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JSCS: Inorganic Chemistry Sub Editor

**Manuscript title: " Synthesis and characterization of copper (II) octaazamacrocyclic complexes with glycine derivatives. *In vitro* antiproliferative and antimicrobial evaluation of Cu(II) and Co(II) analogous"**

Dear Prof. MilošĐuran,

Thank you for useful comments and suggestions on the structure of our manuscript. Accordingly, we revised the manuscript and the detailed corrections are listed below at the point. Corrected sentences are highlighted in the text on the manuscript.

Responsee to the referees` comments

**Inorganic Chemistry Sub Editor**

Comment : Important data on metal complexes geometry are obtained by performing electronic spectra in the polycrystalline state. The structure can also be confirmed by recording mass spectra, X-ray diffraction. Could these determinations be made?

Response: Although the complexes were stable, all efforts to grow single crystals suitable for X-ray analysis failed. The crystals seemed shiny and were of regular size but by prolonged exposition to the open air they loose crystal solvent. Crystals quality was not good enough to perform X-ray single crystal analysis.

Comment: Attention to typographical mistakes (molecular formulas);

Response: Typographical mistakes were corrected in whole paper. All corrections are marked.

Page1/row12-13:"Two new complexes with general formula [Cu2(L)tpmc](ClO4)3•nH2O (L = *N*-methylglycine/*N*,*N*-dimethylglycine), tpmc = *N*,*N’*,*N’’*,*N’’’*-tetrakis(2-pyridylmethyl)-1,4,8,11-tetraazacyclotetradecane) were isolated and ..." is replaced with the following: "Two new complexes with general formula [Cu2(L)tpmc](ClO4)3•nH2O (tpmc = *N*,*N’*,*N’’*,*N’’’*tetrakis(2-pyridylmethyl)-1,4,8,11-tetraazacyclotetradecane, L = *N*-methylglycine, n=3; L=*N*,*N*-dimethylglycine, n = 2) ...".

Page 13/row356-357: "Синтетисана су два нова комплекса општe формулe [Cu2(L)tpmc](ClO4)3•nH2O (L = *N*-метилглицин/*N*,*N*-диметилглицин), tpmc = *N*,*N’*,*N’’*,*N’’’*тетракис(2-пиридилметил)-1,4,8,11-тетраазациклотетрадекан))" is replaced with the following:" Синтетисана су два нова комплекса општe формулe [Cu2(L)tpmc](ClO4)3•nH2O (tpmc = *N*,*N’*,*N’’*,*N’’’*тетракис(2-пиридилметил)-1,4,8,11-тетраазациклотетрадекан, L = *N*-метилглицин, n= 3; L= *N*,*N*-диметилглицин, n =2).... "

Comment: English could need some improvement.

Response: The manuscript was corrected and the english language was improved.

Some expression was corrected:

|  |  |  |
| --- | --- | --- |
| Page/row | Expression/word used | Correct expression/word |
| 1 - title | characterization copper (II) octaazamacrocyclic complexes | characterization of copper (II) octaazamacrocyclic complexes |
| 1- title | evaluation Cu(II) and Co(II) analogous | evaluation of Cu(II) and Co(II) analogous |
| 2/35 | cancer continue to | cancer rates continue to |
| 2/43 | has been receiving significant interest. | received considerable attention. |
| 2/46 | A great number of | Numerous mixed-ligand |
| 2/56 | one or both oxygens | one or both oxygen atoms |
| 3/65 | Continuing this research | As a continuation of this research |
| 3/67 | were proposed on the basis of the spectral | were proposed based on spectral |
| 3/81 | were the products of Sigma-Aldrich | were obtained from Sigma-Aldrich |
| 3/87 | with continuous stirring | under continuous stirring |
| 3/92 | until the precipitation of the solid blue microcrystalline product | until the precipitation of the solid blue microcrystalline product occurred |
| 4/116 | were made | were prepared |
| 4/121 | by comparing it to 0.5 McFarland’s | by comparing it to 0.5 McFarland’s standard |
| 5/126 | Negative control for each plate | For negative control for each plate |
| 5/152 | after the cell adherence | after the cell adhesion |
| 6/173 | adjusted by control of the | adjusted by controlling the |
| 6/179 | are collected in Table I. | are presented in Table I. |
| 6/181 | agree with 1:3 type electrolytes | are in agreement with the values for1:3 type electrolytes |
| 6/182 | The magnetic moment values per Cu(II) complexes | The magnetic moment values for Cu(II) complexes |
| 7/210 | indicates the presence of | indicating the presence of |
| 8/229 | bands were shown to provide useful information | bands provide useful information |
| 9/266 | antimicrobial activity new complexes | antimicrobial activity of the new complexes |
| 10/273 | It can be explained by | This can be explained by |
| 11/ | can be explained by | could be explained by |
| 11/301 | are very interesting because they can be potent antitumor agents. | are very promissing as potential antitumor agents. |
| 12/330 | From all present data it is assumed that | From all the obtained data it could be concluded that |

Some sentences have been modified.

