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Advancing Food Analysis: High-Performance Liquid Chromatography (HPLC) Applications in Ensuring Safety and Quality Standards

Farah Assala Messaoudi

Abstract: In this study, 68 scientific papers were carefully explored from Google Scholar, PubMed, Semantic Scholar, Research Gate, and official websites of high-performance liquid chromatography (HPLC) companies to provide a comprehensive overview of HPLC applications in the food industry. The continued development of the global food industry requires rigorous safety and quality assurance measures. This review examines the role of high-performance liquid chromatography (HPLC) in food analysis, focusing on its main contributions, analytical principles, sample preparation techniques, detectors, and applications. A leading analytical technique developed in the late 1960s, HPLC is revolutionizing food analysis by enabling the accurate detection and quantification of a variety of compounds such as pesticides, mycotoxins, additives, and contaminants. Understanding the principles of HPLC, such as reversed-phase and normal-phase chromatography helps separate compounds based on polarity and hydrophobicity, which are important for different food matrices. Key challenges in HPLC-based food analysis, like matrix complexity, cost considerations, environmental impact, and analyte stability, highlight the need for innovative strategies and optimized methods. This review paper represents a comprehensive data analysis of HPLC applications, describing detailed qualitative and quantitative analysis of various food ingredients, highlighting the versatility and indispensability of HPLC to ensure food safety, quality, and consumer trust.

Keywords: High-Performance Liquid Chromatography (HPLC), Food Analysis, Safety Standard, Quality Control, Analytical Technique, Chromatographic Method.

INTRODUCTION

In the constantly changing environment of the global food industry, it has become more difficult and important to ensure that foods are safe and quality. Due to consumers' demands for transparency, strict regulatory standards, and manufacturers' desire for excellence, the need for precise and reliable analytical procedures has never been greater (Esteki et al., 2019). High-Performance Liquid Chromatography (HPLC) was invented in the late 1960s and early 1970s, it is one of the analytical technologies that provides a full range of capabilities that are crucial for the manufacturing and analysis of food (Reuhs, 2017).

The name "chromatography" is derived from the Greek words "chroma" (color) and "graphein" (to write), therefore chromatography can be described as an ensemble of methods used to separate components in a mixture and present the results in a “Chromatogram” which is a graph (Shukla *et al*., 2023). The primary advantage of HPLC over Gas Chromatography (GC) is that macromolecules can be analyzed because the analytes do not need to be volatile. This method of separation is widely used in the food industry. The HPLC is intended for the detection and quantification of pesticide residues in fruits, vegetables, or cereals. It is essential to ensure that the legally prescribed limits on pesticide levels in food are respected. In addition, it has been instrumental in the identification of mycotoxins such as aflatoxins and ochratoxins, harmful compounds originating from molds that can contaminate cereals, nuts, or other agricultural products (Zhang & Banerjee, 2020). In addition, HPLC is used to assess and quantify food components such as sweeteners, colorants, taste enhancers, and antioxidants as a way to verify that their

Farah Assala Messaoudi, Department of Genetics and Bioengineering, International Burch University, Sarajevo, Bosnia and Herzegovina. E-mail: farah.assala.messaoudi@stu.ibu.edu.ba

concentrations are inside restricted (Nielsen, 2017). This shape of analysis facilitates the detection of pollutants, adulterants, or unlawful substances found in food materials ensuring their authenticity (Nunez *et al*., 2015).

