In silico drug designing against DNA topoisomerase 1 for brain cancer treatment

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ABSTRACT

Drug designing is the approach of finding drugs by design, based on their biological targets. Typically a drug target is a key molecule involved in a particular metabolic or signaling pathway that is specific to a disease condition or pathology, or to the infectivity or survival of a microbial pathogen. The structure of the drug molecule that can specifically interact with the biomolecules can be modeled using computational tools. These tools can allow a drug molecule to be constructed within the biomolecule using knowledge of its structure and the nature of its active site. Construction of the drug molecule can be made inside out or outside in depending on whether the core or the R-groups are chosen first. However many of these approaches are plagued by the practical problems of synthesis. One of the computational tools used in drug designing is "chemsketch", which works with 65% accuracy. The drug structure was downloaded from the drug database and the structure is modified by introducing alcohol, methyl, sodium hydroxide etc. onto the functional groups of the drug. The new drugs obtained undergo various molecular modeling and dynamics to reduce their energy levels. Docking is performed using geometrically optimized molecules as ligands and protein DNA topoisomerase I as protein, converting their chemsketch forms into PDB format using SPDB viewer. HEX software is used for this purpose.
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KEYWORDS

DNA Topoisomerase I;
Chemsketch;
Docking;
Ligand;
PDB;
SPDB.

INTRODUCTION

Cancer is a class of diseases or disorders characterized by uncontrolled division of cells and the ability of these cells to spread, either by direct growth into adjacent tissue trough invasion, or by implantation into distant sites by metastasis (where cancer cells are transported through the blood stream or lymphatic system). Cancer may affect people at all ages, but risk tends to increase with age. It is one of the principal causes of death in developed countries.

Cell division or cell proliferation is a physiological process that occurs in almost all tissues and under many circumstances. Normally the balance between proliferation and programmed cell death is

tightly regulated to ensure the integrity of organs and tissues. Mutations in DNA that lead to cancer disrupt these orderly processes. The uncontrolled and often rapid proliferation of cells can lead to either a benign tumor or a malignant tumor (cancer). A brain tumor is a noncancerous (benign) or cancerous (malignant) growth in the brain, whether it originates in the brain or has spread (metastasized) to the brain from another part of the body. Brain tumors are equally common among men and women, but some types are more common among men and others are more common among women. Brain tumors are occurring with increasing frequency among older people.

DNA topoisomerases play important roles in basic cellular biology. Recently they have been identified as the molecular targets of a variety of pharmaceutical agents. Some of the drugs that target the topoisomerases are anticancer drugs. These anticancer drugs work by a novel mechanism of action. They inhibit the topoisomerase molecule from religating DNA strands after cleavage. This leaves a cell with DNA breaks, which if not repaired, become lethal. In other words, these drugs convert the topoisomerase molecule into a DNA damaging agent. This is a stoichiometric relationship. Each anticancer drug molecule has the potential of interacting with one topoisomerase molecule to cause one DNA lesion. The clinical implication of this mechanism of drug action is that sensitivity to topoisomerase targeting drugs should be dependent on high topoisomerase levels.

Camptothecin is an alkaloid (348 molecular weight) produced by the Chinese tree Camptotheca acuminata and was identified as an antineoplastic agent in the 1960s by Wall and Wani. Early studies with camptothecin indicated that cellular exposure to the drug resulted in DNA breaks; the interaction with topoisomerase I was identified by Liu and colleagues in the 1980s. Because the camptothecin alkaloid is relatively insoluble in aqueous solutions, initial clinical trials with camptothecin used a sodium salt derivative.

Although responses occurred in these trials, severe myelosuppression or cystitis was observed fre-

quently, and the drug was deemed too toxic for clinical use. Subsequently, it was discovered that in the salt derivatives, the lactone at position 20 in camptothecin is hydrolyzed to a carboxylic acid, with this ring opening significantly decreasing the activity of the compound. Further development of camptothecins led to two water-soluble derivatives, topotecan and irinotecan, that can be delivered as lactones and are currently approved for the treatment of cancer. Several other camptothecin analogues are in clinical development.

