Pharmaceutical Sciences, 2022, 28(4), 564-571
doi:10.34172/PS.2022.3
https://ps.tbzmed.ac.ir/

Research Article

In Vitro and *In Silico* Studies on the Anticancer and Antimicrobial Activity of Cu(II), Ni(II) and Co(II) Complexes with Bis (Pyrazolyl) Borate Derivative Ligand

Monireh Ghorbanpour[1], Behzad Soltani[1*], Ommoleila Molavi[2], Elnaz Mehdizadeh Aghdam[2,3]

[1]*Department of Chemistry, Faculty of Basic Science, Azarbaijan Shahid Madani University, Tabriz, Iran.*
[2]*Department of Pharmaceutical Biotechnology, Faculty of Pharmacy, Tabriz University of Medical Sciences, Tabriz, Iran.*
[3]*Molecular Medicine Research Center, Biomedicine Institute, Tabriz University of Medical Sciences, Tabriz, Iran.*

Article Info

Article History:
Received: 16 October 2021
Accepted: 26 January 2022
ePublished: 28 January 2022

Keywords:
-Anticancer activity
-Antimicrobial activity
-Bis(pyrazolyl)borate derivatives
-MDA-MB-231
-Metal complexes
-Molecular docking

Abstract

Background: The development of the pyrazole-based complexes is greatly enhanced due to the identification of their structure as medicinal application. Keeping in view therapeutic and biological activities of pyrazoles based compounds and the potential of transition metals in the antimicrobial application area, we find it vital to join the chemistry of both moieties in designing and developing biometal compounds which could aggressively work against various bacterial species and cancer cells. In this work, we report the synthesis and characterization of copper (II), nickel (II) and cobalt (II) complexes of dihydrobis(pyrazolyl)borate ligands. Also, antibacterial activities, MTT assay and molecular docking of these compounds were investigated.

Methods: A bidentate N-donor pyrazole-based ligand abbreviated as $K[H_2B(Pz^{Me2})_2]$ and corresponding complexes with Cu(II), Ni(II) and Co(II) were synthesized and characterized. The anticancer activities of the synthesized compounds were studied against the (MDA-MB-231) cell lines. The antibacterial investigations of the synthesized compounds against the gram-positive (*B. subtilis*) and the gram-negative (*S. enterica*) bacteria were performed. In addition, molecular docking of the synthesized compounds with YmaH (PDB ID: 3HSB) protein, ecKAS III (PDB ID:1HNJ) protein and DNA dodecamer (PDB ID: 1BNA) as the possible targets was studied.

Results: The result showed among the investigated compounds, complex $[Cu(H_2B(Pz^{Me2})_2)_2]$ indicated the highest cytotoxicity and bacterial inhibition.

Conclusion: In summary, we have synthesized a type of N- donor pyrazole-based ligand and corresponding metal complexes. *In silico* molecular docking along with the experimental MTT assay and antibacterial studies, indicated the metal complexes are more bioactive than free ligand and can be excellent candidates for further evaluations in the biological area.

Introduction

Since the first reportage of poly(pyrazolyl)borates and in especially bis(pyrazolyl)borates by Trofimenko, these compounds have been developed in the inorganic coordination chemistry area and widely employed in various transition metal coordination complexes as anionic N-donor ligands.[1-5] In the pyrazole based ligands, by controlling the number of pyrazolate rings and the substituents of different derivatives, their structure can be modified for the design and synthesis of the ligands with special properties to formation of stable complexes.[6-10] On the other hand, the excellent therapeutic properties of pyrazole-based drugs have attracted the attention of pharmaceutical chemists for the synthesis, development and investigation of various new chemotherapy drugs.[11-14] Development of the pyrazole-based coordination compounds is greatly enhanced due to the identification of their structure as anti-cancer, anti-fungal, anti-bacterial, anti-viral and anti-inflammatory agents.[15-19] On the other hand, Docking simulation can help us to understand the environment around compounds in the active site and allowing medicinal chemists to design and modify compounds. Keeping in view therapeutic and biological activities of pyrazoles based compounds and the potential of transition metals such as copper, cobalt, and nickel in the antimicrobial application area, we find it vital to join the chemistry of both moieties in designing and developing biometal compounds which could aggressively work against various bacterial species and cancer cells.[20-24] In this work, we report the synthesis and characterization of copper (II), nickel (II) and cobalt (II) complexes of dihydrobis(pyrazolyl)borate ligands. Also, antibacterial activities, MTT assay and molecular docking of these compounds were investigated.

*Corresponding Author: Behzad Soltani, E-mail: soltani@azaruniv.ac.ir

Materials and Methods

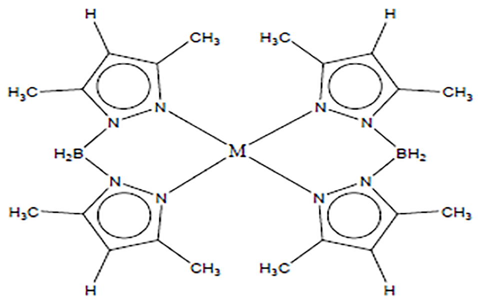
Materials and instrumentation

All chemicals and solvents were obtained from Merck, the solvents were dried according to literature methods when anhydrous conditions were necessary. The MDA-MB-231 cell lines were obtained from Pasteur Institute (Tehran, Iran). Also, the fetal bovine serum (FBS) was bought from HyClone and the RPMI 1640 was obtained from Gibco. FT-IR spectra were recorded on a Bruker instrument in the 400-4000 cm^{-1} range using KBr pellets. The melting points were determined with a Stuart Scientific SMP1 apparatus. Elemental analyses (CHN) were measured in an Elementar Vario ELIII. Disk diffusion method was applied for antibacterial investigations.

