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Antioxidant Capacity and Bioactive Ingredients of Asian Pear

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ABSTRACT

The combinations of soluble sugars, organic acids, and volatile organic compounds (VOCs) are crucial for how food is perceived and accepted. In order to evaluate the volatile organic compounds (VOCs) in Asian pears (*Pyrus pyrifolia*), headspace solid-phase microextraction (HS-SPME) was combined with gas chromatography-mass spectrometry (GC-MS) in this study. Among the 19 aroma compounds identified in the study conducted with a PDMS fiber, acetaldehyde and ethanol were found to be the most abundant. In addition, two more significant organic acids found in Asian pears were found to be malic acid (46.89 mg/100 g) and tartaric acid (45.08 mg/100 g). Glucose (84.70 mg/100 g) and sorbitol (65.75 mg/100 g) were identified in significant concentrations among the soluble sugars that directly affect fruit quality. LC-MS was used to investigate the phenolic content of Asian pears, and important phenolic compounds such as quinic acid (19227 g/L), chlorogenic acid (8445 g/L), procyanidin B2 (3146 g/L), liquiritin (435.1 g/L), and benzoic acid (363.1 g/L) were found.

Asya Armutunun Antioksidan Kapasitesi ve Biyoaktif İçerikleri

Araştırma Makalesi

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ÖZ

Suda çözünen şekerler, organik asitler ve uçucu organik bileşenlerin kombinasyonları, gıdanın algılanma ve kabul edilme süreçlerinde kritik bir rol oynamaktadır. Bu çalışmada, Asya armutları (*Pyrus pyrifolia*) üzerindeki uçucu organik bileşenleri değerlendirmek amacıyla headspace katı faz mikroekstraksiyonu (HS-SPME) yöntemi, gaz kromatografisi-kütle spektrometresi (GC-MS) ile birleştirilmiştir. PDMS fiber kullanılarak gerçekleştirilen çalışma sonucunda belirlenen 19 aroma bileşiği içinde, asetaldehit ve etanol en baskın bileşenler olarak saptanmıştır. Asya armutlarında belirlenen iki önemli organik asidin malik asit (46.89 mg/100 g) ve tartarik asit (45.08 mg/100 g) olduğu gözlemlenmiştir. Meyve kalitesini doğrudan etkileyen çözünür şekerler arasında ise glukoz (84.70 mg/100 g) ve sorbitol (65.75 mg/100 g) önemli konsantrasyonlarda tespit edilmiştir. Asya armutlarının fenolik içeriğini belirlemek için LC-MS kullanılmış ve kuinik asit (19227 g/L), klorojenik asit (8445 g/L), prosiyanidin B2 (3146 g/L), likiritin (435.1 g/L) ve benzoik asit (363.1 g/L) gibi önemli fenolik bileşenlere rastlanmıştır

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1. Introduction

The Asian pear, *Pyrus pyrifolia*, was originally cultivated in China and Japan (Jiang et al., 2016; Golias et al., 2021). Subsequently, it has expanded to Korea and Taiwan, and more recently, it is being grown in the United States, New Zealand, Australia (Golias et al., 2021). Furthermore, Asian pear production in Turkey's fertile lands and plains has been rapidly expanding in recent years. There are private growers in Konya, Çanakkale, and Uşak who cultivate nashi pears. (Yavuz and Parlak, 2018; Kayacan et al., 2022). During the maturation phase, different types of Asian pears display variations in shape, skin color, chemical composition, and ripening characteristics.

Because it is a juicy fruit, Asian pear is low in protein and fats but high in dietary fiber and sugars encompassing fructose, sucrose, and glucose. Additionally, Asian pear is a valuable fruit with health-enhancing properties, including C vitamin, bioactive substances, and minerals (calcium, magnesium, potassium, sodium, zinc, copper) (Yim and Nam, 2016; Jiang et al., 2018).

Consuming antioxidant-rich foods lowers the risk of chronic illnesses and oxidative stress. Fruits and vegetables are high in antioxidants and have health-promoting characteristics. Among such substances, you can find polyphenols, carotenoids, and triterpenoids (Kolniak-Ostek et al., 2020). Fruits and vegetables are reported to contain high amounts of dietary phenolics (Cui et al., 2005). Antioxidant, anti-inflammatory, antiviral, and anticarcinogenic effects are all possessed by phenolic substances (Kolniak-Ostek et al., 2020). The phenolic components of Asian pears have a high correlation with their antioxidant capacity. Asian pears are also susceptible to microbial and enzyme activity because of their high content of bioactive substances, carbohydrates, and moisture (Jiang et al., 2019). On the other hand, Asian pear peels, which are high in dietary fiber, have antioxidant properties as well (Jiang et al., 2016).

Aroma, which is a complex blend of aromatic volatile organic substances, contributes significantly to the flavor of fruits and vegetables in general and varies by fruit variety (Chen et al., 2018; Wang et al., 2019). Given that scent is among the most desirable fruit attributes, volatile organic molecules are probably going to have a significant impact on how customers perceive and accept products. Fruit volatile organic molecules can differ based on fruit variety, age, pre-harvest and post-harvest settings, environmental factors, and analytical methodologies used (Chen et al., 2018). The concentration and detection threshold for each of the volatile organic chemicals, as well as the combined amount of several volatile organic compounds, determine the distinctive scent of each fruit species (Seymour et al., 2012). Terpenoids, phenolic derivatives, lipid derivatives, and derivatives of amino acids are some of the most prominent volatile organic molecules (Schwab et al., 2008). Pears contain volatile substances such as alcohols, ketones, esters, aldehydes, and hydrocarbons. Esters are the primary volatile components in *P. ussuriensis* and *P. communis*, whereas aldehydes, alcohols, and esters are the primary volatile compounds in *P. pyrifolia*. C6 chemicals, which have been considered to be essential components in fruits, have been found in pears (Wang et al., 2019).

