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Design and *in Silico* Studies on Different Dengue Viral Proteins of Some Compounds Based on Acetophenone Skeleton

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ABSTRACT

Aim: In this study, we performed the evaluation of anti-Dengue potential of some acetophenone derivatives and diazine with dihydroxyacetophenone skeleton derivatives, using molecular docking approach, as potential candidates for anti-dengue virus drug discovery.

Material and Methods: For this study 20 synthetic ligands were used. Docking studies were conducting using iGEMDOCK (Generic Evolutionary Method for molecular Docking) software.

Results: The obtained results revealed that all the 20 compounds show good affinity with all the proteins; significant affinity was observed between the tested compounds and Dengue transmembrane domain of NS2A protein.

Conclusion: The conclusion drawn from our virtual screening and docking result reveals that the investigated compounds have a good binding affinity with most of the proteins and some of they can be used as an effective drugs target for Dengue virus.

Keywords: Binding Interaction; Molecular Docking; Dengue Virus; Acetophenone Skeleton; Drug Design

Abbreviations: DVP: Dengue Viral Proteins; AD: Acetophenone Derivatives, iGEMDOCK: Generic Evolutionary Method for Molecular Docking; NS2A Protein: Nonstructural Protein 2A; HIV: Human Immunodeficiency Virus; DHF: Dengue Haemorrhagic Fever; DSS: Dengue Shock Syndrome; E: Envelope; C: Capsid; PreM: Premembrane; NS1 Protein: Nonstructural Protein 1; NS2B Protein: Nonstructural Protein 2B; NS3 Protein: Nonstructural Protein 3; NS4B Protein: Nonstructural Protein 4B; NS5 Protein: Nonstructural Protein 5; NSA Protein: Nonstructural Protein; RNA: Ribonucleic Acid; Mtase: Methyltransferase; PDB: Protein Data Bank; ATPase: Adenosine Triphosphatase; RTPase: Ribonucleic Acid 5' Triphosphatase

Introduction

Highly functionalized acetophenone derivatives are an important class of organic compounds which exhibiting interesting biological properties, such as antimicrobial [1] and anticancer activities [2] being a valuable compounds for supramolecular or medicinal chemistry [3] In last years, 1,2-diazine derivatives have demonstrated to possess a large variety of biological activities: antihypertensive [4], diuretic [5], antiplatelet [6], cardiotonic [7] anticancer [8], anti-HIV [9], anti-inflammatory [10], antimicrobial [11] anti-tuberculosis [12] etc. Dengue virus is part of the family Flaviviridae, genus Flavivirus [13,14]. The Dengue virus causes a range of clinical manifestations, the most severe forms being Dengue hemorrhagic fever (DHF) and dengue shock syndrome (DSS) [15,16]. Currently, there has been no vaccine or effective antiviral drug available against dengue infection. Therefore, many researchers are trying to find an efficient antiviral agent against this virus [17]. There are ten proteins in Dengue virus, three are structural proteins, including envelope (E), capsid (C) and PreM, and seven are nonstructural proteins. The process of viral replication and pathogenesis of Dengue virus is associated with the seven non-structural proteins, namely NS1, NS2A, NS2B, NS3, NS4A, NS4B, and NS5 [18,19]. NS proteins reshape the inner organization of the cell, mature the polyprotein, replicate the viral RNA and help the virus evade the immune system. Among the seven non-structural proteins, NS5 is the largest protein (102 kDa) that encoded by Flavivirus genome.18 NS5 encodes methyltransferase (Mtase) on N-terminal and RNA-dependent RNA polymerase (RdRp) on C-terminal and it can also down-regulate the host immune interferon response [20]. The effective and efficient methods in designing and developing a novel potential drug candidates can be completed through in silico approach because the identification of potential drugs can be obtained more quickly [21]. In recent times, computational drug chemistry is proving to be an efficient tool for identification and prediction of compounds that can have anti dengue activity [22].

Docking analysis is an important aspect for getting the information on the interaction profile of the protein with the ligands. The interaction of the protein with the ligands is given by the binding energy [23]. The compounds containing acetophenone skeleton and diazine with dihydroxyacetophenone scaffold derivatives have wide interest due to their diverse pharmacological properties. In particular, these biological activities make these derivatives more attractive and testing as novel therapeutic compounds. As part of our aim in research of biologically active acetophenone derivatives, the free hydroxyl groups attached on the acetophenone ring has allowed us to introduce some radicals that can improve the biological activity. Looking to the medicinal importance of the acetophenone skeleton, we performed the evaluation of anti-Dengue potential of some acetophenone derivatives and diazine with dihydroxyacetophenone skeleton derivatives, previously synthesized, using molecular docking approach.

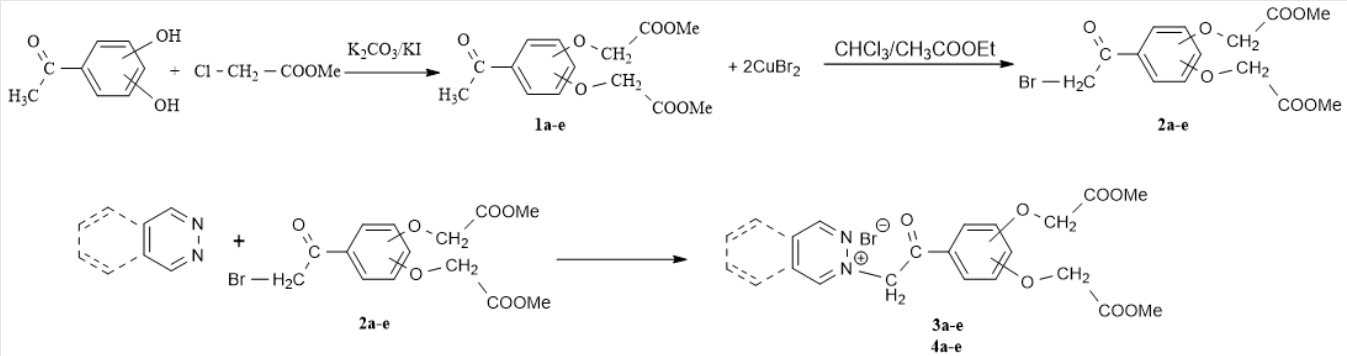
Materials and Methods

Chemistry

The synthesis of the target compounds which were used as ligands were previously reported [24,25]. A series of dihydroxyacetophenone were treated with methylchloroacetate at room temperature. When the reaction occurred in a molar ratio of 1:2, dialkylated derivatives were obtained (1a-e). These derivatives lead to the bromo-bisetherificated acetophenone derivatives (2a-e) in the presence of copper bromide in a mixture of chloroform/ethylacetate. In order to obtain cycloimmonium salts, different nitrogen heterocyclic compounds have been treated with halogenated acetophenone skeleton derivatives with increased reactivity (3a-e and 4a-e).

Spectral Characterization

In Table 1 we present the chemical structure and the name for the synthesized compounds Table 2. shows some physicochemical and spectral characteristics of the derivatives. Synthetic pathway of compounds with acetophenone skeleton is presented in Scheme 1.