Synthesis of the ligand $K[H_2B(Pz^{Me2})_2]$ (1) and complexes $[M(H_2B(Pz^{Me2})_2)_2]$ (M= Cu, Ni, and Co) (2-4)

Similar to earlier described method,[25,26] the synthesized 3,5-dimethyl pyrazole,[23] (0.19 g, 2 mmol) and KBH_4 (0.05 g, 1 mmol) were heated to 100˚C under nitrogen atmosphere and hydrogen evolution started at this point. The temperature is raised to 130˚C gradually and was controlled evolution of hydrogen, after the production of (2 mmol) of hydrogen, the reaction was finished and the resulting white precipitate washed with the mixture of THF (20 mL) and n-hexane (10 mL). Finally, the product ligand **(1)** was dried in vacuum over P_2O_5. For the syntheses of complexes **(2-4),** similar to earlier described method,[25,26] the synthesized ligands **(1)** (0.48 g, 2 mmol) was dissolved in 15 ml dichloromethane and the methanolic solution of metal salts $Cu(CH_3COO)_2.H_2O$, $Ni(CH_3COO)_2.4H_2O$, $Co(CH_3COO)_2.4H_2O$ (1 mmol) were added to the ligand solution, after stirring of the reaction for overnight at room temperature. The resulting precipitate of the metal complexes was collected using vacuum filtration, and then dried in vacuo. The structures of synthesized complex were illustrated in Figure 1.

$K[H_2B(Pz^{Me2})_2]$ (**1**): Yield: 0.16 g (67%). IR: (KBr, cm^{-1}): 2924-2866(w), 2435(m), 1631(w), 1585(m), 1482(w), 1422(s), 1371(w), 1111(s), 1028(m), 979(m), 948(m), 811(m), 766(m). Elem Anal. (%) Calcd for $C_{10}H_{16}BKN_4$: C, 49.59; H, 6.65; N, 23.13. Found: C, 49.55; H, 6.59; N, 23.06. m.p: 210 ºC.



M= Cu (II), Ni (II), Co (II)

Figure 1. Structure of the complexes **(2-4)**.

$[Cu(H_2B(Pz^{Me2})_2)_2]$ (**2**): Yield: 0.38 g (84%). FT-IR (KBr, cm^{-1}): 2958-2859(m), 2451-2276(s), 1534(s), 1495(w), 1450(m), 1416(s), 1363(m), 1321(m), 1173(s), 1110(s), 1053(m), 883(m), 801(s), 779(s), 655(m), 458(w). Elem Anal. (%) Calcd for $C_{20}H_{32}B_2CuN_8$: C, 51.14; H, 6.86; N, 23.85. Found: C, 51.22; H, 6.90; N, 23.81. m.p: >300 ºC (Dec.).

$[Ni(H_2B(Pz^{Me2})_2)_2]$ **(3):** Yield: 0.33 g (72%). FT-IR (KBr, cm^{-1}): 2960-2860(m), 2453-2261(s), 1535(s), 1491(w), 1453(m), 1422(s), 1388(s), 1183(s), 1110(s), 1036(m), 897(m), 788(s), 622(m), 418(w). Elem Anal. (%) Calcd for $C_{20}H_{32}B_2NiN_8$: C, 51.67; H, 6.93; N, 24.10. Found: C, 51.73; H, 6.91; N, 24.05. m.p: >300 ºC (Dec.).

$[Co(H_2B(Pz^{Me2})_2)_2]$ **(4)**[26]**:** Yield: 0.34 g (75%). FT-IR (KBr, cm^{-1}): 2925-2878(m), 2434-2303(m), 1653(m), 1595(s), 1533(m), 1422(m), 1306(s), 1028(s), 855(m), 778(s), 635(m), 480(w),. Elem Anal. (%) Calcd for $C_{20}H_{32}B_2CoN_8$: C, 51.65; H, 6.93; N, 24.09. Found: C, 51.70; H, 6.92; N, 23.98. m.p: >300 ºC (Dec.).

Antibacterial activity

The synthesized compounds (**1-4**) were evaluated for their antibacterial activity against *Bacillus subtilis* (ATCC6633) and *Salmonella enterica* (ATCC14028) by the disk diffusion method. Gentamicin was used as a standard drug and Dimethyl sulfoxide (DMSO) was employed as a negative control. The bacterial cultures were developed by selective nutrient broth at 37 °C for 24 h then, were diluted to 0.5 McFarland standards. Streaking of the lawn growth in Mueller-Hinton Agar medium was performed using a cotton swab was dipped into the nutrient broth medium of the organism. This investigation was performed on Muller Hinton Agar disks with a solution of synthesized ligand and complexes (80 mM) in DMSO. The filter paper disc with a diameter of 6 mm impregnated with 10 µL of synthesized compounds solution and staid on the agar surface. After incubating the plates for 24h at 37 °C, the inhibition zone was obtained using the measurement of minimum dimensions of the zone without the bacterial growth around the disk.[23,27,28] All the tests were performed in triplicate and an average of determination was recorded.

