Supplemental Information

Transient B_{12}-dependent methyltransferase complexes revealed by small-angle X-ray scattering

Nozomi Ando,[1,2] Yan Kung,[2] Mehmet Can,[4] Güneş Bender,[4] Stephen W. Ragsdale,[4] and Catherine L. Drennan[1-3]*

Howard Hughes Medical Institute,[1] Department of Chemistry,[2] and Department of Biology,[3] Massachusetts Institute of Technology, Cambridge, MA 02139

Department of Biological Chemistry,[4] University of Michigan, Ann Arbor, MI 48109

Inventory of Supplemental Information

Figure S1. Individual shape reconstruction of free CFeSP.

Figure S2. Residuals of multi-species fits to titration data under assay conditions.

Figure S3. Singular value decomposition of titration data under both assay and crystallization conditions.

Figure S4. Additional multi-species fits to titration data under assay conditions.

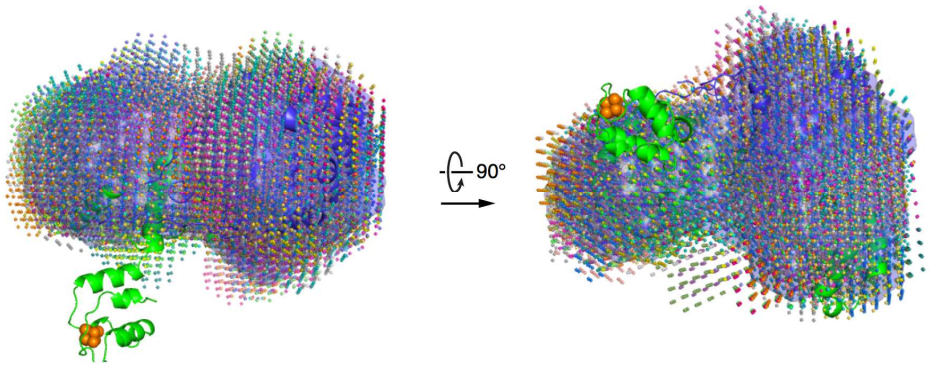

Figure S1. Shape reconstruction of free CFeSP with dammif.[1] The twenty individual models are shown as beads and the averaged model is shown as a surface. When aligned with a crystal structure of CFeSP, the extended Fe_4S_4 domain protrudes from the averaged model. While extended features resembling such a domain are not apparent in the individual reconstructions, there are small lobes in various locations on the surface of the core domains that are removed in the averaging. It is possible that these disordered features represent the Fe_4S_4 domain.

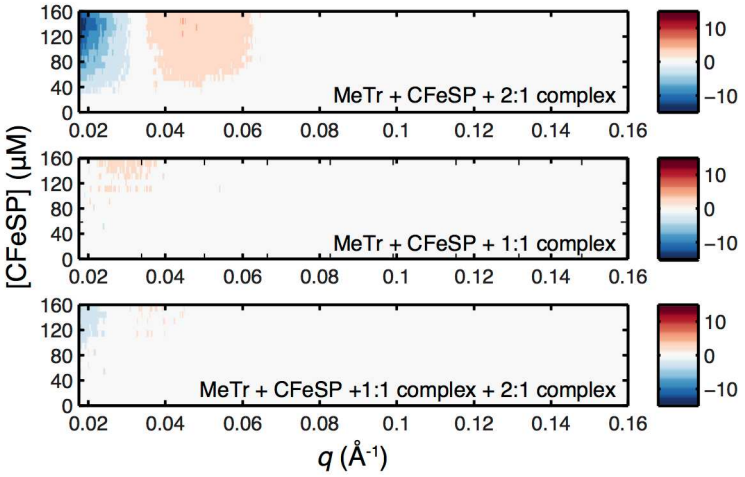

Figure S2. Residuals of multi-species fits to scattering data obtained in the titration of CFeSP (0-150 μM) into MeTr homodimer (fixed at 50 μM) under assay conditions. Residuals of fits to scattering intensity, $I_{data}(q)$-$I_{fit}(q)$, shown as intensity maps are minimized by the inclusion of a 1:1 complex in the fits, while inclusion of the 2:1 complex has a negligible effect.

