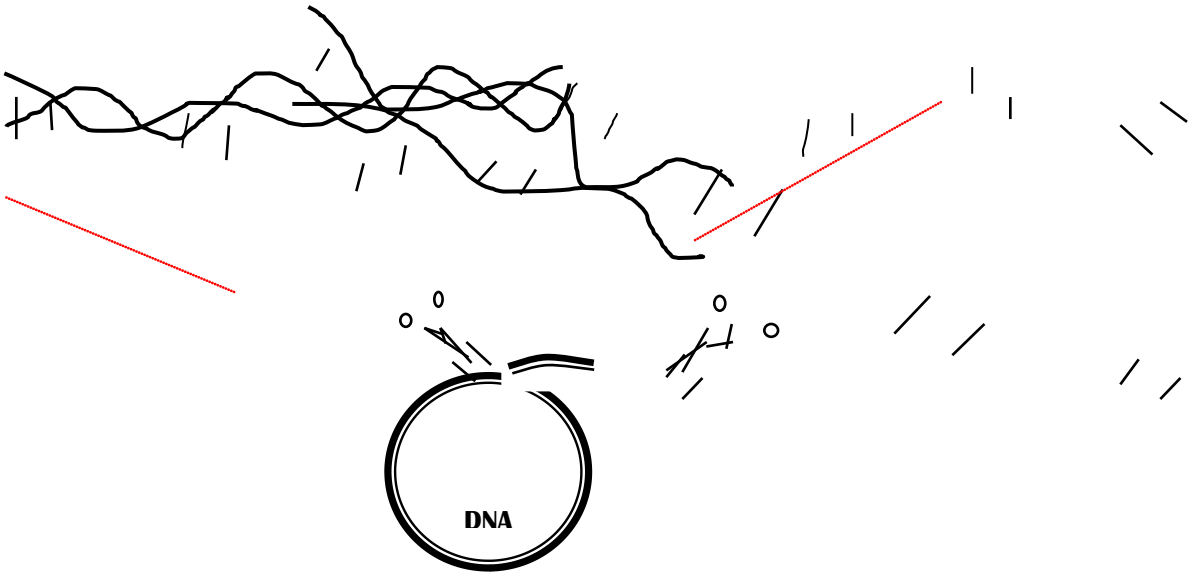
Department of Biological Sciences, KCC, CUNY



KINGSBOROUGH COMMUNITY COLLEGE
of
THE CITY UNIVERSITY OF NEW YORK
COURSE OUTLINE
FOR
BIOLOGY 58

Professor Sarwar Jahangir, *Ph.D.*
Office: Room S 206
Telephone: 718- 368-5743
e-mail: sarwar.jahangir@kbcc.cuny.edu

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Access-Ability Services (AAS) serves as a liaison and resource to the KCC community regarding disability issues, promotes equal access to all KCC programs and activities, and makes every reasonable effort to provide appropriate accommodations and assistance to students with disabilities. Please contact this office if you require such accommodations and assistance. Your instructor will be glad to make the accommodations you need, but you must have documentation from the Access-Ability office for any accommodations.

Access-Ability Services is always looking to hire student aides/federal work study students to help provide certain services for our students with disabilities, if you are interested please stop by D205 to find out more.

Disability Related Services D205

Office Hours:

Monday	9am-5:00pm
Tuesday	9am-8:00pm
Wednesday	9am-8:00pm
Thursday	9am-5:00pm
Friday	9am-5:00pm

By email: AAS@kbcc.cuny.edu By phone: 718-368-5175

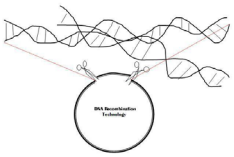
Recombinant DNA Technology Biology 58 CUNY Approved

Course Description

Biology 05800 - Recombinant DNA Biotechnology (4 crs. 6 hrs.)

It is a one semester exciting upper level course teaching the cutting-edge DNA technology who wish to major with A.S. in Biotechnology for a career in the biotechnology industry in NY City and elsewhere. The course is divided into two 2 hr lectures and two 3 hr laboratories each week for 12 weeks. Both the lectures and the laboratory exercises are geared to the study of molecular aspects of recombinant DNA technology, gene cloning, genetic engineering, gene expression in both prokaryotes and eukaryotes including plant, fungi and animal cells. It will emphasize biomedical, environmental, agricultural and aquatic biotechnologies along with ethical, legal, and social concerns. This is a prime course for A.S. in Biotechnology, and students on completion of the program will have the option to transfer to Brooklyn College and complete B.S. or B.A. in Biology.

