

STABILITY STUDY ON PHYSICOCHEMICAL COMPOSITION, BIOACTIVE COMPOUNDS AND ANTIOXIDANT POTENTIAL OF APPLE JAM: INFLUENCE OF DAILY STORAGE CONDITIONS

– Research paper –

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Abstract: The fruits and their derivatives are products known for their excellent energy intake and their beneficial health effects. Jams are an efficient and tasty way of fruit preservation. Jam quality is greatly influenced by storage conditions like temperature and duration. The current investigation aimed to study the evaluation changes of physicochemical characteristics (total sugars (TS), free amino acids (FAA), hydroxymethylfurfural (HMF), and ascorbic acid (AA)), phytochemical composition (carotenoid, total phenolic, and flavonoid contents), and antioxidant ability (DPPH and ferric reducing power) of two brands of commercial apple jam during daily conditions of use. The jams were assessed during the first 30 days after first opening the tins, with testing the effect of 25°C (ambient temperature) and 35°C (hot summer temperature). The findings show that after a month of storage at 25 and 35°C, the decreasing levels for samples were respectively 11-13% (S1) and 12-16% (S2) for TS, 63-68% (S1), and 57-63% (S2) for FAA, 6-7% (S1) and 4-5% (S2) for AA; whereas, HMF revealed increases of 127-143% (S1) and 186-232% (S2). Likewise, significant losses of bioactive substances and antioxidant capacity have been observed for jam samples. Based on the current findings, it can be concluded that storing industrial apple jam under daily conditions of use leads to significant loss of quality and antioxidant parameters. For this reason, it is recommended to consume the apple jam as soon as possible after opening the tin and to store it at relatively low temperatures during use.

Keywords: Physicochemical characteristic, daily storage conditions, bioactive compounds, antioxidant activity, apple jam.

INTRODUCTION

The nutritious value and potential health benefits of fruits make them an excellent dietary addition (Sosa et al. 2012). Fruits are a rich source of bioactive substances that contribute to their potential as antioxidants, including carotenoids, flavonoids, phenolics, and vitamins. The fruits are extremely perishable; therefore, they are processed into a variety of shelf-stable goods in order to limit the loss of fruits that is not eaten fresh, and to enable consumers to benefit from its virtues throughout the year, thus ensuring a consistent intake of their valuable nutrients and bioactive substances (Shinwari and Rao 2018).

Apples are a member of the *Rosaceae* family and are cultivated widely in temperate regions of the world. Because of their wonderful flavor and high nutritional content, apples are one of the most popular fruits consumed globally (Rodríguez et al. 2014; Shalini and Gupta, 2010; Zhang et al. 2021). Apples are one of the most important fruits in a human diet, providing significant energy, antioxidants, vitamins, minerals, and dietary fiber (Han et al. 2023). Epidemiological research has linked the consumption of apples to a reduced risk of diabetes, asthma, several types of cancer, and cardiovascular illnesses (Karaman et al. 2010; Oszmiański et al. 2008; Vieira et al. 2011). Due to the seasonality of production, apple must be preserved to make them available for human consumption all year-round (Vidhya and Narain

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2011). Therefore, in addition to fresh consumption, many methods and processes for preserving of this fruit have been developed, resulting in a range of products such as juice, jam, jelly, marmalade, and nectar.

Jam is one of the most widely consumed fruit-based shelf-stable products. Among various apple derivatives, apple jam serves as a valuable substitute for the fruit's economic exploitation. Fruit jams are commonly used as fillings for cakes, breads, biscuits, and various other baked goods. Jam is typically made from fruit, sugar, pectin, and citric acid. These ingredients are cooked together to create a flavorful product with the right balance of sweetness and good preservation properties. It should be able to be stored for a considerable period of time without the risk of deterioration (Abid et al. 2014; Mohd Naeem et al. 2017; Shinwari and Rao 2018; Wojdyło et al. 2008).

The nutritional qualities, phenolics, flavonoids, and antioxidant activities of jam can all be significantly impacted by storage conditions (Morelli and Prado 2012; Scrob et al. 2022). According to some research, a variety of factors, such as oxygen content, packaging type, and processing and storage circumstances, might impact food quality characteristics. The impact of storage conditions

on the degradation of bioactive substances and antioxidant potential was studied for many jams formulated from different fruits, including strawberry (Patras et al. 2011), blackberry (De Moura et al. 2012), orange, cherry, strawberry, fig, and apricot (Rababah et al. 2011), and lingonberry (Scrob et al. 2022). Most studies on the storage of jams focus on the storage of the initial product, but few investigations have looked at the actual conditions of daily use. Indeed, after opening the containers, jam may begin to degrade as it is exposed to air and moisture, leading to changes in texture, color, and flavor over time, especially when stored in hot temperatures. Additionally, the presence of air, along with the development of certain bacterial and yeast strains due to increased humidity, can promote oxidation, potentially resulting in a loss of nutritional value.

The aim of this research work was to monitor the stability of physicochemical characteristics, phenolic contents (total phenolics and flavonoids), and antioxidant properties of commercial apple jam during a 30-day storage period under daily conditions of use. This involved testing both standard temperature (25°C) and elevated temperature (35°C), simulating the summer season.