Hydroxycamptothecin (SN-38) Camptothecin is a five-ringed heterocyclic alkaloid Certain substitutions in the A ring may augment topoisomerase poisoning, presumably by increasing drug binding to the topoisomerase I-DNA cleavage complex. By contrast, substitutions in the E ring often abrogate activity. Indeed, the stereochemistry of C20 in the E ring is critical, with the (R)-isomer inactive.
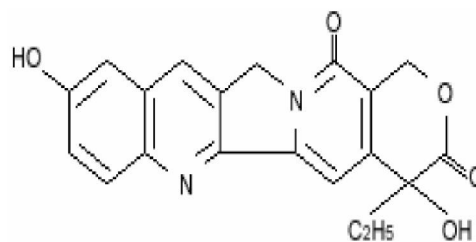
$C_{20}H_{16}N_2O_5$ is the molecular formula of this compound. It has a molecular weight of 364.36 with a formulation of light yellow white crystalline powder and 98% purity. Irinotecan is a prodrug; the piperidino group present at C10 is hydrolyzed by plasma or tissue carboxylesterases to SN-38, which is much more active than irinotecan in inducing topoisomerase I-mediated DNA damage.

There are two other notable derivation strategies that have produced drugs currently in clinical testing: (1) 7 silyl congeners, designed to enhance lipophilicity and stabilize the E ring lactone, and (2) 20 esters, designed as prodrugs to prevent hydrolysis of the E ring. The present findings, as well as other reports that the hydroxy lactone ring of camptothecin is critical for antitumor activity in vivo, correlate with the structure-activity relationships at the level of topoisomerase I and support the hypothesis that antitumor activity is related to inhibition of

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this target enzyme.

This study is concerned with the modeling of a new drug for brain cancer making DNA topoisomerase as a target and camptothecin as the drug used. The drug is modified structurally and its derivatives obtained as such are tested for their affinities in binding with the protein.

MATERIALS AND METHODS

Materials

Chemsketch software, 10-hydroxycamptothecin, Hex 0.8 software.

Methods

The method used for modeling a new drug for brain cancer aiming DNA topoisomerase I as target and 10-hydroxycamptothecin as ligand includes the following steps:

1 Drawing chemical structure of the ligand using chemsketch software
2 Converting it to 3D structure.
3 Measuring bond distances, bond angles and torsion angles.
4 Performing Single point calculation, Geometry optimization by setting the molecular mechanics to force field.
5 Measuring the optimized values.
6 The chemical structure of ligand molecule is then changed by causing variation in its R group; as such 9 new different molecules have been designed.
7 The same procedure that's followed for the ligand molecule is undergone.
8 The protein into which the ligand molecule is fit is considered and the same protein is used for the rest 9 molecules to check the optimization energy that is obtained using force field, when fit into the protein.
9 The ligand derivatives after undergoing optimization along with protein are made devoid of protein and the optimization values are calculated using the force fields
10 These calculated energy values are denoted as 'Y_1'.

11 The optimized ligand molecule and its derivatives are converted to PDB format along with the selected protein and docking is performed using HEX software.
12 The best rankings of the considered molecules are obtained and the fitness energy of each molecule is noted down

Description of the methods mentioned above

Method 1

Chemsketch

It is a versatile molecular modeler and editor and a powerful computational package. It offers many types of molecular and quantum mechanics calculation.

It includes functions like:

• Drawing molecules from atoms and converting them to 3D models.
• Constructing proteins and nucleic acids from standard residues
• Using molecules from other sources like Brookhaven PDB files and rearranging them.
• Setting up and directing chemical calculations including molecular dynamics, by various mechanical or abinitio methods and graphing the results.
• Solvating molecules in a periodic box

Single point calculation

Single point calculations determine the molecular energy and properties for a given fixed geometry. It determines the total energy (in Kcal/mole) and the gradient of a molecular system or of selected atoms in one particular calculation. With a semi empirical or ab- initio method, a single point calculation also determines the electron (charge) distribution in the system. The name "single point" reflects the fact that we calculate a single pre-selected configuration.

Geometry optimization

Geometry optimization is used to find minima on the potential energy surface, with these minimum energy structures representing equilibrium structures. Optimization also is used to locate transition structures, which are represented by saddle points (The

highest point on the pathway between two minima is known as saddle point with the arrangement of atoms being in the transition structure) on the potential energy surface.

Optimization to minima is also referred to as energy minimization. During minimization, the energy of molecules is reduced by adjusting atomic coordinates. It is applied to model-built structures as well as to those derived from coordinate data banks. Energy minimization is done when using either molecular mechanics or quantum mechanics methods and it must precede any computational analyses in which these methods are applied. For example, geometry optimization can be used to

a   characterize a potential energy surface.

b   obtain a structure for a single-point quantum mechanical calculation, which provides a large set of structural and electronic properties.

c   prepare a structure for molecular dynamics simulation - if the forces on atoms are too large, the integration algorithm may fail.

