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Bis(nitrobenzoato)copper(II) Complexes with Nicotinamide, Preparation, Structure and Properties

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Abstract

Reaction of copper (II) salts with nitrobenzoic acids in the presence nicotinamide the nitrobenzoatocopper(II) complexes with nicotinamide are formed. Their composition, spectral, magnetic properties and structure have been studied together with their antimicrobial activity. The stoichiometry of the final product is influenced by the type of nitrobenzoato ligand and 3-nitrobenzoato anion has caused formation of the complex of unusual stoichiometry $[Cu(3\text{-}NO_2bz)_2(nia)_1(H_2O)_2]$ was prepared and studied. The complexes of general formula $[Cu(x\text{-}NO_2bz)_2(nia)_2(H_2O)_y]$ (where x-NO_2 = 2-NO_2, 4-NO_2 and y = 2; or x-NO_2 = 3,5-$(NO_2)_2$ and y = 1) were obtained for other copper(II) nitrobenzoates. Rather unusual stoichiometry of $[Cu(3\text{-}NO_2bz)_2(nia)_1(H_2O)_2]$ has caused unusual magnetic behaviour and the antimicrobial activity different from those ones of composition $[Cu(x\text{-}NO_2bz)_2(nia)_2(H_2O)_y]$.

Keywords: copper complexes, nitrobenzoato ligand, nicotinamide ligand, crystal structure, antimicrobial activity

Introduction

The biomedical activities, chemical and industrial versatility of copper complexes have been the subject of intensive study over the long time. The copper complexes with some ligands can exhibit better biological activity compared to non-coordinated organic ligands e.g. copper(II) complex of 3,5-dimethylpyrazol have shown higher antimicrobial activity (testing on *Staphylococcus aureus*, *Escherichia coli* and *Candida albicans*) than 3,5-dimethylpyrazol (Sokolík 1998). Copper(II) complexes of carboxylato ligands have been the subject of a large number of research studies (Kato 1988, Kato 1998, Melník 1998), presumably, due to the various coordination modes of carboxylato ligands.

Nicotinamide, known also as niacin derivative, or vitamin B_3 component, (nia) is one of nitrogen donor ligand that was in treatment of various skin diseases, e.g. atopic eczema, (Namazi 2003, Makara 1996) psoriasis, or skin cancer (Catz 2005, Gensler 1999, Gensler 1997) and biological activity of its copper complexes were also studied (Kozlevcar 1999).

In the present paper, we report as a part of our investigation of metal ion–drug interaction the complexation of x-nitrobenzoate (x-NO_2bz) (where x = 2, 3, or 4) and/or 3,5-dinitrobenzoate (3,5-$(NO_2)_2$bz) in the presence of nicotinamide (nia). The complexes of [Cu(2-NO_2bz)$_2$(nia)$_2$(H_2O)$_2$] (**1**), [Cu(3-NO_2bz)$_2$(nia)$_1$(H_2O)$_2$] (**2**), [Cu(4-NO_2bz)$_2$(nia)$_2$(H_2O)$_2$] (**3**), and [Cu(3,5-$(NO_2)_2$bz)$_2$(nia)$_2$(H_2O)$_1$] (**4**) have been prepared, and their structure have been solved. Based on the molecular structure, the electronic, infrared, EPR and magnetic data, as well as biological activity are discussed.

Experimental

Preparation

All complexes were prepared in the same way, by reaction of an aqueous solution of copper (II) acetate (1 mmol) with nicotinamide (2 mmol) followed by addition of the appropriate acid (2 mmol). The reaction mixture was stirred until the precipitation of light blue product was finished. The microcrystalline product was filtered off, washed with small portion of water and dried at ambient temperature. The light blue mother liquids obtained after filtration were left for crystallization and blue crystals suitable for X-ray structure determination were obtained after few days. The compositions of powder products were according the elemental analysis the same as the crystals ones (proved by X-ray structure analysis).

Analytical methods

Carbon, hydrogen and nitrogen analysis were carried out on a CHNSO FlashEA™ 1112 (ThermoFinnigan) elemental analyzer. Copper was determined by electrolysis of water solution obtained by the sample mineralization with a mixture of sulfuric acid and potassium peroxodisulfate.

