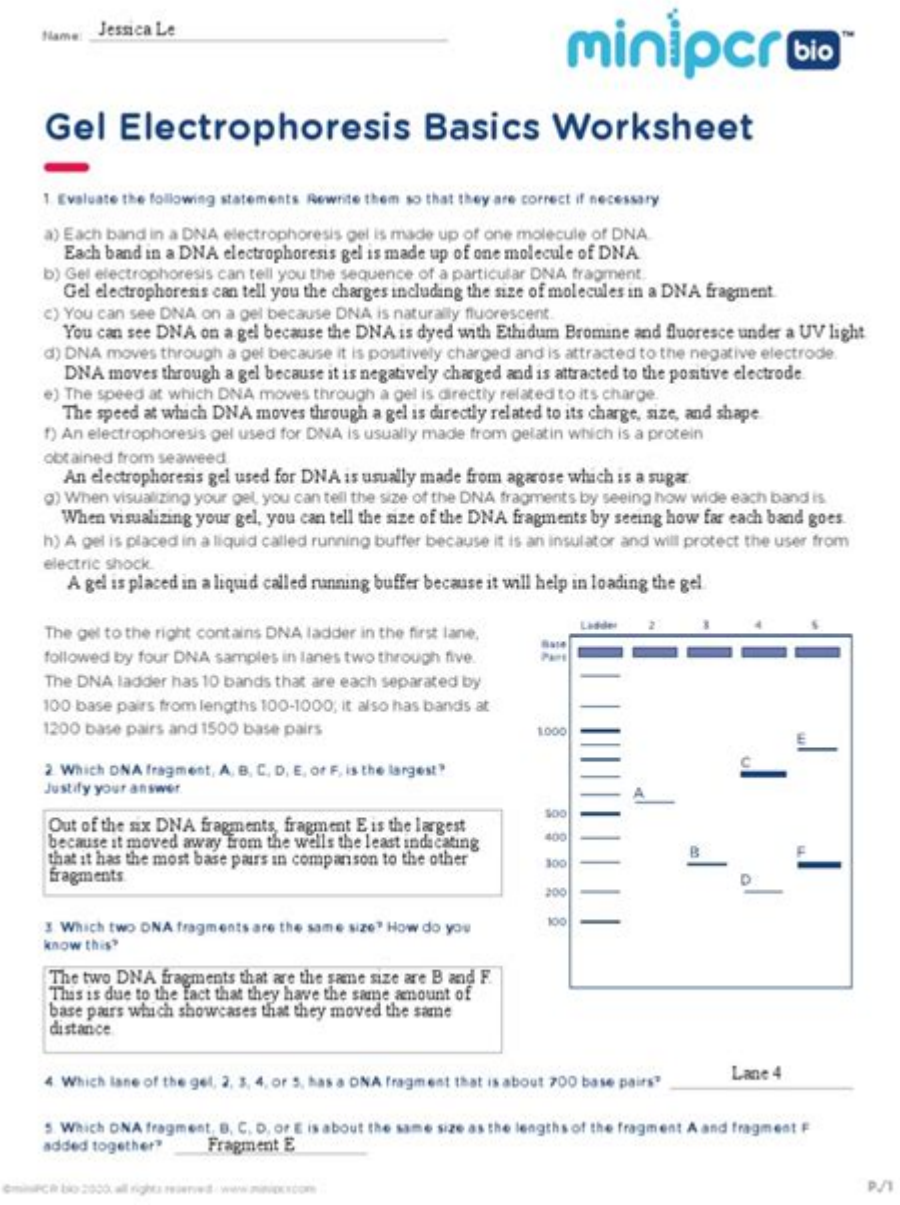
Gel Electrophoresis Basics Worksheet



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Gel Electrophoresis Basics Worksheet

