Molecular modeling of ribavirin - DNA interaction

L.E. Vîjan, and C. M. Topală

Abstract**—**Ribavirin is a purine nucleoside analogue that is active against a number of DNA and RNA viruses. Our experimental results point out that in the ribavirin - nucleic acid complexes, the 1,2,4-triazole-3-carboxamide chromophore is intercalated between the bases of the nucleic acid helix, the carboxamidic group is set outside of the helix toward the major groove, the 2-hydroxymethyl-tetrahydrofuran-3,4-diol fragment being located in the minor groove. In this work, the interactions of two polymorphic modifications of ribavirin (denoted V_1 and V_2) with nucleic acids by the molecular mechanic method were investigated. In order to stress the sequence specificity of drug, some model mono- and double- stranded nucleic acid containing the bases: adenine (A), thymine (T), cytosine (C) and guanine (G) in AAAAAA, TTTTTT, CCCCCC, GGGGGG, ATATAT, CGCGCG, ATCGAT and CGATCG sequences were used. The theoretical results points out that the complexes of ribavirin with nucleic acids are stabilized mainly by van der Waals forces involving the 1,2,4-triazole-3-carboxamide chromophore and the nitrogenous bases from the nucleic acids structure and that the electrostatic term brings a minimal contribution to the binding energy. For both ribavirin conformers, a slight preference for nucleic acids sequences containing adenine and/or thymine bases was found. As a result of the ribavirin – nucleic acid interaction, only the nucleic acids structure is significantly perturbed, the structure of the drug being practically unchanged. In addition, the theoretical calculations in the ribavirin – nucleic acid system predict an increase of the distance between the adjacent bases at the intercalation site level. The turning of the polynucleotidic helix is produced and the "accordion type" motion takes place determining a breaking in the hydrogen bonds between base pairs from double-stranded nucleic acid structure.

Keywords**—** molecular modeling; nucleic acids; ribavirin

I. Introduction

In last years increased attention has been focused on the ways in which many drugs interact with biological systems. In the cell, many drugs, particularly those with planar chromophores, bind to nucleic acids, which are attractive targets for these molecules. Because of the complex structure of deoxyribonucleic acid (DNA) and ribonucleic acid (RNA), different binding modes are possible: intercalation between adjacent base pairs, minor or major groove binding, crosslinking, covalent attachment or some combination of these modalities [1-13].

The intercalation and minor-groove modes have been better characterized compared with other binding modes. In addition, mixed mode of binding is well-known phenomenon. For example, a lot of drugs, such as anthracycline antibiotics (daunomycin, doxorubicin) [14, 15], can intercalate a polycyclic and planar fragment between adjacent base pairs from nucleic acid helix and, also, can bind to the minor-groove of nucleic acid. Detailed information about the structural aspects of binding are given by X-ray diffraction methods or NMR spectroscopy.

The essence of intercalation is the insertion of a planar aromatic chromophore, possessing two or three rings, between adjacent base pairs from nucleic acid, thereby extending and stabilizing the double helix structure. The complexes formed in these cases are stabilized by van der Waals interactions between the drug molecule and the nitrogenous bases from nucleic acid that surround it [6, 7]. There is evidence that the intercalating drugs have a sequence selectivity by two or three base pairs [13].

Drugs that bind in grooves of nucleic acids have several aromatic rings, such as benzene, furan or pyrrole, connected by bonds possessing torsional freedom. The complexes formed in these cases are stabilized by van der Waals forces, hydrophobic interactions and the formation of hydrogen bonds between the groove-binding agent and C2 carbonyl oxygen of thymine/ cytosine or N3 nitrogen of adenine/ guanine [8, 12].

Polycyclic, aromatic and planar drugs can intercalate between the base pairs of nucleic acid structure, drawing them apart from their normal 3.4 Å spacing to 6.8 Å while groove-binding drugs can fit into minor or major groove, sometimes with minimal distortion of the nucleic acid helix [6-9, 12].

Ribavirin is a purine nucleoside analogue that is active against a number of DNA and RNA viruses [16]. Ribavirin, in combination with interferon, has predominantly been applied in the treatment of hepatitis C virus infection and its potential antitumor efficacy has recently become a point of interest. There are numbers of proposed mechanisms of action for ribavirin. These include indirect effects such as inhibition of inosine monophosphate and immunomodulatory effects and direct effects such as polymerase inhibition and interference with viral RNA capping. Recent studies use double or triple combinations of ribavirin with other antiviral drugs, such as oseltamivir or/and amantadine in order to increase the activity against multiple virus strains in vitro [17, 18]. Also, ribavirin is used by our research group in vitro chemotherapy to obtain of grapevine virus-free and potato virus-free plants [18].

Our experimental studies for ribavirin–DNA system [19] have pointed out a complex nature of the binding process. Two binding processes were highlighted: a process of the internal binding, that involves the drug intercalation between the base pairs from nucleic acid structure and a process of the external binding, that involve the drug binding to the grooves from the nucleic acid structure. It was found that the external binding prevails, the binding constant of this process being with an order of magnitude greater than the binding constant of the second process. Also, the dependence of the binding constants on the ionic strength of the medium allowed the dissection of the binding free energy in electrostatic and non-electrostatic contributions. It was found that the non-electrostatic contribution prevails.