PRINCIPLES OF HPLC

The HPLC system consists of several important parts that work together to ensure accurate and efficient analysis. These include a pump that is responsible for controlling precise flow rates of the mobile phase, an injector for introducing samples, a chromatographic column having a stationary phase for separation, a detector sensing as compounds elute from the column, and finally a data system running the entire process of control and analysis of results. Working seamlessly, all these components make HPLC outstanding as an analytical tool in food analysis, it performs better separation, detection, and measuring of liquid compounds with high precision (Akash & Rehman, 2019). In HPLC, a small volume of liquid sample is injected into the system and carried by the mobile phase passes through a column where the components are separated particles (Ganorkar & Shirkhedkar, 2017). The distribution of a component between the liquid mobile phase and the stationary phase determines how well that component is retained within the column. Compounds have different mobilities and therefore leave the column at different times (retention times) and reach the detector (Chormale *et al*., 2023). In HPLC, both reversed phase chromatography (RP-HPLC) and normal phase chromatography (NP-HPLC) play key roles in the separation and analysis. The term RP-HPLC refers to a chromatography mode in which the mobile phase is polar and the stationary phase is nonpolar or hydrophobic. The polarity reversal between the stationary and mobile phases enables the separation of a wide range of substances, particularly nonpolar or hydrophobic analytes. In RP-HPLC, the stationary phase (the component of the column that interacts with the target analyte) often consists of hydrocarbon chains attached to a solid support material, such as Silica particles, which are bonded and nonpolar or hydrophobic (Noman *et al*., 2016). C18 (octadecylsilane), C8 (octylsilane), C4 (butylsilane), phenyl, and cyano are the most widely used stationary phases in RP-HPLC (Abd Elhafeez & Mobarez, 2021). Moreover, the choice of stationary phase in RP-HPLC depends on the analyte type and specific separation goals (Mishra *et al*., 2021). The mobile phase (the liquid that dissolves the target substance) in RP-HPLC is typically a polar solvent or a combination of water plus an organic solvent that is water-miscible, like acetonitrile or methanol. Compounds are eluted through the column through the mobile phase. In numerous industries, including the food industry, RP-HPLC works best for quantifying hydrophobic and nonpolar substances, including lipids, vitamins, and many organic molecules. For example, the analysis of salicylate content in a wide range of products including spices (curry, oregano, red pepper), beverages (beer, brewed tea, milk, wine), lyophilized fruits (apricot, strawberry, watermelon), and vegetables (cucumber, tomato) using RP-HPLC (Szkop *et al*., 2016). NP-HPLC is a chromatography mode in which the mobile phase is nonpolar and the stationary phase is polar or hydrophilic. The stationary phase in NP-HPLC is composed of polar materials that interact with polar or hydrophilic analytes, such as Silica gel or Aluminum Oxide. The mobile phase, on the other hand, is typically a non-polar organic solvent such as hexane or Diethyl Ether. This nonpolar mobile phase helps elute compounds through the column (Sarker and Nahar, 2015). Although RP-HPLC is more common in the food manufacturing industry, NP-HPLC is still useful in some applications where the analysis of polar substances is required (Harvey, 2019).

Retention time in HPLC is the time it takes for a specific compound to travel from the injection point to the detector. It is a critical parameter used to identify compounds in unknown samples by comparing their retention times with those of known standards, compounds with similar retention times are likely the same or structurally similar. HPLC standards are used to calibrate the system and establish a known reference for the identification and quantification of analytes in unknown samples by comparing their behavior to the known standards (Akash & Rehman, 2019). Aside from the retention time, HPLC involves several critical parameters that are crucial for the separation and analysis of compounds such as the peak width, it is based on the dispersion of analytes as they elute from the column, this measurement determines the width of a chromatographic peak at its base. HPLC also includes the resolution; this parameter reflects the column's effectiveness in separating the desired peaks. Higher resolution implies a greater capacity to achieve clear baseline separation between two adjacent peaks (Sabir *et al*., 2016). In addition to the previously mentioned parameters, we have the selectivity, this factor measures the separation of two compounds based on their affinity for the stationary phase. Changing the components of the mobile phase or the stationary phase can alter selectivity, it can also be affected by temperature. Finally, we have the asymmetry factor, which is a measure used in HPLC to quantify the shape of a chromatographic peak, sometimes referred to as the peak tailing factor. An asymmetry factor of 1.0 represents a symmetric peak, while values greater than 1.0 indicate tailing, and values less than 1.0 indicate fronting or peak distortion (Abdu Hussen, 2022).

