chemistry proceedings

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Proceeding Paper

Quince Pomace: A Source of Fiber Products and Polyphenols †

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† Presented at the Exploratory Workshop "Innovative Cross-Sectoral Technologies", Vth Edition, Bucharest, Romania, 22–23 May 2023, Secvent project meeting.

Abstract: Quince fruits are used to make jams, jellies, and juices, resulting in by-products like seeds, pulp, and skin. These by-products contain bioactive compounds such as phenolic acids, flavonoids, and polysaccharides, which offer potential health benefits. Utilizing these by-products to produce fiber-enriched powders and polyphenol-enriched extracts can yield valuable nutrient-rich biomass and reduce waste. This study aimed to assess the potential of quince pomace for producing fiber-enriched powders and polyphenol-enriched extracts, for which this substance's chemical composition, properties, and antioxidant activity were examined. Extraction with water or ethanol was employed, and the polyphenol content in the supernatant was analyzed using high-pressure liquid chromatography (HPLC).

Keywords: extraction; water; ethanol; high-pressure liquid chromatography

Citation: Dimitriu, L.; Preda, D.; Constantinescu-Aruxandei, D.; Oancea, F. Quince Pomace: A Source of Fiber Products and Polyphenols. *Chem. Proc.* **2023**, *13*, 6. https://doi.org/10.3390/chemproc2023013006

Academic Editors: Mihaela Doni and Radu Claudiu Fierăscu

Published: 8 September 2023

1. Introduction

Quince fruits (*Cydonia oblonga*) are used in the production of jams, jellies, preserves, and juices. The industrial processing of quinces generates by-products such as quince seeds, quince pulp, and quince skin, which can be utilized in various fields. Various studies [1–3] have shown that quince fruits are rich sources of bioactive compounds, including phenolic acids, flavonoids, organic acids, and polysaccharides, which have diverse beneficial effects on human health. The by-products obtained from the industrial processing of vegetables and fruits present a major global issue as they transform into waste. However, their utilization as value-added products can contribute to solving this problem and aid in the recovery of valuable nutrient-rich biomass, such as dietary fibers and polyphenols [4]. Dietary fibers from various plant sources are widely recognized for their benefits on the human intestinal tract. These fibers extracted from different sources or using different methods can exhibit varying metabolic and physiological positive effects [5].

Previous studies have investigated the properties of the fiber-rich products from quince (*Cydonia oblonga*) wastes after aqueous or ethanol extraction, but they did not evaluate the properties and possible applications of the resulting extracts [6,7]. A recent study analyzed the acetone extraction of *C. oblonga* fruits, but it did not characterize the resulting substrate [3]. Both extracts and the resulting substrates have been characterized for the peels of Cydonia oblonga but not of other by-products [1,8]. These peel extracts were shown to be rich in polyphenols and have significant antioxidant activity, and the substrate was found to be rich in fiber. Polyphenols are a class of bioactive compounds found in plant

cell walls. They exhibit numerous positive health effects, potentially offering protection against oxidative-stress-induced damage and diseases [9].

The objective of this study was to assess the potential of utilizing quince pomace for producing fiber-enriched powders with beneficial chemical compositions and, at the same time, evaluate the quince pomace extracts obtained by producing fiber-enriched powders. To achieve this, two different fiber-enriched powders and two extract types were obtained through extraction using water or ethanol as solvent. The pomace represents the by-product containing the pulp, seeds, and peel resulting from the processing of quince or other fruit in the food industry. The powders were analyzed for their chemical composition, properties, and antioxidant activity, and the supernatant was analyzed for polyphenol content using HPLC.

2. Materials and Methods

2.1. Material

Quince pomace obtained from the industrial processing of quinces (Qubio Cert, Izvoarele, Romania), which contains seeds, pulp, and peel, was lyophilized and ground with the help of an electric grinder.

