(RESEARCH ARTICLE)

Discovery of antiadhesins of *Helicobacter pylori* from existing drugs and medicines for malaria ventures pathogen box compounds

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Abstract

Background: *Helicobacter pylori* infection is a worldwide problem with more than half of the world's population in both developed and developing countries are infected with this organism. The best-characterized *H. pylori* adhesins, Blood group antigen binding Adhesin (BabA) and Sialic acid binding Adhesin (SabA) are virulent factors which facilitate adhesion of the bacteria to the host cells.

Methods: We determined the binding affinities of selected existing drugs and medicines for malaria venture pathogen box compounds to *H. pylori* adhesin receptors by molecular docking simulations. The 3D crystal structures of *H. pylori* adhesin receptors were obtained from Protein Data Bank (PDB). The receptors were prepared for molecular docking simulations using PyMol 1.3, Chimera 1.9 and AutoDock tools 1.5.6. The 3D structures of the selected existing drugs and Medicines for Malaria Ventures (MMV) pathogen box compounds were obtained from ZINC and PubChem databases. They were prepared for molecular docking simulations using AutoDock tools 1.5.6. Docking protocols were validated by reproducing the PDB crystal structures in silico. Molecular docking simulations were executed with a virtual screening script using AutoDock Vina 1.1.2 on a Linux platform.

Results: Entacapone, sildenafil, gemcitabine, tolcapone, rabeprazole, tolazamide, teriflunomide, sulfamethazine, cefotetan, talbutal, mitotane, tolbutamide, piperazine showed higher average binding affinities than the reference compound nitazoxanide molecular dynamics of one front runner with the reference ligand and protein were done at 1000 ps. Rabeprazole showed lower stability than the reference drug after molecular dynamics simulation.

Conclusion: The identified existing drugs from molecular docking simulations with higher average binding affinities are predicted as possible *H. pylori* multi-target antiadhesins.

Keywords: *Helicobacter pylori*; Anti-adhesins; Molecular docking; Antiadhesins

1. Introduction

Helicobacter organisms were originally placed in the genus *Campylobacter. Helicobacter pylori* (*H. pylori*) infection is a worldwide problem, more than half of the world's population in both developed and developing countries are infected

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with this organism. *Helicobacter pylori* was formerly called *Campylobacter pylori* [1]. By 1989, it was renamed as *Helicobacter pylori* and recognized to be associated closely with antral gastritis (gastric and duodenal ulcers in adults and children). The organisms are 'S' spiral shaped Gram-negative bacteria [2]. Infection by *H. pylori* produces hyper gastrinaemia and gastric acid hyper secretion, the increase in gastrin is what causes increase in acid secretion in the duodenum, therefore resulting in ulceration. *H. pylori* infection may lead to acute gastritis (abdominal pain, nausea and vomiting) within two weeks following infection, many patients infected with the organism have recurrent abdominal symptoms (non-ulcer dyspepsia) without ulcer disease. In the absence of ulcer-inducing medication of non-steroid anti-inflammatory drugs (NSAID) such as aspirin and ibuprofen, *H. pylori* is present in 90% of duodenal ulcer (DU) patients and 70% gastric ulcer (GU) patients [3]. It is highly unlikely that chronic infection with *Helicobacter pylori* could occur in the absence of adhesin-host cell interactions. Also, there is no evidence that any of the serious outcomes of *H. pylori* infection such as gastric and duodenal ulcers, gastric cancer, or mucosa-associated lymphoid tissue (MALT) lymphoma could occur without prior colonization of the gastric epithelium mediated by *H. pylori* adhesins. *H. pylori* is highly adaptable, as evidenced by the fact that it can occupy a single host for decades [4]. An important facet of this adaptability is its ability to physically interact with various types of host cells and with host mucins and extracellular matrix proteins using a number of different adhesins displaying a variety of unique receptor specificities. *Helicobacter pylori* specifically colonizes the human gastric epithelium and is the major causative agent for ulcer disease and gastric cancer development. *H. pylori* is a Gram-negative, micro-aerophilic and spiral bacterium that affects more than half of the world's population. Infection with *H. pylori* is associated with a variety of clinical presentations, from asymptomatic to serious diseases, *H. pylori* colonization can lead to the development of several upper gastrointestinal conditions, including chronic gastritis, increased risks of peptic ulcer, gastric cancer, and mucosa-associated lymphoid tissue lymphoma.

It is estimated that about 90% of the drugs fail during development in phase 1 clinical trial and it takes billions of dollars in investment and average of 15 years to bring a new drug to the market, but this approach (computer aided drug design) of drug discovery saves money, time and unnecessary resources in the drug development processes. Hence, the imperative need for dry lab via molecular docking technology.

The study aims to find existing drugs and Medicines for Malaria Ventures (MMV) pathogen box compounds with antiadhesin potentials in the management of ulcers.

2. Material and methods

2.1. Software and hardware

The following computer softwares were used: PyMol-1.3 (http://pymol.org/ep), Autodockvina 1.5.6 (http://autodock.scripps.edu/resources/adt), Autodock vina 1.1.2 (http://vina.scripps.edu/), Chimera 1.9 (https://www.cgl.ucsf.edu.chimera/), Molinspiration (www.molinspiration.com)**,** Gromacs and Linux operating systems (Ubuntu 12.04) Personal computer hardware used for this work is HP pavilion g series (Windows 7, A4 VISION AMD with 600GB of hard disk and 4GB RAM)

2.2. Databases

The following databases were used: ZINC database (http://zinc.docking.org/search/structure) and Protein Data Bank (PDB) (http://www.rscb.org/pdb/), and MMV database (http://www.mmv.dndi.org).

2.3. Bioinformatics mining- identification of drug target

A search for the drug target, *H. pylori* adhesins was done to identify the potential drug targets. Literature mining was done to have a proper understanding of the individual drug targets.

2.4. Selection and preparation of protein structure

After identification of several targets, literature mining and analysis, a proper target for SabA and BabA receptors were selected, a total of three receptors were selected. The targets were checked in the Protein Data Bank website PDB and the proteins bound to their reference ligands with the lowest resolution were selected. The presence of the bound ligands in ZINC database was also checked. Preparations of the targets were done by downloading the experimental crystal structure of the receptors bound to the drug with PDB accession code from protein data bank (www.rscb.org). The saved files were opened in Chimera-1.10.1 individually and the containing ligand together with other unwanted components were deleted and saved as PDB file. The saved files were opened in Autodock tools vs-1.5.6 and polar hydrogen were added and saved as pdbqt file individually. The grid search spaces were also selected as appropriate [5]

2.5. Selection and preparation of ligand

The mol2 file of the approved ligand that binds to the receptors were downloaded from the ZINC database website (http://zinc.docking.org/) and saved in an appropriate folder. The molecular mass, log p and tPSA for the ligands were noted from the ZINC database. SMILES format of the ligands was copied to Molinspiration (www.molinspiration.com) for biological activity prediction and the following parameters; the ion channel modulator, protease inhibitor kinase inhibitor, G-protein coupled receptor, enzyme inhibitor and the nuclear receptor ligand were noted. The entire data for the bound ligands that were noted was inserted into the bioactivity database containing the data of several ligands. The bound ligand was then used as a standard to sort and select other ligands close to it which has the tendency to interact with the receptor. The mol2 file for the selected ligands were downloaded from ZINC database and saved appropriately. Using Autodock tools, the ligands were prepared by clicking on the ligand option on the tool then clicking on input and then open. The mol2 file of each ligand to be prepared was selected and opened. The ligand option on the toolbar was selected again and torsion tree was selected and then to choose torsion and finally the non-rotatable bonds were made rotatable by clicking on the option. The ligand option on the tool bar was selected again, then to output and saved as PDBQT. The above procedure was repeated for each of the selected ligands.