L.E.Vijan is Associated Professor at the Department of Natural Sciences, Faculty of Science, University of Pitesti, Romania (L.E.Vijan, corresponding author, e-mail loredana.vijan@upit.ro, vloredana2005@yahoo.com)

C.M.Topală is Associated Professor at the Department of Natural Sciences, Faculty of Science, University of Pitesti, Romania (e-mail carmen.topala@gmail.com)

Computer-aided molecular design has become an everyday practice in researches from chemistry, biology, pharmacology, biological systems engineering, and other fields of molecular sciences. It is possible to reproduce experimental information on the hydration energies, the binding energies in different drug – biopolymer systems, the behaviour of water molecules at membrane surfaces, etc., by the molecular simulations. Computational methods can be useful in modeling of the complexes between drugs and nucleic acids to predict the structure and the stereochemistry of recognition.

The present studies represent the second our paper of molecular modeling of interaction of ribavirin with some model nucleic acids in order to determine the structural characteristics of binding of ribavirin to nucleic acid sequences, to estimate the relative contributions to the interaction energy (in van der Waals and electrostatic terms) and to get an insight on the sequence selectivity of the antiviral drug.

II. COMPUTATIONAL DETAILS

The structures of the ribavirin conformers and the sequences of nucleic acids were built within the HyperChem Release version 7.5 program and optimized by the Molecular Mechanics (parameters: MM+ force field, RMS gradient of 0.01 kcal/mol·Å) and the semiempirical AM1 method (parameters: SCF control of 0.01, RHF spin pairing, Polak - Ribiere optimizer, RMS gradient of 0.01 kcal/mol·Å for the ribavirin conformers and the sequences of mono-stranded nucleic acids, RMS gradient of 0.1 kcal/mol·Å for the sequences of double-stranded nucleic acids).

The calculations on the complexes of two ribavirin conformers (denoted V_1 and V_2) with the sequences of mono- and double-stranded nucleic acids were performed in vacuo by the Molecular Mechanics (MM+ force field) method until to RMS gradient of 0.01 kcal/mol·Å. From numerical methods, for optimization we have used the Polak – Ribiere method.

III. RESULTS AND DISCUSSION

A. Conformers of ribavirin

The molecular properties of molecules depend on their three-dimensional structure. It is known that a molecule can adopt due to rotation around σ bonds different spatial arrangements, known as conformations. The study of the energy changes that occur during these rotations defined the conformational analysis. The conformational analysis can be regarded as the analysis of all conformations that a molecule can adopt as a result of single bond rotations, which means searches of all minima for global minimum, local minima or energy barriers.

The conformational search involves the identifying preferred conformations of molecules because the biological activity of molecules is strongly dependent on their conformation. The conformational search done by exploring of the energy surface of a molecule and implying determining the conformations with minimum energy.

Ribavirin has a complex structure (figure 1), comprising a 1,2,4-triazole-3-carboxamide chromophore and a ribose moiety, 2-hydroxymethyl-tetrahydro-furan-3,4-diol. Ribavirin crystallises in two polymorphic forms (denoted V_1 and V_2), which exhibit differences in their IR spectra and melting points, suggesting differences in their hydrogen bonding and crystal packing schemes [20]. X-ray analysis of those forms was undertaken to unequivocally establish the molecular structure and conformation and to elucidate the differences between their properties and their packing patterns [20].

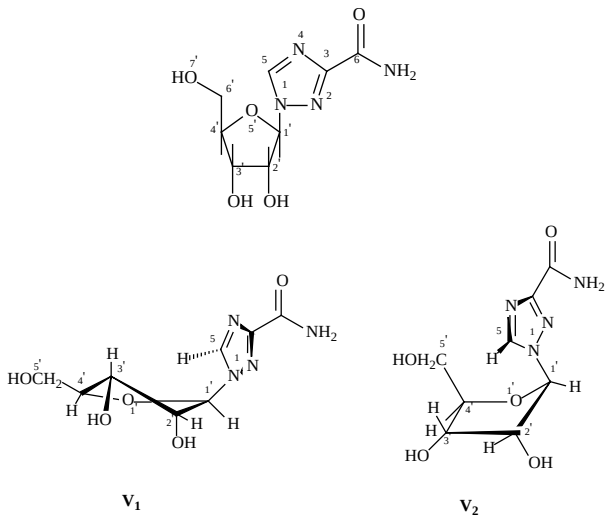


Fig. 1. Chemical structure for ribavirin and its two conformations V_1 ("high *syn*") and V_2 ("high *anti*")

The crystal structures of these forms have presented in literature. Both forms crystallize in orthorhombic system [20], with a=14·863 Å, b=7·512 Å and c=8·788 Å for form V_1 and a=25·034 Å, b=7·719 Å and c=5·289 Å for form V_2, respectively. These two forms were found in different conformations. In V_1, the glycosyl torsion is ,,*normal anti*" (χ=10·4°), the sugar pucker (pseudorotation phase angle P) is $^3T_2$ (P=11·7°), the exocyclic C_4'-C_6' bond torsion is *gauche* and the ribose conformation is 3'-*endo*-2'-*exo* while in V_2, these are ,,*high anti*" (χ=119·0°), $^2T_1$ (P=24·2°), *trans* and the ribose conformation is 2'-*exo*-1'-*endo* [20].

The carboxamide group in V_2 is engaged in hydrogen bonding to the base ring of a symmetry-related molecules whereas there is no interbase hydrogen bonding in V_1 [20]. The usual hydrogen-bonding sites of the base ring in V_1 are involved in hydrogen bonding while the N_2 site of the base in V_2 is not hydrogen bonded [20]. Interestingly, in both forms one of the amino hydrogen atoms is not engaged in hydrogen bonding.

