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Energy Study at Different Temperatures for Active Site of Azurin in Water, Ethanol, Methanol and Gas Phase by Monte Carlo Simulations

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ABSTRACT

The interaction between the solute and the solvent molecules play a crucial role in understanding the various molecular processes involved in chemistry and biochemistry, so in this work the potential energy of active site of azurin have been calculated in solvent by the Monte Carlo simulation. In this paper we present quantitative results of Monte Carlo calculations of potential energies of active site of azurin in water, ethanol, methanol and gas phase at three temperatures 300, 305, 310 K.

According to the obtained results, the potential energy of active site conformation is decreased quickly in water and the greater stability in this study is related to water and then methanol. It has been found that in different solvent media the highest potential energy value and then the least stability correspond to ethanol and also through increasing the dielectric constant of solvent the structural energy values decreased. Thus, the protein environment, which is often aqueous, affects the structure, folding dynamic and stability, and, therefore, the functionality of globular proteins. In fact, solvent-protein interactions, together with the interactions between residues in the protein matrix, facilitate the folding process and establishment of intermolecular interactions with other complex systems. Furthermore, to be properly folded and fully functional, a protein requires a minimum level of hydration.

Keywords: Monte Carlo; Azurin; Potential energy, Solvent effect

INTRODUCTION

Blue copper proteins, like azurin, belong to a class of mononuclear copper proteins that contain a so-called type-1 blue copper site. The metal is strongly bound by the thiolate sulfur of a cysteine and the nitrogen of two histidines, in an approximately trigonal arrangement, while the fourth (axial) coordination position is occupied either by the thioether sulfur of a methionine or the oxygen atom of a glutamine [1].

These proteins are relatively small (8-14 kDa) and function in electron transport.

The structural and electronic properties of the metal sites, which are responsible for the peculiar spectroscopic and redox properties of these species, have been thoroughly investigated and are still subject of extensive experimental and theoretical work [2,3,4].

Unfortunately, a biomolecule-water potential energy surface cannot be constructed from accurate ab initio calculations, even with recent growth in computer power, because too many points are required [4].

The Monte Carlo method is a powerful tool in many fields of mathematics, physics, engineering and chemistry . There are two main directions in

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the development and study of Monte Carlo algorithms. The first is Monte Carlo simulation, where algorithms are used for simulation of real-life processes and phenamena. In this case, the algorithms just follow the corresponding physical, chemical or biulogical processes under consideratinn. In such simulations Monte Carlo is used as a tool for choosing one of many different possible outcomes of a particular process. The second direction is Monte Carlo numerical algorithms [5-6]. Monte Carlo numerical algorithms can be used for solving deterministic problems by modeling variables or random fields. Some Monte Carlo simulations have been succeeded to investigate proteins studies. The Metropolis Monte Carlo was originally developed for calculating equilibrium properties of physical systems [7]. The dynamic interpretation of the MC algorithm for the protein pracess has been widely used in many studies [8-11]. The Metropolis algorithm performs a sample of the configuration space of a system starting from a random conformation and repeating a large number of steps.

The aim of the present work is to describe and characterize the molecular structure, vibrational properties active site of azurin. In this work, the structure of a model coordination compound for the active site of azurin was discussed computationally. Thus, it is worthwhile to collect information on their structures by the means of computational chemistry as well. The ioteraction between the solute and the solvent molecules play a crucial role in understanding the various molecular processes involved io chemistry and biochemistry [12]. Numerous biological processes involve an ioo binding to a nucleic acid or protein and thereby displacing the water hydration. Water play a crucial role for the stability, dynamics, and function of proteins [13]. For this reason Molecular Dynamics (MD), Monte Carlo (MC) and Langevin Dynamic (LD) simulations must account for the effects that this solvent has, both on proteio structure and on protein dynamics [14]. In this paper we present quantitative results of Monte Carlo calculations af potential energies of active site of azurin in water, ethanol, methanol and gas phase

