Baku State University: Journal of Physics & Space Sciences, **2024**, v 1 (2), p. 70-77

journal homepage: http://bsuj.bsu.edu.az/en

Spatial structure of the heptapeptide molecule

Larisa I. Ismailova*, Namik A. Akhmedov

Baku State University, Institute for Physical Problems, Baku, Azerbaijan
Received 26-Apr-2024; Accepted 20-May-2024

Abstract

Studying the three-dimensional structure of peptide molecules is important when creating new drugs. Peptides are the human body's own resource. Understand the mechanism of action of these molecules can be, if you solve the problem of their structural and functional activity. Nociceptins are a new type of regulatory peptides of medical interest. Natural nociceptin reduces motor activity, causes a stress response and modulates spatial attention. This work is devoted to determining the spatial structure and conformational possibilities of the heptapeptide molecule with the amino acid sequence H-Phe1-Gly2-Gly3-Phe4-Pro5-Gly6-Pro7-OH. It is one of the recently synthesized analogues of nociceptin. It was found that the N-terminal tripeptide and tetrapeptide of this molecule are active and very important. Using the method of molecular mechanics, the spatial structure and conformational properties of the heptapeptide molecule were determined. The potential energy of this molecule was estimated as the sum of non-valent, electrostatic, torsion interactions and the energy of hydrogen bonds. 11 low-energy conformations were found for heptapeptide, the values of the dihedral angles of the main and side chains, and the energy of intra- and inter-residue interactions was estimated. It is revealed that low energy conformations of this molecule have the half-folded and folded type of backbone. The side chains of the Phe1 and Phe4 amino acids in low-energy conformations carry out effective interactions and are conformationally labile amino acids, they bring together the regions of the main chain and the side chains of the amino acids included in the heptapeptide. These folded forms bring parts of the backbone and the side chains of the amino acids together, and they result in convenient interactions.

Keywords: nociceptin, structure, molecule, heptapeptide, conformation
PACS: 87.15 Aa; 11.55.-m, 82.80.-d

Corresponding author. Tel.: +994-55-290-22-99
E-mail address: lara.ismailova.52@mail.ru; *ORCID ID*: 0000-0002-2563-0995.

1. Introduction

Currently, new families of the regulatory peptides are being discovered and their properties are being studied. One of these families is nociceptins. Nociceptins are a new type of regulatory peptides of medical interest. For medicine and pharmacology, knowledge of the structure-functional properties of peptide molecules is of great practical importance. The mechanisms of action of the nociceptins are considered anti-opioid. Natural nociceptin reduces motor activity, causes a stress response and modulates spatial attention.

The peptide molecules and thear biological functions in living systems are related with their specific spatial structures. Therefore, to understand the mechanism by which the peptides function it is necessary to know their three-dimensional structures. This scientific work is devoted to study the spatial structure and conformational possibilities of the heptapeptide molecule H-Phe1-Gly2-Gly3-Phe4-Pro5-Gly6-Pro7-OH. This neuropeptide molecule is stable analogue of the nociceptin. It was found that the N-terminal tripeptide and tetrapeptide of this molecule are active [1, 2].

It is known that short linear peptides in solutions do not have a fixed spatial structure. The amino acid sequence and physicochemical properties of the solvent determine the set of low-energy conformations of the peptide molecule. The biologically active conformation of the peptide molecule, which is realized upon interaction with the receptor, is included in the set of low-energy structures that exist in an aqueous solution. Therefore, the study of the spatial structure and conformational capabilities of peptide molecules is of great interest. The peptide molecules and their biological functions in living systems are related with their specific spatial structures. Therefore, to understand the mechanism by which the peptides function it is necessary to know their three-dimensional structures. It is important to know the full complement of low-energy conformational states.

The calculations were carried out by the method of theoretical conformational analysis with regard to nonvalent, electrostatic and torsional interactions, energy of the hydrogen bonds and a special computer program [3]. The low-energy conformations of this molecule and the values of the dihedral angles of the main chain and side chains are found and the energy of the intra- and inter-residue interactions was estimated. The present paper is an extension of our previous investigations of structural and functional organization of peptide molecules [4-10].