Organic acids and soluble sugars, which are important components of fruit quality, might influence fruit aroma. The main organic acids in fresh fruits are malic, citric, quinic, and tartaric acids (Wu et al., 2022). According to studies, organic acids help to extend the shelf life of fresh fruits by stabilizing anthocyanins (Ma et al., 2015). Additionally, it has been reported that citric acid chelates metal ions to avoid browning in fruit wine because of its high antioxidant potential (Tsegay, 2020). Moreover, organic acids that have antibacterial qualities and improve health, such as quinic acid, also affect the features of bitterness and acidity (Wu et al., 2003). As can be observed from the literature studies, organic acids are important to fruits, and the quantity and makeup of organic acids have a substantial impact on fruit aroma (Wu et al., 2022).

An investigation into the effects of various drying methods on the antioxidant potential and phenolic profile of Asian pear fruits cultivated in our country was conducted (Kayacan et al., 2022). The aim of this study, on the antioxidant potential, organic acids, sugars, aroma compounds and phenolic profile of Asian pear (*Pyrus pyrifolia*) fruits cultivated in our country were investigated. In this context, the volatile compounds of Pyrus pyrifolia were thoroughly examined using HS-SPME/GC-MS, and the phenolic compounds were analyzed utilizing LC-MS. Examining the characteristics of this fruit grown in Turkey's unique ecological conditions reveals the unique value of the study.

2. Material and Methods

2.1. Samples and chemicals

Asian pears (*Pyrus pyrifolia* Nashi) were sourced from a local farm in Kozan (Adana), Turkey, and stored at 4 ℃ until used (not more than one week). DPPH (2,2 Diphenyl-1- picrylhydrazyl), ABTS(2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt), $K_2O_8S_2$, DMPD (N,N-Dimethyl-p-phenylenediamine dihydrochloride), $FeCl_36H_2O$, $FeCl_24H_2O$ $CuCI_2$, NH_4SCN, are purchased from Sigma Aldrich. Also, *Escherichia coli* ATCC 11229 was obtained from Microbiologics.

2.2. Antioxidant capacity analyses

This analysis was carried out by making some changes to the method suggested by Tomasa et al. (2018). 5 grams of Asian pear were taken for antioxidant assay, and 5 mL methanol was added. The mixture was homogenized with a homogenizer at 1000 rpm. After 20 minutes in an ultrasonic bath at 25°C, it was centrifuged at 5000 rpm. Subsequently, the upper liquid phase was collected, and the precipitate was extracted again with 5 mL of methanol. Then, the higher liquid phases were mixed. Methanol was used to bring the total volume to 10 mL.

2.2.1. DPPH assay

The DPPH assay was completed using the method described by Makhlouf-Gafsi et al. (2018). A 100µM DPPH solution was prepared for antioxidant determination by the DPPH method. 3.9 mL of DPPH solution was added to each of the Asian pear samples and standards (Trolox and BHT), which

had been produced at different concentrations (2.5, 5.0, 10.0, 20.0, 40.0 μg/mL). The activity of radical scavenging was then measured at 517 nm. After measurement, the % inhibition was calculated by the following equation:

$$\% \text{ inhibition: } ((A_{control} - A_{sample})/A_{control})*100 \quad (1)$$

Then, IC_{50} values ((μg/ mL**)** concentration that inhibits 50% of the radical), Trolox and BHT equivalences were calculated from the calibration curve created with concentrations and % inhibitions. Results were also given as mg standard (Trolox and BHT) equivalent per 5 g sample.

2.2.2. ABTS assay

The Bursal et al. (2013) technique was used to perform the ABTS experiment. For the ABTS radical scavenging activity studies, a 2.45 mM $K_2S_2O_8$ solution and a 2 mM ABTS solution in phosphate buffer at pH 7.4 were combined for 6 hours. The radical solution was then added to different concentrations of Asian pear samples and standards (2.5, 5.0, 10.0, 20.0, 40.0 μg/ mL). After a 30 minute incubation, the absorbance at 734 nm was measured. Following measurement, percentage inhibition was calculated using equation 1. The calibration curve for concentrations and percentage inhibitions was used to generate IC_{50} values (concentrations that inhibit 50% of the radical), Trolox, and BHT equivalents.

2.2.3. CUPRAC assay

The CUPRAC test was performed utilizing the Apak et al. (2007) technique. To the samples prepared at various concentrations, 0.01 M 0.25 mL $CuCl_2$, 0.25 mL $7.5x10^{-3}$ M methanolic neocuproine and 1 M ammonium acetate solutions were added. The absorbance at 450 nm was measured following a half-hour incubation period. A calibration curve was created with concentrations corresponding to absorbance values.

Then, $A_{0.5}$ (concentration at 0.5 absorbance), and Trolox equivalent, BHT equivalent were calculated. Results were also given as mg standard equivalent per 5 g sample.