THEORETICAL BACKGROUND AND COMPUTATIONAL METHOD

In molecular simulations, 'Monte Carlo' is an importance sampling technique. It Makes random move and produce a new conformation and calculate the energy change ΔE for the new conformation and accept or reject the move based on the Metropalis criterion:(Boltzmann factor)

$$P = \exp(-\frac{\Delta E}{kT}) \qquad (1)$$

If $\Delta E<0$, P>1, accept new conformation; Otherwise. P>rand (0,1), accept, else reject[15]. Monte Carlo simulation is widely applied in the fields of chemistry, biology, physics, and eagineering in order to determine the structural and thermodynamic properties of complex systems at the atomic level. Thermodynamic averages of molecular properties can be determined from Monte Carlo methods, as can minimum energy structures [16]. Let (f) represents the average value of some coordinate-dependent property f(x), with x representing the 3N Cartesian coordinates needs to locate all of the N atoms. In the canonical ensemble (fixed N, V and T, with the volume and T the absolute temperature), averages of molecular properties are given by an average of f(x) over the Boltzmann distribution:

$$(f) = \int dx\, f(x) \exp[-\beta U(x)] / \int dx \exp[-\beta U(x)] \qquad (2)$$

where U(x) is the potential energy of the system, $\beta = 1/k_B T$, aad k_B is the Boltzmann constant. If one can compute the thermodynamic average of f(x) it is theo possible to calculate various thermodynamic properties. In tbe canonical ensemble it is most common to calculate E, the intemal energy, and C_V, the constant-volume heat capacity (although other properties can be calculated as well). For example, if we average U(x) over all possible configurations according to Eq. (2) then E and C_V are given by

$$E = 3Nk_B T / 2 + (U) \qquad (3)$$

$$C_V = 3Nk_B / 2 + (U^2) - (U^2) / (k_B T^2) \qquad (4)$$

Tbe first term in each equation represents the contribution of kinetic energy, which is analytically integrable. In the harmonic (low temperature) limit, E given by Eq. (3) will he a linear function of temperature an C_V from Eq (4) will be constant, in accordance with the equipartition theorem.

Water plays a crucial role for the stability, dynamics, and function of proteins. For this reason Monte Carlo (MC) simulations must account for the effects that this solvent has. both on protein structure and on protein dynamics [14-17].

In this study, active site of azurin was simulated using Monte Carlo method with 100 ps steps at three temperatures 300, 305, 310K. Then the system was placed in a box (12 ×12 × 12 nm) containing 24 molecules of water (Fig.1). At next stage, water molecules around the active site of azurin converted to ethanol and methanol by using chem3D program and simulated using Monte Carlo method. It is essential to say that, HyperChem uses the Metropolis method. Kinetic, potential and total energy calculated by Monte Carlo simulation.

Fig. 1. Schematic representation of structural model of active site of azurin in water.

RESULTS AND DISCUSSION

In this paper, we have used Monte Carlo methods to study active site of azurin in confined environments and different temperatures. The Run step and delta max for Monte Carlo simulation were 0.001 and 20000. respectively.

Monte Carlo simulations are commonly used to compute the average thermodynamic properties of a molecule or a system of molecules, and have been employed extensively in the study of the structure and equilibrium properties of molecules [18]. Monte Carlo simulations employ a statistical sampling technique to generate configurations, which represent a trajectory in phase space [19]. Monte Carlo calculation evaluates the averages of the ensemble directly by sampling configurations from the statistical ensemble.

Monte Carlo is generally better in sampling the allowed states of a system. and; thus, can often calculate the average properties more quickly and accurately The total energy of the system, in this method, is called Hamiltonian, which is the sum of kinetic and potential energy: [20]

$$E = E_K + E_P \quad (5)$$

Monte Carlo simulation was carried out on the two systems, gas and solvent active site of azurin. In Table 1, the potential energy is calculated by Monte Carlo simulation in gas, water, ethanol and methanol.

