

BIOINFORMATICS AND PHYSICOCHEMICAL PROPERTIES OF CZCE PROTEIN IN BACTERIA

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ABSTRACT

This study investigated the bioinformatics of heavy metal resistance protein. Mega 6 was used to construct the divergence time and phylogenetic analysis of CzcC and Protparam was used to determine the amino acids, pI and Gravy of CzcE from different bacteria. The results showed the actual bacteria that contain the CzcE protein which can be useful for researchers that wants to work on this protein.

Keywords: CzcE, protein, bioinformatics, Organisms.

INTRODUCTION

Heavy metal resistance protein is a family of metal-binding proteins which are involved in resistance to heavy-metal ions. These proteins form a four-helix hooked hairpin consisting of two long alpha helices each flanked by a shorter alpha helix. CzcE is a periplasmic protein as indicated by pho A fusions. CzcE was purified and identified as a metal binding protein (Grosse et al., 2004). CzcE is a periplasm-located dimeric protein able to bind specifically 4 Cu-equivalent per dimer (Zoropoqui et al., 2008). The oxidized form of CzcE contains Cu (II) atoms, while the reduced form contains four Cu (I) atoms. The ability of CzcE to bind copper and to adapt its conformation to different copper oxidation states could be related to a role in copper sensing for this protein (Petit-Haertlein et al., 2010).

The aim of this study is to investigate evolutionary history of heavy metal resistance protein (CzcE) in certain bacteria having this protein.

METHODOLOGY

Construction of divergence time and phylogenetic tree for heavy metal resistance protein

Phylogenetic tree is a tree-like branch of the graph, used to describe the relationship between species. The phylogenetic tree constructed by comparing the values of biomolecule sequence differences is a molecular phylogenetic tree. Heavy metal resistance protei (CzcE) were selected to construct the respective protein phylogenetic tree. The proteins were first BLAST (<http://blast.ncbi.nlm.nih.gov/Blast.cgi>). Then the NCBI to download these protein FASTA format sequence files. Then the alignment, divergence time and molecular phylogenetic tree were constructed by MEGA 6.0, and the results were analyzed.

Physicochemical analysis of heavy metal resistance protein

The sequence of the heavy metal resistance proteins in different organisms were input into the ExPASy Protparam software, to give the protein parameters to predict the results of physical and chemical properties, including the number of amino acids, the total number of positive and negative charge of the protein contained in the residue with the average hydrophilic (Grand average of hydropathility, GRAVY) and the theoretical-isoelectric point of the protein (theoretical pI).

Result

The divergence time among the bacteria group showed that *Cupriavidus sp* (*C. metallidurans*, *C. gilardii*) (1-5) were the least diverge which means that they are the most primitive among the bacteria group considered. While *Ralstonia mannitolilytica* was the highest diverge which means it was the most advanced group of bacteria having the heavy metal resistance protein (CzcE) (Fig. 1).

For the phylogenetic tree, *Ralstonia insidiosa* showed the highest in the phylogenetic tree, which means it was the most advanced having CzcE protein while the *Cupriavidus metalluridans* showed the lowest, which means it was the most primitive having CzcE protein among the bacteria group (Fig.2).

For the physic-chemical parameters of CzcE in certain groups of bacteria, the pI value for all the bacteria group were within the alkaline range. This can explain why it the CzcE protein can bind copper (Petit-Haertlein et al., 2010). For Gravy value, *Ralstonia insidiosa* has the highest value of 0.375 while *Ralstonia mannitolilytica* has the lowest value of -0.001 (Table 1). This can explain why *Ralstonia mannitolilytica* was the most diverged group of the bacteria considered (Fig 1).

This study indicates the actual bacteria that contain the CzcE protein and the distance apart among them which can be useful for researchers that wants to work on this protein.

	1	2	3	4	5	6	7	8
[1]								
[2]	0.00							
[3]	0.00	0.00						
[4]	0.00	0.00	0.00					
[5]	0.00	0.00	0.00	0.00				
[6]	0.85	0.85	0.85	0.85	0.85			
[7]	1.00	1.00	1.00	1.00	1.00	1.11		
[8]	0.06	0.06	0.06	0.06	0.06	0.85	1.05	

Fig.1. Estimates of Evolutionary Divergence between Sequences .The number of amino acid substitutions per site from between sequences are shown. Analyses were conducted using the Poisson correction model [1]. The analysis involved 8 amino acid sequences. All positions containing gaps and missing data were eliminated. There were a total of 103 positions in the final dataset. Evolutionary analyses were conducted in MEGA6 [2].

