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Simultaneous Determination of Potassium Sorbate and Sodium Benzoate in Processed Food Samples Available in Mashhad Market with a Validated Method Using HPLC-DAD

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Abstract
Measuring preservative concentrations in food is crucial due to their potential health implications. Therefore, it is necessary to develop a reliable and validated analytical method for monitoring. This study focuses on the validation of a simple, sensitive and precise analytical approach using HPLC-DAD to concurrently assess sodium benzoate and potassium sorbate. The validation process resulted in exceptional outcomes, demonstrating strong linearity (R2 > 0.999), precision (RSD < 5%), and accuracy (recovery values ranging from 90.77% to 100.55%). Additionally, the method exhibited low limits of detection and quantification (0.8 and 2.65 mg L-1 for sodium benzoate, and 0.14 and 0.47 mg L-1 for potassium sorbate, respectively). The effectiveness of the validated method on different food metrics was shown by analyzing 110 samples of Olivier salad, dairy products and ketchup sauce from Mashhad, Iran. Comparison of the results with Iran's national standards revealed that the preservative concentrations in most samples were within the acceptable limits set by Iranian regulations.

Keywords
HPLC-DAD, Preservatives, Method validation, Food processed.

1.INTRODUCTION

Food spoilage refers to any changes in food that render it unsuitable for human consumption. The main factors of food spoilage include chemical and physical changes in food, microbial contamination, endogenous enzymatic degradation, and infestation by insects. Food preservation, on the other hand, is a scientific discipline that aims to prevent food decay or spoilage while maintaining its nutritional value, taste, texture, and color. In fact, food preservation stops spoilage, reduces the severity of spoilage and safeguards against food-borne diseases [1]. Preservatives are substances or chemicals used to prevent spoilage, undesired chemical change, and deterioration of quality and nutritional value in various products such as food, drugs, dyes, cosmetics, and biological samples [2]. Methods for preserving food can encompass either physical, chemical, or a combination of both techniques. Physical preservation methods include processes such as dehydration, cooling, freezing, heat treatment, vacuum packing, exposure to ultraviolet radiation, and canning [3-6]. On the other hand, chemical preservation involves the incorporation of preservatives into food. Preservatives are intentionally added to food items to hinder or postpone microbial, enzymatic, or chemical alterations that can lead to nutrient degradation [7]. Common antimicrobial preservatives in the food industry include sodium and potassium salts of benzoic acid and sorbic acid. These preservatives find extensive application in products like sauces, dairy items, baked goods, beverages, meat products, and more [8]. They are valued for their wide-ranging antifungal and antibacterial properties without compromising the sensory qualities of food [9]. The antimicrobial action of these preservatives arises from their ability to permeate cell membranes and acidify the cytoplasm, prompting the cell to utilize ATP to restore cytoplasmic pH, thereby impeding microbial growth. It is worth noting that although potassium sorbate and sodium benzoate are generally considered safe, excessive consumption of these preservatives can pose risks to human health and safety [10]. Studies have shown that

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high intake of these preservatives can trigger allergic reactions such as urticaria, convulsions, and asthma [11-13]. For the use of preservatives in food, there are national and international legislation and guidelines, in which the maximum allowed consumption of these preservatives according to the type of food and conditions of use are precisely mentioned. Therefore, effective food control is required to ensure compliance with the above-mentioned laws and to ensure safety. Various analytical methods such as Electrophoresis [14, 15], Colorimeter [16], UV-visible spectroscopy [17] and HPLC [18, 19], have been proposed for the determination of preservatives. Among these methods, HPLC method has higher accuracy and sensitivity.

In this research, a liquid chromatography technique was developed to concurrently identify and quantify potassium sorbate and sodium benzoate. The method involved a straightforward sample extraction process, followed by separation using RP-HPLC (Reverse Phase High-Performance Liquid Chromatography) and detection through a diode array detector (DAD) at a single wavelength of 254 nm. The quantification of the preservatives was achieved using the calibration curve equation. To validate the proposed method, various parameters including linearity, range, precision, and accuracy, limits of detection (LOD), limits of quantification (LOQ), system suitability, and robustness were assessed.

