

**IN SILICO STUDIES ON DENGUE AND RHINO VIRAL STRUCTURAL PROTEINS
WITH SELECTED *CORIANDRUM SATIVUM* L LEAVES CONSTITUENTS****¹Mahesh K., ¹Nagarjun N., ¹Aniruddha B.S., ¹Ashwini B.M., ²Murugan Rajadurai and ^{*2}Balasubramanian Sathyamurthy**¹Department of Biochemistry, Ramaiah College of Arts, Science and Commerce, Bangalore – 560054.²Professor, Department of Biochemistry, Ramaiah College of Arts, Science and Commerce, Bangalore – 560054.***Corresponding Author: Dr. Balasubramanian Sathyamurthy**

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ABSTRACT

Dengue virus contains two structural proteins. Rhino virus contains four structural proteins. These are considered to be the most effective for drug designing. Phytochemicals present in *Coriandrum sativum* L. Functions as aromatizing agents and has relaxant activity in the alimentary tract. They are found to have antibacterial and anticancerous properties. In this study, the binding efficiency of 4 compounds that are present in the *Coriandrum sativum* L. with all the selected structural proteins were performed through Insilico methods. Our molecular docking result, we found that Dodecanal has the highest binding affinity with the selected dengue and Rhino structural proteins.

KEYWORDS: Coriander, Dodecanal, molecular docking, binding affinities.**1. INTRODUCTION**

Medicinal plants have always been in the forefront for their role in the development of human culture. Medicinal plants have always had a dominant role in health care systems where herbal medicine has been used since ages.^[1] It is an important element of indigenous medical systems all over the world. The ethno botany provides a rich resource for research and development of natural drugs.^[2] Among all the plants *Coriandrum sativum* L, also called as coriander (kitchen herb) plays a great role for its therapeutic uses. Coriander in the language of Sanskrit is called as “Dhanyaka” which is said to have Carminative and Anthelmintic properties.^[3] *Coriandrum sativum* L belonging to family *Apiaceae* is very important for their nutritional and medicinal properties. *Coriandrum sativum* has two varieties i.e. vulgare and microcaprum, the former has larger fruits (3-5mm diameter) while the later has smaller fruits (1.5-3 mm). Their chemical composition varies among different parts of the same plant.^[4] *Coriander sativum* plants are considered as one of the most important source of medicine and drugs with many secondary metabolites and essential oils and they are utilised as formulations for health benefits.^[5] They are recommended for treatment of alzheimers, cancer, dysentery, indigestion, parasitic disease, insomnia, skin disorders, rheumatism, menstrual disorders etc.^[6,7] GC-MS chromatogram of the hydrodistilled extract of *Coriandrum sativum* L herb showed four major peaks: Dodecanal (Synonym: Lauraldehyde), E-2- Dodecanol (Synonym: Dodecan-2-

ol), Decanal (synonym: Decylaldehyde) and E-2-Decenol (synonym: Trans-2-Decen-1-ol) were the major components in the extract. Decanal has the property of Antifungal activity, Aromatic activity and Antimicrobial activity. E-2-Decenol is widely used as a food additive, and has antifungal activities. E-2-Dodecanol is reported to have flavour enhancing properties and is used as a flavouring agent.^[8]

Dengue, a haemorrhagic fever^[9], is caused due to all four serotypes of dengue virus (DENV-1, DENV-2, DENV-3 and DENV-4).^[10] These viruses contain ten proteins out of which three are structural proteins and seven are non structural proteins.^[11] The seven non structural proteins are capsid protein, envelope protein, NS1 protein, transmembrane domain of NS2A, NS2B/NS3 protease, NS3 helicase and NS5 protein. NS2B-NS3 protease is an important enzyme for the viral replication process as it is a hetero dimeric protein of NS2B and NS3 protein.^[12] The N-terminal of the NS3 protein forms association with the NS2B cofactor which is crucial for the viral replication. NS2B/ NS3 protease has an important role in the viral life cycle.^[13] Envelope protein is a structural protein which has a major role in the viral assembly. The protein which is utilised for this study is the envelope protein domain III of the dengue type 4 viruses (strain Dominica / 814669 / 1981). It is classified under structural protein immune system.^[14] The capsid protein is one of the structural proteins, which has a major function in the encapsidation of the viral genome. The capsid protein used for this study was from dengue virus

