

Crystallization trials for CytC apo from Horse Heart

<i>Trials with low ionic strength</i>			
Conditions	Drop 4 μ L	2 μ L ([CytC]=25mg/ml in [Na ₃ PO ₄]=50mM pH7) + 2 μ L (PEG1000 25%w/v in [Na ₃ PO ₄]=50mM pH7)	Drop: We put on cover slip 2 μ L of solution B and after we added 2 μ L of solution C
	Reservoir 700 μ L	PEG1000 30%w/v in [Na ₃ PO ₄]=50mM pH7	The reservoir was made of: 250 μ L of solution A 600 μ L of PEG1000 50% (HR2-523 Hampton) 150 μ L of milliQ water
	Method	Hanging drop (Nextal Plate)	
	Temperature	20°C	

Solution A (*buffer phosphate 200mM pH7*): the solution was made of trisodic phosphate (71908 Fluka) titrated with NaOH (480507 Carlo Erba) and HCl (403912 Carlo Erba) to obtain pH 7.

Solution B (*buffer phosphate 50mM pH7, PEG1000 25%w/v*): the solution was made of 25 μ L of solution A, 50 μ L of PEG 1000 50%w/v (HR2-532 Hampton) and 25 μ L of milliQ water.

Solution C (*[CytC]=25mg/ml, buffer phosphate 50mM pH7, PEG1000 25%w/v*): the solution was made of 10mg of CytC (105201 Sigma Aldrich) stabilized in 100 μ L of solution A. The protein solution was diluted with 300 μ L of milliQ water