Scheme 1: The reaction pathway in synthesis of the target compounds.

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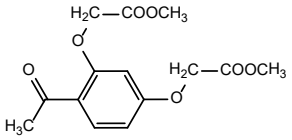
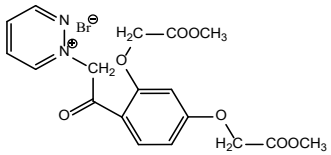
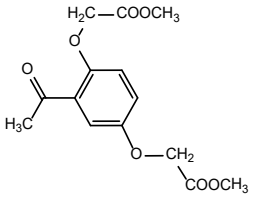
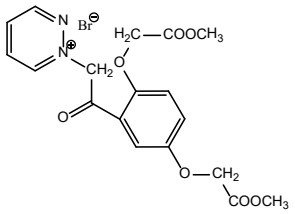
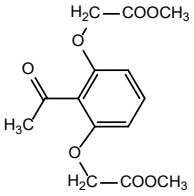
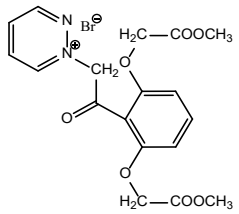
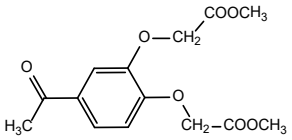
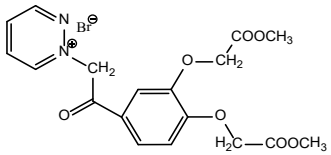
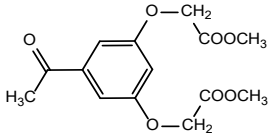
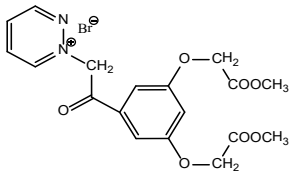
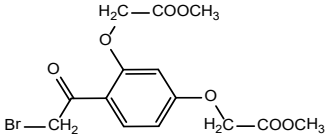
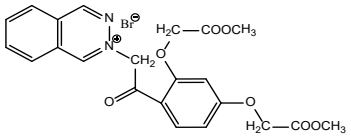
Table 1: Chemical structure of compounds with acetophenone skeleton (1-4).

Comp.	Structure	Comp.	Structure
1a	 dimethyl 2,2'-((4-acetyl-1,3-phenylene) bis(oxy)) diacetate	3a	 1-(2-(2,4-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) pyridazin-1-ium bromide
1b	 dimethyl 2,2'-((2-acetyl-1,4-phenylene) bis(oxy)) diacetate	3b	 1-(2-(2,5-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) pyridazin-1-ium bromide
1c	 dimethyl 2,2'-((2-acetyl-1,3-phenylene) bis(oxy)) diacetate	3c	 1-(2-(2,6-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) pyridazin-1-ium bromide
1d	 dimethyl 2,2'-((4-acetyl-1,2-phenylene) bis(oxy)) diacetate	3d	 1-(2-(3,4-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) pyridazin-1-ium bromide
1e	 dimethyl 2,2'-((5-acetyl-1,3-phenylene) bis(oxy)) diacetate	3e	 1-(2-(3,5-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) pyridazin-1-ium bromide
2a	 dimethyl 2,2'-((4-(2-bromoacetyl)-1,3-phenylene) bis(oxy)) diacetate	4a	 2-(2-(2,4-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) phthalazin-2-ium bromide

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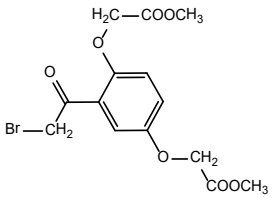
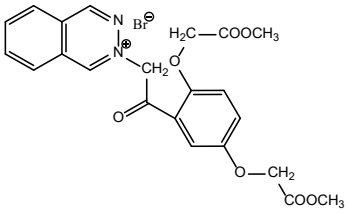
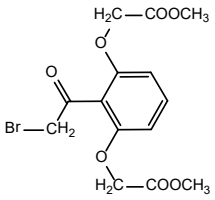
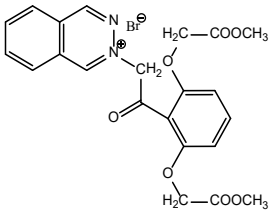
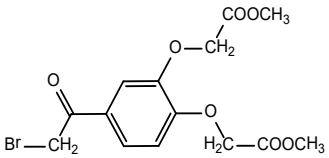
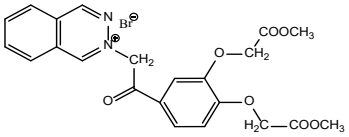
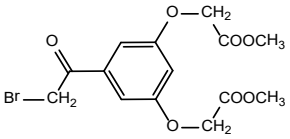
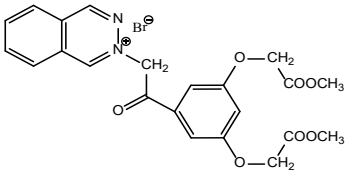
2b	 dimethyl 2,2'-((2-(2-bromoacetyl)-1,4-phenylene) bis(oxy)) diacetate	4b	 2-(2-(2,5-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) phthalazin-2-ium bromide
2c	 dimethyl 2,2'-((2-(2-bromoacetyl)-1,3-phenylene) bis(oxy)) diacetate	4c	 2-(2-(2,6-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) phthalazin-2-ium bromide
2d	 dimethyl 2,2'-((4-(2-bromoacetyl)-1,2-phenylene) bis(oxy)) diacetate	4d	 2-(2-(3,4-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) phthalazin-2-ium bromide
2e	 dimethyl 2,2'-((5-(2-bromoacetyl)-1,3-phenylene) bis(oxy)) diacetate	4e	 2-(2-(3,5-bis(2-methoxy-2-oxoethoxy)-phenyl)-2-oxoethyl) phthalazin-2-ium bromide

Table 2: Physicochemical and spectral characteristics for synthesized compounds.

