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Determination of Lead (Pb) Content in Commercial Fruit Juice Drinks Using Atomic Absorption Spectroscopy with Standard Addition Method

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ABSTRACT

Lead (Pb) is one of the most hazardous heavy metals that may contaminate fruit juice through environmental pollution, raw materials, processing, and packaging. Continuous exposure to Pb poses significant health risks due to its cumulative toxicity. This study aimed to determine the Pb concentration in commercially packaged fruit juice using Atomic Absorption Spectroscopy (AAS). Samples were prepared by wet digestion using nitric acid, followed by Pb determination employing the standard addition method to minimize matrix interference. The analytical method was evaluated through linearity, limit of detection (LOD), limit of quantification (LOQ), precision, and accuracy. The calibration curve exhibited excellent linearity within the investigated concentration range. The Pb concentration in the analyzed sample was found to be 0.15 mg kg^{-1}, which is below the maximum permissible limit established by the Indonesian National Standard (SNI 3719:2014). The method showed satisfactory analytical performance with an LOD of 0.1773 mg L^{-1}, an LOQ of 0.5910 mg L^{-1}, a relative percent difference (RPD) of 2.33%, and a recovery of 105.73%. These results demonstrate that the standard-addition AAS method provides a reliable quantitative determination of Pb in fruit juice matrices.

Keywords: Lead (Pb); Atomic Absorption Spectroscopy; Fruit Juice; Heavy Metal; Standard Addition

1. INTRODUCTION

Food safety has become a major global concern because foods and beverages may serve as important pathways for human exposure to toxic heavy metals. Among these contaminants, lead (Pb) is considered one of the most hazardous due to its persistence, non-biodegradable nature, and tendency to accumulate in biological tissues. Heavy metals, particularly Pb, Cd, and As, have received increasing attention because of their widespread occurrence in food products and their adverse impacts on human health.[1–3] Chronic exposure to Pb may adversely affect neurological development, renal function, hematopoietic processes, and cardiovascular health, while prolonged dietary intake may result in bioaccumulation and increased health risks,

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especially among children and pregnant women.[4,5] Consequently, international organizations and national regulatory agencies have established strict limits for Pb contamination in food products to protect public health.[6,7]

Commercial fruit juice is widely consumed because of its nutritional value, convenience, and pleasant sensory characteristics. Nevertheless, several studies have reported detectable concentrations of heavy metals, including Pb, in commercial fruit juices originating from agricultural practices, contaminated irrigation water, industrial emissions, processing equipment, and packaging materials.[8,9] A recent global meta-analysis involving fruit juices from multiple countries demonstrated that Pb remains one of the most frequently detected toxic elements, although the concentrations generally comply with national regulations.[10] Recent investigations have also confirmed that fruit juices marketed in different countries may contain varying concentrations of Pb depending on fruit origin, environmental pollution, processing technology, and packaging materials.[11] Despite these findings, continuous monitoring remains essential because contamination levels are strongly influenced by environmental conditions, manufacturing processes, and storage systems.[6,12,13]

Atomic Absorption Spectroscopy (AAS) remains one of the most reliable analytical techniques for determining trace concentrations of heavy metals because of its high sensitivity, selectivity, relatively simple operation, and cost-effectiveness.[14–16] In routine quantitative analysis, external calibration is commonly applied to establish the relationship between analyte concentration and instrument response. However, fruit juice represents a chemically complex matrix containing sugars, organic acids, pigments, and various dissolved compounds that may interfere with atomization efficiency and analytical response.[17] Such matrix effects may reduce the accuracy of conventional external calibration methods, particularly when trace metals are present at low concentrations. Therefore, analytical validation is essential to ensure reliable quantitative determination of Pb in complex food matrices.[18,19]

To overcome matrix interference, the standard addition method has been widely recommended because calibration is performed directly within the sample matrix. This approach effectively compensates for signal suppression or enhancement caused by sample composition, resulting in more accurate quantification of trace metals.[20,21] Recent studies have demonstrated that the standard addition technique provides improved analytical performance for Pb determination in complex food and beverage matrices compared with conventional external calibration, particularly when significant matrix effects are present.[22–24] In addition, analytical method validation involving linearity, limit of detection (LOD), limit of quantification (LOQ), precision, and recovery has become an essential requirement to ensure the reliability of quantitative measurements in food safety analysis.