The energy obtained from the potential energy function at the optimized geometry is sometimes called a steric or conformational energy. These energies can be used to calculate differences between stereo isomers and between isologous molecules (i.e., those differing in connectivity but having the same number of each type of functional group). These energies apply to molecules in a hypothetical motionless state at 0 Kelvin. Additional information is needed to calculate enthalpies (e.g., thermal energies of translation, vibration, and rotation) and free energies (i.e., entropy). The geometry of a molecule determines many of its physical and chemical properties. This is why it is very important that we understand the geometry of a molecule when running computations. In computational chemistry we are specifically concerned with optimizing:

•   Bond angles
•   Bond distances (angstroms)
•   Dihedral angles (degrees)

The repulsion forces of the valence electrons directly affect the size of the bond angle. The bond angle is the angle formed by two pairs of valence electrons and the central atom that connects the two.

The stronger the repulsion strength, the larger the bond angle. The torsional energy is defined between every quartet of bonded atoms, and depends on the dihedral angle ö made by the two planes incorporating the first and last three atoms involved in the torsion. Torsional motions are generally hundreds of times less stiff than bond stretching motions. The reason for including torsional energies is to ensure the correct degree of chain rigidity. They mimic the steric hindrance of neighboring atoms and their side-groups to rotation about the chain axis.

The non-bonded energy represents the pair-wise sum of the energies of all possible interacting non-bonded atoms. The non-bonded energy accounts for repulsion, Vander Waals attraction, and electrostatic interactions. The determination of a molecule's geometry has been fairly simple: identify the valence electron pairs and determine the geometry. According to VSEPR model, the geometry of a molecule is determined by the repulsion forces of its valence electron pairs However, the VSEPR model is only a visual model and does not give us the detail needed in computational chemistry. For computational chemistry we need to be more precise by using cartesian coordinates, bond lengths and bond angles to find the optimal molecular geometry.

The arrangement of atoms in the molecules and more specifically thelectrons around the atom determine the energy level of that molecule. In fact, the energy of a molecular system varies even with small changes in its structure. This is why geometry is so important when performing calculations. The objective of a geometry optimization is to find the point at which the energy is at a minimum because this is where the molecule is most stable and most likely to be found in nature. It is, therefore, the purpose of geometry optimizations to locate the minima based on some geometry for the molecule. Programs generally work to find a stationary point, a point on the potential energy surface where the forces are zero. They do this by first calculating the first derivative of the energy (also known as the gradient).

At the minima, of the gradient the derivative of the energy with respect to its coordinates is zero, and has thus reached a stationary point. Geometry

optimization calculations employ energy minimization algorithms to locate stable structures. Two minimization algorithms are provided.

Steepest descent algorithm

The steepest descent algorithm is an old mathematical tool for numerically finding the minimum value of a function, based on the gradient of that function. Steepest descent uses the gradient function (or the scalar derivative, if the function is single-valued) to determine the direction in which a function is increasing or decreasing most rapidly. Each successive iteration of the algorithm moves along this direction for a specified step size, and the re-computes the gradient to determine the new direction to travel. This calculation moves directly down the steepest slope of inter-atomic forces on the potential energy surface. This method makes limited changes to the molecular structure and is useful for correcting bad starting geometry or removing bad contacts. It is most effective when the molecular system is far from minimum, and it is less satisfactory for macro-molecular systems.

Polak Ribiere method

Polak Ribiere method is a conjugate gradient method using one-dimensional searches converging more quickly than steepest descent, but using slightly more memory. RMS gradient - The root-mean-square (RMS) gradient is set to determine the end of the calculations. When the RMS gradient is less than the value we enter, the calculation ends. Cycles - A number is entered to limit the number of search directions. The default value is 15 times the number of atoms. In vacuo - It removes the periodic boundaries from the calculation. Periodic boundary conditions: Uses the periodic boundary conditions that exist for the molecular system.

Molecular mechanics

Four force fields provide computationally convenient methods for exploring the stability and dynamics of molecular systems. Added flexibility of user defined atom types and parameters. Along with MM+, a general purpose force field three specialized bimolecular force fields: Amber, BIO+, and OPLS, Mixed Mode Calculations are used. HyperChem's molecular mechanics methods have many applications to the study of molecular structure and stability. Some typical applications are:

- Calculating relative conformational energies of a series of analogous structures.
- Re-optimizing a peptide after introducing a selective mutation.
- Refining structures prior to more rigorous quantum mechanics calculations.
- Assessing possible steric effects in a reactive intermediate.