Due to the significance of sodium benzoate and potassium sorbate as widely employed preservatives in the food industry, their acceptable levels are governed by both national and international regulations. This study aimed to investigate the presence of sodium benzoate and potassium sorbate in three food categories: dairy products, ketchup, and Olivier salad, encompassing a total of 110 samples. According to Iranian regulations, the use of these preservatives is prohibited in dairy products, whereas their addition is permitted in ketchup and Olivier salad. The Iranian Standard and Industrial Research Institute (ISIRI) has established maximum limits for the total of these preservatives in ketchup sauce (750 mg kg^{-1}) and Olivier salad (150 mg kg^{-1}) and exceeding these limits is considered a violation. It is evident that controlling the levels of these preservatives is vital for compliance with regulations and human health. This study aimed to present an easy, rapid, and accurate method for monitoring these preservatives to address these concerns.

2.EXPERIMENTAL

2.1. Sample collection

The research was carried out using a total of 110 samples collected from Mashhad, Iran. The samples consisted of 50 dairy products, 31 ketchup samples, and 29 Olivier salad samples, each sourced from different brands. The dairy samples encompassed yoghurt, cheese, and doogh, which were conveniently accessible within our department for the purpose of our control activities. It is worth noting that careful consideration was given to selecting samples that are representative of popular choices in the consumer market.

2.2. Chemicals and Materials

For the chromatographic procedure, high-performance liquid chromatography (HPLC) grade solvents, specifically methanol and water, were employed. Other reagents, including Ammonium acetate, Potassium hexacyanoferrate(II)trihydrate, Zinc acetate, and Glacial acetic acid, were of analytical grade and were sourced from Merck, a reputable manufacturer based in Darmstadt, Germany. Deionized water was consistently used throughout the experiments. The necessary standards for the analysis, sodium benzoate, and potassium sorbate, were prepared using materials from Sigma, a trusted supplier of laboratory chemicals and materials.

2.3. Apparatus

The chromatographic analysis was carried out using a High-performance liquid chromatography system from Dionex (UltiMate 3000, US), which was equipped with a diode array detector (DAD), an automated sample injector, and a thermostatted column compartment oven (TCC-100). In this method, a single wavelength (λ) is employed, making it possible to use a UV detector. The chromatographic separation was achieved using an RP-C18 column (4.6 mm × 250 mm, 5 µm) from Thermo (US), which was coupled with the corresponding C18 guard-column. Data integration was performed using Thermo Scientific Chromeleon version 7 software.

2.4. Mobile phase preparation

The mobile phase was composed of 65% ammonium acetate buffer with a pH of 4.2 and 35% HPLC grade methanol. To prepare the ammonium acetate buffer with a pH of 4.2, the following steps were taken:

An accurate weight of 0.30 grams of ammonium acetate was measured. The ammonium acetate was dissolved in approximately 900 ml of HPLC grade water in a 1-liter beaker. 0.5 ml of glacial acetic acid was added to the solution. The pH of the solution was adjusted to 4.2. The volume of the prepared buffer solution was adjusted to 1 liter. The buffer solution intended for HPLC use was then filtered using cellulose acetate filters (47 mm × 0.45 µm) and degassed using ultrasonic treatment.

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2.5. Standard preparation

A stock standards solution with a concentration of 1000 mg L^{-1} was prepared as follows: Exactly 100.00 mg of both sodium benzoate and potassium sorbate were added to a 100.0 ml volumetric flask. The flask was then filled to its volume with deionized water and shaken for 2 minutes to ensure thorough mixing.

For the working mix standard solutions, appropriate volumes of the stock solutions were diluted with a mixture of methanol and water in a ratio of 65:35. Notably, the concentration of sodium benzoate in the working mix standard was five times higher than the concentration of potassium sorbate.

It is essential to store all solutions away from light. For daily use (short-term storage), solutions should be kept at 4°C, and for inter-day (long-term storage), they should be stored at -20°C.

