ORIGINAL PAPER

COMPARATIVE STUDY OF ANTIBACTERIAL AND ANTIFUNGAL ACTIVITIES OF ACETAMIDIC DERIVATIVES

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Abstract. *The antimicrobial activity of the amide derivatives depends on the stability of the complex formed by their interactions with the biological receptor. The interaction between the ligand (amide derivative) and the biological receptor is visualized in the 2D diagrams of the molecular docking method used. In hydrogen-binding interactions are involved the amino acids Gly133, Gly86, Gly156, Gin154, Thr155 from the active sites of Gram-negative Escherichia coli bacteria (PDB ID: 3t88) for the most active compounds. In the case of Gram-positive Staphylococcus aureus bacteria (PDB ID: 3Q8U), the hydrogen bond interactions of the most active compounds involve the amino acids Arg102, Asn112, His115, His52, Thr91. For the Candida albicans (PDB ID: 3WBZ), the hydrogen bonds involve the same amino acids Glu15, Lys14 şi Tyr17. In the case of Aspergillus niger fungi (PDB ID: 1QO7), the hydrogen bonds involve the following amino acids Ala217, Arg219, Arg199, Cys216 şi Thr317. In all cases, the atoms involved in the interaction are nitrogen and oxygen in the molecules of the two participants.*

Keywords: *amide derivatives, antimicrobial activity, informational descriptors.*

1. INTRODUCTION

For a long time, amide groups have been discovered in the structure of drugs. For example, lidocaine is a local anesthetic of amidic type, which is derived from cocaine. Another example is tropicamide, a mydriatic-cycloplegic substance used as an ophthalmic solution. Amide compounds exhibit therapeutic activities including antibacterial, antiviral, antifungal, cardiotonic, anti-tumor, anti-inflammatory, sodium channel blockage [1-13]. A biological response of the body to the action of such a drug appears from its interaction with a particular biological target [14].

The interaction of the ligand (drug) and the biological receptor can be studied using methods of chemical structure - biological activity correlation, as well as molecular docking methods. The latter methods investigate the theoretical possibilities of binding a ligand to the binding site of a biological target, by calculating the intermolecular energies. The interaction of the two, the drug and the biological target, leads to the formation of complexes, the most

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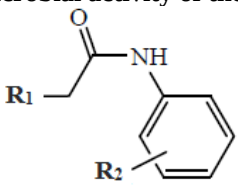
common bonds being hydrogen bonds, electrostatic forces (ion-ion), dipole-ion forces, dipole-dipole forces and reciprocal attractions through van der Waals forces. Hydrogen bonds play an important role, because they are characterized by an advanced specificity, kinetic lability and low binding energy, parameters that make them suitable for complex formation.

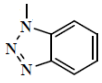
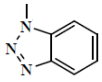
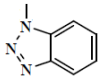
This paper is a correlation study and molecular docking on acetamide derivatives, which show antimicrobial activities against *Escherichia coli*, *Staphylococcus aureus*, *Candida albicans* and *Aspergillus niger*. The paper is a continuation of the study regarding two of the receptors: *Escherichia coli* (PDB ID: 3t88) and *Candida albicans* (PDB ID: 3Q8U) [15].

2. MATERIALS AND METHODS

The inhibitory activities of the 9 acetamide compounds were taken from the literature [16, 17], where their testing is indicated at the concentration of 100 μg/mL in dimethylformamide. The values of biological activities are presented in Table 1.

Table 1. Structure and antimicrobial activity of the studied derivatives [15, 16].



Cmpd	*R_1*	*R_2*	*Antimicrobial activity – Inhibition zones [mm]*			
			E. coli	*S. aureus*	*C. albicans*	*A. niger*
1		H	8.44	-	7.11	-
2		$4CH_3$	26.44	16.19	9.65	15.80
3		4Cl	20.92	29.12	23.90	11.13
4		H	9.16	13.90	14.72	-
5		$4CH_3$	9.49	12.89	8.64	-
6		4Cl	17.38	20.43	11.11	-
7		H	12.19	10.20	9.30	14.07
8		$4CH_3$	10.78	-	7.34	9.51
9		4Cl	11.71	8.06	8.11	10.23

The molecular structures of the studied compounds were used as input data for MOPAC software (Molecular Orbital PACkage) [18]. This is a software designed (University of Texas, USA) to implement semi-empirical quantum chemistry algorithms. The electron populations distributed on atomic or molecular orbitals expressed by MOPAC software were then used for the determination of information parameters, Shannon entropy, Onicescu energy and informational temperature, using specially dedicated programs [19]. These parameters may be used for the characterization of the molecular compounds. To establish acetamide derivative binding patterns in the active sites of the receptors, docking experiments were performed using FlexX docking software [20]. FlexX is an automatic docking tool for flexible ligands, on a highly diverse data set of complexes from the Protein Data Bank, developed by BioSolveIT GmbH, Germany. The receptor molecules were taken from the Protein Data Bank [21], which contains experimentally-determined structures of proteins, nucleic acids and complex assemblies.

