CONJUGATED Zn(II) COMPLEX AS METALLO- β -LACTAMASE MODEL

Raphael H. Ribeiro*, Camilla Abbehausen.

Abstract

This work reports the synthesis, characterization and the interaction with a β -lactam compound of a Zn(II) complex containing imidazole and pyridil groups bearing a free carboxylate for peptide conjugation. The complex was able to interact with the carboxylate next to the β -lactam group of amoxicillin and the conjugation with peptide (glutathione as a model) was successful.

Key words:

metallo- β -lactamase, biomimetics, conjugated complexes.

Introduction

Since the discovery of penicillin in 1928 by Alexander Fleming the wide use of the β -lactams supported the treatment of many bacterial infections. A great part of the success of the use of these compounds is due to its low toxicity in humans. These drugs act by blocking the synthesis of the bacterial wall, causing cellular lysis. At the same time, these compounds exert great evolutionary pressure, leading to the selection of organisms resistant to the treatment with β -lactams. In this direction, the expression of metallo- β -lactamases (M β LS) is one of the mechanisms of bacterial resistance. M β LS are zinc-dependent enzymes capable of hydrolyzing the β -lactam groups, inactivating the antibacterial.

There are many metallic complexes described in the literature that acts as mimetics for M β LS, although they don't reproduce accurately the catalytic sites of the enzymes. In this work, we have synthesized and characterized a Zn(II) complex based on the structure of one of the most common binding sites of M β LS containing three histidine residues coordinated to a Zn(II) ion (Image 1) and the interaction test with a β -lactam commercial compound was evaluated. The conjugation with glutathione (a model peptide) was done in order to introduce a methodology to mimic the influence of the flexible protein loops around the active site, leading to a broader understanding of the mechanism of M β LS aiming to obtain better M β LS mimetics.

Image 1. The Zn-cNNN complex (left) and the glutathione conjugated complex Zn-NNN-Glu (right).

Results and Discussion

The Zn(II) complex, called here as Zn-cNNN, was synthesized from a nucleophilic substitution reaction between histidine and 2-(chloromethyl)pyridine in water, followed by the addition of zinc chloride in methanol, giving an off-white powder with 52% yield. Its characterization was performed with ^1H NMR, ^{13}C NMR and ESI-MS ($m/z=436,03$).

The conjugation was performed as described by Graf et al. (2012) forming Zn-NNN-Glu and its characterization as performed with ESI-MS ($m/z=732,2$).

The interaction between the Zn(II) complex and amoxicillin was verified by the shift of the ^1H NMR signals (from 4,72 to 4,90 ppm) associated to the hydrogen geminal to the carboxylate group present in the β -lactam indicating that the Zn(II) interacts with the carboxylate group (Image 2).

Image 2. ^1H NMR spectra of the interaction test between Zn-cNNN and amoxicillin in DMSO-d₆

Conclusions

The Zn-cNNN and its conjugated were successfully synthesized and the Zn(II) complex interaction test showed that the complex interacts with the β -lactam by its carboxylate group.

Acknowledgement

The authors thank CNPq and FAPESP (2017/12719-0) for financial support.

- Karsisiotis, A. I.; Damblon, C. F. and Roberts, G. C. K. *Metallomics* **2014**, 6, 1181.
 Daumann, L. J., Schenk, G. and Gahan, L. R. *Eur. J. Inorg. Chem.* **2014**, 2869.
 Kaminskaia, N. V., Spingler, B. and Lippard, S. J. *J. Am. Chem. Soc.* **2000**, 122, 6411.
 Chiu, Y. H. and Canary, J. W. *Inorg. Chem.* **2003**, 42, 5107.
 Graf, N., Mokhtari, T. E., Papayannopoulos, I. A. and Lippard, S. J. *J. Inorg. Biochem.* **2012**, 110, 58.
 Kaminskaia, N. V., Spingler, B. and Lippard, S. J. *J. Am. Chem. Soc.* **2001**, 123, 6555.