



ISSN: 0067-2904

## Intensive and Explanatory Statistical Analysis of the Relationship Between Inorganic Phosphate Content and Antioxidant Activity in Fresh and Canned Fruit Juices

Fadheela Al-Salman, Ali Ali Redha\*, Zahra Al-Zaimoor

Department of Chemistry, College of Science, University of Bahrain, Sakhir, Kingdom of Bahrain

Received: 7/3/2021

Accepted: 13/6/2021

Published: 30/5/2022

### Abstract

The inorganic phosphate content and antioxidant activity of fresh fruit juices and canned fruit juices commonly consumed in Bahrain were compared. The fruits considered in this study were kiwi, guava, black grape, strawberry, apple, and pineapple. The inorganic phosphate content of the juices was determined by a colorimetric method using a UV/VIS spectrophotometer. Among the fresh juices, the highest inorganic phosphate content was measured for black grape juice ( $17.330 \pm 0.068$  mg/L), and among the canned juices, the highest inorganic phosphate was measured for black grape canned juice too ( $16.020 \pm 0.141$  mg/L, brand 3). The antioxidant activity was determined in-vitro by measuring the percentage of 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity of the juices. Among the fresh juices, the highest antioxidant activity was measured for fresh guava juice ( $95.98 \pm 0.15\%$ ). Among the canned juices, the highest antioxidant was measured for black grape canned juice (two brands:  $88.69 \pm 0.05$  and  $90.49 \pm 0.12\%$ ). According to the inferential statistical analysis (normality assumption tests and one-way ANOVA), no statistically significant correlation was found between inorganic phosphate content and antioxidant activity (Pearson correlation coefficient = 0.279, p-value = 0.263) at a 5% significance level.

**Keywords:** inorganic phosphate content; antioxidant activity; fresh fruit juices; canned fruit juices

### تحليل إحصائي مكثف وتوضيحي لبيانات محتوى الفوسفات غير العضوي والنشاط المضاد للأوكسدة لعصائر الفاكهة الطازجة والمعلبة

فضيلة السلطان, علي علي رضا\*, زهرة الزيمور

قسم الكيمياء، كلية العلوم، جامعة البحرين، الصخير، مملكة البحرين

### الخلاصة:

تمت مقارنة محتوى الفوسفات غير العضوي والنشاط المضاد للأوكسدة لعصائر الفاكهة الطازجة وعصائر الفاكهة المعلبة التي تستهلك في البحرين. كانت الثمار التي تم بحثها في هذه الدراسة هي الكيوي والجوافة والعنب الأسود والفرولة والتفاح والأناناس. تم تحديد محتوى الفوسفات غير العضوي للعصائر بطريقة القياس اللوني باستخدام مقياس الطيف الضوئي UV / VIS. من بين العصائر الطازجة، تم قياس أعلى محتوى

\*Email: ali96chem@gmail.com

فوسفات غير عضوي لعصير العنب الأسود ( $0.068 \pm 17.330$  مجم / لتر) ، ومن بين العصائر المعلبة ، تم قياس أعلى نسبة فوسفات غير عضوية لعصير العنب الأسود المعلب أيضًا ( $0.141 \pm 16.020$  مجم / لتر) ، العلامة التجارية 3). تم تحديد نشاط مضادات الأكسدة في المختبر عن طريق قياس نسبة 2،2-ثنائي فينيل 1-بيكريل هيدرازيل (DPPH) نشاط الكسح الجذري للعصائر. من بين العصائر الطازجة، كان أعلى نشاط مضاد للأكسدة هو عصير الجوافة الطازج ( $0.15 \pm 95.98$ %، ومن بين العصائر المعلبة ، تم قياس أعلى مضادات الأكسدة لعصير العنب الأسود المعلب (علامتان تجاريتان:  $0.05 \pm 88.69$  ، و  $0.12 \pm 90.49$ %). وفقًا للتحليل الإحصائي الاستنتاجي (اختبارات الافتراض الطبيعي و ANOVA أحادي الاتجاه) ، لم يتم العثور على ارتباط ذي دلالة إحصائية بين محتوى الفوسفات غير العضوي والنشاط المضاد للأكسدة (معامل ارتباط بيرسون =  $0.279$  ، قيمة  $p = 0.263$  ) عند مستوى أهمية 5%.

## 1. Introduction

Fruits are a valuable source of many vital metabolites, vitamins, minerals, and essential elements for humans. One of the favorite ways of fruit consumption by humans is to drink fruit juices. Fruit juices are commonly available in the market as fresh juices or canned juices, and the nutritional value of those is different and ultimately will have a different impact on human health and diet.

Fruits are a natural source of inorganic phosphate and antioxidants. Phosphate is considered as an essential electrolyte and component in the cells of living organisms as it plays a vital role in the synthesis of ATP and DNA [1]. In addition, it also contributes to controlling the pH of the blood and lymph fluid [1]. Yet, excess phosphate in the body can cause disturbed metabolism of minerals, vascular calcification, bone loss, and kidney disorders [2]. Thus, determining the phosphate in food sources is essential to ensure that the right amount of phosphate is consumed. The recommended daily phosphorus intake from food products (average) is 1596 mg/day for men and 1189 mg/day for women [3].

Furthermore, antioxidants also have a significant role in the body as they neutralize the excess of free radicals protecting the cells against the toxic effects of those radicals and preventing diseases such as cancer, cardiovascular disease, and diabetes [4,5]. Antioxidants can inhibit lipid peroxidation, prevent oxidative damage, and act as anti-inflammatory and anti-ageing agents [5]. Compounds such as phenolic acids, flavonoids, isoflavones, carotenoids, and vitamins are known for their antioxidant potential [6]. Fruits such as pomegranate, blueberries, strawberries, raspberries, and blackberries have been reported high antioxidant activity [7–9].

This research compared the inorganic phosphate content and the antioxidant activity of fresh and canned fruit juices. It was previously reported that there is a strong relationship between the concentration of macronutrients, including phosphorus, in the soil and the levels of phenolic and flavonoid compounds and antioxidant activity of tomato and Kacip Fatimah (*Labisia pumila* Benth) [10,11]. Thus, descriptive (mean computation, standard deviation, minimum and maximum values, and box plot) and inferential (normality assumption tests and one-way ANOVA) statistical analyses were conducted to explore the correlation between inorganic phosphate content and the antioxidant activity. The study of this relationship can clarify the relationship in the importance of inorganic phosphate and the presence of antioxidant compounds, such as phenolics (including flavonoids), in different fruit juices. This study considered two types of juices: fresh and canned fruit juices for analysis. It has been reported that the type of juice (fresh or processed) can affect the juice's phenolic content and antioxidant activity. The conclusions of this study were based on intensive and explanatory statistical analysis as it is essential to conduct and apply statistical analysis in such research as it will support obtaining meaningful interpretations and understanding the significance of the findings. The findings and inferences of such a study will only be precise if appropriate statistical tests are conducted [13].

