Potential Metallo-β-lactamase Biomimetic for Antimicrobial Resistance Study: Synthesis, characterization and reactivity study of Zn\textsuperscript{II} complex of the ligand dipicolylglycine.

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Abstract

Synthesis, characterization and reactivity studies were performed for the complex diaqua(bis(2-picolyl)glycinate)zinc(II) nitrate monohydrate (Zn-NNO). This complex is a potential model compound of metallo-β-lactamase (MβL), enzyme responsible for antibacterial resistance. Kinetics studies showed the compound is an effective catalyst of the hydrolysis of nitrocefin and the rates were compared to other complexes prepared by the research group. Gold(I) and gold(III) compounds were evaluated as inhibitors of MβL. Zn-NNO was used as model compound for the study of interaction between gold and zinc enzymes. Nuclear magnetic resonance and mass spectroscopy results suggested the coordination of gold to ligand without displacement of zinc ion.

Key words:
Antimicrobial resistance, metallo-β-lactamases, biomimetics.

Introduction

Antimicrobial resistance (AMR) is the ability of a microorganism to inhibit an antimicrobial activity and can compromise even common surgical procedures.\[1\] Metallo-β-lactamase (MβL) expression in bacteria can develop. This class of β-lactamase depend on Zn\textsuperscript{II} to catalyse hydrolysis of β-lactam antibiotics, like penicillin.\[2\] Hydrolytic activity of enzymes can be studied using mimetics instead of proteins, which are more sensitive to some experimental conditions. Although MβLs mimetics are already described, they do not reproduce the correct coordination sphere of Zn\textsuperscript{II}, solvent accessibility and polarity of MβLs catalytic sites.\[3\] Hence, synthesis, characterization and reactivity studies were performed for the complex diaqua(bis(2-picolyl)glycinate)zinc(II) nitrate monohydrate (Zn-NNO, Figure 1) and its hydrolytic activity was compared to those of the other complexes shown in Figure 1. It was also performed an interaction study with gold compounds, which are potential inhibitors of some zinc enzymes.\[3\]

![Figure 1. In order, structure of Zn-NNO, -NNS and -NNN.](image)

Results and Discussion

Zn-NNO was synthesized by mixing 2-pyridinecarboxaldehyde with glycine ethyl ester hydrochloride in 2.2.2-trifluoroethanol. Potassium carbonate was added to deprotonate the amino group and facilitate the attack to the aldehyde group. NaBH\textsubscript{4} was added to reduce the intermediate imine. The resultant ethyl N-(2-picolyl)glycinate was mixed with 2-pyridinecarboxaldehyde and NaBH\textsubscript{4}CN in acetic acid to afford a brown oil of ethyl ester of NNO. The next steps were executed as described by Aburafag et al. (1995)\[4\] and afforded Zn-NNO as a white amorphous powder. The structural characterization was performed by infrared spectroscopy, elemental analysis, \textsuperscript{1}H and \textsuperscript{13}H NMR and ESI-MS resulting in the following data for \textsuperscript{1}H NMR (250 MHz, D\textsubscript{2}O): δ = 8.678 ppm (d, 2H\textsubscript{a}, py), 8.106 ppm (t, 2H\textsubscript{a}, py), 7.636 ppm (m, 2H\textsubscript{b} and 2H\textsubscript{c}, py), 4.408 ppm (t, 2NCH\textsubscript{3}-py), 3.531 ppm (s, NCH\textsubscript{2}CO). Mass spectra showed a peak (m/z = 320) corresponding to [Zn(NNO)]\textsuperscript{+}.

![Figure 2. Comparison of nitrocefin hydrolysis activity between Zn-NNO and other Zn\textsuperscript{II} complexes.](image)

Zn-NNO was mixed with K[AuCl\textsubscript{4}] or [Ph\textsubscript{3}PAuCl] in DMSO-d\textsubscript{6} and the resultant solution was analyzed by \textsuperscript{1}H NMR along some time. Similar experiment was conducted for \textsuperscript{31}P NMR and ESI-MS analysis, which showed the species [AuZn(NNO)(H\textsubscript{2}O)Cl\textsubscript{3}]\textsuperscript{-} (m/z = 679) and [AuZn(Ph\textsubscript{3})NNOCl]\textsuperscript{-} (m/z = 818).

Conclusions

Zn-NNO compared to Zn-NNS and Zn-NNN showed higher hydrolytic activity of nitrocefin and demonstrated the influence of coordination sphere over zinc ion reactivity. Analyses of gold compound (Au\textsubscript{II}[Au\textsubscript{III}]) and Zn-NNO mixture suggest the coordination of gold ion to the ligand without displacement of zinc.

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