REGULATION OF THE ATPase ACTIVITY OF ABCE1 FROM *PYROCOCCUS ABYSSI* BY Fe-S CLUSTER STATUS AND Mg^{2+}: IMPLICATION FOR RIBOSOMAL FUNCTION.

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Ribosomal function is dependent on multiple proteins. The ABCE1 ATPase, a unique ABC superfamily member that bears two Fe_4S_4 clusters, is crucial for ribosomal biogenesis and recycling. Here, the ATPase activity of the *Pyrococcus abyssi* ABCE1 (*Pab*ABCE1) studied using both *apo*- (without reconstituted Fe-S clusters) and *holo*- (with full complement of Fe-S clusters reconstituted post-purification) forms and is shown to be jointly regulated by the status of FeS clusters and Mg^{2+}. Typically ATPases require Mg^{2+}, as is true for *Pab*ABCE1, but *Pab*ABCE1 for Mg^{2+} also acts as a negative allosteric effector that modulates ATP affinity. Physiological [Mg^{2+}] inhibits the *Pab*ABCE1 ATPase (K_i of ~1 μM) for both *apo*- and *holo*-*Pab*ABCE1. Comparative kinetic analysis of Mg^{2+} inhibition shows differences in degree of allosteric regulation between the *apo*- and *holo*-*Pab*ABCE1 where the apparent ATP K_m of *apo*-*Pab*ABCE1 increases >30 fold from ~30 μM to over 1 mM with Mg^{2+}. This effect would significantly convert the ATPase activity of *Pab*ABCE1 from being independent of cellular energy charge (φ) to being dependent on φ with cellular [Mg^{2+}]. These findings uncover an intricate balance between [Mg^{2+}] and status of FeS clusters playing a joint role in regulating ABCE1's ATPase activity with implications to ribosomal function.