Cytotoxicity assay

The synthesized N-donor ligand **(1)** and corresponding metal complexes **(2-4)** were evaluated for their cytotoxicity against human breast cell line (MDA-MB-231) by using an MTT assay. This colorimetric assay is based on the converting of the yellow (MTT) tetrazolium bromide to a purple formazan derivative mitochondrial succinate dehydrogenase in viable cells. MDA-MB-231 (human breast cell line) was prepared from the National Cell Bank of Iran (Pasteur Institute, Tehran, Iran) and was cultured in RPMI-1640 medium with 10% fetal bovine serum and were incubated in the 5% CO_2 and at 37 °C.[29,30] The cell lines were seeded in a 96-well plate at a density of 1.0×10^4 cells/well and treated for 48h, with various concentrations of the synthesized investigated ligand, corresponding complexes and Paclitaxel (0.5, 1, 2, 4, 8, 16, 31, 62, 125, 250 and 500 μM). At the end of the drug treatment, 100 μL of the MTT solution (5 mg/mL in PBS) were added to each well, then incubated for 4 h.[31] The MTT formazan precipitate was dissolved in 200 μL of DMSO, and the absorbance was recorded at 595 nm by a plate reader. Untreated MDA-MB-231 cell and Paclitaxel drug were used as a negative and positive controls, respectively. This experiment was repeated at least three times independently for each condition. The IC_{50} values were measured from concentration-response curves with GraphPad Prism 8 software and used to measure of cellular sensitivity to a given treatment.

Molecular docking

Based on the literature and due to the fact that these proteins (YmaH, ecKAS III) and DNA have significant inhibition against the studied bacteria and investigated cell line, respectively. So these macromolecules are shown to be the possible targets for anticancer and antibacterial agents.[32-34] The molecular docking of the synthesized compounds with the YmaH (PDB ID: 3HSB), ecKAS III (PDB ID: 1HNJ) protein and DNA dodecamer (PDB ID: 1BNA) d(CpGpCpGpApApTpTpCpGpCpG) as the possible targets was performed by using the AutoDock Tools (ADT) version 1.5.6 and AutoDock version 4.2.6.[35-39] The crystal structures of the macromolecules were obtained from the Protein Data Bank website (RCSB) (http://www.rcsb.org/pdb/). The three-dimensional structures of synthesized ligand and Ni(II) complex were obtained with optimization by M06-2X method with Lanl2DZ and other base sets and the three-dimensional structures of Cu(II) complex and Co(II) complex were obtained from X-ray crystallography.[25,26] First of all, the including water molecules were deleted and the energy calculations were made by genetic algorithms. During *in silico* docking, receptors were set rigid, whereas all the torsional bonds of small molecules were set free. One hundred independent runs with the grid box 126×126×126 Å (grid spacing=0.375Å) was performed to evaluate the binding energy and other results of the inhibitors within the macromolecules.[40,41]

Results and Discussion

FT-IR spectral studies

In the FT-IR spectrum of the ligand, the removal of the ν(N-H) band of pyrazole ring in the 3200 ± 50 cm^{-1} range, also the appearance of the multiple peaks of ν(B-H) bond in the range of 2200- 2500 cm-[1], indicated the formation of ligand. In the FT-IR spectrum of the complexes, the bands in the region of 400-500 cm^{-1} are assigned to the ν(M-N) stretching vibrations bands. The strong aliphatic bands attributed to ν(C-N) vibrations appeared in the ranges of 1321-1388 cm^{-1}. The shifts of other bands of the complexes in compare with the free ligand indicates coordination of the donor atoms to the metal centers.

Table 1. Zone of inhibition of the synthesized compounds (**1-4**).

Compounds	Zone of inhibition (mm)	
	B. subtilis	***S. enterica***
$K[H_2B(Pz^{Me2})2]$ **(1)**	15.33	13.66
$[Cu(H_2B(Pz^{Me2})_2)_2]$ **(2)**	27.66	26.00
$[Ni(H_2B(Pz^{Me2})_2)_2]$ **(3)**	25.00	23.66
$[Co(H_2B(Pz^{Me2})_2)_2]$ **(4)**	24.33	22.00
Gentamicin	29.66	30.00
DMSO	6 (NZ)	6 (NZ)
Filter paper disk	6 (NZ)	6 (NZ)

NZ = no zone of inhibition; the filter disk itself is 6 mm in diameter.

Antibacterial studies

The synthesized compounds, positive and negative controls, were screened separately to evaluate their antibacterial activity against Gram-positive (*B. subtilis*), and Gram-negative (*S. enterica*). The results were summarized in Table 1.

The experimental result of the antibacterial investigation indicated that complexes have considerable activity compared to the free corresponding ligand. The synthesized compounds showed a zone of inhibition ranging from 15.33 to 27.66 mm against *B. subtilis* and 13.66 to 26.00 mm against *S. enterica*. Furthermore, among the metal complexes, complex $[Cu(H_2B(Pz^{Me2})_2)_2]$ (**2**) showed higher antibacterial activity than others. Positive control (Gentamicin) showed significant inhibition zones against the test microorganisms ranging from 29.66 mm to 30.00 mm against Gram-positive and Gram-negative bacteria, respectively. Dimethyl sulfoxide as a negative control revealed no inhibitory effect against any of the bacterial strains. Increasing the antimicrobial activity of coordination compounds in the comparison with the organic ligand can be explained based on chelation theory.[42,43] With the coordination of metal ions to the donor ligands, the polarity of the metal center is reduced to a greater extent due to the overlapping of the ligand orbital and partial sharing of the positive charge of the metal ion with donor groups. Furthermore, delocalization of the π-electrons over the whole chelate ring is increased and lipophilicity of the complexes is enhanced. So the lipid solubility factor is the main factor that controls the antimicrobial activity.[44] Also, the lack of an outer membrane in the Gram-positive strains, which acts as a barrier for penetration, causes the high inhibition zone of investigated compounds against the Gram-positive strains compared with the Gram-negative bacteria.[45] Generally, metal complexes are more bioactive than free organic ligands and can be further applied in the pharmaceutical industry, as an antimicrobial agent.

The MTT assay studies

MTT assay was used for the determination of the cytotoxicity of the synthesized ligand and corresponding copper (II), nickel (II) and cobalt (II) complexes against human breast cancer cell line (MDA-MB-231). The

Table 2. IC_{50} values (μM ± SD) values of the ligand **(1)**, complexes **(2-4)** and Positive control.