The following chemicals were used: Trolox, 97% (Acros Organics, Thermo Fisher Scientific, Pittsburgh, PA, USA); 2,2-Diphenyl-1-picrylhydrazyl (Sigma-Aldrich, Merck Group, Darmstadt, Germany); 2,20-Azino-bis (3-ethylbenzothiazo-line-6-sulfonic acid) diammonium salt, 98%; 2,4,6-tri(2-pyridyl-1,3,5-triazine), 98% (Alfa Aesar, Kandel, Germany); Folin–Ciocalteu's phenol reagent; iron chloride (III) (Merck, Darmstadt, Germany); hydrochloric acid; acetic acid (Chimopar Srl, Bucharest, Romania); sodium acetate; glucose (Scharlau, Barcelona, Spain); HPLC standards, namely, ferulic acid, p-coumaric acid, caffeic acid, quercetin dihydrate (Sigma-Aldrich, Merck Group, Darmstadt, Germany), syringic acid, luteolin, (+)-rutin trihydrate, (Alfa Aesar, Haverhill, MA, USA), apigenin, (−)-epicatechin (Roth, Karlsruhe, Germany), and kaempferol; chlorogenic acid; and myricetin (Cayman Chemical, Ann Arbor, MI, USA).

2.2. Sample Preparation

This study involved the preparation of fiber-enriched samples and extracts rich in bioactives through an adapted version of a method reported in [6]. Two types of samples were prepared: samples prepared via extraction with ethanol (extract: E_QPE and substrate: S_QPE) and those prepared via extraction with water (extract: E_QPW and substrate: S_QPW). For the QPE sample, the quince pomace was mixed with 96% ethanol in a 1:5 ratio (w/v) and subjected to reflux boiling at 80 °C for 30 min. After filtration, the extractable compounds were analyzed using HPLC. For the QPW sample, the quince pomace was mixed with double-distilled water (ddH_2O) in a 1:5 ratio (w/v) and stirred at 50 °C for 30 min. After filtration, the sediment from each extraction was dried, ground, and characterized.

2.3. HPLC Analysis

High-Performance Liquid Chromatography (HPLC) analysis was conducted to identify phenolic acids and flavonoids in the alcoholic and aqueous supernatants obtained from the quince pomace using the treatment procedures mentioned earlier. The analysis was performed using a Dionex Ultimate 3000 system, and the chromatograms were processed with Chromelleon 7.0 software. Chromatographic separation was carried out using a Luna Omega 5 µm Polar C18 100 Å column (250 mm × 4.6 mm).

The phenolic acids were analyzed according to the methods described in [10] with some modifications. This analysis utilized a gradient program with a two-solvent system: A—0.1% aqueous formic acid solution and B—methanol. The following gradient program was applied: 0–25 min, 5% B; 25–33 min, 30% B; 34–40 min, 5% B. The flow rate was set to 1.25 mL/min, and a 10 µL injection volume was used for detecting the phenolic acids at 280 nm.

Flavonoids were identified and quantified following the method outlined in [11]. The analysis involved a gradient elution of two solvents: methanol (solvent A) and 0.5% H_3PO_4 (solvent B). The elution program was set as follows: 0–10 min, 15% A/85% B; 15–25 min, 85% A/15% B; 25–30 min, 60% A/40% B. The flow rate of the mobile phase was 1.5 mL/min, the column temperature was maintained at 25 °C, and the flavonoids were detected at 280 nm.