## 2. Materials and methods

### 2.1. Materials

Ammonium molybdate, copper sulphate, sodium acetate, acetic acid, sodium hydroxide pellets, *p*-methyl aminophenol sulphate, sodium sulphite, trichloroacetic acid, potassium dihydrogen phosphate and 2,2-diphenyl-1-picrylhydrazyl (DPPH) were purchased from Sigma-Aldrich, United States.

### 2.2. Methods

#### 2.2.1. Preparation of Samples

Fresh kiwi, guava, black grapes, strawberry, apple, and pineapple were purchased from the local market and washed with water. Pure fresh juice of each fruit was prepared by blending the fruit using a commercial blender with the addition of 50 mL distilled water (the fruit skin was peeled/discarded before blending, with an exception for strawberry and black grapes). The fruit juices were used immediately after preparation. Canned fruit juices of the same fruits were bought, two samples of popular brands were selected, and they were used before their expiration date.

#### 2.2.2. Estimation of inorganic phosphate content

The inorganic phosphate content was determined according to Al-Salman et al. [14]. A volume of 10 mL of each fruit juice was mixed with 10 mL of 20 g/L trichloroacetic acid and centrifuged at 500 rpm ( $G\text{-force} = 23.8 \times g$ ) for 5 min using a UNIVERSAL 320 centrifuge. A volume of 1.0 mL of each supernatant was added to 3 mL of copper acetate, 0.5 mL of 50 g/L ammonium molybdate and 0.5 mL of reducing agent. The mixture was incubated for 10 minutes at room temperature, and then the sample absorbance was measured at 880 nm using ORION AQUAMATE 8000 UV/VIS spectrophotometer. The inorganic phosphate content was calculated using the calibration curve equation of standard inorganic phosphate solutions (treated like test samples). The blank used for the analysis contained all the reagents without inorganic phosphate.

#### 2.2.3. Determination of antioxidant activity

A 50  $\mu\text{L}$  of each fruit juice sample was added to 2.95 mL of fresh DPPH solution (prepared by dissolving 4.5 mg of DPPH in 100 mL of methanol). The mixture was incubated for 30 minutes at room temperature, and then the sample absorbance was measured at 517 nm using ORION AQUAMATE 8000 UV/VIS spectrophotometer [15,16]. Every sample was tested as triplicates. The antioxidant potential was determined by calculating the percentage potential of scavenging DPPH radical according to the following equation:

$$\% \text{ Antioxidant activity} = \left( \frac{\text{Absorbance of fresh DPPH solution} - \text{Absorbance of test sample}}{\text{Absorbance of fresh DPPH solution}} \right) \times 100\%$$

#### 2.2.4. Statistical analysis

Statistical analysis includes descriptive statistics (central tendency and variability and outliers check), normality assumption testing (by fruit and type), inferential statistics (one-way ANOVA for fruit effect testing and Kruskal-Wallis for type effect test), and correlational analysis (all samples, by fruit, and by type) were performed.

The data collected were organized, appropriately coded, and imported into IBM SPSS version 26 for analysis and the statistical analysis was performed on two levels: descriptive level and inferential level.

The methods used as part of the descriptive statistical analysis were mean computation, standard deviation, minimum and maximum values, and box plot. The inferential statistical analysis method includes:

1. Normality assumption test using Kolmogorov-Smirnov and Shapiro-Wilk normality tests to assess whether the studied data follows a normal distribution which is an essential

assumption when performing parametric tests such as the t test or one-way ANOVA.

2. One-way ANOVA, a parametric test that assumes normality, to statistically test whether there is a mean value difference among the studied variables due to the fruit effect or type. The pairwise mean comparison was tested using Scheffe method for any statistically significant mean difference found. The equivalent nonparametric test for one-way ANOVA is Kruskal-Wallis. It was used if the normality assumption was not verified in the studied variables.

Correlation analysis was performed to assess the direction (positive or negative) and the strength (poor, moderate or strong) linear association between two quantitative variables. The parametric correlation test when the data was found normally distributed is the Pearson method, while the Spearman method was used when the data were not following a normal distribution.

### 3. Results and discussion

#### 3.1. Inorganic phosphate content

The inorganic phosphate content of the fresh juices and canned juices were determined. In the analysis of fresh fruit juices, the highest inorganic content was found in black grape, followed by strawberry, apple, kiwi, pineapple, and guava as shown in Table 1. The inorganic content of canned juices ranged between 0.23-16.02 mg/L. High inorganic phosphate content was determined in black grape canned juice (brand 3, 16.020  $\pm$  0.141 mg/L), pineapple juice (brand 1, 7.823  $\pm$  0.025 mg/L), and strawberry juice (brand 1, 7.720  $\pm$  0.100 mg/L); while the other samples ranged between 0.23-1.63 mg/L. Overall, the inorganic phosphate content in fresh juices is higher than the canned juices. Statistical analysis was conducted to evaluate the significance of these differences (Section 3.3.).

**Table 1** – Inorganic phosphate content of fresh and canned fruit juices. The values are reported as triplicate mean  $\pm$  standard deviation.

Fruit	Inorganic phosphate content (mg/L)		
	Fresh juice	Canned juice, brand 1	Canned juice, brand 2
Kiwi	8.270 $\pm$ 0.095	1.626 $\pm$ 0.002	0.452 $\pm$ 0.006
Guava	3.777 $\pm$ 0.070	1.565 $\pm$ 0.037	0.414 $\pm$ 0.002
Black grape	17.330 $\pm$ 0.068	1.384 $\pm$ 0.036	16.020 $\pm$ 0.141 <sup>a</sup>
Strawberry	10.753 $\pm$ 0.012	7.720 $\pm$ 0.100	0.742 $\pm$ 0.005 <sup>b</sup>
Apple	9.712 $\pm$ 0.040	1.045 $\pm$ 0.004	1.721 $\pm$ 0.013
Pineapple	8.266 $\pm$ 0.067	7.823 $\pm$ 0.025	0.229 $\pm$ 0.013

<sup>a</sup>brand 3, <sup>b</sup>brand 4

#### 3.2. Antioxidant activity

The antioxidant activity of the fruit juices was measured in-vitro based on the scavenging of DPPH radical, and the activity varies among the samples. Guava juice showed the highest antioxidant activity among the fresh fruit juices, followed by black grape, strawberry, pineapple, kiwi, and apple juices, as shown in Table 2. The antioxidant activity of canned juices showed a significant variation, with the lowest activity in kiwi juice (brand 1, 26.50  $\pm$  0.69%) and the highest in black grape juice (brand 4, 90.49  $\pm$  0.12%). Overall, among the canned juices, kiwi juices had the lowest activity (brand 1, 26.50  $\pm$  0.69%; brand 2, 29.00  $\pm$  0.88%), and black grape juices had the highest activity (brand 1, 88.69  $\pm$  0.05%; brand 2, 90.49  $\pm$  0.12%). Statistical analysis was conducted to evaluate the significance of these differences (Section 3.3.).