2.6. Validation of docking protocol

To validate the docking protocol, the experimental crystal structure of the targets bound to their reference drugs were reproduced *in silico*.

2.7. Molecular docking simulations

The docking simulation was done using Vina in Linux platform for the configuration files/scripts used for the simulations. The best docking scores were examined and compared with the experimentally determined structure of the ligand [5]

2.8. Post docking analysis

The analysis of results that were obtained from docking process was done by visualizing the positions of different models of the docked ligands in the binding pocket of the protein in PyMol-v13rl-edu.

2.9. Molecular dynamics

To incorporate biomolecular dynamics in our investigation, molecular dynamics simulations were performed on representative coordinates of the free and target-frontrunner complexes using Groningen Machine for Chemical Simulations (GROMACS) package [5] [6]. The crystal structures of the three *H. pylori* adhesin receptors obtained from PDB [7] with one possible anti-adhesin (Rabeprazole) and reference ligand were subjected to MD simulation.

3. Results

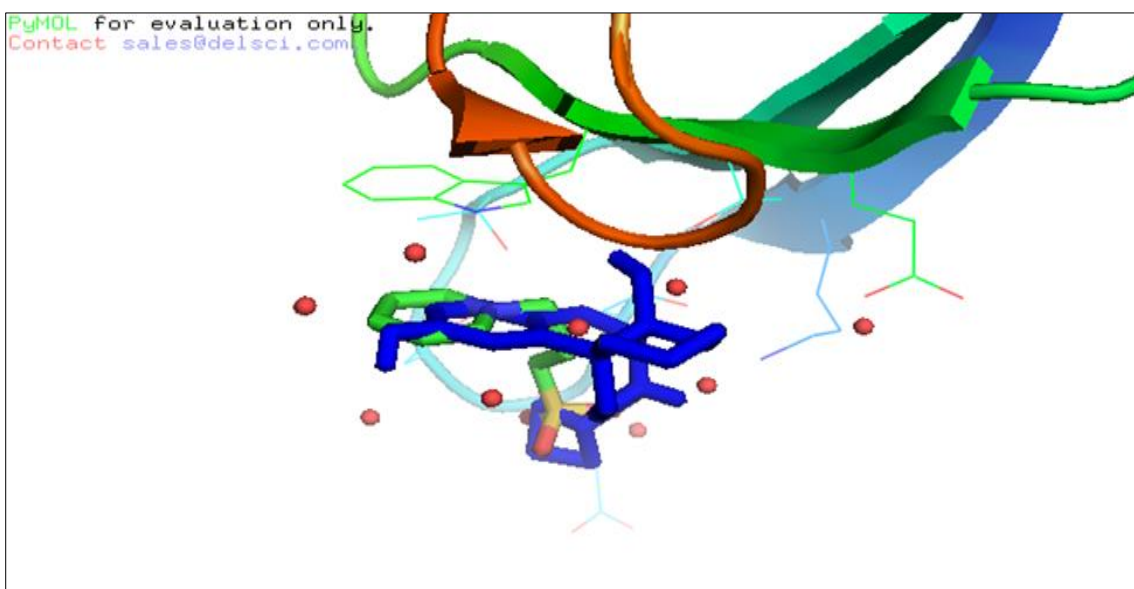## 3.1. Molecular docking results

Table 1 The selected PDB structures of the three targets

The table shows the result of the protein mining from protein data bank. The result showed the PDB codes of the protein, the names and resolutions.

PDB CODE	Name of the targets	Resolution ("Å" angstrom)
4o5j	Crystal structure of SabA from *Helicobacter pylori*	2.2
5f7k	Crystal structure of BabA from *H.pylori*	2.17
5f7n	Blood group antigen binding adhesion of *H.pylori*	2.28

3.2. Result of validation of docking protocol

In order to prove that our docking simulation protocol is able to successfully predict anti-adhesin activity of the tested reference drug (nitazoxanide), validation of molecular docking simulation protocol was carried out. The ability of the protocol to reproduce wet laboratory binding of the reference compounds to the receptor was successfully implemented in *silico.*
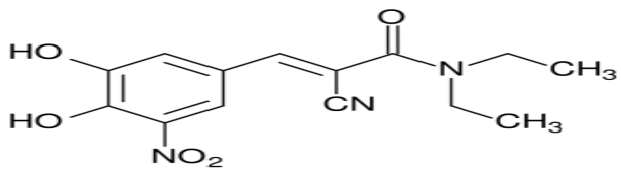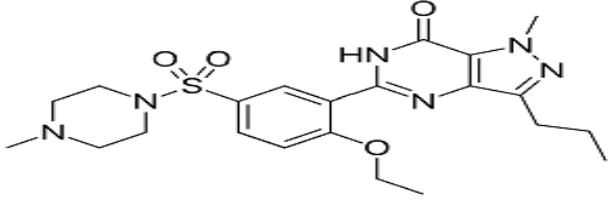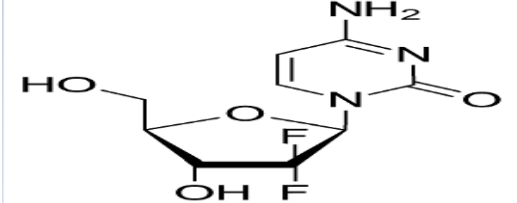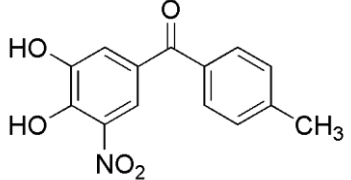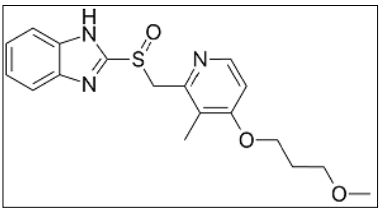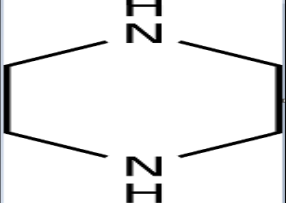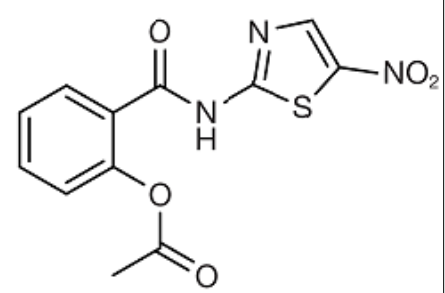
Figure 1 Result of validation of docking protocol

This table shows the *in silico* binding affinities of the selected existing drugs with higher average values than the reference compound (nitazoxanide).

