

Activity- An overview of the process of Whole Genome Sequencing

Subject	Biology
Class	XII
Chapter	Molecular Basis of Inheritance
Topic	Whole Genome Sequencing
Duration	1 ½ hours
Nature of the Activity	Group Activity

What will I learn? (Learning Outcome)

– understand the process and importance of genome sequencing, appreciate the advancements in the field of science

Skills- Analysing, critical thinking

What I have to do? Work in a group and carry out the steps of genome sequencing as per the steps given

What do I require?

Props/ Materials required- 2 sets of alphabets (A-Z), scissors (2), labels of EcoRI and BamHI, long string/ cut outs of YAC/ BAC (2 each), brown tape

What do I need to know?

rDNA Technology; Aim and need for finding genome sequence; the methods of genome sequencing; the steps of genome sequencing; meaning of analogy

- **Teacher's activity-** Teacher will give the instructions along with the protocol (Student's activity)
- **Teacher will explain that the English alphabet(A-Z) is taken to be analogous to a given DNA sequence, say-**

5' AATTCCAATCCGGATCCGGTTACCTTACCGAATTCAATTCC 3'

3' TTAAGGTTAGGCCTAGGCCAATGGAATGGCTTAAGTTAAGG 5'

EcoRI recognises and cuts between G & A in 5'GAATTC 3'

3'CTTAAG 5'

And BamHI recognises and cuts between the 2 G's in 5'GGATCC 3'

3'CCTAGG 5'

Teacher will facilitate the whole activity

How will I proceed?

➤ Student's activity

Assign the roles as-

1. All the labels will be worn as crowns
2. Two volunteers as restriction endonucleases(molecular scissors)- EcoRI, BamHI
3. One volunteer as DNA ligase (molecular glue)
4. Four volunteers as YAC/ BAC
5. One student to read and note the sequence
6. Rest of the group (with more student volunteers) - Two DNA sequences (Each one A-Z)
7. Cutting of the two sequences by two different restriction endonucleases (EcoRI between F & G, BamHI between K & L)

Steps-

To be followed, supported with a slide showing the process

1. Students to stand in 2 lines from A-Z (DNA sequence)
2. EcoRI to cut at the assigned point in one of the lines (A-Z) and generate 2 fragments

BamHI to cut at the assigned point in the 2nd line (A-Z) and generate 2 fragments
3. The broken sequences of line 1 and 2 to be joined to the 4 different YAC, using tape (Volunteer acting as DNA ligase)

(Teacher will explain the next few steps (Cloning these in different vectors -amplification upon insertion in host cell, Arranging sequences based on overlapping regions, Sequences annotated and assigned to different chromosomes))
4. Reading the sequence (equivalent to Automated DNA Sequencing (by Sanger's method))

What have I observed?

The English Alphabet	ABCDEFGHIJKLMNOPQRSTUVWXYZ
Fragments generated by cutting with EcoR1 between F & G	-----, -----
Fragments generated by cutting with BamH1 between K & L	-----, -----
Overlapping Sequence	-----
The given sequence	5' AATTCCAATCCGGATCCGGTTACCTTACCGAATTCAATTCC 3' 3' TTAAGGTTAGGCCTAGGCCAATGGAATGGCTTAAGTTAAGG 5'
Fragments generated by cutting with EcoR1	-----, -----
Fragments generated by cutting with BamH1	-----, -----
Overlapping Sequence	-----

What have I learnt?

Correct sequence of the genome can be generated by comparing the overlapping fragments (like we did for the English Alphabet)

What would I like to know ?

Can genome sequencing of pathogens help find cures to the diseases caused by them?

Let me Evaluate myself-

1. What are the steps of whole genome sequencing?
2. Differentiate between the two methods of genome sequencing- by identifying ESTs and whole genome sequencing

3. Why was sequencing of human genome, a challenging process? Why 24 chromosomes were sequenced under Human Genome Project?
4. Generate a fragment 28bp long, that can be cut by restriction enzymes Sal I and Pvu I. In the given sequence, ensure that only one recognition site of each enzyme is there, with a gap of at least 5bp. How many fragments will be generated if it is cut by –
 - i) Only Sal I
 - ii) Only Pvu I
 - iii) Both Sal I and Pvu I
5. What is the relative time required for genome sequencing of prokaryotes and eukaryotes? How can the technique be useful in finding correct sequence of viral genome, as they keep mutating?