Physical measurements

Electronic spectra (190 – 1100 nm) of the complexes were measured in Nujol suspension with a SPECORD 200 (Carl Zeiss Jena) spectrophotometer, and infrared spectrum in the region of 4000 – 100 cm^{-1} were measured with a Nicolet MAGNA 750 IR spectrometer using both KBr pelet and Nujol suspension technics for 4000 – 400 cm^{-1} region. EPR spectra of powdered sample were recorded at room temperature and at 77 K, on the spectrometer Bruker ESP 300, operating at X-band equipped with an ER 035M Bruker NMR gaussmeter and a HP 5350B Hewlett Packard microwave frequency counter. Magnetic susceptibility measurements in the temperature range of 1.8 – 300 K were carried out on powdered samples of complexes, at the magnetic field 0.5 T, using a Quantum Design SQUID Magnetometer (type MPMS-XL5). Corrections for diamagnetism of the constituting atoms were calculated using the Pascal constants (König 1966), the value of $60\cdot10^{-6}$ cm^3mol^{-1} was used as the temperature-independent paramagnetism of copper(II) ion. The effective magnetic moments were calculated from the susceptibility data.

X-ray crystallography

Data collection and cell refinement of presented complexes were carried out using Kuma KM4 diffractometer (Kuma Diffraction 1998). Intensity data were corrected for Lorenz and polarization factors. The structure was solved by the heavy atom method with *SHELX*86 (Sheldrick 2008), and subsequent Fourier synthesis using *SHELXS*93 (Sheldrick 2008). Geometrical analysis was performed using *SHELXL*97 (Sheldrick 2008) and the structure was drawn using Mercury program.

Antimicrobial assay

Antibacterial and anti-yeasts activities of the tested compounds were evaluated by a micro-dilution method (Jantová 1995) using G^+ bacteria *S. aureus* CCM 3953, G^- bacteria *Escherichia coli* CCM 3988 (both from the Czech Collection of Microorganisms, Masaryk University, Brno, Czech Republic) and the yeasts *Candida parapsilosis* (from the Laboratory of Medical Mycology, Postgraduate Medical Institute, Bratislava, Slovak Republic). The effect of these compounds on filamentous fungi *Rhizopus oryzae* (from the Collection of Microorganisms of Department of Biochemistry and Microbiology, Faculty of Chemical and Food Technology, Slovak University of Technology, Bratislava, Slovak Republic), *Aspergillus fumigatus* CCM F-373 (from the Czech Collection of Microorganisms, Masaryk

University, Brno, Czech Republic) and *Microsporum gypseum* (from the Laboratory of Medical Mycology, Postgraduate Medical Institute, Bratislava, Slovak Republic) was observed by macro-dilution technique on solidified broth medium during static culturing (Dudová 2002).

Chromatographically pure compounds were dissolved in DMSO; its final concentration never exceeded 1.0% vol. in either control or treated samples. Concentration of tested compounds was in the range of 0.01 (for copper complexes) and 0.5 (for free ligands), respectively, to 3.0 mmol L^{-1} in all experiments.

The antimicrobial activity was characterized by the IC_{50} values (concentration of a derivative which in comparison with the control inhibits the growth of microorganisms to 50%) and MIC values (minimal inhibitory concentration of a compound which inhibits microbial growth by 100%). The IC_{50} and MIC values were read from toxicity curves. MIC experiments on subculture dishes were used to assess the minimal microbicidal concentration (MMC). Subcultures were prepared separately in Petri dishes containing appropriate agar medium and incubated at 30 °C for 48 h (bacteria, yeasts) and 25 °C for 96 h (filamentous fungi). The MMC value was taken as the lowest concentration which showed no visible growth of microbial colonies on the subculture dishes.

Results and Discussion

A series including many similar complexes, obtained by the systematic approach to synthesis, offer a good opportunity to study systematically similarity and differences between individual members of the group to find out the most influencing factors for different properties of the complexes.

Structure and Composition

The structural study and analysis of the composition of the prepared complexes could give the information about the main preferences of used ligands to preferentiate some structural type of the complex. Within the group of the nitrobenzoatocomplexes with nicotinamide the same composition and very similar structures and properties were found for the complexes $[Cu(2\text{-}NO_2bz)_2(nia)_2(H_2O)_2]$ (**1**) (Fig. 1) (Stachová 2006) and $[Cu(4\text{-}NO_2bz)_2(nia)_2(H_2O)_2]$ (**3**) (Fig. 2).