To simulate the effects of solvent attenuation of electrostatic interactions, Chemsketch offers a distance-dependent dielectric constant option for selected force fields.

Molecular dynamics

One of the principal tools in the theoretical study of biological molecules is the method of molecular dynamics simulations (MD). This computational method calculates the time dependent behavior of a molecular system. MD simulations have provided detailed information on the fluctuations and conformational changes of proteins and nucleic acids. These methods are now routinely used to investigate the structure, dynamics and thermodynamics of biological molecules and their complexes. They are also used in the determination of structures from x-ray and from NMR experiments.

The molecular dynamics method was first introduced by Alder and Wainwright in the late 1950's (Alder and Wainwright, 1957, 1959) to study the interactions of hard spheres. The first molecular dynamics simulation of a realistic system was done by Rahman and Stillinger in their simulation of liquid water in 1974 (Stillinger and Rahman, 1974).

The first protein simulations appeared in 1977 with the simulation of the bovine pancreatic trypsin inhibitor (BPTI) (McCammon, et al, 1977). Today in the literature, one routinely finds molecular dynamics simulations of solvated proteins, protein-DNA complexes as well as lipid systems addressing a variety of issues including the thermodynamics of ligand binding and the folding of small pro-

teins. The number of simulation techniques has greatly expanded; there exist now many specialized techniques for particular problems, including mixed quantum mechanical - classical simulations that are being employed to study enzymatic reactions in the context of a full protein.

MD is a form of computer simulation wherein atoms and molecules are allowed to interact for a period of time under known laws of physics, giving a view of the motion of the atoms. Because molecular systems generally consist of a vast number of particles, it is impossible to find the properties of such complex systems analytically; MD simulation circumvents this problem by using numerical methods. One of MD's key contributions is creating awareness that molecules like proteins and DNA are machines in motion. MD probes the relationship between molecular structure, movement and function. It is a specialized discipline of molecular modeling and computer simulation based on statistical mechanics.

Biological molecules exhibit a wide range of time scales over which specific processes occur; for example:

1) Local Motions (0.01 to 5 Å, 10^{-15} to 10^{-1} s)
    a   Atomic fluctuations
    b   Side chain Motions
    c   Loop Motions
2) Rigid Body Motions (1 to 10Å, 10^{-9} to 1s)
    a   Helix Motions
    b   Domain Motions (hinge bending)
    c   Subunit motions
3) Large-Scale Motions (> 5Å, 10^{-7} to 10^{4} s)
    a   Helix coil transitions
    b   Dissociation/Association
    c   Folding and Unfolding

Molecular dynamics simulations permit the study of complex, dynamic processes that occur in biological systems. These include, for example,

- Protein stability
- Conformational changes
- Protein folding
- Molecular recognition: proteins, DNA, membranes, complexes
- Ion transport in biological systems and provide

the mean to carry out the following studies,

- Drug Design

Design of a molecular dynamics simulation should account for the available computational power. Simulation size (n=number of particles), time step and total time duration must be selected so that the calculation can finish within a reasonable time period. However, the simulations should be long enough to be relevant to the time scales of the natural processes being studied. To make statistically valid conclusions from the simulations, the time span simulated should match the kinetics of the natural process. Otherwise, it is analogous to making conclusions about how a human walks from less than one footstep. Most scientific publications about the dynamics of proteins and DNA use data from simulations spanning nanoseconds (1E-9s) to microseconds (1E-6s).

For simulating molecules in a solvent, a choice should be made between explicit solvent and implicit solvent. Explicit solvent particles must be calculated expensively by the force field, while implicit solvents use a mean-field approach. The impact of explicit solvents on CPU-time can be 10-fold or more. But the granularity and viscosity of explicit solvent is essential to reproduce certain properties of the solute molecules. In all kinds of molecular dynamics simulations, the simulation box size must be large enough to avoid boundary condition artifacts. Boundary conditions are often treated by choosing fixed values at the edges, or by employing periodic boundary conditions in which one side of the simulation loops back to the opposite side, mimicking a bulk phase.

