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Fluorescent ATP analog mant-ATP reports dynein activity in the isolated *Chlamydomonas* axoneme MARIA FEOFILOVA, JONATHON HOWARD, Yale University — Eukaryotic flagella are long rod-like extensions of cells, which play a fundamental role in single cell movement, as well as in fluid transport. Flagella contain a highly evolutionary conserved mechanical structure called the axoneme. The motion of the flagellum is generated by dynein motor proteins located all along the length of the axoneme. How the force production of motors is controlled spatially and temporally is still an open question. Therefore, monitoring dynein activity in the axonemal structure is expected to provide novel insights in regulation of the beat. We use high sensitivity fluorescence microscopy to monitor the binding and hydrolysis kinetics of the fluorescently labeled ATP analogue mant-ATP (2'(3')-O-(N-methylanthraniloyl) adenosine 5'-triphosphate), which is known to support dynein activity. By studying the kinetics of mant-ATP fluorescence, we identified distinct mant-ATP binding sites in the axoneme. The application of this method to axonemes with reduced amounts of dynein, showed evidence that one of the sites is associated with binding to dynein. In the future, we would like to use this method to find the spatial distribution of dynein activity in the axoneme.

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