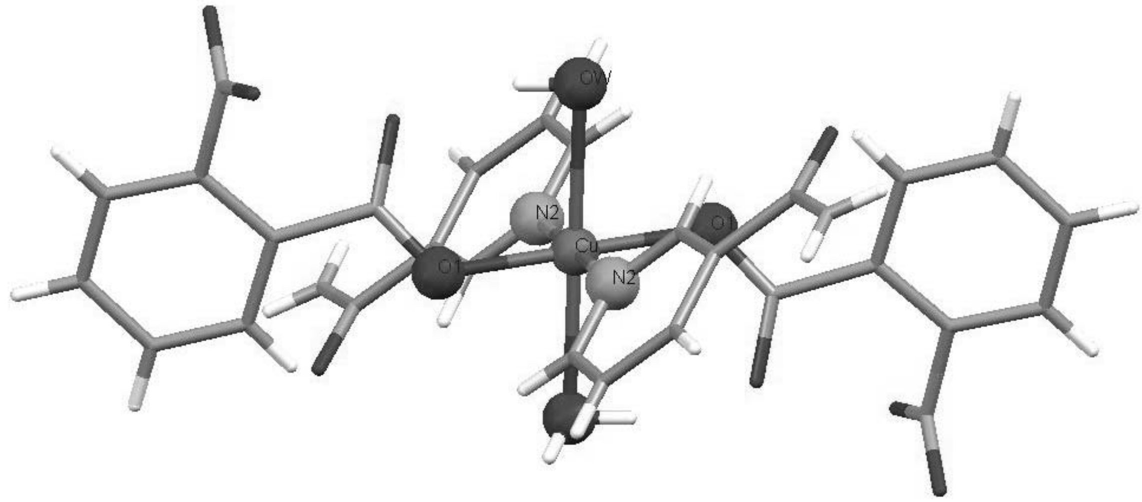

Fig. 1. View of complex [Cu(2-NO_2bz)$_2$(nia)$_2$(H_2O)$_2$] (1) structure

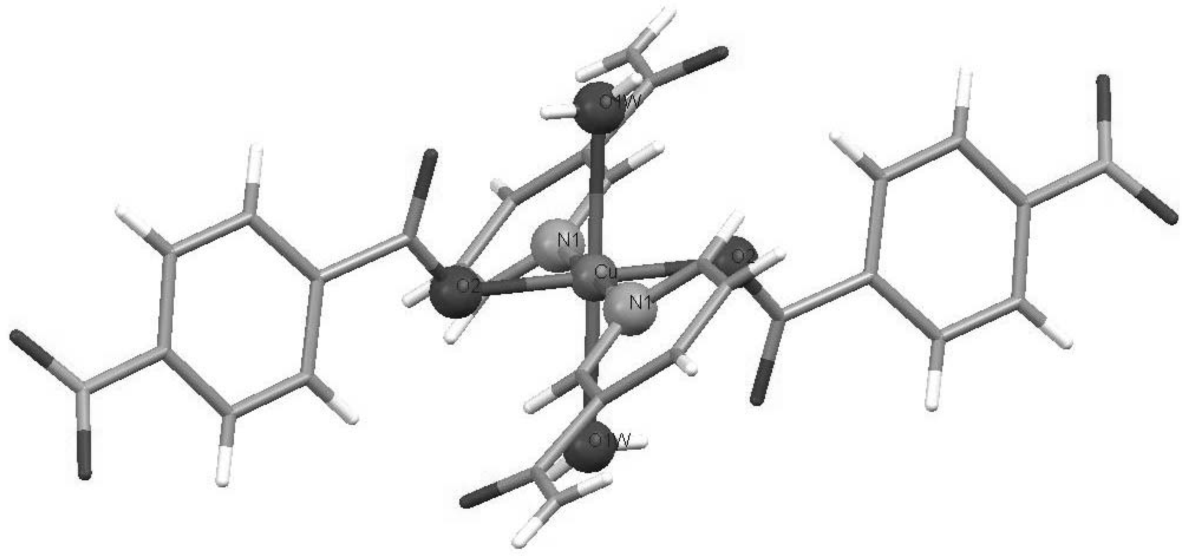

Fig. 2. View of complex [Cu(4-NO_2bz)$_2$(nia)$_2$(H_2O)$_2$] (3) structure

The complex [Cu(3-NO_2bz)$_2$(nia)$_1$(H_2O)$_2$] (**2**) (Fig. 3) differs from previous two ones in the composition, as well as in coordination polyhedra structure. The most striking difference of **2**, in comparison to previous ones, is the presence of nicotinamide in Cu : nia molar ratio 1 : 1 in this nonpolymeric and mononuclear copper (II) complex. In spite of great experimental effort to obtain some 3-nitrobenzoatocopper(II) complexes with different Cu : nia molar ratio the complex [Cu(3-NO_2bz)$_2$(nia)$_1$(H_2O)$_2$] (**2**) is the only product obtainable from this system.