Initial, the molecule of ribavirin was constructed using the HyperChem Release version 7.5 program. In order to find the minimum energy structure we have made the conformational

analysis using Polak – Ribiere method. The conformational searching in torsional space was performed using the multiconformer method [21, 22]. Then, the energy minima for ribavirin were determined by a semi-empirical method AM1 (as implemented in HyperChem Release version 7.5 program). The conformations were confirmed as minima by vibrational analysis.

By conformational analysis, we have obtained a series of conformers of ribavirin presenting the features indicated in literature [20, 23, 24] for the polymorphic forms V_1 and V_2. From all of the conformers obtained, they were selected two optimum conformations, with the minimum energy and corresponding to the crystallographic data presented in literature. The representatives with spheres and the torsion angles for optimized conformers of ribavirin are presented in figures 2 and 3.

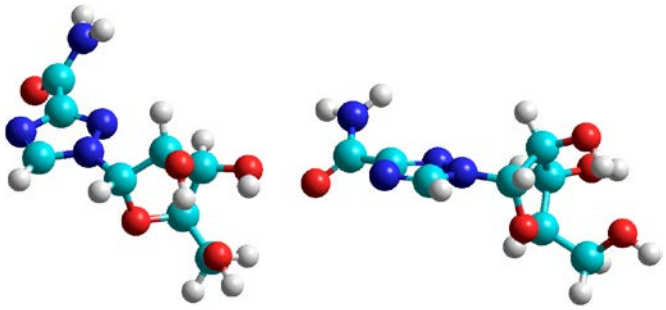


Fig. 2. Ball-and-stick model for V_1 conformer of ribavirin

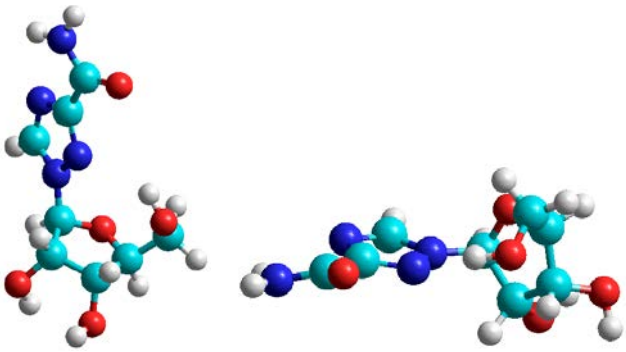


Fig. 3. Ball-and-stick model for V_2 conformer of ribavirin

For both conformers, the charge distribution, the formation energy, the frontier molecular orbitals (HOMO and LUMO), the minimum and maximum values of electrostatic potential were determined [25], the results being presented in table 1.

Table 1. Results of AM1 calculations

Parameter	Ribavirin	
	V_1	V_2
$\Delta H_{formation}$, Kcal/mol	-139.47	-141.25
ε_{homo}, eV	-10.42	-10.47
ε_{lumo}, eV	-0.05	-0.13
V_{min}, Kcal/mol	-41.67	-36.39
V_{max}, Kcal/mol	266.53	246.01

Although the differences between the two conformers of ribavirin are small, it can be said that the V_2 conformer is more stable than the V_1 conformer. The optimized values of the ribavirin conformers energies calculated by the Molecular Mechanics method were used to calculate the drug – nucleic acid interaction energy.

B. Sequences of nucleic acids

Nucleic acids are complex organic molecules that contain the genetic code for the organism [26-28]. The structure, the function and the metabolism of the nitrogenous bases, nucleosides and nucleotides from nucleic acids have attracted attention of researchers in the last years [29, 30]. Nucleic acids can adopt different conformations depending on the nucleotide sequence and other extrinsic factors, such as ionic strength, type of ions, solvents. Deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) are common targets for antiviral, anticancer, and antibiotic drugs and in consequently, the studies of the interaction between drugs and DNA or RNA are of great importance in many areas. It was found that specific atomic sites from DNA or RNA are often the targets for a drug's actions [31-33]. In a broader sense, DNA and RNA can be considered as the macromolecular receptor for drugs.

A remarkable feature of the DNA or RNA molecules is that there are a number of reactive (e.g. nucleophilic) sites uniquely displayed, depending on the sequence, on the surface of the nucleic acid helix. For instance, in the minor groove of deoxyribonucleic acid, the exocyclic N2 amino group of guanine and the N3 atom of both guanine and adenine bases are particularly susceptible to the drugs action. In the major groove, the N7 atom of both guanine and adenine bases is particularly susceptible to drug action [31-33]. Finally, the C4', C5', and C1' atoms of the deoxyribose in the backbone of nucleic acid double-helix are other reactive sites from the nucleic acids sequences [31-33].

Because the literature data pointed out that the intercalating drugs have a sequence selectivity for nucleic acids that does not extend beyond two or three base level [23, 24, 27-29], we have chosen some model mono- and double-stranded nucleic acids containing the AAAAAA, TTTTTT, CCCCCC, GGGGGG, ATATAT, CGCGCG, ATCGAT and CGATCG sequences. Solvent and counterions were not included explicitly for reasons of computational expense. Instead, their effect was simulated using a distance-dependent dielectric constant with $\varepsilon=4r_{ij}$. This formalism was well established in the proteins modeling and has been tested with satisfactory results at nucleic acids [34]. Thus, the nucleic acids sequences were constructed by the charge neutralization of phosphate groups with hydrogen atoms.

The nucleic acids sequences were optimized by the Molecular Mechanics (MM+ force field) and semiempirical AM1 methods and the optimized values of the nucleic acids sequences energies calculated by the Molecular Mechanics method were used to calculate the drug – nucleic acid interaction energy [19, 25].