Prerequisites: Bio 13, Bio 14, Chem 11, Chem 12, and Math 20 or equivalents, and passing CUNY Reading, Writing and Math exams.



DEPARTMENT OF BIOLOGICAL SCIENCES
KINGSBOROUGH COMMUNITY COLLEGE
BROOKLYN, NEW YORK 11235
JANUARY 2012
EDITION 1001

Biology 58: Recombinant DNA Technology

The 20th century was dominated by the discoveries in physics and chemistry, and the 21st century is predicted to be dominated by the discoveries and achievements of biology and biotechnology. The Recombinant DNA Technology is one of the pillars of biotechnology that is applied for gene cloning. KCC students graduating with AS in biology with concentration in biotechnology or AS in biotechnology will have the choice to take this course, and will add an attractive qualification to their already-excellent transcripts.

The Recombinant DNA Technology is a four credit one-semester course. It emphasizes and teaches the theories and techniques for gene splicing and insertion at new locations to achieve a desired goal. Using this technique genes or pieces of DNA may be transferred from one species to another – bacteria to fish, fish to bacteria, animals to plants and plants to animals and all that you can imagine.

Today, many therapeutic molecules including human insulin are produced by bacteria as a product of recombinant DNA technology. Construction of a recombinant DNA molecule involves splicing a DNA fragment with an enzyme(s) and rejoining the fragment in a novel arrangement and location linked by another enzyme(s). This produces a cloned gene or DNA. A gene clone with an appropriate combination may be transferred into a bacterium to produce millions of copies of the same gene. This makes a gene factory, and can become an easy source of a gene on demand for gene therapy, molecular medicine and other usage. Following this procedure, human insulin gene was transferred into a bacterium, *Escherichia coli,* to produce human insulin protein in bacteria. Many other therapeutic molecules are produced today including interferons (alfa-2a and alfa-2b) and granulocyte-macrophage colony-stimulating factor, epidermal growth factor and erythropoietin for tissue repair, human growth hormone, atrial metabolism modulators, vaccines for hepatitis B virus and melanoma, and tissue plasminogen activator, to name a few, using recombinant DNA technology. Soon, therapy for several genetic deficiencies will be available with the help of this technology.

In addition to the health industry, recombinant DNA technology produced insect resistant corn, potato and cotton and frost resistant tomato that alone saved hundreds of millions of dollars worth produce, and at the same time saving the environment from using hazardous insecticides and other chemicals in USA alone.

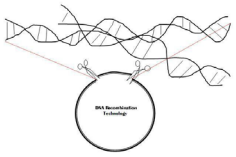
Hence, recombinant DNA Technology already acquired an important role in the field of medicine, food production, environmental protection, industry and business. Unfortunately, we do not have enough well-trained manpower in the pipeline to support the growth of the biotechnology industry today and tomorrow. We attempt in this course to train you in this exciting field so that you are invaluable for the growth of our industry, economy, and the nation.

We encourage you to put every effort to complete all the readings required and the topics assigned for every class lecture and laboratory exercise ahead of time. Coming prepared for every lecture and laboratory experiment will be the key to learn all that is essential and earn a high grade. Make sure to ask your professor that requires further clarification. Please be ***active*** in the class and keep in mind – learning is a two-way process between you and your professor, and you are the ultimate beneficiary of the process.

Your classroom learning may be synergized further by information in the news media, internet and others, outside the classroom. Keep an open eye and read the national newspapers, attend the specialized television programs and science gossips. Our *Public Television* often produces excellent programs on DNA technology and genetic engineering as well as their scope and impacts. The Science Times and the *Scientific American* also produce break through articles on this subject.

We are sure, you will be greatly benefitted taking this course and we will help you all the way in your journey.

Best wishes for you through your career.



Course Goals For Student Learning Outcomes

1. Learn the relationships between genes, DNA, RNA and proteins. Regulation of gene expression.

2. Vector DNA and their unique roles in gene cloning and transfer.

3. Methods of DNA extraction, purification and quantification from living cells.

4. Gene cloning: splice, isolate, insert and ligate DNA fragments into unrelated DNA.

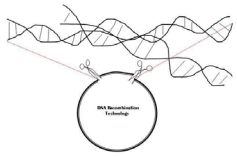
5. Transfer and introduction of DNA into living cells.