2. Section

Neuropeptides play an important role in all nervous systems and structure-functional studies of these peptides is one approach to understanding this role. The heptapeptide molecule H-Phe1-Gly2-Gly3-Phe4-Pro5-Gly6-Pro7-OH has three

amino acids Gly, which has no side chain, two amino acids Phe with have large and labile side chains and two amino acids Pro with a rigid side chain. The potential energy of this molecule was chosen as the sum of the nonvalent, electrostatic and torsional interaction energies and the energy of hydrogen bonds.

In presenting the results of the calculation of the spatial structure of the molecules we used the classification suggested in the work [11]. All structural versions according to it break down into shapes including certain forms of the main chain, each form is represented by a set of conformations. The conformations are determined by the number of rotational degrees of freedom of the side chains of the residues being included in the molecule. The conformational state of each amino residue is conveniently described by the backbone ϕ, ψ, ω and side chain χ_1, χ_2… dihedral angles. The terms "conformation" used in the following analysis will always imply exact quantitative characteristics of residue or fragment geometry. For a stable conformation, the ϕ and ψ dihedral angles are located in low-energy region R, B, L and P of the conformational map. We introduce the notion "form of a residue" to denote the region of its backbone dihedral angle. The conformation of the backbone forms of residue in a given amino acid sequence will specify the backbone form of a fragment. Forms belonging to a particular shape have an analogous peptide chain contour and a similar mutual arrangement of backbones and side chains. Designation's indications of dihedral angles have been measured up to the generally accepted nomenclature [12].

3. Discussion and conclusions

The conformational possibilities of the heptapeptide H-Phe1-Gly2-Gly3-Phe4-Pro5-Gly6-Pro7-OH were studied in fragments. First, the conformational properties of the tripeptides Phe1-Gly2-Gly3 and Pro5-Gly5-Pro5, tetrapeptide Phe1-Gly2-Gly3-Phe4 were determined based on the stable conformations of the monopeptides. And finally, the addition of the tetrapeptide Phe1-Gly2-Gly3-Phe4 and tripeptide Pro5-Gly6-Pro6 allowed us to calculate the spatial structure of the heptapeptide molecule. It is known that the active site of the molecule that activates the receptor is the N-terminal tetrapeptide Phe1-Gly2-Gly3-Phe4. We carried out all of these structures by minimization over all the dihedral angles.

The conformational capabilities of the tripeptide Phe1-Gly2-Gly3 were studied based on the stable conformations of the monopeptides N-acetyl-L-phenylalanine and N-acetyl-L-glycine. For a given tripeptide containing 37 atoms and 10 variable dihedral angles, 4 shapes are possible (ee, ef, fe and ff), represented by 16 forms of the main chain. In total, about 100 conformations were calculated, all of them were minimized in energy, and their geometric and energy parameters were estimated. The calculation revealed the presence of a sharp energy differentiation in the forms of the main chain and shapes.

Just as in the experimental work [2], these conformations can represent four structures. The $B_{21}BL$ conformation has the lowest energy (U_{total} = -2.6 kcal/mol), which belongs to the ef shape. This semi-folded shape is represented by the largest number of low-energy conformations – 12. In the global conformation of $B_{21}BL$ (ΔU_{rel}=0 kcal/mol), the energy of nonvalent interactions is -6.4 kcal/mol, electrostatic 2.3 kcal/mol and torsional 1.5 kcal/mol. The fully unfolded form of the main chain $B_{21}BR$ (U_{total} = -2.0 kcal/mol) of the ee shape is inferior to it by only 0.6 kcal/mol. The conformations of the completely folded shape ff $B_{21}PR$ (U_{total}= -0.6 kcal/mol) are low-energy. More than 3 kcal/mol is inferior to the conformation of another half-folded shape fe $B_{11}PL$ (U_{total} = 0.4 kcal/mol). It should be noted that the main contribution to the energy of low-energy conformations is made by dipeptide and tripeptide interactions. The main energy contribution comes from nonvalence interactions. This contribution varies in the range from -1.1 to -7.0 kcal/mol. All 24 low-energy conformations of the tripeptide molecule were taken into account when calculating the structure of the tetrapeptide Phe1-Gly2-Gly3-Phe4.

