International Journal of Drug Delivery 5 (2013) 81-87

http://www.arjournals.org/index.php/ijdd/index

Original Research Article

ISSN: 0975-0215

Simulation of a Dipeptide boc-Ile-Ile-nhme as a drug carrier

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Abstract

Reverse micelles are discrete nanoscale particles composed of a water core surrounded by surfactant. In this current study the the self assembling properties of the dipeptide Boc-Ile-Ile-NHMe in chloroform to form a stable micelle at various temperatures ranging from 200 to 350 Kelvin has been analysed using insilico methods. The computational analysis was carried out using the steepest descent algorithm, a minimization tool used to study the protein energy level in insilco and it was compared with the thermodynamic parameters determined experimentally. Such reverse micelles finds a vast area of application one of which is drug delivery in nanotechnology. The present dipeptide is shown to carry drugs by insilico methods.

Keywords: Reverse micelles, tetra peptide, minimization, drug delivery, nanotechnology

Introduction

Reverse micelles (RMs) are discrete nanoscale particles composed of a water core surrounded by surfactant. The amount of water within the core of reverse micelles can be easily manipulated to directly affect the size of the reverse micelles [1]. The structure of reverse micelles has been studied by various physical methods *viz* flurorescence, NMR, neutron scattering, X-ray scattering, quasi-elastic light scattering, calorimetry, and infrared and terahertz spectroscopy. In addition, other techniques such as diffusion acoustics and densitometric analysis can be used to gather information on the aggregation and volumetric properties [2]. Peptide modules offer an interesting model to investigate the molecular forces which are involved in the self assembly of complex biological systems. Self assembling peptides are known to form various types of ordered aggregates such as micelles [3], monolayers, membranes and nanotubules [4], in aqueous and nonaqueous solutions. In apolar media, the driving force for aggregate formation is attributed to the high solvophobic property of the -NHC (=O)]-groups [5]. The strong directional nature of the amide groups, combined with their ability to form hydrogen bonds, may result in interesting entropy–enthalpic interactions. Therefore, a search for the peptide components containing solvophobic methyl groups which can form persistent packing motifs is expected to be worthwhile [6]. These models will also be useful in exploring the contribution of hydrogen bonding and solvophobic interactions to the free energy change involved in the pre-assembly of peptides. Individual surfactant molecules that are in the system but are not part of a micelle are called "monomers." Lipid micelles represent a molecular assembly in which the individual components are thermodynamically in equilibrium with monomers of the same species in the surrounding medium. In a micelle, the hydrophobic tails of several surfactant molecules assemble into an oil-like core, the most stable form of which has no contact with water. The extent of lipid solubility is determined by the unfavorable entropy contribution due to the ordering of the water structure according to the hydrophobic effect [7].

The physicochemical studies of the peptide aggregates and understanding the nature of hydrodynamic characteristics are necessary for rationalizing the biochemical activities of such peptides. The work also gains importance as aggregates of peptides are the main causative reason for amyloid related diseases[8]. Although the interiors of folded proteins, or protein domains, are compact and well-packed[9-11], the packing of aminoacid residues is not perfect (i.e., proteins are compressible). In addition to imperfect packing, a significant fraction of protein compressibility originates from the finite rigidity of the intermolecular interaction potentials. Gekko and Nogushi [12] pointed out that, in addition to imperfect packing, the presence of internal cavities [13] contributes positively to protein adiabatic (isoentropic) compressibility as well as volume changes. When such voids are large enough to accommodate water molecules, internal water can then act as a structure stabilizer [14] by maintaining good hydrogen bonds between the domains and filling sites of imperfect packing. As a universal solvent, water interacting at the protein surface contributes negatively to protein volume and compressibility. Difficulties in interpreting the precise changes in protein volume and compressibility induced by hydration at protein-water interfaces have led to empirical estimates of the intrinsic protein volume, that is, the volume that cannot be penetrated by the solvent [15]. There is no consensus on the exact value of the compressibility of the solvent-inaccessible protein core, or intrinsic protein compressibility, due to the diverse approaches used by different investigators [16].

