

“To Cross-Seed or Not To Cross-Seed”: A Pilot Study Using Metallo- β -lactamases

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Supporting Information

ABSTRACT: Knowledge of protein structures is of central importance in modern drug discovery and molecular biology, but to be useful the structures, including those obtained in the crystalline state, must be biologically relevant. Small variations in crystallization conditions can lead to alternative crystal forms, conformations, and oligomerization states, causing changes which can lead to altered fold and active site architectures. In the determination of protein structures by X-ray crystallography, crystallization is an essential prerequisite and remains a major bottleneck in drug discovery. Although many methods have been tried in an attempt to improve the production of protein crystals, it is still largely a “trial and error” process. To our knowledge, crystallization by cross-seeding using homologous proteins has previously only been successful for proteins with greater than 61–74% sequence identity. In the study presented here, we explore the effect of low sequence similarity on cross-seeding using metallo- β -lactamases with sequence identities and sequence similarities as low as 24% and 36%, respectively, but with homologous core folds. Despite the low sequence identities, the results show that micro-cross-seeding matrix screening can increase the number of hits obtained and can shorten crystallization time. It can also help in the identification of new crystallization conditions and different crystal forms.