type 2 (strain Puerto Rico/PR159-S1/1969).^[15] The protein used for this study was the trans-membrane domain of the NS2A of dengue virus type 2. NS2A is a non structural protein and it is a component of viral replication complex which is functionally active in the assembly of the virion and also it acts as an antagonist to the host immune response.^[16] NS3 helicase belongs to the non-structural and a multi-domain dengue virus replication protein.^[17] The protein used for this study is the non-structural 5 (NS5) protein from the dengue virus type 3 (strain Sri Lanka / 1266 / 2000). This protein is classified under the transferases. The RNA dependent RNA polymerase (RdRp) domain of the NS5 protein plays a crucial part in the replication of the viral genome. RNA is synthesized via “de novo” by NS5 protein.^[18]

Rhinoviruses can effectively infect upper and lower respiratory tract and cause common cold in humans. Though common cold is relatively mild in nature, it is a global socioeconomic burden.^[19, 20] Rhinoviruses are also associated with severe respiratory tract illnesses such as pneumonia.^[21] Cystic fibrosis^[22], bronchitis^[23], chronic obstructive pulmonary disease^[24], asthma^[25] and whizzing illnesses in infants.^[26] Rhinoviruses are small, non-enveloped, single-stranded RNA viruses with a positive sense genome of 7200 bases. The genome contains a single open reading frame (ORF) which is flanked by 59 and 39 untranslated regions (UTR). The ORF encodes a single polyprotein that is post-translationally cleaved into four structural proteins (VP1, VP2, VP3 and VP4) and seven non-structural proteins (2A, 2B, 2C, 3A, 3B, 3C and 3D). Sequences of various sub-genomic regions such as 59UTR, VP4/VP2, VP1, 3D polymerase and partial 2A have been used to study evolutionary relatedness of HRV-A, -B and -C strains/isolates.^[27]

Bioinformatics is an interdisciplinary branch of science which utilizes statistics, computer and mathematics to analyse biological data.^[26] Bioinformatics is now used for many researches to identify many aspects such as evolution. Protein Data Bank (PDB) is a protein storage bioinformatics tool. It contains the structures of large numbers of proteins, ligands and other macromolecules.^[28] Docking analysis can be conducted for a particular protein and the ligand to analyse the fitness and the interaction with each other in the form of energy. This interaction could be used as the pharmaceutical approach for drug production.^[29]

The aim of our study is to compare the best docking fit for the selected *Coriandrum sativum* L leaves constituents with the Dengue and Rhino structural proteins.

2. MATERIALS AND METHODOLOGIES

2.1. Preparation of viral proteins

The protein data bank (PDB) was used to obtain the three-dimensional image of the macromolecule. PDB contains large number of proteins which are experimentally determined and stored in this site. The structures are downloaded and saved either in mmCIF or in PDB format. Proteins of dengue and rhino were used for this study. Only those proteins are used for the study whose PDB ID is available. The 3D structure of all the five proteins were downloaded from PDB and saved in PDB format. The downloaded proteins were viewed in Rasmol viewer.^[30]

2.2. Preparation of ligands

Ligands selected were from the previous studies on GCMS analysis on *Coriandrum sativum* L herb extract.^[8] 4 ligands were used for the study. Ligands were constructed using Chem Sketch.^[31] The constructed ligands were optimized to add the hydrogen bonds and the obtained structures were saved in mol for docking analysis and named as A, B and C and D respectively.

2.3. Docking study

Docking studies were conducted using iGEMDOCK software. iGEMDOCK (Generic Evolutionary Method for molecular Docking) is a graphical-automatic drug design system for docking, screening and post-analysis.^[32] The proteins and the ligands were loaded and the output path was set as desired. The Standard docking parameters used for docking the ligands with the Dengue and rhino protein used was - (population size=200, generations =70 and Number of solutions =2). The docking process was initiated. After the docking process, the best docking pose for the individual ligands can be obtained two dengue and three rhino viral proteins. The output path of the best binding pose, the binding affinity and the total binding energy values were saved in the output folder. The saved files were visualized in Rasmol viewer.^[33]

3. RESULTS

3.1. Total Binding Energy (kcal/mol) profile for Dengue and rhino virus proteins with 4 ligands.

Table – 1: The Total Binding Energy (kcal/mol) profile for Dengue and Rhino virus structural proteins with 4 ligands.