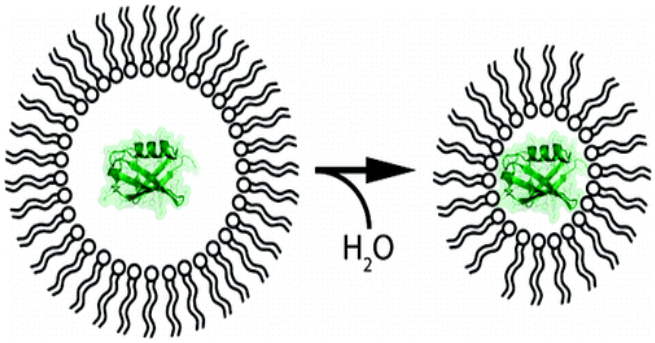

Figure 1: The structure of Reverse micelles with drug

Computer simulation, especially molecular dynamics simulation, has become an important and widely used tool in the study of biomolecular systems with the growing availability of high speed desktop computers and cluster computing. However, as computational resources grow and algorithms and theory improve, we can strive to develop truly accurate macromolecular computer experiments[17]. The structure of Reverse micelles with drug shown in the Figure 1. The tetrapeptide Boc-Ile-Gly-Met- Thr(Bzl)-OBzl is found in the β-structure region of proteinase of HIV-1 (RP 93-96). The micelle formation of the above tetrapeptide is analysed in terms of the thermodynamic functions.

In this current study we report here that the dipeptide Boc-Ile-Ile-NHMe self assembles in chloroform to form stable Reverse Micelles at various temperatures ranging from 200 to 350 Kelvin. The computational analyses were carried out using the steepest descent algorithm a minimization tool used to study the protein energy level in Insilco. One of the important application of reverse micelles is in drug delivery. So the drugs were selected from the Drug bank, based on the inner volume of reverse micelles and three different drug molecules were screened. They are Ciprofloxacin, Dapsone and Anastrozole. These drugs were packed in the inner core of Reverse Micelles. The drug molecules were packed using Packmol package. The drug with Reverse Micelles was minimized and energy also calculated using Discovery Studio2.0

Materials and Methodology

Construction of Dipeptide water and Chloroform using ChemSketch

The structure of Boc-Ile-Ile NHMe dipeptide was drawn using Chemsketch tool. Structure mode is used for drawing chemical structure. Similarly the structures of water and chloroform were also drawn using the chemical formula. The structure of the elements is readily available in the chemsketch tool. Template window offers the structures of Alkaloids, carbohydrates, DNA/RNA, steroids and sugars. Using the template structures, the chemical structure of the above mentioned dipeptide was drawn. SMILES notations of this structure was generated using the 'Generate SMILES notation' option in chemsketch.

Selection of Drugs from Drug Bank

The inner volume of the Reverse Micelles were calculated using insilico methods. Drugs with volume lesser than the volume of the inner core of the RMs were selected. The selected drugs are: (i) Ciprofloxacin which is an antibacterial drug also used in the treatment and prevention of anthrax, (ii) Dapsone which is also an antibacterial drug used in the treatment of Mycobacterium leprae (leprosy) and (iii) Anastrosole which is used in the treatment of breast cancer after surgery and also for the metastasis in pre and post menopausal women.

Conversion of chemical structures to 3D format using Smi2Depict

Smi2Depict is available in the ChemDatabase. It generates 2D images from the SMILES notation, from which the structure can be easily converted to 3D format. The SMILES notation of the dipeptide structure was provided as input for Smi2Depict. The obtained 2D format is converted to 3D format with the help of the options provided in Smi2Depict.

Packmol Program

The reverse micelle structure of the Boc-Ile-Ile.NHMe was created using the packmol package. First, the path for the packmol program was set in the specified directory.

