## Molecular mechanisms of chromatin recognition and remodeling by the ATAD2/B proteins Karen C. Glass, Ph.D., University of Vermont

## **Preliminary Results:**

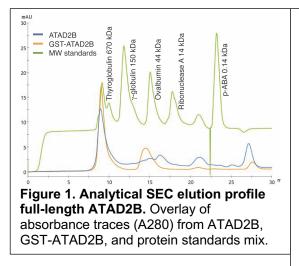


Figure 2. Pull-down assays with GST-ATAD2B full-length protein and histone H3-H4 tetramer. SDS-PAGE gel of GST-ATAD2B protein (MW 149 kDa, top arrow) bound to Glutathione Agarose and incubated with the purified recombinant *X. laevis* histone H3-H4 tetramer (MW 15.3 and 11.2 kDa).

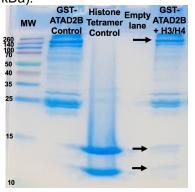
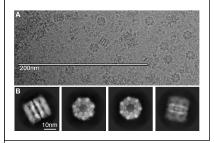
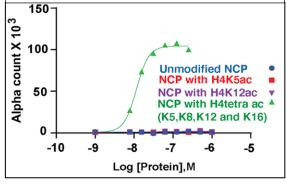


Figure 3. Cryo-EM images taken of ATAD2B on a 300 kV TFS Titan Krios at the NCCAT. A. A raw micrograph of full-length GST-ATAD2B. B. Representative 2D class averages of the GST-ATAD2B complex processed with CryoSPARC.



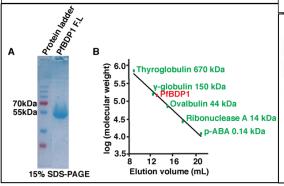
**Figure 4.** dCypher nucleosome binding assay of PfBDP1 with recombinant nucleosome core particles (NCPs) containing unmodified or acetylated H4 histones.



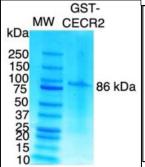
**Table 1.** Binding affinities of full-length PfBDP1 and PfBDP1-BRDs with histone ligands measured by ITC.

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Histone Peptide (1-24)	PfBDP1 Full length (1-488) K <sub>D</sub> (µM)	PfBDP1-BRD (303-488) Κ <sub>D</sub> (μΜ)	PfBDP1-BRD (333-456) Κ <sub>D</sub> (μΜ)
Unmodified H4	No binding	No binding	No binding
H4K5acK8ac	100 ± 5.30	1510 ± 168	No binding
H4K5acK12ac	214 ± 14.8	621 ± 28.3	No binding
H4K5acK8acK12acK16ac	48.8 ± 3.63	173 ± 11.3	No binding

**Figure 5.** (A) SDS-PAGE gel of purified full-length PfBDP1. (B) Analytical SEC of full-length PfBDP1 with protein standards.



**Figure 6**. SDS-PAGE gel of the purified GST-CECR2 protein (residues 4-538).



**Figure 7**. dCypher nucleosome binding assay of the CECR2 bromodomain with recombinant nucleosome core particles (NCPs) containing unmodified or acetylated H4 histones.