SAMPLE PREPARATION

Food samples are complex heterogeneous matrices in which all analytes are randomly distributed. Sampling, homogenization, and sample preparation are steps in the food analysis process that improve the precision and accuracy of the analysis. For example, the analysis of pesticide residues in fruits and vegetables typically involves a multistep process. First, representative samples are collected and homogenized to create a uniform sample for analysis. To create a homogenous sample, the edible parts of the fruits and vegetables are either blended or crushed (Wahab *et al*., 2022). Then, Solid-Liquid Extraction (SLE) is used to extract the pesticide residues from the homogenized sample. Usually, acetonitrile or ethyl acetate is used as the suitable solvent. To help get pesticides into the solvent phase, the extraction mixture is vigorously agitated. The extract is next filtered or centrifuged to separate it from the solid residue (Salman and Ahmed, 2017). To ensure compatibility with HPLC analysis, the resultant liquid extract is subsequently filtered to remove any remaining particles. If necessary, it can also be further concentrated or cleaned up (Vasconcelos *et al*., 2019). The procedure for preparing samples for mycotoxin analysis in fruits and vegetables using High-Performance Liquid Chromatography (HPLC) encompasses various stages, including extraction, purification, and concentration of the mycotoxins from the sample. A widely adopted method for this purpose is QuEChERS (Quick, Easy, Cheap, Effective, Rugged, and Safe), as extensively employed according to Abdulra'uf *et al*. (2022). This sample preparation technique streamlines the process, emphasizing efficiency and cost-effectiveness while ensuring the robust and safe extraction of mycotoxins from complex matrices like fruits and vegetables. The process initiates with the collection and weighing of representative samples, which are subsequently homogenized to attain a uniform matrix. An accurately measured portion of the homogenized sample is then placed in an extraction vessel, and a suitable volume of acetonitrile, often incorporating internal standards for quantification, is introduced. The addition of QuEChERS salts, typically a blend of magnesium sulfate and sodium acetate, induces phase separation, thereby enhancing the efficiency of mycotoxin extraction. Following rigorous shaking, the sample undergoes centrifugation to facilitate the separation of the acetonitrile phase, containing the extracted mycotoxins, from the solid matrix. The resulting supernatant is transferred to another clean tube, and dispersive solid-phase extraction (d-SPE) utilizing specific sorbents like C18 and PSA is employed to eliminate interfering compounds. The cleaned extract undergoes a subsequent round of centrifugation, followed by concentration, an essential step for efficient sample analysis and to mitigate the risk of sample loss to remove excess solvent and reduce the sample volume. The concentrated extract is reconstituted in an appropriate solvent, such as water or acetonitrile, and subjected to filtration to eliminate any remaining particulates (Yu *et al*., 2023). For carbohydrates, the level of preparation required for analysis depends on the characteristics of the food being studied. Certain foods, like nuts, cereals, fruits, bread, and vegetables, contain carbohydrates closely connected with other elements, making it necessary to separate carbohydrates for precise analysis. The specific approach for carbohydrate isolation is influenced by factors such as the type of carbohydrate, the composition of the food matrix, and the analytical objectives. There are many crucial steps in the sample preparation procedure for HPLC carbohydrate analysis. Initially, filtering is used to remove solid particles and contaminants, allowing the HPLC equipment to accurately analyze carbs. Then, reverse osmosis is used to eliminate salts and other ions that may interfere with the analysis. Following this, dialysis is utilized to eliminate small particles like salts and sugars by passing the sample through a semi-permeable membrane. Subsequently, the extraction step ensues, involving the isolation of carbohydrates from the sample matrix using a solvent such as 80% alcohol (McClements, 2019). The extraction of carbohydrates is achieved through the process of boiling the sample with the solvent, and the carbohydrates are then separated from the solvent for subsequent analysis. Post-extraction, the sample undergoes concentration to eliminate surplus solvent and reduce volume, streamlining the analytical process. The concentrated sample gets injected into the HPLC system for analysis, where carbohydrates are separated according to their molecular structure and size. The detection method is evaporative light scattering, which provides information about the molecular weight and structure of carbohydrates. This complex process ensures both accuracy and reliability of carbohydrate analysis (Yan, 2014). A variety of procedures are employed to prepare samples for lipid extraction intended for HPLC analysis, these may range from liquid-liquid extraction (LLE), microwave-assisted technology (MAT), and solid-phase extraction among others. MAE is an effective technique used to extract lipids from various types of food items which include microalgae, fish, and other food commodities (Tanzi *et al*., 2017). The use of this technique has different advantages for lipid extraction such as shorter extraction durations, greater lipid production, and the possibility of using green or low-cost solvents (Ferrara *et al*., 2023). It is a method that involves microwave radiation to hasten the extraction process that starts with sample preparation through grinding or homogenizing, and then the use of a proper solvent that can effectively extract lipids out of food matrices as well as perform