2.2.4. FRAP assay

The FRAP assay was performed following the method outlined by Gülçin (2012). The volumes of Asian pear and standards (BHT and Trolox), obtained at various concentrations (10.0, 20.0, 40.0, 60.0, 80.0 μg/mL), were adjusted to 0.5 mL using 0.3 M buffer (acetate) to assess antioxidants using the FRAP method. Subsequently, a 20 mM solution of $FeCl_3$ and the reagent for FRAP were added in sequence. After 10 minutes, the absorbances at 593 nm were measured. A calibration curve was generated using concentrations that corresponded to absorbance readings. Then, $A_{0.5}$ (concentration at

0.5 absorbance), Trolox equivalent, and BHT equivalent were calculated also given as mg standard equivalent per 5 g sample.

2.2.5. DMPD assay

The DMPD test was performed utilizing the Fogliano et al. (1999) technique. A radical cation ($DMPD^{\bullet+}$) was produced by mixing 0.1 M DMPD solution with 100 mL of buffer (pH: 5.3) to measure the $DMPD^{\bullet}$ radical scavenging activity of Asian pears. Then, 1 mL of the $DMPD^{\bullet+}$ solution was added to solutions prepared at different concentrations of Asian pear, BHT, and Trolox (10.0, 20.0, 40.0, 60.0, 80.0 µg/mL). After a 50 minute incubation at room temperature, the absorbances were taken at 505 nm. Results were calculated as mg standard (Trolox and BHT) equivalent per 5 g sample.

2.2.6. Total antioxidant analysis

The total antioxidant activity of Asian pear was found using the thiocyanate method (Yen and Chen, 1995). The volume of samples prepared at different concentrations was adjusted to 2.5 mL with buffer (pH: 7.4). Afterwards, 2.5 mL of 0.017 M linoleic acid emulsion was added to each tube. The tubes were then incubated in the dark at 37°C. Every 24 hours, 50 µl was taken from the test tubes and transferred to test tubes containing 2.35 mL of methanol. Then, 50 µl of Fe^{2+} solution and 50 µl of NH_4SCN solution were added sequentially. The absorbances at 500 nm of Asian pear, BHT, and Trolox solutions(10.0, 20.0, 40.0, 60.0, 80.0 µg/mL) were measured. In this method, the oxidation prevention capacity was determined as a percentage.

2.3. Organic acid analysis

The organic acids in Asian pear were determined using a modified approach based on Arzani et al., 2008. A Waters Alliance e2695 HPLC (High-Performance Liquid Chromatography) apparatus with a PDA (Photo-Diode Array) detector was used for the analysis at 210 nm. To prepare the samples, 5 grams of fruit were ground and then diluted with 20 mL of water. After filtration through a 0.45 µm filter, 5 mL of the filtrate was taken and diluted to 20 mL with 0.01 M $K_2HP0_4.3H_2O$ (0.01 M, pH = 2.6). The resulting solution was filtered again through a 0.45 µm filter. Ultimately, a sample volume of 20 µl was introduced into the HPLC with column conditions a flow rate of 0.7 mL at 30 degree.

2.4. Sugar analysis

Ramchoun et al.'s (2017) approach was modified to analyze the sugar content of Asian pears. Samples of 2 g were weighed and extracted with 20 mL of pure water for 30 min. After this, the mixture was centrifuged at 4 °C and 5500 rpm for 15 min. Then, the supernatant was filtered through a 0.45 µm pore size membrane filter before injection. The analysis was performed on an Alliance e2695 HPLC (High-Performance Liquid Chromatography) instrument. Refractive Index Detector (RID) and InertSustain NH_2 column (250 x 4.6 mm, 5 µm) were used in the analysis. The mobile phase used was

75/25 acetonitrile-water. The column temperature was set at 35 °C, the injection volume was 10 µl, and the flow rate was 1 mL/min (Ramchoun et al., 2017).

2.5. Phenolic compounds

LC-HRMS (Liquid Chromatography-High-Resolution Mass Spectrometry) analyses were carried out using a system consisting of a DIONEX UltiMate 3000 RS autosampler, pump, and a column oven. The mass spectrometry was conducted with an Exactive Plus Orbitrap system with a heated electrospray ionization interface from Thermo Fisher Scientific. Using an automated syringe injector (Thermo Fisher Scientific, USA), positive (Pierce™ LTQ Velos ESI Positive Ion Calibration Solution) and negative (Pierce™ Negative Ion Calibration Solution) calibration solutions were used to set the Orbitrap-MS apparatus. Samples of 150 g were weighed and extracted with 100 mL of methanol for 30 min. After this, the mixture was centrifuged at 4 °C and 5500 rpm for 15 min. Then, the supernatant was filtered through a 0.45 µm pore size membrane filter before injection. The analyses were performed using a Phenomenex® Gemini® 3µm NX-C18 110 Å column (100 mm × 2 mm), with a column oven temperature of 30 °C. The elution gradient comprised 2% (v/v) glacial acetic acid in the mobile phase A and methanol in the mobile phase B. The Orbitrap HRMS with a heated electrospray ionization interface, was operated in both positive (Full MS/AIF) and negative (Full MS/AIF) modes. The ionization interface settings included a sheath gas flow rate of 35, spray voltage of 3.5 kV, auxiliary gas flow rate of 7, capillary temperature of 350 °C, auxiliary gas temperature of 350 °C, and S-lens RF level set at 50. The mass spectrometry parameters included a search range of 60-800 m/z, a resolution of 17500, an automatic gain control (AGC) target of 3.106, a maximum ion trap time (IT) of 2 ms, and collision energy (CE) at 25 V. The total analysis time was set to 20 minutes, and each analysis was performed in triplicate (Ercan, 2024).

