Mechanobiology of stator remodeling in the bacterial flagellar motor

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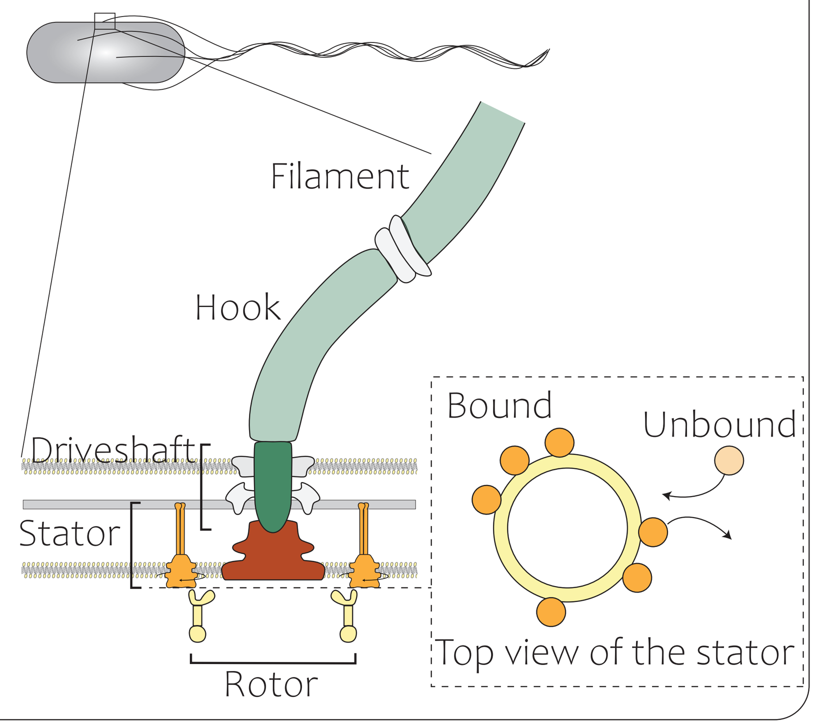
Background

Many bacteria swim by rotating helical filaments, which are driven by a highly complex molecular machine called the bacterial flagellar motor. The motor is powered by a stator which applies torque on a rotor to drive flagellar rotation. The stator consists of up to 11 stator stator units that exchange with a pool of free units in the inner membrane.

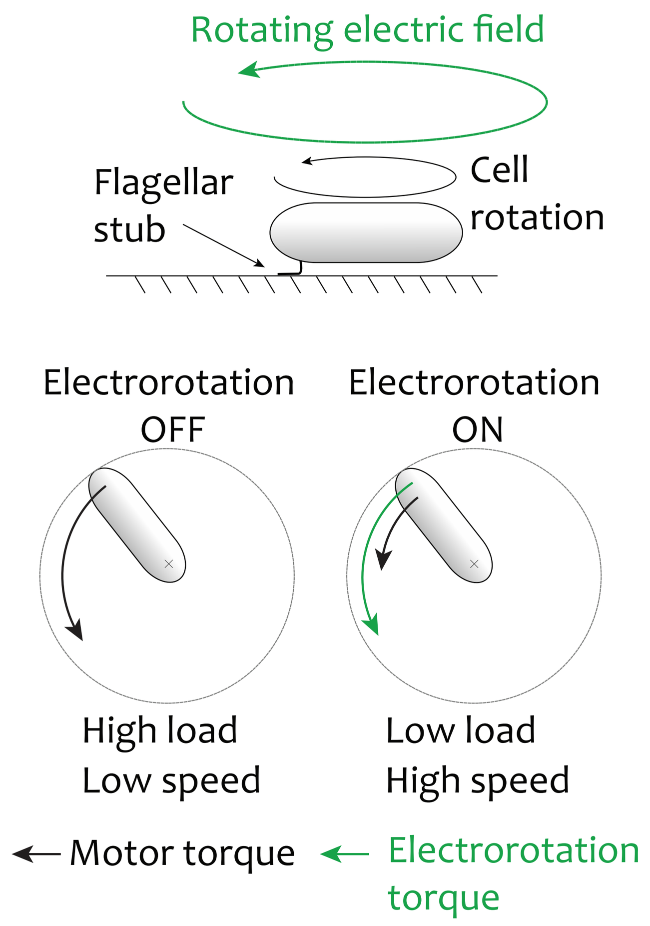


Motivation

Bacteria inhabit many complex enviroments, where they are exposed to different fluid viscosities, surface conditions, and crowding. These result in the flagellar motor working at many different levels of mechanical load. How does the motor deal with changes in mechanical load?

