

Capillary Electrophoresis in Pharmaceutical and Chemical Analysis: Principles, Techniques, and Applications

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Commentary

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INTRODUCTION

Capillary electrophoresis (CE) is a high-resolution analytical technique widely employed in pharmaceutical, chemical, environmental, and clinical research. It separates charged molecules based on their size-to-charge ratio under the influence of an electric field within narrow capillaries. CE offers several advantages, including high separation efficiency, minimal sample consumption, rapid analysis, and compatibility with various detectors. Its applications include the analysis of drugs, biomolecules, impurities, and metabolites. This article provides a comprehensive overview of CE, covering its principles, types, instrumentation, methodologies, and applications, emphasizing its significance in modern analytical and pharmaceutical sciences.

Capillary electrophoresis; Pharmaceutical analysis; Analytical chemistry; Drug quantification; Biomolecule separation; Impurity profiling; Electrophoretic mobility; High resolution; Microfluidics; Quality control; Method validation; CE instrumentation; Separation techniques; Rapid analysis; Trace analysis

Capillary electrophoresis (CE) has emerged as a versatile and powerful analytical tool, particularly in the analysis of charged molecules. Unlike traditional chromatographic techniques, CE separates analytes in narrow capillaries based on their electrophoretic mobility under an applied electric field. This mobility depends on the charge and size of the molecules, providing high-resolution separation with minimal sample and reagent consumption.

In pharmaceutical analysis, CE is widely used for drug assay, impurity profiling, enantiomer separation, and metabolite quantification. It provides rapid and reproducible results, making it suitable for routine quality control and research applications. CE is also applied in environmental monitoring, clinical diagnostics, and biomolecular research due to its ability to separate nucleic acids, proteins, peptides, and other charged biomolecules. Regulatory agencies, including the

International Council for Harmonisation (ICH) and the U.S. Food and Drug Administration (FDA), recognize CE as a validated method for analytical applications, emphasizing its reliability and precision.

The evolution of CE has led to the development of several variants, including capillary zone electrophoresis (CZE), micellar electrokinetic chromatography (MEKC), capillary gel electrophoresis (CGE), and capillary isoelectric focusing (CIEF). These methods expand the range of analytes and enhance the technique's versatility, enabling high-throughput and trace-level analysis in complex sample matrices.

DESCRIPTION

Principles of Capillary Electrophoresis

The principle of CE is based on the movement of charged particles in an electric field. Key concepts include:

- Electrophoretic Mobility:** The velocity of an ion is proportional to its charge and inversely proportional to its size. Small, highly charged ions move faster than larger, less charged molecules.
- Electroosmotic Flow (EOF):** In CE, the bulk movement of the buffer solution inside the capillary generates a flow that

influences the migration of all analytes. EOF allows neutral molecules to be carried through the capillary and can be manipulated by adjusting the buffer composition or capillary surface properties.

3. Separation Efficiency: CE provides extremely high separation efficiency due to narrow capillaries and uniform electric fields, resulting in sharp peaks and minimal band broadening.

Types of Capillary Electrophoresis

- Capillary Zone Electrophoresis (CZE): The simplest form of CE, separating analytes based on their charge-to-size ratio in a free solution. Widely used for small ions, drugs, and biomolecules.
- Micellar Electrokinetic Chromatography (MEKC): Incorporates surfactants in the buffer to create micelles, allowing separation of neutral compounds along with charged molecules.
- Capillary Gel Electrophoresis (CGE): Uses a gel-filled capillary to separate biomolecules, especially proteins and nucleic acids, based on molecular size.
- Capillary Isoelectric Focusing (CIEF): Separates amphoteric molecules like proteins based on their isoelectric points (pI) in a pH gradient within the capillary.

Instrumentation of Capillary Electrophoresis

A typical CE system consists of:

- Fused-silica capillary: Narrow (25–100 μm internal diameter) to provide high-resolution separation.
- High-voltage power supply: Creates the electric field for analyte migration.
- Buffer reservoirs: Maintain the electrochemical environment and EOF.
- Sample injector: Introduces minute volumes (nanoliters) into the capillary.
- Detectors: Common detectors include UV-Visible absorbance, fluorescence, and mass spectrometry for sensitive and selective detection.

Applications of Capillary Electrophoresis

1. Pharmaceutical Analysis: CE is employed for drug assay, impurity profiling, chiral separation, and monitoring of degradation products. Its minimal sample requirement and high efficiency make it ideal for high-throughput analysis.
1. Biomolecular Separation: CE is widely used for proteins, peptides, nucleic acids, and oligonucleotides. Techniques like CIEF and CGE allow precise characterization of biomolecules in research and clinical laboratories.
1. Environmental Monitoring: CE detects heavy metals, ionic pollutants, and trace compounds in water and soil samples.
1. Clinical Diagnostics: CE quantifies metabolites, therapeutic drugs, and biomarkers in biological fluids, supporting therapeutic monitoring and disease diagnostics.
2. Research and Development: CE assists in drug discovery, metabolite profiling, and structural characterization of molecules in pharmaceutical and chemical research.

Advantages of Capillary Electrophoresis

- High resolution and efficiency, enabling separation of closely related compounds.
- Minimal sample and reagent consumption, reducing operational costs.
- Rapid analysis with short run times.
- Versatile for a wide range of analytes, including small ions and large biomolecules.
- Compatible with multiple detection techniques for enhanced sensitivity and specificity.

Challenges and Limitations

- Limited loading capacity compared to HPLC, affecting preparative applications.
- Sensitivity depends on detector type; UV detection may be less sensitive for low-absorbing compounds.
- Requires careful control of buffer composition, pH, and temperature for reproducibility.
- Method development can be complex for multi-component or highly heterogeneous samples.

CONCLUSION

Capillary electrophoresis is a powerful analytical technique offering high-resolution, rapid, and versatile separation of charged and

neutral molecules. Its application spans pharmaceutical, chemical, environmental, and clinical research, providing reliable data for drug analysis, quality control, biomolecular characterization, and trace-level detection.

CE's advantages include minimal sample consumption, high separation efficiency, compatibility with various detectors, and applicability to a wide range of analytes. Its ability to perform chiral separation, impurity profiling, and biomolecule characterization makes it particularly valuable in pharmaceutical and clinical laboratories.

Despite challenges such as limited sample loading and sensitivity concerns with certain detectors, advancements in instrumentation, microfluidic integration, and coupling with mass spectrometry have significantly expanded the scope and performance of CE. Regulatory agencies recognize CE as a validated, reliable, and reproducible technique for analytical applications, emphasizing its importance in quality control and research.

In conclusion, capillary electrophoresis represents a cornerstone technique in modern analytical science, combining precision, efficiency, and versatility. Its continued evolution ensures it remains integral to pharmaceutical analysis, biomolecular research, and environmental monitoring, supporting innovation, quality assurance, and public health.

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