2.6. Aroma compounds with Headspace-SPME-GC/MS

The volatile fraction of asian pear was characterized using a protocol modified slightly from Silva and Camara (2013). In the analysis of aroma compounds, a "SHIMADZU QP2020" GC-MS instrument with a DB-HEAVYMAX capillary column (60 m x 0.25 mm x 0.25 um) was employed. The oven temperature was initially set at 40 °C, and the injector temperature was set to 250 °C. After a 2-minute hold, the temperature was raised by 3 °C per minute until it achieved 80 °C, where it was held for 1 minute. Then, the temperature was increased by 5 °C per minute until it achieved 240 °C, and it was held at this temperature for 6 minutes. The carrier gas, helium, was employed at a flow rate of 1.05 milliliters per minute. The temperature of the ion source was 230 °C, the temperature of the MS transfer line was 250 °C, and the ionization energy of the mass spectrometer was 70 eV. The extraction time for headspace sampling was 40 minutes, and the extraction temperature was set at 70 °C with a mixing speed of 250 rpm. A 100 µm PDMS fiber was used in the analysis.

2.7. Statistical Data Analysis

The data were subjected to one-way ANOVA by utilizing SPSS software (Version 24.0; SPSS Inc., Chicago, IL, USA).

3. Results and Discussion

3.1. Antioxidant capacity

In our study, the results of DPPH, ABTS, DMPD, CUPRAC, and FRAP assays in terms of Trolox equivalent were determined to be 52.44±0.03, 266.35±0.04, 1298.10±0.02, 333.72±0.01, and 159.86±0.02 mg Trolox/ 5 g sample, respectively (Table 1). When Kayacan et al. (2022) looked at how different drying methods affected the antioxidant capacity of Asian pears, they discovered that ultrasonic-assisted vacuum drying (USVD) outperformed hot air drying (HAD) and infrared drying (IRD) in terms of antioxidant capacity. According to their investigation, fresh samples had the highest DPPH and CUPRAC values (765.58 and 920.87 mg/TE 100g DM), whereas freeze-dried samples had the lowest findings (559.41 and 859.73 mg/TE 100g DM). The breakdown of phenolic components and a drop in the samples' antioxidant activity might have been caused by heat processes and oxidative reactions (Kim et al., 2021). In our study, DPPH and CUPRAC values were found to be slightly higher than in the study conducted by Kaycan et al (2022).

Table 1. Asian Pear's antioxidant capacity data (mg/5g samples)

Methods	**DPPH**	**ABTS**	**DMPD**	**CUPRAC**	**FRAP**	**Total Antioxidant Analysis**
Trolox Equivalent	52.44±0.03	266.35±0.04	1298.10±0.02	333.72±0.01	159.86±0.02	273.57±0.01
BHT equivalent	113.56±0.03	480.99±0.02	-	186.62±0.02	256.34±0.03	174.73±0.01

In this study, Table 2, Figure 1, and Figure 2 show the IC_{50} values (the concentration that scavenges 50% of the radical), $A_{0.5}$ values for FRAP and CUPRAC studies, and percentage inhibition values for the total antioxidant assay for *Pyrus pyrifolia* and the standards (BHT, Trolox). In the study conducted by Ma et al. (2012) on *Pyrus pyrifolia* cv. Punguoli, found that the IC_{50} value for pear peel extract's DPPH radical scavenging activity was 38.3 mg/mL, which was significantly higher than the highest concentration of 50 mg/mL used for the meat extract. Lee et al. (2015), investigated the antioxidant activity differences between fruit peel and pulp in *Pyrus pyrifolia* Nakai cv. Chuhwangbae and Niikata species found that the DPPH radical scavenging effect was higher in the fruit peel. When ABTS, FRAP, DPPH, CUPRAC and total antioxidant values of Asian Pear and standards (Bht and trolox) were compared with one-way ANOVA, $p<0.5$ was found.

Table 2. IC_{50} and $A_{0.5}$ values of Asian Pear, BHT, and Trolox are given in Table 2.

	IC_{50} (µg/mL)			$A_{0.5}$ (µg/ mL)		% reduction (at 72 hour)
Sample and Standarts	**DPPH**	**ABTS**	**DMPD**	**CUPRAC**	**FRAP**	**Total Antioxidant**
Asian Pear	420.05±1.9	55.72±1.5	33.00±6.0	56.18±0.01	84.44±0.01	15.53±0.06
BHT	47.70±0.1	26.79±0.5	-	10.48±0.09	21.64±0.05	88.86±0.01
Trolox	22.02±0.2	14.84±0.1	42.14±2.0	18.75±0.08	13.50±0.02	56.75±0.03

