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Evolution of robustness to protein mistranslation by accelerated protein turnover

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Translational errors occur at high rates, influence organism viability and the onset of genetic diseases. To investigate how organisms mitigate the deleterious effects of protein synthesis errors during evolution, a mutant yeast strain was engineered to translate a codon ambiguously (mistranslation). It thereby overloads the protein quality control pathways and disrupts cellular protein homeostasis. This strain was used to study the capacity of the yeast genome to compensate the deleterious effects of protein mistranslation. Laboratory evolutionary experiments revealed that fitness loss due to mistranslation can rapidly be mitigated. Genomic analysis demonstrated that adaptation was primarily mediated by large-scale chromosomal duplication and deletion events, suggesting that errors during protein synthesis promote the evolution of genome architecture. By altering the dosages of numerous, functionally related proteins simultaneously, these genetic changes introduced large phenotypic leaps that enabled rapid adaptation to mistranslation. Evolution increased the level of tolerance to mistranslation through acceleration of ubiquitin-proteasome mediated protein degradation and protein synthesis. As a consequence of rapid elimination of erroneous protein products, evolution reduced the extent of toxic protein aggregation in mistranslating cells. However, there was a strong evolutionary trade-off between adaptation to mistranslation and survival upon starvation: the evolved lines showed fitness defects and impaired capacity to degrade mature ribosomes upon nutrient limitation. Moreover, as a response to an enhanced energy demand of accelerated protein turnover, the evolved lines exhibited increased glucose uptake by selective duplication of hexose transporter genes. We conclude that adjustment of proteome homeostasis to mistranslation evolves rapidly, but this adaptation has several side-effects on cellular physiology. Our work also indicates that translational fidelity and the ubiquitin-proteasome system are functionally linked to each other and may, therefore, co-evolve in nature.

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Effect of essential oils and their main components on biofilm formation and quorum sensing of food-related microorganisms

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Food contamination and food poisoning are serious problems in the food industry and also in health care. Bacterial biofilms are a community of bacteria imbedded in a self-synthesized extracellular matrix. It can form easily on food surfaces and in processing environment, representing a problem of post-processing contamination and cross-contamination. These structures can be formed by bacteria and yeast as well, but the most common forms in nature are the mixed-species biofilms. Biofilm-forming bacterial cells are able to communicate by the density-dependent cell-to-cell communication mechanism called quorum sensing (QS). A variety of different molecules can be used as signals for communication. Common classes of signaling molecules are oligopeptides in Gram-positive bacteria, N-acyl homoserine lactones (AHLs) in Gram-negative bacteria and autoinducer-2 (AI-2) in both Gram-negative and Gram-positive bacteria. Potentially, the resistance of biofilm-forming microorganisms could be influenced by controlling QS and biofilm formation.

In the food industry conventional preservatives often produce unpleasant by-products. Therefore, it is important to find effective natural antimicrobial agents which replace the synthetic ones at least partially. Among others, various essential oils (EOs) could be used against food spoilage- and food-borne pathogens. EOs are volatile liquids obtained from herbs, spices and different plants mainly by steam distillation. Some of them have more than 50 different constituents. In some cases these oils can help prevent the formation of biofilms of both food-spoilage and food-borne pathogens and could be used as natural preservatives for the extension of shelf life of foods.

The present study focuses on the anti-biofilm forming and anti-QS effect of six EOs (cinnamon, clary sage, juniper, lemon, marjoram and thyme) and their main components (cinnamaldehyde, α-pinene, limonene, linalool, terpinene-4-ol and thymol) against five food spoilage bacteria, a food borne pathogen, a yeast and their mixed cultures. Besides lemon, EOs showed good anti-biofilm forming effect and also inhibited QS of *Chromobacterium violaceum* in most cases. Scanning electronmicroscopy images showed the disappearance of biofilm-specific structures, furthermore, the lab-on-a-chip measurements revealed quantitative changes in the protein profile of bacteria after these treatments. The biofilm formation of the food borne pathogen *Listeria monocytogenes* was also inhibited on chicken breast fillets after marinating it with thyme EO and thymol. Additionally, the oils added also had a pleasant flavour effect. In conclusion, the EOs tested are good candidates for food preservation and represent alternatives to synthetic additives.

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