In Indonesia, the maximum allowable Pb concentration in commercial fruit juice is regulated by the Indonesian National Standard (SNI 3719:2014), which specifies a maximum limit of 0.20 mg kg^{-1}. Although numerous studies have investigated heavy metal contamination in beverages, relatively few studies have comprehensively evaluated both the analytical performance of the AAS method, including linearity, limit of detection (LOD), limit of quantification (LOQ), precision, and accuracy, and the application of the standard addition approach for Pb determination in commercially available fruit juice products.[25–27] Therefore, this study aimed to determine Pb concentration in packaged fruit juice using AAS coupled with the standard addition method and to evaluate the analytical performance of the developed method. The obtained Pb concentration was further compared with the maximum permissible limit established by SNI 3719:2014 to assess the safety of the analyzed product and to support continuous monitoring of heavy metal contamination in processed beverages.

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Unlike previous studies that primarily focused on determining Pb concentration using external calibration, this study integrates comprehensive analytical method validation with the standard addition approach to minimize matrix interference in commercial fruit juice samples. This work provides more reliable quantitative data and contributes to the application of validated AAS methods for food safety monitoring in accordance with Indonesian quality standards. Furthermore, the study supports current efforts to strengthen food safety surveillance through the application of validated analytical methods for heavy metal determination in commercial beverages.

2. EXPERIMENTAL

2.1. Chemicals, Equipment and Instrumentation

Analytical-grade nitric acid (HNO_3, 65%, Merck, Darmstadt, Germany), hydrochloric acid (HCl, 37%, Merck), lead standard solution (Pb, 1000 mg L^{-1}, Merck Certipur®), and deionized water were used throughout this study without further purification. Commercial packaged fruit juice samples were purchased from a local supermarket in Yogyakarta, Indonesia.

Lead determination was performed using an Atomic Absorption Spectrophotometer (AAS, PerkinElmer AAnalyst 300) equipped with a hollow cathode lamp for Pb analysis. Sample preparation employed an analytical balance (Ohaus PA224), a programmable muffle furnace, hot plate, porcelain crucibles, volumetric flasks, volumetric pipettes, micropipettes, beakers, desiccator, and other laboratory glassware.

2.2. Sample Collection

Commercial packaged fruit juice samples were randomly purchased from a local retail market in Yogyakarta, Indonesia. The products were transported to the laboratory in their original packages and analyzed before the expiration date. Before analysis, the samples were homogenized by gentle shaking to ensure uniform distribution of the liquid matrix.

2.3. Sample Digestion

A 10 mL aliquot of fruit juice was transferred into a porcelain crucible and evaporated on a hot plate until no visible fumes remained. The residue was subsequently ashed in a muffle furnace at 450 ± 5°C until white ash was obtained. If gray ash indicating residual carbon was observed, approximately 2.5 mL of concentrated nitric acid was added dropwise, followed by further heating on the hot plate and reheating in the furnace until complete mineralization was achieved.

The resulting ash was dissolved in 2.5 mL of 6 N hydrochloric acid, followed by the addition of 5 mL of 0.1 N nitric acid. The solution was quantitatively transferred into a 25 mL volumetric flask and diluted to volume with deionized water.

2.4. Preparation of Standard Solutions

A working standard solution of 50 mg L^{-1} Pb was prepared by diluting the stock standard solution with deionized water. Calibration standards containing 0.00, 0.10, 0.25, 0.50, 1.00, 1.50, and 2.00 mg L^{-1} Pb were prepared by appropriate dilution of the working standard solution using 25 mL volumetric flasks.

2.5. Standard Addition Procedure

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The standard addition method was applied to minimize matrix interference during Pb determination. Aliquots of the prepared sample solution (1 mL) were transferred into a series of 25 mL volumetric flasks, followed by the addition of Pb standard solution to obtain final concentrations of 0.00, 0.10, 0.25, 0.50, 1.00, 1.50, and 2.00 mg L^{-1}. Each flask was diluted to volume with deionized water and mixed thoroughly before instrumental analysis.

2.6. Instrumental Analysis

Lead concentration was determined using Atomic Absorption Spectrophotometry at a wavelength of 283.3 nm under the manufacturer's recommended operating conditions. Calibration curves were established by plotting absorbance against Pb concentration. Sample concentrations were calculated from the regression equation obtained by the standard addition method. The Pb concentration in the fruit juice sample was calculated using Equation (1):

$$\text{Pb concentration (mg kg}^{-1}) = \frac{C \times V}{W}$$

where:
C = Pb concentration obtained from the calibration curve (mg L^{-1})
V = final volume of the digested solution (mL)
W = sample weight (g)

2.7. Method Validation

The analytical performance of the method was evaluated by determining linearity, limit of detection (LOD), limit of quantification (LOQ), precision, and accuracy. Linearity was assessed from the coefficient of determination (R^2) of the calibration curve. LOD and LOQ were calculated based on the standard deviation of the regression and the slope of the calibration curve according to ICH guidelines. Precision was evaluated as the Relative Percent Difference (RPD) between duplicate measurements, while accuracy was assessed using the standard addition recovery method.

