Up Close and Personal: The Process of Mycobacterial Cell Division

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Abstract

Bacterial cell growth and division occurs as a sequence of events for which many processes remain only partially understood. Simple questions such as "do single cells grow linearly or exponentially", or "do cells grow symmetrically or asymmetrically" are still a subject of intense debate. What governs the position and timing of cell division is still largely unclear for many bacteria. In most bacterial models, FtsZ-ring formation is believed to be the first event leading to division. Nucleoid segregation, septum formation, cytokinesis, and physical cell separation are all successive events in the process of division [2]. To date, many of these processes have been characterized using static methods. Establishing the sequence and possible dependence of these processes however requires time resolved live-cell imaging [3, 4], at high resolution. Here we present a concise time-sequence of events describing division of Mycobacterium smegmatis, a non-pathogenic cousin of Mycobacterium tuberculosis. We used a combination of multi-day time-lapse atomic force microscopy (AFM), time-lapse fluorescence microscopy with real time measurements of the cell separation at timescales down to 10s of milliseconds. Combining the nanoscale 3D information from AFM and the biochemical specificity from fluorescence microscopy we have characterized cell division from the early stage pre-selection of division sites, to assembly and subsequent disassembly of the FtsZ ring, localization of Wag31 and cytokinesis all the way to the rapid cell separation. Contrary to what is believed to be true in many bacterial cell types, cell separation in Mycobacterium smegmatis isn't a gradual event, but occurs abruptly within 10s of milliseconds, resembling more a mechanical fracture than a cellular remodeling process. Using mechanical stimulation, we demonstrate that the build-up of mechanical stress governs the time and place of cell separation. By applying additional mechanical stress, we were able to initiate cell separation at times in the cell cycle well before it would occur in the non-stimulated case. These observations suggest a new model for the late stages of cell division in Mycobacterium smegmatis, where cell mechanical properties and local stress concentration govern the timing and place of cell separation.

References

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