Danger lurking in the "unknowns": structure-to-function studies of hypothetical protein Bleg1_2437 from Bacillus lehensis G1 alkaliphile revealed an evolutionary divergent B3 metallo-beta-lactamase

ABSTRACT

The effectiveness of β-lactam antibiotics as chemotherapeutic agents to treat bacterial infections is gradually threatened with the emergence of antibiotic resistance mechanism among pathogenic bacteria through the production metallo-β-lactamase (MBL). In this study, we discovered a novel hypothetical protein (HP) termed Bleg1_2437 from the genome of alkaliphilic Bacillus lehensis G1 which exhibited MBL-like properties of B3 subclass; but evolutionary divergent from other circulating B3 MBLs. Domain and sequence analysis of HP Bleg1_2437 revealed that it contains highly conserved Zn2+-binding residues such as H54, H56, D58, H59, H131 and H191, important for catalysis, similar with the subclass B3 of MBL. Built 3-D Bleg1_2437 structure exhibited an αββα sandwich layer similar to the well-conserved global topology of MBL superfamily. Other features include a ceiling and floor in the model which are important for accommodation and orientation of β-lactam antibiotics docked to the protein model showed interactions at varying degrees with residues in the binding pocket of Bleg1_2437. Hydrolysis activity towards several β-lactam antibiotics was proven through an in vitro assay using purified recombinant Bleg1_2437 protein. These findings highlight the presence of a clinically important and evolutionary divergent antibiotics-degrading enzyme within the pools of uncharacterized HPs.

Keywords: Antibiotic resistance; Beta-lactam antibiotics; Hypothetical proein; Metallo-beta-lactamase; Bacillus lehensis G1