# The nucleating agents, crystallophores, to boost your crystal production.

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The crystallophore is a lanthanide complex combining phasing and nucleating properties [1], successfully exploited to produce crystalline forms free of crystal defects often encountered by crystallographers such as low-resolution diffracting samples or crystals with twinning [2], to generate crystals can from enriched fractions containing several proteins [2] leading to the structure determination of a protein complex [3] and to induce nucleation directly from the protein solution, as exemplified by the crystallization of hen egg white lysozyme in water [4].

Time-resolved Serial Crystallography (TR-SX) allows to obtain structural dynamics information and observe biological macromolecules in action by capturing transient intermediates along a biological pathway. [5–8] From an experimental point of view, serial crystallography brings new constraints on crystal preparation as it intrinsically requires a large amount of samples to make sure to collect a complete diffraction data set. Moreover optimal time-resolved experiments require crystalline samples with a narrow size distribution in order to ensure a uniform triggering of the reaction under study through the entire crystal volume.

The nucleating properties of the crystallophore have been challenged for the production of crystals with the appropriate size for either SX experiments or electron diffraction of 3D nanocrystals [9]. Moreover, these crystals were generated in the minute time-scale opening new opportunities in TR-SX. We will also show the input of crystallophore variants bearing chemical modifications to expand the possibility to control the number and size of crystals [10]. Finally, to facilitate crystal detection, a crystallophore with optimized imaging property (Fig. 1) completes the toolbox [11].



###### **Figure 1**. HEWL crystal co-crystallized with TbXo4PhOMeNMe2 (1mM).

###### Transmission imaging (left) and spectral imaging (λexcitation = 700 nm, Plaser=10%) (right).

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