Compound	IC_{50} (μM)
$K[H_2B(Pz^{Me2})2]$ **(1)**	40.25 ± 1.66
$[Cu(H_2B(Pz^{Me2})_2)_2]$ **(2)**	18.66 ± 0.54
$[Ni(H_2B(Pz^{Me2})_2)_2]$ **(3)**	22.47 ± 0.17
$[Co(H_2B(Pz^{Me2})_2)_2]$ **(4)**	21.39 ± 0.85
Positive control	18.84 ± 0.15

obtained result was evaluated according to cell inhibition which is illustrated as IC_{50} values (summarized in Table 2). The results showed that the IC_{50} value of complex $[Cu(H_2B(Pz^{Me2})_2)_2]$ **(2)** is much lower than free ligand **(1)** and other investigated complexes (**3,4**), also this complex showed almost the same activity with the Paclitaxel as a drug against the investigated cell line. Generally, the coordination compounds have high cytotoxicity compared with free organic compounds and it can be explained by chelation theory which is the reason for the high cytotoxic character of the inorganic compounds.[46,47] Our experimental findings, along with molecular docking studies, suggest that the copper complex could be an excellent candidate for further anticancer studies.

Molecular docking analysis

In silico Molecular docking studies were carried out by AutoDock software for a synthesized ligand and their Cu(II), Ni(II), and Co(II) complexes with the DNA and two selected proteins (YmaH and ecKAS III). The predicted binding energy and other results are summarized in Tables 3, 4 and 5. According to the obtained results of these studies, the investigated receptors showed better interactions with most of the synthesized metal complexes compared to the free ligands. As can be seen in Table 3, the complex $[Co(H_2B(Pz^{Me2})_2)_2]$ **(4)** bound to DNA with the binding energy of -6.05 kcal/mol and the complex $[Cu(H_2B(Pz^{Me2})_2)_2]$ **(2)** bound to YmaH and ecKAS III proteins with a binding energy of -7.48 kcal/mol and -6.47 kcal/mol, respectively and showed the highest binding energy among the studied compounds. Figure 2 shows the 3D molecular docking modeling between complex $[Co(H_2B(Pz^{Me2})_2)_2]$ **(4)** and DNA dodecamer. Figures 3 and 4 presented the molecular docking modeling between

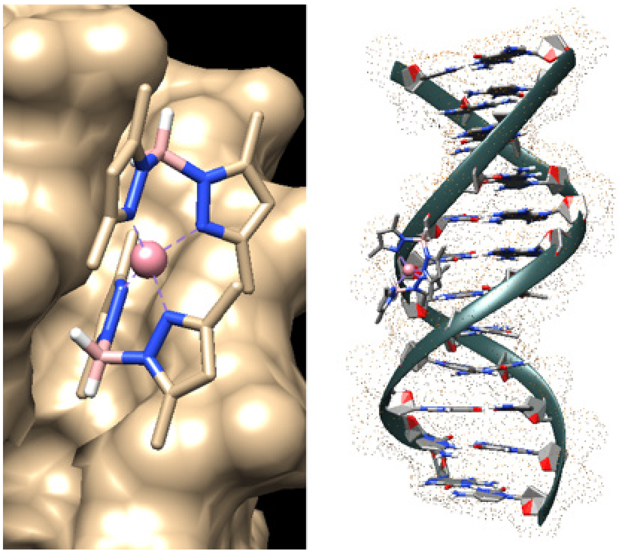


Figure 2. Molecular docking modeling between complex $[Co(H_2B(Pz^{Me2})_2)_2]$ (**4**) and DNA dodecamer.

Table 3. The obtained results of the docking of compounds (**1-4**) and DNA dodecamer (PDB ID: 1BNA).

Compound	Binding Energy	Intermol energy	(Van der Waals + Hbond + desolv) energy	Electrostatic energy	Total internal	Unbound energy	Torsional energy	Ki
1	-5.82	-6.42	-6.27	-0.14	-0.8	-0.8	0.6	54.1
2	-5.81	-5.81	-5.72	-0.1	0	0	0	55.03
3	-5.69	-5.69	-5.65	-0.04	0	0	0	67.28
4	-6.05	-6.05	-5.84	-0.21	0	0	0	36.6

Table 4. The obtained results of the docking of compounds (**1-4**) and YmaH (PDB ID: 3HSB) protein.

Compound	Binding Energy	Intermol energy	(Van der Waals + Hbond + desolv) energy	Electrostatic energy	Total internal	Unbound energy	Torsional energy	Ki
1	-5.95	-6.54	-6.46	-0.08	-0.54	-0.54	0.6	43.78
2	-7.48	-7.48	-7.45	-0.03	0	0	0	3.28
3	-7.22	-7.22	-7.2	-0.02	0	0	0	5.14
4	-7.01	-7.01	-6.96	-0.05	0	0	0	7.27

Table 5. The obtained results of the docking of compounds (**1-4**) and ecKAS III (PDB ID: 1HNJ) protein.

