Metal Cations Effect on Membrane Crystallized Lysozyme

<u>Silvia Simone</u>^{a,b}, Efrem Curcio^{a,c}, Gianluca Di Profio^{a,c}, Enrico Drioli^{a,c}, Marta Ferraroni^d, Andrea Scozzafava^d, ^aITM-CNR, Cosenza, Italy. ^bDepartment of Chemistry, University of Calabria. ^cDepartment of Chemical Engineering and Materials, University of Calabria. ^dDepartment of Chemistry, University of Florence. E-mail: s.simone@unical.it

Microporous hydrophobic membranes are innovative tools in protein crystallization [1]. In this work, effects of $CoCl_2$ and $CuCl_2$, used as precipitant agents, on membrane-crystallization of hen egg white lysozyme (HEWL), are described. The HEWL*Co²⁺ complex gave rise to a new P2₁2₁2₁ orthorhombic form (a= 36.81 Å, b= 77.56 Å, c= 80.38 Å) beside the ordinary tetragonal one. Literature reports only another similar case: a P2₁2₁2₁ orthorhombic form of an HEWL*Ni²⁺ compound, grown under strong magnetic field [2]. Membrane crystallization of HEWL with CuCl₂ allowed to observe new coordination positions of Cu²⁺ to lysozyme (Asp18, Asp87) respect to ones already described (Asp52, Leu129, Arg14) [3]. DSC tests showed cobalt coordination increases lysozyme stability, while copper binding by oxygen atoms is unfavourable and decreases crystals melting point. HEWL specific activity increases after crystallization owing to its further purification and seems to be more affected by copper coordination.

Curcio E., Di Profio G., Drioli E., *Jnl. Crys. Growth*, 2003, 247, 166. [2]
Yin D.C., Oda Y., Wakayama N.I., Ataka M., *Jnl. Crys. Growth*, 2003, 252, 618. [3]
Teichberg V.I., Sharon M., Moult J., Smilansky A., Yonath A., *Jnl. Mol. Biol.*, 1974, 87, 357.

Keywords: crystal growth apparatus design, bioinorganic chemistry, protein crystallization