3. RESULTS AND DISCUSSION

Informational quantities (Shannon entropy S, Onicescu energy O and information temperature T) are defined as convex functions on probability fields generated by the distribution of the electronic population on the quantum levels in each atom after the formation of chemical bonds [19]. For the calculation of these quantities the Lowdin process was used for the distribution of the electronic population in the molecule, as it is closer to reality since the distribution of the electronic densities in the area between atoms takes into account their center of gravity, in accordance with the chemical intuition that a more electronegative atom will receive a higher electron density than a less electronegative atom. The information quantities were regressively correlated with antimicrobial activities and the following results were obtained (Table 2).

Table 2. Regression results for the information quantities

Parameter	***R^2 [%]***			
	E. coli	***S. aureus***	***C. albicans***	***A. niger***
S	19.6	20.0	16.6	25.9
O	7.4	3.7	2.5	1.9
T	10.4	11.2	29.6	14.3

The antimicrobial efficacy of all studied compounds has been subjected to the docking technique to explore the way they are binding to the biological receptors. Thus, the interactions between drug compounds and *Escherichia coli* (PDB ID: 3t88) and *Staphylococcus aureus* (PDB ID: 3Q8U) bacteria are shown in Figs. 1 and 2.

The correlation coefficients are very close in the case of the four types of antimicrobial activities and are higher in the case of informational entropy. The explanation would be that informational entropy is defined on a probability field generated by the electronic distribution on the quantum levels of each atom. Each atom has a certain electronic configuration and therefore a specific electronic distribution, meaning that the size S be a fingerprint for each atom in the molecule [22]. MenB from *Escherichia coli* is 1,4-dihydroxy-2-naphthoyl-CoA synthase, a naphthoquinone that functions as a redox active cofactor in the electron transport chain of some Gram negative and most Gram-positive bacteria. Mammalian cells cannot synthesize menaquinone, and thus the enzymes in the biosynthetic pathway of bacterial menaquinone are potential targets for novel antibacterial drug [16].