Compound	Binding Energy	Intermol energy	(Van der Waals + Hbond + desolv) energy	Electrostatic energy	T o t a l internal	Unbound energy	Torsional energy	Ki
1	-5.64	-6.23	-6.17	-0.06	-0.66	-0.66	0.6	73.83
2	-6.47	-6.47	-6.39	-0.08	0	0	0	18.1
3	-6.27	-6.27	-6.2	-0.07	0	0	0	25.36
4	-5.94	-5.94	-5.87	-0.07	0	0	0	44.44

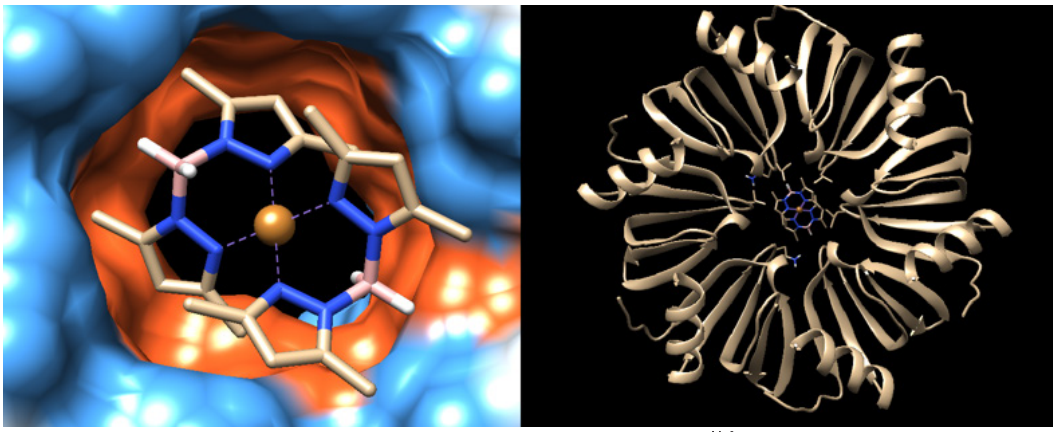


Figure 3. Molecular docking modeling between complex [Cu($H_2B(Pz^{Me2})_2)_2$] (**4**) and YmaH protein.

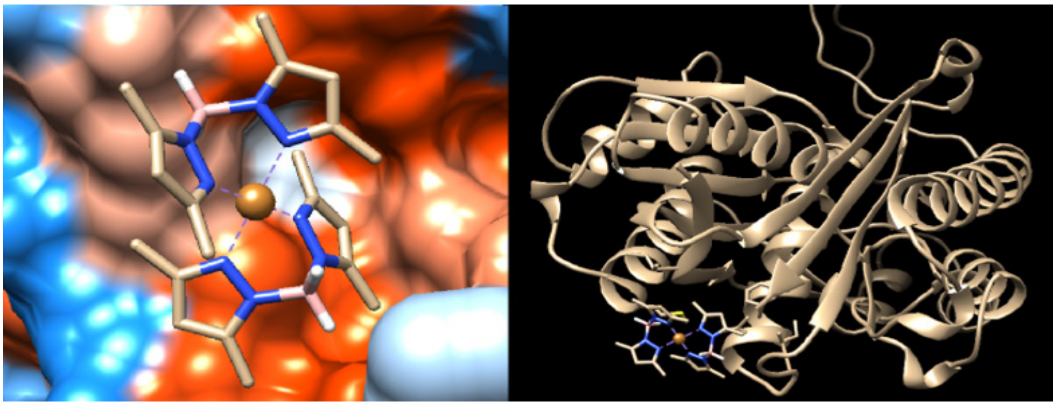


Figure 4. Molecular docking modeling between complex [Cu($H_2B(Pz^{Me2})_2)_2$] (**4**) and ecKAS III protein.

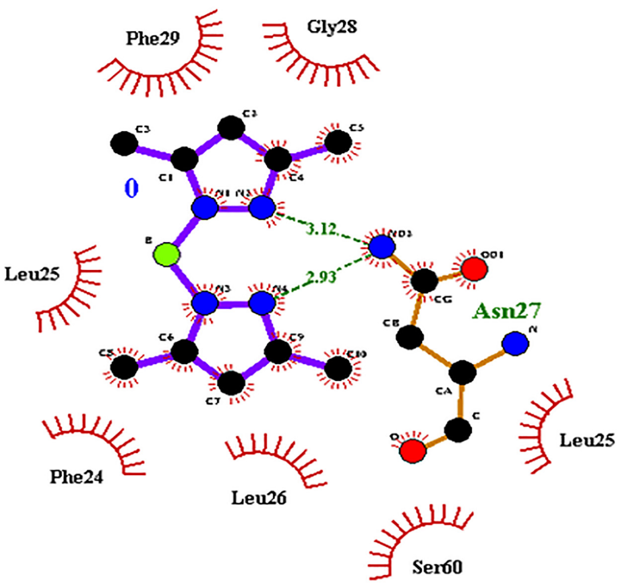


Figure 5. Binding mode of ligand K[$H_2B(Pz^{Me2})_2$] (**1**) and the YmaH protein, the H-bond (green line) is displayed as line.

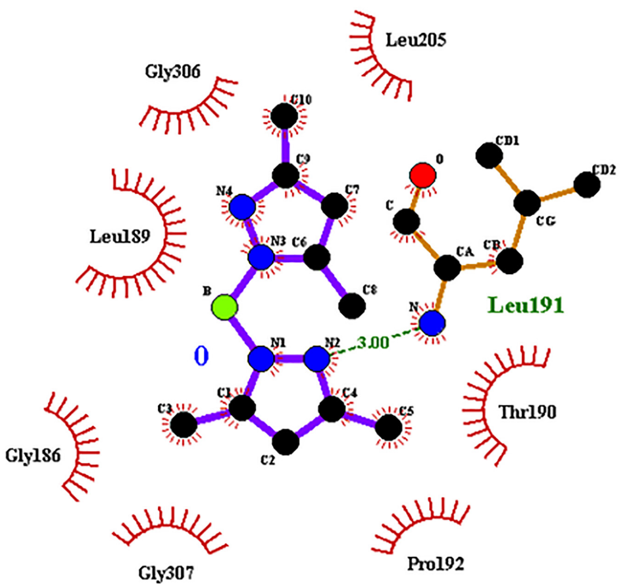


Figure 6. Binding mode of ligand K[$H_2B(Pz^{Me2})_2$] (**1**) and the ecKAS III protein, the H-bond (green line) is displayed as line.

complex [Cu($H_2B(Pz^{Me2})_2)_2$] **(2)** and the YmaH and ecKAS III protein, respectively. Uncoordinated free nitrogen atoms in the structure of ligand cause the formation of the hydrogen bonding with receptors, As can be seen in Figure 5, ligand K[$H_2B(Pz^{Me2})_2$] **(1)** presented two strong hydrogen bond between the two N atom from the ligand and Asn27 group of YmaH of Bacillus substilis protein. Also, the hydrogen bond of ligand and Leu191 group of ecKAS III (PDB code: 1HNJ) protein was shown in Figure 6. Our findings from molecular docking investigation and biological assay data suggest that these synthesized complexes are the potential antibacterial and anticancer inhibitory.