2.6. Simultaneous analysis of sodium benzoate and potassium sorbate

2.6.1 Sample preparation

A sample weighing 10.00 ± 0.01 g was accurately measured and placed in a volumetric flask. Subsequently, 40 ml of methanol were added to the flask. Following that, 2 ml of potassium ferrocyanide solution and 2 ml of zinc acetate solution were introduced into the mixture. The resulting solution was subjected to stirring by a magnetic stirrer for 15 minutes. Afterward, the solution was brought to its designated volume with deionized water and stirred for an additional 2 minutes. To eliminate any particulate matter, the solution underwent a filtration process. Initially, it was filtered using filter paper, and then it was further passed through a polytetrafluoroethylene (PTFE) syringe filter with a pore size of 0.45 µm. The filtered solution was subsequently transferred into a vial for injection. For HPLC analysis, an autosampler injected 20 µl of the prepared solution. The peaks corresponding to sodium benzoate and potassium sorbate were identified based on their respective retention times (ISIRI No. 2454, 2015; ISIRI No. 17813, 2018).

2.6.2 HPLC conditions

The analytes, sodium benzoate and potassium sorbate, were separated using reversed-phase high-performance liquid chromatography (RP-HPLC). Eluent A was a 65% aqueous ammonium acetate buffer, and eluent B was composed of 35% methanol. The separation process occurred at a flow rate of 1 mL min^{-1}, and the column temperature was maintained at 30 °C.

To optimize data collection, each sample was analyzed over a 12-minute period. The injection volume for each sample was set at 20 µL. The peaks corresponding to sodium benzoate and potassium sorbate were detected and quantified at a wavelength of 254 nm.

2.6.3. Qualitative and quantitative analysis

In order to identify the preservatives of sodium benzoate and potassium sorbate, their retention time was compared with the standard solution of the respective. Preservatives were detected at 254 nm wavelength. Quantitative analysis was conducted utilizing the external standard plot method. This involved injecting various concentrations of the standard mix solution containing potassium sorbate and sodium benzoate. To determine the concentration of the analytes, a calibration curve was generated by plotting the peak area against the concentration using the equation of the regression line.

2.7. Method validation

2.7.1. Linearity and range

The studied method was validated based on the international guidelines of ICH (International Conference on Harmonization) [20]. In order to evaluate linearity, a calibration curve was plotted for sodium benzoate in the concentration range of 2.5 -100 mg L^{-1} and for potassium sorbate in the concentration range of 0.5 - 20 mg L^{-1}. Standard solutions were prepared as a mixture of sodium benzoate and potassium sorbate by diluting the stock standard solution. According to the obtained correlation coefficient, it was observed that there is a good linearity between the obtained signal (analyte peak area) and the analyte concentration. In method validation, range was evaluated and the minimum specified ranges of 70 to 130% of the test concentration were considered. The results unequivocally demonstrated that this method exhibits a satisfactory level of linearity for samples containing analyte quantities falling within the specified range or at their maximum concentration.

2.7.2. Precision and accuracy

Method precision (repeatability) was assessed by conducting seven sets of test samples on the same day, representing intraday precision. Precision was evaluated by calculating the average of repeated results and the Relative Standard Deviation (RSD). Accuracy was assessed by determining the recovery of the method at various concentrations. To accomplish this, a sample without preservatives was selected, and known quantities of the working solutions (containing the analytes) were added to it. Three different levels of spiking concentrations were investigated for each analyte, aiming to cover the entire range of interest. Each spiking level was tested with three replicates. Accuracy was evaluated based on the repeatability of the method,

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calculating the average and RSD for each set of replicates.

2.7.3. Detection limit and quantitation limit

The Limit of Detection (LOD) is the lowest amount of analyte in a sample that can be detected but not precisely quantified. In contrast, the Limit of Quantitation (LOQ) represents the minimum analyte quantity in a sample that can be both accurately and precisely measured.

To determine the LOD and LOQ values, a specific calibration curve was constructed using samples containing the analyte within the studied range. These values were derived from either the residual standard deviation or the standard deviation of the y-intercepts of the regression lines. The LOD was calculated as 3.3 times the standard deviation, while the LOQ was defined as 10 times the standard deviation. In essence, the LOD serves as a threshold for detection, while the LOQ offers a more robust measure for accurate and precise quantification.

2.7.4. System suitability

System suitability testing plays a pivotal role in the validation of analytical methods, as it assesses the overall performance of the integrated system, encompassing equipment, electronics, analytical procedures, and the samples intended for analysis. Its primary purpose is to verify that the system is operating correctly and capable of generating dependable and precise results. In the context of HPLC methods, specific default criteria are employed for system suitability evaluation. These criteria encompass the separation factor, which should exceed 1.1, the resolution factor, which should surpass 2, and the tailing factor, which should be less than 2 [21]. These values serve as indicators of the efficiency of the separation, the extent of peak resolution, and the symmetry of the resulting peaks, respectively. The findings from this study affirm that the system meets the requisite criteria, underscoring its suitability for the intended analytical purpose. In the context of this study, the results obtained suggest that the system utilized is indeed suitable for the intended purposes.