6. DNA as a means of identification – Southern hybridization, restriction enzyme mapping, DNA fingerprinting and barcoding.

7. Polymerase Chain Reaction (PCR) as a means of DNA amplification, forensics, DNA fingerprinting and DNA barcoding.

8. Methods for DNA sequencing and their applications.

9. Scope of recombinant DNA technology in biotechnology: medicine, forensics and agriculture.



FACULTY NAME

Professor Sarwar Jahangir

Office: S 206, Tel: 718-368-5743
Bio Office 718- 368-5502

PROFESSOR

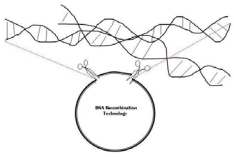
Name: _____ Office: _____
Office Hours: _____ Telephone: _____

PROFESSOR

Name: _____ Office: _____
Office Hours: _____ Telephone: _____

TUTOR

Name: _____ Location: _____
Hours: _____ Telephone: _____



CONTENTS

Week	Lecture Topic	
1	Introduction. Study of genes, DNA, RNA, and proteins. Gene transcription, translation and packaging.	12
2	Gene cloning. Application of gene cloning in medicine, environment, agriculture and research. Ethics of gene cloning.	13
3	Gene cloning vectors or vehicles: plasmids and viruses.	14
4	Methods of DNA extraction, purification, and quantification from living cells.	15
5	Lecture Exam 1. Scissors and wrenches for gene manipulation – enzymes.	16
6	Scissors and wrenches for gene manipulation – enzymes – continued.	17
7	Transfer and introduction of DNA into living cells.	18
8	Gene expression regulation. Cloning as means to investigate gene and genome.	19
9	Lecture Exam 2 Introduction to polymerase chain reaction (PCR).	20
10	*In vitro* amplification of gene using PCR and real-time PCR (RT-PCR).	21
11	Artificial chromosome: reconstruction and expression in heterologous genome. Its application in medicine, pharmaceuticals and gene therapy.	22
12	Application of recombined genes in medicine, agriculture, food industry and environment.	23
13	Final examination. Cumulative… A^+ A^+ A^+ A^+	

TEXTBOOK AND GRADING POLICIES

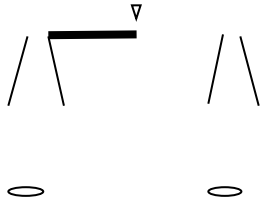
Textbook:

Gene Cloning and DNA Analysis: An Introduction, 5th edition, 2006. Author: Terence A. Brown. Blackwell Science Ltd. Pp 386. ISBN 978-1-4051-1121-8.

Laboratory Manual:

Laboratory DNA Science: An Introduction to Recombinant DNA Techniques and Methods of Genome Analyis. 1995. Mark V. Bloom, Greg A. Freyer and David A. Miklos. Benjamin Cummings, NY. Pp 434. ISBN 0-8053-3040-2.

GRADING POLICIES



Lecture:

Two Lecture Exams @ 10% X 2	= 20%
Written review: Frontiers in biotechnology	= 10%
One Final Exam @ 20% X 1	= 20%
Total	= 50%

Laboratory:

Laboratory reports	= 25%
Laboratory Quizzes 6.25% x 4	= 25%
Total	= 50%

Lecture (50%) + Laboratory (50%) = 100%

It is the policy of the school that every student must wear a laboratory coat as they enter the laboratory each week. The coat must be of 100% cotton, knee high, full sleeve and buttoned in front. In addition, each student must carry a pair of laboratory goggles and hand gloves, and wear them as necessary.

Students must prepare their lab reports neat and clean and bind them at the end of the semester for getting the full laboratory grade.

EXTRA CREDIT- FIVE POINTS: Students may earn extra credit by submitting a term paper at the end of the 9th regular semester week. The term paper must be:

1. At least 5 typed or printed - double spaced pages.
2. Topic to be selected with the approval of the instructor.
3. Use bibliographic format and at least three (5) sources.
4. It must be your original work. Presenting someone else's work, ideas or words as your own is plagiarism. You must give credit to those authors and researchers, whose ideas, words and research you used, in your term paper.

STUDENT RESPONSIBILITIES

Absence Policy

A student who has been absent 15% of the total number of instructional hours in BIOLOGY 58 is deemed to be excessively absent. The 15% excessive absence policy applies to either the laboratory or lecture component. Each time late will be counted as ½ hour of absence. Excessive absences may result in the instructor assigning either a lower grade or a "WU" grade for that course.