Ligand	Compound name	Dengue virus		Rhino virus		
		Capsid protein	Envelope protein	VP1	VP2	VP4
A	Dodecanal	-69.6	-60.9	-62.3	-67.1	-64.1
B	E-2-Dodecanol	-69.2	-73.1	-76	-67.1	-72.3
C	Decanal	-66.8	-62.9	-69.9	-61.1	-63.5
D	E-2-Decenol	-62.8	-59.3	-81.9	-62.4	-69.0

3.2. H – Bond profile for Dengue and Rhino virus protein with 4 ligands.

Table – 2: H – bond profile for Dengue and Rhino virus structural proteins with 4 ligands.

Ligand	Compound name	Dengue virus		Rhino virus		
		Capsid protein	Envelope protein	VP1	VP2	VP4
A	Dodecanal	-	H-M	H-S	H-M	-
B	E-2-Dodecanol	H-M	H-S	-	H-M	-
			H-M		H-S	
C	Decanal	H-S	H-S	-	H-S	H-S
D	E-2-Decenol	-	H-S	H-M	H-M	H-M
			H-M	H-S		

3.3. Amino acid position profile for Dengue and Rhino virus protein with 4 ligands

Table – 3: Amino acid position profile for Dengue and Rhino virus structural proteins with 4 ligands.

Ligand	Compound name	Dengue virus		Rhino virus		
		Capsid protein	Envelope protein	VP1	VP2	VP4
A	Dodecanal	-	Gly(628), Arg(629)	His(41)	Ala(88)	-
B	E-2-Dodecanol	Lys(73)	Ile(618), Val(626)	-	Gln(38), Gln(42)	-
C	Decanal	Arg(41)	Arg(619)	-	Lys(30)	His(215)
D	E-2-Decenol	-	Glu(638), Ser(642), Thr(644)	Ile(265) Lys(264)	Val(219)	Tyr(28)

4. DISCUSSION

Considering all the tables from Table – 1, to Table - 3, the 3D structure coordinates of two proteins of dengue and four proteins of Rhino virus are optimized and 4 compounds from *Coriandrum sativum* L herb extract were identified. The total binding energy of the compounds with all the five proteins was calculated using iGEMDOCK. Evaluations of binding conformation of these 4 compounds with two dengue as well as four Rhino virus proteins are performed using iGEMDOCK. From docking study, we listed binding affinities of 4 compounds based on ligand binding energy (Table- 1). The binding pose for each ligand molecule into the dengue and rhino virus proteins are analyzed and the one having lowest ligand binding energy with these proteins among the different poses are generated. The lower total binding energy scores represent better protein-ligand target binding affinities as compared to higher total binding energy scores. Considering the structural proteins of Dengue virus, among the 4 analogs, compound “B” is found to have lower ligand binding energy (binding energy value= -73.1kcal/mol), than other analogs for Envelope protein. Compound “B” has least binding energy score with Capsid protein (binding energy value= -69.2 kcal/mol). The structural proteins of

Rhino virus had following binding energies, VP1 (‘D’ binding energy value= -81.9 kcal/mol), VP2 (‘A &B’, binding energy value= -67.1 kcal/mol) and VP4 (‘B’, binding energy value= -72.32 kcal/mol). We further analyzed the docked pose for finding the binding mode of compound “B” in to two dengue and three rhino virus proteins to validate the reasonable binding conformations.

4.1. Structural proteins of Dengue virus

4.1.1. The Total Binding Energy for Dengue virus Capsid protein with 4 ligands

From Table – 1, Table – 2 and Table – 3, the docking simulation of 4 ligands were performed for Dengue virus Capsid protein. From the docking study, we observed that compound B has best binding affinity with the target Capsid protein with the binding energy value of -69.2 kcal/mol. Interaction analysis of binding mode of compound B in dengue virus. Capsid protein reveals that it forms one hydrogen bond with low energy, with Lys (73) residue. A close-up view of the Total Binding Energy (kcal/mol) profile for Dengue virus Capsid protein with 4 ligands: is shown in Fig.1.