2.7.5. Robustness

Robustness refers to the capability of an analytical method to maintain its reliability and accuracy even in the presence of small variations in method parameters. These variations can occur intentionally or unintentionally during the validation or routine usage of the method. Within the scope of this study, the robustness of the method was evaluated by investigating how variations in two critical factors, namely the pH of the mobile phase and the composition of the mobile phase, impacted the results. Through purposefully introducing minor alterations in these parameters, the study sought to determine whether the method remained resilient and if the results consistently held up

3.RESULTS AND DISCUSSION

This study presents a rapid and straightforward method for quantifying sodium benzoate and potassium sorbate in ketchup, dairy, and Olivier salad samples. Simultaneous measurement of these two preservatives in the samples was achieved using an HPLC system equipped with a UV/diode array detector (DAD) and a C18 column measuring 4.6 mm × 250 mm with a particle size of 5 µm. For the analysis, an isocratic mobile phase was employed, consisting of 65% ammonium acetate buffer (A) and 35% HPLC-grade methanol (B). The flow rate was maintained at 1 mL min^{-1}, and the column temperature was held at a constant 30 °C during the analysis. 20 µL sample was injected into the system, and the detection of sodium benzoate and potassium sorbate was carried out at a wavelength of 254 nm. The chromatogram, presented in "Figure 1," illustrates the clear separation of the sodium benzoate and potassium sorbate peaks. The retention times for sodium benzoate and potassium sorbate were determined to be 7.67 and 10.02 minutes, respectively.

3.1 Method validation

In order to validate the methodology employed in this research, various parameters including linearity, range, precision, accuracy, limits of detection (LOD), limits of quantification (LOQ), system suitability, and robustness were assessed, as recommended by relevant guidelines [22-24].

The linearity of the method was determined through the calculation of the square of the correlation coefficients (R^2) obtained from the calibration curves. To establish these calibration curves, which depict the relationship between peak area and concentration, a series of working solutions was prepared. These solutions contained sodium benzoate at levels of 2.5, 5, 10, 20, 40, and 100 mg L^{-1}, and potassium sorbate at levels of 0.5, 1, 2, 4, 5, and 20 mg L^{-1}, as illustrated in "Figure 2". As indicated in the results presented in "Table 1," correlation coefficients greater than 0.999 were obtained for both sodium benzoate and potassium sorbate. This demonstrates an excellent linear correlation between peak area and the concentration of these preservatives.

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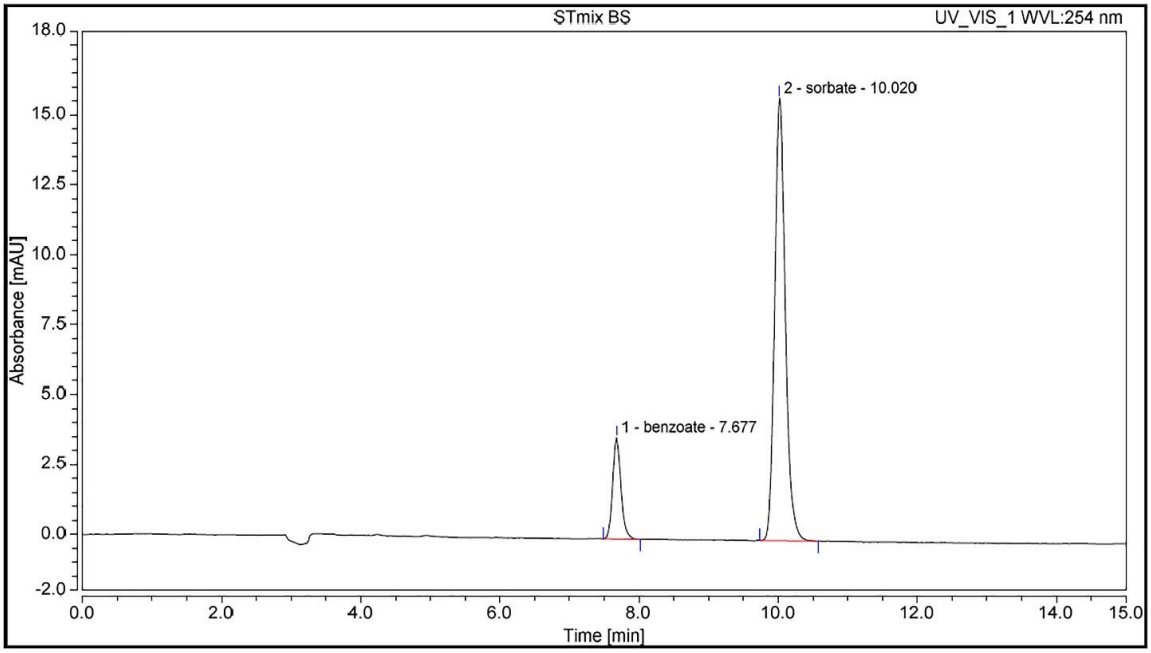