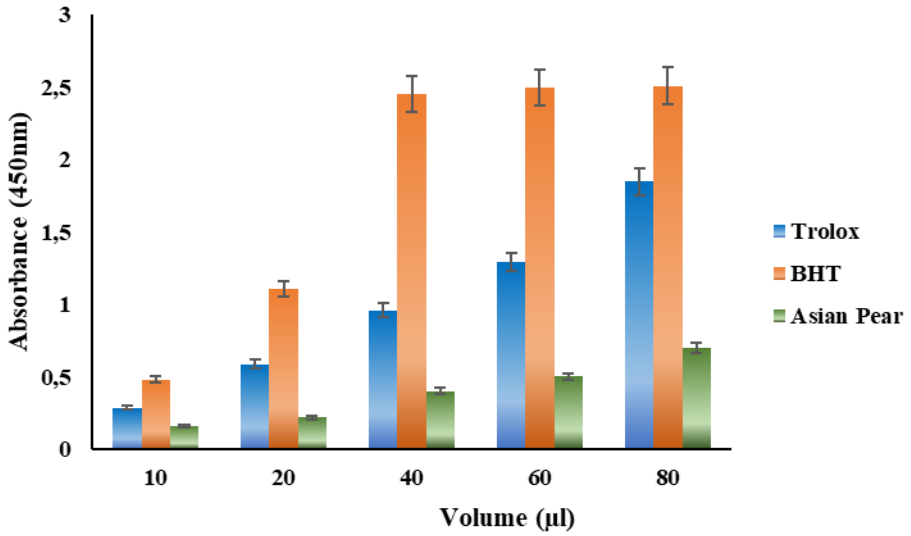

Figure 1. CUPRAC test results of Asian Pear (500 mg/ mL concentration) and standards (1 mg/ mL concentration)

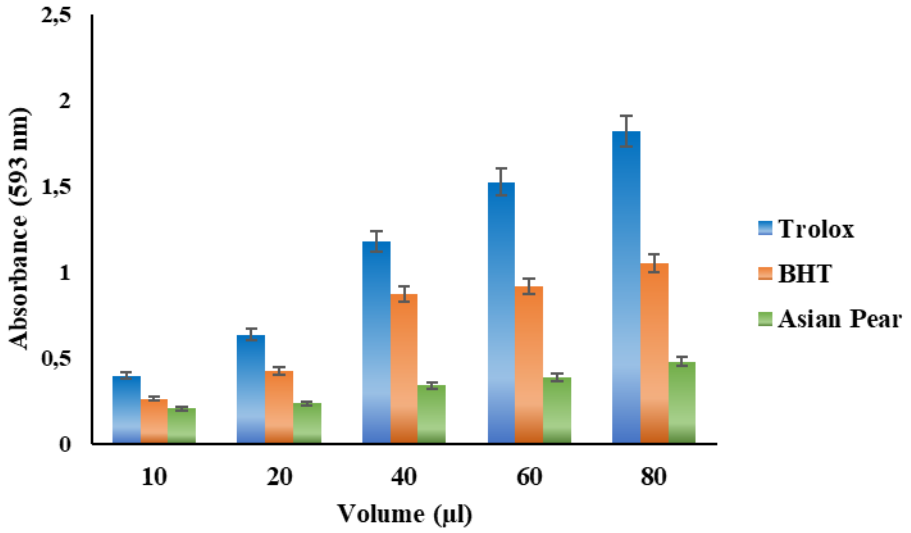

Figure 2. FRAP test results of Asian Pear (500 mg/ mL concentration) and standards (1 mg/ mL concentration)

3.2. Organic acids

Fruit taste and quality are largely influenced by the amount and ratio of organic acids present (Zhang et al., 2012). Fruit species typically differ in their organic acid concentration and makeup. For

example, while malic acid is the primary acid in apples, kiwifruit contains a high amount of citric acid, and watermelon primarily contains malic, citric, and oxalic acids (Wu et al., 2022).

According to our investigation, we determined that the organic acid content was in the following order: malic acid > tartaric acid > succinic acid > quinic acid > citric acid > oxalic acid. In this study, malic acid (46.89 mg/100 g) and tartaric acid (45.08 mg/100 g) were determined to be the significant major components (Table 3). Duan et al. (2020) identified eight organic acids in six different *Pyrus pyrifolia* varieties (Cuiyu, Xuefeng, Hosui, Kousui, Wonhuwang, Hwangkumbae. These compounds were ranked as malic acid > citric acid > quinic acid > oxalic acid > shikimic acid > lactic acid > fumaric acid > maleic acid. Another study by Sha et al. (2011) identified a total of ten organic acids in 10 varieties of P. pyrifolia. Malic, citric, quinic, oxalic, shikimic, and fumaric acids were discovered in all 10 varieties, while tartaric acid was found in 2 varieties, and acetic acid was only found in 1 variety. In their study, malic acid and citric acid were determined to be the important major components. The range of malic acid was 0.61 to 2.11 mg/g, and citric acid was found at levels of 0.36 to 1.48 mg/g. The fruit included quinic as a minor organic acid (ranging from 0.12 to 0.44 mg/g) and oxalic acids (ranging from 0.01 to 0.17 mg/g). Acetic, shikimic, succinic, fumaric, tartaric, and lactic acids were found to have comparatively low content.

Table 3. The organic acid analysis result of Asian pears (mg/100 g)

Sample	Malic	Tartaric	Citric	Oxalic	Quinic	Succinic
Asian pear	46.89±0.71	45.08±0.12	12.47±0.08	10.05±0.04	15.58±0.12	35.12±0.30

3.3. Sugar

Sugars have a direct impact on fruit quality and perform many significant roles in plant metabolism. The fruit's soluble sugar content has a major impact on the quality of pears (Kou et al., 2016). Pear fruit contains four different kinds of sugars: sorbitol, fructose, glucose, and sucrose (Pilando and Wrolstad, 1992). Among the most significant sugars in fruits are sucrose, glucose, and fructose, which are produced in photosynthetic source tissues (Kou et al., 2016). The composition and total amount of sucrose, fructose, and glucose directly affect the perceived sweetness of the fruit. The total sugar concentration is one of the important markers of fruit ripening, which is a complicated process (Pilando and Wrolstad, 1992).

