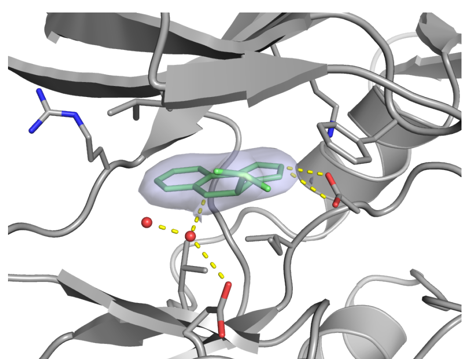

When sending your samples for crystallization:

In order to set up crystallization screens for crystal structure determination we generally need 0.2-0.5 ml of a 10 mg/ml protein solution, although we can start with less protein or more dilute samples if material is scarce or less soluble. With this amount we can set up a battery of crystallization screens (many hundred drops) at different temperatures, initially 20 °C and 4 °C. Our state-of-the-art robotic equipment allows us to set up 96-well crystallization plates with as little as 15 microL protein solution per plate. The protein solution can be sent in a variety of ways, most commonly frozen and shipped on dry ice (using FedEx or similar). If the sample is sensitive to freezing (which sometimes causes aggregation when the sample is thawed), glycerol may be added prior to freezing. The protein may also be sent in a diluted state, if dilution does not denature it. Upon arrival of the sample we apply an extensive protein characterization procedure to assess solubility, stability and polydispersity of the protein. We use the methods of dynamic light scattering and differential scanning fluorimetry (Thermofluor). Prior to the crystallization trials we may dialyze the sample to a new buffer and concentrate it if required. Crystallization screens and crystallization conditions optimization can take as little as 3-6 weeks, although longer times may be necessary depending on the kinetics of crystallization. Data collection from well-diffracting crystals and subsequent structure determination are usually very quick. In cases when a close homologue with known three-dimensional structure (generally above 40-50% sequence identity) exists in the Protein Data Bank, the method of molecular replacement may by used for structure determination. For "de novo" protein structure determination, where no structural homologues are available, we strongly recommend selenomethionine (SeMet) labeled protein material, which we can produce if the customer is not able to. This enables rapid structure determination using synchrotron X-rays. Alternatively we may use halide soaking of the crystals. If, instead, the traditional method of heavy metal soaking has to be used, the structure determination may take longer.