HPLC analysis. Hexane or an eco-friendly solvent can be utilized for this purpose. Next, the prepared sample is mixed with the chosen solvent, after exposure to microwaves, which heat the mixture making the extraction process faster (Medina *et al*., 2015). Additionally, the mixture is filtered yielding an organic phase concentrated with lipids. Solvent evaporation may be used to collect and concentrate lipid-rich phases (Kataoka, 2018). LLE is a widely accepted and effective technique for extracting lipids before HPLC analysis. This method uses a special solvent (usually a combination of chloroform and methanol) to separate lipids from a complex sample matrix. After the separation process, the lipid-rich phase is isolated and a washing step is performed to remove unwanted impurities. The concentrated lipid content is then extracted by evaporating the solvent, then concentrated lipid extract can be reconstituted if desired before introduction into the HPLC system. To enable accurate quantification of lipids in HPLC analysis, LLE provides an efficient and reliable approach for differentiating lipids from complex matrices (Reichl *et al*., 2020). In HPLC, sample preparation is a crucial process that has a big impact on the accuracy and reliability of analytical results. The prepared sample should be an aliquot that is compatible with the HPLC procedure and will not damage the column, and it should be relatively clear of interferences (H. Snow, 2017).

HPLC DETECTORS

The de-tector of the HPLC system is place-d right at the end. It's there- to examine the solution coming out from the- column. Each part of the mixture create-s an electronic signal, which is linked to the-concentration of each analyte compone-nt. HPLC uses many detectors for the- job of measuring and quantifying chemicals in food analysis:

Fluorescence HPLC Detectors

HPLC fluorescence detectors are very sensitive and specific. It is used in measuring the fluorescence of material after absorbing light at a specific wavelength. These detectors are particularly appropriate for the detection of vitamins like riboflavin, and chlorophyll as well as diverse food additives. Dansyl chloride fluorescence derivatives may be used to detect compounds with poor or no fluorescent properties (Sunil *et al*., 2018). This approach is especially helpful in the analysis of lipids because it makes possible the identification of fluorescent lipids. The fluorescence detectors provide high sensitivity and selectivity, which makes them an important tool in a variety of testing protocols including food safety and nutritional analysis.

Mass Spectrometry (MS)

MS detectors identify substances based on their mass-to-charge ratios and provide information about their molecular structure. An MS detector may be able to directly detect a compound since its mass spectrum is like a fingerprint and corresponds to that compound. Mass spectrometers are powerful detectors for food analysis Ideal for complex matrices, they can be used to identify and quantify a variety of substances such as pesticides, mycotoxins, and food contaminants (De Girolamo *et al*., 2022).

UV-Visible Detector (UV-Vis)

UV-Vis detectors operate by measuring the absorption of ultraviolet and visible light by analytes as they elute from the column (different compounds absorb light at characteristic wavelengths), they are based on electronic transitions within molecules. They are suitable for compounds with chromophores (double bonds or aromatic rings) and are commonly used in food analysis for quantifying organic molecules like vitamins, pigments, and flavor compounds (Nie and Nie, 2019).

Refractive Index Detector (RI or RID)

Refractive index detectors (RIDs), which are frequently used in High-Performance Liquid Chromatography (HPLC) for food analysis, work by detecting changes in the refractive index of eluting compounds. This detector is especially helpful for quantifying substances like sugars and other non-chromophoric analytes that have no inherent UV-Vis absorption or fluorescence properties (Başaran *et al*., 2017). The RID provides an output signal, typically in milli-refractive index units (mRIU) or micro-refractive index units (μRIU), which corresponds to the concentration of the analyte, this signal is used for quantification in HPLC analysis (Al-Sanea and Gamal, 2022).