MATERIALS AND METHODS

Chemicals

Sodium carbonate was purchased from Sigma-Aldrich (Switzerland); Folin-Ciocalteu's phenol reagent, ethanol, and methanol were provided by Biochem, Chemopharma (Montreal, Quebec); 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,6-dichlorophenol-indophenol were obtained from Sigma-Aldrich (Germany); potassium ferricyanide and aluminum chloride were acquired from Biochem, Chemopharma (Georgia, USA); gallic acid was from Prolabo (Montreuil, France).

Samples and extracts preparation

Two different brands of industrial apple jams (two tins of each brand from two randomly selected batches) were purchased from a local retail market in Bejaia (Algeria) designated as sample 1 (S1) and sample 2 (S2). The analyzed samples, S1 and S2, are characterized by titratable acidity of 0.462 and 0.530%, with total soluble solids content of 68.36 and 73.15°Brix, respectively. Samples were obtained within the first month of production, with a shelf life of 3 years indicated on the package. The ingredients of jam, as stated on the label, are

fruits, sugar (sucrose), pectin, and citric acid. After opening the tins, the samples were stored at room temperature (25°C) to represent standard storage conditions, and at 35°C to simulate the effects of hot temperatures, such as those experienced during the summer period. The jams were periodically assessed over a 30-day storage period, with evaluations conducted at 0, 3, 6, 9, 12, 15, and 30 days. The experimental methodology employed in this study is outlined in Figure 1 and explained in more depth later in this section.

Physicochemical characterization

The phenol-sulfuric acid method was used to determine the total sugar concentration (Dubois et al. 1956). Briefly, after mixing 0.5 mL of phenol solution (5%) to the diluted sample (0.5 mL), 2.5 mL of concentrated sulfuric acid was added. The absorbance was measured at 490 nm after incubation at 105°C for 5 min. The total sugar content was calculated using a standard curve of sucrose.

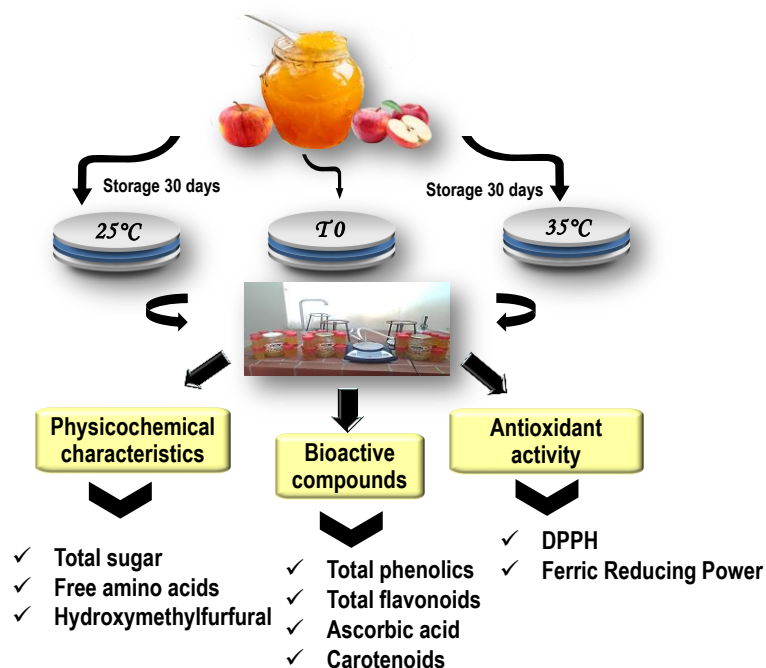


Figure 1. Diagram illustrating the experimental approach used for storing and assessing jam.

The method of Yemm and Cocking (1955) was used for free amino acids determination. 1 mL of jam aqueous solution was mixed with 0.5 mL of citrate buffer (0.2 M, pH 5), 1 mL of potassium cyanide (0.01 M), and 200 μ L of 1% ninhydrin. After 15 min of incubation at 100°C, 2.3 mL of 60% ethanol was added to the reaction mixture. The absorbance was measured at 570 nm, and the results were reported as mg glycine equivalent per 100g of jam referring to a calibration curve.

The HMF content was determined according to the method described by White (1979). A volume of 100 μ L of Carrez I (15% potassium ferricyanide) and Carrez II (30% zinc acetate) reagents was added to the homogenized sample and distilled water was used to get the volume down to 10 milliliters. After the supernatant was paper-filtered, 1 mL of the filtrate was placed in each of test tubes along with 1 mL of either distilled water (a sample) or 0.2% bisulfite (a reference). Equation 1 was used to determine the HMF content:

$$\text{HMF (mg/100g)} = (A_{284} - A_{336}) \times 74.85 \quad [1]$$

where, A_{284} , A_{336} : absorbance values at 284 and 336 nm; 74.85: specific factor of dilution and conversion.