**Table 2** – Antioxidant activity of fresh and canned fruit juices. The values are reported as triplicates mean  $\pm$  standard deviation.

Fruit	Antioxidant activity (%)		
	Fresh juice	Canned juice, brand 1	Canned juice, brand 2
Guava	95.98 $\pm$ 0.15	74.91 $\pm$ 1.61	82.60 $\pm$ 1.37
Kiwi	44.44 $\pm$ 1.97	26.50 $\pm$ 0.69	29.00 $\pm$ 0.88
Pineapple	55.28 $\pm$ 0.93	36.54 $\pm$ 1.32	34.70 $\pm$ 1.35
Strawberry	65.69 $\pm$ 1.61	49.44 $\pm$ 1.23	34.88 $\pm$ 0.41 <sup>a</sup>
Black grape	74.32 $\pm$ 0.39	88.69 $\pm$ 0.05	90.49 $\pm$ 0.12 <sup>b</sup>
Apple	40.25 $\pm$ 2.08	45.95 $\pm$ 0.27	49.85 $\pm$ 0.37

<sup>a</sup>brand 3, <sup>b</sup>brand 4

### 3.3. Statistical analysis

#### 3.3.1. Descriptive statistics

##### 3.3.1.1. Central tendency

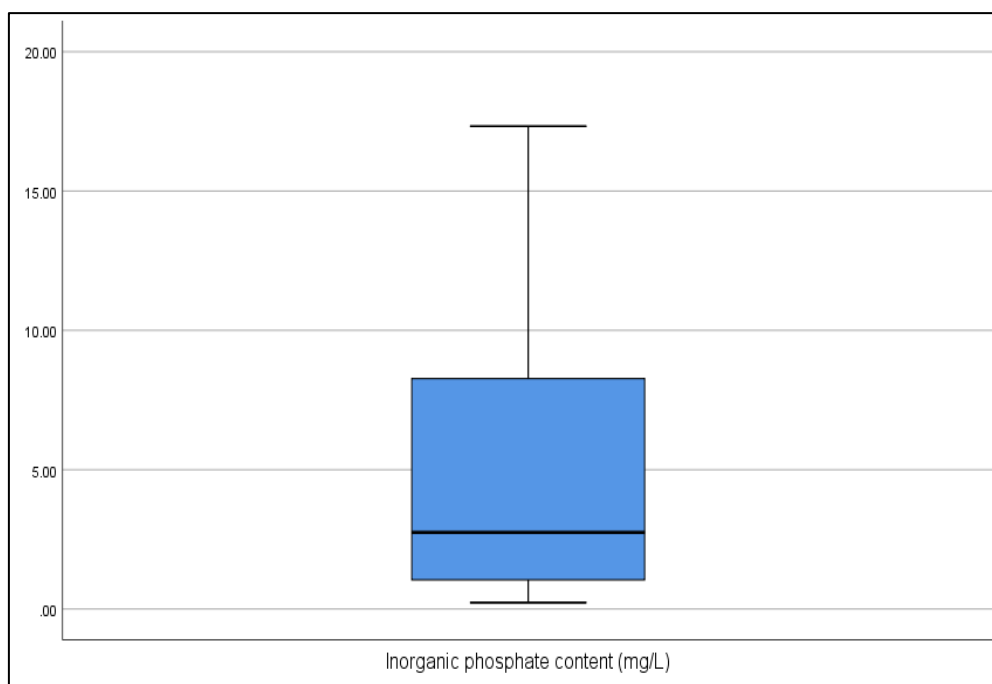
The summary statistics, including central tendency measures: mean and median, and dispersion measures: standard deviation, minimum and maximum are shown in Table 3. The mean value of the total sample observation collected for the inorganic phosphate content was 5.49, lower than the median value of 2.75. This mean difference compared to median values may indicate that that outlier values influenced the mean to be higher. Similarly, the mean value for antioxidant activity was 56.64%, compared to a lower median value of 49.65. In this case, the median value would be better and fair to describe the central tendency of both variables of interest. The standard deviation value was 5.47 for inorganic phosphate content and 22.73 for antioxidant activity. Usually, the standard deviation value is compared with one which is the value under a normal distribution. Since both variables had a higher standard deviation value than 1, this indicates a large variability around the mean value. It might suggest that the distribution is skewed.

**Table 3** – Central tendency analysis

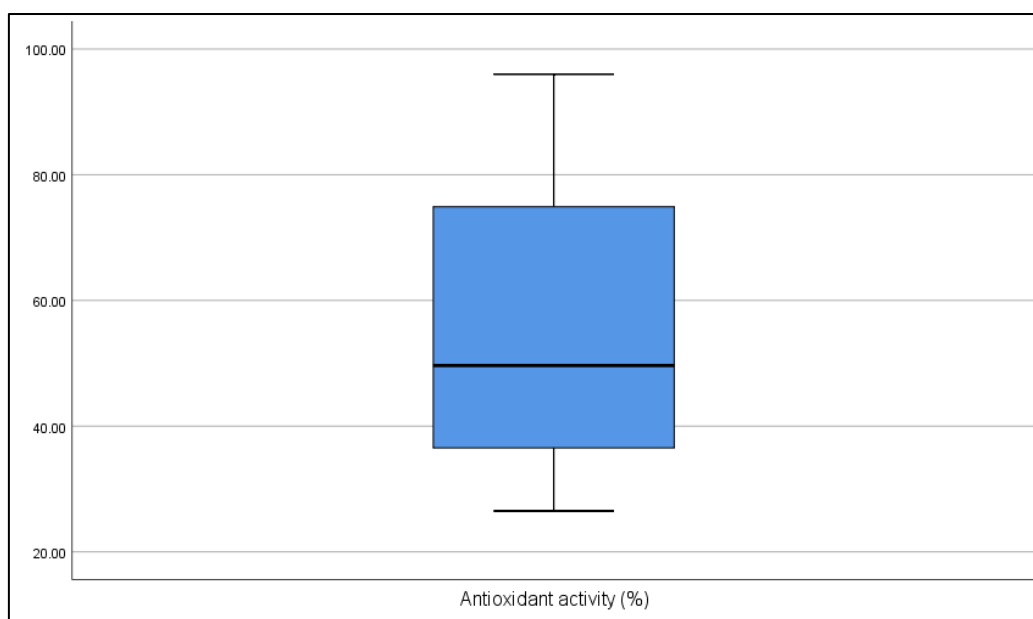
	Mean	Median	Std. Deviation	Minimum	Maximum
Inorganic phosphate content (mg/L)	5.49	2.75	5.47	0.23	17.33
Antioxidant activity (%)	56.64	49.65	22.73	26.50	95.98