3. RESULTS AND DISCUSSION

3.1. Calibration Curve and Linearity

The calibration curve for Pb determination was established using seven standard solutions with concentrations of 0.00, 0.10, 0.25, 0.50, 1.00, 1.50, and 2.00 mg L^{-1}. The absorbance increased proportionally with increasing Pb concentration, indicating that the Atomic Absorption Spectrophotometer (AAS) exhibited a stable analytical response throughout the investigated concentration range. Linear regression analysis produced the calibration equation $y = 0.2692x + 0.0181$ with a coefficient of determination (R^2) of 0.995.

The high coefficient of determination demonstrates a strong linear relationship between Pb concentration and absorbance, indicating that the analytical response obeyed the Beer–Lambert law within the selected concentration range. According to the ICH Q2(R2) guideline, an R^2 value of 0.995 or higher is generally considered acceptable for quantitative analytical methods, confirming that the proposed calibration model is suitable for Pb determination.[28] In addition, a highly linear calibration curve minimizes systematic error during

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concentration prediction and improves the reliability of quantitative analysis, particularly for trace metal determination in food samples.[29,30]

The linearity obtained in this study is comparable with recent studies reporting R^2 values greater than 0.995 for Pb determination by flame AAS in fruit juices and beverage samples, demonstrating that AAS remains a robust analytical technique for routine heavy metal monitoring. Similar findings were also reported by Patriani et al. (2023)[31] in Indonesia, who demonstrated excellent linearity for Pb analysis in canned beverages using the standard addition approach, highlighting the applicability of AAS for complex beverage matrices. The excellent linearity obtained in this study therefore confirms that the developed analytical method is suitable for quantitative determination of Pb in commercial fruit juice.

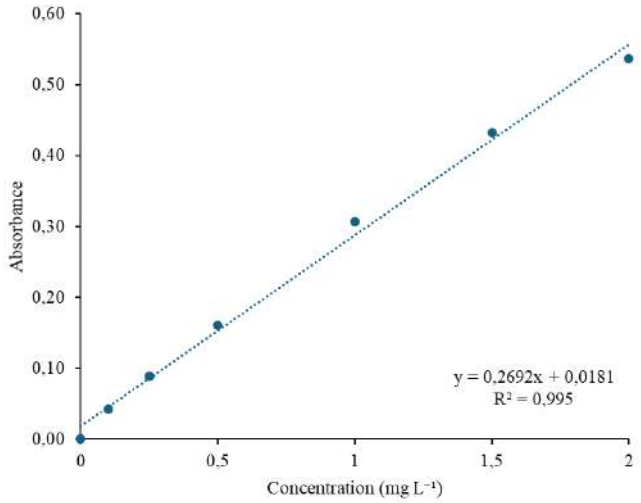

Figure 1. Calibration curve of Pb standard solutions

3.2. Standard Addition Analysis

Commercial fruit juice contains various organic constituents, including sugars, organic acids, vitamins, pigments, and phenolic compounds, which may interfere with atomization efficiency during AAS measurement. Such matrix components can suppress or enhance the analytical signal, resulting in inaccurate quantification when conventional external calibration is applied.[32] Consequently, the standard addition method was selected to compensate for matrix effects and improve analytical accuracy.

As presented in Table 1, the absorbance increased consistently with increasing concentrations of the added Pb standard solution, indicating that the analytical response remained linear even in the presence of the fruit juice matrix. The proportional increase in absorbance demonstrates that the added Pb ions were successfully atomized without significant signal distortion, confirming the suitability of the standard addition method for complex beverage samples.