C. Molecular modeling of ribavirin - nucleic acid complexes

The optimized conformers of ribavirin and the optimized sequences of mono- and double-stranded nucleic acids were utilized in the optimization of the drug - nucleic acid complexes. For the optimization of the drug - nucleic acid complexes by the Molecular Mechanics (MM+ force field) method, the solvent effect was not considered. The starting structures of the drug - nucleic acid complexes were built by the docking procedure. Initially, several restraints were imposed, so that the 1,2,4-triazole-3-carboxamide fragment to be oriented parallel to the bases from the nucleic acid helix, the carboxamidic group to be set outside of the helix toward the major groove and the 2-hydroxymethyl-tetrahydrofuran-3,4-diol moiety to be located in the minor groove of nucleic acids structure. After optimization of the drug - nucleic acid complexes until the required gradient, the restraints were eliminated and the complexes were optimized again.

In figures 4 and 5 are presented the optimized geometries for two complexes of ribavirin V_1, respectively V_2 with the mono-stranded nucleic acids sequences, being presented the nucleic acid structure before and after the interaction with the antiviral agent.

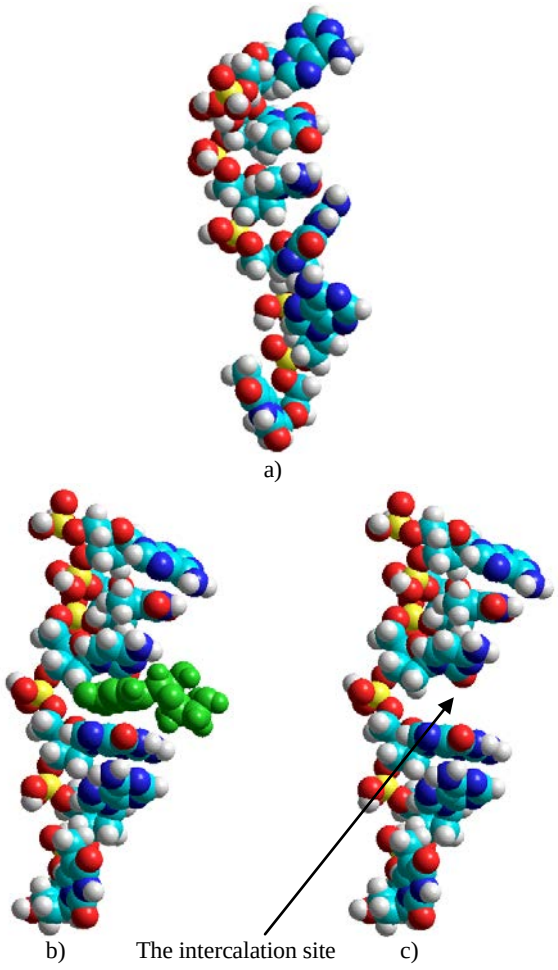


Fig. 4. The optimized geometry of ATCGAT (a).
The optimized geometry of ribavirin V_1-ATCGAT complex (b).
The intercalation site of the ribavirin V_1 to ATCGAT sequence (c).

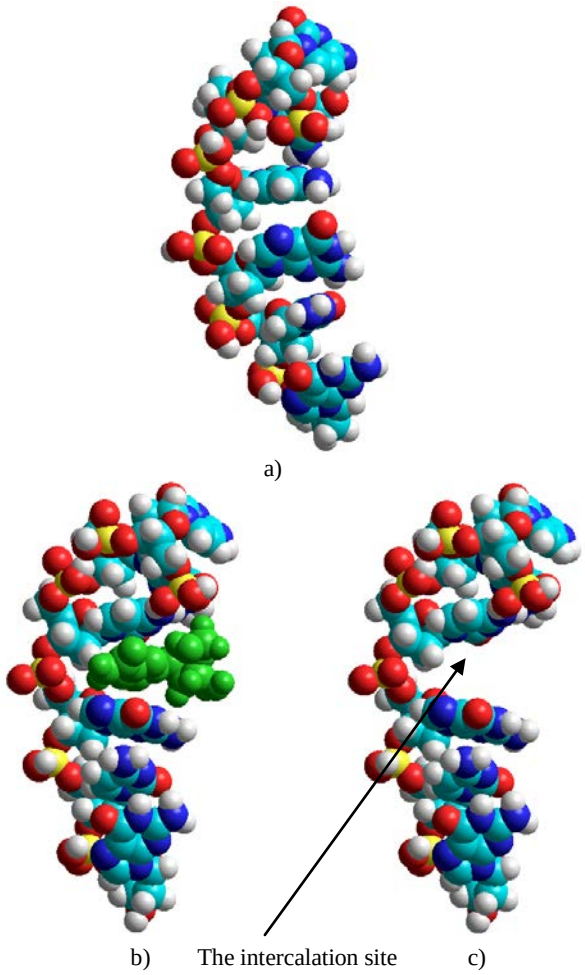
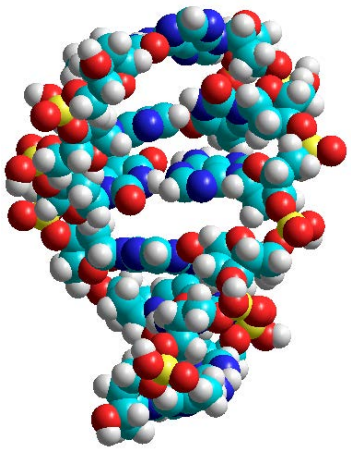
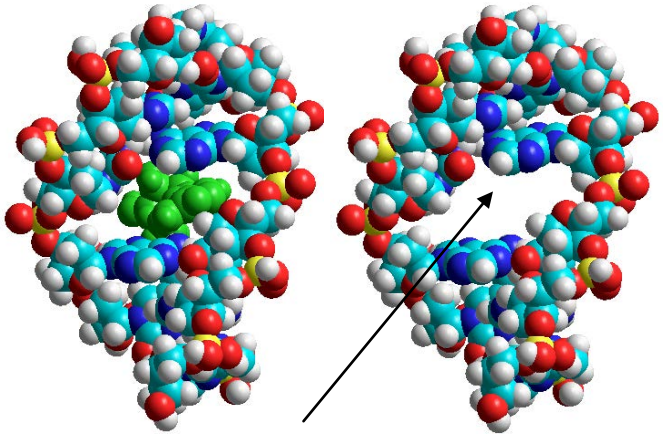