The amino acid sequence of the tetrapeptide molecule included two amino acids each phenylalanine and glycine. Tetrapeptide molecule Phe1-Gly2-Gly3-Phe4 contains 57 atoms and 15 variable dihedral angles. The specificity of the amino acid side chains of the tetrapeptide molecule determined the number of initial approximations. In total, over 150 conformations were calculated, belonging to 64 forms of the main chain and 8 possible shapes for this molecule. All of them were minimized in energy, and their geometric and energy parameters were estimated. The calculation showed the presence of a sharp energy differentiation of conformations. Representatives of 22 forms of the main chain fall into the energy range 0 – 4 kcal/mol.

The low-energy conformations of the tetrapeptide Phe1-Gly2-Gly3-Phe4 and the C-terminal tripeptide Pro5-Gly6-Pro7 were taken into account when calculating the spatial structure of the entire heptapeptide molecule Phe1-Gly2-Gly3-Phe4-Pro5-Gly6-Pro7. In total, over 300 initial approximations were compled, which were then minimized in energy. Heptapeptide molecule contains 92 atoms and 22 variable dihedral angles. It is known that for any amino acid residue (with the exception Gly) present in front of Pro having all conformations with R-form of main chain are high-energy therefore such state for Phe4 is excluded from the calculation. The relative energy of the conformations of the heptapeptide molecule varied within the range 0 – 10 kcal/mol. There are the energy differentiation both in respect of the conformations, and forms of the main chain and shapes.

The low-energy conformations of the heptapeptide molecule are presented in Table 1. The global conformation of this molecule (U_{rel}=0 kcal/mol) is $B_{11}LPB_{11}RBR$. The contribution of the stabilizing nonvalent to this conformation is (–24.4) kcal/mol, where as electrostatic interactions account for (-4.4 kcal/mol) and torsion for 3.0 kcal/mol. This conformation is efficient both in nonvalent and in electrostatic interactions forming hydrogen bonds between atoms of main chain which

make contribution (-2.1 kcal/mol) in total energy. The main contributions of the interresidual interactions in this conformation were dipeptide contributions (–4.6) kcal/mol, tripeptide (–3.0) kcal/mol, tetrapeptide (–4.2) kcal/mol, pentapeptide (–8.7) kcal/mol, hexapeptide (-2.1) kcal/mol and heptapeptide (-6.9) kcal/mol.

Table 1. The energy parameters: relative energy (U_{rel}) and energy contributions of nonvalent (U_{nv}), electrostatic (U_{el}), torsion (U_{tors}) interactions of optimal conformations of the heptapeptide

№	Shapes	Conformation	U_{rel}	Energy range, kcal/mol		
				U_{nv}	U_{el}	U_{tors}
1	effeee	B_{11} R R $B_{1\,1}$R L R	5.5	-23.8	0.8	2.8
2	effeef	B_{11} R R B_{11} R P R	4.8	-23.7	0.6	2.2
3	feeefe	B_{11} P L B_{11} B L R	3.5	-23.0	-2.6	3.4
4	fffefe	B_{11} P R B_{11} R B R	1.3	-22.4	-5.1	3.1
5		B_{11} L P B_{11} R B R	0.0	-24.4	-4.4	3.0
6		B_{21} L P B_{11} R B R	3.4	-21.7	-3.8	3.3
7		B_{31} L P B_{11} R B R	4.4	-20.2	-4.1	3.0
8		B_{11} L P B_{21} R B R	2.5	-22.4	-4.4	3.5
9		B_{21} L P B_{21} R B R	3.4	-20.7	-5.0	3.4
10		B_{21} L P B_{31} R B R	3.2	-20.8	-4.8	3.5
11	fffeff	B_{11} L P B_{21} R R R	3.3	-23.4	-4/1	5.1

In this conformation, amino acid residues Phe1-Gly2-Gly3-Phe4-Pro5-Gly6-Pro7 form folded structure. The geometric parameters of the three low-energy conformations of the heptapeptide molecule are presented in Table 2. It is revealed that

Table 2. Geometric parameters (degree) of the optimal conformations of Phe-Gly-Gly-Phe-Pro-Gly-Pro heptapeptide molecule