Table 2 Molecular docking results of the selected existing drugs with *H. pylori* adhesin three receptors

S/N	Drugs	4o5j	5f7k	5f7n	Mean ± SD
1	Entacapone3_35342784	-8.4±0.0	-8.3±0.1	-5.3±0.0	-7.3±1.8
2	Sidenafil2_38595321	-7.9±0.0	-7.7±0.1	-6.3±0.0	-7.3±0.9
3	Gemcitabine4_6532	-7.9±0.0	-8.3±0.1	-4.9±0.1	-7.0±1.9
4	Entacapone1_35342787	-7.8±0.0	-7.6±0.1	-5.2±0.1	-6.8±1.4
5	Tolcapone_35342789	-7.4±0.0	-7.8±0.1	-5.3±0.0	-6.8±1.4
6	Entacapone2_5121	-7.4±0.0	-7.7±0.1	-5.4±0.1	-6.8±1.2
7	Rabeprazole_12496506	-7.1±0.0	-7.1±0.1	-5.8±0.1	-6.6±0.8
8	Tolazamide_57512	-7.4±0.0	-7.5±0.1	-4.7±0.1	-6.5±1.6
9	Teriflunomide_13512456	-7.1±0.0	-6.8±0.1	-5.5±0.0	-6.4±0.8
10	Sulfamethazine_57494	-7.4±0.0	-7.4±0.1	-4.50.1	-6.4±1.7
11	Cefotetan_3830441	-7.7±0.0	-6.2±0.1	-4.8±0.0	-6.2±1.5
12	Talbutal2_12503087	-7.3±0.0	-7.2±0.1	-4.0±0.1	-6.2±1.9
13	Mitotane2_1530726	-7.1±0.0	-7.0±0.1	-4.4±0.0	-6.2±1.5
14	Mitotane1_1530725	-7.4±0.0	-6.9±0.1	-4.2±0.1	-6.1±1.7
16	Tolbutamide_1530703	-7.0±0.0	-6.7±0.1	-4.5±0.1	-6.0±1.3
17	Piperazine4_3886212	-6.7±0.0	-6.8±0.1	-4.6±0.0	-6.0±1.2
18	Nitazoxanide_3956788	-7.4±0.1	-6.6±0.0	-4.0±0.1	-6.0±1.8

Table 3 Structures and functions of some selected existing drugs

S/N	Drugs	Functions	Structures
1.	Entacapone	Medication used for the treatment of Parkinson's disease	
2.	Sildenafil	Used for the treatment of erectile dysfunction	
3.	Gemcitabine	Used for the treatment of cancer	
4.	Tolcapone	Medication used to treat Parkinson's disease	
5.	Rabeprazole	Used for the treatment of ulcer	
6.	Piperazine*	Used for the treatment of intestinal worms	
7.	Nitazoxanide	Used for the treatment of various helminthic, protozoal and viral infections	

This table shows the *in-silico* binding affinities of the MMV compounds with higher average values than the reference compound (nitazoxanide).

Table 4 Molecular docking results of the MMV pathogen box compounds with *H. pylori* adhesin three receptors

S/N	MMV Drugs	4o5j	5f7k	5f7n	Mean ± SD
1	Emin_MMV099637	-8.4±0.0	-8.3±0.1	-6.6±0.1	-7.8±1.0
2	Emin_MMV659010	-8.4±0.0	-8.3±0.1	-6.6±0.1	-7.8±1.0
3	Emin_MMV676269	-8.4±0.0	-8.3±0.1	-6.6±0.1	-7.8±1.0
4	Emin_MMV676472	-8.4±0.0	-8.3±0.1	-6.6±0.1	-7.8±1.0
5	Emin_M mMV676605	-8.4±0.0	-8.3±0.1	-6.6±0.1	-7.8±1.0
6	Emin_MMV1037162	-7.9±0.0	-8.3±0.1	-6.6±0.1	-7.6±0.9
7	Emin_MMV009054	-7.9±0.0	-8.3±0.1	-6.6±0.1	-7.6±0.9
8	Emin_MMV019807	-7.9±0.0	-8.3±0.1	-6.3±0.1	-7.5±1.1
9	Emin_MMV020710	-7.9±0.1	-8.3±0.0	-6.3±0.0	-7.5±1.1
10	Emin_MMV024035	-7.9±0.1	-8.3±0.0	-6.3±0.0	-7.5±1.1
11	Emin_MMV102872	-7.9±0.1	-8.3±0.0	-6.3±0.0	-7.5±1.1
12	Emin_MMV661713	-7.9±0.1	-8.3±0.0	-6.3±0.0	-7.5±1.1
13	Emin_MMV676270	-7.9±0.1	-8.3±0.0	-6.3±0.0	-7.5±1.1
14	Emin_MMV676474	-7.9±0.1	-8.3±0.0	-6.3±0.0	-7.5±1.1
15	Emin_MMV676877	-7.9±0.0	-7.8±0.0	-6.3±0.0	-7.3±0.9
16	Emin_MMV687251	-7.9±0.0	-7.8±0.0	-6.3±0.0	-7.3±0.9
17	Emin_MMV687801	-7.9±0.0	-7.8±0.0	-6.3±0.0	-7.3±0.9
18	Emin_MMV688327	-7.9±0.0	-7.8±0.0	-5.8±0.1	-7.2±1.2
19	Emin_MMV688470	-7.9±0.0	-7.8±0.1	-5.8±0.1	-7.2±1.2
20	Emin_MMV688756	-7.9±0.0	-7.8±0.1	-5.8±0.1	-7.2±1.2
21	Emin_MMV688854	-7.9±0.0	-7.8±0.1	-5.8±0.1	-7.2±1.2
22	Emin_MMV689028	-7.9±0.0	-7.8±0.1	-5.8±0.1	-7.2±1.2
23	Emin_MMV085499	-7.8±0.1	-7.8±0.1	-5.8±0.1	-7.1±1.2
24	Emin_MMV658993	-7.8±0.1	-7.8±0.1	-5.8±0.1	-7.1±1.2
25	Emin_MMV676204	-7.8±0.1	-7.8±0.1	-5.8±0.1	-7.1±1.2
26	Emin_MMV676468	-7.8±0.1	-7.8±0.1	-5.8±0.1	-7.1±1.2
27	Emin_MMV676603	-7.8±0.0	-7.8±0.1	-5.8±0.1	-7.1±1.2
28	Emin_MMV062221	-7.7±0.0	-7.8±0.1	-5.5±0.1	-7.0±1.3
29	Emin_MMV560185	-7.7±0.0	-7.8±0.1	-5.5±0.1	-7.0±1.3
30	Emin_MMV676064	-7.7±0.0	-7.8±0.1	-5.5±0.1	-7.0±1.3
31	Emin_MMV676412	-7.7±0.0	-7.7±0.1	-5.5±0.1	-6.9±1.3
32	Emin_MMV676571	-7.7±0.0	-7.7±0.1	-5.5±0.1	-6.9±1.3
33	Emin_MMV000016	-7.7±0.0	-7.7±0.0	-5.5±0.1	-6.9±1.3
34	Emin_MMV690103	-7.7±0.0	-7.7±0.0	-5.5±0.1	-6.9±1.3