##### 3.3.1.2. Variability and outliers check

By sample: Box plots for both inorganic phosphate content and antioxidant activity are shown in Figures 1 and 2, and the box plots show no outlier detected in both variables. Also, since the thick bold black line in the blue box representing the median value was found not in the middle of the box, the distribution of both variables is skewed.

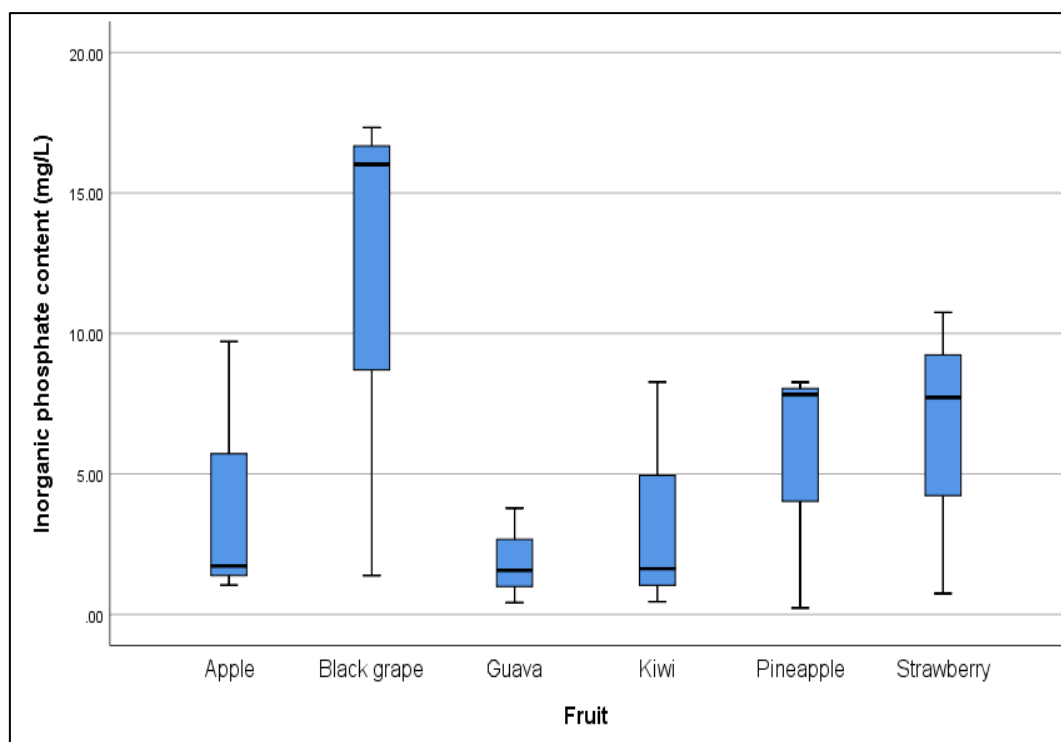


**Figure 1** – Box plot for inorganic phosphate content

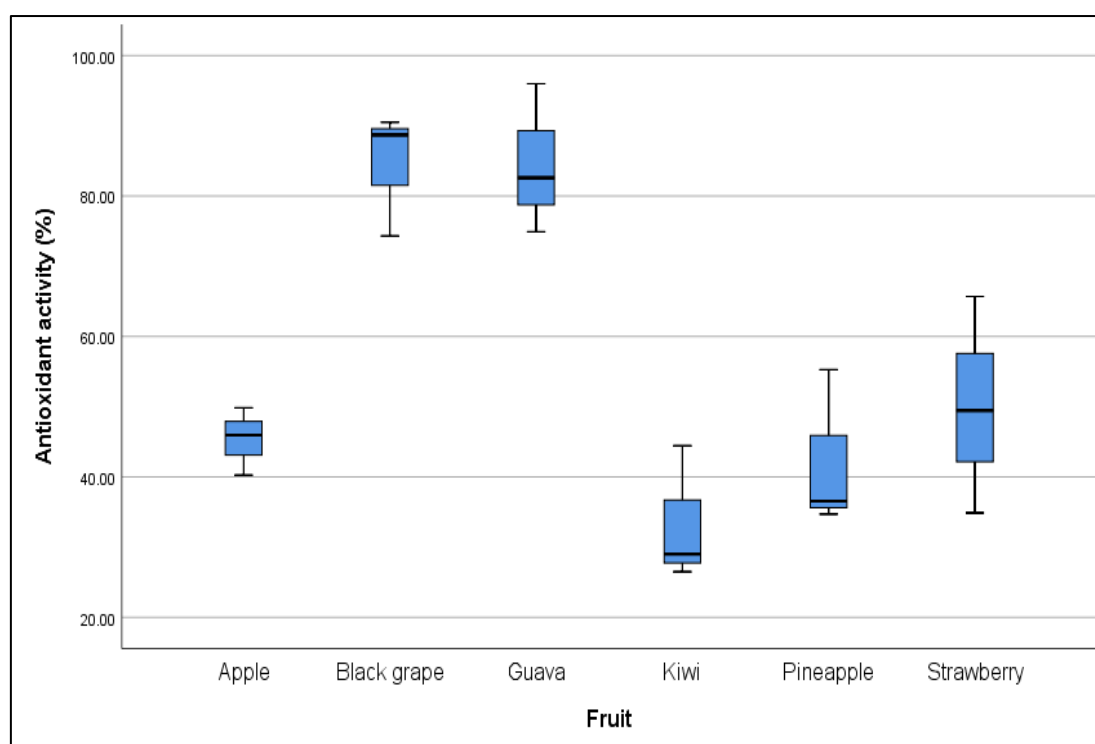


**Figure 2** – Box plot for antioxidant activity

By fruit: Box plot by fruit was produced as shown in Figures 3 and 4 for both variables of interest. The Box plot of Figure 3 shows the difference in the central tendency found in inorganic phosphate content across the six different fruits. However, it might be noticed that the box plots are overlapping. This might indicate that the differences are not statistically significant, and one-way ANOVA will be later performed to confirm this. The Box plot in Figure 4 shows a clear non-overlapping difference found across Black grape and Guava, compared to the remaining fruits in the antioxidant activity. This might suggest a statistically significant difference, which was later verified using one-way ANOVA test.



