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15th World Congress on

BIOTECHNOLOGY AND BIOTECH INDUSTRIES MEET & 2nd International Conference on ENZYMOLOGY AND MOLECULAR BIOLOGY

March 20-21, 2017 Rome, Italy

A new approach to obtain the catalytic sites region of human sACE with correct fold and activity

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A ngiotensin-converting enzyme I (ACE) is a membrane-bound that catalyzes the conversion of angiotensin I to the potent vasopressor angiotensin II. ACE is a key part of the renin-angiotensin system, which regulates blood pressure and is widely distributed throughout the body. There are two isoforms of human ACE, including the somatic ACE (sACE) present in somatic tissue and the testicular ACE (tACE) present in male germinal cells. The sACE possesses two domains, N- C- domains, with catalytic sites which exhibit 60% sequence identity. These domains differ in terms of chloride-ion activation profiles, rates of peptide hydrolysis of angiotensin I, bradykinin, Goralatide, Luliberin, substance P, angiotensina, beta-amyloid peptide and sensitivities to various inhibitors. A more detailed analysis shows that these regions are composed of HEMGH and EAIGD sequences that bind zinc ions to facilitate catalytic activity (Fig. 1). Our question is: If the synthesis of catalytic sites with corrects structure and activity could be a good model *per si* to study new drugs. The objective was to obtain the Ala³⁶¹ a Gli⁴⁶⁸ and Ala⁹⁵⁹ to Ser¹⁰⁶⁶ catalytic regions sACE in a structural conformation that resembles its native form. The catalytic regions were obtained from bacterial system; the expression of this protein in soluble form enables completion of the solubilization/purification steps without the need for refolding. The characterization of Ala⁹⁵⁹ to Ser¹⁰⁶⁶ region shows that this has an α -helix and β -strand structure, Fig. 1b, which zinc ion (essential for its activity) binds to, and with enzymatic activity was successful.

Biography

Regina Affonso has experience in the field of Biochemistry, with emphasis on protein in the area of molecular biology, working on the following topics: RNA extraction, RT-PCR, PCR, cloning, expression and purification of recombinant human proteins in bacterial system and cell culture. In the structural area, she is interested in: circular dichroism, fluorescence, crystallography, and bioinformatics.

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