Conclusion

In summary, we have synthesized a type of N- donor pyrazole-based ligand and corresponding copper (II), nickel (II), and cobalt (II) complexes. The *in vitro* antibacterial studies against Gram-positive (*B. subtilis*), and Gram-negative (*S. enterica*) and MTT assay against MDA-MB-231 (human breast cancer) cell line carried out for the ligand as well as for the complexes. The result showed among the investigated compounds, complex $[Cu(H_2B(Pz^{Me2})_2)_2]$ **(2)** indicated the highest cytotoxicity and bacterial inhibition. The molecular docking studies of synthesized compounds with the YmaH, ecKAS III protein and DNA dodecamer as the possible targets showed the complex $[Co(H_2B(Pz^{Me2})_2)_2]$ **(4)** bound to DNA with the binding energy of -6.05 kcal/mol and the complex $[Cu(H_2B(Pz^{Me2})_2)_2]$ **(2)** bound to YmaH and ecKAS III proteins with a binding energy of -7.48 kcal/mol and -6.47 kcal/mol, respectively and showed the highest binding energy among the studied compounds. Generally, our finding of *in vitro* experimental data and *in silico* molecular docking indicated the metal complexes displayed good biological properties for their activity response in antibacterial and anticancer studies in compare with free ligand, which can be explained by chelation theory. Our studies results suggest that the developed pyrazolyl borate based complexes can be the good candidates for further evaluations in biological areas.

Acknowledgments

The authors are grateful to the Azarbaijan Shahid Madani University for financial support of this study.

Author Contribution

MG: Participated in the design of the study, analysis, revising the manuscript, interpretation of data and drafting the manuscript. BS: Participated in the design of the study, analysis, revising the manuscript, interpretation of data, drafting the manuscript and final approval of the version to be published. OM: Participated in the design of the study, revising the manuscript, interpretation of data and final approval of the version to be published. EMA: Participated in the design of the study, analysis, revising the manuscript, interpretation of data and final approval of the version to be published.

Conflict of Interest

The authors report no conflicts of interest.

References

1. Pettinari C, Pettinari R. Metal derivatives of poly (pyrazolyl) alkanes: II. Bis (pyrazolyl) alkanes and related systems. Coord. Chem Rev. 2005;249(5-6):663-91. doi:10.1016/j.ccr.2004.08.017
2. Trofimenko S. Recent advances in poly (pyrazolyl) borate (scorpionate) chemistry. Chem Rev. 1993;93(3):943-80. doi:10.1021/cr00019a006
3. Soellner J, Pinter P, Stipurin S, Strassner T. Platinum (ii) complexes with bis (pyrazolyl) borate ligands: Increased molecular rigidity for bidentate ligand systems. Angew Chem. 2021;60(7):3556-60. doi:10.1002/anie.202011927
4. Ossinger S, Naggert H, Bill E, Näther C, Tuczek F. Electronic structure, vibrational spectra, and spin-crossover properties of vacuum-evaporable iron (ii) bis (dihydrobis (pyrazolyl) borate) complexes with diimine coligands. Origin of giant raman features. Inorg Chem. 2019;58(19):12873-87. doi:10.1021/acs.inorgchem.9b01813
5. Abernethy RJ, Foreman MRS-J, Hill AF, Smith MK, Willis AC. Relative hemilabilities of H2B(az)2 (az = pyrazolyl, dimethylpyrazolyl, methimazolyl) chelates in the complexes [M(η-C3H5)(CO)2{H2B(az)2}] (M = Mo, W). Dalton Trans. 2020;49(3):781-96. doi:10.1039/C9DT03744F
6. Fujisawa K, Ono T, Ishikawa Y, Amir N, Miyashita Y, Okamoto K-i, et al. Structural and electronic differences of copper (i) complexes with tris (pyrazolyl) methane and hydrotris (pyrazolyl) borate ligands. Inorg Chem. 2006;45(4):1698-713. doi:10.1021/ic051290t
7. Marques N, Sella A, Takats J. Chemistry of the lanthanides using pyrazolylborate ligands. Chem Rev. 2002;102(6):2137-60. doi:10.1021/cr010327y
8. Santini C, Pellei M, Lobbia GG, Papini G. Synthesis and properties of poly (pyrazolyl) borate and related boron-centered scorpionate ligands. Part a: Pyrazole-based systems. Mini Rev Org Chem. 2010;7(2):84-124. doi:10.2174/157019310791065519
9. Sadr MH, Mohammadi VM, Soltani B, Jamshidi-Ghaleh K, Mousavi SZ. Nonlinear optical responses of $MoS_4Cu_4(Pz^{me3})_6Cl_2$ under low power CW He-Ne laser excitation. Optik. 2016; 127(15):6050-5. doi:10.1016/j.ijleo.2016.04.051
10. Soltani B, Sadr MH, Engle JT, Ziegler CJ, Joo SW, Hanifehpour Y. Synthesis and structural characterization of three dinuclear copper(II) complexes incorporating pyrazolyl-derived ligands. Transit Met Chem. 2012; 37(8):687-94. doi:10.1007/s11243-012-9639-7
11. Manna F, Chimenti F, Fioravanti R, Bolasco A, Secci D, Chimenti P, et al. Synthesis of some pyrazole derivatives and preliminary investigation of their affinity binding to p-glycoprotein. Bioorg Med Chem Lett. 2005;15(20):4632-5. doi:10.1016/j.bmcl.2005.05.067
12. Monteiro M, Lechuga G, Lara L, Souto B, Viganó M, Bourguignon S, et al. Synthesis, structure-activity relationship and trypanocidal activity of pyrazole-imidazoline and new pyrazole-tetrahydropyrimidine hybrids as promising chemotherapeutic agents for chagas disease. Eur J Med Chem. 2019;182:111610. doi:10.1016/j.ejmech.2019.111610
13. Keter FK, Darkwa J. Perspective: The potential of