Molecular dynamics simulations compute classical trajectories for molecular systems. Quantum forces can be used to model reactive collisions. Heating, equilibration, and cooling periods can be employed for simulated annealing and for studies of other temperature dependent processes. Both constant energy and constant temperature simulations are available. Temperature is an important parameter to be considered in MD simulations as molecules behaviour is highly dependent on it. Commonly we have experience with macroscopic temperatures,

which involve a huge number of particles. But temperature is a statistical quantity.

If there is a large enough number of atoms, statistical temperature can be estimated from the instantaneous temperature, which is found by equating the kinetic energy of the system to $nk_BT/2$ where n is the number of degrees of freedom of the system. A temperature-related phenomenon arises due to the small number of atoms that are used in MD simulations. In the canonical ensemble, moles (N), volume (V) and temperature (T) are conserved. It is also sometimes called constant temperature molecular dynamics (CTMD).

In NVT, the energy of endothermic and exothermic processes is exchanged with a thermostat. A variety of thermostat methods are available to add and remove energy from the boundaries of an MD system in a realistic way, approximating the canonical ensemble. Popular techniques to control temperature include the Nosé-Hoover thermostat and Langevin dynamics.

Method 2

Docking

Three-dimensional molecular structure is one of the foundations of structure-based drug design. Often, data are available for the shape of a protein and a drug separately, but not for the two together. Docking is the process by which two molecules fit together in 3D space. In the field of molecular modeling, docking is a method which predicts the preferred orientation of one molecule to a second when bound to each other to form a stable complex. Knowledge of the preferred orientation in turn may be used to predict the strength of association or binding affinity between two molecules using for example scoring functions.

Docking is frequently used to predict the binding orientation of small molecule drug candidates to their protein targets in order to in turn predict the affinity and activity of the small molecule. Hence docking plays an important role in the rational design of drugs. Molecular docking can be thought of as a problem of "lock-and-key", where one is interested in finding the correct relative orientation of

the "key" which will open up the "lock" . Here, the protein can be thought of as the "lock" and the ligand can be thought of as a "key". Molecular docking may be defined as an optimization problem, which would describe the "best-fit" orientation of a ligand that binds to a particular protein of interest.

However since both the ligand and the protein are flexible, a "hand-in-glove" analogy is more appropriate than "lock-and-key". During the course of the process, the ligand and the protein adjust their conformation to achieve an overall "best-fit" and this kind of conformational adjustments resulting in the overall binding is referred to as "induced fit". The focus of molecular docking is to computationally stimulate the molecular recognition process.

The aim of molecular docking is to achieve an optimized conformation for both the protein and ligand and relative orientation between protein and ligand such that the free energy of the overall system is minimized.

Method 3

HEX (HEXADECIMAL 0.8)

HEX (hexadecimal) is a genetic algorithm for docking flexible ligands into protein binding sites. *Hex* is an interactive protein docking and molecular superposition program, written by Dave Ritchie. *Hex* understands protein and DNA structures in PDB format, and it can also read small-molecule SDF files. As of October 2013, there have been about 33,000 downloads. It provides all the functionality required for docking ligands into protein binding sites from *prepared* input files and it is meant to be used in conjunction with a modeling program since we will be required to create and edit starting models, e.g. add all hydrogen atoms, including those necessary for defining the correct ionization and tautomeric states of the residues. Input files will also need to be created in the appropriate format and the results visualized in third party software. Commonly used molecular modeling environments include SYBYL and Insight II.

Binding free energy

This energy is the minimum energy required by

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R = C₂H₅
Molecular weight = 367.30amu
Molecule 1

