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Impact of Gemini Surfactants on the Stability of Insulin using Computational Tools

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Abstract

Herein, a theoretical model has been designed to study the aggregation behaviour of native bovine insulin using computational tools. Herein, self-association and aggregation of insulin in the presence of Gemini surfactant was investigated. The data showed the noteworthy interaction and stabilization of insulin due to the Gemini surfactants. Using Gemini surfactants having spacer of two carbons i.e., ethylene group, it was found that Gemini surfactant, 71 found to be best for the stabilization of insulin. This conclusion is based on the energy contributed due to hydrogen bonding, electrostatic interaction and Van der Waals interactions. Further, Gemini surfactant, 71 has been proven a better stabilizer when the results were compared with the reported ligands as in PDB files (1ZNI, 2HR7 and 2OLY). Further, its comparison was done with conventional surfactants. It was found that Gemini surfactant, 71 is more potent than the conventional surfactants.

Keywords: iGemdock; Insulin; Gemini surfactants; Aggregation

Introduction

Insulin is a very popular protein and it is known as metabolic hormone. It has been synthesized and secreted from beta cells of pancreas of human being. Insulin plays an important role in opening of the cell in the body and permits the glucose to be used as an energy source. With increases in the glucose levels in the plasma of the blood, an increase in the uptake and metabolism by the pancreas beta cells has been observed and it leads the insulin secretion. Many diabetic patients are advised to take insulin so they can the level of sugar and can avoid complications caused [1-8]. Insulin is stored as granules and it consists of six units. They are loosely connected by various forces e.g. hydrophobic interaction. Different modifications in the structure of insulin have been made to affect insulin [9-21]. Insulin has the tendency to undergo for the structural transformation and results in aggregation and formation of insoluble insulin fibrils. It has been the most motivating and thoroughly studied problem. The absolute mechanism of the formation of the fibril is still unclear. Therefore, most popular methods for the stabilization of the insulin against fibrillation contribute to counteract associated insulin from being disassembled [2,9-13,22]. It is evident that the stabilization mechanism is consistent with the destabilising role attributed to hydrophobic surfaces [14-26].

In view of this, surfactants have interesting properties like their interfacial and bulkiness and used to stabilize various biomolecules. Conventional surfactant has a single hydrophobic tail connected to an ionic or polar head group, whereas a Gemini surfactant has in sequence a long hydrocarbon chain, an ionic group, a spacer, a second ionic group and another hydrocarbon tail. Gemini surfactants are considerably more surface-active than conventional surfactants. Therefore, insulin can be stabilized by using different types of Gemini surfactant. In the present work, authors hypothesized that addition of Gemini surfactants to insulin can suppressed the self-aggregation tendency by decreasing the hydrophobic interactions. In this study, authors studied the effects of Gemini surfactants on structural stability of the insulin using computational tools.

Experimental Methodology

Molecular interactions are useful for identifying lead compounds and understanding ligand binding mechanisms for a therapeutic target. These interactions are often inferred from a set of active compounds that were acquired experimentally. Docking program is most likely coupled the stages of structure based docking/screening and post-analyzing modules contain several components to make the screening/analyzing procedure. The iGEMDOCK is computational tools useful generates for protein compound interaction profiles of electrostatic (E), hydrogen bonding (H), and Van der Waals interaction. It can sequentially be applied to four computational phases and it includes target and database preparation, molecular docking and post-docking analysis. If protein-ligand interactions have low energy (negative energy) indicates a stable system and thus a likely binding interaction [27-32].

Docking

It is a powerful approach for structure based drug design and structural hypotheses, how the ligands interact with the target? The ligand-protein docking is to predict the predominant binding modes of a ligand with a protein of known three-dimensional structure.

Ligand preparation

Gemini Surfactants (GS) were drawn using ChemDraw Ultra 12.0 and a library of 100 GS has been made as in Table 1 based on Figure 1. These iGEMDOCK do not accept 2D structures and takes only in MDL MOL, SYBYL MOL2 and PDB format. It is recommended

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to prepare the GS in MDL MOL format then optimize Gaussian 9.0. Then, the optimized molecules will be taken as the input of iGEMDOCK. After using iGEMDOCK best pose of GS suggested in Table 2 [27-32].