Fig. 1. HPLC chromatogram obtained for mixed standard solution.

Table 1. Validation data of the proposed HPLC method

Analyte	Regression equation	Correlation coefficient (R^2)	repeatability (% RSD[a]) (n = 7)	LOD[b] (mg L^{-1})	LOQ[c] (mg L^{-1})	Measurement Uncertainty (%)
Sodium benzoate	Y= 0.1183X + 0.0223	1.00	3.81	0.80	2.65	1.07
Potassium sorbate	Y= 3.2544X – 0.361	0.9998	3.69	0.14	0.47	0.50

[c]Limit of quantitation

[a]Relative standard deviation.

[b]Limit of detection.

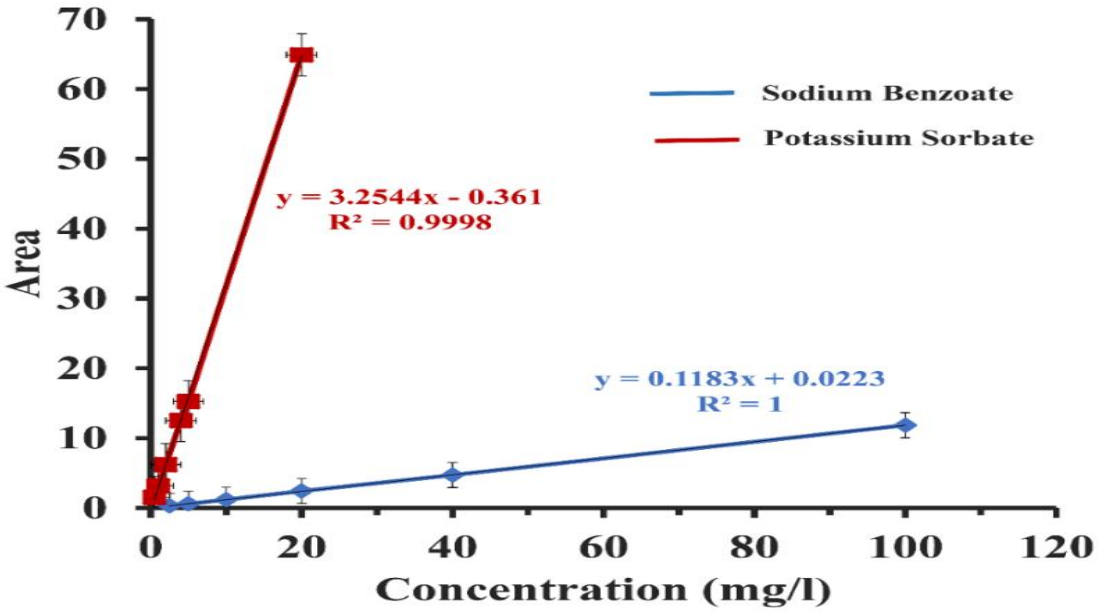

Fig. 2. Standard calibration curve of sodium benzoate and potassium sorbate at 254 nm.

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To assess the precision and repeatability of the method, seven replicate measurements of the preservative amounts were conducted on the test sample within a single day. The relative standard deviation (RSD) was then calculated for these replicate measurements.