The ascorbic acid content (AAC) was measured using the methodology outlined by Mau et al. (2005). The sample (1g) was homogenized with 10 mL of 1% oxalic acid for 30 min. Following centrifugation, 100 μ L of the supernatant was added to 900 μ L of 2,6 dichlorophenol-indophenol. The absorbance was measured at 515 nm and AAC was reported as milligrams of ascorbic acid

equivalents per 100g of jam using calibration curve made with different concentration of pure ascorbic acid.

Phytochemical composition

The extraction process of total carotenoid was described by Choi et al. (2002). 10 mL of a 50:25:25 (v/v/v) mixture of hexane, acetone, and ethanol were mixed with 1g of jam. Once the upper layer of hexane containing the carotenoids was collected using centrifugation for 10 min at 5000 rpm, the absorbance was measured at 570 nm. With reference to a calibration curve, the results were reported as mg of β -carotene equivalent per 100g of jam (mg CE/100g).

For the total phenolic and flavonoids content as well as antioxidant activity an acetonic extract was performed. Concisely, 1g of the sample was mixed with 20 mL of 50% acetone. The extract was collected after extraction by paper-filtration and centrifugation at 5000 rpm for 15 min.

The total phenolic content (TPC) was evaluated using the Folin-Ciocalteu reagent (Singleton and Rossi, 1974). Two hundred microliters of acetonic extract were mixed with 0.75 mL of diluted Folin-Ciocalteu reagent (1/10, v/v). Four minutes later, 400 μ L of 7.5% sodium carbonate was added. Following 90 min of incubation at room temperature, the mixture's absorbance at 765 nm was determined. The results were given as milligrams of gallic acid equivalents per 100 grams of jam (mg GAE/100 g).

The total flavonoid content (TFC) was determined using the procedure described by Quettier-Deleu et al. (2000). Equal volumes of acetonic extract and 2% aluminum chloride solution were mixed. After 10 minutes of incubation, the mixture's absorbance was measured at 410 nm. TFC were expressed as mg quercetin equivalents per 100 g of jam (mg QE/100g).

Antioxidant activity

The DPPH[•] radical scavenging assay is employed as previously described (Tezcan et al. 2009). Briefly, 1 mL of methanolic DPPH solution was mixed with 200 μ L of the extract. Prior to measuring the absorbance at 517 nm, the test tubes were placed in the dark for 30 minutes. The antioxidant activity was expressed as mg gallic acid equivalents per 100 g of jam (mg GAE/100g). The procedure described by Oyaizu (1986) was used to measure the ferric reducing power (FRP). In brief, 100 μ L of extract was mixed with 250 μ L

of potassium ferricyanide (1%), and 250 μ L of phosphate buffer (0.2 M, pH 6.6). Then, 850 μ L of distilled water, 250 μ L of 10% trichloroacetic acid and 170 μ L of 0.1% ferric chloride were added to the mixture after 20 min incubation at 50°C. The absorbance was measured at 700 nm. The results were reported as milligrams of gallic acid equivalents per 100g of jam (mg GAE/100g).

Statistical analysis

INFOSTAT software was used to analyze the data. The effect of temperature and storage duration were incorporated in the model, and analysis of variance was carried out using the ANOVA approach with two factors. At a least significant difference of $P < 0.05$, means were separated using LSD analysis. Three duplicates of each experiment were run. Correlations were conducted at three distinct significance levels (0.05, 0.01 and 0.001) using the STATISTICA 5.5 software.

RESULTS AND DISCUSSION

Influence of storage time and temperature on nutritional characteristic

Total sugars content

The most significant constituent of fruit products is sugar, which plays a crucial role in flavor development and acts as a natural food preservative (Pavlova et al. 2013). It is clear from Table 1 that temperature and storage duration had an impact on the total sugar content. After 30 days of storage at 25 and 35°C, the initial total sugar content of the jam decreased by 10.5 and 13.4% for S1, and by 11.8 and 16.3% for S2, respectively. Statistical analysis revealed that the interaction between temperature and storage duration had a significant impact ($p < 0.05$) on the total sugar content. The coconut jam's total sugar content decreased by 0.56% and 0.87% following six months of storage at 28 and 37°C. The reducing sugars could be due to their implication in the formation of HMF and non-enzymatic browning products.

Free amino acids content

The evaluation of the impact of storage conditions on amino acid content in jams has been the subject of only a few investigations. Amino acids are

crucial food components that not only increase the nutritional quality of food but also have several health benefits. Additionally, they serve a variety of purposes including providing energy, serving as a source of protein precursors, and adding to the sensory quality of foods; therefore, it is increasingly important to determine how they affect the quality of fruit-derived products (Dajanta et al. 2011; Odriozola-Serrano et al. 2013).

Under the experimental conditions applied, the results shown in Table 1 demonstrated that the total FAA content was impacted during storage. Hence, during conservation, there was a notable reduction. Extended storage duration and heightened temperature led to greater decreases. The total FAA content diminished by 63.3 and 68.2% for S1 and 57.1 and 63.4% for S2 after 30 days of storage at 25 and 35°C, respectively. These findings concur with those of Touati et al. (2014), who found that after two months of apricot jam storage at 25 and 37°C, total FAA dropped by 34 and 46%, respectively. Likewise, Djaoudene and Louaileche (2016a) recorded losses ranging from 50 to 60% during orange jam storage. This decrease may be caused by the involvement of amino acids in the non-enzymatic browning process as Maillard reactions.

