DNA and RNA Extraction

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History

- The very first DNA isolation was done by a Swiss physician, Friedrich Miescher in 1869 [8].
- He hoped to solve the fundamental principles of life, to determine the chemical composition of cells. He tried to isolate cells from lymph nodes for his experiment but the purity of lymphocytes was hard and impossible to be obtained in sufficient quantities.
- Therefore, he switched to leucocytes, where he obtained them from the pus on collected surgical bandages.

History

- Initially, Miescher focused on the various type of protein that make up the leukocytes and showed that proteins were the main components of the cell's cytoplasm.
- During his tests, he noticed that a substance precipitated from the solution when acid was added and dissolved again when alkali was added. This was, for the first time he had obtained a crude precipitate of DNA.

History

- To separate DNA from the proteins in his cell extracts, Miescher developed new protocol to separate the cells' nuclei from cytoplasm and then isolated DNA.
- However, his first protocol failed to yield enough material to continue with further analysis.
- He had to develop a second protocol to obtain larger quantities of purified nuclein, which had been named as 'nucleic acid' later by his student, Richard Altman [8].

Current Tendency

- After the fated event where Miescher managed to obtain DNA from cell, many others have followed suit which lead to further advancement in the DNA isolation and purification protocol.
- The initial routine laboratory procedures for DNA extraction were developed from density gradient centrifugation strategies. Meselson and Stahl used this method in 1958 to demonstrate semiconservative replication of DNA [3].
- Later procedures made use of the differences in solubility of large chromosomal DNA, plasmids, and proteins in alkaline buffer [3].

Current Tendency

- Currently, there are many specialized method of extracting out pure DNA or RNA. Generally, they are divided into
- solution-based or
- column-based protocols.

Most of these protocols have been developed into commercial kits that ease the biomolecules extraction processes.

Type of Nucleic Acid Extraction

1. Conventional Method solution-based

- Guanidinium Thiocyanate-Phenol-Chloroform Extraction
- Alkaline Extraction Method
- CTAB Extraction Method
- Ethidium Bromide-Cesium Chloride Gradient Centrifugation
- Purification of Poly RNA by Oligp(dT)-Cellulose Chromatography

Type of Nucleic Acid Extraction

2. Solid-phase Nucleic Acid Extraction

- Silica Matrices
- Glass Particle
- Diatomaceous Earth
- Magnetic Bead Based Nucleic Acid Purification
- Anion-Exchange Material

Type of Nucleic Acid Extraction

3. All-in-One Biomolecules Extraction

4. Automated Extraction System

Possible Future Direction

Biomolecules extraction is the first step that needs to be performed for the following analysis or manipulation process. The liquid handling requirement is the most challenging aspect. Therefore, any automatic system must include not only automatic equipment for each extraction step but also equipment for automating the transfer of liquid between machines. Automation has aided in increasing the throughput and improving the reliability of the process, but these systems are still designed for use in a laboratory environment only. Some of the nucleic acid extraction system that are available in the market are large and require manual pre-processing stages by laboratory staff with technical expertise [54]. Therefore, robotic workstations for nucleic acid extraction should fulfill a true “walk-away” automation, which means a fully automated process [49].

Possible Future Direction

It is often inconvenient that targeted biomolecules sample from an animal, plant or even a clinical sample must be sent to a laboratory for it to be extracted and analyzed [54]. The samples, especially clinical sample such as blood, need to be refrigerated and transferred to the nearest laboratory for extraction and analyzing. Hence, a portable biomolecules extraction system, which brings several advantages such as reduced labour, reduced waste and increased speed of extracting process, can be a potential development in the future [54]. The combination of portable extraction system with DNA, RNA, or protein analyzer can be build up in the future to help researchers in reducing working time and increasing the work efficiency.

Possible Future Direction

Continued improvement in miniaturization will be the future trend of robotic automation in the laboratory [28]. Many clinical laboratories are performing workflow analysis and finding that smaller systems with lower throughput are more consistent with clinical laboratory workload. Besides, this automation system can be implemented at relatively low cost, improving the turnaround times and also reduce the labor costs [55].

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