Comp.	Melting point (°C)	IR (cm^{-1})
1a	114–115	3102, 3077, 3050 (C–H arom), 2963, 2945, 2920 (C–H aliph), 1765 (C=O ester), 1668 (C=O keto), 1600, 1492, 1451, 1427(C–C arom), 1265, 1250, 1225, 1198 (C–O–C)
1b	95–96	3070, 3048, 2997 (C–H arom),2953, 2928 (C–H aliph), 1765, 1742 (C=O ester), 1665 (C=O keto), 1590, 1545, 1499, 1429 (C–C arom), 1303, 1282, 1224, 1203, 1169 (C–O–C)
1c	80-81	3100, 3048, 3011 (C–H arom), 2957, 2917 (C–H aliph), 1767, 1751 (C=O ester), 1697 (C=O keto), 1599, 1470, 1441 (C–C arom), 1248, 1211, 1130, 1086 (C–O–C)
1d	89–90	3077, 3036, 3011 (C–H arom),2982, 2959, 2920 (C–H aliph), 1777, 1763 (C=O ester), 1674 (C=O keto), 1591, 1521, 1437, 1412 (C–C arom), 1298, 1276, 1202, 1144 (C–O–C)
1e	102–103	3094, 3045, 3007 (C–H arom),2974, 2959, 2943 (C–H aliph), 1755 (C=O ester), 1684 (C=O keto), 1595, 1460, 1439, 1381 (C–C arom), 1303, 1276, 1219, 1167, 1088 (C–O–C)
2a	71–72	3093, 3072, 3018 (C–H arom), 2965, 2918 (C–H aliph), 1763, 1738 (C=O ester), 1674 (C=O keto), 1606, 1502, 1452, 1433, 1417 (C–C arom), 1296, 1215,1165, 1080 (C–O–C), 608 (C–Br)
2b	84–85	3094, 3071, 3017 (C–H arom), 2963, 2920 (C–H aliph), 1763, 1737 (C=O ester), 1674 (C=O keto), 1601, 1503, 1451, 1435, 1417 (C–C arom), 1296, 1215, 1165, 1080 (C–O–C), 608 (C–Br)
2c	57-58	3093, 3052 (C–H arom), 2994, 2947, 2911 (C–H aliph), 1749 (C=O ester), 1676 (C=O keto), 1595, 1470, 1435, 1377 (C–C arom), 1220, 1182, 1145, 1124 (C–O–C), 613 (C–Br)

2d	89–90	3091, 3022 (C–H arom), 2956, 2915 (C–H aliph), 1745 (C=O ester), 1637 (C=O keto), 1615,1514, 1433, 1400 (C–C arom), 1246, 1155, 1116, 1063 (C–O–C), 617 (C–Br)
2e	75–76	3090, 3013 (C–H arom), 2964, 2951 (C–Haliph), 1751 (C=O ester), 1705 (C=O keto), 1595, 1460, 1439, 1379 (C–C arom), 1219, 1166, 1093 (C–O–C), 654 (C–Br)
3a	83–84	3093, 3028 (C–H arom), 2972, 2955 (C–H aliph), 1741, 1739 (C=O ester), 1690 (C=O keto), 1598, 1502, 1492, 1438, 1423 (C–C arom, C–N arom), 1213, 1176, 1084, 1061 (C–O–C)
3b	127–128	3098, 3028 (C–H arom), 2970, 2952, 2920 (C–H aliph), 1748, 1739 (C=O ester), 1690 (C=O keto), 1599, 1513, 1495, 1437, 1423 (C–C arom, C–N arom), 1213, 1177, 1084, 1060 (C–O–C)
3c	127-128	3095, 3032 (C–H arom), 2969, 2949 (C–H aliph), 1753 (C=O ester), 1690 (C=O keto), 1597, 1470, 1437, 1423 (C–C arom, C–N arom), 1253, 1219, 1130 (C–O–C)
3d	188–189	3071, 3044 (C–H arom), 2978, 2931 (C–H aliph), 1767, 1748 (C=O ester), 1682 (C=O keto), 1597, 1586, 1514, 1433 (C–C arom, C–N arom), 1279, 1219, 1180, 1159 (C–O–C)
3e	100–101	3096, 3065, 3018 (C–H arom), 2978, 2955, 2931 (C–H aliph), 1749, 1732 (C=O ester), 1692 (C=O keto), 1601, 1438, 1387 (C–C arom, C–N arom), 1294, 1230, 1169 (C–O–C)
4a	148–149	3091, 3061 (C–H arom), 2990, 2957 (C–H aliph), 1751 (C=O ester), 1680 (C=O keto), 1614, 1591, 1495, 1452, 1423 (C–C arom, C–N arom), 1288, 1209, 1173, 1076 (C–O–C)
4b	130–131	3087, 3055, 3011 (C–H arom), 2976, 2955 (C–H aliph), 1757, 1737 (C=O ester), 1686 (C=O keto), 1605, 1573, 1495, 1435, 1418 (C–C arom, C–N arom), 1283, 1215, 1176, 1082 (C–O–C)
4c	130-131	3080, 3056 (C–H arom), 2955, 2936 (C–H aliph), 1751 (C=O ester), 1687 (C=O keto), 1597, 1470, 1437, 1395 (C–C arom, C–N arom), 1281, 1219, 1130, 1080 (C–O–C)
4d	151–152	3090, 3039 (C–H arom), 2990, 2938 (C–H aliph), 1768, 1748 (C=O ester), 1676 (C=O keto), 1597, 1582, 1528, 1441 (C–C arom, C–N arom), 1298, 1256, 1193, 1139 (C–O–C)
4e	165–166	3096, 3065, 3019 (C–H arom), 2978, 2932 (C–H aliph), 1749, 1732 (C=O ester), 1692 (C=O keto), 1601, 1473, 1439, 1387 (C–C arom, C–N arom), 1294, 1263, 1231, 1169, 1086 (C–O–C)

Molecular Docking

Preparation of Dengue Viral Proteins: The protein data bank (PDB) was used to obtain the three-dimensional structure 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 mm CIF or PDB format. Proteins of dengue virus were used for this study. The 3D structure of all the seven proteins were downloaded from PDB and saved in PDB format. The downloaded proteins were viewed in Py-Mol viewer.

Preparation of Ligands: The compounds selected from both natural and synthetic sources can be considered as ligands. 20 synthetic ligands were used for this study (Table 1). Ligands were constructed using ChemSketch. 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 respectively.

Docking Study: Docking studies were conducting using iGEMDOCK software. IGEMDOCK (Generic Evolutionary Method for molecular Docking) is a graphical-automatic drug design system for docking, screening and post-analysis. The proteins and the ligands were loaded, and the out path was set. Standard docking parameters were used for docking (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 for all the seven dengue viral proteins. 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 Py-Mol viewer.

Results and Discussion

Chemistry

The designed dihydroxyacetophenone derivatives (1a-e, 2a-e) and diazine with dihydroxyacetophenone skeleton derivatives (3a-e, 4a-e) were synthesized according to scheme 1. The procedures and conditions of the reactions were previously presented [24,25].

In Silico Molecular Docking

Using molecular docking approach, we performed the evaluation of anti-Dengue potential of five bis-etherificated acetophenone derivatives, five α-bromo-bisetherificated acetophenone derivatives and ten cycloimmonium salts with acetophenone skeleton, previously synthesized. So, 20 compounds, which were used as ligands, were selected for this study (Table 1). The fitness and the interaction profile (the total binding energy, Van der Waal's Force and H-bond energy, in kcal/mol) of the dengue virus proteins with the ligands are presented in Tables 3-5. The lower energy scores represent better target protein-ligand binding affinity compared to higher energy score. The cluster interaction tables which include H-Bond profile and amino acid position profile for the dengue virus proteins with the ligands are presented below (Tables 6 & 7). A close-up view of binding mode of ligands with the

seven selected proteins is shown in Figures 1-7. From Tables 3-5, the 3D structure coordinates of protein of dengue virus are optimized and 20 compounds are identified. Their total binding energy was calculated using iGEMDOCK. Evaluations of binding conformation of 20 compounds with dengue viral proteins are performed using iGEMDOCK. From docking study, we listed binding affinity of 20 compounds based on ligand binding energy (Tables 3-5). The binding pose for each ligand molecule into the dengue viral protein is analyzed and the one having lowest ligand binding energy with these proteins among the different poses are generated.

