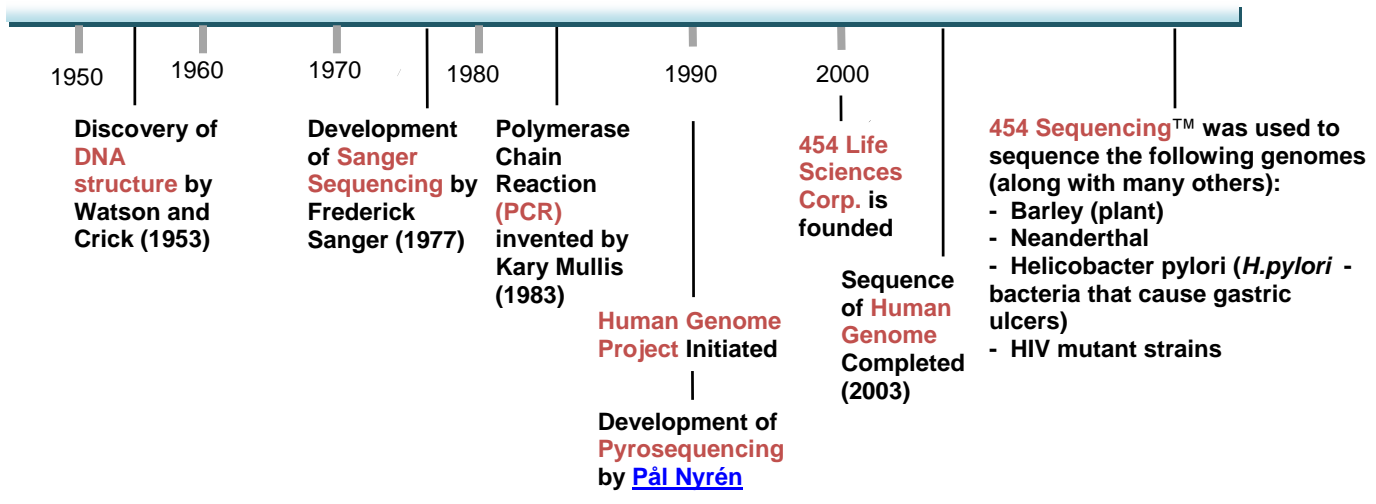


History of Genome Sequencing – The Sanger Method

The sequencing of DNA molecules began in the 1970s with development of the Maxam-Gilbert method, and later the Sanger method. Originally developed by Frederick Sanger in 1975, most DNA sequencing that occurs in medical and research laboratories today is performed using sequencers employing variations of the Sanger method. Termed the chain-termination method, it involves a reaction where chain-terminator nucleotides are labeled with fluorescent dyes, combined with fragmented DNA, DNA sequencing primers and DNA polymerase. Each nucleotide in the DNA sequence is labeled with a different dye color and a chromatogram is produced, with each color representing a different letter in the DNA code – A, T, C, or G. Advances in sequencing technology and computer programming enabled relatively fast and cost efficient DNA sequencing. However, sequencing of entire genomes of organisms was difficult and time consuming. At the time the Human Genome Project was officially started in 1990, it was thought that sequencing the human genome would take fifteen years. The sequence was released in 2003, although some gaps still exist. The estimated project cost for the entire Human Genome Project is **around \$3 billion**, although this figure represents a wide range of scientific activities that went into the project, not exclusively genome sequencing. Optimally, current sequencers are able to sequence approximately 2.8 million base pairs per 24 hour period. However, even the smallest organisms such as bacteria are hundreds of millions of base-pairs long and the human genome is about 3 billion (3,000,000,000) base pairs. At this rate, using the most modern Sanger sequencers, it takes almost **three years** to sequence the human genome.

History of Genome Sequencing



Source: U.S. Department of Energy Office of Science, Systems Biology for Energy and the Environment, Human Genome Project Information
 Base URL: <http://genomics.energy.gov/>

Race to the Finish Line – Celera’s “Shotgun” Method

The race to sequence of the human genome heated up in 1998 when Rockville, Maryland-based Celera Genomics, a private company under the leadership of Dr. Craig Venter, entered the scene. Celera Genomics adopted an alternative “shotgun” method to sequence the human genome: this technology provided a faster means to analyze the entirety of genetic data, although some researchers argued less reliable. Celera received their validation in early 2000 when they successfully sequenced the *Drosophila* (fruit fly) genome. A year later, Celera would tie with the Human Genome Project in successfully sequencing the human genome; however, compared to the government-based Human Genome Project, Celera accomplished their goal in three years as opposed to 13 and in a far more viable way. The Celera genome project ended up costing around \$300 million dollars, a 90% price reduction when compared to the estimated \$3 billion dollars for the HGP.

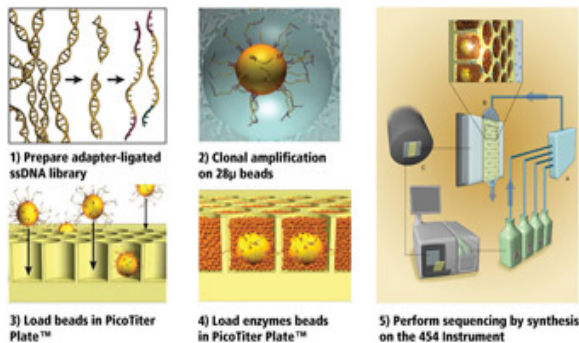
Human Genome Project’s open-access policy opened the floodgates for academic circles and researchers of genetic disease, while Celera opted for a more commercially-oriented approach of academic non-disclosure. However, Celera eventually opened its data to the public community and remains to this to day one of the most important factors in making genome sequencing into a financial and scientific reality.



Source: U.S. Department of Energy Office of Science, Systems Biology for Energy and the Environment, Human Genome Project Information
Base URL: <http://genomics.energy.gov/>

Development of 454 Sequencing™

Pyrosequencing was originally developed by [Pål Nyrén](#) in the 1990s. This method is commonly referred to as sequencing-by-synthesis. Essentially, a DNA strand is sequenced by synthesizing the complementary strand. Each time a nucleotide, (A, T, C or G) is incorporated, a series of enzymatic reactions result in a light signal that can then be read by an analyzer. 454 Life Sciences further optimized this method by utilizing the well-known Polymerase Chain Reaction (PCR). In short, 454 Sequencing™ involves breaking up genomic DNA into fragments, placing the individual fragments onto specially designed microbeads where the many copies of each fragment are made via PCR. The amplified fragments are then loaded onto very small wells on a special plate (PicoTiterPlate™). As the wells are loaded with samples, reagents are pumped across the plate. The addition of the reagents result in an enzymatic reaction between complimentary bases in the DNA fragments, and a light signal is created and read by the Genome Sequencer FLX analyzer. The Genome Sequencer FLX from 454 Life Sciences can sequence 100 million bases per 8 hour instrument run or approximately 300 million base pairs per 24 hour period. At this rate the human genome can be sequenced in about **ten days**.



454 Sequencing™

Sources:

454 Life Sciences Corporation

Nyrén, P., [The History of Pyrosequencing](#), Methods Mol Biol., 2006; 373:1-14

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