Table 1. Total sugar (TS), free amino acids (FAA) and HMF changes during apple jam storage.

Storage time (Days)	TS (g/100g)				FAA (mg/100g)				HMF (mg/100g)			
	25°C		35°C		25°C		35°C		25°C		35°C	
	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2
0	69.04± 2.03 ^a	66.91± 1.20 ^a	69.04± 2.03 ^a	66.91± 1.20 ^a	4.47± 0.08 ^a	13.14± 0.16 ^a	4.47± 0.08 ^a	13.14± 0.16 ^a	3.50± 0.62 ^f	0.69± 0.11 ^g	3.50± 0.62 ^f	0.69± 0.11 ^f
3	68.83± 2.04 ^{ab}	66.82± 0.65 ^a	67.15± 2.07 ^{ab}	64.93± 0.60 ^{ab*}	3.79± 0.19 ^b	12.34± 0.09 ^b	3.52± 0.15 ^{b*}	11.74± 0.20 ^{b*}	4.11± 0.26 ^c	1.03± 0.05 ^f	4.86± 0.24 ^{e*}	1.17± 0.10 ^{e*}
6	67.79± 1.20 ^{abc}	65.90± 0.45 ^a	66.52± 2.60 ^{abc}	64.07± 1.01 ^b	3.69± 0.09 ^b	11.65± 0.12 ^c	3.39± 0.17 ^{b*}	11.30± 0.12 ^{c*}	5.17± 0.39 ^d	1.15± 0.03 ^e	5.54± 0.64 ^d	1.27± 0.05 ^{de*}
9	67.27± 0.71 ^{bc}	63.17± 1.15 ^b	65.05± 1.70 ^{bc}	63.00± 0.63 ^{bc}	3.38± 0.12 ^c	10.26± 0.13 ^d	3.11± 0.17 ^{c*}	9.77± 0.08 ^{d*}	5.61± 0.54 ^d	1.27± 0.04 ^d	6.10± 0.58 ^{c*}	1.36± 0.11 ^d
12	66.81± 0.35 ^c	63.11± 0.27 ^b	64.68± 1.27 ^{bc}	61.90± 1.16 ^c	2.51± 0.07 ^d	9.01± 0.30 ^e	2.27± 0.11 ^{d*}	8.48± 0.32 ^e	6.31± 0.60 ^c	1.37± 0.04 ^c	6.59± 0.65 ^c	1.48± 0.05 ^{c*}
15	66.74± 0.35 ^c	63.17± 1.10 ^b	64.25± 0.75 ^{c*}	61.75± 0.25 ^{c*}	2.42± 0.06 ^d	8.66± 0.20 ^f	2.20± 0.17 ^{d*}	8.08± 0.19 ^{f*}	6.86± 0.88 ^b	1.57± 0.06 ^b	7.40± 0.78 ^b	1.82± 0.04 ^{b*}
30	61.80± 1.22 ^d	59.00± 0.50 ^c	59.82± 0.27 ^{d*}	56.05± 0.29 ^{d*}	1.64± 0.30 ^e	5.64± 0.17 ^g	1.42± 0.32 ^{e*}	4.81± 0.09 ^{g*}	7.94± 0.92 ^a	1.97± 0.05 ^a	8.49± 0.72 ^{a*}	2.29± 0.10 ^{a*}

In the same column, means ± SD with the same letters (a–g) have no significant differences ($P \leq 0.05$). Asterisk (*) indicate significant differences ($P \leq 0.05$) between the results obtained at 25°C and 35°C. LSD interaction temperature-time of TS, FAA and HMF of sample 1 (S1) and sample 2 (S2) are 2.22, 0.20, 0.50, and 2.03, 0.26, 0.10, respectively.

Hydroxymethylfurfural content

In a variety of foods, including honey, jam, fruit juices, and milk, HMF is recognized as a sign of poor storage and quality degradation. This aldehyde is produced as an intermediate in the Maillard process, from reaction of amino acids and reducing sugars. When jams are stored in improper circumstances throughout marketing, their HMF concentration may increase (Anese and Suman 2013; Korus et al. 2015; Lee et al. 2019; Severin et al. 2010). The initial HMF content of the jam was 3.5 mg/100 g for S1 and 0.69 mg/100 g for S2. A significant increase in HMF content during storage was observed at both tested temperatures 25 and 35°C (126.9 and 142.6% for S1, and 185.5 and 231.9% for S2, respectively). This finding demonstrated that the generation of HMF was highly dependent on storage temperature (Table 1). According to statistical analysis, the time-temperature interaction had a substantial ($p < 0.05$) impact on HMF content. These findings were in line with the research conducted by Rada-Mendoza et al. (2004), who similarly found that peach jam's HMF level increased significantly after a year of storage, reaching 316 and 577% at 20 and 35°C, respectively. According to Duru et al. (2012), rosehip nectars stored for eight months showed a considerable rise in HMF content, reaching 771 and 182% at 25 and 35°C, respectively. Additionally, the increase in HMF content found in this study was similar than those shown by previous investigations on orange jam (Djaoudene and Louaileche 2016a) and on apple pekmez products (Kuşçu and Bulanteki, 2016). The breakdown of sugars and amino acids in an acidic environment may be responsible for the accumulation of HMF.