Table 3: The total binding energy (kcal/mol) profile for Dengue virus proteins with the investigated ligands.

Ligand	Capsid protein	Envelope protein	NS1 protein	NS2A protein	NS3 protease protein	NS3 helicase protein	NS5 protein
1a	-89.7	-89.06	-106.15	-635.01	-89.87	-94.95	-98.66
1b	-87.9	-85.47	-88.3	-580.07	-101.2	-89.7	-96.04
1c	-94.48	-89.71	-104.68	-564.22	-95.4	-106.8	-97.45
1d	-86.76	-87.5	-100.04	-629.62	-107.78	-94.87	-103.4
1e	-94.67	-88.84	-100.94	-589.19	-97.33	-96.63	-90.41
2a	-88.09	-86	-104.58	-613.36	-98.19	-96.92	-94.53
2b	-103.8	-95.49	-110.91	-591.18	-95.85	-94.08	-97.45
2c	-84.3	-82.71	-94.61	-566.75	-98.64	-101.93	-97.61
2d	-86.46	-86.94	-106.3	-588.14	-95.65	-93.21	-111.26
2e	-86.66	-92.06	-100.77	-637.089	-100.83	-101.88	-99.91
3a	-95.37	-104.65	-102.33	-655.9	-119.72	-106.6	-98.68
3b	-88.9	-104.62	-104.19	-648.73	-97.6	-113.61	-113.87
3c	-94.27	-106.92	-106.62	-672.03	-110.38	-109.35	-99.27
3d	-107.53	-104.59	-106.82	-714.91	-100.03	-110.31	-107.81
3e	-98.29	-102.47	-118.08	-692.88	-103.11	-125.27	-108.72
4a	-110.76	-99.69	-112.89	-719.34	-124.58	-102.09	-118.23
4b	-104.18	-105.47	-125.63	-732.67	-103.07	-100.71	-112.29
4c	-107.54	-98.52	-104.44	-673.19	-118.92	-115.73	-133.29
4d	-102.82	-96.26	-130.32	-701.34	-111.48	-100.82	-103.09
4e	-100.03	-113.81	-105.08	-677.67	-98.56	-114.16	-102.55

Table 4: Van der Waal's Force (kcal/mol).

Ligand	Capsid protein	Envelope protein	NS1 protein	NS2A protein	NS3 protease protein	NS3 helicase protein	NS5 protein
1a	-87.58	-68.2	-89.65	-582.54	-69.11	-79.71	-88.33
1b	-70.42	-71.77	-75.32	-575.02	-87.24	-74.77	-85.54
1c	-81.47	-72.27	-73.67	-535.97	-81.4	-89.5	-89.18
1d	-78.45	-62.39	-77.73	-258.57	-81.92	-83.18	-82.99
1e	-80.94	-76.35	-82.5	-557.23	-76.69	-80.15	-72.85
2a	-78.32	-77.24	-81..35	-491.54	-72.68	-76.67	-78.05
2b	-88.16	-73.27	-80.78	-466.02	-81.26	-77.76	-71.15
2c	-62.02	-66.38	-87.36	-562.92	-88.14	-80.94	-95.11
2d	-71.09	-70.43	-99.3	-588.14	-82.3	-74.72	-101.76
2e	-69.62	-68.53	-79.58	-580.68	-81.34	-82.23	-84.05
3a	-81.39	-84.05	-85.71	-655.9	-89.62	-89.94	-78.78
3b	-73.65	-91.81	-93.07	-648.73	-85.06	-96.4	-98.62
3c	-73.46	-99.92	-87.54	-617.15	-94.55	-87.85	-85.89
3d	-98.25	-86.32	-84.57	-714.91	-87.56	-88.29	-93.47

3e	-92.02	-83.38	-97.23	-686.79	-92.62	-103.94	-89.06
4a	-94.99	-82.04	-102.15	-668.27	-100.67	-80.4	-102.41
4b	-95.96	-87.36	-106.51	-655.21	-98.59	-94.03	-97.92
4c	-96.82	-79.71	-85.18	-568.33	-103.69	-89.5	-105.1
4d	-83.71	-89.26	-105.77	-697.84	-105.14	-87.25	-98.61
4e	-80.3	-99.27	-85.85	-624.79	-85.06	-97.99	-89.29

Table 5: H-bond energy (kcal/mol) profile for Dengue virus proteins with the investigated ligands.

Ligand	Capsid protein	Envelope protein	NS1 protein	NS2A protein	NS3 protease protein	NS3 helicase protein	NS5 protein
1a	-2.08	-20.85	-16.5	-52.47	-20.75	-15.25	-10.33
1b	-17.48	-13.71	-12.99	-5.05	-14	-14.93	-10.5
1c	-13.01	-17.44	-31.01	-28.25	-14	-17.3	-8.27
1d	-8.31	-25.11	-22.31	-47.05	-25.86	-11.69	-20.41
1e	-13.73	-12.5	-18.44	-23.96	-20.64	-15.4	-17.55
2a	-9.77	-8.76	-23.23	-121.82	-25.51	-17.25	-16.48
2b	-15.64	-2.22	-30.13	-125.17	-14.59	-16.31	-26.3
2c	-22.28	-16.33	-7.25	-3.83	-10.5	-20.99	-2.5
2d	-15.36	-16.51	-7	0	-13.35	-18.49	-9.5
2e	-23.04	-23.53	-21.19	-56.41	-19.49	-19.65	-15.85
3a	-13.98	-20.61	-16.62	0	-30.1	-16.66	-19.9
3b	-15.36	-12.81	-11.12	0	-12.9	-17.29	-15.25
3c	-20.81	-7	-19.08	-57.89	-15.83	-21.5	-13.38
3d	-9.28	-18.27	-22.24	0	-12.47	-22.01	-14.34
3e	-6.27	-19.09	-20.84	-6.09	-10.5	-21.33	-19.65
4a	-15.77	-17.65	-10.74	-51.07	-23.92	-21.69	-15.82
4b	-8.22	-18.11	-19.13	-77.46	-10.48	-6.69	-14.37
4c	-10.73	-18.81	-19.26	-104.86	-15.23	-26.23	-27.88
4d	-19.11	-7	-24.56	-3.5	-6.34	-13.57	-4.49
4e	-19.73	-14.54	-19.23	-52.85	-13.5	-22.17	-13.26

Table 6: H-bond profile for Dengue virus proteins with the investigated ligands.