The absent student must take the initiative for remaining up to date in the course and is responsible for all covered material and assigned work. The student must discuss absences from laboratory activities with his or her professor. This should be done prior to an anticipated absence or immediately following a missed activity session.

Cell Phones and Beepers

The use or ringing of cell phones and beepers in the classroom during class sessions is a disruption of class and a violation of the Henderson Rules. Violation of this policy may result in a disciplinary referral**.**

Written Assignments

Based on your experiment, you will prepare a laboratory reports each week. The problems and questions will be based on the observations and experiments that you complete. Although, you will be completing experiments in groups, **your reports must demonstrate your independency in observations, interpretations, and conclusions**.

Plagiarism

You may find the CUNY and KCC's Academic Integrity Policies are in:

- Kingsborough Community College Catalogue
- Kingsborough Community College Student Handbook
- www.kingsborough.edu/**Academic**_Integrity_Policy.pd

"Plagiarism **as a violation of academic integrity** is the intentional use of another's intellectual creation(s) without attribution. Determination and penalty—ranging from grade reduction to course failure—is at the sole discretion of the faculty member."

In addition, your instructor may inform you of his or her policy regarding academic integrity at the beginning of the semester.

Assessment

You will be given a 20 minutes assessment test at the last part of the semester in order to test the effectiveness of teaching and learning in this course. Your instructor will decide of the content of this test and may assign a credit value, if appropriate.

Please see the Course Outline for further details

Prof. Sarwar Jahangir, Ph.D.
Office S 206; Tel: 368-5743; Office Hr: M & W 11:00 AM -12:00 PM

BIOLOGY-58 W01 Lec; M T W & Th; Room M119

9:00 AM-10:35 AM: Winter 2020, Tentative Schedule.

Tentative Date	Major Lecture Topics	Text Reading Assignments
Jan 06 & 07	Introduction. Study of genes, DNA, RNA, and proteins. Gene transcription and translation.	Biology 9th Ed. Chapter 17 Campbell, 2010
Jan 08 & 09	Applications of recombinant DNA technology in gene cloning and the ethical issues surrounding gene cloning.	Textbook Chapter 1; pages: 3-13. See syllabus for more information.
Jan 13 & 14	Gene cloning vectors: plasmids and viruses.	Textbook Chapters 2 & 6; pages: 14-27 and 107-131.
Jan 15 &16	Methods of DNA extraction, purification and quantification from living cells.	Textbook Chapter 3; pages: 28- 53; See syllabus for additional information.
Jan 21 & 22	**Lecture Exam 1. Jan 21** "Scissors and Wrenches (Enzymes) for gene/DNA manipulation".	Textbook Chapter 4; pages: 54-86.
Jan 23 & 27	Scissors and wrenches for gene manipulation – enzymes – continued.	Textbook Chapter 4; pages: 54-86.
Jan 28 & 29	Transfer and introduction of DNA into living cells.	Textbook Chapter 5; pages: 87–106. See syllabus for additional information.
Jan 30 & Feb 03	Followed by, how to obtain a recombinant DNA clone.	Textbook Chapter 8; pages: 158-180.
Feb 04 & 05	**Lecture Exam 2; Feb 04** Polymerase Chain Reaction (PCR) and DNA sequencing.	Textbook Chapters 9 & 10; pages: 181-195 and 199-218.
Feb 06 & 10	Molecular means for genetic identification.	Textbook Chapter 8 & 11; pages: 239-250. See syllabus for additional information.
Feb 11 & 13	Recombinant DNA cloning in Medicine.	Textbook Chapter 14; pages: 302- 322.
Feb 18 & 19	Recombinant DNA in agriculture, forensics and archeology.	Textbook Chapter 15; pages: 323-345.
Feb 24 Mon	**Final Examination**; **Cumulative of Lectures.** *Room TBA*	**Cheer-up**! ☺☺☺

Grading: Please see the course syllabus handed to you for the grading procedure in detail. This course is graded by adding your scores in the lecture and in the laboratory. The exams in the lecture will comprise 50% and the lab will comprise another 50% for the final grade. There will be two lecture exams and one cumulative final lecture exam. The lecture exams will carry 15% each and the final lecture exam will carry 20% and, thus, 50% for calculating the final grade in this course.

Pleasure to have wonderful students like you in the class.

Week 1

Introduction. Study of genes, DNA, RNA, and proteins. Gene transcription and translation.

