Summary for the general public

Looking through the microscope on bacterial cells taken from a certain environment we could be astonished how similar in shape and size they are. Two very similar bacterial cells arise after division of a mother cell, however before this happens, bacterial genetic material has to be faithfully replicated and separated to the daughter cells. In addition, many bacterial species can multiply so fast that the period between subsequent cell divisions is two-fold shorter than the time required to duplicate their genome. This is possible because grandmother cell initiates synthesis of the genetic material that in its complete form, becomes a heritage of her granddaughter. But even living at such a speed, bacteria are still able to flawlessly replicate their DNA, segregate it and keep the characteristic size of progeny cells! Equally astonishing is the fact that despite knowing a plethora of molecular details of bacterial cells, we still don't understand how they really do it. The question how bacteria coordinate DNA replication with cell growth and division is a classic in microbiology, however the clear answers are still missing. Finding answers to this question is important not only for our understanding how various processes in the cell cooperate to make one living system. It has also practical implications, because destroying these coordinating mechanisms could impede survival and division of pathogenic bacteria in their host organism. Thus, these mechanisms are attractive targets for the development of new antibacterial therapeutics. This is of particular importance in the era of fast increase in the occurrence of multidrug-resistant bacteria. In our project we propose to investigate one of the mechanism that may be crucial for bacterial cells to choose the right moment to start duplication of their genetic material while the cell grows. It relies on interaction of one of the products of biochemical reactions that occur in bacterial cell (metabolite) with one of their proteins known to take part in the regulation of DNA replication, dubbed DiaA. Changes of the concentration of this metabolite, which is also a substrate to synthesize nucleotides (components of DNA) and cell envelope, could be a signal to trigger DNA replication, while DiaA could transmit this signal to protein machinery which carries out the duplication of the genetic material. To verify this hypothesis we will use both classical biochemical methods and modern biophysical approaches which will enable us to estimate the impact of binding of the metabolite by DiaA on the activity of this protein. Moreover we plan to test whether by changing the intracellular concentration of the metabolite we can affect various parameters of the cell cycle. To achieve this we will use flow cytometry, a method which allows to monitor and statistically analyze processes in many cells simultaneously. We will also employ a modern method (tandem gas chromatography-mass spectrometry) to measure how the level of most metabolites present in the cell changes during the cell cycle, from the time when a new cell is born to the next division.