Influence of storage time and temperature on phytochemical composition

Ascorbic acid content

Vitamin C is an essential component for human health and may help prevent a number of degenerative disorders, due to its strong antioxidant properties and ability to protect the body from free radical damage (Esteve et al. 2005). Ascorbic acid is an unstable substance when stored under certain conditions, such as exposure to air and light, and at high temperatures. The loss of ascorbic acid may be both a critical factor for the shelf life and quality indicator, since the degradation reactions of ascorbic acid are frequently the cause of major quality changes that occur during foods storage. The degradation of vitamin C has been observed to produce a variety of reactive decomposition products, which can

then react with amino acids to make brown pigments (Duru et al. 2012). As per the findings displayed in Figure 2A, storage at 25°C resulted in a significant decrease of 6 and 4% of AA content, while storage at 35°C induced a decrease of 7% in S1 and 5% in S2. In this line, Patras et al. (2011) obtained similar findings when examining variations in AA of strawberry jams stored for 28 days at 15°C. AAC in black plum (*Syzygium cumini*) jam decreased over the storage period as mentioned by Aslam et al. (2019). The authors reported that this reduction might be caused by the reaction of conversion of ascorbic acid into dehydroascorbic acid under the effect of the oxidation. Additionally, Mazur et al. (2014) reported that the concentration of AA considerably reduced over the course of strawberry jam's six-month storage period at 20°C. However, the study reported by Pavlova et al. (2013) revealed that the vitamin C content of jams made from raspberry and peach stored for 90 days was retained its vitamin C content.

Total carotenoid content

Carotenoids are a class of naturally occurring pigments that are lipid-soluble, widely distributed in nature, and have a variety of structural properties as well as multiple vital functions for human health (Gama and Sylos 2005). These bioactive substances are the precursors of vitamin A, which the body converts to retinol. The functional relevance of carotenoids is due to their antioxidant effect in addition to its role as a pro-vitamin A (Plaza et al. 2011). However, because carotenoids are extremely susceptible to changes in light, heat, air, and other factors, the storage conditions may result in degradation or loss. Figure 2B illustrates the changes in total carotenoid content of the apple jam during storage. Before the storage, the carotenoid content of S1 and S2 jam samples was measured and found to be 0.52 and 0.61 mg/100g, respectively. At the end of storage, it was found that the carotenoid contents of the studied samples decreased by 17 and 19% for S1, and 23 and 26% for S2, respectively. In this regard, Igual et al. (2013) revealed that the carotenoid content of grapefruit jam exhibited a decreasing trend throughout the duration of 90 days of storage. According to Bernás et al. (2023), storage duration is one parameter that influences the amount and profile of carotenoids, potentially leading to the loss of carotenoids during processing and storage. The loss in carotenoid content during storage might be resulted to the isomerization reactions and/or fragmentation and oxidation.

Total phenolic compounds

Due to their advantageous health effects, phenolics have been found in a number of studies to have a significant impact on human health, particularly when they are present in food, especially fruits. In addition to their antioxidant qualities, phenolic compounds have a wide range of biochemical traits and may help prevent the onset of illnesses including cancer and cardiovascular diseases.

From Figure 2C, the statistical analysis show that temperature and duration of storage exhibited significant decrease of total phenolic content. The samples stored at 25 and 35°C induced significant decrease which occurred after 30 days with 11, 12% for S1 and 13, 15% for S2 of total phenolic content, respectively. These results corroborated with those of Pioana et al. (2011) and De Moura et al. (2012), who found that strawberry and blackberry jam's phenolic contents decreased after three months (20°C) and 180 days (10 and 20°C), respectively. In a similar vein, Rababah et al. (2011) found that orange jam that was kept at 25°C for five months had less total phenolics. Furthermore, our findings agreed with those of Daminiani et al. (2017), who reported that marolo jam's total polyphenol content greatly reduced after a year of storage at 25°C. Oxidation and polymerization reactions may be the main causes of the decrease in polyphenol content.

Total flavonoid compounds

Flavonoids, as one of the most diverse and prevalent classes of phenolics, play essential roles in the growth, development, and defense mechanisms of plants. These compounds display a number of chemical and biological activities and are known to have significant positive effects on human nutrition and health (Abd Ghafar et al. 2010).

Figure 2D shows the change of the flavonoid content of apple jam over storage. The total flavonoid content decreased during one month of storage at 25 and 35°C resulting in losses of 22 and 27% for S1, and 43 and 47% for S2, respectively. These results are consistent with those reported in previous studies on orange jam (Djaoudene and Louaileche 2016b). The study of Zafrilla et al. (2001) showed changes in flavonols content of strawberry jam stored for six months at 20°C. Furthermore, during 90 days of storage at room temperature, Igual et al. (2013) discovered that grapefruit jam lost 29 to 39 % of its flavonoids content.