1- *C. metallidurans*, 2- *C. gilardi*, 3- *C. metallidurans*, 4- *C. metallidurans*, 5- *C. metallidurans* 6- Burkholderiaceae bacterium 7- *Ralstonia insidiosa*, 8- *Ralstonia mannitolilytica*

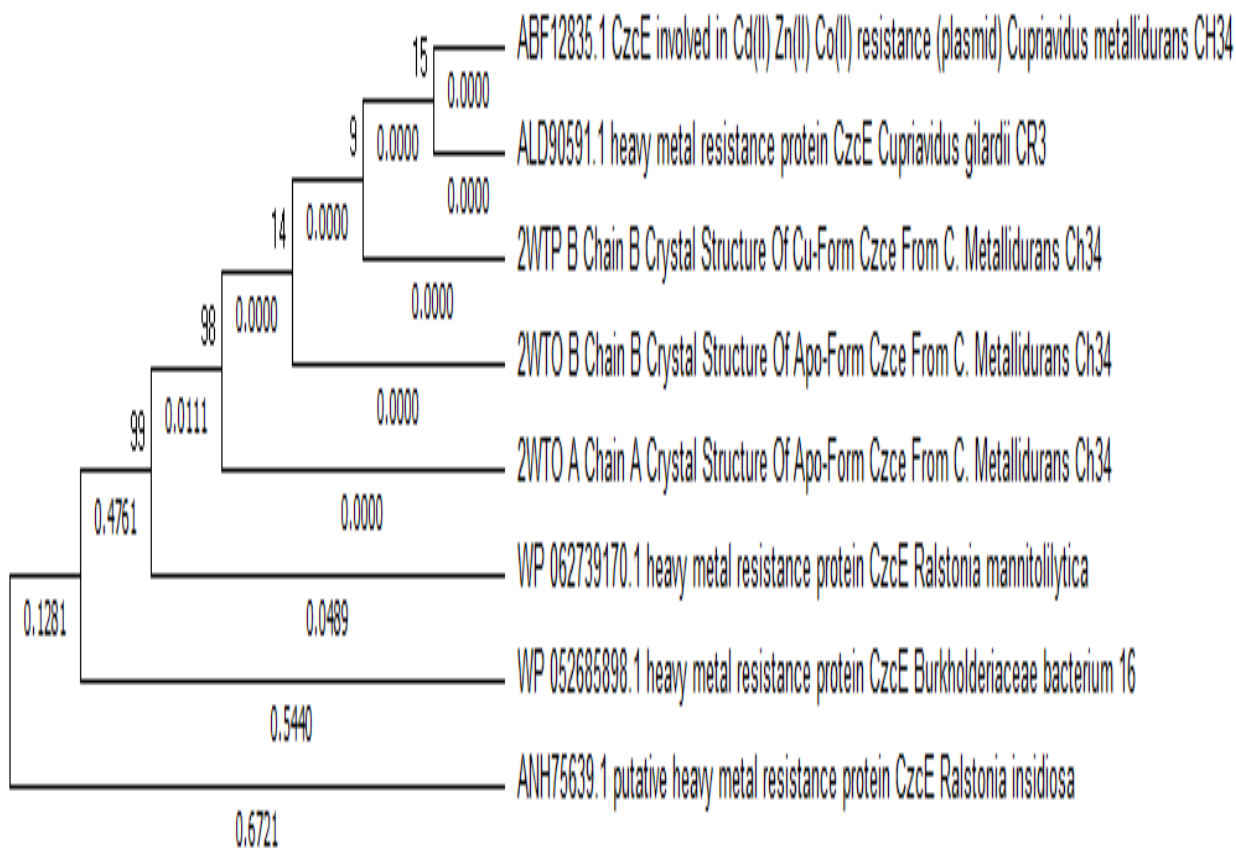


Fig.2 Phylogenetic tree for CzcE protein.

CzcE in Bacteria	No of amino acid	P ₁	Asp+ Glu	Arg + Lys	Gravy
CzcE in (<i>C.metallidurans</i>)	133	9.34	11	14	0.086
CzcE in (<i>Ralstonia mannitolilytica</i>)	113	9.57	9	13	-0.001
CzcE in (<i>Burkholderiaceae</i> bacterium)	127	9.99	5	10	0.292
CzcE in (<i>Ralstonia insidiosa</i>)	124	9.51	7	9	0.375

Table 1: Physico-chemical parameters of CzcE protein in certain group of Bacteria

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