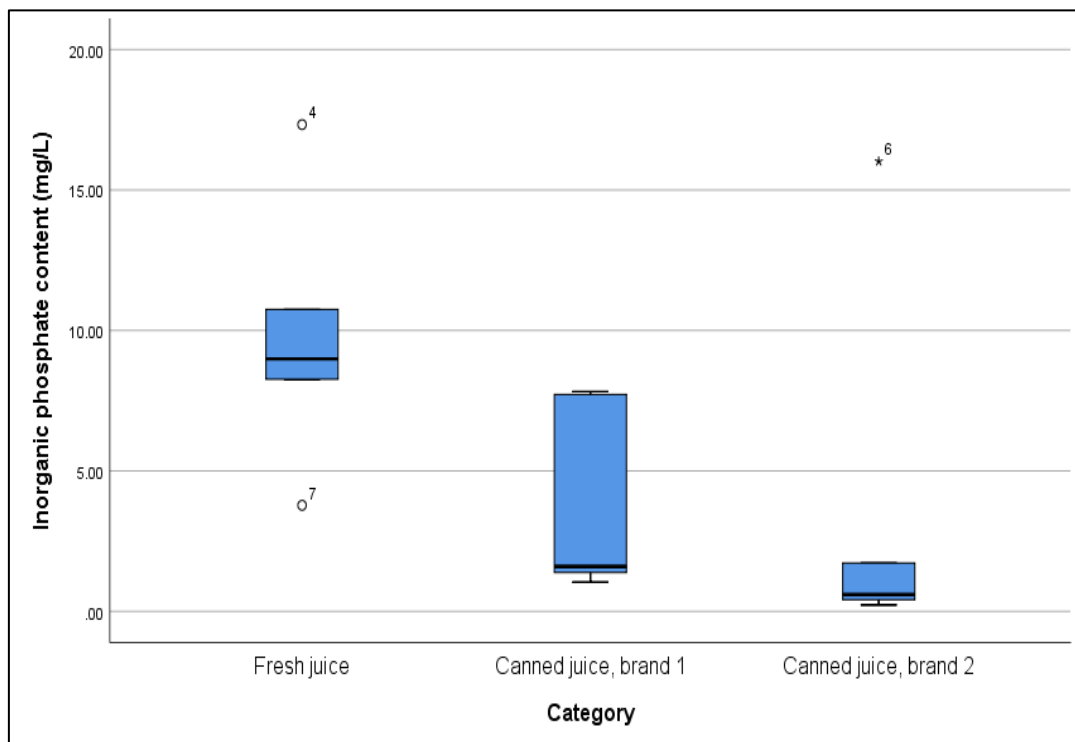
**Figure 3** – Box plot of inorganic phosphate content by fruit



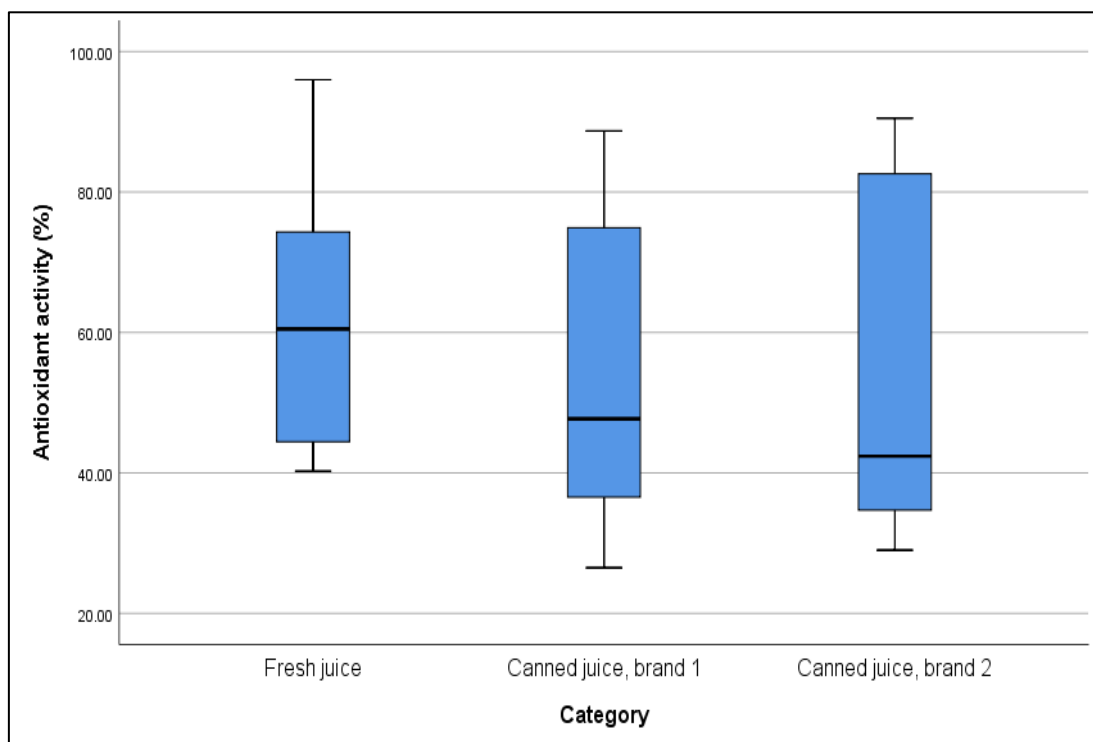
**Figure 4** – Box plot of antioxidant activity by fruit

By type: Box plot by type was produced as in Figures 5 and 6 for both variables of interest. The box plot in Figure 5 shows the difference in the central tendency found in inorganic phosphate content across the three different types. There is no overlapping found between fresh juice from one side and the other side, both canned juice, brand one and canned juice, brand 2. This might indicate that the differences are statistically significant. One-way ANOVA will be later performed to confirm this. The box plot of Figure 6 shows the difference in the central

tendency found in antioxidant activity across the three different types. However, it might be noticed that the box plots are overlapping. This would indicate that the differences are not statistically significant. One-way ANOVA was later performed to confirm this.