READING ASSIGNMENT

You should access Biology, 8th Ed, Chapter 17, Campbell et al. 2008. In addition, a handout will be circulated for additional information.

LEARNING OBJECTIVES

You should have a clear concept on the topic and be able to:

1. Define and describe a gene, DNA, RNA and protein.

2. Understand the flow of genetic information. State the differences and relationships with clarity between a gene, DNA, RNA and protein.
3. How a gene is transcribed, translated into proteins.

4. One gene one polypeptide or one protein.

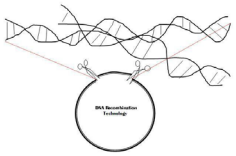
5. Understand processing of messenger RNA, triplet codes, codon dictionary, start and stop codons and a reading frame.

6. Describe the degenerative natures of codons, nearly universal nature of codons with differences between eukaryotic and prokaryotic codons.

7. Explain what is a promoter, a terminator, and a transcription unit.

8. Post transcriptional modifications.

9. Follow what is a point mutation, insertion and deletion, base pair substitution, missense and nonsense mutations.



Week 2

Application of recombinant DNA technology in gene cloning and ethics of gene cloning.

READING ASSIGNMENTS

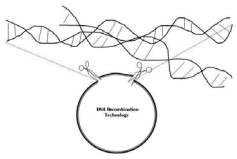
1. Textbook pages 1-12;
2. Human Genome Project Information. Sponsored by the US Department of Energy, Office of Biological and Environmental Research. http://www.ornl.gov/sci/techresources/Human_Genome/elsi/cloning.shtml

3. CRISPR and Biotechnology. http://www.sciencemag.org/topic/crispr
4. Some lecture handouts will be provided.

LEARNING OBJECTIVES

You should be fully aware of and explain:

1. What is cloning and gene cloning? Are there different types of cloning?
2. What is Recombinant DNA Technology or DNA Cloning**.**
3. How can cloning technologies be used?
4. What animals have been cloned?
5. Can organs be cloned for use in transplants?
6. What are the risks of cloning?
7. Should humans be cloned?
8. What are its biological, environmental, economic and social implications.



Week 3

Gene cloning vectors or vehicles: plasmids and viruses.

READING ASSIGNMENT

Textbook pages: 14-27 and 132-157.

LEARNING OBJECTIVES

You should know the:

1. Prokaryotic Vectors for Gene Cloning: Plasmids and Bacteriophages.
    a. Plasmids: Basic features of plasmids, Size and copy number, Conjugation and compatibility. Classification of plasmids and non bacterial plasmids.
    b. Bacteriophages. Basic features of bacteriophages, lysogenic phages, 8 DNA organization, linear and circular forms. M13 filamentous phage as a cloning vector.
2. Cloning Vectors for Eukaryotes
    a. Vectors for yeast and other fungi.
        i. Yeast episomal plasmids (Yep),
        ii. Yeast Artificial chromosomes (YAC) and applications.
    b. Cloning vectors for higher plants:
        i. *Agrobacterium tumefaciens* – nature's smallest genetic engineer.
        ii. Ti plasmid to introduce new genes into a plant cell.
        iii. Production of transformed plants with the Ti plasmid.
        iv. The Ri plasmid. Limitations of cloning with *Agrobacterium* plasmids.
    c. Cloning vectors for animals –
        i. insects, p-elements, baculovirus.
        ii. mammals – adenovirus, adeno-associated virus, papillomavirus, retrovirus, and simian virus-40.

Week 4

Methods of DNA extraction, purification and quantification from living cells.

READING ASSIGNMENT

1. Textbook pages 28- 53; and
2. Jahangir, ZMG Sarwar. Isolation of nuclear DNA from experimental fishes. *In* A study of euteleost phylogeny based on the genomic diversity of ribosomal DNA… 1995. Ph.D. Thesis, CUNY. Pp 67-71. You may request it using interlibrary loan program from the Graduate School and University Center, CUNY, through the KCC Library.

LEARNING OBJECTIVES

You should be able to:

Describe the extraction and purification of DNA from Living Cells.

Prokaryotic cells:

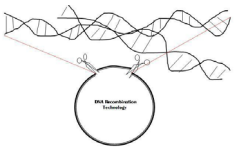
Preparation of total cell DNA, grow and harvest bacteria in culture, prepare a cell extract, purify DNA from the extract, remove contaminants by organic extraction and enzyme digestion. Ion-exchange chromatography to purify DNA from cell extracts. How to measure DNA concentrations in samples? Other methods for the preparation of total cell DNA.