Protein preparation

Protein preparation or selecting is most important think for accurate result in molecular docking; therefore MMV 2.5 was used to see the insulin-GS interactions.

Parameters set in iGEMDOCK

Parameters in iGEMDOCK were set for the successful screening of drug like molecules are as follow: initial step sizes (r=0.8 and w=0.2), family competition length (L=2), population size (N=300), and recombination probability (pc=0.3). Optimization is set to generate 70 iterations for which it generated 1200 solutions in one generation process and if exceeded then it terminated after 84,000 solutions [27-32].

Molecular docking

All the optimized structures of Gemini surfactants were generated in .pdb format and then used for the docking to determine their energies. Molecular docking was performed using iGEMDOCK, a computational tool to determine the binding between the insulin and Gemini surfactant in terms of energy as mentioned in Table 2.

Results and Discussion

Physical or chemical degradation and aggregation of the insulin found to be the main cause of the immunogenicity. Formation of aggregates may cause the altered release profile of insulin in the body and further it may cause the normoglycemia [33-40]. Interaction between insulin and the Gemini surfactants was studied and total energy was calculated. It is the summation of the energy contributed by Van der Waals interaction, hydrogen bonding and electrostatic interactions. The binding of Gemini surfactants with the insulin is assumed to prevent the aggregation of insulin and thereon, its denaturation. Herein, the main role is played by Van der Waals interaction on binding of Gemini surfactants and insulin but other factor, energy contributed by hydrogen bonding is also

S. No.	Anion	Alkyl group (-R)	Compound no.
1	Br	-CnH2n+1 N=1-20	1-20
2	Cl		21-40
3	I		41-60
4	BF_4		61-80
5	OTf		81-100

Table 1: Details of the parent, side chain and anions in the Gemini surfactants.

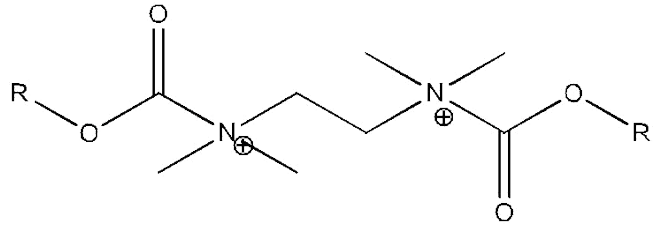


Figure 1: Structure of parent Gemini surfactant.