Fig. 5. The optimized geometry of CGCGCG (a).
The optimized geometry of ribavirin V_2-CGCGCG complex (b).
The intercalation site of the ribavirin V_2 to CGCGCG sequence (c).

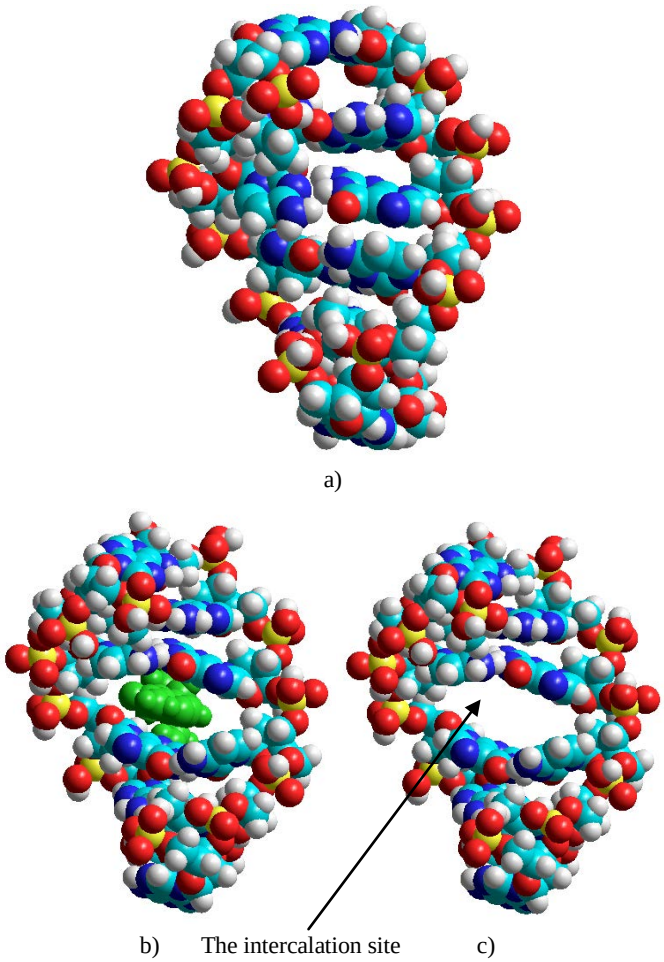
In figures 6 and 7 are presented the optimized geometries for two complexes of ribavirin V_1, respectively V_2 with the double-stranded nucleic acids sequences, being presented the nucleic acid structure before and after the interaction with the antiviral agent.





b) The intercalation site c)

Fig. 6. The optimized geometry of ATATAT-TATATA (a). The optimized geometry of ribavirin V_1-ATATAT-TATATA complex (b). The intercalation site of the ribavirin V_2 to ATATAT-TATATA sequence (c).



a)

b) The intercalation site c)

Fig. 7. The optimized geometry of ATCGAT-TAGCTA (a). The optimized geometry of ribavirin V_2-ATCGAT-TAGCTA complex (b). The intercalation site of the ribavirin V_2 to ATCGAT-TAGCTA sequence (c).

In all drug - nucleic acid complexes, the formation of some intercalation complexes was observed. Initially, the nucleic acid undergoes a conformational change that leads to the obtaining of an intercalation site. In this step, the nitrogenous bases from the nucleic acid structure were separated to constitute the cavity in which the drug will be intercalated. Then, in the second step an external binding of drug at the nucleic acid sequence occurs and in the third step the drug intercalation between the nitrogenous bases from the nucleic acid structure appears.

The energies of the drug - nucleic acid complexes were used for the evaluation of the following quantities [14, 15, 35]:
- the interaction energy:

$$E_{\text{int eraction}} = E_{complex} - (E_{drug} + E_{DNA})_{optimized} \quad (1)$$

- the binding energy:

$$E_{binding} = E_{complex} - (E_{drug} + E_{DNA})_{frozen\ in\ complex} \quad (2)$$

- the perturbation energy:

$$E_{perturbation} = E_{i\,nteraction} - E_{binding} \quad (3)$$

The values of the binding energies of the ribavirin - nucleic acids sequences calculated by the Molecular Mechanics method are presented in table 2. The van der Waals (VdW) contribution to the binding energy are also included in table 2.

