Supporting Information for:

Unraveling the Mystery of ATP Hydrolysis in Actin Filaments

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Restraint on Coarse-grained Variables

A conformational change occurs in actin monomers upon polymerization. One of the main changes is a flattening of the subdomain 2-1-3-4 dihedral angle. We chose to mimic the F-actin environment by simulating an actin monomer in an initial F-actin geometry and restraining the coarse-grain (CG) variable of the 2-1-3-4 dihedral angle. This was done in CP2K by creating a collective variable containing the center-of-mass of the Cα positions of the four subdomains and restraining that variable to be at a value of -0.55° with a harmonic force constant of 228.8 kcal/mol.

Additional QM/MM Metadynamics Simulation Details

Two collective variables were biased in the QM/MM simulations of ATP hydrolysis in actin. The coordination number between P_γ and O_β describes the making and breaking of the $\mathrm{P}_\gamma - \mathrm{O}_\beta$ bond and allows for recombination with any of the O_β atoms. This is depicted as CV1 in Figure S1. The mathematical form of the coordination number is:

$$CN = \sum_{i \in \mathrm{O}_\beta\ atoms} \frac{1 - \left(\frac{\left| r_i - r_{P_\gamma} \right|}{\sigma} \right)^{NN}}{1 - \left(\frac{\left| r_i - r_{P_\gamma} \right|}{\sigma} \right)^{ND}}$$

where $\sigma = 4.5\ Bohr$ was chosen to describe the making and breaking of the P-O bonds and NN=6 and ND=12 are CP2K default values.

The second collective variable biased in the metadynamics simulation is the coordination number between P_γ and both O_γ and QM water oxygens. This variable is chosen to describe the associative pathway of hydrolysis in which the lytic water adds to the gamma phosphate. This variable is depicted as CV2 in Figure S1 and the mathematical form is the same as above.

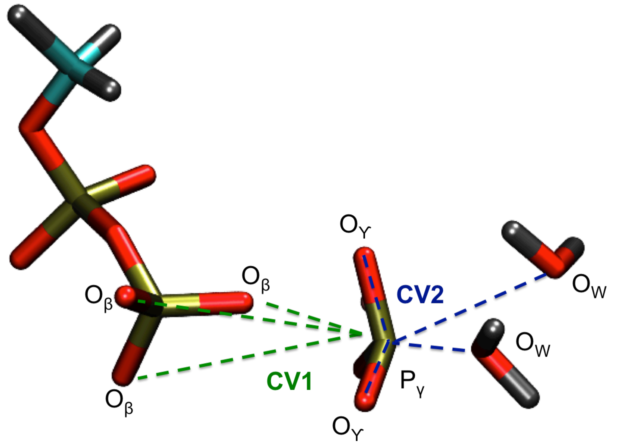

Figure S1. A depiction of the two collective variables biased in metadynamics simulations of ATP hydrolysis in actin.

Comparison of Starting Structures of G- and F-actin

The starting structures for G- and F-actin were taken from equilibrated classical molecular dynamics (MD) simulations. G-actin simulations were started from the crystal structure (pdb code 1NWK). The F-actin system was started from the Oda model (pdb code 2ZWH) in a 13mer periodic filament. Both systems were neutralized, solvated and equilibrated and simulated for at least 50 ns. In order to insure that the starting structures were indeed different, the residue by residue RMSD relative to each other was computed (Figure S2a). Numerous areas (residues 50-75 and 225-250) have RMSD values of 8 Å or more suggesting that the two structures are indeed quite different. The RMSD values for the 10 amino acids identified in the QM region are highlighted with red dots in Figure S2a. These areas do not have extremely large RMSD values but values of 1-2 Å still suggest slight differences in the QM region that may lead to differences in ATP hydrolysis.

In order to ensure that the F-actin structure is not only different than the equilibrated G-actin structure but also to the G-actin crystal structure, the residue by residue RMSD as compared to the crystal structure was computed (Figure S2b). The trends seen here are similar to the above with regions such as residues 50-75 and 225-250 showing large RMSD values.