The obtained values, as shown in "Table 1", indicate good repeatability of the method, demonstrating consistent and reproducible results for the analysis of sodium benzoate and potassium sorbate.

To assess the method's precision, we calculated the recovery of spiked samples. In this process, a known quantity of sodium benzoate and potassium sorbate was introduced into a blank sample (sample without preservative) and analyzed using the provided methodology. The outcomes are summarized in "Table 2," revealing recovery rates ranging from 90.77% to 100.55% for sodium benzoate and 94.51% to 99.26% for potassium sorbate. These elevated recovery rates demonstrate that the method remains unaffected by sample matrix effects and maintains a high level of accuracy.

In accordance with "Table 1," the LOD values for sodium benzoate and potassium sorbate within the samples were determined to be 0.80 mg kg^{-1} and 0.14 mg kg^{-1}, respectively. Furthermore, the LOQ values for sodium benzoate and potassium sorbate were established at 2.65 mg kg^{-1} and 0.47 mg kg^{-1}, respectively.

Table 3. System suitability parameters of the proposed HPLC method.

Parameters	DAD[a]
Separation factor	1.28 ± 0.02
Resolution factor	2.78 ± 0.26
Tailing factor (Sodium benzoate)	1.06 ± 0.06
Tailing factor (Potassium sorbate)	1.05 ± 0.05
Number of theoretical plats (Sodium benzoate)	8121 ± 23
Number of theoretical plats (Potassium sorbate)	3000 ± 19

[a]Diode array detector.
The results is expressed as "mean ± SD" (n=7).

Prior to each validation phase, we conducted a system suitability assessment of the chromatographic system. Seven sample replicates were prepared in accordance with the prescribed method and then subjected to HPLC. Key system suitability parameters, including separation factor, resolution factor, number of theoretical plates, and tailing factor, were evaluated, and the results are documented in "Table 3."

The method's robustness was assessed through alterations in chromatographic parameters, including varying the pH of the acetate buffer (from 4.2 to 4.1 and 4.3) and adjusting the mobile phase composition (from 65:35 v/v to 64:36 and 66:34 v/v). By computing the relative standard deviation (all of which were below 5%) and the resolution factor (each exceeding 2), it was determined that modifications in these conditions did not lead to significant deviations in the results. Consequently, the method presented in "Table 4" demonstrates a strong level of robustness.

Table 4. Robustness parameters of the proposed HPLC method.

Parameters	Sodium benzoate		Potassium sorbate	
	PH changes	mobile phase changes	PH changes	mobile phase changes
RSD[a] - Retention time (%)	4.22	1.46	1.94	1.59
RSD - Peak Area (%)	1.12	5.00	2.90	3.33
Resolution factor (mean)	2.54 ± 0.14	2.30 ± 0.13	2.54 ± 0.14	2.30 ± 0.13

[a]Relative standard deviation.
The results related to resolution factor are reported as "mean ± SD" of three measurements.

3.2 Analysis of samples

In this research, a validated method was applied to assess the levels of sodium benzoate and potassium sorbate in a total of 110 food samples collected from supermarkets in Mashhad. These samples were categorized into three groups: Olivier salad (n = 29), dairy products (n = 50), and ketchup (n = 31). A summary of the findings is presented in "Table 5." In accordance with the Iranian National Standard (ISIRI No. 17813, 2018), the maximum allowable concentration of sodium benzoate and potassium sorbate, whether individually or in combination, in Olivier salad is set at 150 mg kg^{-1}. Analyzing the results from "Table 5" and "Figure 3," it becomes evident that in 20.69% of the Olivier salad samples, this concentration exceeds the specified limit.

Under Iranian regulations, dairy products are not permitted to contain any preservatives. The study results reveal that the majority of the analyzed dairy samples were free from preservatives, demonstrating compliance with regulatory requirements. Only 8% of the dairy samples were found to contain preservatives, which does not adhere to the guidelines established by the Institute of Standard and Industrial Research of Iran (ISIRI).

It is worth noting that a minimal amount of sodium benzoate was detected in all dairy products. Previous research has suggested that low levels of sodium benzoate may naturally occur in milk and fermented milk derivatives due to the metabolic

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activity of certain microorganisms acting on hippuric acid [25, 26]. This natural contamination can arise from the metabolic activity of certain microorganisms acting on hippuric acid.