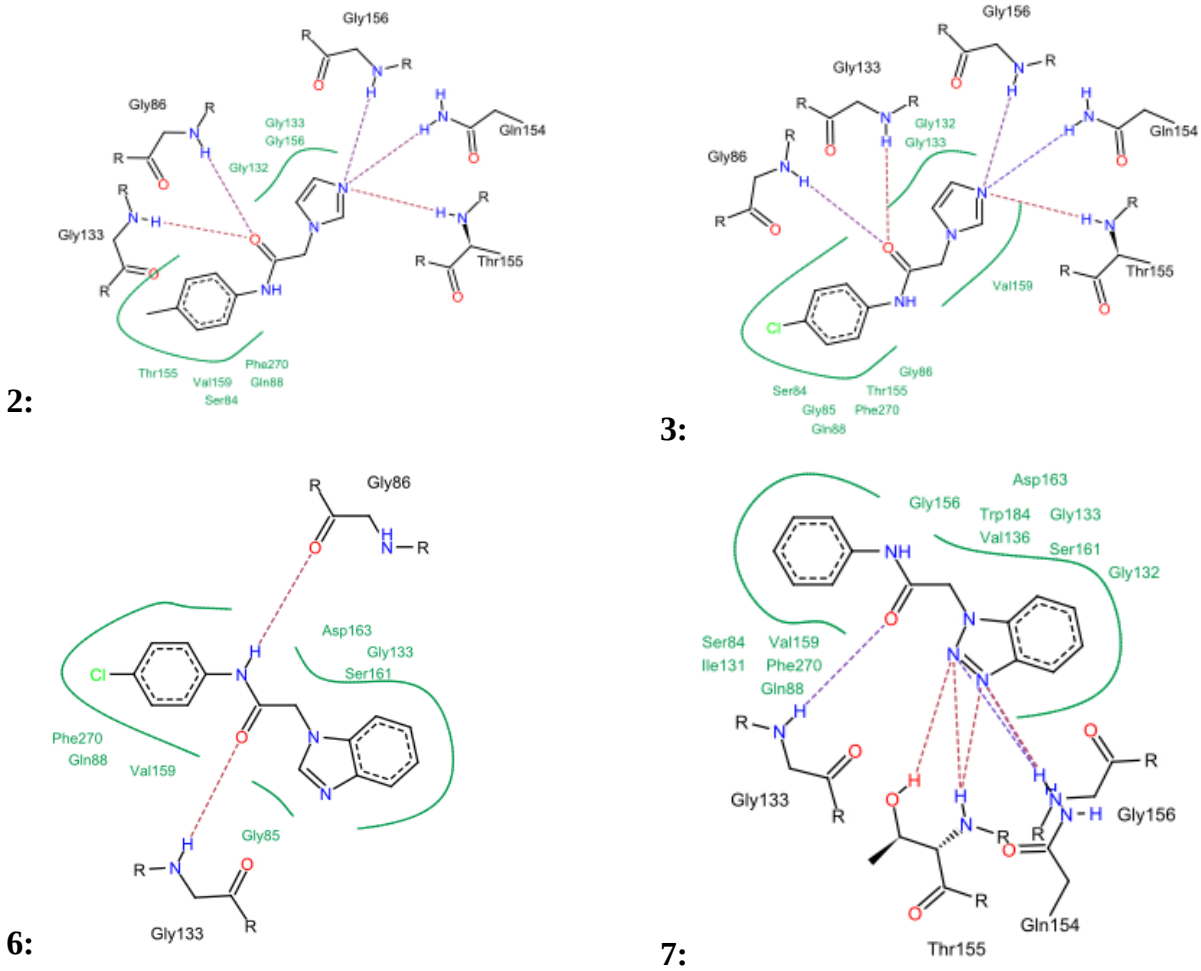


Figure 1. Diagram 2D for the most active inhibitors (2, 3, 6, 7) inside the receptor-binding site of *Escherichia coli*.

From the docking studies it is shown that the active sites of Gram-negative *E. coli* bacteria (PDB ID: 3t88) containing amino acids Gly133, Gly86, Gly156, Gin154, Thr 155, Gly132, Gin88, Val159, Val136, Ser84, Ser161, Phe270, Ile131, Asp163 and Trp184 are approximately the same interaction sites for the top four most active compounds (2, 3, 6, 7). Hydrogen-binding interactions involve the amino acids Gly133, Gly86, Gly156, Gin154, Thr155 (Fig. 1).

Nucleoside di-phosphate kinase (NDK) from *Staphylococcus aureus* has a catalytic mechanism governed by a histidine that coordinates a magnesium ion at the active site [22-24]. The active sites of Gram-positive *S. aureus* bacteria (PDB ID: 3Q8U) contain the amino acids Arg102, Asn112, His115, His52, Thr91, Phe54, Leu61, Gly110, Tyr49, Phe57, Asp118, His115 (Fig. 2), and the hydrogen bond interactions of the first four most active compounds (3, 6, 2, 4) involve the amino acids Arg102, Asn112, His115, His52, Thr91.