QUALITATIVE AND QUANTITATIVE ANALYSIS

Quantitative Analysis

In quantitative analysis, HPLC allows precise evaluation of a variety of food components. For the regulation of food quality, regulatory compliance, and proper labeling, this quantitative precision is essential. Food additives concentration including artificial colorants, preservatives, and sweeteners are frequently measured using HPLC. For instance, a study measured the quantities of synthetic food dyes present in various confectionery products. The accurate measurement of these compounds ensures that food items adhere to labeling requirements and regulatory guidelines (Catenza and Donkor, 2021). In this analysis, HPLC can also detect and quantify contaminants in food, including pesticides, mycotoxins, and other organic contaminants. For example, in the analysis of pesticide residues in vegetables, HPLC can quantify the levels of multiple pesticide residues, ensuring that they are within permissible limits and pose no risk to consumers. Quantitative analysis also appears when detecting essential nutrients like vitamins, amino acids, and fatty acids. An example is the quantification of vitamin C in various fruit juices, a critical aspect of assessing the nutritional content and quality of these products (Cortés-Herrera *et al*., 2018).

Qualitative Analysis

In qualitative analysis, HPLC is crucial for the identification and characterization of complex compounds, such as flavors, fragrances, and natural pigments. By comparing their retention times and spectral data, it is possible to pinpoint the presence of target compounds. HPLC can determine whether a product aligns with its claimed origin or characteristics. For instance, in the analysis of olive oils, HPLC can assess the levels of phenolic compounds to verify the oil's variety and origin, ensuring consumers receive authentic and high-quality products (Benincasa *et al*., 2022).

CHALLENGES AND LIMITATIONS OF HPLC IN FOOD ANALYSIS

Matrix Complexity

The challenges presented by the matrix complexity in HPLC for food analysis are numerous. Food matrices are complex mixtures of different substances, including carbohydrates, sugars, proteins, lipids, and additives. These substances have the potential to result in matrix effects, which may interfere with chromatographic separation and detection. Matrix effects manifest as changes in retention times, decreased chromatographic resolution, or analyte signal suppression or enhancement (Nasiri *et al*., 2021). The matrix's intricacy can make obtaining accurate and exact measurements difficult, especially when studying trace-level components or determining the presence of contaminants among an array of food elements (Raposo and Barceló, 2021). To reduce matrix interferences and improve the accuracy of analytical results, intensive sample preparation methods like extraction, clean-up, and sample dilution are frequently needed. Furthermore, the use of selective chromatographic techniques and detectors capable of separating between analytes and matrix constituents is essential in overcoming matrix complexity challenges in HPLC-based food analysis (Steiner *et al*., 2020).

Cost Considerations

The challenge of cost considerations for HPLC in the food industry has been a topic of discussion in recent years. According to a report by Technavio, one of the major challenges to the growth of the global HPLC market is the high cost of HPLC equipment, including systems, detectors, columns, pumps, and other equipment. Additionally, ongoing operating costs include maintenance, consumables (solvents, standards), and repeated calibration, all of which contribute to overall costs (Soyseven *et al*., 2023). Operating costs increase due to the need for trained personnel to operate and maintain equipment (Chen, 2022). However, it is important to note that although HPLC systems are expensive, they offer benefits such as accurate detection of product quality issues, high-throughput analysis automation, and regulatory compliance regulations, which can help avoid costly product recalls and protect the trademark reputation of the food industry (Farré and Barceló, 2020). Therefore, to establish and use HPLC systems in the food sector, it is imperative to understand their cost-effectiveness in the context of food analysis.

Environmental Impact

The environmental impact of HPLC in the food sector is a complex issue that includes many different factors, such as waste generation, energy consumption, and use of hazardous solvents (Kannaiah *et al*., 2021). Organic solvents such as acetonitrile and methanol commonly used in HPLC can contribute to air and water pollution if not properly treated and disposed of (Armenta *et al*., 2022). Additionally, the energy used to operate HPLC equipment such as pumps, detectors, and cooling systems contributes to the

industry's overall carbon footprint (Nowak *et al*., 2023). Many different materials are used in the manufacture of HPLC columns and instruments, some of which can have negative environmental impacts during extraction, processing, and disposal (Majors, 2015). The adoption of greener solvents, recycling or appropriate solvent disposal, and the development of energy-efficient processes and equipment are all necessary to reduce the environmental impact of HPLC in the food industry (Nakov *et al*., 2023). Additionally, finding alternative environmentally friendly column materials and techniques that limit solvent consumption and waste generation is essential to address these environmental issues.