Table 4. The sugar analysis result of Asian pears (mg/100 g)

	Fructose	Glucose	Sorbitol	Sucrose
Asian pear	35.29±0.89	84.7±1.03	65.75±0.96	10.61±0.10

In your study, the sugar ranking was determined to be glucose > sorbitol > fructose > sucrose (Table 4). Duan et al. (2020) conducted a study on six different varieties of *Pyrus pyrifolia* (Cuiyu, Xuefeng, Hosui, Kousui, Wonhuwang, Hwangkumbae) and found that the ranking of soluble sugars was

fructose > sorbitol > glucose > sucrose. Yim et al. (2016), in their study of ten varieties from four species grown in Korea (*Pyrus bretschneideri, Pyrus ussuriensis, Pyrus pyrifolia* and *Pyrus communis*), found that fructose content to be more abundant than other sugars in all four species (ranging from 4.21-6.80 g/100 g FW). They also noted variations in the ranking of soluble sugars within different *Pyrus pyrifolia* varieties, such as fructose > sucrose > glucose > sorbitol in Wonwhang, fructose > glucose > sucrose > sorbitol in Niikata, fructose > sorbitol > glucose > sucrose in Hanreum, and fructose > glucose > sorbitol > sucrose in Chuwhang Jiang et al. (2023) conducted a study on *Pyrus pyrifolia* during fruit ripening, performing QTL mapping and transcriptome analysis of sugar content. They discovered that fructose (varying from 35.4% to 45.7%) made up the greatest fraction of all sugars, with sorbitol (ranging from 28.5% to 32.8%) coming in second. Similar to our study, they reported that sucrose content was the lowest (ranging from 2.7% to 17.5%). They also noted a significant accumulation of sucrose and fructose in sweet varieties and a significant decrease in sorbitol content during fruit ripening. Durán-Soria et al. (2020) conducted another study and found that glucose content increased gradually until maturity. In our study, glucose has the highest proportion. The fruit's overall sugar content and the proportion of each type of sugar determine how sweet it is (Jiang et al., 2023).

3.4. Phenolic compounds

Asian pears' phenolic components have a substantial correlation with their antioxidant properties (Jiang et al., 2019). In the results obtained in our study, major phenolic compounds were identified as quinic acid (19227 µg/L), chlorogenic acid (8445 µg/L), procyanidin B2 (3146 µg/L), liquiritin (435.1 µg/L), and benzoic acid (363.1 µg/L) (Table 5).

In the study by Kayacan et al. (2022), they identified chlorogenic acid (18.65 mg/L) as the main phenolic compound. Chlorogenic acid was followed by epicatechin (10.60 mg/L), trans-cinnamic acid (3.60 mg/L), and 3,4-dimethoxybenzaldehyde (3.60 mg/L). While they found 2.31 mg/L of catechin, your study detected 31.50 µg/L of catechin. In a study by Jian et al. (2019) on *Pyrus pyrifolia* Nakai (Asian pear), they detected arbutin (122.0 – 94.00 mg/100 g), chlorogenic acid (10.02-9.01 mg/100 g), and p-coumaric acid (20.01-18.68 mg/100 g). In another study by Cui et al. (2005) on various varieties of *Pyrus pyrifolia* (Huangjin, Xinxue, Xueli, Daguo Shuijing, and Housui), they found varying amounts of chlorogenic acid in the skin (ranging from 0.066 mg/g to 0.522 mg/g), in the core (ranging from 0.163 mg/g to 2.730 mg/g), and in the flesh (ranging from 0.004 mg/g to 0.096 mg/g). In pears, chlorogenic acid is thought to be the most significant antioxidant-active ingredient. Recent research has reported its physiological activities, including anticancer activity, reducing the harmful effects of chemotherapy drugs, immune system enhancement, affecting the sleep-wake cycle, and antioxidant capacity (Cui et al., 2005).

In the study by Zhang et al. (2007), which examined the impact of growth on phenols in three Asian pear varieties (Hosui, Niitaka, and Chuwhangbae), arbutin was found to be high in the early growth

stage in the seed, flesh, and skin. Arbutin, chlorogenic acid, and epicatechin were detected in all three varieties during early growth. Caffeic acid, catechin, and 4-hydroxybenzoic acid were found in the seed and skin during ripening. Similarly, in our study, catechin and 4-hydroxybenzoic acid were detected. Additionally, in this study, procyanidin B2 was the most abundant compound after quinic acid and chlorogenic acid. Similarly, Jeong et al. (2012) isolated and identified procyanidin B2 from *Pyrus pyrifolia* Nakai cv. Chuhwangbae. Proanthocyanidins are found in lots of food resources, including fruits, nuts, and beans, and numerous biological benefits, such as antioxidant, antibacterial, antiallergic, anticancer, and anti-obesity properties, have been described for them (Jeong et al., 2017). Liquiritin (4′,7-Dihydroxyflavanone 4′-glucoside) and narcissin are two other important compounds identified in this study. Liquiritin, a flavonoid compound, has been found to exhibit several pharmacological actions, including anti-myocardial fibrosis, antioxidant, and neuroprotective properties (Zhai et al., 2019).