**Figure 5** – Box plot of inorganic phosphate content by type



**Figure 6** – Box plot of antioxidant activity by type



### 3.3.2. Normality assumption testing

By fruit: Prior to proceeding with the inferential testing, the normality assumption was tested for inorganic phosphate content by fruits, as shown in Table 4. The normality test null hypothesis states that: the variable follows a normal distribution, while the alternative hypothesis states that: the variable does not follow a normal distribution. Table 4 shows that the p-values results from the Kolmogorov-Smirnov test were zero, while the Shapiro-Wilk normality test for inorganic phosphate content by fruit had p-values greater than 0.05. Therefore, the null hypothesis was not rejected, and it is concluded that the normality assumption is verified.

**Table 4** – Tests of normality by the fruit of inorganic phosphate content

	Fruit	Kolmogorov-Smirnov <sup>a</sup>		Shapiro-Wilk	
		Statistic	p-value	Statistic	p-value
Inorganic phosphate content (mg/L)	Apple	0.360	0.000	0.808	0.134
	Black grape	0.359	0.000	0.811	0.141
	Guava	0.249	0.000	0.968	0.656
	Kiwi	0.334	0.000	0.860	0.267
	Pineapple	0.368	0.000	0.791	0.094
	Strawberry	0.268	0.000	0.951	0.573

a. Lilliefors Significance Correction

\* significant at 5% (p-value < 0.05); \*\* significant at 1% (p-value 0.01)

Normality assumption was tested for antioxidant activity by fruits, as shown in Table 5. Table 5 shows that the Shapiro-Wilk normality test for antioxidant activity by fruit had p-values greater than 0.05. Therefore, the null hypothesis was not rejected, and it is concluded that the normality assumption is verified. Therefore, the parametric test one-way ANOVA would be applied to test the effect of fruits on the two studied variables.

**Table 5** – Tests of normality by the fruit of antioxidant activity

	Fruit	Kolmogorov-Smirnov <sup>a</sup>		Shapiro-Wilk	
		Statistic	p-value	Statistic	p-value
Antioxidant activity (%)	Apple	0.216	0.000	0.988	0.794
	Black grape	0.348	0.000	0.832	0.194
	Guava	0.237	0.000	0.976	0.705
	Kiwi	0.338	0.000	0.852	0.246
	Pineapple	0.356	0.000	0.816	0.154
	Strawberry	0.181	0.000	0.999	0.940

a. Lilliefors Significance Correction

\* significant at 5% (p-value < 0.05); \*\* significant at 1% (p-value 0.01)

By type: Normality assumption was tested for inorganic phosphate content by type as shown in Table 6. Table 6 shows that Kolmogorov-Smirnov and Shapiro-Wilk normality tests for inorganic phosphate content by type had p-values greater than 0.05 for fresh juice. However, it was less than 0.05 for both canned juice, brand one and canned juice, brand 2 (p-value = 0.006, and 0.001/0.000 respectively) at 1% significance test. Therefore, the null hypothesis

was not rejected for fresh juice, and it is concluded that the normality assumption is verified. At the same time, the two remaining types were found to be not normally distributed.

**Table 6** – Tests of normality by type of inorganic phosphate content

	Category	Kolmogorov-Smirnov <sup>a</sup>		Shapiro-Wilk	
		Statistic	p-value	Statistic	p-value
Inorganic phosphate content (mg/L)	Fresh juice	0.238	0.200*	0.924	0.533
	Canned juice, brand 1	0.385	0.006**	0.695	0.006**
	Canned juice, brand 2	0.430	0.001**	0.565	0.000**

\*. This is a lower bound of the true significance.

a. Lilliefors Significance Correction

\* significant at 5% (p-value < 0.05); \*\* significant at 1% (p-value 0.01)

Table 7 shows that Kolmogorov-Smirnov and Shapiro-Wilk normality tests for antioxidant activity by type had p-values greater than 0.05. Therefore, the null hypothesis was not rejected, and it is concluded that the normality assumption is verified. Since there was a mix of normally distributed variables and not commonly distrusted variables, the nonparametric test Kruskal-Wallis as does not assume normality, will be applied to assess whether or not there is a mean difference effect due to type.

**Table 7** – Tests of normality by type of antioxidant activity

	Category	Kolmogorov-Smirnov <sup>a</sup>		Shapiro-Wilk	
		Statistic	p-value	Statistic	p-value
Inorganic phosphate content (mg/L)	Fresh juice	0.144	0.200*	0.950	0.743
	Canned juice, brand 1	0.238	0.200*	0.936	0.625
	Canned juice, brand 2	0.259	0.200*	0.836	0.120

\*. This is a lower bound of the true significance.

a. Lilliefors Significance Correction

\* significant at 5% (p-value < 0.05); \*\* significant at 1% (p-value 0.01)

### 3.3.3. Inferential statistics: One-way ANOVA for fruit effect testing

One-way ANOVA was performed to investigate any statistically significant mean difference due to fruit on both studied variables. The results of one-way ANOVA are reported in Tables 8, 9 and 10. Table 8 shows the mean values per fruit and the standard deviation and 95% confidence interval for the mean. Table 9 shows the homogeneity of variance test required for testing the pairwise mean comparisons. Table 10 shows the one-way ANOVA test findings. Since the p-value for inorganic phosphate content was greater than 0.05, then it is concluded that there is no statistically significant mean difference due to fruit at a 5% significance level. However, a statistically significant mean difference was due to fruit found in antioxidant activity (F value = 13.169, p-value = 0.000) at a 1% significance level. Therefore, and for further analysis, pairwise mean comparisons using the Scheffe method were reported for an antioxidant activity to test which pair of fruits were found statistically different on average.