R = OH
Molecular weight = 353.31amu
Molecule 2

R = H
Molecular weight = 337.37amu
Molecule 3

R = Cl
Molecular weight = 371.76amu
Molecule 4

R = CH₂OH
Molecular weight = 367.34amu
Molecule 5

R = CCl₂OH
Molecular weight = 436.23amu
Molecule 6

R = NH3
Molecular weight = 353.33amu
Molecule 7

R = CH₂CH₂CH₃
Molecular weight = 379.39amu
Molecule 8

R = CF₂OH
Molecular weight = 402.31amu
Molecule 9

R = CH₃
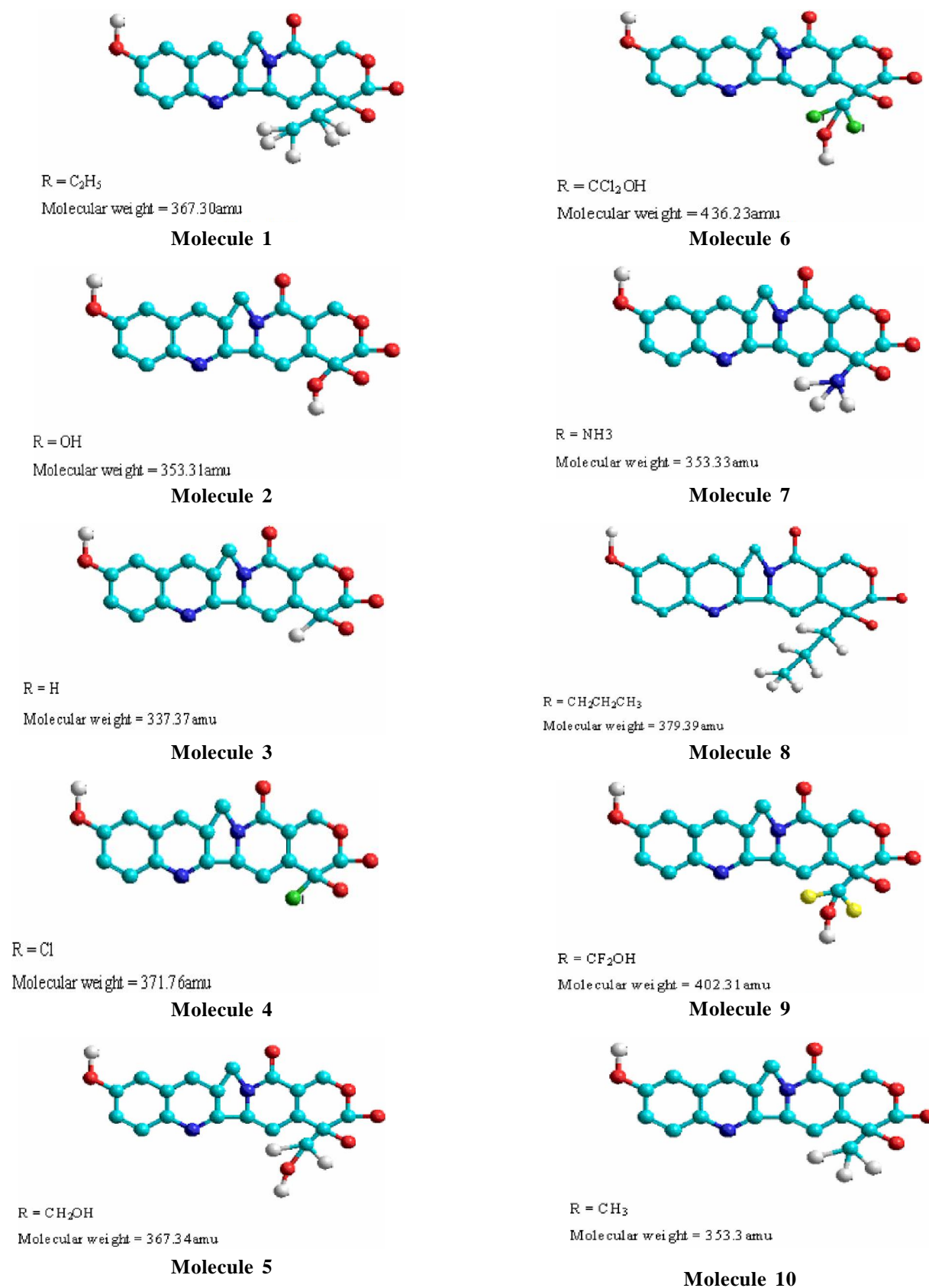Molecular weight = 353.3amu
Molecule 10

Figure 1 : Molecular weight, R-group and 3-D strctures of ligands (Molecules 1-10)

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the ligand molecule to bind to the target with maximum stability. The stable state of the molecule obtained with the protein-ligand optimization is used for the calculation of binding free energy. The most rapid methods for estimation of binding free energies are so-called empirical or knowledge-based (statistical) scoring approaches, which are based on very simple energy functions or on the frequency of occurrence of different atom-atom contact pairs in complexes of known structure, respectively. The simplicity of the energy function along with the lack of conformational sampling and explicit water treatment makes these approaches very fast, but usually at the cost accuracy.

Binding affinity

In biochemistry, a ligand is a molecule that is able to bind to and form a complex with a biomolecule to serve a biological purpose. In a narrower sense, it is an effector molecule binding to a site on a target protein, by intermolecular forces such as ionic bonds, hydrogen bonds and Van der Waals forces. The docking (association) is usually reversible (dissociation).

