Sequencing DNA at the Advanced Center for Genome Technology, Univ. of Oklahoma

1. Genomic DNA from organism of interest

2. Nebulization to physically shear genomic DNA into small random fragments

3. Ligation of 2-4Kbp fragments into a vector, pUC18, to form a circular plasmid, electro-transformation to move the plasmid into *E. coli* strain XL1blue-MRF

4. White *E. coli* colonies, which carry the plasmid, are picked using the Flexys robot to inoculate broth for growth of cell in an aerated incubator called the HiGro

5. Plasmid DNA isolation is done semiautomatically on a Biomek 2000 or a Hydra, agarose gel electrophoresis is performed to examine the yield of plasmid/template DNA

6. DNA templates containing the subfragments are used to generate nested sets of ssDNA with Fluorescently labeled dideoxynucleotide terminators using Taq polymerase

7. Automated sequencing on ABI 3700, ABI377, Megabace, or Basestation of the labeled ssDNA: gel or capillary electrophoresis, detection of the 4 fluorescent dyes and base identification in the short ~500 base sequences

8. Assembly of short DNA sequence data using the computer program, Phrap, into a single large sequence representing the original genomic DNA.

Locating gene regions on the complete DNA sequence using the computer to compare to other known genes or predicting genes.
1859 Darwin’s Origin of Species

The first printing of Charles Darwin’s book, *On the Origin of Species by Means of Natural Selection*, presented the idea of evolutionism to the world. Darwin described his idea in a narrative form, emphasizing the gradual change in species over time due to natural selection. His ideas were well received, and his work laid the foundation for modern evolutionary biology.

1865 Mendel’s peas

Mendel’s work, published in 1865, described the principles of heredity. He studied pea plants, which have clear and easily observable traits, and conducted experiments to understand how traits are passed from parent to offspring. Mendel’s experiments showed that traits are inherited in a predictable manner, and his work is now considered the foundation of modern genetics.

1953 DNA double helix

Francis Crick and James Watson described the double helix structure of DNA. By the time Watson and Crick turned their attention to solving the chemical structure of DNA, it was known to possess a helical structure. They proposed that DNA consists of two strands that wind around each other, held together by hydrogen bonds between specific pairs of bases.

1975-77 DNA sequencing

Sanger and his colleagues developed methods for sequencing DNA, which allowed scientists to determine the exact sequence of DNA bases in a sample. This was a major breakthrough in understanding the molecular basis of heredity and paved the way for genetic research.

1999 Chromosome 22

In December 1999, the Human Genome Project completed the first full-length sequence of a human chromosome—chromosome 22. This accomplishment demonstrated the ability of the Human Genome Project to map the human genome with unprecedented accuracy and provided a blueprint for understanding human genetics.

2003 Planned completion of human sequencing

Researchers aim to sequence the entire human genome, which will provide a comprehensive understanding of human genetics and the basis for personalized medicine and genetic therapies.
REVIEW QUESTIONS:

1. What are the three components of DNA? ______________________
   ______________________________________________________

2. What is the human genome? Why is it important to study the human genome?
   ______________________________________________________
   ______________________________________________________
   ______________________________________________________

3. Which other organisms’ genome might be important to know? __________
   ______________________________________________________

4. What are the four bases contained in DNA? What does Dr. Roe call his lab?
   ______________________________________________________
   ______________________________________________________

5. How many chromosome pairs does a human have? ________________
   ______________________________________________________

6. In what year did Watson and Crick describe the double helix structure of DNA?
   ______________________________________________________

7. Which was the first full-length human chromosome sequence to be completed?
   ______________________________________________________

8. Dr. Roe’s lab sequences DNA by attaching colored markers to the growing ends of DNA chains. What is this method of sequencing called? (i.e., who is it named for?)
   ______________________________________________________
The Human Genome Project

and

Ethical, Legal, and Social Issues

The information contained in these pages was obtained from the following web sites:


Additional information was obtained from an educational CD-ROM published by Nature.

"The United States Department of Energy and the National Institute of Health have devoted 3% to 5% of their annual Human Genome Project budgets toward studying the ethical, legal, and social issues surrounding availability of genetic information. This represents the world's largest bioethics program, which has become a model for ELSI programs around the world."

The ethical, legal, and social issues program was established coinciding with the inception of the Human Genome Project. The ELSI program has several goals for their research:

9. Examine the issues surrounding the completion of the human DNA sequence and the study of human genetic variation.

10. Examine issues raised by the integration of genetic technologies and information into health care and public health activities.

11. Examine issues raised by the integration of knowledge about genomics and gene-environment interactions into non-clinical settings.

12. Explore ways in which new genetic knowledge may interact with a variety of philosophical, theological, and ethical perspectives.
13. Explore how socioeconomic factors and concepts of race and ethnicity influence the use and interpretation of genetic information, the utilization of genetic services, and the development of policy.

The following questions are examples of the research questions facing the ELSI Research and Planning Group, and are intended to encourage examination of a variety of ethical, legal, and social issues.

1. Will the discovery of DNA polymorphisms influence current concepts of race and ethnicity?
2. How will individuals and groups respond to potential challenges to or affirmations of their racial and/or ethnic self-identification, based on new genetic information?
3. What are the clinical and societal implications of identifying common polymorphisms that predict disease susceptibility or resistance?
4. Will genetic testing promote risky behavior in persons found to be genetically resistant to particular pathogens, such as HIV, or environmental hazards, such as cigarette smoke?
5. How will individuals be benefited or harmed by the integration of genetic information into individual medical records, managed care organization records, and public health registries?
6. What are appropriate and inappropriate uses of genetic testing in the employee setting?
7. Are there conditions under which it might be ethical and/or legal to use genetic testing to identify those employees who may have had a susceptibility to workplace hazards?
8. What implications does the American with Disabilities Act have for such testing?
9. What are the implications of obtaining genetic information for use in adoption proceedings and establishment of child custody and child support?
10. What are the potential uses and abuses of genetic information in educational settings?
11. Is placement of students on the basis of genetic data any more or less beneficial or harmful than tracking on the basis of traditional categories or classifications?

12. What are the implications of genetic enhancement technologies for conceptions of humanity?

13. As new genetic technologies and information provide additional support for the central role of evolution in shaping the human species, how will society accommodate the challenges that this may pose to traditional religious and cultural views of humanity?

14. What ethical or theological challenges might be posed by the ability to alter the genetic makeup of future generations?

15. How have past misuses of genetic science and information influenced perceptions of genetic research and services among individuals from diverse communities and groups?

16. Will particular communities and groups be more vulnerable to employment discrimination based on genotype?

17. In what ways are access to, and use of, genetic information and services affected by ethnicity, race, or socioeconomic status?

18. What are the most effective strategies to ensure that genetic counseling and other genetic services are culturally sensitive and relevant?