C. No.	Total Energy	VDW	H Bonding	Electrostatic
1	-75.01	-65.124	-10.789	0.90312
2	-71.589	-59.735	-12.048	0.1931
3	-73.419	-62.988	-9.2836	-1.1475
4	-73.8	-70.924	-2.5	-0.376
5	-83.485	-76.168	-7	-0.3173
6	-79.782	-71.157	-7.8833	-0.741
7	-80.111	-76.613	-3.8496	0.35163
8	-78.935	-74.806	-4.1293	0
9	-90.56	-86.67	-3.2961	-0.5942
10	-92.242	-92.217	-0.025	0
11	-104.05	-104.31	0	0.25297
12	-63.705	-62.171	-1.1899	-0.3448
13	-83.174	-80.852	-2.3222	0
14	-77.79	-73.641	-4.149	0
15	-84.785	-82.285	-2.5	0
16	-59.109	-55.609	-3.5	0
17	-75.233	-75.233	0	0
18	-73.528	-73.528	0	0
19	-90.162	-90.162	0	0
20	-88.983	-86.483	-2.5	0
21	-80.3064	-61.7031	-18.7257	0.122473
22	-89.81	-71.9431	-18.4496	0.582682
23	-98.7727	-86.4864	-11.9172	-0.36904
24	-96.3598	-87.5631	-8.90503	0.108329
25	-85.2168	-81.5472	-4.14294	0.473347
26	-106.546	-99.8422	-7.00153	0.297971
27	-88.8314	-73.0716	-15.7128	-0.04699
28	-99.428	-94.9088	-5.49479	0.975581
29	-103.727	-99.3615	-3.5	-0.8658
30	-93.1124	-88.1008	-4.85669	-0.15492
31	-90.5653	-90.5653	0	0
32	-78.0673	-73.4816	-4.5857	0
33	-71.5466	-69.7623	-1.78424	0
34	-59.1551	-59.1551	0	0
35	-75.5205	-75.5205	0	0
36	-76.5186	-76.5186	0	0
37	-91.0314	-88.5314	-2.5	0
38	-76.6346	-76.6346	0	0
39	-84.3433	-84.3433	0	0
40	-76.7104	-76.7104	0	0
41	-71.6326	-57.4975	-14.9127	0.777556
42	-78.234	-65.2309	-12.6083	-0.39487
43	-73.4814	-63.3934	-10.0821	-0.00595
44	-87.6183	-80.1645	-8.0813	0.627548
45	-88.7133	-84.2053	-3.97973	-0.52836
46	-83.6325	-73.92	-9.33729	-0.37527
47	-89.5599	-77.1916	-13	0.631608
48	-78.0492	-73.7588	-4.29035	0
49	-86.2523	-80.7371	-7.19778	1.68258
50	-92.3237	-85.4902	-7.26585	0.43241
51	-86.6249	-81.3812	-4.95276	-0.29102
52	-72.8342	-71.016	-1.53894	-0.27923
53	-75.9693	-69.0417	-6.92759	0
54	-82.0408	-82.0408	0	0
55	-85.1673	-85.1673	0	0
56	-71.5218	-70.1207	-1.40109	0
57	-88.5331	-85.0331	-3.5	0
58	-83.1368	-79.6368	-3.5	0
59	-76.8322	-76.2996	-0.53262	0
60	-92.8352	-92.8352	0	0

61	-75.7682	-63.6437	-13.0438	0.919312
62	-74.8403	-64.9929	-9.9732	0.125746
63	-76.4007	-72.727	-3.42943	-0.24433
64	-81.715	-72.5823	-9.42207	0.28939
65	-90.376	-74.9617	-15.9175	0.503192
66	-90.447	-81.0105	-9.5	0.063488
67	-94.79	-88.5488	-7	0.758825
68	-92.2301	-89.3785	-3.5	0.648383
69	-90.3794	-87.201	-3.5	0.321634
70	-105.548	-98.3625	-6.84109	-0.34401
71	-119.349	-109.449	-9.68788	-0.21164
72	-90.2708	-85.2303	-6.04628	1.0058
73	-95.3002	-91.8563	-3.44388	0
74	-86.1489	-83.6905	-2.45831	0
75	-85.8036	-79.6864	-6.1172	0
76	-83.1418	-77.1522	-5.98959	0
77	-87.9433	-87.9433	0	0
78	-82.4975	-79.207	-3.29047	0
79	-74.4999	-74.4999	0	0
80	-75.6408	-75.6408	0	0
81	-90.95	-76.6472	-11.9105	-2.39233
82	-96.5938	-72.7324	-20.958	-2.9035
83	-91.7757	-76.8895	-14.8862	0
84	-86.8595	-71.314	-11.9785	-3.56703
85	-115.174	-94.0959	-19.0745	-2.00357
86	-87.8546	-81.8534	-8.2625	2.26134
87	-100.295	-87.7388	-11.5823	-0.9739
88	-97.5295	-83.1953	-12.4086	-1.9255
89	-96.31	-89.31	-7	0
90	-98.9215	-92.5169	-5.60638	-0.79819
91	-69.7308	-63.7347	-5.99608	0
92	-56.3002	-56.3002	0	0
93	-75.52	-75.4384	-0.08161	0
94	-82.6149	-82.6149	0	0
95	-87.8567	-84.3567	-3.5	0
96	-80.5996	-80.5996	0	0
97	-73.8782	-73.8782	0	0
98	-87.0241	-87.0241	0	0
99	-83.3306	-83.3306	0	0
100	-87.7629	-86.2864	-1.47657	0

Table 2: Total energy, energy contributes due to Van der Waals interaction, hydrogen bonding and electrostatic interaction.

important up to decide the potential candidate. Therefore, it has been observed that the interaction between insulin and Gemini surfactant is not only driven by the hydrophobic interaction between the long chain of the surfactant and the polar amino acids of the insulin but also due to the contribution from the interactions through the polar heads. This theoretical model is based on interactions due to binding of Gemini surfactants with insulin and this approach extends the accepted criteria that association between insulin and Gemini surfactants is preferentially guided by hydrophobic interactions.