For example

C:\set path= packmol

The Structure file and the required parameters along with structure file of the water and chloroform molecules were typed in a wordpad. The parameter used

```
tolerance 2.0
structure water.pdb
        number 1
        inside sphere  0. 0. 0. 6.
       end structure
structure kumu.pdb
                    number 20
                    atoms 26
                    inside sphere  0. 0. 0. 7.
              end atoms
 atoms 12
   outside sphere  0. 0. 0. 12.
              end atoms
end structure
structure chlor.pdb
        number 400
        inside box  -30. -30. -30.   30. 30. 30.
 outside sphere  0. 0. 0. 14.
  end structure
output 20mol.pdb
```

Save the wordpad with .inp extension.

Running packmol

The saved wordpad file was run in the command line. The command to run the file
packmol<filename.inp

A similar process of packmol was carried out for the drug with reverse micelles.

Simulation of Reverse Micelle

The constructed reverse micelles and the drugs Ciprofloxacin, Dapsone, Anastrozole with reverse micelles were minimized using the Steepest Descent Method algorithm and their respective energies were calculated using Acclerys Discovery studio2.0.

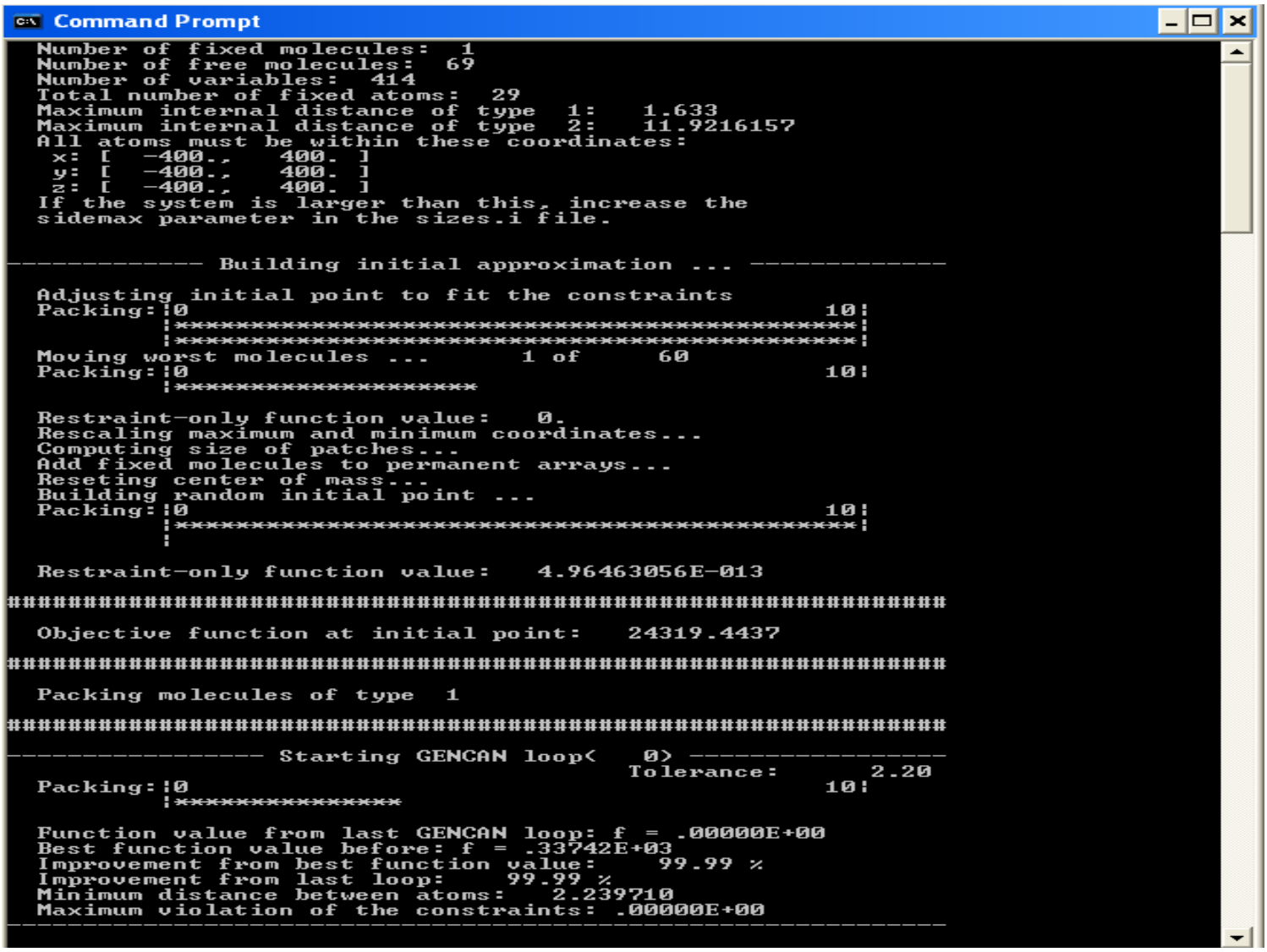

Figure 2:Running packmol in command line

Results

Construction of peptide

The chemical structures of Boc-Ile-Ile-NHMe group were drawn using Chemsketch and the SMILES notation was generated for structure. The SMILES notations were further submitted to ChemDB to retrieve their original chemical structures. The chemical structure of the dipeptide is given in the Figure 3.

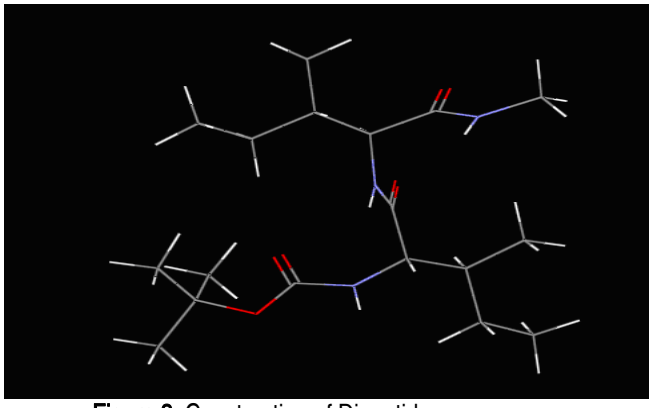

Figure 3: Construction of Dipeptide

Constructed Reverse Micelle Using Packmol

The reverse micelle structure of the Boc-Ile-Ile.NHMe was created using the packmol package. Seven different aggregation such as 20 dimer, 43 dimer, 57 dimer, 61dimer, 58dimer, 50 dimer, 40 dimer of the peptide was constructed by using packmol is shown in the Figure 4. Constructed Reverse Micelle structure is viewed using Discovery Studio 2.0

Minimization of Reverse Micelle with and without drug

The seven different aggregates of the constructed dipeptide and the reverse micelle structure of the Boc-Ile-Ile.NHMe was minimized using the Steepest Descent Method algorithm this algorithm works in such way of slow to converge the structures ,