Residues	Shape (conformation)		
	fffefe (B_{11}LPB_1RBR)	**feeefe (B_{11}PLB_{11}BLR)**	**fffeff (B_{11}LPB_{21}RRR)**
Phe 1	-178 159 179 64 89	-179 164 174 64 87	-174 158 -177 66 82
Gly2	76 87 -179	82 -59 -176	86 81 -176
Gly3	94 -51 -175	118 64 180	82 -76 178
Phe4	-134 150 -171 63 100	-159 151 -177 60 151	-92 135 -168 204 75
Pro5	-60 -46 179	-60 134 -170	-60 -41 -175
Gly6	-75 180 -175	73 71 175	-55 -75 -177
Pro7	-60 -54	-60 -44	-60 -54
U_{rel} (kcal/mol)	0	3.5	3.3

Note: The values of dihedral angles are given in the sequence ϕ, ψ, ω, χ^1, χ^2,...

low energy conformations of this molecule have the folded and half folded types of backbone. These folded forms bring parts of the backbone and the side chains of the amino acids together, and they result in convenient interactions.

The results can be used to study the spatial structure of heptapeptide molecule as well as to study the conformational capabilities of side chains of the Phe1 and Phe4 when interacting with receptor molecules. The side chains of these residues have conformational freedom in the low-energy structures of the heptapeptide molecule. Thus, the theoretical conformational analysis of this peptide molecule led to such structural organizations of molecules that do not exclude the realization by the molecule of a number of various functions that require strictly specific interactions with various receptors.

Figures 1(a, b, c) represent schematically the backbone forms and positions of residues in low-energy conformations $B_{11}LPB_{11}RBR$, $B_{11}PLB_{11}BLR$ and $B_{11}LPB_{21}RRR$ of the heptapeptide molecule. The figures show that this molecule has folded N-terminal fragment of the molecule. Conformational possibilities of side chains of Phe1 and Phe4 in the best low energy conformations of peptide molecule have been investigated by plotting conformational maps. The conformational maps show that side chains of this residues have conformational freedom.

We have studied in detail the spatial structure and conformational properties of heptapeptide molecule Phe1-Gly2-Gly3-Phe4-Pro5-Gly6-Pro7. The relative energy of the conformations of the heptapeptide molecule varied within the range 0– 10 kcal/mol. Study of the spatial structure of this molecule have almost 11 low-energy

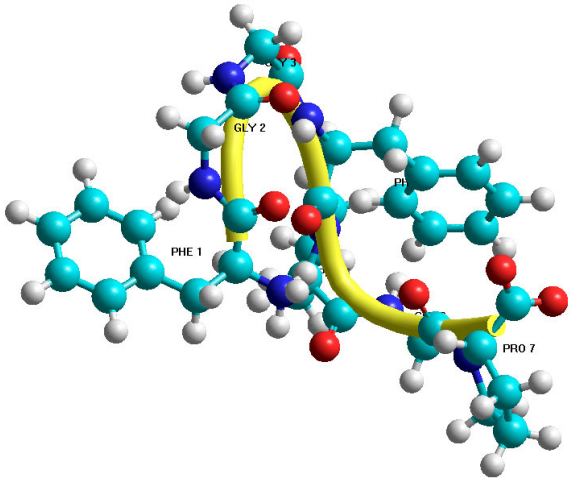

Fig. 1a. Spatial structure of the low energy conformation $B_{11}LPB_{11}RBR$ of the heptapeptide molecule

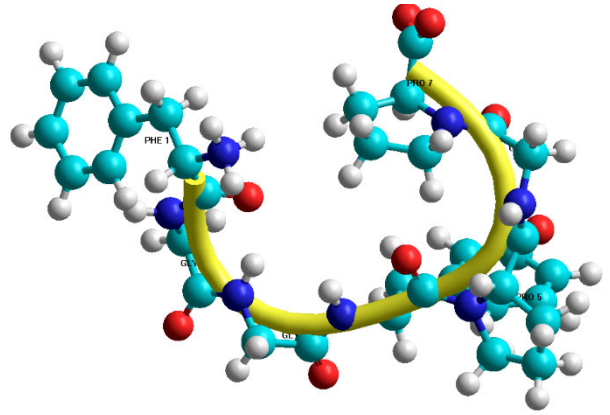

Fig. 1b. Spatial structure of the low energy conformation $B_{11}PLB_{11}BLR$ of the heptapeptide molecule