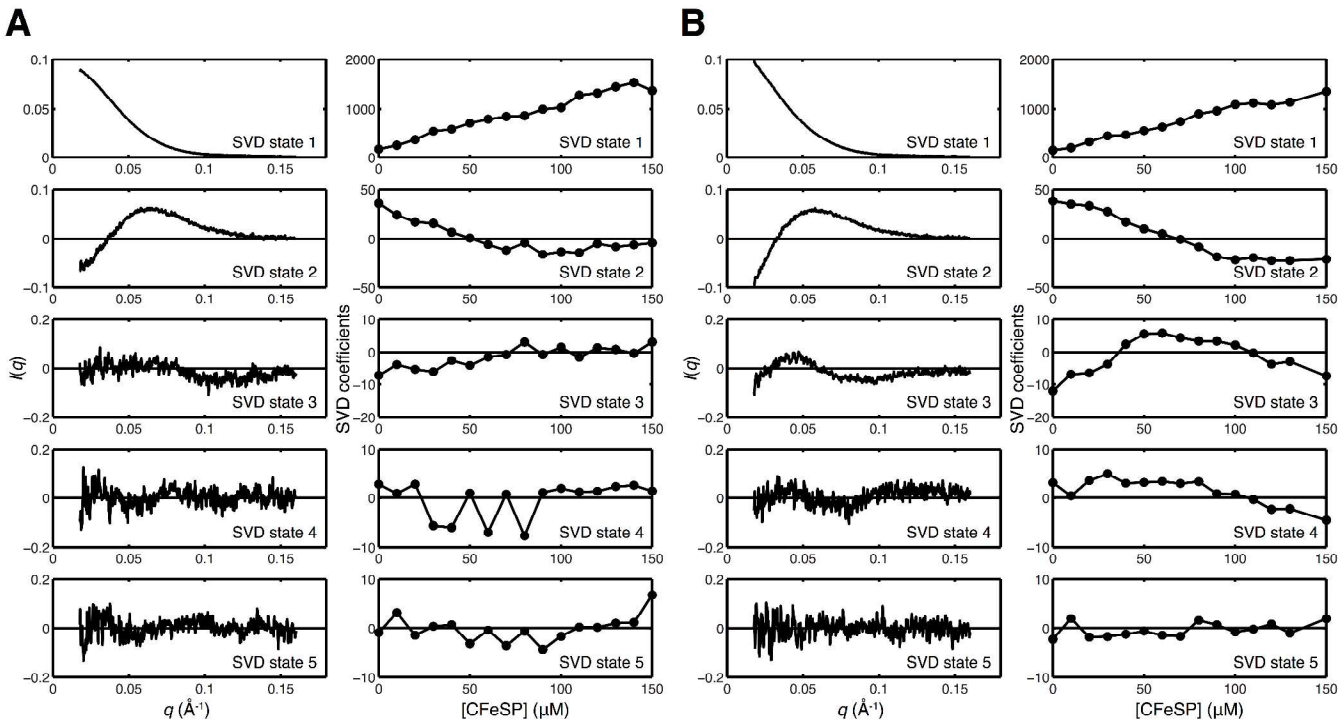

Figure S3. Singular value decomposition (SVD) analysis of scattering data obtained in the titration of CFeSP (0-150 μM) into MeTr homodimer (fixed at 50 μM). (A) Under assay conditions, three SVD states show meaningful changes in the SVD coefficients as a function of the titrated CFeSP concentration, [CFeSP]. The first SVD state accounts for the overall increase in scattering intensity due to the increase in total protein concentration. The coefficients for the second and third SVD states show meaningful trends with respect to [CFeSP], changing sign near the equimolar point (50 μM), thus account for changes in the distribution of species. Higher order SVD states display coefficients with no apparent correlation to [CFeSP]. (B) Under crystallization conditions, four SVD states show meaningful changes with [CFeSP]. Again, the first SVD state accounts for the overall increase in scattering intensity. The coefficients for the second to fourth states display changes in sign and/or slope with respect to [CFeSP] and thus accounts for changes in shape due to changes in the distribution of species. Higher order SVD states display coefficients with no apparent correlation to [CFeSP].