Analyte Stability

Analyte stability is a major challenge when using HPLC in the food industry, and the stability of the analyte (the compound of interest), is critical to obtaining accurate and reliable results. Food matrices are complex and dynamic, containing a variety of molecules that can degrade or change over time due to various factors such as temperature, pH, light exposure, and enzyme activity (Mohammed *et al*., 2023). Such changes can lead to analyte instability, affecting the molecular structure and concentration of the analyte, and ultimately affecting the results of HPLC analysis. For example, vitamins (Nestola and Thellmann, 2015), antioxidants (Cheng *et al*., 2023), or volatile compounds in food samples can be degraded during sample preparation or analysis, resulting in potentially inaccurate quantification or identification. Additionally, sample storage conditions play an important role in analyte stability (Ohnmacht *et al*., 2019). Improper handling, prolonged storage, or exposure to unsuitable environments can worsen the challenge. To address this challenge, HPLC-based food analysis requires careful sample preparation, immediate post-extraction analysis, and the use of stabilizing agents controlled storage conditions are essential to maintain analyte stability and ensure the reliability of results (Vasconcelos *et al*., 2018).

RESULTS AND DISCUSSION

Table 1 shows different food products analyzed by the HPLC. The quantification of the sugar substances (Glucose, Fructose, Sucrose) in onions using HPLC-RID (Magwaza and Opara, 2015), (Davis *et al*., (2007). Since sugars don't include UV chromophores, RI detection is preferred for sugar analysis over UV-based detection. However, because sugars exhibit UV absorption below 200 nm, this restriction does not eliminate UV-based detection (Jalaludin and Kim, 2021). According to Trani *et al* (2017), the qualification of Lactose using NP-HPLC and RID as a detector is crucial since this substance should not be contained in Lactose-free milk, this substance can be harmful to people with lactose intolerance (LI) which is A genetically based condition linked to lactase deficiency (Malik and Panuganti, 2023).

Organic acids can also be quantified using HPLC and UV-Vis as a detector in a tomato sample, these organic acids are significant elements that regulate tomato fruit flavor and crucial breeding qualities, however the quantity of Citric acid in a tomato should be under limits that vary from 0.3% to 0.6% (Felföldi *et al*., 2021).

Like fruits and vegetables, juices can also be quantified using HPLC, for instance, cranberry juice contains acids (Quinic Malic, Ascorbic, Shikimic, Citric) that can be analyzed, quantified using HPLC and detected with a UV-VIS detector ensuring that they are within permissible limits claimed by the manufacturer (Sai Lakshmi *et al*., 2022). The studies conducted by Abdullah AlFaris et al. (2020), Lee et al. (2007), and Tuzimski and Rejczak (2016) showcase the diverse applications of HPLC in the analysis of various substances in complex matrices. In 2020 Abdullah AlFaris *et al* investigated the presence of aflatoxins, including AFB1, AFB2, AFG1, and AFG2, in a variety of cereals and nuts, such as white almond, almond, walnut, peanut, broad bean, and pistachio. The analytical approach employed a Methanol (CH3OH): water solvent system and a Zorbax 5 mm Eclipse-XDB-C18 column with an 80 Å particle size in conjunction with HPLC. The detection of aflatoxins was achieved using fluorescence, showcasing the sensitivity of the method in identifying these mycotoxins. Lee *et al*'s work in 2007 emphasized the detection of several antibiotics in different matrices. According to **Table 1,** the antibiotic detected was Tylosin in beef using a C18 column and Acetonitrile (CH3CN): Methanol (CH3OH): Ammonium phosphate: ((NH4)2PO4) solvent system, employing UV-Vis detection. HPLC can also detect pesticides according to Tuzimski and Rejczak. Their study was focused on the analysis of different pesticides in olive oils using RP-HPLC with ZORBAX Eclipse XDB-C18 column and Diode Array Detector (DAD).

