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Extraction of DNA from Onion Cells

Objective

The objective of this experiment was to extract DNA from onion cells and demonstrate how cell membranes and other cellular components can be disrupted and separated in order to isolate and visualize DNA.

Introduction

DNA, or deoxyribonucleic acid, contains the genetic information of living organisms. Although DNA is normally microscopic and cannot be seen in individual cells, it can be isolated and collected in larger quantities using laboratory techniques.

In this experiment, onion cells were treated with a homogenizing solution containing sodium chloride, EDTA, and SDS. The onion mixture was heated and blended to help disrupt the cells and release their contents. The mixture was then filtered to remove larger cellular materials. Finally, ice-cold ethanol was added to precipitate the DNA. Because DNA becomes visible when enough of it precipitates together, the extracted DNA could be observed as white, thread-like material and collected using a Pasteur pipet hook.

Materials and Methods

Approximately 25 g of diced onion was placed into a 250 mL beaker. 50 mL of homogenizing solution containing 0.15 M NaCl, 0.1 M EDTA-Na_2, and 1% SDS was added to the onion. The mixture was gently stirred and placed in a 60°C water bath for 15 minutes.

After heating, the mixture was transferred to an ice bath and cooled to approximately 15–20°C. It was then blended for 30–60 seconds at medium speed to further lyse the onion cells. The blended mixture was allowed to rest in an ice bath for another 15 minutes.

The mixture was filtered through four layers of cheesecloth into a clean beaker. Ten milliliters of the filtered solution was transferred into a 50 mL conical centrifuge tube and chilled in an ice

bath. 10 mL of ice-cold 95% ethanol was carefully added down the inside wall of the tube to form a separate layer.

After the ethanol was added, white, thread-like DNA became visible. A Pasteur pipet hook was inserted into the tube and gently rotated to spool the DNA onto the hook.

Results

Observations

Step	Observation
Onion + homogenizing solution	Onion was mixed with the homogenizing solution.
Heating	Mixture was heated in a 60°C water bath.
Blending	Onion mixture became more homogenized after blending.
Filtration	Larger pieces and foam were removed from the mixture.
Addition of ice-cold ethanol	A separate ethanol layer formed above the filtered onion solution.
DNA precipitation	White, thread-like material became visible at the interface of the layers.
DNA spooling	White strands of DNA could be collected using the Pasteur pipet hook.

The laboratory procedure specifically states that DNA should appear as white, thread-like material after the addition of ice-cold ethanol and that the DNA can then be spooled onto the hook.

Discussion/Conclusion

The experiment successfully demonstrated the extraction and visualization of DNA from onion cells. The onion cells were disrupted through heating, blending, and treatment with the homogenizing solution. These steps allowed the DNA contained inside the cells to be released into the solution.

The addition of ice-cold ethanol was an important step because it caused the DNA to precipitate, making it visible as white, thread-like material. The DNA could then be collected by gently rotating the Pasteur pipet hook through the precipitated material.

Overall, the experiment demonstrated that DNA, although normally invisible within individual cells, can be isolated and observed using a series of physical and chemical separation techniques. The experiment also demonstrated the importance of cell disruption, filtration, and precipitation in DNA extraction.

References

DNA Extraction Lab: Onion Cell Edition Laboratory Protocol.