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## Structural adaptation of late embryogenesis abundant protein from *Artemia* under molecular crowding conditions

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### Abstract

Late embryogenesis abundant (LEA) proteins are crucial for protecting organisms like *Artemia* against abiotic stresses, such as high salinity and drought [1,2]. Although their protective roles are well-documented, the underlying molecular mechanisms, particularly regarding their structural adaptations under stress, remain poorly understood. This study investigates the structural behavior of LEA proteins in response to molecular crowding agents and membrane mimetics, which replicate conditions similar to those encountered during environmental stress. LEA protein from *Artemia urmiana* was expressed in *E. coli* BL21(DE3) and purified. To mimic the molecular crowding and stress conditions, we selected compounds such as polyethylene glycol (PEG), glycerol, trifluoroethanol (TFE), and sodium dodecyl sulfate (SDS). PEG and glycerol simulate molecular crowding, while TFE induces the formation of helical structures, and SDS serves as a membrane mimic, as LEA proteins are known to protect cell membranes under stress [3,4,5]. We used spectroscopic techniques-UV absorption, circular dichroism (CD), and fluorescence spectroscopy-to examine the structural transitions of LEA proteins in the presence of these compounds. Our results show that in its native, hydrated state, the LEA protein predominantly adopts a random coil conformation. However, in the presence of PEG, glycerol, and TFE, the protein underwent a structural shift toward increased helicity, indicating a transition to a more compact conformation. This helical formation was most pronounced in the presence of TFE, suggesting that LEA proteins may utilize such structural changes to stabilize cellular structures under stress.

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Additionally, in the presence of SDS, which mimics a membrane environment, LEA proteins showed enhanced folding, supporting their known membrane-protective role.

**Key words:** LEA proteins, Molecular Crowding, Trifluoroethanol, SDS, Artemia

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