All simulations were performed at three temperatures 300. 305, 310 K. Each solvent system was immersed in a periodic water box, and the structures of water molecules were maintained. A 100 ps time step was used in all the simulations. The diagram of potential energy has been drawn as functions of temperature and environment (Fig.2).

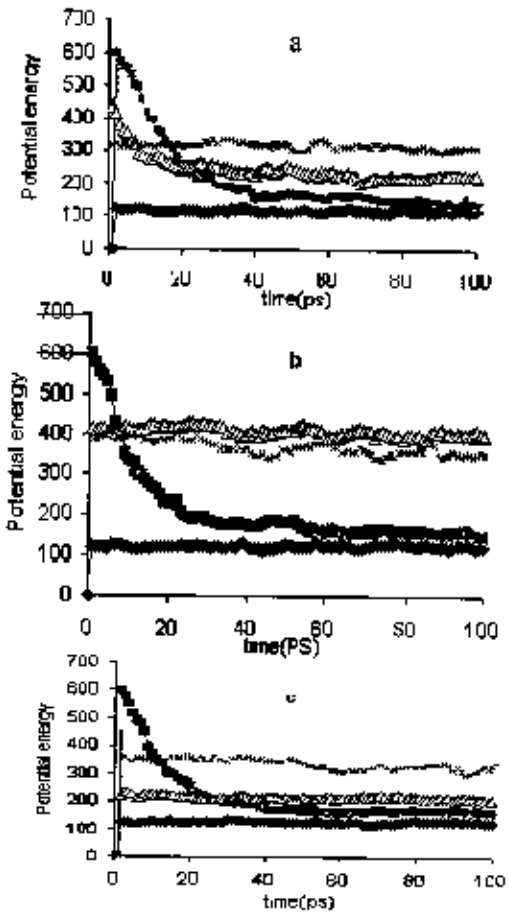

Fig. 2. The potential energy (kcal/mol) via time (ps) during Monte Carlo (MC) simulation at 300K(a), 310K(b), 315K(c) in gas, water methanol and ethanol environments.

It can be seen from Fig. 2 that, the potential energy of active site conformation is decreased quickly at 20 ps time step MC in water.

Figures have shown the function of the reduced temperature and environment. Low temperatures promote complex structure stability, whereas high temperatures oppose it. The reduction in energy translates into structure predictions of increased accuracy [21].

Solute-solvent pair interaction of potential energies was shown that the greater stability of solvent observed over all states investigated in this study is related to water and then methanol. The reduction in energy results in improved accuracy of the corresponding protein structure predictions [22-23].

Results are presented in Table 1(a,b) that was indicated potential energies of active site of azurin in three temperatures and various dielectric constants. By changing the dielectric constant from water toward ethanol (Fig.3-a), the highest peak on the graphs of potential energy versus dielectric constant which is corresponded to ethanol at three temperatures and after passing that region the decreasing trend has been obvious and in water solvent the peak reaches to the lowest value.

From Fig. 3-b we can see that by changing the dielectric constant from water toward methanol in three temperatures, the highest values of potential energy have been yielded in the range of 58 up to 68 of dielectric constant and the highest peak on the graphs of potential energy versus dielectric constant is corresponded to D = 63.2.

Table1. Potential energy calculation (kcal/mol) in three temperatures via dielectric constant for Azurin (a , b)

ε	T=300 K	T=318 K	T=315 K
78.39	149.14	150.95	165.65
73.9	160.87	138.55	155.75
69.4	157.6	163.27	155.62
64.9	165.63	170.5	178.06
60.4	199.59	179.87	183.39
55.96	200.99	197.3	199.34
51.5	240.13	204.89	206.9
47	240.55	221.42	225.57
42.5	254.39	218.44	256.32
38	250.71	256.51	241.66
33.5	317.88	270.32	268.72
29	285.87	264.49	277.42
24.55	383.08	355.53	342.81
1	125.92	119.47	124.17

(a)