35	Emin_MMV000011	-7.5±0.0	-7.7±0.0	-5.5±0.0	-6.9±1.3
36	Emin_MMV004168	-7.5±0.0	-7.7±0.0	-5.5±0.0	-6.9±1.3
37	Emin_MMV011511	-7.5±0.0	-7.7±0.0	-5.4±0.0	-6.9±1.3
38	Emin_MMV020165	-7.5±0.0	-7.7±0.0	-5.4±0.0	-6.9±1.3
39	Emin_MMV021660	-7.5±0.0	-7.7±0.0	-5.4±0.0	-6.9±1.3
40	Emin_MMV024406	-7.5±0.0	-7.7±0.0	-5.4±0.0	-6.9±1.3
41	Emin_MMV687703	-7.5±0.0	-7.7±0.0	-5.4±0.0	-6.9±1.3
42	Emin_MMV688122	-7.5±0.0	-7.7±0.0	-5.4±0.0	-6.9±1.3
43	Emin_MMV688361	-7.5±0.0	-7.7±0.0	-5.4±0.0	-6.9±1.3
44	Emin_MMV688514	-7.5±0.0	-7.7±0.0	-5.3±0.0	-6.8±1.3
45	Emin_MMV688771	-7.5±0.0	-7.7±0.0	-5.3±0.0	-6.8±1.3
46	Emin_MMV688936	-7.5±0.0	-7.7±0.1	-5.3±0.0	-6.8±1.3
47	Emin_MMV689255	-7.5±0.0	-7.7±0.1	-5.3±0.0	-6.8±1.3
48	Emin_MMV690102	-7.5±0.0	-7.6±0.0	-5.3±0.1	-6.8±1.3
49	Emin_MMV090930	-7.4±0.0	-7.6±0.0	-5.3±0.1	-6.7±1.3
50	Emin_MMV188296	-7.4±0.1	-7.6±0.0	-5.3±0.1	-6.7±1.3
51	Emin_MMV659004	-7.4±0.1	-7.6±0.0	-5.3±0.1	-6.7±1.3
52	Emin_MMV671636	-7.4±0.1	-7.6±0.0	-5.3±0.1	-6.7±1.3
53	Emin_MMV676260	-7.4±0.1	-7.6±0.0	-5.3±0.1	-6.7±1.3
54	Emin_MMV676377	-7.4±0.1	-7.6±0.1	-5.3±0.1	-6.7±1.3
55	Emin_MMV676470	-7.4±0.1	-7.5±0.1	-5.3±0.1	-6.7±1.3
56	Emin_MMV676480	-7.4±0.1	-7.5±0.1	-5.3±0.1	-6.7±1.2
57	Emin_MMV676604	-7.4±0.1	-7.5±0.1	-5.3±0.1	-6.7±1.2
58	Emin_MMV687146	-7.4±0.1	-7.5±0.1	-5.3±0.1	-6.7±1.2
59	Emin_MMV002817	-7.4±0.1	-7.5±0.1	-5.3±0.1	-6.7±1.2
60	Emin_MMV006372	-7.4±0.1	-7.5±0.1	-5.2±0.1	-6.7±1.3
61	Emin_MMV006833	-7.4±0.1	-7.5±0.1	-5.2±0.1	-6.7±1.3
62	Emin_MMV010576	-7.4±0.1	-7.5±0.1	-5.2±0.1	-6.7±1.3
63	Emin_MMV011765	-7.4±0.1	-7.5±0.1	-5.2±0.0	-6.7±1.3
64	Emin_MMV012074	-7.4±0.1	-7.4±0.1	-5.2±0.0	-6.6±1.3
65	Emin_MMV020081	-7.4±0.0	-7.4±0.1	-5.2±0.0	-6.6±1.3
66	Emin_MMV020291	-7.4±0.1	-7.4±0.1	-5.2±0.0	-6.6±1.3
67	Emin_MMV020321	-7.4±0.1	-7.4±0.1	-5.1±0.0	-6.6±1.3
68	Emin_MMV021052	-7.4±0.1	-7.4±0.1	-5.1±0.0	-6.6±1.3
69	Emin_MMV022236	-7.4±0.1	-7.4±0.1	-5.1±0.0	-6.6±1.3
70	Emin_MMV023183	-7.4±0.1	-7.4±0.0	-5.1±0.0	-6.6±1.3
71	Emin_MMV024195	-7.4±0.1	-7.4±0.0	-5.1±0.0	-6.6±1.3
72	Emin_MMV024829	-7.4±0.1	-7.4±0.0	-5.1±0.1	-6.6±1.3

73	Emin_MMV026020	-7.4±0.1	-7.4±0.1	-5.1±0.1	-6.6±1.3
74	Emin_MMV687696	-7.4±0.1	-7.4±0.1	-4.9±0.0	-6.5±1.5
75	Emin_MMV687729	-7.4±0.1	-7.4±0.1	-4.9±0.0	-6.5±1.5
76	Emin_MMV687747	-7.4±0.1	-7.4±0.1	-4.9±0.0	-6.5±1.5
77	Emin_MMV687812	-7.4±0.1	-7.4±0.1	-4.9±0.0	-6.5±1.5
78	Emin_MMV688125	-7.4±0.1	-7.4±0.1	-4.9±0.0	-6.5±1.5
79	Emin_MMV688179	-7.4±0.1	-7.4±0.0	-4.9±0.0	-6.5±1.5
80	Emin_MMV688350	-7.4±0.1	-7.2±0.0	-4.9±0.0	-6.5±1.5
81	Emin_MMV688364	-7.4±0.1	-7.2±0.0	-4.8±0.0	-6.5±1.5
82	Emin_MMV688372	-7.4±0.1	-7.2±0.0	-4.8±0.0	-6.5±1.5
83	Emin_MMV688474	-7.4±0.0	-7.2±0.1	-4.8±0.0	-6.5±1.5
84	Emin_MMV688547	-7.4±0.0	-7.2±0.1	-4.8±0.1	-6.5±1.5
85	Emin_MMV688550	-7.4±0.0	-7.2±0.1	-4.8±0.1	-6.5±1.5
86	Emin_MMV688763	-7.4±0.0	-7.2±0.1	-4.8±0.1	-6.5±1.5
87	Emin_MMV688774	-7.4±0.0	-7.2±0.1	-4.8±0.1	-6.5±1.5
88	Emin_MMV688793	-7.4±0.0	-7.2±0.1	-4.7±0.1	-6.4±1.5
89	Emin_MMV688891	-7.4±0.0	-7.1±0.1	-4.7±0.1	-6.4±1.5
90	Emin_MMV688939	-7.4±0.0	-7.1±0.1	-4.7±0.1	-6.4±1.5
91	Emin_MMV688942	-7.4±0.0	-7.1±0.1	-4.7±0.1	-6.4±1.5
92	Emin_MMV689061	-7.4±0.0	-7.1±0.0	-4.7±0.1	-6.4±1.5
93	Emin_MMV689480	-7.4±0.0	-7.1±0.1	-4.7±0.1	-6.4±1.5
94	Emin_MMV689758	-7.4±0.0	-7.1±0.1	-4.7±0.1	-6.4±1.5
95	Emin_MMV003152	-7.4±0.0	-7.1±0.1	-4.7±0.1	-6.4±1.5
96	Emin_MMV010764	-7.4±0.0	-7.1±0.1	-4.7±0.1	-6.4±1.5
97	Emin_MMV020120	-7.4±0.0	-7.1±0.0	-4.6±0.1	-6.3±1.5
98	Emin_MMV021057	-7.4±0.0	-7.1±0.0	-4.6±0.1	-6.3±1.5
99	Emin_MMV024311	-7.4±0.0	-7.0±0.0	-4.6±0.1	-6.3±1.5
100	Emin_MMV687699	-7.4±0.0	-7.0±0.0	-4.6±0.1	-6.3±1.5
101	Emin_MMV687813	-7.4±0.0	-7.0±0.1	-4.6±0.1	-6.3±1.5
102	Emin_MMV688352	-7.4±0.0	-7.0±0.1	-4.6±0.1	-6.3±1.5
103	Emin_MMV688508	-7.4±0.0	-7.0±0.1	-4.6±0.1	-6.3±1.5
104	Emin_MMV688766	-7.4±0.1	-7.0±0.0	-4.6±0.1	-6.3±1.5
105	Emin_MMV688921	-7.4±0.1	-7.0±0.0	-4.6±0.1	-6.3±1.5
106	Emin_MMV689243	-7.4±0.1	-6.9±0.1	-4.6±0.0	-6.3±1.5
107	Emin_MMV001561	-7.3±0.1	-6.9±0.1	-4.5±0.0	-6.2±1.5
108	Emin_MMV007920	-7.3±0.1	-6.9±0.0	-4.5±0.0	-6.2±1.5
109	Emin_MMV019742	-7.3±0.1	-6.9±0.0	-4.5±0.0	-6.2±1.5
110	Emin_MMV020623	-7.3±0.1	-6.9±0.0	-4.5±0.0	-6.2±1.5