Ligand	Capsid protein	Envelope protein	NS1 protein	NS2A protein	NS3 protease protein	NS3 helicase protein	NS5 protein
1a	-	H-S H-M	H-S H-M	H-M	H-S	H-S H-M	H-S
1b	H-S H-M	H-S H-M	H-M	H-S	H-S H-M	H-S H-M	H-S H-M
1c	H-S H-M	H-M	H-S	H-S	H-S	H-S	H-S
1d	H-S H-M	H-S H-M	H-S	H-S H-M	H-S H-M	H-S	H-S H-M
1e	H-S H-M	H-S	H-S	H-S	H-S H-M	H-S H-M	H-S H-M
2a	H-S	H-M H-S	H-S H-M	H-M	H-S H-M	H-S H-M	H-S H-M

2b	H-S	H-S H-M	H-S H-M	H-M	H-S H-M	H-S H-M	H-S H-M
2c	H-S H-M	H-S	H-M	H-S	H-S	H-S H-M	H-S
2d	H-S H-M	H-M	H-M	-	H-S H-M	H-S	H-S
2e	H-S H-M	H-S H-M	H-S H-M	H-S	H-S H-M	H-M	H-S
3a	H-S H-M	H-M	H-S H-M	-	H-S H-M	H-S H-M	H-M
3b	H-S H-M	H-M	H-S H-M	-	H-S H-M	H-S H-M	H-S H-M
3c	H-S H-M	H-S H-M	H-S	H-S H-M	H-S H-M	H-S H-M	H-S H-M
3d	H-S	H-M	H-S H-M	-	H-S H-M	H-S H-M	H-S H-M
3e	H-S	H-S H-M	H-S	H-S H-M	H-S H-M	H-S H-M	H-S H-M
4a	H-S	H-M	H-S H-M	H-M	H-S H-M	H-S H-M	H-S
4b	H-S	H-S H-M	H-S	H-S	H-S	H-S	H-S H-M
4c	H-S	H-S H-M	H-S	H-M	H-S H-M	H-S H-M	H-S H-M
4d	-	H-M	H-S H-M	H-S	H-M	H-S	H-M
4e	H-S H-M	H-S H-M	H-S H-M	H-S H-M	H-S H-M	H-S	H-S H-M

Note: H-M is the hydrogen bonding between the viral protein and the main chain of the aminoacid;

H-S is the hydrogen bonding between the viral protein and the side chain of the aminoacid

Table 7: Amino-acid position/profile for Dengue virus proteins with the investigated ligands.

Ligand	Capsid protein	Envelope protein	NS1 protein	NS2A protein	NS3 protease protein	NS3 helicase protein	NS5 protein
1a	-	ARG(619)/-11.9 SER(577)/-3.3	LYS(221)/-7 SER(185)/ GLY(199)/-3.5	PHE(15)/-3	ARG(60)/-7	THR(289)/ THR(317)/-2.5 ALA(290)/ THR(289)/-3.5	ASN(69)/ ARG(581)/-3.5

1b	ARG(41)/-3.5 LEU(44)/ LYS(45)/ LEU(46)/ PHE(47)/-3.5	LYS(625)/-7 ILE(618)/-3.5	GLY(259)/-7	ASP(1)/-2.5	ASN(152)/-7 LEU(149)/-3.5	ASP(470)/-2.5 ARG(463)/ ASN(464)/-3.5	ARG(352)/-3.5 THR(270)/-3.5
1c	ARG(41)/-7 LEU(44)/-3.1	GLY(628) /-7	ARG(294)/-12.9	THR(7)/-2.5	ASN(152)/-10.5	ARG(460)/-7	LYS(253)/-8.3
1d	ARG(41)/-3.5 PHE(47)/-4.6	ARG(619)/-11.1 PRO(635)/-2.9	GLN(253)/-9.9	ASP(1)/-2.5 GLY(3)/-2.9	ASN(105)/-9.5 SER(75)/-3.5	ASN(329)/-3.5	LYS(253)/-7 ASP(254)/-3.5
1e	ARG(32)/-4 LEU(29)/-2.7	THR(667)/-5	ARG(294)/-9.4	ARG(18)/-7	ARG(107)/-7 LEU(74)/ GLU(90)/ ASN(105)/-3.5	THR(450)/-2.8 THR(450)/-3.5	ASN(272)/-7 ASP(596)/-2.8
2a	ARG(68)/-6.3	TYR(578)/-3.5 THR(667)/-2.5	TRP(232)/-6.2 TRP(210)/-3.5	GLY(5)/-7	ARG(24)/-7 ASP(58)/-3.5	HIS(485)/-6.9 THR(389)/-3.5	ARG(361)/-6.1 THR(270)/-3.5
2b	ARG(41)/-10.3	THR(634)/-6.7 GLY(628) /-5	GLN(253)/-6 LYS(227)/-6.9	GLY(5)/-7	TRP(83)/-3.5 LEU(149)/-3.5	ARG(387)/-6.7 PHE(610)/-2.6	ASP(131)/-9.2 LYS(105)/-5.5
2c	THR(25)/-3.9 ARG(22)/-4.5	ARG(619)/-10	GLY(259)/-3.5	ARG(18)/-3.8	ASN(152)/-7	ARG(418)/-11.8 ASN(464)/-3.5	TYR(90)/-2.5
2d	ARG(41)/-12.2 PHE(47)/-3.2	GLY(628)/ ILE(630)/ ARG(629)/ THR(634)/-3.5	AS(180)/ SER(185)/-3.5	-	ASN(152)/-3.5 LEU(149)/-3.5	ARG(463)/ ASN(464)/-3.5 GLY(462)/ ARG(463)/-3.5	ARG(581)/ ASN(69)/-3.5
2e	THR(62)/-5 VAL(23)/ SER(24)/ THR(25)/-3.5	LYS(625)/-3.5 ARG(629)/-7	GLN(253)/-6.2 LYS(227)/-3.3	THR(7)/- -4.5	ARG(107)/-10.1 SER(75)/-3.4	ARG(418)/-7 ARG(418)/-3.5	HIS(53)/ -9.9
3a	LYS(67)/-7 ARG(100)/-7	ARG(629)/ ILE(630)/-7	ASN(234)/-7 SER(228)/-3.5	-	ARG(107)/-9.5 ASN(105)/-7	ARG(418)/-8.8 GLY(196)/-5.4	TYR(606)/-10.5
3b	THR(71)/-6.3 ARG(100)/-6.9	GLY(628)/ ILE(630)/-3.5	ARG(336)/-15.3 GLY(292)/-6.2	-	ASN(152)/-5.9 ALA(166)/ ASN(167)/-3.5	ARG(418)/-3.5 GLY(196)/-8.3	THR(104)/-5.7 THR(105)/-5.6
3c	ARG(85)/-10.5 ASN(93)/-3.3	LYS(625)/ -3.5 GLY(628)/-3.5	ASN(234)/-5.3	THR(7)/-2.7 TYR(8)/-3.2	ASN(105)/-6.3 SER(75)/ ASN(105)/-3.5	THR(289)/-12 THR(289)/-3.5	HIS(110)/-3.9 GLU(111)/ VAL(132)/-3.5