The data in "Table 5" indicates that sodium benzoate and potassium sorbate preservatives were found in the majority of the ketchup samples, either individually or in combination. However, the concentrations of these preservatives did not exceed the acceptable limit set by Iran's standard, which specifies that the total content of sodium benzoate and potassium sorbate should not exceed 750 mg kg^{-1} (ISIRI No. 2454, 2015).

In a separate study conducted on mayonnaise samples in the Iranian market, it was determined that the levels of potassium sorbate and sodium benzoate complied with the acceptable limits specified by the national standard of Iran. This suggests that the analyzed mayonnaise products met regulatory requirements regarding preservative content [27, 28]. However, in another study involving tomato paste samples in Iran, it was observed that in some cases, the concentration of sodium benzoate exceeded the acceptable limits outlined by Iran's legislation. This raises concerns about potential non-compliance with regulatory requirements in those specific samples, emphasizing the need for enhanced monitoring and control in the tomato paste industry to ensure adherence to safety standards [29]. Furthermore, a study carried out on sauce samples in Urmia, Iran, indicated that the levels of preservatives in all the analyzed sauce samples fell within the acceptable limits set by Iran's standard [19]. Additionally, a study on Iranian doogh samples revealed that potassium sorbate was detected in 13% of these samples [30]. Collectively, these studies on doogh, tomato paste, and sauce samples from various regions of Iran shed light on the compliance of these food products with national standards for preservative content. The results underscore the importance of regular monitoring and enforcement of regulations to ensure the safety and quality of food products in the market.

Table 5. Abundance of samples with and without detected sodium benzoate and potassium sorbate and samples containing preservatives higher than MPL[a]

Food	Samples Analyzed (No.)	Samples without preservative (%)	Samples with sodium benzoate (%)	Samples with potassium sorbate (%)	Samples with both preservative (%)	Samples < MPL (%)	Samples > MPL (%)
Olivier salad	29	0	65.52	100	65.52	79.31	20.69
Dairy	50	92	0	8	0	92	8
Ketchup	31	12.91	45.16	80.65	38.71	100	0

[a]Maximum permitted levels.

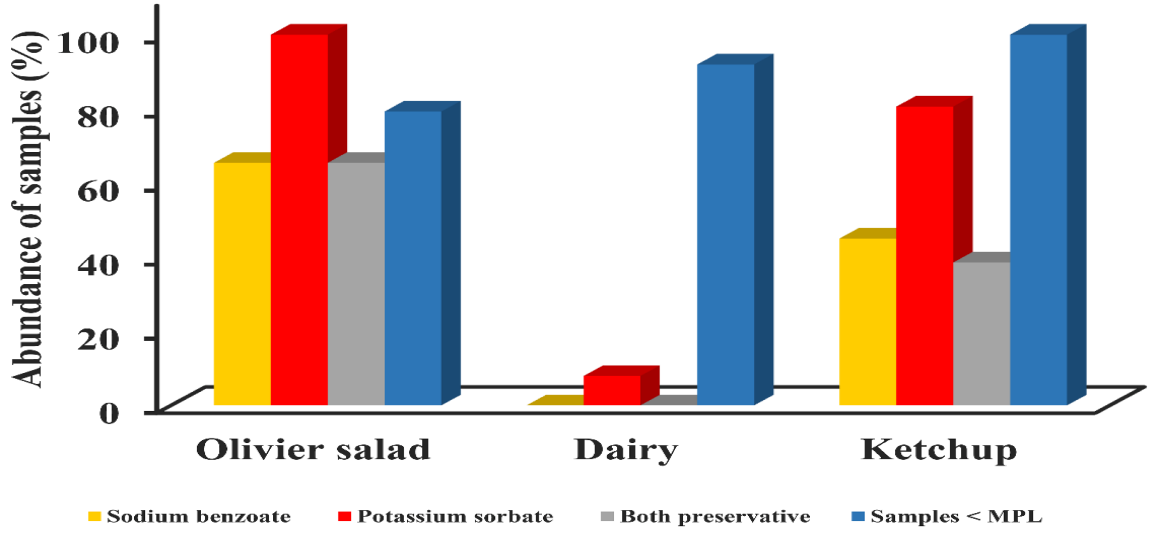

Fig. 3. The comparison between the amount of preservatives in Olivier salad, dairy and ketchup samples.