According to **Table 1**, the mobile phase and the column changing depend on the nature of the analytes and their compatibility with certain columns or mobile phases due to chemical interactions or adsorption. The selection of column and mobile phase can have significant effects on the analysis's sensitivity. Tailoring these parameters helps in the optimization of the conditions for achieving lower detection limits in quantification or for identifying trace compounds in qualitative analysis. In quantitative analysis,

having well-separated peaks is crucial for accurate quantification, by changing the column and mobile phases, the resolution can be enhanced between the target compounds, reducing the risk of co-elution and ensuring precise quantification. In qualitative analysis, better resolution aids in the identification of compounds with precision. The chemical characteristics of many food molecules vary, including polarity, charge, and size. by modifying the column and mobile phase, enhancing the separation and selectivity for the particular analytes of interest can be achieved (Žuvela *et al*., 2019). These investigations underscore the versatility of HPLC in diverse fields, ranging from food safety to pharmaceuticals, with variations in column types, solvent systems, and detection methods tailored to specific analytical needs. Conclusions drawn from this comparative analysis could

highlight the importance of selecting appropriate HPLC parameters based on the targeted analytes and matrices to achieve accurate and reliable results in analytical research.

Table 1: Applications of high-performance liquid chromatography to measure various analytes of different food products (quantitative/qualitative analyses)

Product	**Analyte**	**Mobile phase (eluent)**	**Column**	**Detector**	**Technique**	**Reference**
Onions	Sucrose, Glucose, Fructose	Acetonitrile (CH3CN): water	Waters Carbohydrate	RID	HPLC	Magwaza and Opara (2015) Davis *et al*. (2007)
Lactose-free milk	Lactose	Acetonitrile (CH3CN): water	Spherisorb Amino (NH2) column 80 Å	RID	NP-HPLC	Trani *et al*. (2017)
Tomato	organic acids: Citric acid, Tricarballylic acid (IS)	A: Ammonium phosphate: ((NH4)2PO4) B: Ammonium phosphate: ((NH4)2PO4) in 10% Acetonitrile (CH3CN)	Nucleodur C18ec, 5 µm, 250x 4.6 mm; 25°C	UV-Vis	RP-HPLC	Agius *et al*. (2018)
Cranberry juice	Acids: Quinic, Malic Ascorbic, Shikimic, Citric	potassium phosphate buffer (K3PO4)	PerkinElmer Brownlee Validated Aqueous C18	UV-Vis	HPLC	Sai Lakshmi *et al*. (2022)
Cereals, nuts (white almond, almond, walnut, peanut, broad bean, pistachio)	Aflatoxins (AFB1, AFB2, AFG1, and AFG2)	Methanol (CH3OH): water	Zorbax, 5 mm Eclipse-XDB-C18, 80 Å	fluorescence	HPLC	Abdullah AlFaris *et al*. (2020)
Beef (small intestine)	Antibiotic (Tylosin)	Acetonitrile (CH3CN): Methanol (CH3OH): Ammonium phosphate: ((NH4)2PO4)	C18 (5µm, 250mm×4.6 mm)	UV-Vis	HPLC	Lee *et al*. (2007)
Olive oils	Pesticide	Acetonitrile (CH3CN): water	ZORBAX Eclipse XDB-C18 (5µm, 150 mm×4.6 mm)	DAD	RP-HPLC	Tuzimski & Rejczak, (2016)

CONCLUSION

HPLC facilitates the precise quantification of various food components, including additives, contaminants, and essential nutrients The flexibility of HPLC, with its ability to adapt columns and mobile phases for specific analytes, makes it an indispensable tool in the food industry, where diverse food matrices and compounds are encountered. Simultaneously, it empowers qualitative analysis by aiding in compound identification, characterization, and the authentication of food products, contributing to transparency and trust in the food supply chain. As the food industry continues to grow and evolve, HPLC remains at the forefront, ensuring the integrity and safety of the products we consume.

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CONFLICTS OF INTEREST

The author declares no conflicts of interest regarding the publication of this review paper. There are no financial, personal, or professional interests that could be construed as influencing the content or conclusions presented herein. The author is committed to maintaining objectivity and integrity in all aspects of the research and publication.

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