Preparation of plasmid DNA: Separation of plasmid DNA based on sizes and conformation; alkaline denaturation of DNA; ethidium bromide–cesium chloride density gradient centrifugation for DNA separation.

Preparation of bacteriophage DNA, grow bacterial cells in cultures to obtain a high 8 titer; prepare non-lysogenic 8 phages; collect phages from an infected culture. Purification of DNA from 8 phage particles and M13 DNA.

Prokaryotic cells:

Isolation and purification of nuclear DNA from eukaryotic cells with examples from teleost fishes. Isolation and purification of mitochondrial DNA from euteleost fishes as an example.



Week 5

Lecture Exam 1. Followed by: "Scissors and Wrenches (Enzymes) for gene manipulation".

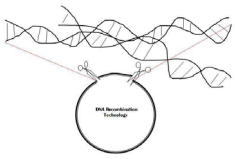
READING ASSIGNMENT

Textbook pages 54-86.

LEARNING OBJECTIVES

You should be able to fully understand the theory and principles of:

1. Manipulation of DNA using enzymes: The range of DNA manipulative enzymes – nucleases, ligases, polymerases, topoisomerases, and restriction endonucleases.
2. The discovery and function of restriction endonucleases. Type II restriction endonucleases cut DNA at specific nucleotide sequences, blunt ends and sticky end cutters, frequency of recognition sequences in a DNA molecule; and performing a restriction digest in the laboratory.
3. Analyzing the result of restriction endonuclease cleavage: separation of molecules by gel electrophoresis; visualizing DNA molecules by staining a gel; visualizing DNA molecules by autoradiography; estimation of the sizes of DNA molecules; Mapping the positions of different restriction sites in a DNA molecule.
4. Ligation – joining DNA molecules together, the mode of action of DNA ligase, sticky ends increase the efficiency of ligation, putting sticky ends onto a blunt-ended molecule using linkers and adaptors.



Week 6

Scissors and wrenches for gene manipulation – enzymes – continued.

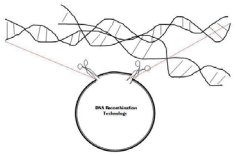
READING ASSIGNMENT

Textbook pages 54-86.

LEARNING OBJECTIVES

You should be able to fully understand the theory and principles of:

1. Manipulation of DNA using enzymes: The range of DNA manipulative enzymes – nucleases, ligases, polymerases, topoisomerases, and restriction endonucleases.
2. The discovery and function of restriction endonucleases. Type II restriction endonucleases cut DNA at specific nucleotide sequences, blunt ends and sticky end cutters, frequency of recognition sequences in a DNA molecule; and performing a restriction digest in the laboratory.
3. Analyzing the result of restriction endonuclease cleavage: separation of molecules by gel electrophoresis; visualizing DNA molecules by staining a gel; visualizing DNA molecules by autoradiography; estimation of the sizes of DNA molecules; Mapping the positions of different restriction sites in a DNA molecule.
4. Ligation – joining DNA molecules together, the mode of action of DNA ligase, sticky ends increase the efficiency of ligation, putting sticky ends onto a blunt-ended molecule using linkers and adaptors.



Week 7

Transfer and introduction of DNA into living cells.

READING ASSIGNMENT

1. Textbook- pages 87 – 106; and
2. Eckhardt, R.A. and ZMG S. Jahangir, 1996. Molecular Tagging of Lake Sturgeon as a Means of Identification for Use in Developing a Domesticated Breeding Stock, for Use in Population Re-Establishment, and for Use in Conservation Enforcement Programs. NA36FD0384; NMFS NUMBER: 92-NER-009 National Marine Fisheries Service, Northeast Region, One Blackburn Drive, Gloucester, MA 01930-2298. Will be made available in the KCC Library.

LEARNING OBJECTIVES

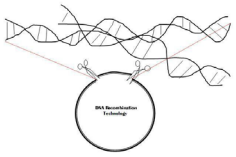
You should be able know the theories behind:

1. Introduction of DNA into Living Cells: transformation and the uptake of DNA by bacterial cells. How to prepare competent *Escherichia coli* cells? Select transformed cells and identification of recombinants. Recombinant selection with pBR322 – insertional inactivation of an antibiotic resistance gene, inactivation of no- antibiotic gene.

2. Introduction of phage DNA into bacterial cells: transfection, *In vitro* packaging of 8 cloning vectors, visualization of infected phage plaques on an agar medium.

