

REVERSIBLE UNFOLDING OF RECOMBINANT PORCINE S100A12

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Porcine S100A12 is a member of the S100 protein family, a group of small acidic calcium-binding proteins characterized by the presence of two EF-hand motifs. These proteins are involved in many cellular events such as the regulation of protein phosphorylation, enzymatic activity, protein-protein interaction, Ca²⁺ homeostasis, inflammatory processes and intermediate filament polymerization. In addition, members of this family bind Zn²⁺ or Ca²⁺ with cooperative effect on ion binding. In this study, the gene sequence encoding porcine S100A12 was obtained by the synthetic gene approach using *E. coli* codon bias, resulting in the production of large amounts of the recombinant protein. Additionally, we report a thermodynamic study of the recombinant S100A12 using far-UV circular dichroism, fluorescence and isothermal titration calorimetry (ITC). The results of urea and temperature induced unfolding and refolding processes indicated a reversible two-state process. Also, the ANS fluorescence studies showed that in presence of divalent ions the protein exposes hydrophobic sites which could facilitate the interaction with other proteins and trigger the physiological responses.

Keywords: S100A12; calcium-binding protein; S100 family, Circular dichroism (CD); Fluorescence spectroscopy; protein unfolding.

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