Effect of Gemini surfactant binding on the conformational stability of insulin

Potential of Gemini surfactants was studied to prevent the agitation-induced denaturation of insulin and it is of interest for the development of novel pharmaceutical formulations. An attempt to predict the ability of Gemini surfactant on the conformational stability of insulin was made. It was found that surfactant 71 gave the maximum stability to the insulin as in Table 2 and mentioned in Figure 2. Interacted amino acids with the Gemini surfactant, 71 is mentioned in Table 3 and it indicates that GS, 71 interacts with PHE, GLN, HIS, PRO, LYS and GLU of insulin (Figure 3). Further, the results obtained by the interaction of potential Gemini surfactant, 71 was compared with the ligands presents in reported PDB, 1ZNI, 2HR7 and 2OLY taken from the biological databank, RCSB are 3Zn+3Cl, Eight sulphate anions and 2Zn+5Cl+7 Urea respectively (Table 4). It was found that Gemini surfactant, 71 was better than the others as in Table 4. The energy contributed by 101-103 is only due to Van der Waals interactions and no contribution by hydrogen bonding and electrostatic interactions. Further, the potential of Gemini surfactant, 71 was compared with molecules (104-106), still 71 was found to be better stabilizing agents and more strongly interact with insulin. Interaction of conventional surfactants (107-118) with insulin was also studied and it was found that Gemini surfactant, 71 showed best interactions (Table 5). Although, compound number 104 has also shown good interactions based on the total energy but it is still less than energy contributed by 71 [41-47].

Conclusion

In the present work, authors has reported the an efficient, time saving model to study the interactions between insulin and the Gemini surfactants at the atomic level based on results from MD simulations with the computational tools i.e., iGEMDOCK. Interaction of the Gemini surfactants with the hormone, insulin predominantly via the dioxyethylene groups of their polar heads i.e., amino group and ester functionality through hydrogen bonds and Van der Waals interactions, in addition to hydrophobic interactions through the alkyl i.e., hydrocarbon tails. The docked pictorial view is in agreement of the favorable Van der Waals interaction between insulin and Gemini surfactant, 71. Further, it has been found that the Gemini surfactant, 71 bind to non-specific sites of the insulin. It has been suggested that the formation of these clusters of Gemini surfactant, 71 around the insulin structure could increase the protein conformational stability. Further, Gemini surfactants, 71 has been proven a better stabilizer than the cofactors (1ZNI, 2HR7 and 2OLY) mentioned in the reported PDB files as available on biological databank, RCSB. Even, Compound no. 71 was found even more potent than the conventional surfactants (107-118) on the basis of total energy.