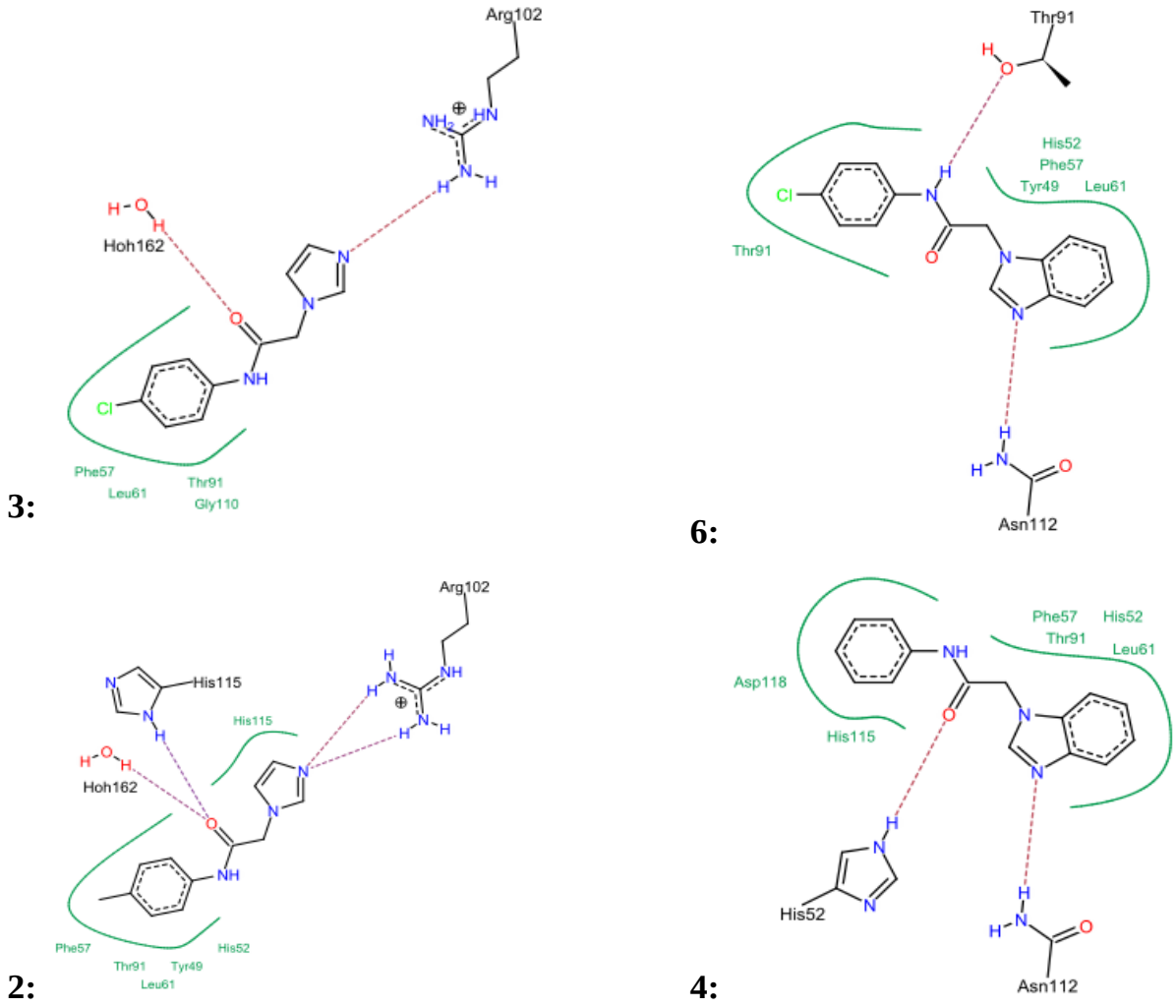


Figure 2. Diagram 2D for the most active inhibitors (3, 6, 2, 4) within the receptor-binding site of *Staphylococcus aureus.*

Guanylyltransferase from *Candida albicans* (CaThg1), like its other polymerase relatives, has subunits that can be described as a hand shape, comprising a palm domain that includes a catalytic core (residues 1–137) and a finger domain (residues 138–268) [25, 26].

The interactions between drug compounds and biological targets of *Candida albicans* (PDB ID: 3WBZ) and *Aspergillus niger* (PDB ID: 1QO7) respectively are shown in Figs. 3 and 4.

In the case of *C. albicans* (PDB ID: 3WBZ), the most active acetamide compounds (3, 4, 6, 2) are orientated into the same active site of the receptor containing the following amino acid residues: Glu15, Lys14, Tyr17, Glu13 şi Asn16. Hydrogen bonds involve the same amino acids Glu15, Lys14 şi Tyr17 (Fig. 3).

Epoxide hydrolases are important in the protection of cells against epoxides that can be potentially harmful. Transformation of epoxides into diols that are less toxic and easily excreted is an effective strategy. Some microorganisms employ the same chemistry to use epoxides as a carbon source.

The active site of microsomal epoxide hydrolase has a classical catalytic triad composed of Asp226, Glu404 and His431 [27], and also a glutamic acid residue and two tyrosines that probably help in catalysis [28].