pyrazole-based compounds in medicine. Biometals. 2012;25(1):9-21. doi:10.1007/s10534-011-9496-4

14. Ghorbanpour M, Soltani B, Ziegler CJ, Jamshidi-Ghaleh K. Novel pyrazolate-bridged binuclear Ni (ii), Cu (ii) and Zn (ii) complexes: Synthesis, x-ray crystal structure and nonlinear optical studies. Inorg Chim Acta. 2021;514:119957. doi:10.1016/j.ica.2020.119957
15. Rahimizadeh M, Pordel M, Bakavoli M, Rezaeian S, Sadeghian A. Synthesis and antibacterial activity of some new derivatives of pyrazole. World J Microbiol Biotechnol. 2010;26(2):317-21. doi:10.1007/s11274-009-0178-0
16. Akbas E, Berber I, Sener A, Hasanov B. Synthesis and antibacterial activity of 4-benzoyl-1-methyl-5-phenyl-1h-pyrazole-3-carboxylic acid and derivatives. Farmaco. 2005;60(1):23-6. doi:10.1016/j.farmac.2004.09.003
17. Bandgar BP, Gawande SS, Bodade RG, Gawande NM, Khobragade CN. Synthesis and biological evaluation of a novel series of pyrazole chalcones as anti-inflammatory, antioxidant and antimicrobial agents. Bioorg Med Chem. 2009;17(24):8168-73. doi:10.1016/j.bmc.2009.10.035
18. Hassan SY. Synthesis, antibacterial and antifungal activity of some new pyrazoline and pyrazole derivatives. Molecules. 2013;18(3):2683-711. doi:10.3390/molecules18032683
19. Bouabdallah I, M'Barek LA, Zyad A, Ramdani A, Zidane I, Melhaoui A. Anticancer effect of three pyrazole derivatives. Nat Prod Res. 2006;20(11):1024-30. doi:10.1080/14786410600921441.
20. Xu Y, Shi Y, Lei F, Dai L. A novel and green cellulose-based schiff base-cu (ii) complex and its excellent antibacterial activity. Carbohydr Polym. 2020;230:115671. doi:10.1016/j.carbpol.2019.115671.
21. Chang EL, Simmers C, Knight DA. Cobalt complexes as antiviral and antibacterial agents. Pharmaceuticals. 2010;3(6):1711-28. doi:10.3390/ph3061711
22. Chohan ZH, Rauf A, Noreen S, Scozzafava A, Supuran CT. Antibacterial cobalt (II), nickel (II) and zinc (II) complexes of nicotinic acid-derived schiff-bases. J Enzyme Inhib Med Chem. 2002;17(2):101-6. doi:10.1080/14756360290024209.
23. Soltani B, Ghorbanpour M, Ziegler CJ, Ebadi-Nahari M, Mohammad-Rezaei R. Nickel (II) and cobalt (II) complexes with bidentate nitrogen-sulfur donor pyrazole derivative ligands: Syntheses, characterization, x-ray structure, electrochemical studies, and antibacterial activity. Polyhedron. 2020;180:114423. doi:10.1016/j.poly.2020.114423.
24. Kirschner S, Wei Y-K, Francis D, Bergman JG. Anticancer and potential antiviral activity of complex inorganic compounds. J Med Chem. 1966;9(3):369-72. doi:10.1021/jm00321a026.
25. Ghorbanpour M, Soltani B, Molavi O, Shayanfar A, Mehdizadeh Aghdam E, Ziegler CJ. Copper (II) complexes based bis (pyrazolyl) borate derivatives as efficient anticancer agents: Synthesis, characterization, x-ray structure, cytotoxicity, molecular docking and qsar studies. Chem Pap. 2022. doi:10.1007/s11696-022-02288-9
26. Hossaini Sadr M, Soltani B, Engle JT, Ziegler CJ, Taheri M. Bis [bis (3, 5-dimethyl-1h-pyrazol-1-yl) borato] cobalt (II). Acta Crystallogr A. 2011;67(Pt 7):m866. doi:10.1107/S1600536811020976
27. Sanla-Ead N, Jangchud A, Chonhenchob V, Suppakul P. Antimicrobial activity of cinnamaldehyde and eugenol and their activity after incorporation into cellulose-based packaging films. Packag Technol Sci. 2012;25(1):7-17. doi:10.1002/pts.952
28. Gopikrishna V, Kandaswamy D, Jeyavel RK. Comparative evaluation of the antimicrobial efficacy of five endodontic root canal sealers against *Enterococcus faecalis* and *Candida albicans*. J Conserv Dent. 2006;9(1):2. doi:10.4103/0972-0707.41303
29. Ermakova S, Sokolova R, Kim S-M, Um B-H, Isakov V, Zvyagintseva T. Fucoidans from brown seaweeds sargassum hornery, eclonia cava, costaria costata: Structural characteristics and anticancer activity. Appl Biochem Biotechnol. 2011;164(6):841-50. doi:10.1007/s12010-011-9178-2.
30. Franco-Molina MA, Mendoza-Gamboa E, Sierra-Rivera CA, Gómez-Flores RA, Zapata-Benavides P, Castillo-Tello P, et al. Antitumor activity of colloidal silver on mcf-7 human breast cancer cells. J Exp Clin Cancer Res. 2010;29(1):148. doi:10.1186/1756-9966-29-148
31. Dong K, Liu Z, Li Z, Ren J, Qu X. Hydrophobic anticancer drug delivery by a 980 nm laser-driven photothermal vehicle for efficient synergistic therapy of cancer cells in vivo. Adv Mater. 2013;25(32):4452-8. doi:10.1002/adma.201301232
32. Zha G-F, Leng J, Darshini N, Shubhavathi T, Vivek H, Asiri AM, et al. Synthesis, sar and molecular docking studies of benzo [d] thiazole-hydrazones as potential antibacterial and antifungal agents. Bioorganic Med Chem Lett. 2017;27(14):3148-55. doi:10.1016/j.bmcl.2017.05.032
33. Gupta RK, Sharma G, Pandey R, Kumar A, Koch B, Li P-Z, et al. DNA/protein binding, molecular docking, and in vitro anticancer activity of some thioether-dipyrrinato complexes. Inorg Chem. 2013;52(24):13984-96. doi:10.1021/ic401662d
34. Cheng K, Zheng Q-Z, Qian Y, Shi L, Zhao J, Zhu H-L. Synthesis, antibacterial activities and molecular docking studies of peptide and schiff bases as targeted antibiotics. Bioorg Med Chem. 2009;17(23):7861-71. doi:10.1016/j.bmc.2009.10.037
35. Cheng K, Xue J-Y, Zhu H-L. Design, synthesis and antibacterial activity studies of thiazole derivatives as potent ecKAS III inhibitors. Bioorg Med Chem Lett. 2013;23(14):4235-8. doi:10.1016/j.bmcl.2013.05.006
36. Dubey N, Sharma MC, Kumar A, Sharma P. A click chemistry strategy to synthesize geraniol-coupled 1, 4-disubstituted 1, 2, 3-triazoles and exploration of their

