Adaptive Laboratory Evolution of Antibiotic Resistance Using Different Selection Regimes Lead to Similar Phenotypes and Genotypes

Antibiotic resistance is a global threat to human health, wherefore it is crucial to study the mechanisms of antibiotic resistance as well as its emergence and dissemination. One way to analyze the acquisition of de novo mutations conferring antibiotic resistance is adaptive laboratory evolution. However, various evolution methods exist that utilize different population sizes, selection strengths, and bottlenecks. While evolution in increasing drug gradients guarantees high-level antibiotic resistance promising to identify the most potent resistance conferring mutations, other selection regimes are simpler to implement and therefore allow higher throughput. The specific regimen of adaptive evolution may have a profound impact on the adapted cell state. Indeed, substantial effects of the selection regime on the resulting geno- and phenotypes have been reported in the literature. In this study we compare the geno- and phenotypes of Escherichia coli after evolution to Amikacin, Piperacillin, and Tetracycline under four different selection regimes. Interestingly, key mutations that confer antibiotic resistance as well as phenotypic changes like collateral sensitivity and cross-resistance emerge independently of the selection regime. Yet, lineages that underwent evolution under mild selection displayed a growth advantage independently of the acquired level of antibiotic resistance compared to lineages adapted under maximal selection in a drug gradient. Our data suggests that even though different selection regimens result in subtle genotypic and phenotypic differences key adaptations appear independently of the selection regime.