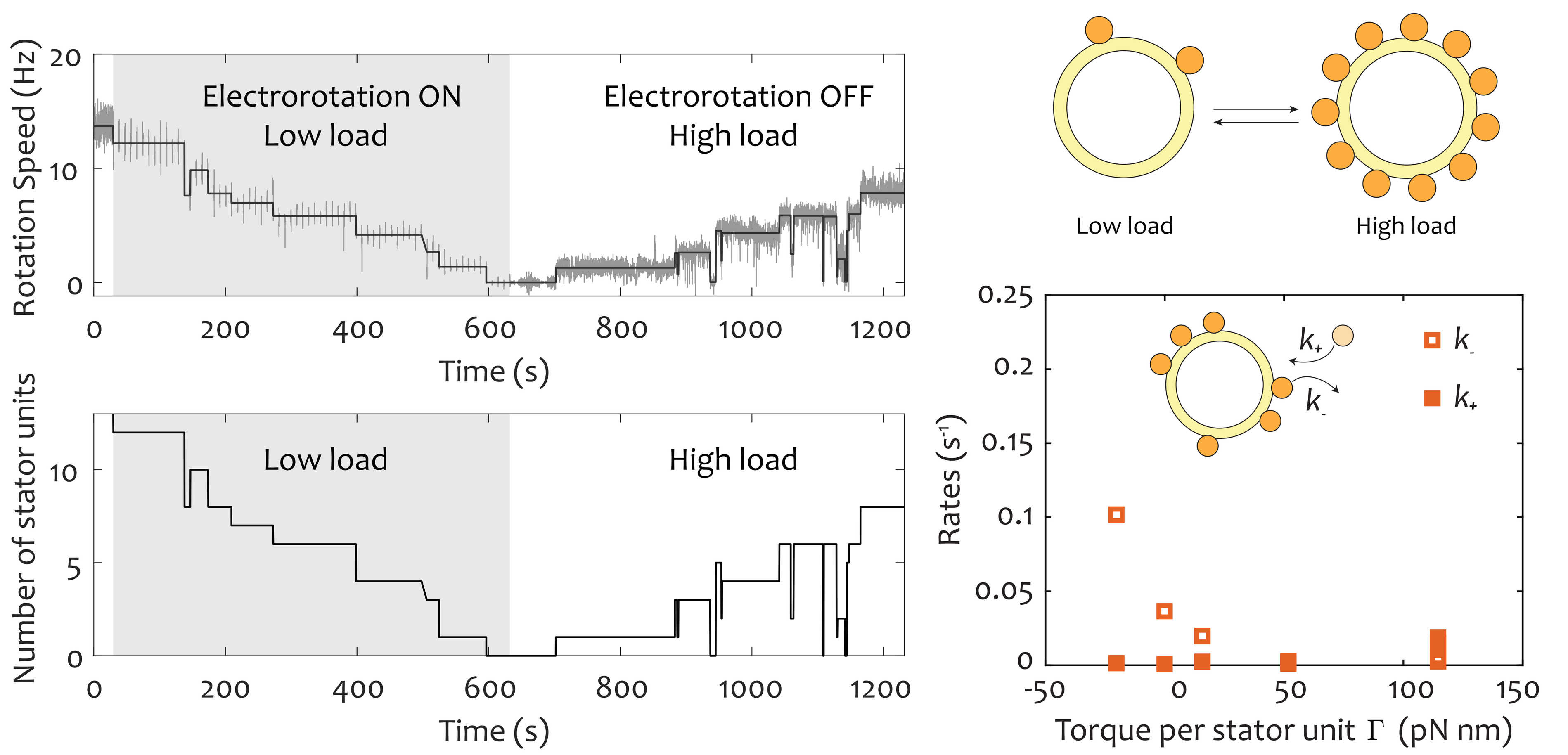
Methods

We performed *in vivo* load manipulation using electrorotation. We tethered *E. coli* to a surface via a short flagellar stub. In tethered cells, the motor rotates the cell body under high load. We then applied a high frequency rotating electric field, which exerted an external *assistive* torque on the cell. The external torque reduced motor load and increased cell rotation speed. Turning the field off increased load again. The amount of external torque could be tuned by changing the electric field strength. This technique enabled us to change the load on the motor instantaneously, reversibly, and controllably.