3d	ARG(68)/-9.3	GLY(628)/ ILE(630)/ ARG(629)/ THR(634)/-3.5	THR(262)/-4.5 ILE(242)/ LYS(245)/-3.5	-	THR(118)/ THR(120)/-2.5 ALA(166)/-3.5	ARG(387)/-10.3 ASP(603)/-2.9	HIS(52)/-3.5 THR(50)/ THR(51)/ -3.5
3e	ARG(55)/-6.3	LYS(625)/-6.2 ILE(618)/-2.9	ARG(294)/-8.3	ASP(1)/-3.5	LYS(74)/ ASN(152)/-3.5 LEU(149)/-3.5	ASP(284)/-4.6 LEU(193)/-3.5	THR(51)/-4.7 THR(51)/ GLN(693)/ TRP(694)/-3.5
4a	ARG(41)/-12.3	ILE(630)/-6.2	ARG(336)/-5.6 ASN(293)/-7	GLY(5)/-3.2	ARG(107)/-4.5 ASN(105)/-7	ARG(463)/-4.3 GLY(196)/-6.9	THR(51)/-7.5
4b	ARG(55)/ LYS(45)/-3.5	THR(644)/-4.7 LEU(636)/ GLU(638)/-3.5	ASN(234)/-6.5	THR(7)/-8	ARG(55)/-6.1	ARG(427)/-3.8	HIS(52)/ ASP(690)/-2.5 ASP(690)/-2.9
4c	ARG(41)/-10.7	THR(634)/-5.3 GLY(628)/ ILE(630)/ ARG(629)/-3.5	SER(228)/-4.2	GLY(5)/-7.4	ARG(55)/-3.5 VAL(59)/ GLU(86)/ VAL(146)/-3.5	ARG(387)/-10 VAL(544)/-3.5	THR(51)/-9.5 THR(51)/-3.5
4d	-	GLY(628)/ ILE(618)/-3.5	SER(239)/-2.5 GLU(238)/-5.6	ARG(18)/-3.5	VAL(155)/-2.8	ARG(274)/-10.1	GLY(743)/-2.6
4e	THR(25)/-3.4 VAL(23)/ SER(24)/-3.5	LYS(625)/-3.5 ILE(618)/-3.5	ARG(336)/-6.7 SER(228)/-3.5	THR(7)/-2.5 TYR(8)/-3	ARG(107)/-6.9 SER(75)/-3.5	ARG(387)/-10.3	TRP(418)/-6.3 GLY(743)/-3.5

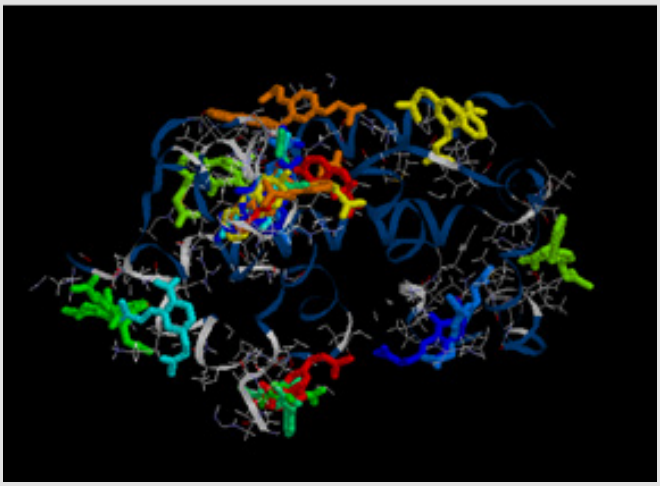


Figure 1: Interaction of all compounds with Capsid protein.

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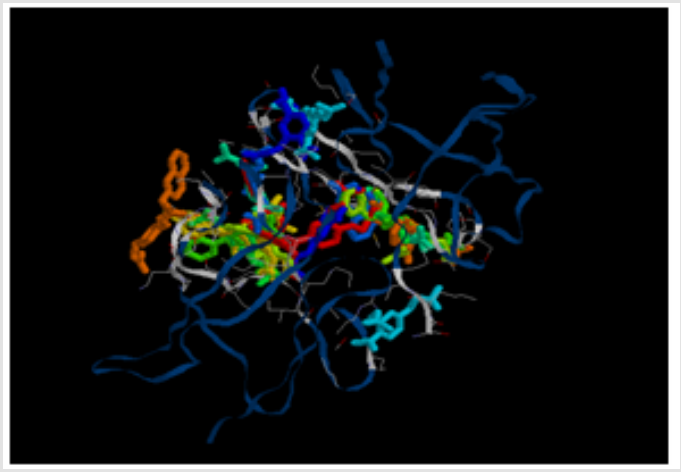

Figure 2: Interaction of all compounds with Envelope protein.

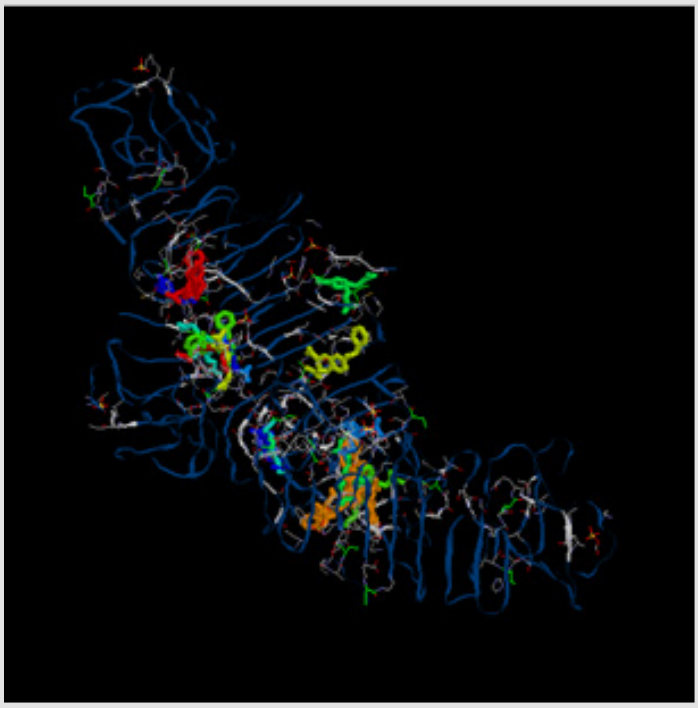

Figure 3: Interaction of all compounds with NS 1 protein.

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Figure 4: Interaction of all compounds with Transmembrane Domain of NS2A protein.

Figure 5: Interaction of all compounds with NS3 Protease protein.