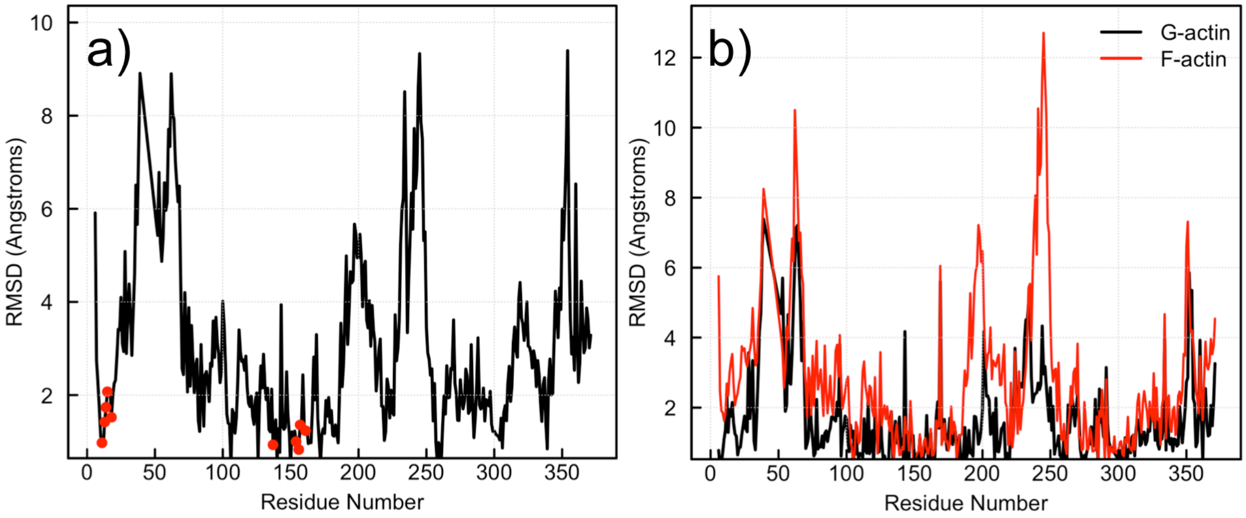

Figure S2. The RMSD of starting structures for G- and F-actin. a) The RMSD per residue of the F-actin starting structure relative to the G-actin starting structure. b) The RMSD of G-actin (black) and F-actin (red) relative to the G-actin crystal structure.

Discussion of Sources of Error.

A barrier height reduction of 8 kcal/mol is computed for ATP hydrolysis in F-actin as compared to G-actin from metadynamics QM/MM simulations. In comparison, the experimentally measured value is 7 kcal/mol. The 1 kcal/mol discrepancy is reasonable given the assumptions and approximations in our model. One obvious source of error is the density functional chosen. The barrier height discrepancy with experiment is also similar to the mean unsigned error of 1.8 kcal/mol (in solvent) for the barrier height for phosphodiester hydrolysis computed using DFT with the PBE functional.[1] Previous hydrolysis simulations using both PBE and BLYP functionals reported similar barrier height magnitudes to those reported in this study.[2-6]

Correlation Between Biased Collective Variables and Proton Transfer

The correlation between the two collective variables biased in the metadynamics simulations and the proton transfer pathway was monitored. The proton transfer variable chosen was that of the protonation state of the lytic water. It was found that a change in both collective variables (CV1 and CV2 described above) lead to proton transfer (Figure S3) suggesting a strong coupling between these degrees of freedom. Additionally, the time-scale of the proton transfer process is much more rapid than the change in two collective variables we chose to include in our metadynamics bias.

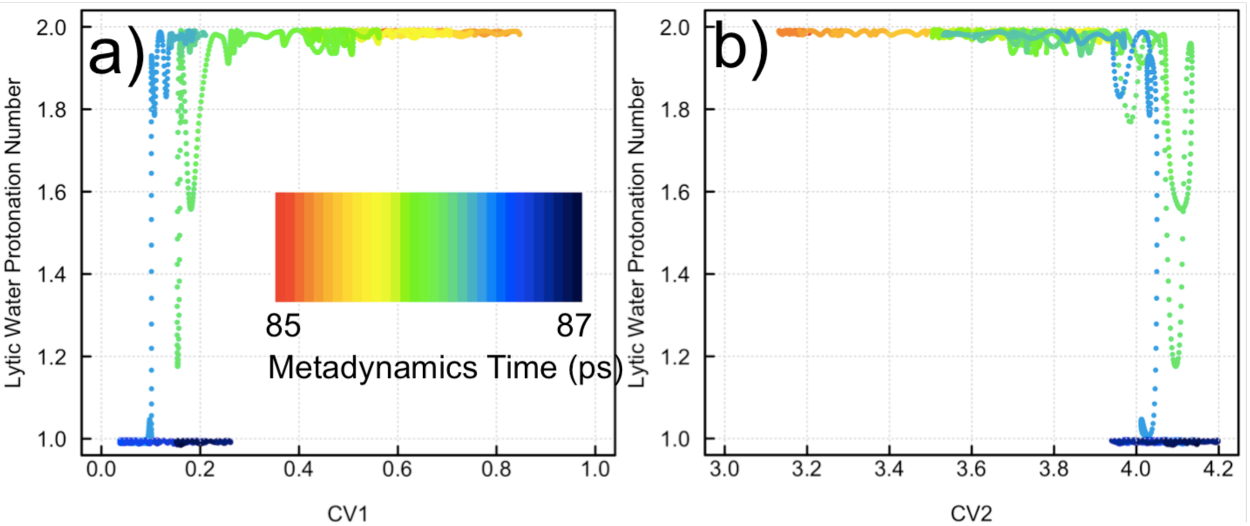

Figure S3. The correlation of metadynamics biased collective variables (CV1 in (a) and CV2 in (b)) and protonation of lytic water for F-actin. The metadynamics time of the given ordered pair is color-coded from red (prior to hydrolysis) to blue (post hydrolysis). The change in collective variable, in both cases, leads to a rapid deprotonation of the lytic water.

Supplemental References

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