Table 5. Phenolic compound analysis results by LCMS.

No	Target Compounds	Quantification (μg/L)
1	Quinic acid	19227±150
2	Taxifolin	30.5±1.02
3	Protocatechuic acid	35.5±3.00
4	Protocatechuic aldehyde	25.9±1.96
5	Esculin hydrate	11.4±0.51
6	4-Hydroxybenzoic acid	16.8±1.20
7	Catechin	31.5±2.37
8	Gentisic acid	14.9±0.99
9	Procyanidin B2	3146±26.00
10	3-hydroxyphenylacetic acid	11.6±1.68
11	Chlorogenic acid	8445±43.00
12	3-hydroxybenzoic acid	11.2±0.89
13	2,4-dihydroxybenzoic acid	3.7±0.05
14	Vanillic acid	119±11.02
15	3,4-Dihydroxyphenylacetic acid	120±15.00
16	Ethyl gallate	4.8±0.75
17	Vicenin 2	29.1±2.03
18	Daidzin	2.9±0.48
19	Vanillin	6.9±0.87
20	Luteolin 7-glucoside	6.7±0.21
21	*m*-Coumaric acid	51.3±1.58
22	Ferulic acid	86.0±3.20
23	Sinapic acid	3.5±0.34
24	Liquiritin (4′,7-Dihydroxyflavanone 4′-glucoside)	435±12.02
25	Benzoic acid	363±10.11
26	Hesperidin	21.5±0.97

27	Neohesperidin	21.7±1.03
28	Naringin	19.8±2.00
29	Eriodictyol (3,4,5,7-Tetrahydroxyflavanone)	11.6±0.54
30	Rutin hydrate	43.4±2.14
31	Phloridzin	0.46±0.00
32	Rosmarinic acid	0.45±0.01
33	Nicotiflorin (Kaempferol 3-O-β–rutinoside)	5.7±0.38
34	Astragalin (Kaempferol 3-glucoside)	3.7±0.05
35	Kuromanine (Cyanidin 3-glucoside chloride)	3.8±0.02
36	Salicylic acid	15.9±1.30
38	Hyperoside (Quercetin 3-D-galactoside)	59.3±3.82
39	Isoquercitrin (Quercetin 3-glucoside)	59.3±3.21
40	Narcissin (Isorhamnetin 3-rutinoside)	274±11.02
41	Luteolin	0.8±0.00
42	Quercetin	1.5±0.45
43	Isorhamnetin (Quercetin 3′-methyl ether)	15.4±0.28

Narcissin, a natural flavonoid obtained from edible and traditional medicinal herbs, has been found to have a variety of biological roles and possible therapeutic benefits on illnesses such as hypertension, cancer, and Alzheimer's (Gao et al., 2023). In a study by Zhang et al. (2006) on three Asian pear varieties, they were unable to detect ferulic acid and sinapic acid. In our study, ferulic acid was detected at 86.0 μg/L, and sinapic acid was detected at 3.5 μg/L.

3.5. Aroma compounds

One of the most significant sensory quality factors for fruits and fruit products is aroma compounds (Jiang et al., 2019). In our study, 19 aroma compounds were identified, as illustrated in Table 6 and Figure 3. The most common substances were aldehydes (45.3%), esters (24.11%), and alcohols (27.66%). While esters are the main volatile chemicals in *P. communis* and *P. ussuriensis*, aldehydes predominate in *P. pyrifolia*, followed by alcohols and esters (Wang et al., 2019).