The change in bioactive compounds during storage of jam depends on various factors like storage conditions (time, temperature, oxygen), residual enzyme activity, water activity, pH and product composition (sugars, pectin). Shinwari et al. (2018) reported that lower water activity, enzymes such as PPO and POD (peroxidases), and the pH of jam might be responsible for the degradation of anthocyanins through direct condensation reactions, or through the formation of hydroxymethyl furfurals (HMFs), as well as the hydrolysis of anthocyanin glycosides.

Influence of storage time and temperature on antioxidant capacity

Due to the complexity of phytochemicals, it is impossible to adequately assess the total antioxidant capacity using a single test. Consequently, numerous assays have been developed to assess the antioxidant activity. These techniques often assess an antioxidant's capacity to sequester metal ions, remove reactive oxygen species, or prevent lipid peroxidation. The antioxidant capacity of apple jam was assessed in the current study using two antioxidant activity assays (DPPH and FRP).

DPPH radical scavenging activity

The evolution of DPPH[•] scavenging activity of apple jam upon storage is shown in Table 2. It was found that storage conditions had obviously affected the DPPH[•] scavenging activity of two brands of apple jam. The DPPH[•] scavenging activity decreased significantly toward the end of storage; the initial capacity was reduced by 16 and 20% in S1, and 17 and 20% in S2 under 25 and 35°C, respectively. This trend is consistent with the changes observed for the antioxidant activity against DPPH reported in the study performed on jam made from blue honeysuckle berry, there was a significant decrease after 30 days of storage at 6°C (Kalisz et al. 2023). The findings of our investigation were also in line with those of Wicklund et al. (2005), who claimed that strawberry jam's antioxidant activity had decreased. Black carrot jam's radical scavenging activity dropped by 13–70% of its initial value when stored at 25°C (Kamiloglu et al. 2015). Rababah et al. (2011) noted a 50% loss in orange jam that had been kept at 25°C for five months. Furthermore, these results corroborated those of Istrali et al. (2013), who reported a decrease in DPPH[•] scavenging activity during 10 days of goji fruit jam storage.