Actual irreversible covalent binding between a ligand and its target molecule is rare in biological systems. ligand binding to receptors alters the chemical conformation, i.e. the three dimensional shape of the receptor protein. The conformational state of a receptor protein determines the functional state of a receptor. The tendency or strength of binding is

TABLE 1 : List of chemical names and R-groups of ligands

Ligand	R-Group	Chemical Name
1	C_2H_5	[2,3] dihydro-1 H-pyrrolo [3,4-b] quinoline [1,4] ethyl-3-(hydroxymethyl) pyridio-2(1H)-one
2	CCl_2OH	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,4]-[dichloro(hydroxyl)methyl]-3-(hydroxymethyl)pyridine-2(1H)-one
3	CF_2OH	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,4]-[difloro(hydroxyl)methyl]-3-(hydroxymethyl)pyridine-2(1H)-one
4	$CH_2CH_2CH_3$	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,3]-(hydroxymethyl)-4-propylpyridin-2(1H)-one
5	CH_2OH	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,3],4-bis(hydroxymethyl) pyridine-2(1H)-one
6	CH_3	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,3]-(hydroxymethyl-4-methylpyridine-2(1H)-one
7	Cl	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,3]-chloro-3-(hydroxymethyl)pyridine-2 (1H)-one
8	H	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,3]-(hydromethyl)pyridine-2(1H)-one
9	NH_3	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,3]-(hydroxymethyl)-4-propylpyridin-2-(1H)-one
10	OH	2-methyl-2,3-dihydro-1 H-pyrrolo[3,4-b]quinoline[1,4]-hydroxy-3-(hydroxymethyl)pyridine-2-(1H)-one

TABLE 2 : Molecular characteristics of ligands

Ligand	R-Group	Molecular Formula	Molecular Weight	Ligand Optimization Energy With Protein	Total Energy (kcal/mol)
1	C_2H_5	$C_{18}H_{16}N_2NaO_2$	315.321	13.167852	37.5337
2	CCl_2OH	$C_{17}H_{12}CL_2N_2NaO_3$	386.184	13.689668	62.2183
3	CF_2OH	$C_{17}H_{12}F_2N_2NaO_3$	353.274	12.201728	77.191
4	$CH_2CH_2CH_3$	$C_{19}H_{18}N_2NaO_2$	329.347	10.784	65.4239
5	CH_2OH	$C_{17}H_{14}N_2NaO_3$	317.293	11.320295	58.973
6	CH_3	$C_{17}H_{14}N_2NaO_2$	301.294	10.331671	50.9474
7	Cl	$C_{16}H_{11}ClN_2NaO_2$	321.712	10.677284	56.155
8	H	$C_{16}H_{12}N_2NaO_2$	387.267	7.587224	53.8309
9	NH_3	$C_{19}H_{18}N_2NaO_2$	329.347	8.764965	58.1226
10	OH	$C_{16}H_{12}N_2NaO_3$	303.267	12.136456	57.687

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TABLE 3 : Measurement of bond distances, bond angles and torsion angles for a 3D structure of ligand and its derivatives