111	Emin_MMV023969	-7.3±0.1	-6.9±0.0	-4.5±0.1	-6.2±1.5
112	Emin_MMV687246	-7.3±0.1	-6.9±0.0	-4.5±0.1	-6.2±1.5
113	Emin_MMV687798	-7.3±0.1	-6.8±0.0	-4.5±0.1	-6.2±1.5
114	Emin_MMV688283	-7.3±0.0	-6.8±0.0	-4.5±0.1	-6.2±1.5
115	Emin_MMV688467	-7.3±0.0	-6.8±0.0	-4.5±0.1	-6.2±1.5
116	Emin_MMV688754	-7.3±0.0	-6.8±0.0	-4.5±0.0	-6.2±1.5
117	Emin_MMV688852	-7.3±0.0	-6.8±0.0	-4.5±0.1	-6.2±1.5
118	Emin_MMV688994	-7.3±0.0	-6.8±0.0	-4.5±0.1	-6.2±1.4
119	Emin_MMV003270	-7.1±0.0	-6.8±0.0	-4.5±0.1	-6.1±1.4
120	Emin_MMV011229	-7.1±0.0	-6.8±0.0	-4.5±0.1	-6.1±1.4
121	Emin_MMV020136	-7.1±0.1	-6.8±0.0	-4.5±0.1	-6.1±1.4
122	Emin_MMV021375	-7.1±0.1	-6.8±0.1	-4.5±0.1	-6.1±1.4
123	Emin_MMV024397	-7.1±0.1	-6.8±0.1	-4.5±0.0	-6.1±1.4
124	Emin_MMV687700	-7.1±0.1	-6.8±0.1	-4.5±0.0	-6.1±1.4
125	Emin_MMV688029	-7.1±0.1	-6.8±0.1	-4.5±0.0	-6.1±1.4
126	Emin_MMV688360	-7.1±0.1	-6.8±0.1	-4.5±0.0	-6.1±1.4
127	Emin_MMV688509	-7.1±0.1	-6.8±0.1	-4.5±0.0	-6.1±1.4
128	Emin_MMV688768	-7.1±0.0	-6.8±0.1	-4.5±0.0	-6.1±1.4
129	Emin_MMV688934	-7.1±0.0	-6.8±0.1	-4.5±0.0	-6.1±1.4
130	Emin_MMV689244	-7.1±0.0	-6.8±0.1	-4.5±0.0	-6.1±1.4
131	Emin_MMV001499	-7.1±0.0	-6.8±0.1	-4.5±0.0	-6.1±1.4
132	Emin_MMV007803	-7.1±0.0	-6.7±0.1	-4.5±0.0	-6.1±1.4
133	Emin_MMV019721	-7.1±0.0	-6.7±0.1	-4.5±0.0	-6.1±1.4
134	Emin_MMV020591	-7.1±0.0	-6.7±0.1	-4.5±0.1	-6.1±1.4
135	Emin_MMV023953	-7.1±0.0	-6.7±0.1	-4.5±0.1	-6.1±1.4
136	Emin_MMV200748	-7.1±0.0	-6.7±0.1	-4.5±0.1	-6.1±1.4
137	Emin_MMV675968	-7.1±0.0	-6.7±0.1	-4.5±0.1	-6.1±1.4
138	Emin_MMV676379	-7.1±0.0	-6.7±0.1	-4.5±0.1	-6.1±1.4
139	Emin_MMV676492	-7.1±0.0	-6.7±0.1	-4.5±0.1	-6.1±1.4
140	Emin_MMV687170	-7.1±0.1	-6.7±0.1	-4.5±0.1	-6.1±1.4
141	Emin_MMV687243	-7.1±0.1	-6.6±0.1	-4.5±0.1	-6.1±1.4
142	Emin_MMV687796	-7.1±0.1	-6.6±0.1	-4.4±0.1	-6.0±1.4
143	Emin_MMV688279	-7.1±0.1	-6.6±0.1	-4.4±0.1	-6.0±1.4
144	Emin_MMV688466	-7.1±0.1	-6.6±0.1	-4.4±0.1	-6.0±1.4
145	Emin_MMV688704	-7.1±0.1	-6.6±0.1	-4.4±0.1	-6.0±1.4
146	Emin_MMV688846	-7.1±0.1	-6.6±0.1	-4.4±0.1	-6.0±1.4
147	Emin_MMV688991	-7.1±0.1	-6.6±0.1	-4.4±0.1	-6.0±1.4
148	Emin_zincNitazoxanide_3956788	-7.1±0.1	-6.3±0.1	-3.7±0.1	-5.7±1.8

Binding of one of the front runner (rabeprazole) to *H. pylori* adhesin receptor 5f.
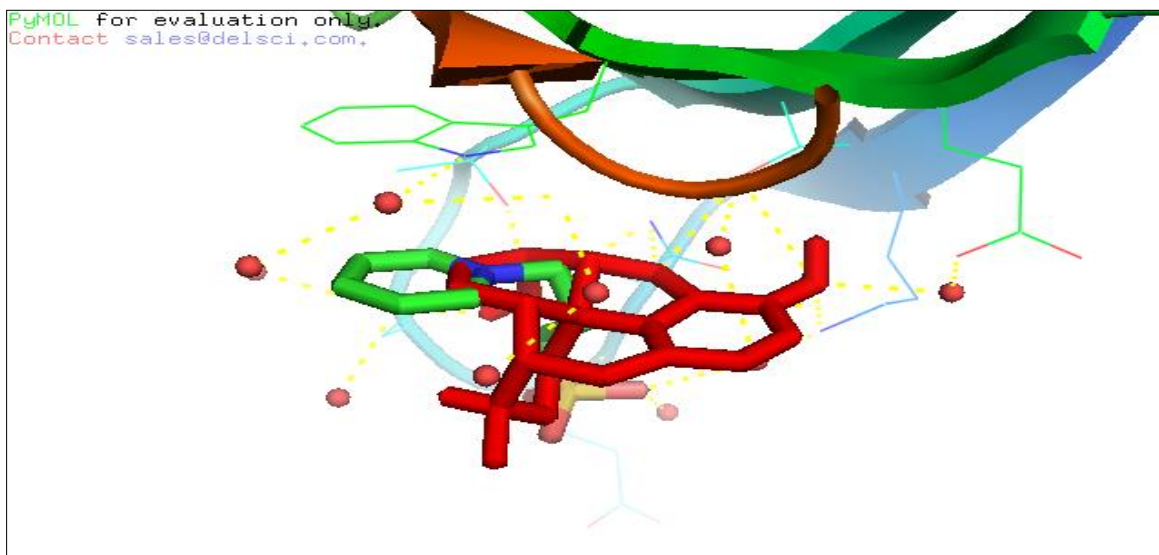

Figure 2 The binding of the front runner (Rabeprazole) to *H. pylori* adhesin receptor 5f7n

3.3. Molecular Dynamics Results

Results from the trajectories of energy minimization, position restrain dynamics simulation and production run were analysed and presented below:

3.3.1. Energy minimization results

From the figure below, the ligands and receptor are in their ground states (0).
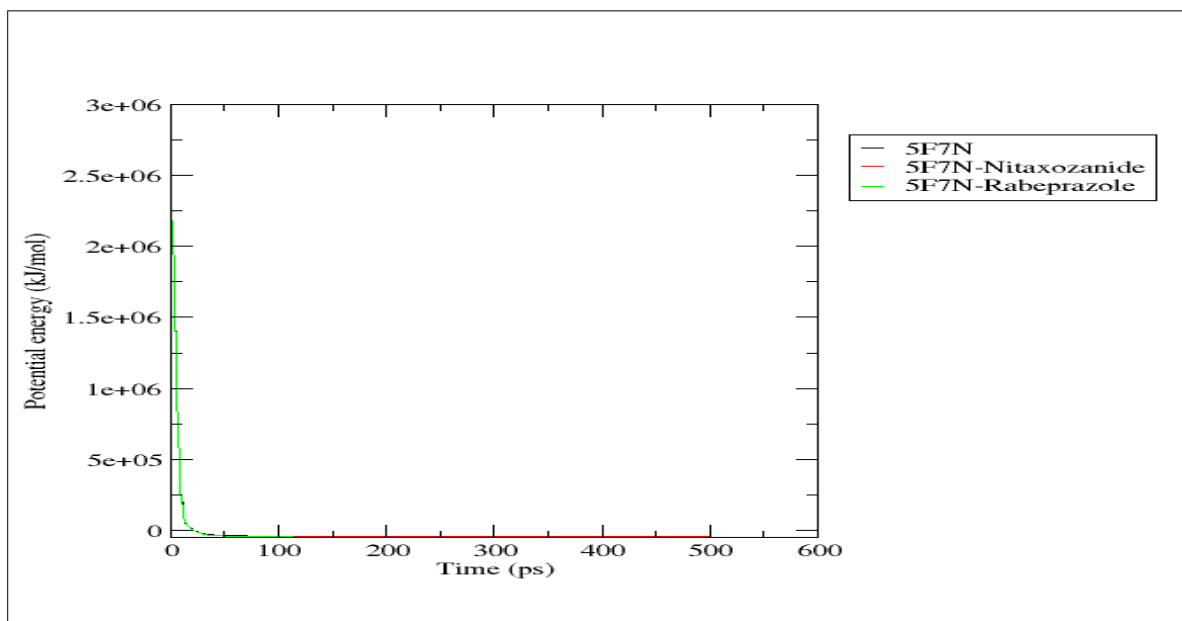
Figure 3 *In vacuo* energy minimization results of the ligands and receptor
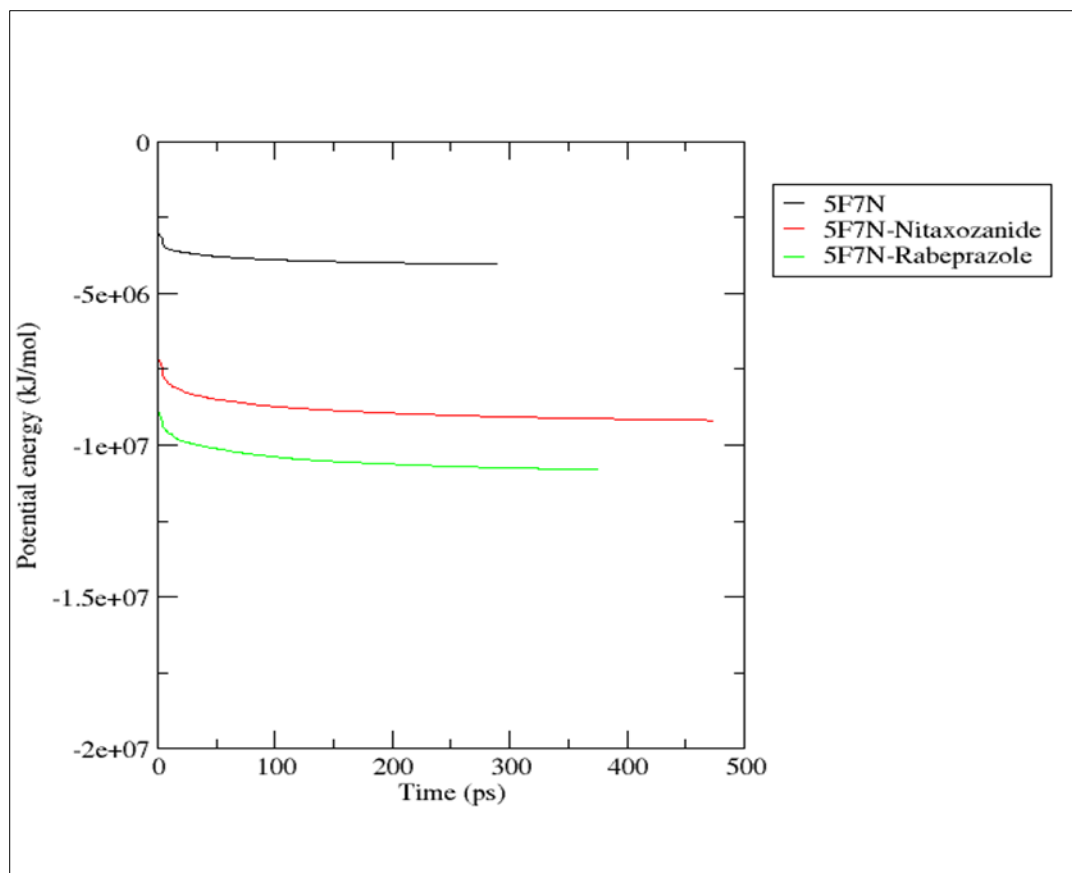
Figure 4 Energy minimization after salvation and neutralization for the ligands and receptor

3.3.2. Position restrain dynamics simulation results

Position restraint dynamics was done to avoid drastic rearrangement of critical parts of the system
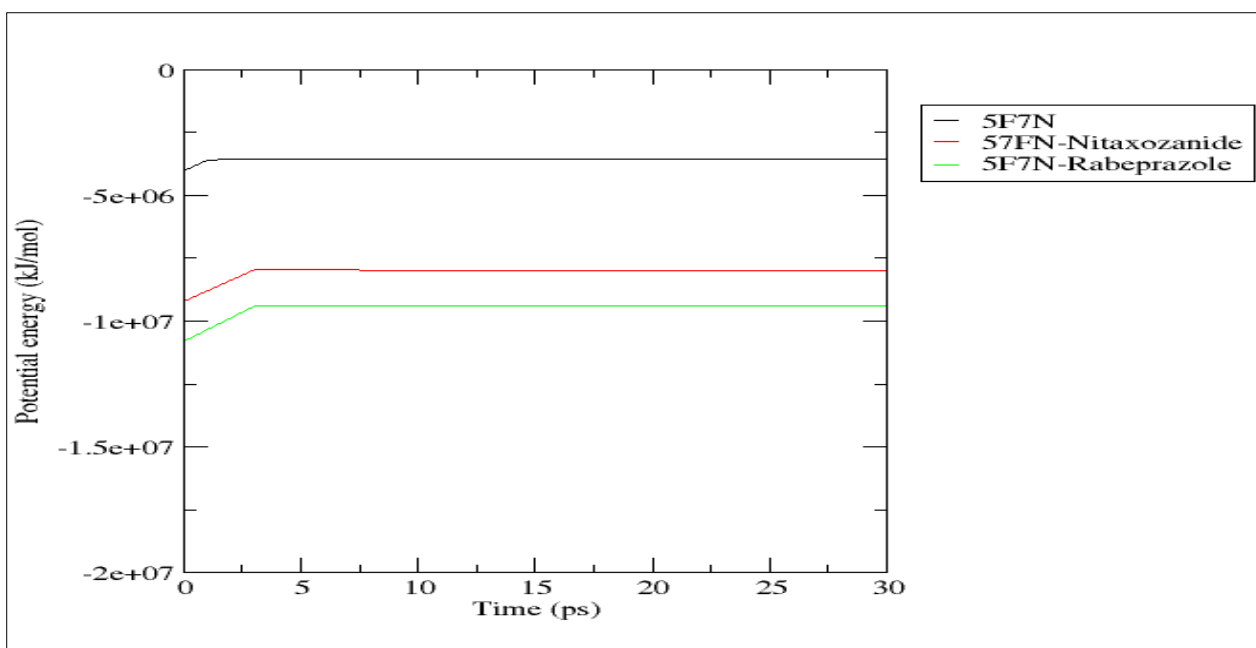