1. Evaluate the following statements. Rewrite them so that they are correct if necessary.

a) Each band in a DNA electrophoresis gel is made up of one molecule of DNA.
Each band in a DNA electrophoresis gel is made up of one molecule of DNA.
b) Gel electrophoresis can tell you the sequence of a particular DNA fragment.
Gel electrophoresis can tell you the charges including the size of molecules in a DNA fragment.
c) You can see DNA on a gel because DNA is naturally fluorescent.
You can see DNA on a gel because the DNA is dyed with Ethidum Bromine and fluoresce under a UV light.
d) DNA moves through a gel because it is positively charged and is attracted to the negative electrode.
DNA moves through a gel because it is negatively charged and is attracted to the positive electrode.
e) The speed at which DNA moves through a gel is directly related to its charge.
The speed at which DNA moves through a gel is directly related to its charge, size, and shape.
f) An electrophoresis gel used for DNA is usually made from gelatin which is a protein obtained from seaweed.
An electrophoresis gel used for DNA is usually made from agarose which is a sugar.
g) When visualizing your gel, you can tell the size of the DNA fragments by seeing how wide each band is.
When visualizing your gel, you can tell the size of the DNA fragments by seeing how far each band goes.
h) A gel is placed in a liquid called running buffer because it is an insulator and will protect the user from electric shock.
A gel is placed in a liquid called running buffer because it will help in loading the gel.

The gel to the right contains DNA ladder in the first lane, followed by four DNA samples in lanes two through five. The DNA ladder has 10 bands that are each separated by 100 base pairs from lengths 100-1000; it also has bands at 1200 base pairs and 1500 base pairs.

2. Which DNA fragment, A, B, C, D, E, or F, is the largest? Justify your answer.

Out of the six DNA fragments, fragment E is the largest because it moved away from the wells the least indicating that it has the most base pairs in comparison to the other fragments.

3. Which two DNA fragments are the same size? How do you know this?

The two DNA fragments that are the same size are B and F. This is due to the fact that they have the same amount of base pairs which showcases that they moved the same distance.

4. Which lane of the gel, 2, 3, 4, or 5, has a DNA fragment that is about 700 base pairs? Lane 4

5. Which DNA fragment, B, C, D, or E is about the same size as the lengths of the fragment A and fragment F added together? Fragment E

Gel electrophoresis basics worksheet is an essential resource for students and researchers who wish to understand the fundamental concepts and applications of gel electrophoresis in molecular biology. This technique plays a crucial role in the separation and analysis of macromolecules, primarily nucleic acids and proteins. In this article, we will explore the principles of gel electrophoresis, its components, how to prepare and run a gel, and common applications of this invaluable laboratory technique.

Understanding Gel Electrophoresis

Gel electrophoresis is a widely used laboratory method for separating

molecules based on their size and charge. It is particularly useful in the fields of genetics, biochemistry, and molecular biology. The basic premise of this technique is that charged molecules will migrate through a gel matrix when an electric field is applied, allowing for their separation and subsequent analysis.

Principles of Gel Electrophoresis

1. Charge: Molecules such as DNA and proteins carry an electric charge. DNA molecules, for example, are negatively charged due to their phosphate backbone, which means they will move towards the positive electrode when an electric field is applied.

2. Size: The gel matrix, usually made of agarose or polyacrylamide, acts as a sieve that impedes the movement of larger molecules more than smaller ones. As a result, smaller molecules migrate faster and further through the gel compared to larger counterparts.

3. Electric Field: The electric field is generated by connecting the gel to a power supply. The strength and polarity of the electric field can affect the speed and distance that molecules travel through the gel.

Components of Gel Electrophoresis

To perform gel electrophoresis, several key components are required:

- Gel Matrix:
- Agarose Gel: Commonly used for DNA separation, with concentrations typically ranging from 0.7% to 2% depending on the size of the DNA fragments.
- Polyacrylamide Gel: Used for protein separation and smaller DNA fragments, offering higher resolution than agarose.

- Buffer Solution:
- Provides ions that carry the current and maintains a stable pH during electrophoresis. Common buffers include Tris-acetate-EDTA (TAE) and Tris-borate-EDTA (TBE).

- Power Supply:
- Generates the electric field required for the migration of the molecules through the gel.

- Sample Loading Dye:
- Added to samples to track the progress of electrophoresis and to increase the density of the sample, ensuring it sinks into the wells of the gel.

- Staining Agent:
- Used to visualize the separated molecules post-electrophoresis, such as

ethidium bromide for DNA or Coomassie Brilliant Blue for proteins.

Preparing and Running a Gel

The process of gel electrophoresis involves several steps, from preparing the gel to visualizing the results.

1. Preparing the Gel

- Choose the Gel Type:
- Determine whether you need agarose or polyacrylamide based on the size of the molecules you are analyzing.