R-group	Bond angle		Bond Distance	
	3D structure	Optimized structure	3D structure	Optimized structure
C_2H_5	H-O-C =109.47° H-C-C =119.99° C=C-H=119.89° C-N=C=117.58° C-C-H =111.93°	C-O-H =120.13° C-C-H =120.07° C-C-H =120.3° C-N=C=116.34° C=C-H=120.53°	C-O =1.36A° C=C =1.33A° C-N =1.408A° C=O=1.22A° C-C =1.45A°	C-C =1.405A° C=C =1.416A° N=C =1.36A° N-C =1.37A° C=O =1.23A°
CCl_2OH	H-C-O=109.09° C-O-C=109.54° C-C-Cl=109.47° C-O-H=109.47° Cl-C-O=109.47°	C-N=C=120° O=C-C=120.36° Cl-C-O=108.27° C-C-Cl=111.39°	C-N =1.320A° C=O=1.22A° C-H =2.143A° C-O =1.43A° C-C l=1.76A°	C-N=1.488A° C=N=1.511A° C-Cl=1.76A°
CF_2OH	H-O-C =109.47° C-N=C =117.58° C-C-F =109.47° F-C-F =109.47°	C-O-H =108.441° H-O-C =108.76° C-C-F =110.20° F-C-F =108.76°	C-H=1.43A° N-C=1.49A° C-F=1.36A C-O=1.43A	C-H =1.08A° N-C =1.37A° C-F =1.49A° C-O =1.41A°
$CH_2CH_2CH_3$	H-O-C =109.471° H-C-C =119.999° C=C-H =119.89° C-N=C =117.581° N=C=O=121.893° H-C-H =109.47	C-O-H =120.135° C=C-H =119.726° C-N=C =116.3456° C=C-H =120.539° N=C=O=120.247° H-C-H = 108.35°	C-H=1.09A° N-C=1.49A° C-O=1.43A°	C-N = 1.353A° C-O =1.41A°
CH_2OH	H-O-C =109.47° O-C=C=119.71° C=C-H=120.98° C=C-N=121.22° H-C-N=119.38°	C-O-H =120.09° O-C=C=120.64° C=C-H=119.26° H-C-N=120.52° C-N=C=121.13°	O-H =0.960A° O-C =1.36A° C-H =1.08A° C=N =1.520A°	O-H=0.959A° O-C=1.350A° C=N=1.305A° C-N=1.41A°
CH_3	C-N=C=117.58° C-C-H=109.09° C-O-C=109.49° H-O-C=109.47° H-C-H=109.47°	C=N-C =120.394° C=C-H =119.871° H-C-H =108.99° C-O-H =108.441 °	C-O=1.36A° C-N=1.408A° C-H=1.09A°	C-H =1.08A° C-O =1.41A° C-N = 1.353A°
Cl	C-O-H =109.47° O-C-Cl =109.39° N=C=O=121.76° C-C-H =111.90°	N=C=O=120.42° O-C-C =108.98° C-O-H =108.58° H-C-C =122.65° O-C-Cl =109.76°	C=N=1.511A° C=O=1.22A° C-O=1.43A° C-Cl=1.76A°	C=N=1.520A° C-O=1.36A°
H	C-N=C =120° N=C=O=121.76° C-C=O =121.76° O-C-O =109.39°	N-C-H =111.71° C-C-N =106.15° N=C=O=120.42° C-C=O =120.72°	C=N =1.511A° C-H =2.143A° C-O =1.43A°	C-H=1.08A° C=N=1.305A° C=O=1.23A°
NH_3	H-C-C=119.99° C-N=C=117.58° N=C=O=121.89° C-C-N=109.47° H-N-H=109.47° H-N-C=109.47°	C-C-H=111.996° C=N-C=120° C-C- N=109.00° N-C-O=107.24° H-N-H=106.82° H-N-C=111.90°	C-N=1.07A° C=O =1.22A° C-H =1.43A°	C-N=1.04A° H-C=1.08A°
OH	H-O-C=109.47° O-C=C=119.71° C=C-H=119.38° H-C-N=119.38° C-C=N=118.58° N=C=O=121.76°	N-C-H=111.71° C-C-N=106.15° N=C=O=120.42° C-C-H=110.92° O-C-C=108.98° C-O-H=108.58°	O-C=1.36A° C-H=1.08A° C=N=1.520A°	O-C=1.350A° C-H=1.08A° C=N=1.305A°

Regular Paper

called affinity. Ligands include substrates, inhibitors, activators, and neurotransmitters.

RESULTS AND DISCUSSION

The measurement of bond distances, bond angles and torsion angles for a 3D structure of ligand and its derivatives along with the molecular dynamics and monte carlo simulations followed by optimisation energy values are tabulated and noted as follows:

CONCLUSION

Molecular modeling method has been used for modeling a new molecule for brain cancer using 10hydroxycamptothecin, a drug which's already designed. This drug is drawn using chemsketch, and its R group is modified by replacing different functional groups like OH, NH_3, H, CH_2OH, F, Cl, CH_3 etc in its place. The molecules designed as such are optimized using different algorithms and their affinity is checked with the protein. The binding free energy of the protein is calculated by performing docking process. The molecule with minimum binding energy will have the maximum binding affinity. The binding free energy is calculated by the formula Z = Sum of the energy of optimized ligand devoid of solvation parameters and the energy of the protein-ligand optimization. The binding free energy of the designed molecules is obtained by eliminating the energy of the main molecule i.e., 10hydroxycamptothecin. From the results obtained its clear that ligand 5 and 3 have the maximum binding affinity. So these molecules are determined as the best lead molecules targeting DNA topoisomerase I for curing brain cancer computationally.

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