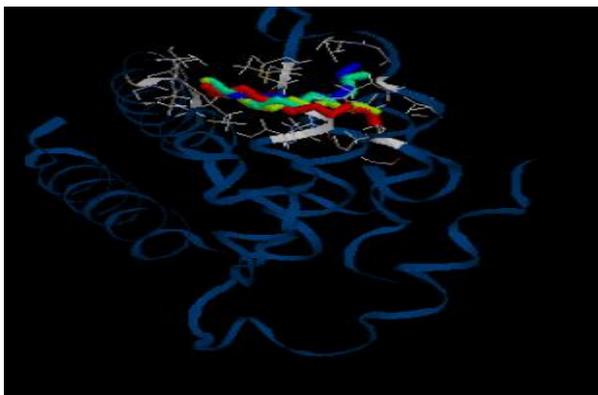


Fig.1: The Total Binding Energy for Dengue virus Capsid protein with 4 ligands.

4.1.2. The Total Binding Energy for Dengue virus envelope protein with 4 ligands

From Table – 1, Table – 2 and Table – 3, the docking simulation of 4 ligands were performed for Dengue virus envelope protein. From the docking study, we observed that compound B has best binding affinity with the target envelope protein with the binding energy value of -73.01 kcal/mol. Interaction analysis of binding mode of compound B in dengue virus. Envelope protein reveals that it forms two hydrogen bonds with low energy, with Val(626) and Ile(618) residues. A close-up view of the Total Binding Energy (kcal/mol) profile for Dengue virus envelope protein with 4 ligands: is shown in Fig.2.

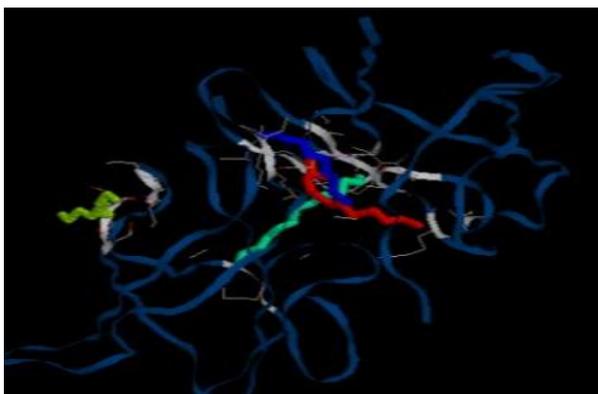


Fig.2: The Total Binding Energy for Dengue virus envelope protein with 4 ligands.

4.2. Structural proteins of Rhino virus

4.2.1. The Total Binding Energy for Rhino virus VP1 protein with 4 ligands

From Table – 1, Table – 2 and Table – 3, the docking simulation of 4 ligands were performed for Rhino virus VP1 protein. From the docking study, we observed that compound D has best binding affinity with the target VP1 protein with the binding energy value of -81.9kcal/mol. Interaction analysis of binding mode of compound B in dengue virus. VP1 protein reveals that it forms two hydrogen bonds with low energy, with His (625) and Lys (264) residues. A close-up view of the Total Binding Energy (kcal/mol) profile for Rhino virus VP1 protein with 4 ligands: is shown in Fig.3.

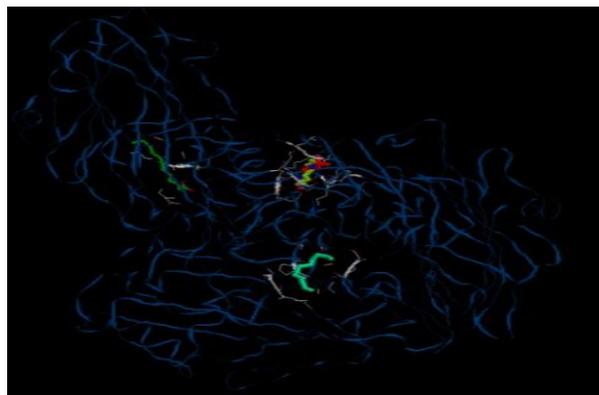


Fig.3: The Total Binding Energy for Rhino virus VP1 protein with 4 ligands.