microbicidal and antioxidant potential with molecular docking profile. Med Chem Res. 2015;24(6):2717-31. doi:10.1007/s00044-015-1329-5

37. Pages BJ, Ang DL, Wright EP, Aldrich-Wright JR. Metal complex interactions with DNA. Dalton Trans. 2015;44(8):3505-26. doi:10.1039/C4DT02700K
38. Haribabu J, Jeyalakshmi K, Arun Y, Bhuvanesh NS, Perumal PT, Karvembu R. Synthesis, DNA/protein binding, molecular docking, DNA cleavage and in vitro anticancer activity of nickel (ii) bis (thiosemicarbazone) complexes. RSC Adv. 2015;5(57):46031-49. doi:10.1039/C5RA04498G
39. Shahabadi N, Falsafi M, Maghsudi M. DNA-binding study of anticancer drug cytarabine by spectroscopic and molecular docking techniques. Nucleosides Nucleotides Nucleic Acids. 2017;36(1):49-65. doi:10.1080/15257770.2016.1218021
40. Rai A, Gupta TK, Kini S, Kunwar A, Surolia A, Panda D. Cxi-benzo-84 reversibly binds to tubulin at colchicine site and induces apoptosis in cancer cells. Biochem Pharmacol. 2013;86(3):378-91. doi:10.1016/j.bcp.2013.05.024
41. Ermakova E. Structural insight into the glucokinase-ligands interactions. Molecular docking study. Comput Biol Chem. 2016;64:281-96. doi:10.1016/j.compbiolchem.2016.08.001
42. Arunadevi A, Porkodi J, Ramgeetha L, Raman N. Biological evaluation, molecular docking and DNA interaction studies of coordination compounds gleaned from a pyrazolone incorporated ligand. Nucleosides Nucleotides Nucleic Acids. 2019;38(9):656-79. doi:10.1080/15257770.2019.1597975
43. Panchal PK, Parekh HM, Pansuriya PB, Patel MN. Bactericidal activity of different oxovanadium (IV) complexes with schiff bases and application of chelation theory. J Enzyme Inhib Med Chem. 2006;21(2):203-9. doi:10.1080/14756360500535229
44. Raman N, Thalamuthu S, Dhaveethuraja J, Neelakandan M, Banerjee S. DNA cleavage and antimicrobial activity studies on transition metal (II) complexes of 4-aminoantipyrine derivative. J Chil Chem Soc. 2008;53(1):1439-43. doi:10.4067/S0717-97072008000100025
45. Malanovic N, Lohner K. Gram-positive bacterial cell envelopes: The impact on the activity of antimicrobial peptides. Biochim Biophys Acta. 2016;1858(5):936-46. doi:10.1016/j.bbamem.2015.11.004
46. Adam MSS, Elsawy H. Biological potential of oxo-vanadium salicylediene amino-acid complexes as cytotoxic, antimicrobial, antioxidant and DNA interaction. J Photochem Photobiol B: Biol. 2018;184:34-43. doi:10.1016/j.jphotobiol.2018.05.002
47. El-Sherif AA, Eldebss TM. Synthesis, spectral characterization, solution equilibria, in vitro antibacterial and cytotoxic activities of Cu(II), Ni(II), Mn(II), Co(II) and Zn(II) complexes with Schiff base derived from 5-bromosalicylaldehyde and 2-aminomethylthiophene. Spectrochim Acta A Mol Biomol Spectrosc. 2011;79(5):1803-14. doi:10.1016/j.saa.2011.05.062