4.CONCLUSION

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Chemical preservatives play a crucial role in food preservation within the food industry, especially with the increasing production and consumption of ready-to-eat foods. Regulatory bodies set permissible levels for these preservatives in various food types, necessitating precise measurement in food analysis. This study introduces a validated analytical method for quantifying sodium benzoate and potassium sorbate preservatives simultaneously. The analysis was conducted using RP-HPLC, with a solution containing acetate buffer at pH 4.2 and methanol. Detection was performed at a wavelength of 254 nm, utilizing a C18 column for separation. This method enabled the efficient separation of these preservatives within a 12-minute runtime. The method validation followed the guidelines provided by the International Conference on Harmonization (ICH) and yielded outstanding results for validation parameters, including strong linearity (R^2 > 0.999), precision (RSD < 5%), accuracy (recovery ranging from 90.77% to 100.55%), as well as LOD and LOQ (0.8 and 2.65 mg L^{-1} for sodium benzoate and 0.14 and 0.47 mg L^{-1} for potassium sorbate, respectively). It also exhibited low measurement uncertainty, with values of 1.07% for sodium benzoate and 0.50% for potassium sorbate, respectively. This method underwent full validation for the simultaneous determination of preservatives in three sample matrices: Olivier salad, dairy products, and ketchup.

Results indicate the detection rate of potassium sorbate was higher than sodium benzoate in samples, and both preservatives were detected simultaneously in 65.52% of Olivier salad and 38.71% Ketchup. Most samples adhered to permissible limits set by the Iranian Standard and Industrial Research Institute (ISIRI), with only 0.9% exceeding the limit. According to the results, it is recommended that food quality, particularly in commonly consumed food items such as sauces, salads, and dairy products, should be continuously monitored by relevant regulatory bodies. Ensuring that the concentration of preservatives in food stays within approved safety thresholds for human consumption is imperative. This is vital to guarantee that preservative levels in food products fall within the permissible range. The method used in this study has several advantages, including easy, accurate, and sensitive analysis, a simple extraction method, short analysis runtime, and simultaneous separation of both preservatives in a single analytical run, enhancing efficiency and reducing time and resources required for analysis. This method correctly passed all validation parameters.

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Notes

The authors declare no conflict of interest.

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یک روش ساده، حساس و اعتباربخشی شده با HPLC-DAD برای تعیین همزمان محتوای بنزوات سدیم و نگهدارنده‌های سوربات پتاسیم در مواد غذایی فرآوری شده

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چکیده

اندازه گیری میزان مواد نگهدارنده در غذا به دلیل پیامدهای بالقوه آنها برای سلامتی بسیار مهم است .بنابراین، توسعه یک روش تحلیلی قابل اعتماد و معتبر برای پایش ضروری است .این مطالعه بر اعتبار سنجی یک رویکرد تحلیلی ساده، حساس و دقیق با استفاده از HPLC-DAD برای ارزیابی همزمان بنزوات سدیم و سوربات پتاسیم تمرکز دارد .فرآیند اعتبار سنجی منجر به نتایج استثنایی شد که خطی بودن قوی($R^2 > 0.999$) ، دقت($RSD < 5$) ٪، و صحت (مقادیر بازیابی از ۹۰.۷۷٪ تا ۱۰۰.۵۵٪) را نشان داد .علاوه بر این، مقادیرحد تشخیص و حد اندازه گیری این روش پائین می باشد (به ترتیب ۰.۸ و ۲.۶۵ میلی‌گرم در لیتر برای بنزوات سدیم، و ۰.۱۴ و ۰.۴۷ میلی‌گرم در لیتر برای سوربات پتاسیم) .اثربخشی روش اعتبار بخشی شده برروی ماتریس های مختلف غذایی با تجزیه و تحلیل ۱۱۰ نمونه سالاد اولویه، محصولات لبنی و سس کچاپ از مشهد، ایران نشان داده شد. مقایسه نتایج با استانداردهای ملی ایران نشان داد که غلظت مواد نگهدارنده در اکثر نمونه ها در محدوده قابل قبول تعیین شده توسط مقررات ایران است.

کلید واژه ها

HPLC-DAD؛ مواد نگهدارنده؛ اعتبار بخشی روش؛ مواد غذایی فرآوری شده.

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