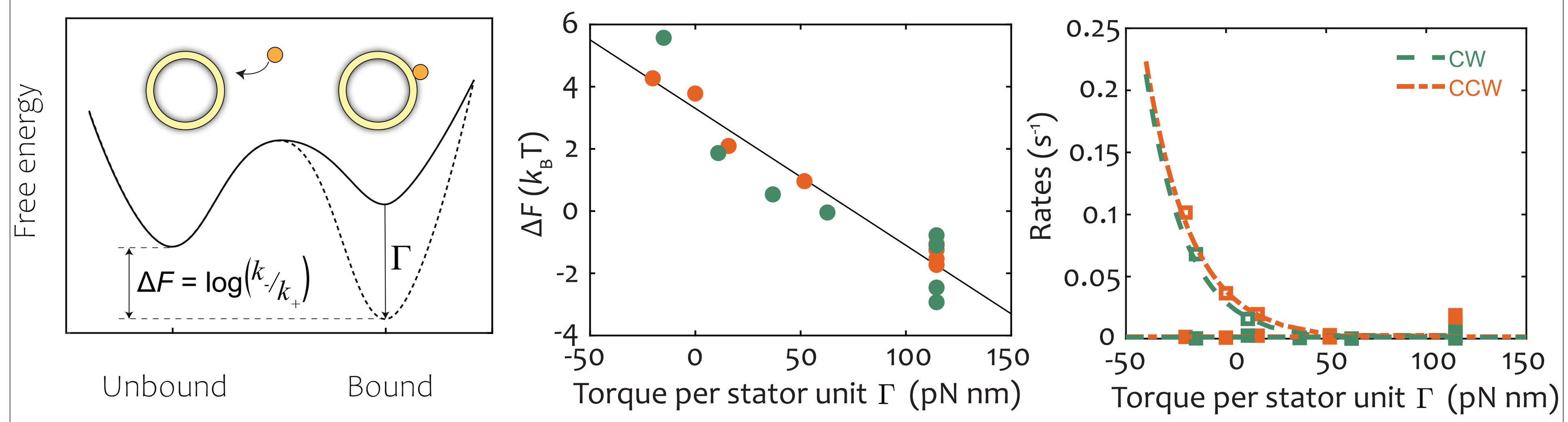
Load change triggers stator remodeling

1. A reduction in load triggers the release of stator units from the motor while an increase in load promotes stator unit recruitment.
2. The off rate of bound stator units decreases with torque.



Motor torque controls stator remodeling

1. Torque lowers the free energy of bound stator units, making the bound state more favorable.
2. Remodeling kinetics from clockwise (CW) and counterclockwise (CCW) rotating motors collapse on to the same curve if plotted against torque.
3. These data suggest that torque is the primary parameter that controls stator remodeling.



15s-Summary

Question: How does the bacterial flagellar motor deal with changes in mechanical load?

Method: We used electrorotation to experimentally manipulate the load on the motor and observed its response.

Results: 1. The motor gains or releases torque-generating stator units with an increase or decrease in load.
2. The unbinding rate decreases with motor torque, making the stator more stable at high load.
3. This mechanosensitive remodeling of the flagellar motor is independent of its direction of rotation.

Discussion: The flagellar motor is a complex biological machine, which tunes its assembly to the external mechnical environment. We now understand the physical rules that govern its dynamic self-assembly.

Discussion

In this research, we have investigated how a large macromolecular machine, the bacterial flagellar motor, adapts to changes in mechanical load by remodeling itself. Unbinding of torque-generating stator units from the motor is tuned by their torque production. This mechanism enables auto-regulation of stator assembly, ensuring that the motor's output (torque) keeps up with demand (load). Autonomous control of self-assembly may be a general strategy for tuning the functional output of protein complexes. This strategy has the advantage that the assembly/disassembly can be controlled directly, allowing the cell to quickly respond and adapt to sudden changes in the environment.

References

1. Wadhwa, N., Phillips, R. and Berg, H.C., 2019. Torque-dependent remodeling of the bacterial flagellar motor. *PNAS*, 116(24), 11764-11769.
2. Wadhwa, N., Tu, Y. and Berg, H.C., 2021. Mechanosensitive remodeling of the bacterial flagellar motor is independent of direction of rotation. *bioRxiv*, 2021.01.19.427295