**Table 8** – Mean values per fruit along with the standard deviation and 95% confidence interval for the mean

		N	Mean	Std. Deviation	95% Confidence Interval for Mean	
					Lower Bound	Upper Bound
<b>Inorganic phosphate content (mg/L)</b>	Apple	3	4.16	4.82	-7.82-	16.13
	Black grape	3	11.58	8.85	-10.41-	33.57
	Guava	3	1.92	1.71	-2.33-	6.16
	Kiwi	3	3.45	4.22	-7.02-	13.92
	Pineapple	3	5.44	4.52	-5.78-	16.66
	Strawberry	3	6.41	5.13	-6.35-	19.16
	Total	18	5.49	5.47	2.77	8.21
<b>Antioxidant activity (%)</b>	Apple	3	45.35	4.83	33.36	57.34
	Black grape	3	84.50	8.86	62.49	106.51
	Guava	3	84.50	10.66	58.01	110.98
	Kiwi	3	33.31	9.72	9.18	57.45
	Pineapple	3	42.17	11.39	13.88	70.46
	Strawberry	3	50.00	15.41	11.72	88.29
	Total	18	56.64	22.73	45.33	67.94

**Table 9** – Homogeneity of variance test

		Levene Statistic	df1	df2	p-value
<b>Inorganic phosphate content (mg/L)</b>	Based on Mean	2.613	5	12	0.080
	Based on Median	0.258	5	12	0.928
	Based on Median and with adjusted df	0.258	5	6.290	0.921
	Based on trimmed mean	2.201	5	12	0.122
<b>Antioxidant activity (%)</b>	Based on Mean	0.701	5	12	0.634
	Based on Median	0.263	5	12	0.925
	Based on Median and with adjusted df	0.263	5	9.667	0.923
	Based on trimmed mean	0.661	5	12	0.660

**Table 10** – One-way ANOVA test findings

		Sum of Squares	df	Mean Square	F	p-value
<b>Inorganic phosphate content (mg/L)</b>	Between Groups	169.779	5	33.956	1.205	0.364
	Within Groups	338.124	12	28.177		
	Total	507.903	17			
<b>Antioxidant activity (%)</b>	Between Groups	7431.302	5	1486.260	13.169	0.000**
	Within Groups	1354.360	12	112.863		
	Total	8785.662	17			

\* significant at 5% (p-value < 0.05); \*\* significant at 1% (p-value 0.01)

Scheffe pairwise mean comparisons (Table 11) showed that Black grape and Guava had statistically significant higher mean values of antioxidant activity compared to Apple (p-value = 0.021, 0.21 respectively) at a 5% significance level. At the same time, there was no statistically significant pair mean comparison found otherwise. This confirms what was found in the box plot earlier.

**Table 11** – Scheffe pairwise mean comparisons

Dependent Variable	(I) Fruit	(J) Fruit	Mean Difference (I-J)	Std. Error	p-value
Antioxidant activity (%)	Apple	Black grape	-39.15*	8.67423	0.021*
		Guava	-39.15*	8.67423	0.021*
		Kiwi	12.04	8.67423	0.850
		Pineapple	3.18	8.67423	1.000
		Strawberry	-4.65-	8.67423	0.997
	Black grape	Apple	39.15*	8.67423	0.021*
		Guava	0.00	8.67423	1.000
		Kiwi	51.19*	8.67423	0.003**
		Pineapple	42.33*	8.67423	0.013*
		Strawberry	34.50*	8.67423	0.047*
	Guava	Apple	39.15*	8.67423	0.021*
		Black grape	0.00	8.67423	1.000
		Kiwi	51.18*	8.67423	0.003**
		Pineapple	42.32*	8.67423	0.013*
		Strawberry	34.49*	8.67423	0.047*
	Kiwi	Apple	-12.04-	8.67423	0.850
		Black grape	-51.19*	8.67423	0.003**
		Guava	-51.18*	8.67423	0.003**
		Pineapple	-8.86-	8.67423	0.952
		Strawberry	-16.69-	8.67423	0.608
	Pineapple	Apple	-3.18-	8.67423	1.000
		Black grape	-42.33*	8.67423	0.013*
		Guava	-42.32*	8.67423	0.013*
		Kiwi	8.86	8.67423	0.952
		Strawberry	-7.83-	8.67423	0.972
	Strawberry	Apple	4.65	8.67423	0.997
		Black grape	-34.50*	8.67423	0.047*
		Guava	-34.49*	8.67423	0.047*
Kiwi		16.69	8.67423	0.608	
Pineapple		7.83	8.67423	0.972	

\*. The mean difference is significant at the 0.05 level.

\* significant at 5% (p-value < 0.05); \*\* significant at 1% (p-value 0.01)

### 3.3.4. Kruskal-Wallis for a type effect test

Nonparametric Kruskal-Wallis, which is equivalent to the parametric one-way ANOVA, was performed to investigate any statistically significant mean difference due to type on both studied variables. The results of Kruskal-Wallis are reported in Table 12 and Table 13. Since the p-value for antioxidant activity was greater than 0.05, then it is concluded that there is no statistically significant effect found due to type at 5% significance level. However, a statistically significant effect was found due to type found in inorganic phosphate content (Kruskal-Wallis H = 7.871, p-value = 0.020) at a 5% significance level. Therefore, and for further analysis, mean and median values were computed for antioxidant activity by type. Table 13 shows that both mean and median values were higher in the fresh juice compared to both canned juice, brand one and canned juice, brand 2.

**Table 12** – Test statistics (A)

	Kruskal-Wallis H	df	p-value
Inorganic phosphate content (mg/L)	7.871	2	0.020*
Antioxidant activity (%)	0.889	2	0.641

a. Grouping Variable: Category

\* significant at 5% (p-value < 0.05); \*\* significant at 1% (p-value 0.01)

**Table 13** – Test statistics (B)

	Category					
	Fresh juice		Canned juice, brand 1		Canned juice, brand 2	
	Mean	Median	Mean	Median	Mean	Median
Inorganic phosphate content (mg/L)	9.68	8.99	3.53	1.60	3.26	0.60

### 3.3.5. Correlational analysis

All samples: Correlational analysis using Pearson's parametric method was performed in Table 14. Table 14 shows no statistically significant correlation between inorganic phosphate content and antioxidant activity (Pearson correlation coefficient = 0.279, p-value = 0.263) at a 5% significance level.

By fruit: For further analysis, the correlational analysis was produced for each fruit separately as shown in Table 4. There was no statistically significant correlation between inorganic phosphate content and antioxidant activity among each fruit sample individually at a 5% significance level.

By type: The correlational analysis was produced for each type separately for further analysis. There was no statistically significant correlation between inorganic phosphate content and antioxidant activity among each type of sample at a 5% significance level.