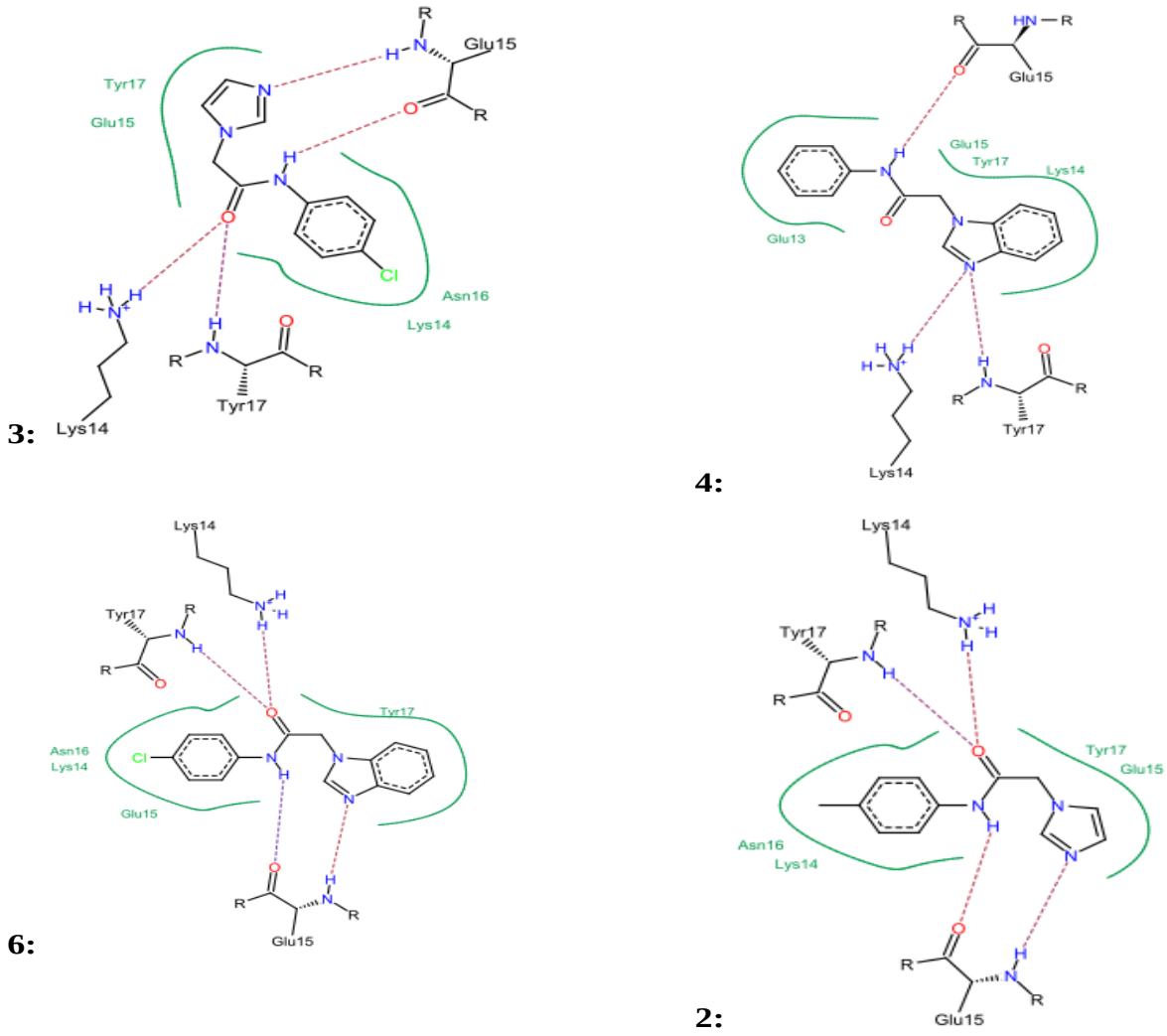


Figure 3. Diagram 2D for the most active inhibitors (3, 4, 6, 2) within the receptor-binding site of *Candida albicans.*

In the case of *A. niger* fungi (PDB ID: 1QO7), the most active acetamide compounds are 2, 7, 3, 9. They are orientated into the receptor active sites containing the following amino acid residues: Ala217, Arg219, Arg199, Cys216, Thr317, Met218, Met245, Arg119, Leu334, Leu166, Leu200, Leu213, Leu330 şi Phe196. Hydrogen bonds involve the following amino acids Ala217, Arg219, Arg199, Cys216 şi Thr317 (Fig. 4).