3. Identification of recombinant phages, insertional inactivation of a lacZ´ gene carried by the phage vector, insertional inactivation of the 8 cI gene; selection using the Spi phenotype; section based on 8 genome size.

3. Introduction of DNA into non-bacterial cells and transformation of whole organisms using liposomes, electroporation, microinjection and biolostics.



Week 8

How to obtain a recombinant DNA specific gene clone.

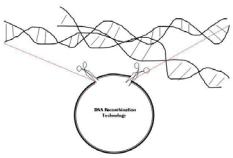
READING ASSIGNMENT

Textbook pages: 158-180.

LEARNING OBJECTIVES

You should be able to know the techniques to determine a recombinant DNA specific gene clone.

1. Learn the problems of selection: There are two basic strategies for obtaining the clone you want.
    a. Direct selection.
        i. Marker rescue extends the scope of direct selection.
        ii. The scope and limitations of marker rescue.
    b. Identification of a clone from a gene library. Gene libraries; expression of genes in the library; mRNA clone as complementary DNA.
2. Methods for clone identification. Hybridization of complementary nucleic acid strands; colony and plaque hybridization; Examples of the practical use of hybridization probing.
    a. Abundancy probing to analyze a cDNA library; Oligonucleotide probes for genes.
    b. Heterologous probing.
3. Identification methods based on detection of the translation product of the cloned gene. Immunological detection methods and problems.



Week 9

The Polymerase Chain reaction and DNA sequencing.

READING ASSIGNMENT

Textbook pages – 181- 195 and 207-219.

LEARNING OBJECTIVES

You should be able to master the theory behind PCR chain reaction and DNA sequencing.

1. The Polymerase Chain Reaction. Outline, the nature of *Taq* DNA polymerase, primer design, determination of appropriate PCR reaction temperatures; determination of PCR products by electrophoresis and cloning.

2. DNA sequencing. The Sanger–Coulson method – chain-terminating nucleotides. The Maxam–Gilbert method – chemical degradation of DNA.

The primer DNA, synthesis of complementary strand, four types of reaction termination; reading the DNA sequence from the autoradiograph. What are the appropriate DNA polymerases for sequencing? Automated DNA sequencing, sequencing and PCR.

Building up a long DNA sequence; and the achievements of DNA sequencing.

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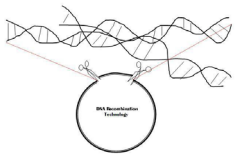


Week 10

Molecular means for genetic identification. Southern and Northern hybridization, restriction enzyme mapping, DNA fingerprinting, microsatellite DNA polymorphism and DNA Barcoding.

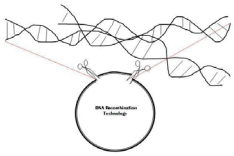
READING ASSIGNMENT

1. Textbook - pages 199- 203; 225; 261-264; 317-318; 347-350.
2. A 3 page handout on "Microsatellite DNA Methodology" will be supplied. http://www.bio.davidson.edu/COURSES/genomics/method/microsatellite.html.
3. A four page "DNA barcoding protocol" with rationale will be supplied. http://www.coreocean.org/Dev2Go.web?id=255158.

LEARNING OBJECTIVES

You should be able to lean the use of DNA as a molecular means for identification:

1. Principles of Southern hybridization and Northern hybridization.
2. Techniques rationale of restriction enzyme mapping.
3. Principles of DNA finger printing.
4. Microsatellite DNA as a means of population identification.
5. Mitochondrial DNA for barcoding and species identification.



Week 11

Recombinant DNA cloning in Medicine.

READING ASSIGNMENTS

Textbook pages 302- 322.

LEARNING OBJECTIVES

You will be responsible to learn all the achievements made in medicine by using recombinant DNA as follows:

1. Production of recombinant pharmaceuticals.
    a. Recombinant insulin. Synthesis and expression of artificial insulin genes.
    b. Synthesis of human growth hormones in *Escherichia coli*.
    c. Recombinant factor VIII.
    d. Synthesis of other recombinant human proteins.
    e. Recombinant vaccines.
        i. Producing vaccines as recombinant proteins.
        ii. Recombinant vaccines in transgenic plants.
        iii. Live recombinant virus vaccines.
2. Identification of genes responsible for human diseases.
    a. How to identify a gene for a genetic disease.
        i. Locating the approximate position of the gene in the human genome.
        ii. Identification of candidates for the disease gene.
3. Gene therapy.
    a. Gene therapy for inherited diseases.
    b. Gene therapy and cancer.
    c. Ethical issues raised by gene therapy

Week 12

Recombinant DNA in agriculture, forensics and archeology.

