Determination of Structural Parameters of α-Trypsin in Aqueous-Organic Media

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INTRODUCTION: α-trypsin is a serine-protease with a polypeptide chain of 223 amino acid residues and six disulfide bridges. The use of enzymes in organic media has allowed the occurrence of reactions using aqueous-immiscible substrate. This work studied the effect of monoalcohols concentration on thermodynamic parameters of bovine α- trypsin. **MATERIALS AND METHODS:** The structural changes in α-trypsin were tested in aqueous solutions of ethanol and monitored using spectroscopic methods, like circular dichroism, dynamic scattering light, fluorescence and UV-VIS spectroscopy. The protein solution was prepared in buffer and ethanol, with the following concentrations of solvent: 20%, 40%, 60% and 80% v/v. The final protein solution concentration was in range from 0.01 to 0.5 $mg.mL^{-1}$. **RESULTS AND DISCUSSION:** The DLS assay showed an increase of diameter directly related to the increase in ethanol concentration, suggesting protein aggregation. The fluorescence results demonstrate that has occurred increase of the emission intensity and a displacement of wavelength for infrared region with the ethanol concentration increase, indicating unfolding and suggesting protein aggregation. The circular dichroism demonstrated a small conformational change at organic solvent range tested. **CONCLUSION:** These results show that the effect of organic solvents on α-trypsin structure induces small conformational change and the formation of aggregates.

Word Keys: α-trypsin, structural parameters, spectroscopy, ethanol.

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