Title
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Evolution and Stability in a Syntrophic Community

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Mutualistic relationships between species may often originate from an opportunistic exchange of byproducts produced by each species, but few have addressed how microorganisms evolve in response to these interactions. We used experimental evolution to observe this process in real time in an interaction between the bacterium *Desulfovibrio vulgaris* and the archeaon, *Methanococcus maripaludis*. This mutualism based on the exchange of hydrogen represents an interaction that is fundamental to many microbial communities. After 300 generations of evolution , we compared 20 independently evolved cocultures to cocultures with ancestral *D. vulgaris* and *M. maripaludis*. All but one of the evolved cocultures could grow significantly faster and obtain a much higher yield from the same resource. We tested whether these changes could be attributed to *D. vulgaris*, *M. maripaludis*, or both species by analyzing mixtures of evolved and ancestral populations and found that increases in coculture yield were caused by the combined adaptations of both species. Whether *D. vulgaris*, *M. maripaludis*, or both contributed to improvements in coculture growth rate depended on the heterogeneity of the evolution environment. Several cocultures had evolved in tubes that were not shaken, so that gradients of hydrogen could form during incubation. Both *D. vulgaris* and *M. maripaludis* that evolved in this environment were capable of enhancing syntrophic growth rate when paired with the ancestor, but *M. maripaludis* that evolved in a constantly shaken homogeneous environment had little if any effect. The evolutionary process leading to these adaptations involved dramatic changes in stability of cocultures over time. After ~30 generations of evolution, cocultures began occasionally entering phases of very slow growth, but eventually these phases stopped, suggesting that one or both species fixed mutations that would stabilize the coculture. Together, these results show that the *D. vulgaris* and *M. maripaludis* strains we studied have a capacity for rapid evolutionary improvements in syntrophic growth, but the process enabling this change depends on the heterogeneity of the environment, and may cause instability in the community.