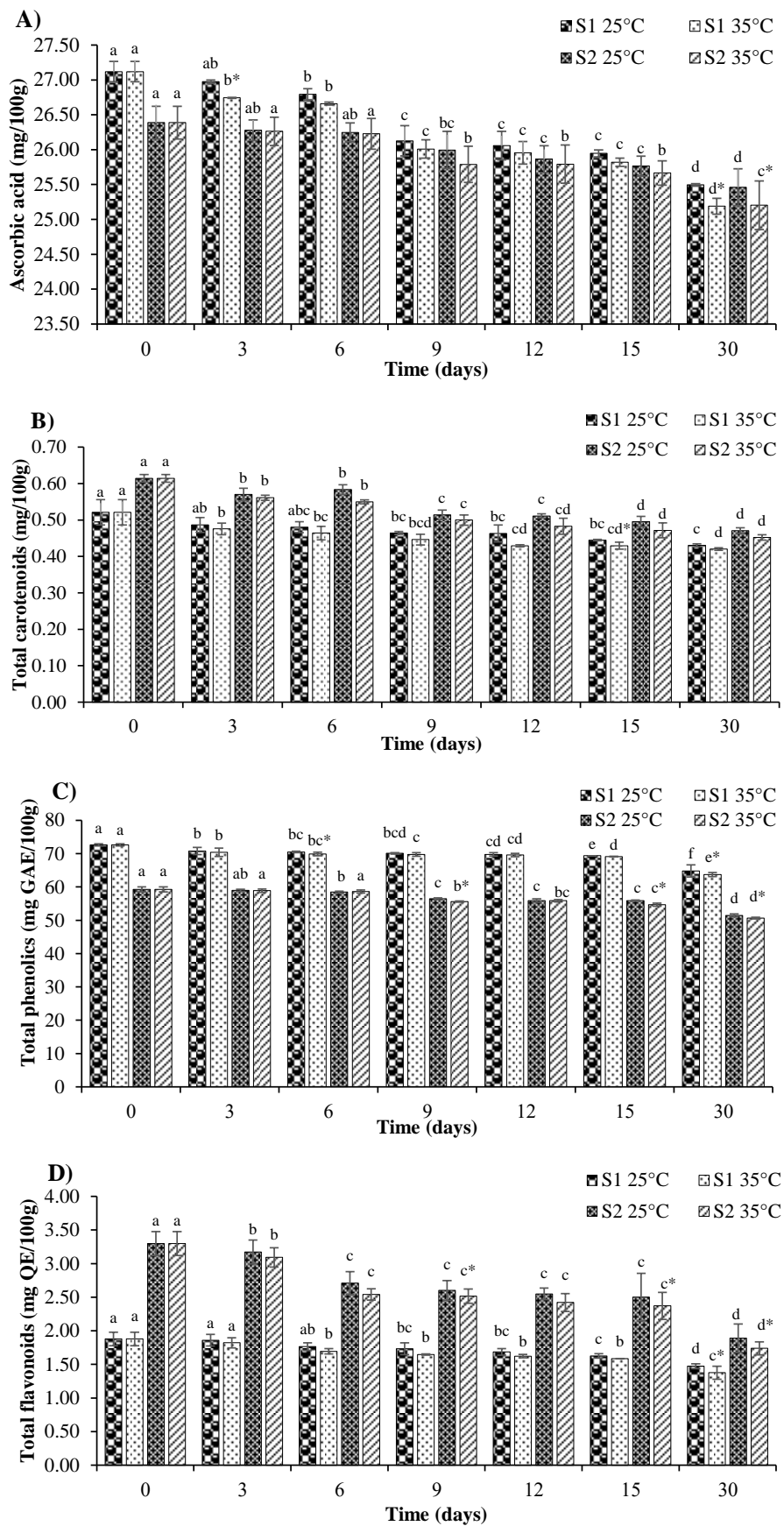


Figure 2. Influence of storage conditions (time and temperature) on total carotenoids (A), ascorbic acid content (B), total phenolics (C) and total flavonoids (D) of sample 1 (S1) and sample 2 (S2) apple jam.

In the same column, means \pm SD with the same letters (a–f) have no significant differences ($P \leq 0.05$); Asterisk (*) indicate significant differences ($P \leq 0.05$) between the results obtained at 25°C and 35°C; LSD interaction temperature-time of ascorbic acid, carotenoids, total phenolics and flavonoids of S1 and S2 are 0.28, 0.04, 0.69, 0.12 and 2, 0.3, 0.65, 0.25, respectively.

Table 2. Impact of storage conditions on antioxidant potential of apple jam.

Storage time (Days)	DPPH scavenging (mg GAE/100g)				FRP (mg GAE/100g)			
	S1		S2		S1		S2	
	25°C	35°C	25°C	35°C	25°C	35°C	25°C	35°C
0	4.44± 0.32 ^a	4.44± 0.32 ^a	5.46± 0.11 ^a	5.46± 0.11 ^a	38.89± 0.06 ^a	38.89± 0.06 ^a	29.69± 0.64 ^a	29.69± 0.65 ^a
3	4.32± 0.29 ^a	4.27± 0.32 ^{ab}	5.43± 0.09 ^{ab}	5.29± 0.05 ^{b*}	38.62± 0.12 ^{ab}	37.64± 0.24 ^{b*}	29.65± 0.31 ^b	29.58± 0.44 ^a
6	4.29± 0.26 ^{ab}	4.29± 0.28 ^{ab}	5.41± 0.03 ^{ab}	5.28± 0.04 ^{b*}	37.98± 0.60 ^{bc}	37.05± 0.20 ^{c*}	29.47± 0.25 ^{bc}	29.32± 0.26 ^a
9	4.08± 0.36 ^{bc}	4.05± 0.37 ^{bc}	5.34± 0.04 ^b	5.26± 0.06 ^b	37.81± 0.51 ^c	36.37± 0.29 ^{d*}	28.95± 0.44 ^c	29.25± 0.67 ^{a*}
12	4.05± 0.35 ^{cd}	4.00± 0.40 ^c	5.18± 0.03 ^c	5.13± 0.03 ^{c*}	36.89± 0.42 ^d	35.85± 0.17 ^{d*}	27.76± 0.30 ^c	27.88± 0.63 ^b
15	3.85± 0.20 ^{de}	3.74± 0.11 ^d	4.77± 0.10 ^d	4.67± 0.03 ^d	36.00± 0.86 ^e	35.23± 0.41 ^{e*}	26.98± 0.52 ^d	26.02± 0.19 ^{c*}
30	3.71± 0.17 ^e	3.57± 0.19 ^{d*}	4.51± 0.03 ^e	4.37± 0.08 ^{e*}	34.91± 1.39 ^f	33.88± 1.31 ^{f*}	24.63± 0.48 ^e	23.53± 0.37 ^{d*}

In the same column, means ± SD with the same letters (a–f) have no significant differences ($P \leq 0.05$); Asterisk (*) indicate significant differences ($P \leq 0.05$) between results obtained at 25°C and 35°C; LSD interaction temperature-time DPPH radical scavenging activity and reducing power of sample 1 (S1) and S2 are 0.22, 0.62 and 0.10, 0.67, respectively.

Ferric reducing power

The reducing capacity was another assay utilized to evaluate the apple jam’s antioxidant potential. The test relied on the presence of reductants, which lead to the reduction of the Fe³⁺/ferricyanide complex to the ferrous form; the Fe²⁺ was measured by observing the development of Perl’s Prussian blue at 700 nm.

Based on the results presented in Table 2, the analyzed jam exhibited an initial value of 39 and 30 mg GAE/100g for S1 and S2, respectively. After 30 days of storage, apple jam stored at 25 and 35°C exhibited a significant decrease; losses of 10 and 13% in S1 and 17 and 21% in S2, respectively. The results previously presented by Wicklund et al. (2005) and Poiana et al. (2012) concurred with our findings. Poiana et al. (2011) also reported that the reducing power of jams made with strawberries and sweet/sour cherries, decreased by 11 to 19.2%.