Table 1. Absorbance standard addition method

Concentration (mg L^{-1})	Volume of sample added (mL)	Absorbance			Average
		1	2	3	
0	1	0.0070	0.0040	0.0060	0.0057

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0.1	0.0380	0.0410	0.0390	0.0393
0.25	0.0890	0.0900	0.0880	0.0890
0.5	0.1650	0.1640	0.1740	0.1677
1	0.3070	0.3060	0.3100	0.3077
1.5	0.4340	0.4370	0.4460	0.4390
2	0.5620	0.5590	0.5550	0.5587

Unlike external calibration, the standard addition technique performs calibration directly within the sample matrix, thereby correcting signal suppression or enhancement caused by dissolved organic compounds and other interfering substances. This approach has been widely recommended for trace metal determination in beverages, fruit products, and other food matrices because it provides more accurate quantitative results, particularly when matrix effects cannot be eliminated during sample preparation.[33,34]

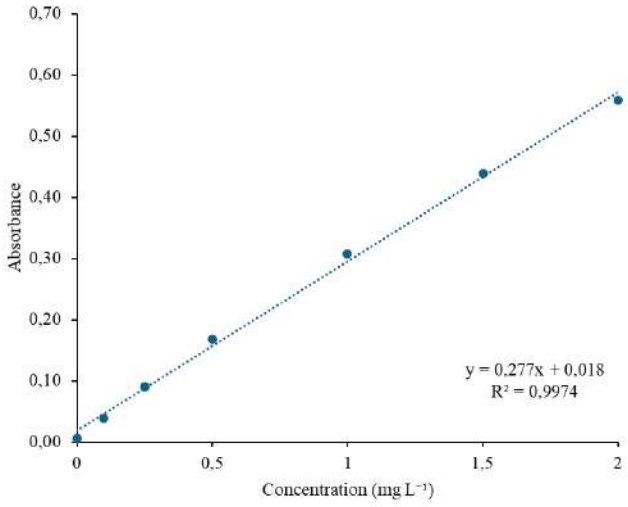

Figure 2. Standard addition calibration plot

The analytical behavior observed in this study agrees with previous investigations reporting that the standard addition method significantly improves the accuracy and reliability of Pb determination in fruit juices, canned beverages, dairy products, and other complex food matrices.[35] Furthermore, recent studies have shown that combining wet digestion with standard addition and flame AAS effectively minimizes analytical bias while maintaining satisfactory precision and recovery, making this approach appropriate for routine food safety monitoring.

The linear increase in absorbance shown in Table 1 also indicates that no significant chemical interference occurred after sample digestion, suggesting that the digestion procedure effectively decomposed the organic matrix before instrumental analysis. Consequently, the developed method can be considered reliable for Pb determination in commercial fruit juice and suitable for application in food quality control laboratories.

3.3. Lead Concentration in Commercial Fruit Juice

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The Pb concentration determined by the standard addition method was 0.15 mg kg^{-1}. This result indicates that Pb was present in the analyzed commercial fruit juice sample. The presence of Pb in fruit beverages may originate from contaminated agricultural soils, irrigation water, atmospheric deposition, industrial emissions, food processing equipment, or packaging materials. Previous studies have reported that Pb contamination in fruit juice is generally associated with environmental pollution during cultivation rather than intentional contamination during manufacturing.

Although Pb was detected, the measured concentration remained below the maximum permissible limit established by the Indonesian National Standard (SNI 3719:2014), which specifies a maximum concentration of 0.20 mg kg^{-1} for fruit juice beverages. The Pb concentration obtained in this study was also comparable with values reported in commercial fruit juices from several countries, where Pb concentrations generally ranged between non-detectable levels and approximately 0.18 mg kg^{-1} (Table 2).

Table 2. Comparison of Pb concentration in commercial fruit juices reported in recent studies

Study	Country	Sample	Method	Pb concentration (mg kg^{-1})
This Study	Indonesia	Commercial fruit juice	FAAS (Standard Addition)	0.15
Patriani et al. (2023)[31]	Indonesia	Canned beverage	AAS	0.09-0.17
Borges et al. (2023)[27]	Brazil	Fruit juice	FAAS	ND-0.18
Paudel et al. (2024)[36]	Nepal	Fruit juice	FAAS	ND
Amheisen et al. (2025)[37]	Libya	Fruit juice	AAS	ND

Table 2 shows that the Pb concentration obtained in this study (0.15 mg kg^{-1}) is comparable to those reported in recent studies on commercial fruit juices. Although variations in Pb concentration have been observed among different countries, most studies reported Pb levels below the maximum regulatory limits. These differences are primarily associated with geographical origin, agricultural practices, environmental pollution, fruit species, manufacturing processes, and analytical methods employed. Similar observations have been reported in recent studies, indicating that environmental contamination during cultivation remains the dominant source of Pb in fruit-based beverages, while contamination from processing and packaging generally contributes to a lesser extent.