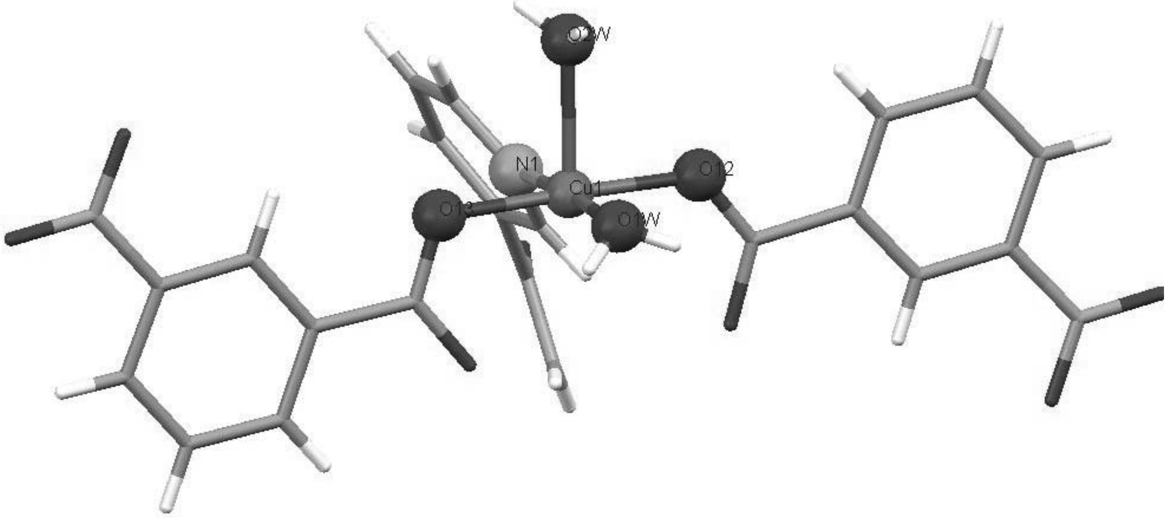

Fig. 3. View of complex [Cu(3-NO_2bz)$_2$(nia)$_1$(H_2O)$_2$] (2) structure

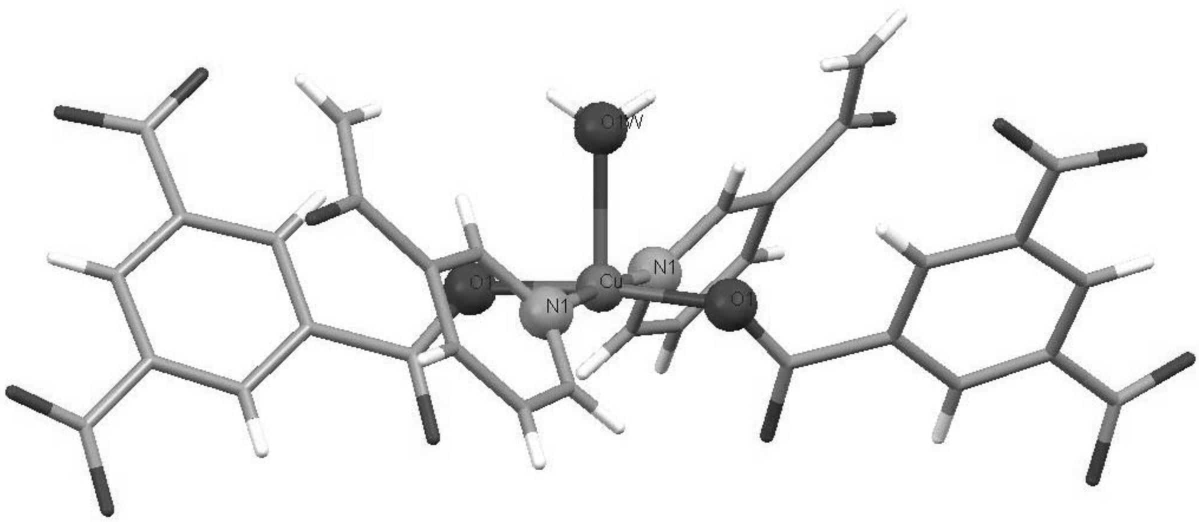

Fig. 4. View of complex [Cu(3,5-(NO_2)$_2$bz)$_2$(nia)$_2$(H_2O)$_1$] (4) structure

The complex [Cu(3,5-(NO_2)$_2$bz)$_2$(nia)$_2$(H_2O)$_1$] (**4**) differs from the complexes **1** and **2** but only in solvent molecule presence within its copper(II) coordination polyhedron. There are some other differences in solid state supramolecular structure.