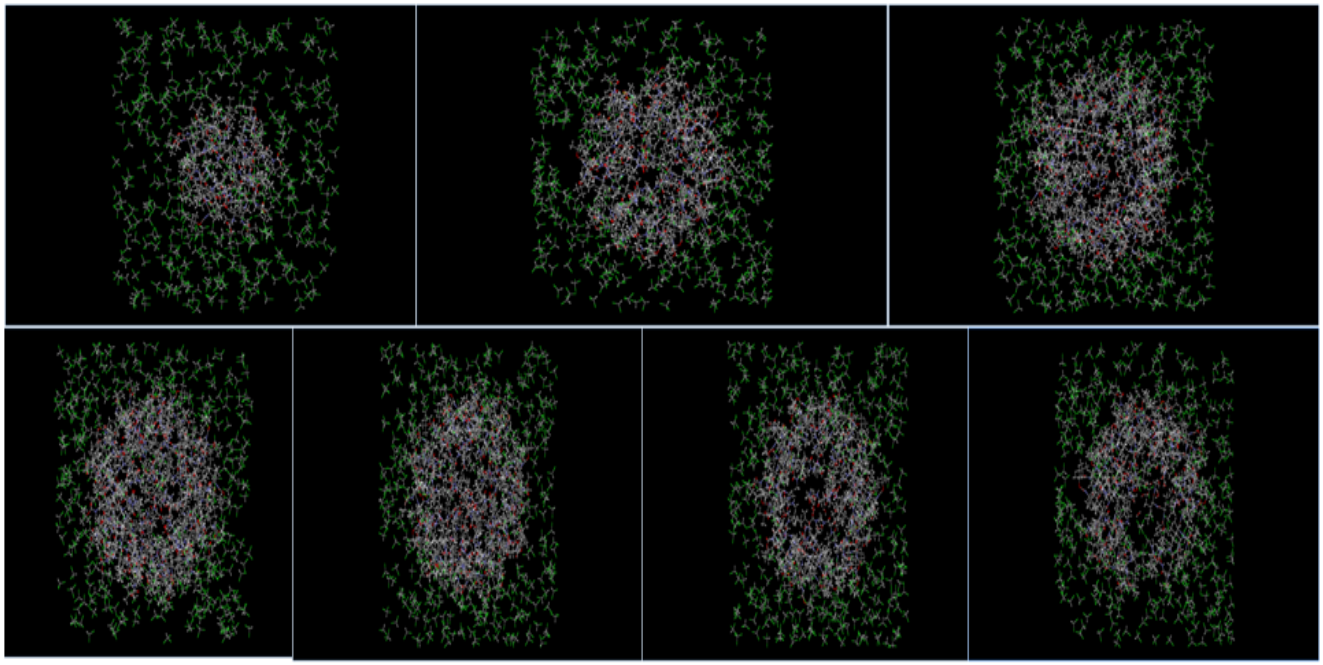

Fig ure 4 : Reverse micelles constructed using packmol in Seven different forms such as 20 dimer, 43 dimer, 57 dimer, 61dimer, 58dimer, 50 dimer, 40 dimer

and requires large number of energy evaluations is shown in the Figure 5. The energy of seven different aggregates entropy values both experimental and theoretical values are compared and tabulated in Table 1. Further the drugs such as Ciprofloxacin, Dapsone, Anastrozole is incorporated with Reverse Micelle and the minimization of the aggregates were carried out same as like that of reverse micelles without drug. The drug with reverse micelles is shown in the Figure 6, 7 and 8 respectively.