Table 2. MM results of drug – nucleic acid interaction

Nucleic acid	Ribavirin V_1		Ribavirin V_2	
	$E_{binding}$, Kcal/mol	% VdW	$E_{binding}$, Kcal/mol	% VdW
AAAAAA	-19,05	86,31	-17,85	69,47
TTTTTT	-21,24	79,81	-24,37	78,05
ATATAT	-23,75	71,98	-20,02	78,12
CCCCCC	-20,26	77,84	-18,77	73,73
GGGGGG	-15,25	93,38	-16,39	88,41
CGCGCG	-18,92	81,61	-17,85	88,63
ATCGAT	-14,24	80,76	-15,12	74,67
CGATCG	-14,23	83,91	-12,95	69,88
AAAAAA-TTTTTT	-27,97	78,48	-26,68	86,58
ATATAT-TATATA	-23,91	82,93	-24,39	83,81
CCCCCC-GGGGGG	-25,14	70,59	-28,27	72,56
CGCGCG-GCGCGC	-30,13	80,95	-27,11	86,09
ATCGAT-TAGCTA	-22,77	96,25	-23,24	89,51
CGATCG-GCTAGC	-20,46	79,62	-22,74	75,45

In all cases, the binding energies have negative values reflecting the drug - nucleic acid interaction. The results underline the significant van der Waals contribution (>70%) to the binding energy and, consequently, the low percentage of the electrostatic interactions, in agreement with our previous experimental data. A slight preference for the sequences containing adenine and/or thymine bases can be noticed for both ribavirin conformers with nucleic acids sequences.

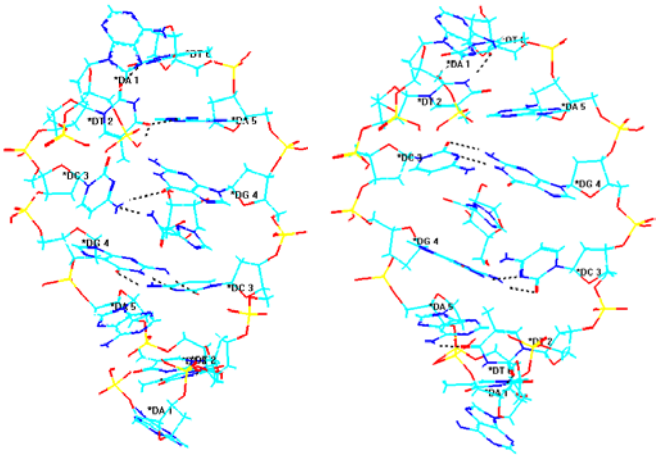
In table 3 are presented the values for the interaction and perturbation energies, characteristic to the inclusion processes of the two ribavirin conformers in the nucleic acids structures. It is noted that the intercalation of ribavirin conformers in the nucleic acids helix causes a small disturbance in the drug structure and a big disturbance in the structure of nucleic acids.

Table 3. MM results of drug – nucleic acid interaction

Ribavirin – nucleic acid complexes		$E_{interaction}$, kcal/mol	$E_{perturbation}$, kcal/mol	
			total	nucleic acid
RIBAVIRIN V_1	AAAAAA	-3,96	15,09	14,74
	TTTTTT	-9,67	11,57	10,76
	ATATAT	-16,19	7,56	6,79
	CCCCCC	-13,61	6,63	6,32
	GGGGGG	-2,71	12,54	12,12
	CGCGCG	-7,79	11,13	10,84
	ATCGAT	-12,76	1,48	0,64
	CGATCG	-13,41	0,82	0,73
	AAAAAA-TTTTTT	-25,75	2,22	1,06
	ATATAT-TATATA	-22,54	1,37	0,22
	CCCCCC-GGGGGG	-20,32	4,82	3,59
	CGCGCG-GCGCGC	-11,87	18,26	17,52
	ATCGAT-TAGCTA	-11,81	10,96	10,21
	CGATCG-GCTAGC	-15,99	4,47	2,95
RIBAVIRIN V_2	AAAAAA	-3,78	14,07	13,59
	TTTTTT	-15,88	8,49	7,81
	ATATAT	-11,31	8,71	8,11
	CCCCCC	-5,9	12,87	9,73
	GGGGGG	-2,7	13,69	12,84
	CGCGCG	-9,96	7,89	7,33
	ATCGAT	-9,67	5,45	4,61
	CGATCG	-12,04	0,91	0,44
	AAAAAA-TTTTTT	-24,39	2,29	0,55
	ATATAT-TATATA	-25,55	2,22	1,06
	CCCCCC-GGGGGG	-16,05	12,22	10,91
	CGCGCG-GCGCGC	-23,13	3,98	3,3
	ATCGAT-TAGCTA	-13,48	9,76	8,83
	CGATCG-GCTAGC	-11,38	11,36	10,34

A lot of features of the dyes and drugs intercalation [14, 15, 36-40] between the purine and pyrimidine bases from the nucleic acids structure were found, as follows:
i) the distortion in nucleic acid structure by the angle opening of the phosphate groups for allowing the drug intercalation,
ii) the lengthening of the helix by approx. 3.4 Å which causes a conformational change of some sugar moieties involved,
iii) the increase in the distance between nitrogenous bases at the intercalation site level.

