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Cell cycle dependent oscillations of gene expression in E. coli

A long-standing hypothesis sees DNA replication control in *E. coli* as a central cell cycle oscillator at whose core is the DnaA protein. The consensus is that the activity of the DnaA protein, which is dependent on its nucleotide bound state, is an effector of initiation of DNA replication and a sensor of cell size. However, while several processes are known to regulate the change in DnaA activity, the oscillations in DnaA production and DnaA activity have never been observed at the single cell level, and their correlation with cell volume has yet to be established. Here, we measured the volume-specific production rate of a reporter protein under control of the *dnaA*P2 promoter in single cells. By a careful dissection of the effects of DnaA-ATP- and SeqA-dependent regulation of dnaAP2 promoter activity two distinct cell cycle oscillators emerge. The first one, driven by both DnaA activity and SeqA repression, is strongly coupled to cell cycle and cell size, and its minima show the same behaviour as initiation events, following each other by a constant added size. The second, a reporter of DnaA activity, is still coupled with cell size but not to the time of cell division and the added size between its minima is dependent on the cell size. These findings suggest that while DnaA activity and gene dosage perform volume sensing, SeqA repression primes the DnaA oscillator to follow initiation of DNA replication, thus setting the cell size of initiation of the next replication round in the daughter cells.

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