Figure 5 Position restrain dynamics simulation results of the drug target and bound front runner ligands
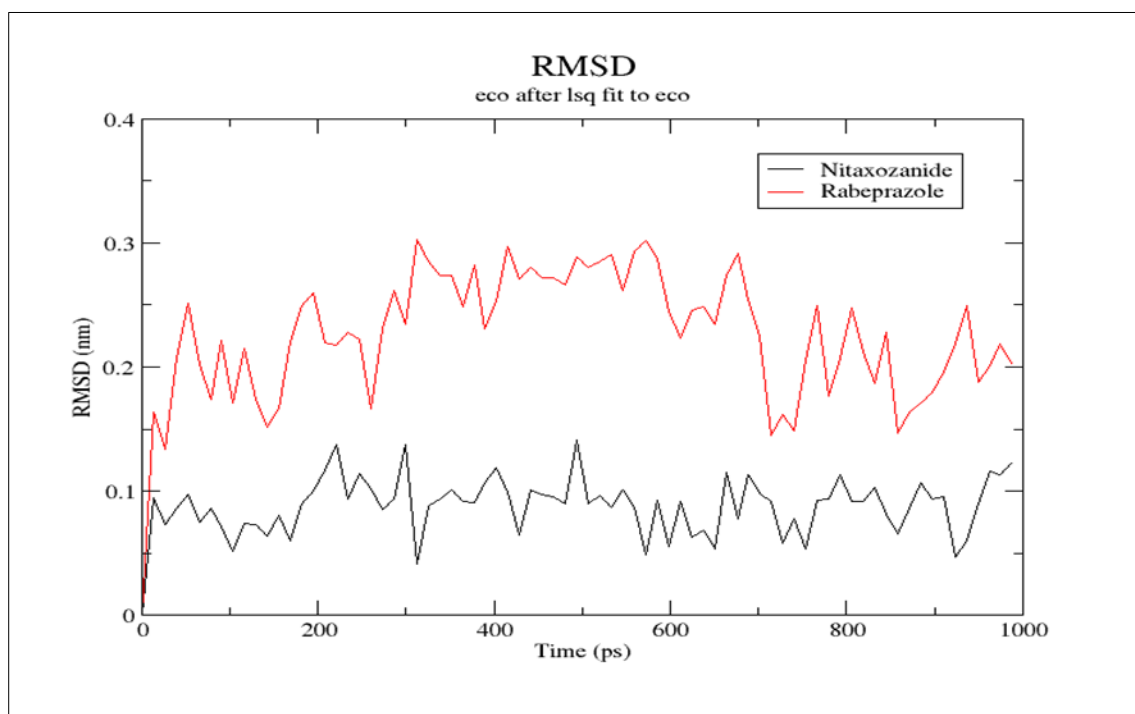
3.3.3. Production run results

Different results from the production run were computed from the production run trajectories. The results includes: stability profile analysis, flexibility profiles, radius of gyration and interaction.

Stability profile analysis
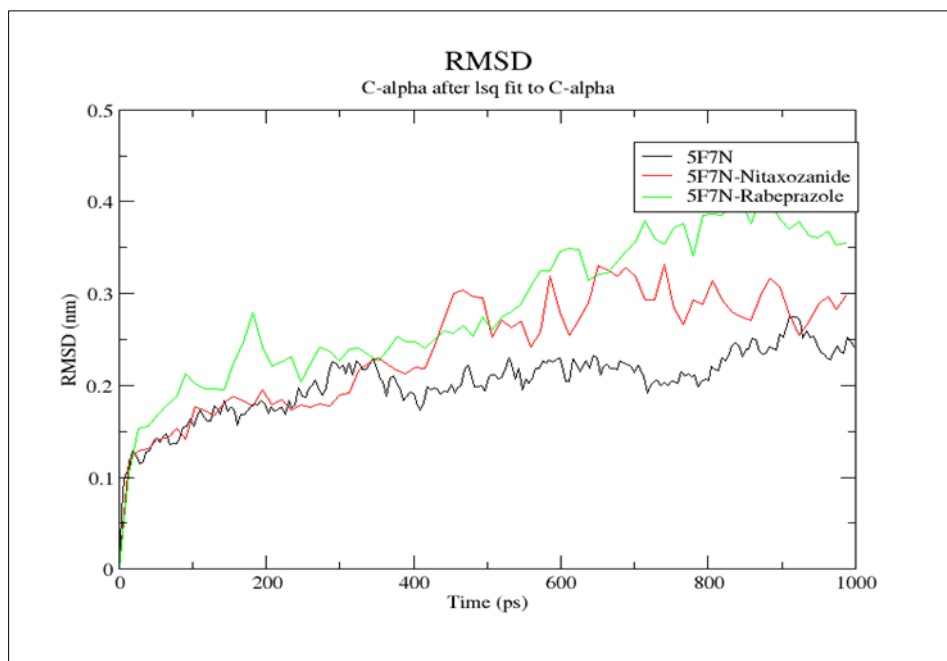
Figure 6 RMSD plot of the molecular dynamics simulation of the drug target and ligands

Figure 7 RMSD plot of the molecular dynamics simulation of the front runner ligands
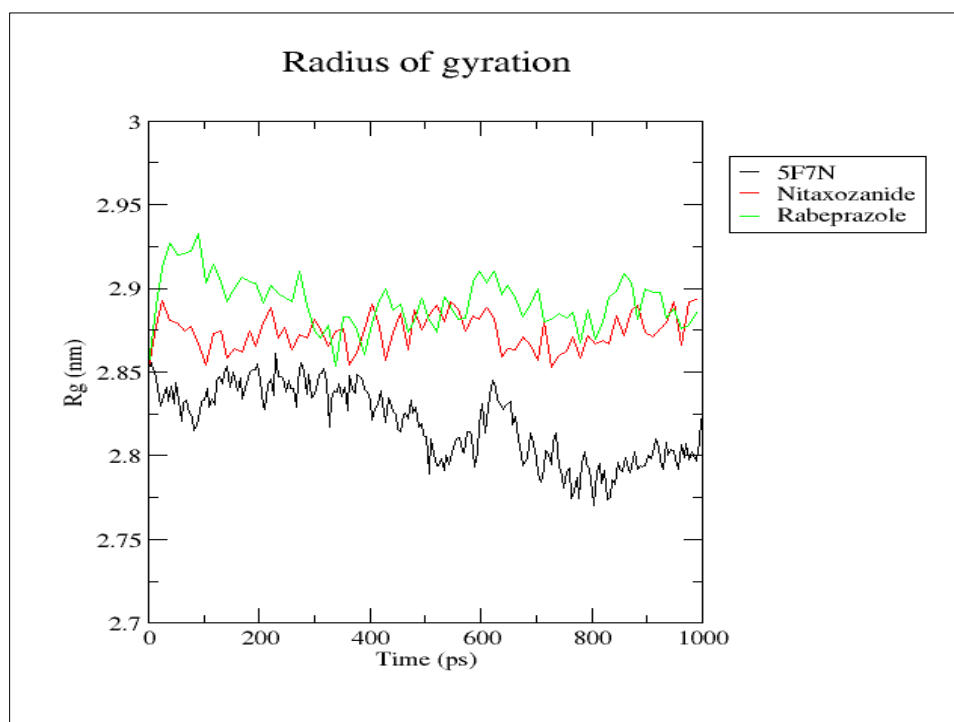
Radius of gyration