Page 4/row99: “The complexes at room temperature are well soluble in CH3OH and insoluble in H2O“

was replaced with:“At room temperature, the complexes are soluble in CH3OH and insoluble in H2O“

Page 5/row 126: “Negative control for each plate was medium used only.“ was replaced with: “For a negative control for each plate the medium was used only.“

Page6/row173: “The reaction conditions were carefully adjusted by control of the pH and temperature“

was replaced with: “The reaction conditions were carefully adjusted by controlling the pH and temperature.“

Page 9/row 255: “The participation of the nitrogen of co-ligand atom is excluded.“ was replaced with: “The participation of co-ligand's nitrogen atom is excluded.“

Page 10/row276: “Literature data show that bimetallic complexes of sarcosine with Zn(II) and Sn(IV) showed more activity against Gram-(+) bacterial strain than Gram-(-) bacteria.“was replaced with: “The literature data show that bimetallic complexes of sarcosine with Zn(II) and Sn(IV) are more active against Gram-(+) bacterial strain than against Gram-(-) bacteria.“

The section: page 10/row277-284: “Several studies have used a classification based on the MIC results to evaluate the antimicrobial activity new compounds as: good, MIC inferior to 100 μg/mL; moderate: MIC between 100 and 500 μg mL-1; weak: MIC between 500 and 1000 μg mL-1; and inactive when the MIC is superior to 1000 μg mL-1.32 This way, it was possible to evaluate the antimicrobial activity of the examined metal complexes as moderate (Table III). Further, when the MBC/MIC ratio is less than or equal to 4.0, the agent will be considered bactericidal, and when this relation is over 4.0 should be considered bacteriostatic. Examined compounds showed bactericidal effect to *S. aureus* and *B. subtilis.*“;

was replaced with: “Several studies have used a classification based on the MIC results to evaluate the antimicrobial activity of new compounds as: good, MIC less than 100 μg mL-1; moderate: MIC between 100 and 500 μg mL-1; weak: MIC between 500 and 1000 μg mL-1; and inactive when the MIC value is more than 1000 μg mL-1.32 In this way, it was possible to evaluate the antimicrobial activity of the examined metal complexes as moderate (Table III). Further, when the MBC/MIC ratio is less than or equal to 4.0, the investigated agent would be considered as bactericidal, and when this ratio is more than 4.0 should be considered as bacteriostatic.“

The section: page 10/row 291-297:“The obtained IC50 values are given in Table IV along with the activity of cisplatin as referent cytostatic. All four compounds have promoted significant decreases in the metabolic activity of the HeLa, K562, MDA-MB-453, and MRC-5,which occurred in a dose-dependent fashion (cell survival, *S* vs concentration of compounds, Fig 2). The IC50 values (concentration of compounds that induced a 50% decrease in cell survival) of the complexes were in the range of 8.80–66.1 µM against four tested cell lines, while for cisplatin lied in the range 5.82–8.63 µM (Table IV).“;

was replaced with: “The obtained IC50 values (concentration of compounds that induced a 50% decrease in cell survival) are given in Table IV along with the activity of cisplatin as the referent cytostatic drug. All four compounds have promoted significant decrease in the metabolic activity of the HeLa, K562, MDA-MB-453, and MRC-5, which occurred in a dose-dependent manner (cell survival, *S* vs concentration of compounds, Fig 2). The IC50 values of the complexes were in the range of 8.80–66.1 µM against four tested cell lines, while for cisplatin were in the range of 5.82–8.63 µM (Table IV).“

Page 11/row 303: “In the Table IV it can be seen that the values for IC50 on the cell line MRC-5 is as close as those obtained for cisplatin.“ was replaced with : “The data presented in the Table IV, show that IC50 values for the MRC-5 cell line are similar to the ones obtained for cisplatin“

Page11/row 309: “Due to similarity of the structure of Co (II) and Cu (II) complexes, different antimicrobial activity can be explained by electronic and steric factors11/“was replaced with : “Due to the similarity of Co (II) and Cu (II) complexes structure, the different antimicrobial activity could be explained by electronic and steric factors.“

The sentence in line 267 : “The Cu (II) and Co (II) complexes with amino acid derivatives, did not show activity against the Gram (-) bacteria and the yeast *C. albicans*. “ was moved after table II.

We hope that our paper is now suitable for publication as original scientific paper in Journal of the Serbian Chemical Society.

August 10th, 2019. Sincerely yours,

Dr Branka Dražić