4.2.2. The Total Binding Energy for Rhino virus VP2 protein with 4 ligands

From Table – 1, Table – 2 and Table – 3, the docking simulation of 4 ligands were performed for Rhino virus VP2 protein. From the docking study, we observed that compound A & B has best binding affinity with the target VP2 protein with the binding energy value of -67.1kcal/mol. Interaction analysis of binding mode of compound B in dengue virus. VP2 protein reveals that it forms two hydrogen bonds with low energy, with Ala (88), Gln (38) and Gln (42) residues. A close-up view of the Total Binding Energy (kcal/mol) profile for Rhino virus VP2 protein with 4 ligands: is shown in Fig.4.

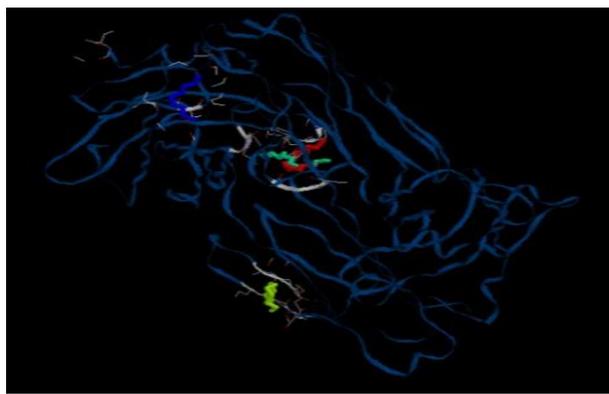


Fig.4: The Total Binding Energy for Rhino virus VP2 protein with 4 ligands.

4.2.3 The Total Binding Energy for Rhino virus VP4 protein with 4 ligands

From Table – 1, Table – 2 and Table – 3, the docking simulation of 4 ligands were performed for Rhino virus VP4 protein. From the docking study, we observed that compound B has best binding affinity with the target VP4 protein with the binding energy value of -72.32kcal/mol. Interaction analysis of binding mode of compound B in dengue virus. VP4 protein reveals that it forms one hydrogen bond with low energy, with His (215) residues. A close-up view of the Total Binding Energy (kcal/mol) profile for Rhino virus VP4 protein with 4 ligands: is shown in Fig.6.

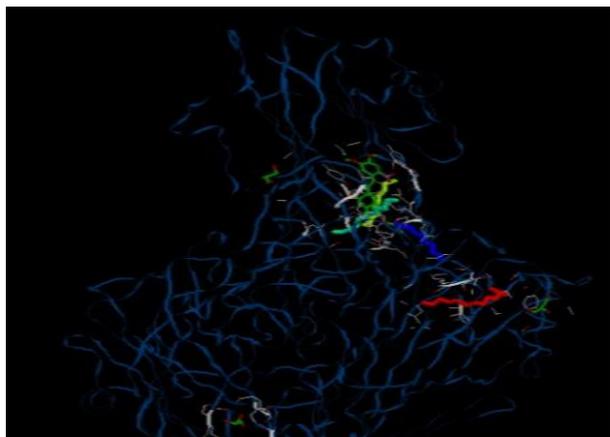


Fig.6: The Total Binding Energy for Rhino virus VP4 protein with 4 ligands.

5. CONCLUSION

Our molecular docking studies explored the possible binding modes of 4 compounds that are present in *Coriandrum sativum* L herb with two proteins of Dengue virus and three proteins of rhino virus. Dengue virus consists of envelope protein and Capsid protein. Rhino virus consists of VP1, VP2 and VP3 coat structural proteins. It revealed that all the 4 compounds show minimum affinity with all the proteins. The compound B (E-2 Dodecanal) shows best results compared to other compounds. On comparing the binding energy and the binding site residues, we found that all the compounds will differ in either of them for hydrogen bond formation. The conclusion which is drawn from our virtual screening and docking result are that the Compound B has highest binding affinity with most of the structural proteins of Dengue virus as well as with the most of the structural proteins of rhino virus. Hence, the Compound B may be considered as the effective drug target for both dengue and rhino virus because it can effectively bind to most of the proteins of both the viruses. Though, there are many reports on the *in vitro* analysis of these compounds and its medicinal and toxic properties, there are no *in silico* studies that predict the binding and active regions especially with these proteins. Our study is an attempt to predict the binding site and the binding residues. However, validation of our results through *in vivo* and *in vitro* experiments and also with animal models will throw light for the future development of more potent drugs for the treating of Dengue and common cold.

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