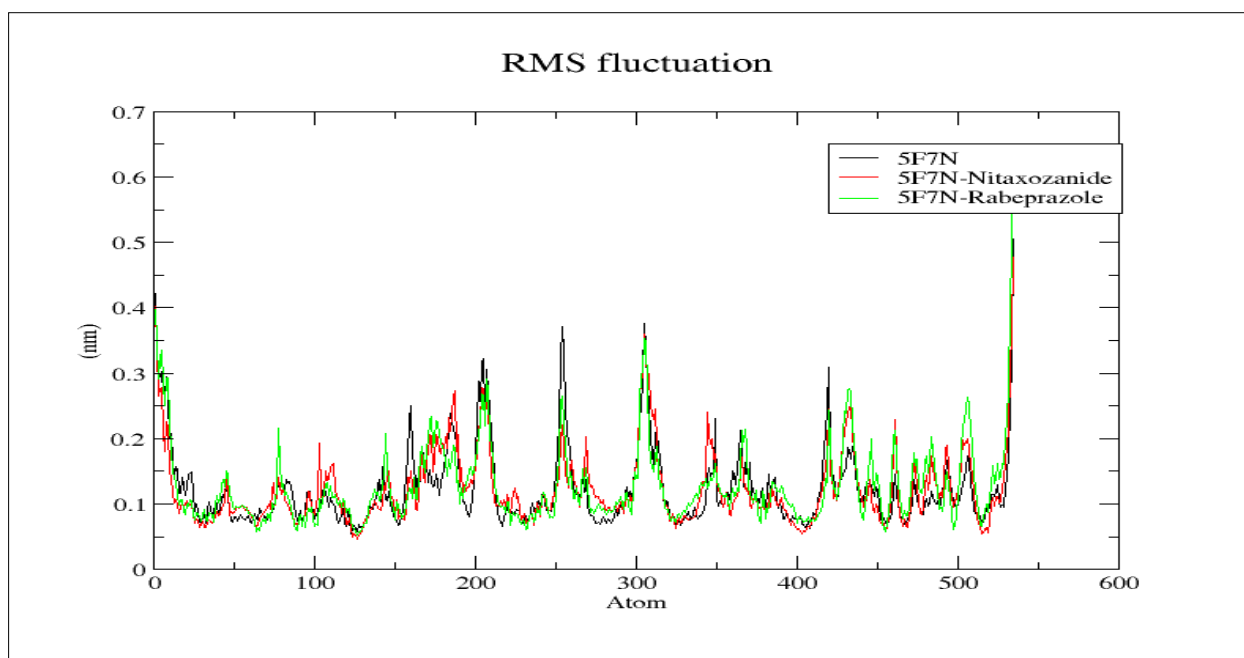**Figure 8** Radius of gyration plot

Flexibility profile analysis

Figure 9 RMSF plot of the molecular dynamics simulation of the drug target and ligands

4. Discussion

Table 1, showed the selected PDB structures of *H. pylori* adhesins. The selection was done based on the resolution. The resolution suggests or measures the quality of the data collected on the crystal containing the protein. This means that

the targets with low resolution values close to 1Å suggests the quality data thereby predicting the possibility of no missing residues on the crystalline structure. 4O5J has a resolution of 2.2Å, 5F7K 2.17Å and 5F7N 2.28Å.

Table 2, Showed molecular docking results of the selected existing drugs with *H. pylori* adhesin three receptors. From the results obtained, There are seventeen (17) existing drugs with higher average values than the reference compound (nitazoxanide) and can be predicted as potential *H. pylori* anti-adhesins.

Table 3, Shows the functions and chemical structures of some selected existing drugs.

Table 4, Showed molecular docking results of the MMV compounds with *H. pylori* adhesin three receptors. From the results obtained, There are one hundred and forty-seven (147) MMV compounds with higher average values than the reference compound (nitazoxanide) and can be predicted as potential *H. pylori* anti-adhesins.

Fig.1. shows the result of validation of docking protocol, to prove that our docking simulation protocol can successfully predict anti-adhesin activity of the tested reference drug (nitazoxanide), validation of molecular docking simulation protocol was carried out. The ability of the protocol to reproduce wet laboratory binding of the reference compounds to the receptor was successfully implemented in *silico.*

Figure 3 and Figure 4 shows the *in-vacuo* energy minimization results and energy minimization of solvated molecular systems respectively. This indicates successful removal of restraining forces in the molecular coordinates and systems at global energy minima. From the graph it can be seen that the systems (drug targets and ligand) are low, that is tending towards zero on the y-axis. This means that the geometry of the drug target and its bound frontrunner were optimized and brought to a relatively global energy minima.

From the results in fig 5. 30 pico seconds (ps) position restrain dynamics simulation of the molecular systems showed well soaked systems by removing restraining forces on the target and target front-runner complexes and allowing water molecules to move.

Fig 6 and 7 Shows the stability profile of the protein ligand complex with that of the protein alone and protein-ligand complex in terms of root mean standard deviation (RMSD). It can be deduced from the graph that rabeprazole in red is less stable than the reference ligand and the protein alone.

Fig 8, Shows Radius of gyration plot. The radius of gyration measures the structure compactness profile of the ligand and shows various degree of fluctuation. The lesser the fluctuation the higher the stability. From the plot it can be deduced that the reference ligand in red is more stable than the rabeprazole in green.

Fig 9, Residues contributing to complex structural fluctuations can be accessed by root mean square fluctuations (RMSF) of each residue. Analysis of the RMSF shows the amino acid residues involved in the complex and their differences at different residue. It can however be deduced from the graph that rabeprazole is less stable than the reference ligand.

It can however be deduced from the graph that rabeprazole is less stable than the reference ligand. Molecular docking simulations of three receptors (4o5j, 5f7k and 5f7n) and several ligands were done and front runner drugs were obtained from the results which are predicted to be antiadhesins: Entacapone3 -7.3kcal/mol, Sildenafil2 -7.3kcal/mol, Gemcitabine4 -7.0kcal/mol, Entacapone1 -6.8kcal/mol, Tolcapone -6.8kcal/mol, Entacapone2 -6.8kcal/mol, Rabeprazole -6.6kcal/mol, Tolazamide -6.5kcal/mol, Teriflunomide -6.4kcal/mol, Sulfamethazine -6.4kcal/mol, Cefotetan -6.2kcal/mol, Talbutal2 -6.2 kcal/mol, Mitotane2 -6.2kcal/mol, Mitotane1 -6.1kcal/mol, Tolbutamide -6.0kcal/mol, Piperazine4 -6.0 kcal/mol showed higher average binding affinities than the reference compound Nitazoxanide -6.0kcal/mol.

Molecular dynamics simulations of one of the front runners (rabeprazole) showed lower stability than the reference drug.

5. Conclusion

Molecular docking simulations of the three receptors gave us front runners which had higher binding affinities than the reference ligand and can be predicted to be potential antiadhesins of *Helicobacter pylori*.

Molecular dynamics simulation using rabeprazole further showed less stability than the reference compound. It can be concluded from this work that the selected existing drugs and the MMV compounds from molecular docking simulations with higher average binding affinities can be predicted as possible *H.pylori* antiadhesins. They can also be predicted as multi-target inhibitors of *H.pylori* adhesins.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare no conflict of interest.

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