Physical properties

All presented complexes are of light blue color and their infrared spectral properties correspond to the ligand bonding mode. However, the EPR spectra have shown the small but significant difference between $[Cu(3\text{-}NO_2bz)_2(nia)_1(H_2O)_2]$ (**2**) complex in comparison to those ones of the complexes **1**, **3** and **4**. In addition to that, the complex **2** exhibits different magnetic behaviour in the temperature dependence of the magnetic susceptibility (Fig. 5).

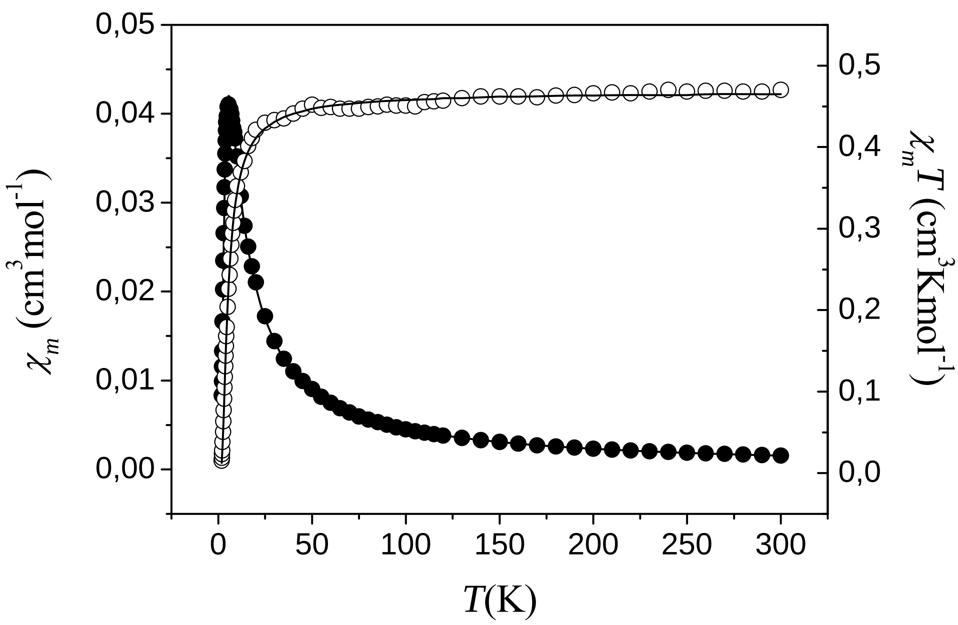

Fig. 5. Temperature dependence of χm (●) and χmT (○) of (2)

Antimicrobial activities

The results of determination of antimicrobial activity (characterized by the IC_{50} and MIC values; $mmol \cdot L^{-1}$) of the complexes from **1-4** and some other structurally not characterized complexes, as well as free nitrobenzoic acids and their salts containing nicotinamidium cation, are summarized in Table 1.

While the free nitrobenzoic acids and their nicotiamidium salts have not influenced the growth of model bacteria, yeasts and filamentous fungi with only a few exceptions (IC_{50} > 3.0 mmol L^{-1}), IC_{50} values for the copper(II) nitrobenzoates and copper(II) complexes with nicotinamide have been found in the wider range of 0.3-3.0 mmol L^{-1}, and it can be supposed that the presence of copper atom in the tested compound enhances in the most cases its antimicrobial activity. On the other hand it should be stressed that presence of nicotinamide in the most of nitrobenzoatocopper(II) complexes under study slightly decreases the inhibition activities. There is only one exception from this statement – the complex of unusual composition $[Cu(3\text{-}NO_2bz)_2(nia)_1(H_2O)_2]$ (**2**) and its analogue showing smaller water

Table 1 Antimicrobial activity of tested compounds characterized by IC_{50} and MIC ($mmol \cdot L^{-1}$)