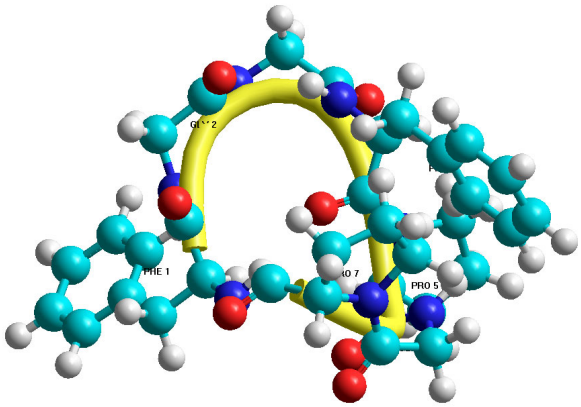

Fig. 1c. Spatial structure of the low energy conformation $B_{11}LPB_{21}RRR$ of the heptapeptide molecule

conformations. It can be assumed that the low-energy folded form of the main chain of the molecule provides intramolecular interactions. Amino acid Phe1 and Phe4 lack the ability to interact with the receptor. Therefore, the lack of activity of the heptapeptide molecule can be associated with the folded form of the molecule and the lack of conformational freedom of amino acid residues.

References

[1] Shamakina I.Yu., Shagiakhmetov F., Anokhin P.K., Kohan V.S., Davidiva T.V. The role of nociceptin in opioid regulation of brain functions. *Biomeditsinskaya Khimiya,* 2021, vol. 67, N 1, pp. 5-16.

[2] Ivanova E.A., Sarycheva N.Yu., Dubynin V.A., Malyshev A.V. et.al. Behavioral effects of original tetrapeptide an analog of N-terminal nociceptin fragment. *Bulletin of Experimental Biology and Medicine* 2012, vol. 13, N 2, pp. 851-857.

[3] Maksumov I. S., Ismailova L. I., Godjaev N. M. A computer program for calculation of conformations of molecular systems, *J. Struc. Chem*. 1983, vol. 24, pp. 147-148.

[4] Ismailova] L.I., Akverdieva G.A., Demukhamedova S.D., Akhmedov N.A. Molecular Modelling of Pro-Gly gliproline and its complexes. *Moscow University Physics Bulletin.* 2023, vol. 78, N 5, pp. 668-680.

[5] Akhmedov N.A., Gadjiyeva Sh.N., Abbasli R.M. Structural organization of Asp-Pro- Lys-Gln- Asp-Phe-Met-Arg-Phe-NH_2 molecule*, Cur.Top. in Pept.&Prot.Res.* 2009, vol.10, pp. 57-62.

[6] Akhmedov N.A., Ismailova L.I., Agayeva L.N., Gocayev N.M. The spatial structure of the cardio active peptides, *Cur.Top. in Pept.&Prot. Res.,* 2010, vol.11, pp. 87-93.

[7] Akhmedov N.A., Ismailova L.I., Abbasli R.M., Agayeva L.N., Akhmedova S.R. Spatial structure of Octarphin molecule*, IOSR JAP*, e-ISSN:2278-4861. 2016, vol. 8, pp. 66-70.

[8] Akhmedov N., Agayeva L., Akverdieva G., Abbasli R., Ismailova L. Spatial structure of the ACTH-(6-9)-PGP molecule*. J.Chem.Soc.Pak.,* 2021, vol. 43. N 5, pp. 500-504.

[9] Akhmedov N., Agayeva L., Akhmedova S., Abbasli R., Ismailova L. Spatial structure of the β-Casomorphin-7 Molecule. *IOSR Journal of Applied Physics (IOSR-JAP)*, 2021, vol 13, Issue 5, pp.62-67.

[10] Ismailova L.I., Abbasli R.M., Akhmedov N.A. Computer modeling of the spatial atructure of nonapeptide molecule. Proceedings of the 7-th International Conference on Control and Optimization with Industrial Applications, 2020, vol. I, pp. 218-220.

[11] Popov E.M. An Approach to calculations of the problem of structure functional organization of natural peptides*, Mol. Biol.,* 1985, vol. 19, pp. 1107-1138.

[12] IUPAC-IUB, Quantity, Units and Symbols in Physical Chemistry, 1988, vol. 39, Blackwell Scientific Publications, Oxford.