Correlations

The relationship between various antioxidant parameters was explored using linear regression analysis, as illustrated in Figure 3. The results revealed that total phenolics exert a highly significant influence on the antioxidant activities of apple jam, demonstrating a correlation coefficient of about 0.88. Notably, total flavonoids exhibited an even stronger and significantly positive correlation ($p < 0.001$) with both DPPH and FRP antioxidant activities, with correlation coefficients of 0.95 and 0.98, respectively. This underscores the paramount importance of flavonoids in contributing to the overall antioxidant capacity of the jam. Furthermore, strong relationships were observed between total phenolic and flavonoid contents and between DPPH and FRP activities. These findings were in line with earlier studies, reinforcing the robustness of the observed associations (Patras et al. 2011; Poiana et al. 2012; Rababah et al. 2011).

CONCLUSIONS

Daily storage conditions significantly affect the physicochemical properties and antioxidant capacity of apple jam, with higher temperatures exacerbating these changes. The study underscores the importance of prompt consumption and proper storage, particularly at lower temperatures, to

mitigate quality loss and preserve antioxidant parameters.

These findings provide valuable insights for consumers and manufacturers alike, highlighting the need for informed decisions regarding the handling and storage of apple jam to maintain its nutritional and health benefits over time.

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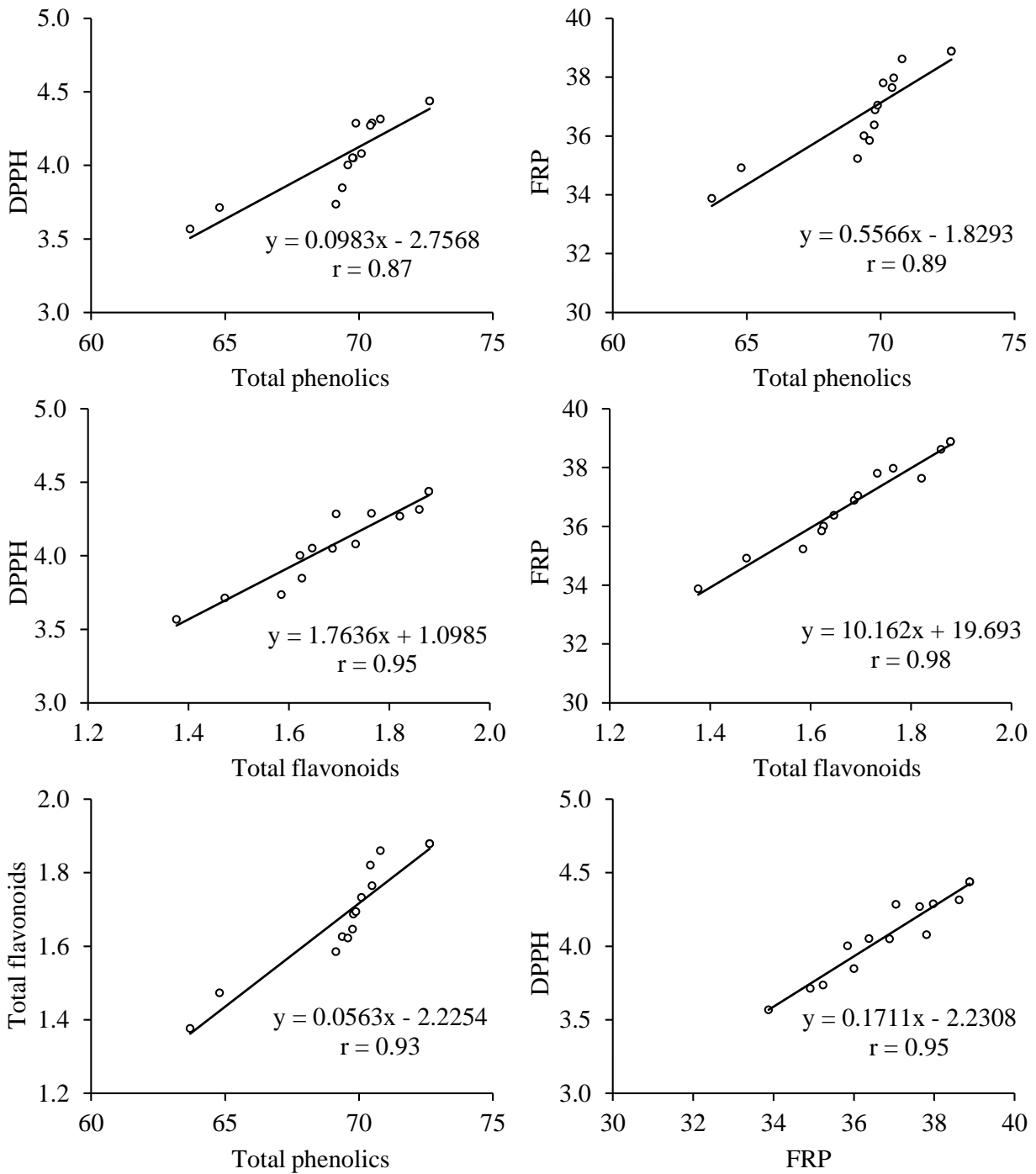


Figure 3. Correlations between antioxidant parameters of apple jam.
 ***: extremely significant correlation ($p < 0.001$).

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