**Table 14** – Correlational analysis of all samples, by fruit and by sample

		Inorganic phosphate content (mg/L)	
<b>All sample</b>	Antioxidant activity (%)	Pearson Correlation	0.279
		p-value	0.263
<b>Apple</b>	Antioxidant activity (%)	Pearson Correlation	-0.884
		p-value	0.309
<b>Black grape</b>	Antioxidant activity (%)	Pearson Correlation	-0.476
		p-value	0.684
<b>Guava</b>	Antioxidant activity (%)	Pearson Correlation	0.757
		p-value	0.453
<b>Kiwi</b>	Antioxidant activity (%)	Pearson Correlation	0.964
		p-value	0.171
<b>Pineapple</b>	Antioxidant activity (%)	Pearson Correlation	0.608
		p-value	0.584
<b>Strawberry</b>	Antioxidant activity (%)	Pearson Correlation	0.968
		p-value	0.163
<b>Fresh juice</b>	Antioxidant activity (%)	Spearman's rho	-0.086
		Correlation Coefficient	
<b>Canned juice, brand 1</b>	Antioxidant activity (%)	p-value	0.872
		Spearman's rho	-0.429
<b>Canned juice, brand 2</b>	Antioxidant activity (%)	Correlation Coefficient	
		p-value	0.397
		Spearman's rho	0.543
		Correlation Coefficient	
		p-value	0.266

## 4. Conclusions

The inorganic phosphate content and antioxidant activity of selected fruit juices commonly consumed in Bahrain were determined and compared. Black grape juice had the highest inorganic phosphate content among the fresh juices, and guava juice had the highest antioxidant activity. Black grape juices had the highest phosphate content and antioxidant

activity among the canned juices. Statistically, based on inferential statistical analysis (normality assumption tests and one-way ANOVA), no significant correlation was found between the inorganic phosphate content and antioxidant activity at a 5% significance level.

### Acknowledgements

We would like to thank Mr Jalal Naser for his assistance in the statistical analysis.

### References

- [1] S. Berchmans, T. B. Issa, and P. Singh, 'Determination of inorganic phosphate by electroanalytical methods: A review', *Anal. Chim. Acta*, vol. 729, pp. 7–20, Jun. 2012, doi: 10.1016/j.aca.2012.03.060.
- [2] J. Uribarri and M. S. Calvo, 'Dietary Phosphorus Excess: A Risk Factor in Chronic Bone, Kidney, and Cardiovascular Disease?', *Adv. Nutr.*, vol. 4, no. 5, pp. 542–544, Sep. 2013, doi: 10.3945/an.113.004234.
- [3] National Institute of Health, 'Phosphorus', *U.S. Department of Health & Human Services, National Institutes of Health*, 2021. <https://ods.od.nih.gov/factsheets/Phosphorus-HealthProfessional/#en2> (accessed May 06, 2021).
- [4] F. Mehraban et al., 'Molecular insights into the effect of ozone on human hemoglobin in autohemotherapy: Highlighting the importance of the presence of blood antioxidants during ozonation', *Int. J. Biol. Macromol.*, vol. 119, pp. 1276–1285, Nov. 2018, doi: 10.1016/j.ijbiomac.2018.08.028.
- [5] Z. Zou, W. Xi, Y. Hu, C. Nie, and Z. Zhou, 'Antioxidant activity of Citrus fruits', *Food Chem.*, vol. 196, pp. 885–896, Apr. 2016, doi: 10.1016/j.foodchem.2015.09.072.
- [6] H. G. Bulama, D. Dahiru, and J. O. Madu, 'Investigation of the Anti-cataract and Antioxidant Activities of *Cnidioscolus aconitifolius* Leaves Extract In vitro', *Iraqi J. Sci.*, vol. 62, no. 1, pp. 28–38, 2021.
- [7] A. A. Ali Redha, A. M. Hasan, and Q. Mandeel, 'Phytochemical Determinations of Pomegranate (*Punica granatum*) Rind and Aril Extracts and their Antioxidant, Antidiabetic and Antibacterial Activity', *Nat. Prod. Chem. Res.*, vol. 06, no. 04, 2018, doi: 10.4172/2329-6836.1000332.
- [8] B. Baby, P. Antony, and R. Vijayan, 'Antioxidant and anticancer properties of berries', *Crit. Rev. Food Sci. Nutr.*, vol. 58, no. 15, pp. 2491–2507, Oct. 2018, doi: 10.1080/10408398.2017.1329198.
- [9] H. Akhavan, M. Barzegar, H. Weidlich, and B. F. Zimmermann, 'Phenolic Compounds and Antioxidant Activity of Juices from Ten Iranian Pomegranate Cultivars Depend on Extraction', *J. Chem.*, vol. 2015, pp. 1–7, 2015, doi: 10.1155/2015/907101.
- [10] O. E. Aina, S. O. Amoo, L. L. Mugivhisa, and J. O. Olowoyo, 'Effect of organic and inorganic sources of nutrients on the bioactive compounds and antioxidant activity of tomato', *Appl. Ecol. Environ. Res.*, vol. 17, no. 2, pp. 3681–3694, 2019, doi: 10.15666/aer/1702\_36813694.
- [11] M. Ibrahim, H. Jaafar, E. Karimi, and A. Ghasemzadeh, 'Impact of Organic and Inorganic Fertilizers Application on the Phytochemical and Antioxidant Activity of Kacip Fatimah (*Labisia pumila* Benth)', *Molecules*, vol. 18, no. 9, pp. 10973–10988, Sep. 2013, doi: 10.3390/molecules180910973.
- [12] K. H. Wern, H. Haron, and C. B. Keng, 'Comparison of total phenolic contents (TPC) and antioxidant activities of fresh fruit juices, commercial 100% fruit juices and fruit drinks', *Sains Malaysiana*, vol. 45, no. 9, pp. 1319–1327, 2016.
- [13] Z. Ali and S. B. Bhaskar, 'Basic statistical tools in research and data analysis', *Indian J. Anaesth.*, vol. 60, no. 9, p. 662, 2016, doi: 10.4103/0019-5049.190623.
- [14] F. Al-Salman, A. Ali Redha, and Z. Al-Zaimoor, 'Inorganic Analysis and Antioxidant Activity of Shilajit', *Int. J. Sci. Res. Chem. Sci.*, vol. 7, no. 3, pp. 5–10, 2020.
- [15] A. Ali Redha, 'Phytochemical Investigations of Nerium Oleander L. Leaves and Flowers', *Int. J. Sci. Res. Chem. Sci.*, vol. 7, no. 4, pp. 1–4, 2020.
- [16] A. H. Al-basheer and S. M. Al-wandawi, 'In Vitro Assessment of the Antioxidant and Antitumor Potentials of Biogenic Silver Nanoparticle', *Iraqi J. Sci.*, vol. 61, no. 6, pp. 1253–1264, 2020.