Effect of TELSAM-Target Protein Linkers on Crystal Formation and Quality

Celeste Litchfield[1], Parag Gajjar[2], Miles Callahan[3], Nathan Redd[4], James Moody[5]

[1]Brigham Young University [2]Brigham Young University, [3]Brigham Young University, [4]Brigham Young University, [5]N/A

maisonceleste@gmail.com

Understanding the limitations and characteristics of crystal formation from TELSAM polymers is important in using TELSAM to crystallize proteins that cannot feasibly be crystallized on their own. One variable in TELSAM polymerization that needs to be understood is the effect of the linker between TELSAM and the target protein. To better understand this factor, we attempted to crystallize TELSAM and CMG2 connected with 4 different linkers. We observed the effect of the linker on the conditions under which the proteins crystallized as well as the resolution of the diffraction data from each crystal. Threonine-threonine linkers did not form any crystals. Of the alanine-alanine linkers, only 1 crystal diffracted; it had decent resolution (2.6Å). Threonine-valine linkers produced crystals with the best resolution upon diffraction (as low as 2.3Å typically around 3.5Å) followed closely by alanine-valine(as low as 2.6Å, but typically around 3.5Å). This is consistent with the theory that a threonine-threonine linker would be most rigid and an alanine-alanine linker would be most flexible with the alanine-valine and threonine-valine linkers having intermediate flexibility; and that the best linkers for TELSAM have some flexibility to shift as the polymer forms, but are stiff enough to hold their position and stay in a crystal formation. The diffraction data is currently being refined, and in the future, we intend to test different lengths of linkers as well as different target proteins.