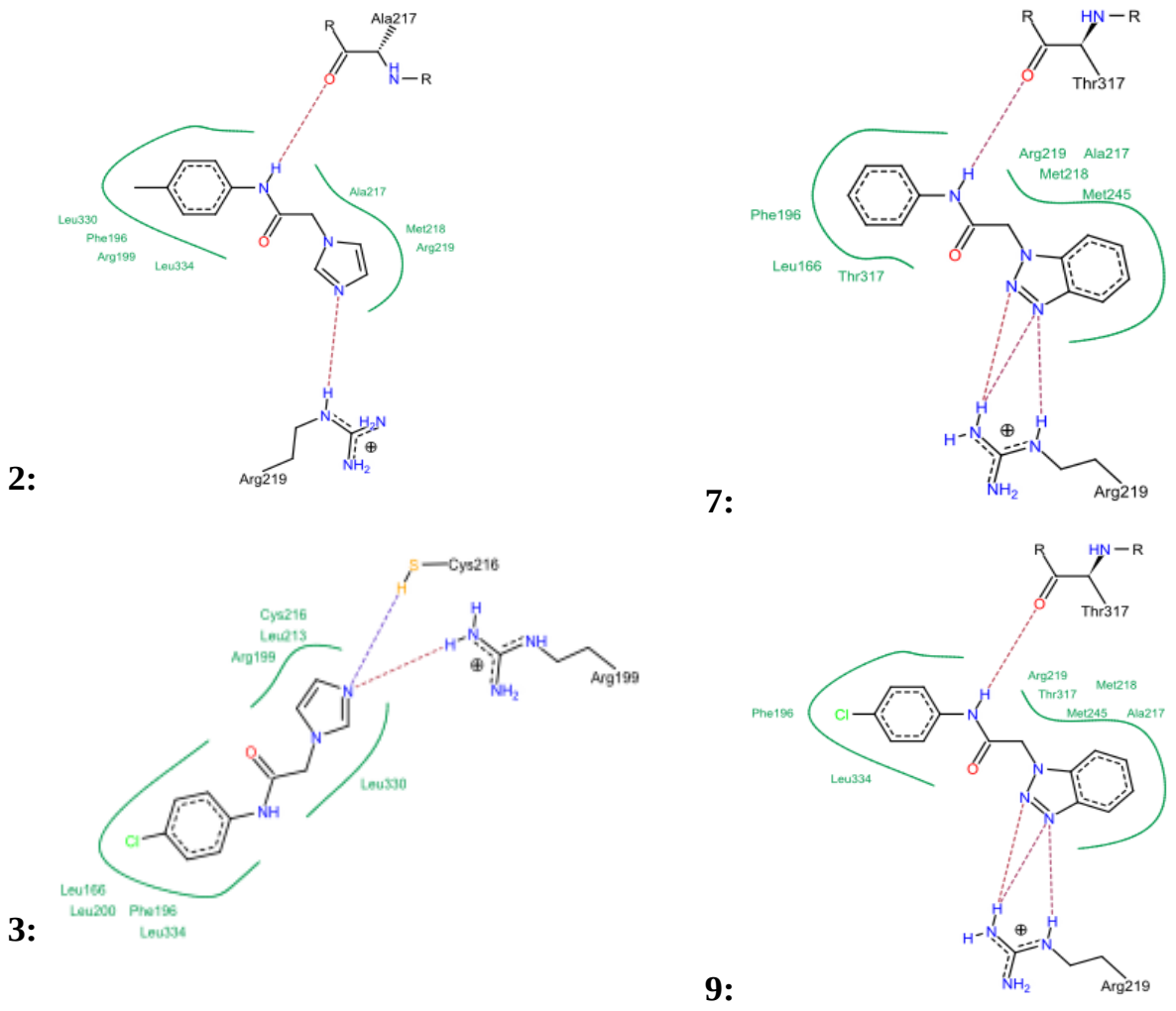


Figure 4. Diagram 2D for the most active inhibitors (2, 7, 3, 9) within the receptor-binding site of *Aspergillus niger*.

In all cases, the groups of atoms involved in the formation of hydrogen bonds contain nitrogen and oxygen atoms.

4. CONCLUSIONS

The molecular recognition of the ligand (drug) by the biological receptor depends on the steric accommodation of the two and on the intensity of the interaction, which is all the more pronounced as the ligand and the receptor have a proper distribution of electrical charges on atoms. Informational quantities (Shannon entropy and molecular temperature) describe well the antimicrobial activities of the studied acetamide compounds. An interesting result is that the values of the correlation coefficients for these parameters are very close as value in the case of the four different biological receptors. Using docking studies we conclude that the atoms most involved in ligand-receptor interaction are the oxygen atom belonging to the amide group and the nitrogen atom from position 3 of the imidazolic nucleus, respectively the nitrogen atoms from positions 2 and 3 of the triazolic heterocycles of the molecules of the studied compounds.

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