3.4. Method Validation

Method validation was performed to evaluate the analytical performance of the proposed Atomic Absorption Spectroscopy (AAS) method for Pb determination in commercial fruit juice. The validation parameters included sensitivity, expressed as the limit of detection (LOD) and limit of quantification (LOQ), as well as precision and accuracy. These parameters were assessed following internationally accepted analytical validation guidelines to ensure the reliability of the proposed method for routine analysis.

3.4.1. Sensitivity (LOD and LOQ)

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The sensitivity of the analytical method was evaluated by determining the limit of detection (LOD) and limit of quantification (LOQ). The calculated LOD and LOQ values were 0.1773 mg L^{-1} and 0.5910 mg L^{-1}, respectively (Table 3).

Table 3. LOD & LOQ Observation Data of Samples

Concentration (mg L^{-1})	Average absorbance	y_1	$y-y_1$	$(y-y_1)^2$
0	0.0000	0.0181	-0.0181	0.0003
0.1	0.0427	0.0450	-0.0024	0.0000
0.25	0.0883	0.0854	0.0029	0.0000
0.5	0.1600	0.1527	0.0073	0.0001
1	0.3070	0.2873	0.0197	0.0004
1.5	0.4320	0.4219	0.0101	0.0001
2	0.5370	0.5565	-0.0195	0.0004
			SD	0.0159
			LOD	0.1773
			LOQ	0.5910

The obtained values indicate that the developed method possesses sufficient analytical sensitivity for the determination of Pb in commercial fruit juice samples. The measured Pb concentration (0.15 mg kg^{-1}) was above the instrumental detection capability, demonstrating that Pb could be detected and quantified reliably under the experimental conditions.

The analytical sensitivity obtained in this study is comparable to previous studies employing flame AAS for Pb determination in food and beverage matrices, indicating that the proposed analytical procedure is suitable for routine monitoring of heavy metal contamination. Moreover, adequate LOD and LOQ values are essential for ensuring compliance with food safety regulations, particularly when trace concentrations of toxic metals are determined.

3.4.2. Precision

Method precision was evaluated by duplicate measurements and expressed as the Relative Percent Difference (RPD). The calculated RPD was 2.33% (Table 4). The low RPD value indicates excellent repeatability of the analytical procedure. According to commonly accepted analytical criteria, RPD values below 5% demonstrate good precision for duplicate analyses of trace metals in food samples. The small variation observed between replicate measurements confirms that the proposed analytical method provides consistent and reproducible results. Good analytical precision is particularly important for routine food quality control because it ensures that the measured Pb concentration is not significantly affected by random analytical errors.

Table 4. Sample Precision Observation Data

Sample	Average absorbance	Pb Concentration (mg L^{-1})	%RPD
1	0.0037	-0.0537	2.33
2	0.0040	-0.0525	

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3.4.3. Accuracy

Method accuracy was assessed using the standard addition recovery approach. The recovery obtained was 104.29% (Table 5), indicating satisfactory analytical accuracy. Recovery values within the range of 80–120% are generally considered acceptable for trace metal analysis in food matrices according to internationally recognized analytical validation guidelines. The satisfactory recovery confirms that matrix interference was effectively compensated by the standard addition method, allowing accurate quantification of Pb in the analyzed fruit juice sample.

Table 5. Sample Accuracy Observation Data

Solution	Concentration (mg L^{-1})	Theoretical Concentration (mg L^{-1})	%R
Sample	-0.0531	0.1	105.73
Spike solution	0.0526		

Overall, the satisfactory values of LOD, LOQ, precision, and accuracy confirm that the developed AAS method is valid and suitable for routine determination of Pb in commercial fruit juice, supporting its application in food safety monitoring and quality control programs.

4. CONCLUSION

Lead (Pb) in commercial fruit juice was successfully determined using Atomic Absorption Spectroscopy coupled with the standard addition method. The analytical method exhibited satisfactory performance, as demonstrated by good linearity, acceptable sensitivity (LOD = 0.1773 mg L^{-1}; LOQ = 0.5910 mg L^{-1}), excellent precision (RPD = 2.33%), and satisfactory recovery (105.73%). The Pb concentration in the analyzed sample was 0.15 mg kg^{-1}, which is below the maximum permissible limit specified by the Indonesian National Standard (SNI 3719:2014). These findings indicate that the proposed method is suitable for routine Pb determination in fruit juice and can support food safety monitoring programs.

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