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Structure, Charged Residue, and Temperature's Effect of Bacterial Flagellar Motors

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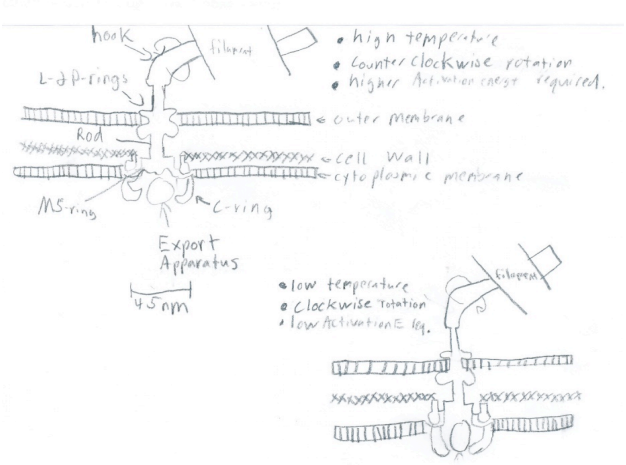
Bacteria with flagella produce movement by rotating small helical filaments, each driven by a reversible rotary motor. This motor is about 45 nanometers in diameter and is assembled via 20 different parts. It is powered by an ion flux. Scientists are studying this bacterial motor in order to figure out how such an efficient motor works. Also, if they are able to figure out how it works, they intend on answering questions concerning how to stop or destroy it. These motors power bacterial movement, which is also a way that the bacterial can avoid chemicals, such as antibiotics. Advancements in the understanding of bacterial flagellar motors will hopefully lead to advancements in stopping or slowing of infections. What is the structure? What charged residues affect its function? How does temperature effect the motor? These are questions that the researchers in the articles in analysis are asking. The major obstacle in answer these question is the size of these bacterial motors. They are smaller than most microscopes can resolve, therefore extremely advanced techniques have been employed.

Introduction

Charged residues play a huge role in the bacterial flagellar motor. We understand that the flagellum uses chemical energy to power the motor, but the molecular mechanisms to power it are not fully understood. Within the charged residues article, they find, through genetic analyses, that there are a few of the 50 proteins are closely involved with the torque generation. It was found through mutation testing that the FliG Mot- protein is the most directly involved to the torque generation, which means there are charge residues within that protein that are related. The charged residues on that protein, Arg279 Asp286 Asp287, were found to be directly involved in torque generation. The second article is concerned with the effect of temperature and the motor. It is understood that chemotaxis is the main factor in controlling the direction of rotation of the flagellar motor, but the motor spins both clockwise and counterclockwise. What isn't well understood is the switching, in particular, the factors and mechanisms that control the switching of directions. In this article, the scientists focus on the effect of temperature by removing the chemotaxis signaling proteins and observing the direction of rotation at different temperatures. In the last article, researchers work towards revealing the structure of the rotor. Revealing the structure of the rotor has encountered a major obstacle, the maximum ability of resolution of microscopy. Visible microscopy's maximum resolution is 200 nanometers, which is set by the diffraction limit. These scientists implemented more advanced microscopy techniques, Electron Cyromicroscopy and Single-particle Image Analysis in order to gain a better understanding about the function of the bacterial flagellar motor. Most research performed on the bacterial flagellar motor has been causality based. For example, creating mutations in the genetic code and observing its effect. Therefore, in the pursuit of obtaining more definitive knowledge about the proteins in question, direct observation must occur.

Recent Progress

Charged residues Arg279, Asp286, and Asp287 were believed to be all jointly involved in torque generation. Mutation of just two of the residues still resulted in the cell being motile, but when researchers performed a triple mutation of FliG, no motile cells were observed. Interestingly, these mutations only kept the motor from rotating. Triple mutation did not discourage the flagellar assembly. Some mutations of other proteins affected motility as well, but usually only about 10% of the impact that the triple mutation of Arg279, Asp286, and Asp287. "Although the precise function of those three residues are not known, but it is clear that their charges are important." (Lloyd SA, Blair DF 735) Advances in microscopy are required in order to study the structure of the FliG protein to gain a better understanding of these residues. Results of temperature's effect on bacterial flagellar motors found that at room temperature the cells spun only in the CCW (counterclockwise) direction. Motor directions began to change below 10 degrees C. At 12 degrees Celsius it was 100% CCW, at -2 degrees it was 100% CW(clockwise). It is concluded that activation energy plays a large role in what direction the flagellar spins. In the basis of standard free energy and standard enthalpy and entropy changes, a lower energy can be obtained by lowering the temperature. Therefore, since CCW only happens at a higher temperature, it seems that CW requires a higher activation energy. Although a direct effect of temperature seems likely, there is still a possibility that there is an indirect effect due to changes in the physiological state of the cell. This is an interesting conclusion because it suggests that bacterial have no internal control over their flagella and that external temperature is the main factor. That would be like your legs not being able to function because it is cold outside. Research concerning the structure of the rotor of the flagellar motor was the first to discover "new" aspects about the structure of the bacterial flagellar motor. In fact, it is the first to resolve 3



dimensional structure pictures of the Flif and FliG rings by single particle image analysis. They were not able to find the exact position of FliG. It is believed that the FliFG ring is quite flexible, which causes the position to be disordered and harder to find. The research in this area is not lost; however, it infers that the there could be some elastic element that is part of the torque generation.

Discussion

The results of the temperature began a good step in the right direction to find what variables cause which direction of rotation to occur. Activation energy is a critical part of many processes that support life. When activation energy is brought up, it is a good idea to acknowledge enzymes and their ability to lower required activation energy. Some of the signaling chemotaxis proteins were removed in order to observe the effect of temperature, but what if those proteins were indirectly partnered with an enzyme that may lower the activation energy? Therefore, the temperatures said to cause CCW, 12 degrees, may actually be a bit higher than the bacterial require in the wild. Secondly, charged residue research results may have been a small step in understanding the functionality of the flagella motor, but conclusively have been a large step in the pharmaceutical goal to stop or slow infections. The study did not conclude exactly how the residues work together, or how the other residues effect the motor, or even why the other residues effected the motor's capacity to a lesser magnitude, but it did give causal data that can be used in figuring out a way to render an aggressive bacteria's motility. Lastly, rotor structural research's discovery of the alpha helixes mutual interlocking of the axial part of the flagellum is important because it may be a critical factor to the stability of its section. That part of the flagellum would be under a lot of kinetic stress when the rotor is spinning, so it must be extra durable to withstand the forces. Although they were not able to find the position of FliG, inability of being able to pinpoint its location gave a clue that maybe it is because the position is disordered which infers it to be part of an elastic part. This makes sense because when something is spinning and absorbing stress an elastic piece to absorb stress would be more effective than a rigid section unable to absorb stress. Further microscopy advancement is imperative to the success of learning the truth about what actually exists through direct observation.

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