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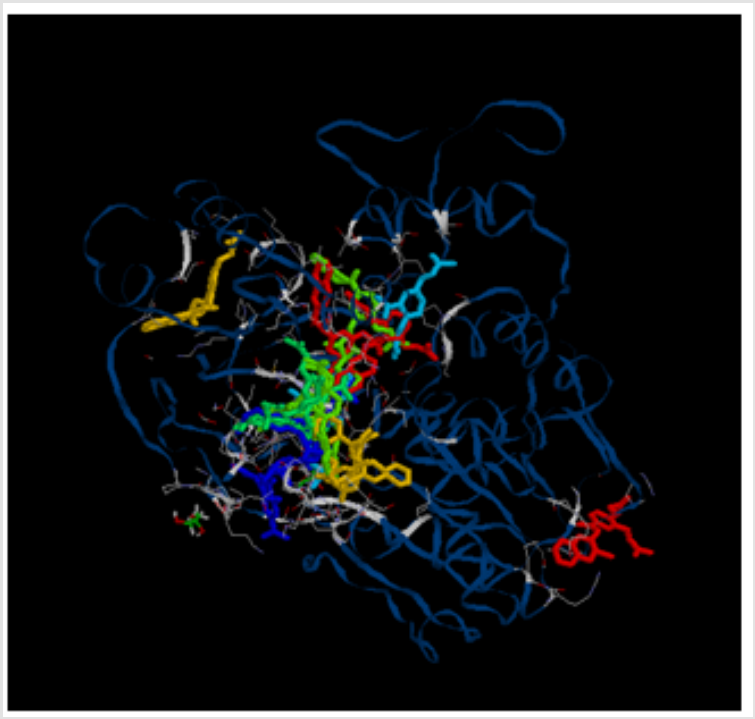

Figure 6: Interaction of all compounds with NS3 Helicase protein.

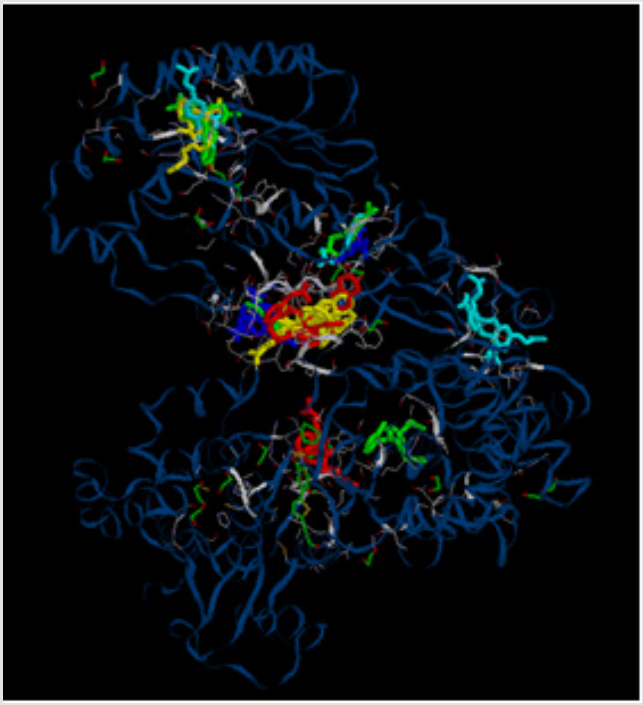

Figure 7: Interaction of all compounds with NS 5 protein.

The lower energy scores represent better protein-ligand target binding affinity compared to higher energy score. The obtained results showed significant affinity between the tested compounds and Dengue transmembrane domain of NS2A protein. NS2A protein, a membrane protein, is one of the proteins found in the replication complex of flaviviruses. This protein possesses specific recognition sites for certain proteases [26]. Compound 4b was found to have the lower ligand binding energy compared to the binding energy at the other analogs for transmembrane domain of NS2A protein (binding energy value = -732.67 kcal/mol), The amino acid positions for each ligand were studied (Table 7). Interaction analysis of binding mode of compound 4b for transmembrane domain of NS2A protein revealed that it forms one hydrogen bonds at positions THR (7) with bond energies -8 kcal/mol, Among the 20 analogs, compound 4a was found to have lower ligand binding energy than other analogs for Capsid protein (binding energy value= -110.76kcal/mol), and for NS3 Protease protein (binding energy value= -124.58kcal/mol). Evaluations of the results showed that, the compound 4a 2-(2-(2,4-bis(2-methoxy-2-oxoethoxy) phenyl)-2-oxyethyl)phthalazin-2-ium bromide forms one hydrogen bond at positions ARG(41), with bond energies, -12.3kcal/mol. We further analyzed the docked pose for finding the binding mode of compound 4a in to Capsid protein and for NS3 Protease protein to validate the reasonable binding conformations. Compound 4c was found to have the lower ligand binding energy for NS5 protein with a value of -133 [27]. kcal/mol and it appear two hydrogen bonds at positions THR(51) with different bond energies (-9.5 kcal/mol and -3.5 kcal/mol respectively). For Envelope protein compound 4ewas found to have lower ligand binding energy (binding energy value=-113.81kcal/mol), than other analogs.

Compound 4e 2-(2-(3,5-bis(2-methoxy-2-oxoethoxy)phenyl)-2-oxyethyl)phthalazin-2-ium bromide forms two hydrogen bonds at positions LYS(625) and ILE(618), with bond energies, -3.5kcal/mol. NS3 protein is a particularly interesting molecular target for antiviral compounds because of it central role in the viral life cycle. Dengue virus NS3 is a multifunctional protein in which the N-terminal 180 amino acids encode a protease that mediates viral polyprotein processing [28]. This protein endowed with protease, helicase, adenosine triphosphatase (ATPase) and RNA 5' triphosphatase (RTPase) activities [29]. In examining the binding interaction and position of the compound 4a with NS3 Protease protein ligand binding site predicted, it was found that two hydrogen bonds are formed, at positions ARG(107) and ASN(105) with bond energies, -4.5 kcal/mol and -7 kcal/mol, respectively. The lower ligand binding energy for NS3 Helicase protein was proven to be compound 3e. The evaluation of amino acid positions reveals that this compound, 1-(2-(3,5-bis(2-methoxy-2-oxoethoxy) phenyl)-2-oxyethyl)pyradazin-1-ium bromide, forms two hydrogen bonds at positions ASP(284) and LEU(193).Dengue virus NS1 is a glycoprotein essential for viral replication with an unknown mechanism.29NS1 is considered a unique "viral toxin" in dengue disease [30]. Compound 4dwas found to have lower ligand binding energy (binding energy value= -130.32kcal/mol), for NS1 protein. The compound 4d 2-(2-(3,4-bis(2-methoxy-2-oxoethoxy)phenyl)-2-oxyethyl)phthalazin-2-ium bromide forms two hydrogen bonds at positions ARG(336) and SER(228) with bond energies, -6.7 kcal/mol and -3.5kcal/mol respectively.

Conclusion

Our molecular docking studies explored the possible binding modes of 20 synthesized compounds, as ligands, with different Dengue virus proteins which include capsid protein, envelope protein, NS1 protein, transmembrane domain of NS2A, NS3 protease protein, NS3 helicase protein and NS5 protein. Several interactions were observed between the ligands and the protein binding pocket like the total binding energy, Van der Waal's Force and H-bond energy, in kcal/mol. The obtained results revealed that all the 20 compounds show good affinity with all the proteins; significant affinity was observed between the tested compounds and Dengue transmembrane domain of NS2A protein. On comparing the binding energy and the binding site residues, we found that all compounds differ either in their binding modes or with the binding site residues for hydrogen bond formation. The conclusion drawn from our virtual screening and docking result was that the compound 4a has highest binding affinity with most of the proteins and it can be used as an effective drug target for Dengue virus. Though, there are many reports on the in vitro analysis of these compounds and its antioxidant properties, but there are no in silico studies that predict the binding and active regions especially with these proteins.