ε	T=300 K	T=310 K	T=315 K
78.39	149.14	150.95	165.65
74.57	155.97	131.028	145.4
70.77	221.333	170.73	163.67
66.95	223.499	172.126	178.36
63.14	7451.8	7414.2	7397.9
59.33	3058.5	3040.1	3055.2
55.52	180.155	192.19	176.99
51.69	181.434	187.97	179.96
47.88	173.88	177.54	180.1
44.87	198.121	189.99	183.41
48.26	210.716	200.93	183.88
36.44	205.94	223.16	202.63
32.63	221.35	223.43	192.62
1	125.92	119.47	124.17

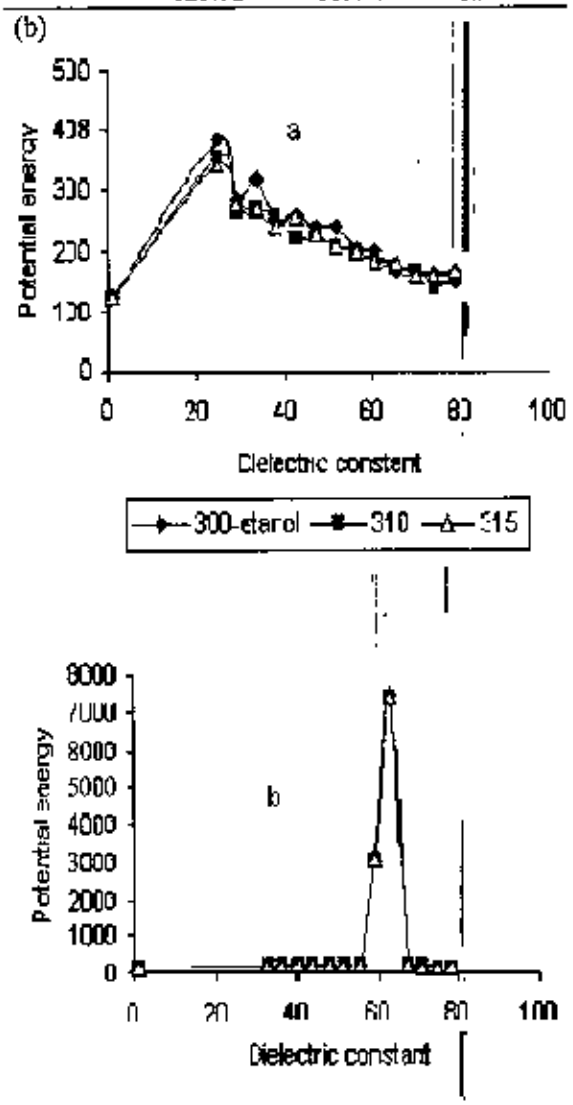

Fig. 3. The potential energy (kcal/mol) via dielectric constant during Monte Carlo (MC) simulation at 300K, 310K, 315K (a) in gas, water and ethanol, (b) in gas, water and methanol environments.

CONCLUSION

The stability of active site of azurin in aqueous solution was investigated by means of Monte Carlo simulation, with the aim of determining the thermodynamic stability of the protein and of characterizing the thermally induced conformational changes of its active site.

The solvent– induced effect on conformational energies and structural stability of active site of azurin is a critically important feature that should be seriously identified in order to find out the unique physico–chemical properties of this protein Structural investigations of active site of azurin in general show a relation between the solvent and the structural stability of similar biological compounds [24]. These results are in agreement with the common chemical concepts.

It has been found that in different solvent media the highest potential energy value and then the least stability correspond to ethanol and also through increasing the dielectric constant of solvent the structural energy values decreased.
In the case of different dielectric constants effect of methanol on potential energy the result show that the highest value of potential energy have been obtained in the range of 58 up to 68.

Thus, the protein environment, which is often aqueous, affects the structure, folding dynamic and stability, and, therefore, the functionality of globular proteins [24-27]. In fact, solvent-protein interactions, together with the interactions between residues in the protein matrix, facilitate the folding process and establishment of intermolecular interactions with other complex systems. Furthermore, to be properly folded and fully functional, a protein requires a minimum level of hydration [4].

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