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Building a minimal cell-The JCVI design-build-test cycle for synthetic cells

The synthetic biology group at the J. Craig Venter Institute is building a bacterial cell that contains only the core set of genes required for life in the laboratory. Our group has worked on the complete chemical synthesis and installation of bacterial genomes for a number of years. In the course of this work we developed methods for designing, building, and testing bacterial genomes. Many of these methods can be applied to natural genomes as well as synthetic ones. A key to our approach is the cloning of complete bacterial genomes as extra chromosomes in yeast. For example, we can insert a yeast vector composed of a yeast centromere, a yeast origin of DNA replication, and a yeast selectable marker, into the bacterial genome. Then the genome can be released from the bacterial cell, and introduced into spheroplasts of yeast cells, by PEG mediated transformation. In yeast the bacterial genome is propagated stably as a yeast centromeric plasmid (YCp), which is essentially a circular yeast artificial chromosome. This method was developed using mycoplasmas, which are bacteria with very small genomes and lacking cell walls, and we have recently extended it to several other bacterial species. We have also been able to assemble complete mycoplasma genomes from synthetic DNA pieces, using yeast recombination. These methods for cloning bacterial genomes in yeast allow us to bring the power of yeast genetics to bear on engineering bacterial genomes. We also developed a method that we call genome transplantation, which can be used to reinstall the genome of the mycoplasma *M. mycoides*, isolated from yeast, into a bacterial cell. This allows us to test a genome engineered in yeast for viability. These methods comprise a design-build-test cycle that we are using to produce a *M. mycoides* cell with a minimized genome.

Biography

Clyde Hutchison is distinguished investigator at the J. Craig Venter Institute in San Diego, California, where he is a member of the Synthetic Biology Group headed by Hamilton Smith. He is also Chair of the Scientific Advisory Board of Synthetic Genomics, Inc. In 1995 he was elected to membership in the National Academy of Sciences.

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