Conflict of Interest

The authors declare no conflict of interest.

References

1. Balan A, Florea O, Moldoveanu C, Zbancioc G, Iurea D, et al. (2009) Diazinium salts with dihydroxyacetophenone skeleton: Syntheses and antimicrobial activity. Eur J Med Chem 44(5): 2275-2279.
2. Syam S, Abdelwahab S, Al Mamary M, Mohan S (2012) Synthesis of Chalcones with Anticancer Activities. Molecules 17(6): 6179-6195.
3. Zbancioc G, Florea O, Jones P, Mangalagiu I (2012) An efficient and selective way to new highly functionalized coronands or spiro derivatives using ultrasonic irradiation. Ultrasonics Sonochem 19(3): 399-403.
4. Demirayak S, Karaburun A, Beis R (2004) Some pyrrole substituted aryl pyridazinone and phthalazinone derivatives and their antihypertensive activities. Eur J Med Chem 39(12): 1089-1095.
5. Akahane A, Katayama H, Mitsunaga T, Kato T, Kinoshita T, et al. (1999) Discovery of 6-Oxo-3-(2-phenylpyrazolo[1,5-a]pyridin-3-yl)-1(6H)-pyridazinebutanoic Acid (FK 838): A Novel Non-Xanthine Adenosine A1 Receptor Antagonist with Potent Diuretic Activity. J Med Chem 42(5): 779-783.
6. Saracoglu M, Kandemirli F (2009) The Structure-AChE Inhibitory Activity Relationships Study in a Series of Pyridazine Analogues. Med Chem 5(4): 325-335.

7. Asif M, Singh A, Siddiqui A (2011) The effect of pyridazine compounds on the cardiovascular system. Med Chem Res 21(11): 3336-3346.
8. Zbancioc A, Zbancioc G, Tanase C, Miron A, Ursu C, et al. (2010) Design, Synthesis and In Vitro Anticancer Activity of a New Class of Bifunctional DNA Intercalators. Lett Drug Des Discov 7(9): 644-649.
9. De Clercq E (2005) New Approaches toward Anti-HIV Chemotherapy. J Med Chem 48(5): 1297-1313.
10. Refaat H, Khalil O, Kadry H (2007) Synthesis and anti-inflammatory activity of certain piperazinylthienylpyridazine derivatives. Arch Pharm Res 30(7): 803-811.
11. Butnariu R, Mangalagiu I (2009) New pyridazine derivatives: Synthesis, chemistry and biological activity. Bioorg Med Chem 17(7): 2823-2829.
12. Mantu D, Cătălina Luca M, Moldoveanu C, Zbancioc G, Mangalagiu I (2010) Synthesis and antituberculosis activity of some new pyridazine derivatives. Part II. Eur J Med Chem 45(11): 5164-5168.
13. Rodenhuis-Zybert I, Wilschut J, Smit J (2010) Dengue virus life cycle: viral and host factors modulating infectivity. CMLS 67(16): 2773-2786.
14. Powers C, Setzer W (2016) An In-Silico Investigation of Phytochemicals as Antiviral Agents Against Dengue Fever. Comb Chem 19(7): 516-536.
15. Hosseinzadeh S, Jafarikukhdan A, Hosseini A, Armand R (2015) The Application of Medicinal Plants in Traditional and Modern Medicine: A Review of Thymus vulgaris. Int J Clin Med 06(09): 635-642.
16. Prakash P, Gupta N (2005) Therapeutic uses of Ocimum sanctum Linn (Tulsi) with a note on eugenol and its pharmacological actions: A short review. Indian J Physiol Pharmacol 49(2): 125-131.
17. Qaddir I, Majeed A, Hussain W, Mahmood S, Rasool N (2020) An in silico investigation of phytochemicals as potential inhibitors against non-structural protein 1 from dengue virus 4. Brazilian J Pharm Sci 56: 17420.
18. Lim S, Noble C, Shi P (2015) The dengue virus NS5 protein as a target for drug discovery. Antiviral Res 119: 57-67.
19. Tambunan U, Nasution M, Azhima F, Parikesit A, Toepak E, et al. (2017) Modification of S-Adenosyl-l-Homocysteine as Inhibitor of Nonstructural Protein 5 Methyltransferase Dengue Virus Through Molecular Docking and Molecular Dynamics Simulation. Drug Target Insights 11: 117739281770172.
20. Thisyakorn U, Thisyakorn C (2013) Latest developments and future directions in dengue vaccines. Ther Adv Vaccines 2(1): 3-9.
21. Kumar A, Voet A, Zhang K (2012) Fragment Based Drug Design: From Experimental to Computational Approaches. Curr Med Chem 19(30): 5128-5147.
22. Ganguly S (2014) Molecular Docking Studies and ADME Prediction of Novel Isatin Analogs with Potent Anti-EGFR Activity. Med Chem 4(8): 558-568.
23. Sathyamurthy B, Sushmitha H (2018) In Silico drug designing studies on Dengue Virus NS1 Protein. Pharma Tutor 6(10): 31.
24. Zbancioc A, Miron A, Tuchilus C, Rotinberg P, Mihai C, et al. (2014) Synthesis and In vitro Analysis of Novel Dihydroxyacetophenone Derivatives with Antimicrobial and Antitumor Activities. Med Chem 10(5): 476-483.
25. Zbancioc G, Zbancioc A, Mangalagiu I (2014) Ultrasound and microwave assisted synthesis of dihydroxyacetophenone derivatives with or without 1,2-diazine skeleton. Ultrasonics Sonochem 21(2): 802-811.
26. Nemésio H, Villalaín J (2014) Membrane Interacting Regions of Dengue Virus NS2A Protein. J Phys Chem B 118(34): 10142-10155.
27. Muller D, Young P (2013) The flavivirus NS1 protein: Molecular and structural biology, immunology, role in pathogenesis and application as a diagnostic biomarker. Antiviral Res 98(2): 192-208.
28. Wang C, Huang Z, Chiang P, Chen C, Wu H (2009) Analysis of the nucleoside triphosphatase, RNA triphosphatase, and unwinding activities of the helicase domain of dengue virus NS3 protein. FEBS Lett 583(4): 691-696.
29. Swarbrick C, Basavannacharya C, Chan K, Chan S, Singh D, et al. (2017) NS3 helicase from dengue virus specifically recognizes viral RNA sequence to ensure optimal replication. Nucleic Acids Res 45(22): 12904-12920.
30. Chen H, Lai Y, Yeh T (2018) Dengue virus non-structural protein 1: a pathogenic factor, therapeutic target, and vaccine candidate. J Biomed Sci 25(1): 58-69.

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