Compounds	Bacteria				Yeasts		Filamentous fungi					
	S. aureus		*E. coli*		*C. parapsilosis*		*R. oryzae*		*A. fumigatus*		*M. gypseum*	
	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC
2-NO_2bzH	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3
($C_6H_5ON_2$)(2-NO_2bz)	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3
[Cu_2(2-NO_2bz)$_4$(H_2O)$_2$].2H_2O	0.7	3^s	>3	>3	0.8	2^s	1.1	2^s	1.7	>3	2.3	3^c
[Cu(2-NO_2bz)$_2$(nia)$_2$(H_2O)$_2$] (**1**)	1.5	>3	>3	>3	1.4	3^s	1.5	3^s	1.8	>3	2.6	>3
3-NO_2bzH	>3	>3	>3	>3	>3	>3	>3	>3	2.7	>3	>3	>3
($C_6H_5ON_2$)(3-NO_2bz)	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3
Cu(3-NO_2bz)$_2$(H_2O)$_2$	1.8	>3	>3	>3	1.9	>3	2.5	3^s	2.5	>3	>3	>3
[Cu(3-NO_2bz)$_2$(nia)$_1$(H_2O)$_2$] (**2**)	2.4	>3	>3	>3	2.5	>3	2.2	3^c	>3	>3	2.9	>3
Cu(3-NO_2bz)$_2$(nia)$_1$(H_2O)$_1$	0.8	>3	>3	>3	1.5	>3	0.9	2^s	2	>3	1.3	2^c

^{s} microbistatic effect, ^{c} microbicide effect

Table 1. (Continued)

Compounds	Bacteria				Yeasts		Filamentous fungi					
	S. aureus		*E. coli*		*C. parapsilosis*		*R. oryzae*		*A. fumigatus*		*M. gypseum*	
	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC	IC_{50}	MIC
3,5-$(NO_2)_2$bzH	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	2.1	3^c
$(C_6H_5ON_2)$(3,5-$(NO_2)_2$bz)	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3
Cu(3,5-$(NO_2)_2$bz$)_2(H_2O)_2$	1.4	>3	>3	>3	2.2	>3	1.5	3^s	2.5	>3	2.1	3^c
[Cu(3,5-$(NO_2)_2$bz$)_2$(nia$)_2(H_2O)_1$] (**4**)	1.3	>3	2.1	>3	2.0	>3	0.8	3^s	2.5	>3	>3	>3
Cu(3,5-$(NO_2)_2$bz$)_2$(nia$)_2(H_2O)_2$	1.5	>3	>3	>3	2.7	>3	1.4	3^s	2.4	>3	2.3	3^c
4-NO_2bzH	>3	>3	>3	>3	>3	>3	>3	>3	>3	>3	1.4	2^c
$(C_6H_5ON_2)$(4-NO_2bz)	>3	>3	>3	>3	>3	>3	>3	>3	2.6	>3	1.4	2^c
Cu_2(4-NO_2bz$)_4$(4-NO_2bz$)_2$	$<0.5^c$	$<0.5^c$	1	3^c	0.3	1^c	0.5	1^s	1.2	2^s	0.7	1^c
Cu_2(4-NO_2bz$)_4$(EtOH$)_2$	2.5	>3	2.1	>3	1.8	3^c	0.6	1^s	2.3	3^s	0.7	1^c
[Cu(4-NO_2bz$)_2$(nia$)_2(H_2O)_2$] (**3**)	>3	>3	>3	>3	1.5	2^c	1.1	2^c	1.1	2^c	0.7	1^c

^{s} microbistatic effect, ^{c} microbicide effect content which exhibit greater inhibition activities than the corresponding copper 3-nitrobenzoate. The high inhibition activities of both complexes were probably caused by their unusual Cu : nia = 1 : 1 stoichiometry.

The eukaryotic model fungi *C. parapsilosis*, *R. oryzae*, *A. fumigatus* and *M. gypseum* were more sensitive to the tested compounds than prokaryotic *S. aureus* and *E. coli*, respectively. The growth of G^+ bacteria (*S. aureus*) was influenced more than the growth of G^- bacteria (*E. coli*).

Conclusion

In conclusion, we have prepared and characterized six nitrobenzoatocopper(II) complexes containing nicotinamide and four of them were structurally characterized. All complexes were tested for their antimicrobial activity and it was shown that the novel molecular complex $[Cu(3\text{-}O_2Nbz)_2(nia)_1(H_2O)_2]$ (**1**) of unusual composition that possessing square pyramidal structure of each copper(II) atom exhibit increased antimicrobial activity too. This increased activity is related to its unusual composition, structure and to its unusual magnetic behaviour. The other complexes containing nicotinamide have shown decreased antimicrobial activity in comparison to the corresponding complexes without nicotinamide ligand.

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