	3	111.035
Tetradesmus(3)	169.227	81.0252	81.0252	0.00997	−5.21477	3.86543	4	94.2794
Tetradesmus(2)	4.63E+09	1.28E+10	1.28E+10	0.00931	−16.7696	2.06886	1	166.954
Tetradesmus(2)	4.63E+09	1.28E+10	1.28E+10	0.00931	−16.7696	2.06886	2	154.852
Tetradesmus(2)	4.63E+09	1.28E+10	1.28E+10	0.00931	−16.7696	2.06886	3	151.356
Tetradesmus(2)	4.63E+09	1.28E+10	1.28E+10	0.00931	−16.7696	2.06886	4	145.022
Tetradesmus(2)	4.63E+09	1.28E+10	1.28E+10	0.00931	−16.7696	2.06886	5	126.874
Tetradesmus(2)	4.63E+09	1.28E+10	1.28E+10	0.00931	−16.7696	2.06886	6	121.566
Tetradesmus(2)	4.63E+09	1.28E+10	1.28E+10	0.00931	−16.7696	2.06886	7	112.027
Tetradesmus(2)	4.63E+09	1.28E+10	1.28E+10	0.00931	−16.7696	2.06886	8	94.0851
CagA	−13,919.60	43.835	43.835	1.12101	−2,481.29	260.438		
2. VacA								
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	1	158.603
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	2	138.674
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	3	133.924
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	4	130.397
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	5	128.84
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	6	120.626
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	7	117.805
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	8	115.054
Tetradesmus(3)	−295.572	0.01	−295.572	−5.21509	0.00738	3.86676	9	113.631

Table 7 (continued)

Virulence protein vs. algae	Initial potential energy	Initial RMS gradient	Potential energy	RMS gradient	Van der Waals energy	Bond energy	Pose Number	Libdock score
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	10	112.362
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	11	111.442
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	12	107.868
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	13	105.028
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	14	104.657
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	15	102.445
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	16	101.433
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	17	100.149
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	18	98.3665
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	19	97.8358
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	20	97.2203
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	21	93.048
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	22	91.6868
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	23	91.4846
Tetradesmus(3)	− 295.572	0.01	− 295.572	− 5.21509	0.00738	3.86676	24	82.9004
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	1	140.739
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	2	140.455
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	3	138.105
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	4	137.644
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	5	137.322
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	6	135.981
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	7	131.023
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	8	128.53
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	9	128.235
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	10	127.863

Table 7 (continued)

Virulence protein vs. algae	Initial potential energy	Initial RMS gradient	Potential energy	RMS gradient	Van der Waals energy	Bond energy	Pose Number	Libdock score
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	11	123.154
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	12	121.385
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	13	120.817
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	14	116.387
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	15	116.189
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	16	113.95
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	17	110.837
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	18	110.477
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	19	107.836
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	20	107.771
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	21	107.399
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	22	107.068
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	23	106.464
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	24	105.606
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	25	101.847
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	26	101.213
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	27	100.591
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	28	100.396
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	29	98.5927
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	30	98.4803
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	31	91.888
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	32	91.1216
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	33	86.5332
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	34	83.4544
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	35	83.1895

Table 7 (continued)

Virulence protein vs. algae	Initial potential energy	Initial RMS gradient	Potential energy	RMS gradient	Van der Waals energy	Bond energy	Pose Number	Libdock score
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	36	81.8417
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	37	81.3287
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	38	76.5339
Tetradesmus(2)	− 387.059	0.00933	− 387.059	− 16.7687	0.00734	2.06844	39	72.6597
Tetradesmus(1)	16,358.40	13,956.70	− 225.97	− 24.6201	0.03148	2.32487	1	162.273
Tetradesmus(1)	16,358.40	13,956.70	− 225.97	− 24.6201	0.03148	2.32487	2	140.112
Tetradesmus(1)	16,358.40	13,956.70	− 225.97	− 24.6201	0.03148	2.32487	3	115.105
Tetradesmus(1)	16,358.40	13,956.70	− 225.97	− 24.6201	0.03148	2.32487	4	94.6154
VacA	− 10,460.10	994.226	− 46,328.30	− 4,102.72	0.40513	454.658		
NIPP-2	− 341.699	0.00951	− 341.699	− 29.4392	0.00734	2.35601	1	125.31
NIPP-2	− 341.699	0.00951	− 341.699	− 29.4392	0.00734	2.35601	2	119.634
NIPP-2	− 341.699	0.00951	− 341.699	− 29.4392	0.00734	2.35601	3	114.32
NIPP-2	− 341.699	0.00951	− 341.699	− 29.4392	0.00734	2.35601	4	84.2202
NIPP-2	− 341.699	0.00951	− 341.699	− 29.4392	0.00734	2.35601	5	66.3894
NIPP-2	− 341.699	0.00951	− 341.699	− 29.4392	0.00734	2.35601	6	64.8138
NIPP-2	− 341.699	0.00951	− 341.699	− 29.4392	0.00734	2.35601	7	64.6833
NIPP-2	− 341.699	0.00951	− 341.699	− 29.4392	0.00734	2.35601	8	62.6854
NIPP-1	− 301.886	0.00991	− 301.886	− 24.835	0.00779	2.2159	1	134.878
NIPP-1	− 301.886	0.00991	− 301.886	− 24.835	0.00779	2.2159	2	132.048
NIPP-1	− 301.886	0.00991	− 301.886	− 24.835	0.00779	2.2159	3	120.157
NIPP-1	− 301.886	0.00991	− 301.886	− 24.835	0.00779	2.2159	4	110.074
NIPP-1	− 301.886	0.00991	− 301.886	− 24.835	0.00779	2.2159	5	105.985

Table 7 (continued)

Virulence protein vs. algae	Initial potential energy	Initial RMS gradient	Potential energy	RMS gradient	Van der Waals energy	Bond energy	Pose Number	Libdock score
NIPP-1	− 301.886	0.00991	− 301.886	− 24.835	0.00779	2.2159	6	105.837
3.HtrA								
Tetradesmus(3)	− 295.572	0.01		− 5.21509	0.00738	3.86676	1	89.397
HtrA	− 13,956.70	111.212		− 2,845.14	0.48696	313.222		

CagA: input ligands, 8; conformers generated, 35; ligands failed to dock, 4; poses docked, 12

VacA: input ligands, 8; conformers generated, 222; ligands failed to dock, 2; poses docked, 81

HtrA: input ligands, 8; conformers generated, 222; ligands failed to dock, 7

Conclusion

There is a tremendous need for novel antimicrobial agents from different microbes in general and microalgae in particular provides important sources of chemical compounds and secondary metabolites which have many therapeutic applications. In this study, microalgal peptide derived from *Tetradesmus* sp. exhibited as antibacterial and inhibitor of *H. pylori*. It almost acts on all the three selected virulence factors, *VacA*, *CagA*, and *HtrA* through antagonizing effect. These peptides could be lead molecules to treat the *H. pylori*-induced diseases as well as to develop a vaccine.

Acknowledgements DM is thankful to the SEED/AUN-NET JICA through Special Program on Research Against Corona (SPRAC 2020) program. DM and RS are thankful to BSACIST funded through Crescent Seed Money (CSD/CSM/3 dated 11/03/2021).

Author Contribution DM: Corresponding author, guidance, and data analysis. TA: Data acquisition and manuscript writing. RS: Data analysis. NI: Data analysis.

Data Availability Not applicable.

Declarations

Ethics Approval Not applicable.

Consent to Participate Not applicable.

Consent for Publication Not applicable.

Conflict of Interest The authors declare no competing interests.

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