- Prepare the Gel Solution:
- For an agarose gel, dissolve the appropriate amount of agarose powder in buffer solution by heating it until fully dissolved.

- Casting the Gel:
- Pour the molten agarose into a gel casting tray and insert a comb to create wells. Allow the gel to solidify, which usually takes about 30 minutes.

2. Preparing the Samples

- Mix Samples with Loading Dye:
- Combine your samples with the loading dye to ensure they are dense enough to stay in the wells and to monitor the migration during electrophoresis.

- Load the Samples into the Gel:
- Carefully remove the comb from the solidified gel and load the samples into the wells using a micropipette.

3. Running the Gel

- Connect the Power Supply:
- Place the gel in the electrophoresis chamber filled with buffer, ensuring that the wells are closest to the negative electrode (cathode).

- Set the Voltage:
- Adjust the power supply to the desired voltage, typically between 80-150 volts, depending on the gel type and size.

- Monitor the Progress:
- Run the gel for a specific duration, usually 30 minutes to several hours,

depending on the size of the fragments and the gel concentration.

4. Visualizing the Results

- Staining the Gel:
- After running the gel, stain it with an appropriate dye to visualize the separated bands. For DNA, ethidium bromide is commonly used, while proteins can be stained with Coomassie Brilliant Blue.

- Imaging:
- Use a UV transilluminator or a gel documentation system to capture images of the gel for analysis.

Applications of Gel Electrophoresis

Gel electrophoresis has numerous applications in research, clinical diagnostics, and forensic science. Some notable uses include:

- DNA Analysis:
- Used in techniques such as PCR (Polymerase Chain Reaction) to separate and analyze DNA fragments.

- Protein Separation:
- Essential in proteomics for analyzing complex protein mixtures and understanding protein interactions.

- Genetic Research:
- Important for genotyping, sequencing, and studying genetic variation among populations.

- Forensic Science:
- Utilized in DNA fingerprinting for criminal investigations and paternity testing.

- Quality Control:
- Employed in laboratories to verify the quality and purity of samples in various fields, including pharmaceuticals and biotechnology.

Conclusion

A gel electrophoresis basics worksheet serves as a practical guide for understanding the fundamental principles and procedures involved in this technique. Whether you are a student embarking on your scientific journey or a researcher seeking to refine your skills, mastering gel electrophoresis is crucial for success in the molecular biology field. With its wide range of

applications and the ability to separate molecules effectively, gel electrophoresis remains an indispensable tool in modern science.

Frequently Asked Questions

What is gel electrophoresis and how does it work?

Gel electrophoresis is a laboratory technique used to separate DNA, RNA, or proteins based on their size and charge. The sample is placed in a gel matrix, and an electric current is applied, causing the molecules to migrate through the gel. Smaller molecules move faster and further than larger ones.

What materials are typically used to create the gel for electrophoresis?

The most common materials used to create gels are agarose for DNA/RNA separation and polyacrylamide for protein separation. Agarose gels are easier to prepare and are used for larger DNA fragments, while polyacrylamide gels provide better resolution for smaller proteins.

What are the main components of a gel electrophoresis setup?

A gel electrophoresis setup typically includes a gel tray, a comb to create wells, electrophoresis buffer, a power supply, and a gel box. The gel tray holds the gel, the comb forms wells for sample loading, and the buffer provides the medium for conducting electricity.

How can you visualize the results after running gel electrophoresis?

After running gel electrophoresis, the gel can be stained with a DNA-binding dye, such as ethidium bromide or SYBR Green, which allows visualization under UV light. This staining highlights the bands of separated molecules, which can be analyzed for size and quantity.

What are some common applications of gel electrophoresis?

Gel electrophoresis is widely used in molecular biology for DNA cloning, genetic fingerprinting, and analyzing PCR products. It is also used in protein research for determining protein size, purity, and molecular weight.

What safety precautions should be taken when performing gel electrophoresis?

Safety precautions include wearing gloves and goggles to protect against

chemical exposure from staining dyes, using appropriate lab attire, and handling electrical equipment carefully to avoid electric shock. Additionally, ethidium bromide is a mutagen, so proper disposal and handling procedures should be followed.

Gel Electrophoresis Basics Worksheet

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