EXPRESSION, CHARACTERIZATION AND ENZYMATIC ACTIVITY OF THE C-CATALYTIC SITE OF THE ANGIOTENSIN-CONVERTING ENZYME I

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Angiotensin converting enzyme I (ACE) is a membrane-bound, zinc dependent dipeptidase that catalyzes the conversion of the angiotensin I to the potent vasopressor angiotensin II. ACE is well known as a key part of the renin angiotenisn system that regulates blood pressure, and its inhibitors have potential for the treatment of hypertension. In particular, the catalytic mechanisms of the two active sites of somatic ACE in the cleavage of angiotensin I and bradykin in are different. Therefore, it would likely provide a new way for exploiting novel ACE inhibitors with fewer side-effects by specifically-targeting the individual active sites of somatic ACE.

This study aims to express in bacterial system, purify and characterize the Ala^{959} to Ser^{1066} region (c-ACE) that corresponds to the c-Catalytic domain of human somatic ACE.

The methodology used was the cDNA amplification of this by PCR reactions, cloning and expression in bacterial system. The purification was used the His Tag column and the characterization of the cACE catalytic site was by circular dichroism, fluorescence and enzymatic activity assay.

We amplified the 324 bp sequence, cloned into pET28 vector and the cACE catalytic site was expressed at 37°C for 5h. The expression this protein was in soluble form and its purification was made in one step by His Tag column, it produced the cACE pure. Its secondary structure was confirmed by CD and this protein owns activity

In conclusion, the strategic used for production the cACE catalytic site was very efficient and it produced the protein with the secondary structure with enzymatic activity.

Keywords**:** ACE I; c-Catalytic site, enzymatic activity

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