Bacterial genome plasticity and its impact on adaptation to metal contamination

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Metal homeostasis is important for all bacteria since they have to react swiftly to both scarcity and excess of either essential or toxic metals. To defend themselves against high metal toxicity, bacteria depend on multiple resistance mechanisms. In addition, bacteria continuously evolve to survive different environmental challenges. This evolution results from natural selection acting on variability in populations.

We scrutinized the adaptive potential of *Cupriavidus metallidurans* CH34 to increased zinc concentrations. *C. metallidurans* CH34 is a well-studied metal-resistant β-proteobacterium that contains a very high number of genes involved in the resistance and processing of metals.

First, an adaptive laboratory evolution experiment was performed to obtain derivatives adapted to grow in the presence of zinc concentrations above the minimal inhibitory concentration for the parental strain. Genetic characterization revealed the inactivation of a DeoR-type transcriptional repressor by insertion of the insertion sequence element IS*1088*, resulting in the constitutive expression of the neighboring ABC-type sugar transporter.

Next, the adaptive potential of a CH34 derivative lacking the main zinc resistance determinants was scrutinized. Again, this derivative appeared to be able to increase its zinc resistance level. Late-occurring zinc resistant variants likely arose in response to the selective conditions, as they were enriched in adaptations caused by specific insertion sequence elements whose transposase expression was found to be zinc-responsive. Finally, further deletion of the latter adaptation potential still enabled adaptation by transposition of insertion sequence elements (IS*Rme5* and IS*1086*) that provided outward-directed promoters.

Thus, our results indicated that *C. metallidurans* is able to adapt to increased zinc concentrations, even if its dedicated zinc resistance mechanisms are sequentially removed. In addition, transposition of insertion sequence elements play a multifaceted, pivotal role in adaptation and can even be induced by stress conditions.