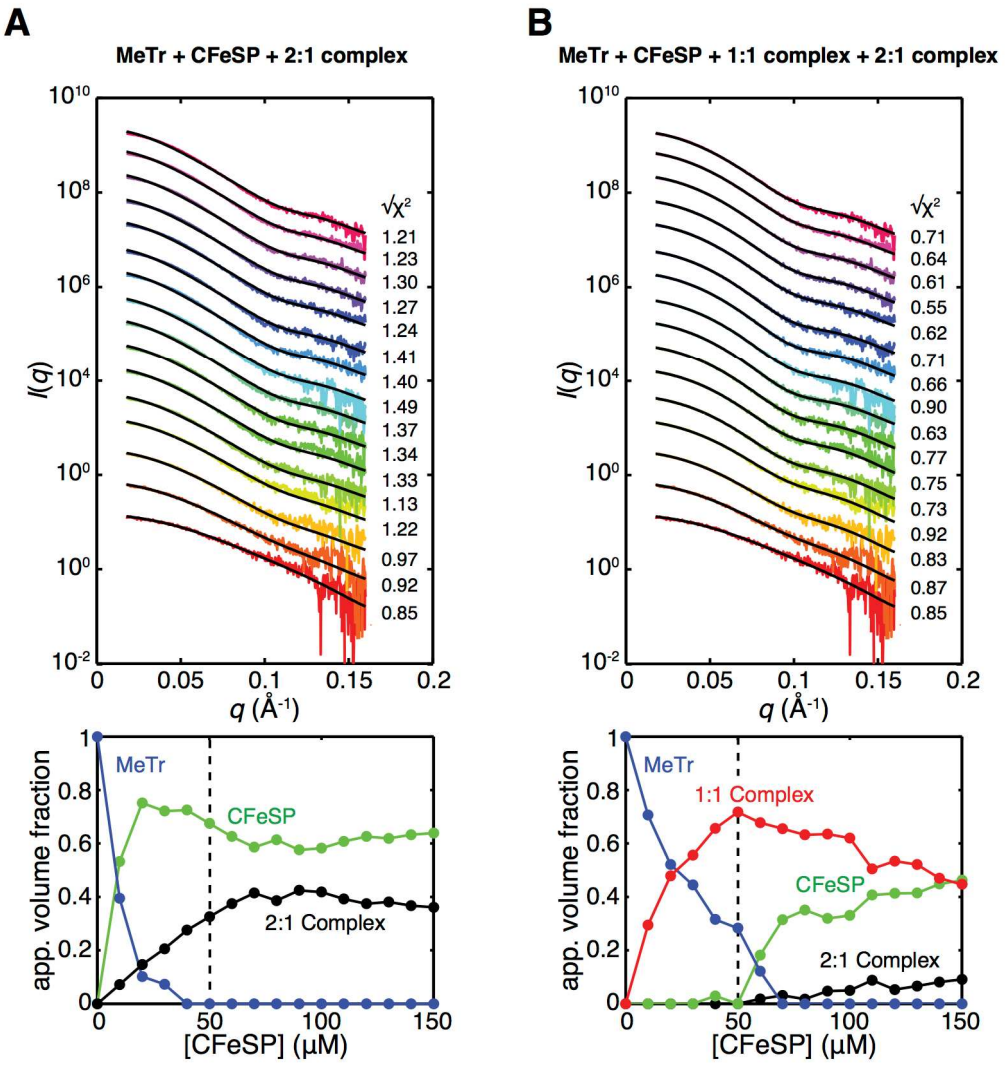

Figure S4. Additional multi-species fitting to scattering data obtained in the titration of CFeSP (0-150 μM) into MeTr homodimer (fixed at 50 μM) under assay conditions. Profile colors range from red to violet (bottom to top) and indicate increasing CFeSP concentrations. Multi-species fits (shown in black) and corresponding $\sqrt{\chi^2}$ values were obtained with the program OLIGOMER.[2] (A) An example of fitting an incorrect combination of species (free MeTr, free CFeSP, and the 2:1 complex). While the fits may not appear poor by eye, the apparent volume fractions display trends that are far from physically reasonable. For example, CFeSP should not be observed in large excess below the equimolar point (dotted line). (B) Fitting free MeTr, free CFeSP, the 1:1 complex, and the 2:1 complex yield similar results to that obtained without the 2:1 complex, suggesting that the 2:1 complex is not a significant species under these conditions.

References

(1) Franke, D.; Svergun, D. I. *J. Appl. Cryst.* **2009**, *42*, 342.

(2) Konarev, P. V.; Volkov, V. V.; Sokolova, A. V.; Koch, M. H. J.; Svergun, D. I. *J. Appl. Cryst.* **2003**, *36*, 1277.