Although some features of the intercalation are found in the ribavirin - nucleic acid system, however there is a remarkable difference, determined by the "accordion type" motion (specified in the model Lerman [1, 2]) that occurs with the breaking of some hydrogen bonds between the purine and pyrimidine bases from the nucleic acids structure. In addition, the formation of new hydrogen bonds between ribavirin and the nitrogenous bases from the nucleic acids sequences at the intercalation site level (figure 8) was observed.



a) b)

Fig. 8. Hydrogen bonds in ribavirin V_1 (a), respectively ribavirin V_2 (b) - ATCGAT-TAGCTA complexes

IV. CONCLUSIONS

The results of the molecular modeling points out that the complexes of ribavirin with nucleic acids are stabilized mainly by van der Waals forces involving the 1,2,4-triazole-3-carboxamide moiety and the bases from the nucleic acids structure and that the electrostatic term brings a minimal contribution (<20%) to the binding energy. For both ribavirin conformers, a slight preference for nucleic acids sequences containing adenine and/or thymine bases was found. As a result of the ribavirin – nucleic acid interaction, only the nucleic acids structure is significantly perturbed, the structure of the drug being practically unchanged.

In all ribavirin – nucleic acids complexes, an increase of the distance between the adjacent bases at the intercalation site level was observed. The turning of the polynucleotidic helix was produced and the "accordion type" motion determined a breaking in the hydrogen bonds between base pairs from double-stranded nucleic acid helix.

ACKNOWLEDGMENT

This work was supported by a grant of the Romanian National Authority for Scientific Research, CNDI-UEFISCDI, project number 104/2012 (PN-II-PT-PCCA-2011-3.1

REFERENCES

[1] L.S. Lerman, "Structural considerations in the interaction of DNA and acridines", J. Mol. Biol., vol. 3, pp. 18-30, 1961.
[2] L.S. Lerman, "The structure of the DNA-acridine complex", Proc. Natl. Acad. Sci. U.S.A., vol. 49 (1), pp. 94-102, 1963.
[3] S. Satyanarayana, J.C. Dabrowiak, and J.B. Chaires, "Tris(phenanthroline)ruthenium(II) enantiomer interactions with DNA: Mode and specificity of binding", Biochemistry, vol. 32 (10), pp. 2573-2584, 1993.
[4] D. Reha, M. Kabelac, F. Ryjacek, J. Sponer, J.E. Sponer, M. Elstner, S. Suhai, and P. Hobza, "Intercalators. 1. Nature of stacking interactions between intercalators (ethidium, daunomycin, ellipticine, and 4/,6-diamidine-2-phenylindole) and DNA base pairs. Ab initio quantum chemical, density functional theory, and empirical potential study", J. Am. Chem. Soc., vol. 124, pp. 3366-3376, 2002.