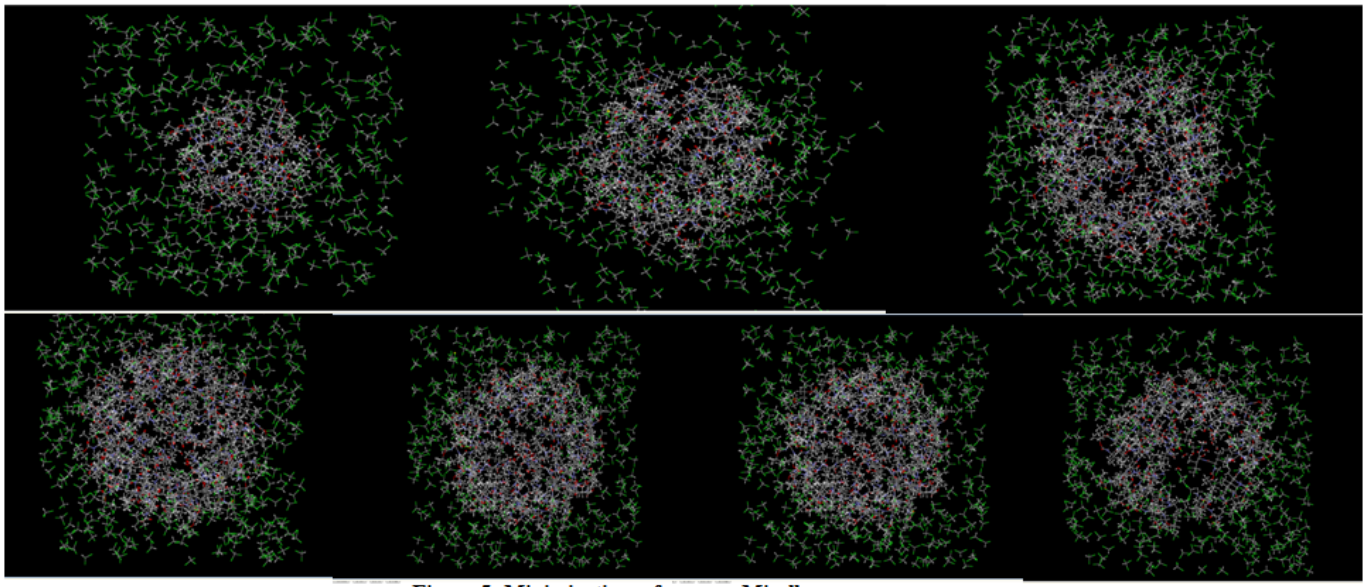

Figure 5 :Minimization of reverse Micelles

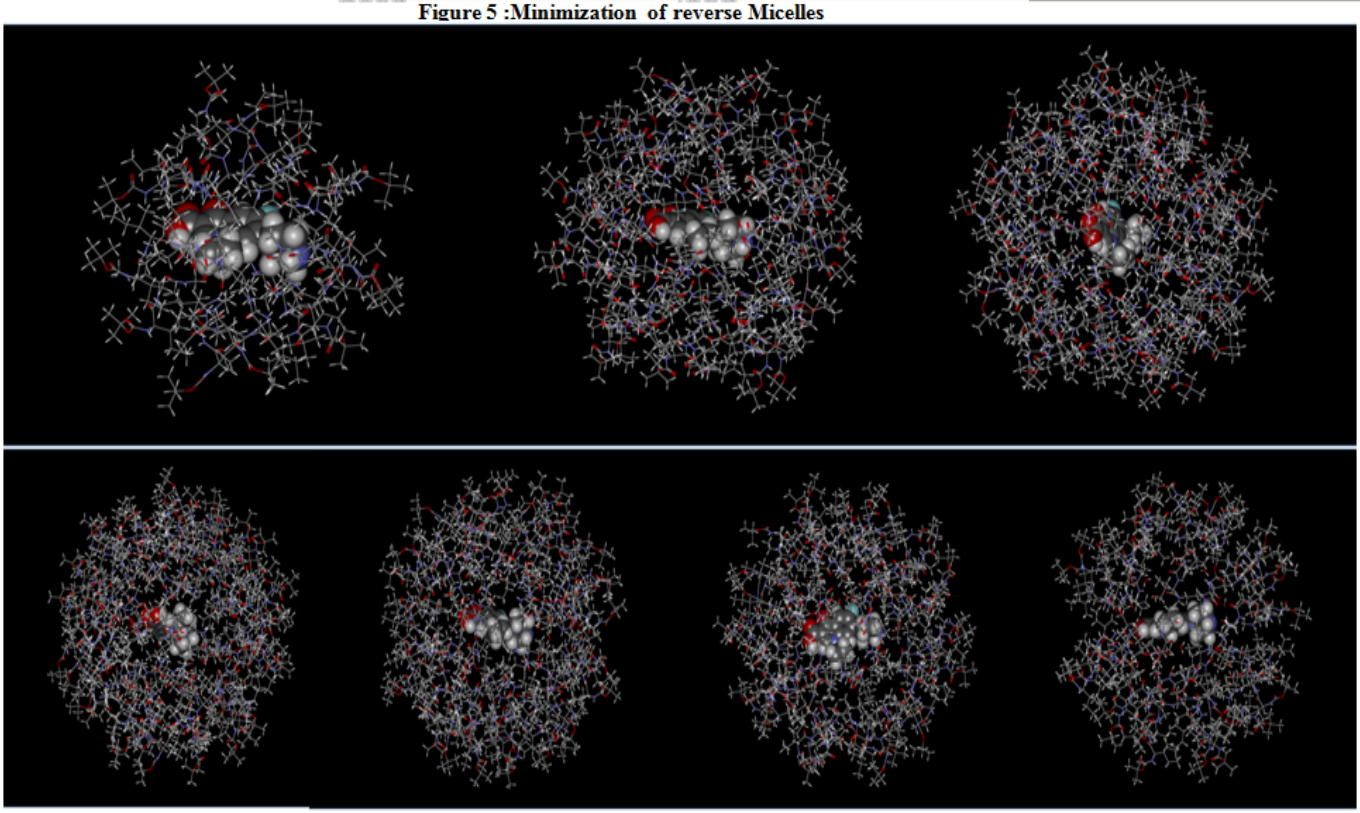

Figure 6 : Reverse Micelles with Ciprofloxacin drug

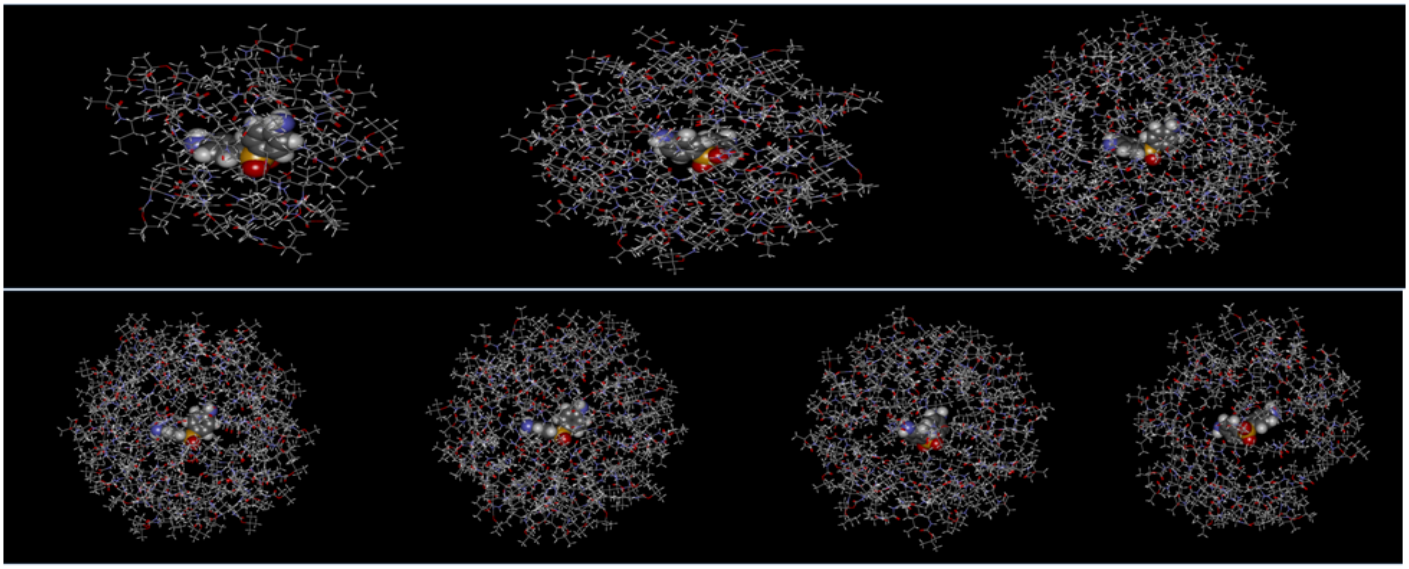

Figure 7: Reverse Micelles with Dapsone drug

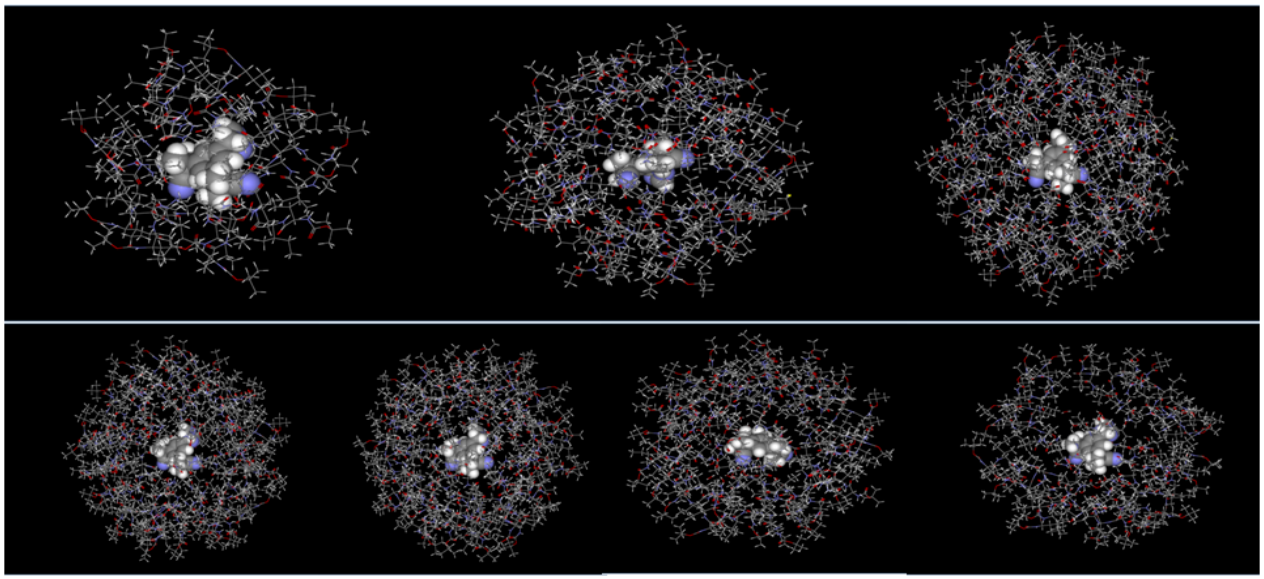

Figure 8 : Reverse Micelles with Anastrozole drug

Table 1: The free energy value of the Reverse micelles

Aggregation Number	Free Energy	
	Experimental Value* in KJ/Mol	Computational Value in KJ/Mol
20	-16.36	-16.205
43	-14.456	-14.819
57	-13.307	-13.317
61	-13.178	-13.158
58	-14.096	-14.978
50	-14.592	-14.813
40	-15.188	-15.360

*The values along with other thermodynamic parameters have been determined in wet lab and published [18].

Discussion

The Reverse Micelles of the dipeptide Boc-Ile-Ile-NHMe was constructed with seven different (20 dimer, 43 dimer, 57 dimer, 61dimer, 58dimer, 50 dimer, 40 dimer) aggregation numbers. Water and chloroform were added in the inner core region and outer surface of Reverse Micelles respectively. The initial configuration was done using packmol package. After the initial configuration, the reverse micelle structures were minimized using Discovery Studio 2.0 and the energy values like initial energy, potential energy, electrostatic energy, van der waals energy, solvation energy, RMS gradient, nonpolar term, and free energy values were calculated. The free energy values calculated in Discovery Studio 2.0 were found to be in good agreement with the experimental value shown on the Table 1. The free energy of the dipeptide RM and other thermodynamic parameters have been determined in the wet lab study and the results are published [18].

Conclusion

Hence, The Reverse Micelles with different Aggregation numbers were constructed and their free energy was calculated. Computational value of Free energy was shown to be same as experimental value. The constructed Reverse Micelles were then constructed with insertion of drugs. In future, this work is yet to be continued to study the delivery of the drugs using the Reverse Micelles at different $_{P}H$ condition and temperature.

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