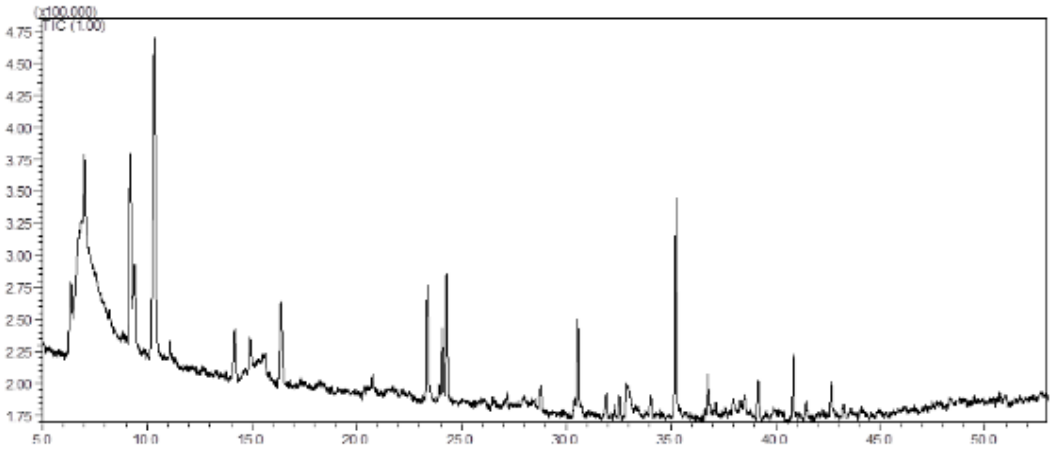

Figure 3. Volatile component analysis chromatography by GC-MS

In our study, acetaldehyde (27.46%) and ethanol (21.87%) were detected in the highest concentrations. Similarly, in a study by Golias et al. (2022) on 'Conference' and 'Yali' pear varieties, they found very high concentrations of both acetaldehyde and ethanol in both varieties. Jiang et al. (2019) also detected 18,30% ethanol in freeze-dried powdered of Asian pear and 8.37% ethanol in hot air-dried powdered of Asian pear. Acetaldehyde is naturally found in coffee, bread, and ripe fruits and is produced by plants (Sadighara et al., 2020). Compounds like acetaldehyde, known as C6 compounds, are considered green leaf volatiles contribute to the herbaceous aroma of fruits (Wang et al., 2019). Additionally, in a study by Golias et al. (2021) on various *Pyrus pyrifolia* Nakai varieties, they also detected higher levels of aldehydes, alcohols, and ester compounds. In our study, it was observed that the majority of ester compounds were composed of ethyl acetate. Similarly, in a study by Wang et al. (2019), they found that high concentrations of acetates were the main ester components. They also found that ethanol was the dominant compound among alcohols.

The activity of many key enzymes (lipoxygenase, pyruvate decarboxylase, alcohol dehydrogenase, and alcohol O-acetyltransferase) might change greatly due to variable shelf life or storage circumstances in the production of volatile aroma molecules in pear fruit (Lara et al., 2003).

Table 6. Volatile component results of Asian pear

LRI	Volatile Compounds	Peak Area %	LRI	Volatile Compounds	Peak Area %
381	Acetaldehyde	27.46±1.01	1420	Ethyl octanoate	0.92±0.02
577	Ethyl Acetate	9.57±0.95	1498	Decanal	1.03±0.01
888	Methanol	4.88±0.01	1519	(*E*)-2-Nonenal	9.24±0.04
926	Ethanol	21.87±0.05	1591	(*E*,*Z*)-2,6-Nonadienal	2.30±0.01
1064	Ethyl 2-methyl butanoate	1.59±0.01	1605	Methyl benzoate	0.74±0.00
1172	Ethyl 2-butanoate	0.81±0.01	1666	2-Furanmethanol	0.91±0.01
1200	2-pentyl furan	0.69±0.00	1677	Ethyl benzoate	1.95±0.03
1220	Ethyl hexanoate	2.99±0.02	1725	α-Farnesene	2.23±0.03
1245	Ethyl tiglate	5.54±0.12	1797	(*E*,*E*)-2,4-Decadienal	1.44±0.06
1380	Nonanal	3.84±0.08			

3.6. Antimicrobial Activity

In our study, the pear extract at a concentration of 100 mg/ mL inhibited an 8.0 mm inhibition zone on *E. coli*, a gram-negative bacterium belonging to the Enterobacteriaceae family. In comparison, the positive control, rifampin (at 5 µg/ mL), created an 11.5 mm inhibition zone *E. coli* is a pathogenic species known to cause intestinal infections and other infections (Omerovic M et al., 2017). In our study, Asian pear extract at 100 mg/ mL showed a 69.56% inhibition effect when compared to rifampin at 5 µg/ mL (Figure 4). This highlights the antimicrobial properties of a natural fruit, taking into consideration the side effects of antibiotics.

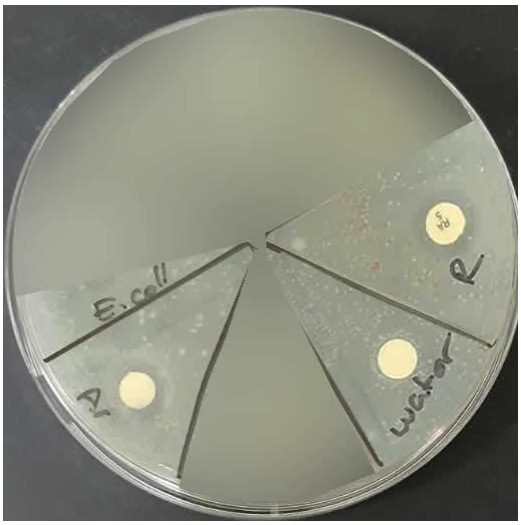

Figure 4. Antimicrobial activity of Asian pear

4. Conclusion

Aroma, volatile organic compounds, organic acid compositions, and sugar blends are the primary components that influence the intricate composition of food acceptance and perception. Within this complex tapestry, certain compounds emerge as pivotal players. In this research, the distinct effect of certain compounds stands out. While acetaldehyde and ethanol stand out as the aroma compounds found in the highest amounts in this comprehensive study, the presence of important organic acids such as malic acid and tartaric acid in Asian pear is also noteworthy. Nonetheless, it has been found that significant sugars like sorbitol and glucose, which have a direct impact on fruit quality, are present in significant amounts. When an in-depth examination is made on the composition of Asian pear, the presence of major phenolic compounds such as quinic acid, chlorogenic acid, procyanidin B2 and liquiritin becomes evident. These thorough research results illuminate the complex interactions between volatile chemicals and phenolic compounds, adding to our understanding of the subject while also acting as a beacon of guidance for future investigations. Also, a comprehensive approach like this has the potential to progress the fields of food innovation and flavor science, which will ultimately improve customer pleasure and gastronomic experiences.

Author's Contribution

The contributions of the authors are equal.

Conflict of Interest

The authors have declared there are no conflict of interest.

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