[5] D.E. Graves, and L.M. Velea, "Intercalative binding of small molecules to nucleic acids", Curr. Org. Chem., vol. 4, pp. 915-929, 2000.
[6] H.M. Berman, and P.R. Young, "The Interaction of Intercalating Drugs with Nucleic Acids", Annu. Rev. Biophys. Bioeng., vol. 10, pp. 87-114, 1981.
[7] W.D. Wilson, and R.L. Jones, "Intercalating drugs: DNA binding and molecular pharmacology", Adv. Pharmacol. Chemother., vol. 18, pp. 177-222, 1981.
[8] B.S.P. Reddy, S.K. Sharma, and J.W. Lown. "Recent developments in sequence selective minor groove DNA effectors", Curr. Med. Chem., vol. 8, pp. 475-508, 2001.
[9] R. Eckel, R. Ros, A. Ros, S.D. Wilking, N. Sewald, and D. Anselmetti. "Identification of binding mechanisms in single molecule-DNA complexes", Biophys. J., vol. 85, pp. 1968-1973, 2003.
[10] A. Larsson, C. Carlsson, M. Jonsson, and B. Albinsson. "Characterization of the binding of the fluorescent dyes YO and YOYO to DNA by polarized light spectroscopy", J. Am. Chem. Soc., vol. 116, pp. 8459-8465, 1994.
[11] M.J. Waring, "DNA Modification and Cancer", Ann. Rev. Biochem., vol. 50, pp. 159-192, 1981.
[12] D. Suh, and J.B. Chaires, "Criteria for the mode of binding of DNA binding agents", Bioorg. Med. Chem., vol. 3(6), pp. 723-728, 1995.
[13] S. Neidle, and T.C. Jenkins, "Molecular modeling to study DNA interaction by anti-tumor drugs", Methods in enzymology, vol. 203, pp. 433-458, 1991.
[14] L. E. Vîjan, E. Volanschi, and M. Hillebrand, "Molecular modeling of anthracycline-DNA interaction", Progress in Colloid and Polymer Science, vol. 122, pp. 67-72, 2003.
[15] L.E. Vîjan, "The theoretical study of the interaction of some anthracycline drugs with nucleic acids", Analele Universităţii din Bucureşti, vol. I-II, pp. 377-382, 2005.
[16] E. De Clercq, "Toward Improved Anti-HIV Chemotherapy: Therapeutic Strategies for Intervention with HIV Infections", Adv. Virus Res., vol. 42, pp.1–55, 1993.
[17] J.T. Nguyen, J.D. Hoopes, D.F. Smee, M.N. Prichard, E.M. Driebe, D.M. Engelthaler, M.H. Le, P.S. Keim, R.P. Spence, and G.T. Went, "Triple combination of oseltamivir, amantadine, and ribavirin displays synergistic activity against multiple influenza virus strains in vitro", vol. 53, pp. 4115–4126, 2009, doi:10.1128/AAC.00476-09
[18] I.C. Guţă, E.C. Buciumeanu, E.M. Vişoiu, and C.M. Topală, "Modifications of Some Biochemical Compounds of Virus Infected Grapevine *in vitro* Cultured on Media with Chemotherapeutic Agents", Rev. Chim. (Bucharest), vol. 65(8), pp. 884-887, 2014.
[19] L.E. Vîjan, and C.M. Topală, "Study of ribavirin - nucleic acids interaction", Chemistry and biology, to be published
[20] P. Prusiner, and M. Sundaralingam, "The Crystal and Molecular Structures of Two Polymorphic Crystalline Forms of Virazole (1-β-D-Ribofuranosyl-l,2,4-triazole-3-carboxamide). A New Synthetic Broad Spectrum Antiviral Agent", Acta Cryst, B32, pp. 419-426, 1976.
[21] M. Lipton, and W.C. Still, "The multiple minimum problem in molecular modeling. Tree searching internal coordinate conformational space", J. Comput. Chem., vol. 9 (4), pp. 345-355, 1988.
[22] D. Bartulewicz, K. Bielawski, A. Bielawska, and A. Rozanski, "Synthesis, molecular modelling, and antiproliferative and cytotoxic effects of carbocyclic derivates of distamycin with chlorambucil moiety", Eur. J. Med. Chem., vol. 36, pp. 461-467, 2001.
[23] J. Karle, and I.L. Karle, "The symbolic addition procedure for phase determination for centrosymmetric and non-centrosymmetric crystals", Acta Cryst., vol. 21, pp. 849-859, 1966.
[24] D.T. Cromer, and J.T. Waber, "Scattering factors computed from relativistic Dirac–Slater wave functions", Acta Cryst., vol. 18, pp. 104-109, 1965.
[25] L.E. Vîjan, and C.M. Topală, "Molecular modeling of interaction between ribavirin and nucleic acids", proceedings of the 2nd WSEAS International Conference on MATHEMATICS and COMPUTERS in SCIENCES and INDUSTRY, Sliema, Malta, August 17-19, 2015.
[26] Y. Wang, Q. Zhang, W. Wang, and D. Huang, "A continuum model for condensation of DNA in solution", WSEAS TRANSATIONS on MATHEMATICS, vol. 13, pp. 704-713, 2014.
[27] Y. Yamada, and K. Satou, "Prediction of genomic methylation status on CpG islands using DNA sequence featurea", WSEAS TRANSACTIONS on BIOLOGY and BIOMEDICINE, vol. 5(7), pp. 153-162, 2008.
[28] J. Savkovic-Stevanovic, "Genetic information in biopolymeric chain", proceedings of the 2nd WSEAS International Conference BIOMEDICAL ELECTRONICS and BIOMEDICAL INFORMATICS, ID 618-337, Moscow, Russia, August 20-22, 2009, pp. 122-126.
[29] J.B. Savkovic-Stevanovic, "Nucleotides recombination states", WSEAS TRANSACTIONS on BIOLOGY and BIOMEDICINE, vol. 7(3), pp. 253-262, 2010.
[30] J. Savkovic-Stevanovic, "Human tissue information processing", proceedings of the 2nd WSEAS International Conference BIOMEDICAL ELECTRONICS and BIOMEDICAL INFORMATICS, ID 618-272, Moscow, Russia, August 20-22, 2009, pp. 64-69.
[31] X.L. Yang, and A.H. Wang, "Structural studies of atom-specific anticancer drugs acting on DNA", Pharmacol. Ther., vol. 83, pp. 181-215, 1999.
[32] F. Gago, "Stacking interactions and intercalative DNA binding", Methods, vol. 14, pp. 277-292, 1998.
[33] Y.M. Chang, C.M. Chen, and M.H. Hou, "Conformational changes in DNA upon ligand binding monitored by circular dichroism", Int. J. Mol. Sci., vol. 13, pp. 3394-3413, 2012.
[34] M. Orozco, C.A. Laughton, P. Herzyk, and S. Neidle, "Molecular-mechanics modelling of drug-DNA structures; the effects of differing dielectric treatment on helix parameters and comparison with a fully solvated structural model", J. Biomol. Struct. Dyn., vol. 8 (2), pp. 359-373, 1990.
[35] L. Mammino, and M.M. Kabanda, "Ab initio study of nodifloridin-A and nodifloridin-B in vacuo and in solution", WSEAS TRANSACTIONS on BIOLOGY and BIOMEDICINE, vol. 6(4), pp. 79-88, 2009.
[36] J. Ren, and J.B. Chaires, "Sequence and structural selectivity of nucleic acid binding ligands", Biochemistry, vol.38, pp. 16067-16075, 1999.
[37] W.D. Wilson, S. Mizan, F.A. Tanious, S. Yao, and G. Zon, "The interaction of intercalators and groove-binding agents with DNA triple-helical structures: the influence of ligand structure, DNA backbone modifications and sequence", J. Mol. Recognit., vol. 7, pp. 89-98, 1994.
[38] K. Sandström, S. Wärmländer, J. Bergman, R. Engqvist, M. Leijon, and A. Gräslund, "The interaction of intercalators and groove-binding agents with DNA triple-helical structures: the influence of ligand structure, DNA backbone modifications and sequence", J. Mol. Recognit., vol. 14, pp. 277-285, 2004.
[39] M. Cervero, and F. Mendicuti, "Inclusion complexes of dimethyl 2,6-naphthalenedicarboxylate with alpha- and beta-cyclodextrins in aqueous medium: Thermodynamics and molecular mechanics studies", J. Phys. Chem. B, vol. 104, pp. 1572-1580, 2000.
[40] B.A. Neto, and A.A. Lapis, "Recent developments in the chemistry of deoxyribonucleic acid (DNA) intercalators: principles, design, synthesis, applications and trends", Molecules, vol. 14, pp. 1725-1746, 2009.