READING ASSIGNMENT

Textbook pages: 323-345; 346-361.

LEARNING OBJECTIVES

You will be responsible to learn the achievements made in agriculture, forensics and archeology by using recombinant DNA as follows:

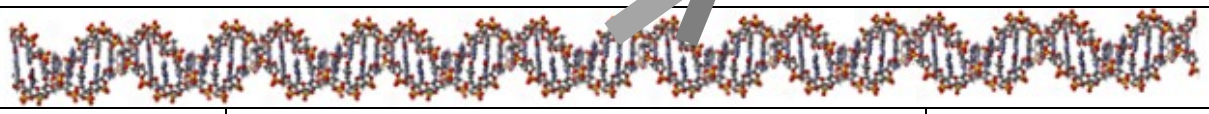
1. Gene Cloning and DNA Analysis in Agriculture.
    a. plant genetic engineering.
    b. Plants that make their own insecticides.
    c. Cloning ∗-endotoxins of *Bacillus thuringiensis* in maize, and chloroplasts,
    d. Countering insect resistance to d-endotoxin crops.
2. Herbicide resistant crops.
    a. Roundup Ready' crops.
    b. A new generation of glyphosate resistant crops.
3. Problems with genetically modified plants.
    a. Safety concerns with selectable markers.
    b. The terminator technology.
    c. The possibility of harmful effects on the environment.
4. Gene Cloning and DNA Analysis in Forensic Science and Archaeology.
    a. DNA analysis in the identification of crime suspects.
    b. Genetic fingerprinting by hybridization probing.
    c. DNA profiling by PCR of short tandem repeats.
    d. Studying kinship by DNA profiling.
    e. DNA profiling and the remains of the Romanovs.
    f. Identification of missing children and sex identification by DNA analysis.
5. Archaeogenetics – using DNA to study human evolution.
    a. The origins of modern humans.
    b. DNA analysis challenged the multiregional hypothesis.
    c. Neanderthals are not the ancestors of modern Europeans.
    d. DNA can also be used to study prehistoric human migrations.
    e. The spread of agriculture into Europe.
    f. Mitochondrial DNA depicts past human migrations into Europe.

Department of Biological Sciences, Kingsborough Community College

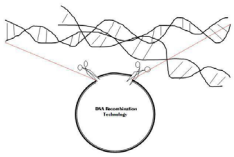
The City University of New York

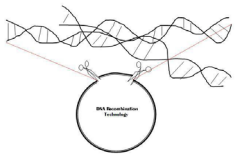
BIOLOGY 58 Lab W301 T & TH S114

Tentative Winter 2020 Schedule: 11:00 AM-2:00 PM.



Week Tentative	Exercises	Experiments in the Lab Manual & Readings Assigned
Jan 07	Measurements, pipetting, micropipetting and sterile techniques. Prepare LB broth and LB Agar plates.	3-12 390-391
Jan 9	Culture of bacterial cells in plates and broth	13-30
Jan 14	Transformation of *Escherichia coli* with plasmid DNA - pGlo plasmid	73-87
Jan 16	**Quiz 1 on Exercises 1-3.** Isolation, purification and identification of plasmid DNA – *p*Glo	89-107 Bio-Rad Handout
Jan 21	Restriction Enzyme Digestion of pGlo and separation by agarose gel electrophoresis. Restriction enzyme mapping.	31-53
Jan 23	Isolation, purification and identification of GFP	Bio-Rad Brochure
Jan 28	Visit to a Biotech Lab; **Tentative.**	NY/NJ
Jan 30	**Quiz 2 on Exercises 4-6.** Recombination of antibiotic resistance genes. Transformation of *E. coli* with recombined DNA	111-123 125-140
Feb 04	Extraction of recombined plasmid; Identification of the plasmid by Agarose gel electrophoresis and	31-53
Feb 06	Polymerase chain reaction. **Quiz 3 on Exercises 7-9.**	281-297
Feb 11	Extraction of nuclear DNA from fish blood and Set-up DNA digestion with restriction endonucleases.	Handout will be supplied.
Feb 13	Southern blotting Southern hybridization **Quiz 4 on Exercises 10-12.**	191-208 209-215
Feb 18	***Last day to submit the Final Laboratory Report***	




DNA Recombination
Technology