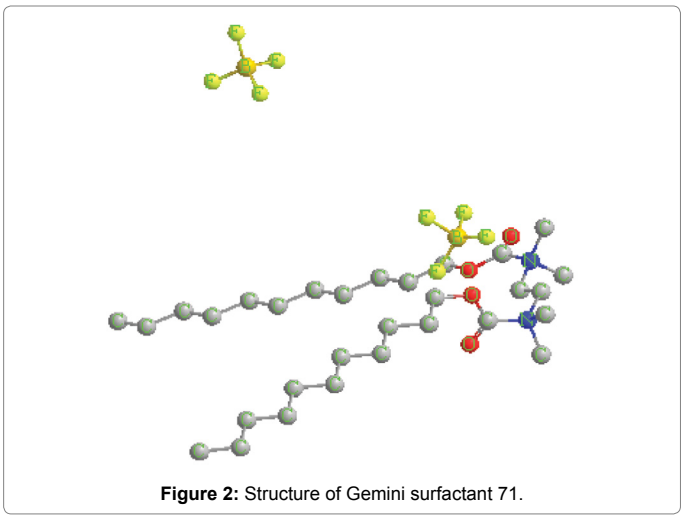
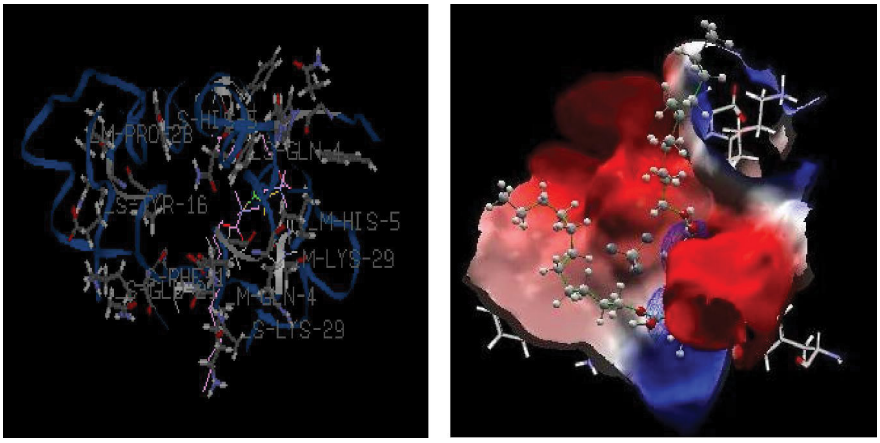


Figure 2: Structure of Gemini surfactant 71.

Surfactant		Energy due to interaction of amino-acid of insulin with GS, 71									
	Energy	V-S-PHE-	V-M-	V-S-	V-M-HIS-	V-S-HIS-	V-M-	V-M-	V-S-LYS-	V-S-TYR-	V-S-
GS	-	1	GLN-4	GLN-4	5	5	PRO-28	LYS-29	29	16	GLU-21
71	-90.6	-6.23047	-6.59286	-9.83831	-5.34681	-5.52889	-7.69984	-7.25254	-7.17315	-4.92231	-6.2785

Table 3: Interaction of Gemini surfactant, 71 with the following amino-acids of insulin.



(a) (b)

Figure 3: (a) Docked view and (b) electrostatic pictogram obtained of the interaction of Gemini surfactant 71 with insulin.

C. No.	Ligand	Total Energy	VDW	H Bond	Elec
101	3Zn + 3Cl	-30.0985	-30.0985	0	0
102	Eight sulphate anions	129.959	129.959	0	0
103	2Zn + 5Cl + 7 Urea	-1.55076	-1.55076	0	0
104	CRSALEN	-115.243	-94.9151	-20.3276	0
105	Curcumin	-95.7769	-82.5779	-13.199	0
106	Glucose	-79.6889	-35.8629	-43.8261	0

Table 4: Energy contributed by the interaction of reported molecules with insulin [41-45].

C. No.	Ligand	Total Energy	VDW	H Bond	Elec	Aver Con Pair
107	Cocamide MEA	-79.1584	-65.9245	-13.2339	0	24.6471
108	Cocamide DEA	-83.499	-68.0345	-15.4645	0	24.4
109	Sodium tridecylsulfate	-75.519	-65.2683	-7	-3.25072	22.4211
110	Cetrimonium bromide	-64.5552	-63.7413	0	-0.813863	23.125
111	Cetylpyridinium chloride	-74.0267	-72.3196	-1.7071	0	23.9444
112	Benzethonium vhloride	-88.8475	-81.8924	-6.42475	-0.530353	23.5161
113	Dimethyldioctadecylammonium bromide	-80.8777	-80.8777	0	0	13.6585
114	Sodium dioctylsulfosuccinate	-100.54	-88.7953	-11.7444	0	19.6897
115	Sodium lauryl sulfate	-90.2307	-87.7307	-2.5	0	19.069
116	Sodium undecylbenzene sulfate	-88.2449	-69.3291	-17.5	-1.41576	19.913
117	Cocoamidopropylhydroxysultaine	-79.6999	-51.9538	-24.4868	-3.25927	17.1304
118	Lauramidipropylbetaine	-83.4929	-80.3036	-3.5	0.310748	21.625

Table 5: Energy contributed by the interaction of conventional surfactants with insulin [37,38,44-47].

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