2.4. Physico-Chemical Characterization of Quince-Fiber-Enriched Powder

The swelling capacity (SC) of quince fiber samples was analyzed using the adapted method described in [6]. Briefly, 0.2 g of dried sample was placed in a tube, and 10 mL of ddH_2O was added; then, the sample was left to hydrate for 18 h. The final volume reached by the sample after 18 h was measured. The swelling capacity was calculated using the equation given below:

$$\text{SC (mL/g)} = \frac{\text{volume occupied by the sample}}{\text{initial sample weight}}. \quad (1)$$

Water retention capacity (WRC) represents the amount of water bound to hydrated fibers after the application of an external force (such as centrifugal force). A WRC analysis was performed following the method described in [12]. This method involved weighing 1.000 g of the sample, which was then placed in a tube, followed by the addition of 30 mL of distilled water. After 18 h of hydration, the sample was centrifuged, and the supernatant was separated. The wet residue was then weighed and dried at 100 °C for 2 h.

$$\text{WRC} = \frac{\text{weight of wet residue} - \text{weight of dry residue}}{\text{weight of dry residue}}. \quad (2)$$

2.5. Determination of Lignin and Polysaccharide Content

The two fiber-enriched substrates (S_QPE and S_QPW) and the raw quince pomace sample (QPU) were used to determine the content of cellulose, lignin (as described in [6]), and total carbohydrates. For the determination of lignin content, a gravimetric method was employed after the acid hydrolysis of cellulose and non-cellulosic polysaccharides. A sample weighing 0.6 g was treated with 4.16 mL of 72% sulfuric acid and continuously agitated for 3 h at room temperature.

Then, water was added to reach a concentration of 1 M of sulfuric acid (final volume of 25 mL). The prepared sample was heated at 100 °C for 2.5 h. After cooling, the supernatant was separated to analyze the total carbohydrate content using the phenol–sulfuric acid method described below. The remaining residue was washed with distilled water, dried, weighed, and reported as the total lignin content. In the second procedure, 0.6 g of the sample was mixed with 4.16 mL of 72% sulfuric acid, followed by immediate addition of water to reach a concentration of 1 M sulfuric acid (final volume of 25 mL), and the mixture was heated at 100 °C for 2.5 h. After cooling, the supernatant was separated to analyze the total non-cellulose polysaccharide content described below, and the residue was washed, dried, weighed, and reported as the total cellulose + lignin content. The cellulose content was calculated as (cellulose + lignin) − lignin.

The Total Carbohydrate and Non-Cellulose Polysaccharides

The total carbohydrate and non-cellulosic carbohydrate content was determined using the phenol–sulfuric acid method described in [13] with some modifications. In this method, 25 µL of the sample or standard solution was mixed with 25 µL of 5% phenol (in H_2O), followed by the addition of 125 µL H_2SO_4. The prepared samples were then incubated on a Digital heat block (BenchMark, Sayreville, NJ, USA) at 95 °C for 15 min. The calibration curve was constructed using a glucose solution of 1 mg/mL for the total carbohydrate content, covering concentrations from 50 to 600 µg/mL. For non-cellulosic carbohydrates, a calibration curve was constructed using a xylose solution of

1 mg/mL, covering concentrations from 50 to 400 µg/mL. The results were expressed as mg glucose/xylose equivalent/100 g of sample.

2.6. Total Polyphenol Content and Antioxidant Activities

To determine the total polyphenol content and the antioxidant activity of the substrates, 1 g of each sample was mixed with 10 mL of 50% ethanol. Subsequently, the mixture underwent ultrasonication in an ultrasonic bath for 30 min. After ultrasonication, the samples were centrifuged (Hettich Universal 320R, Hettich, Bäch, Switzerland) at 7350 RCF for 30 min to separate the supernatant, which was then subjected to further analysis.

2.6.1. Total Polyphenol Content

Total Polyphenol Content (TPC) was measured using the Folin–Ciocalteau method. The extracts were analyzed spectrophotometrically, following the procedure described in [14]. The samples or standard solutions of gallic acid were mixed with the reagents, and absorbance values were recorded at 765 nm. A calibration curve was constructed using gallic acid standards in the range of 2–10 µg/mL. The results were expressed as milligrams of gallic acid equivalent (GAE) per 100 g of the substrates of quince pomace (S_QPE and S_QPW) and untreated quince pomace (QPU).

2.6.2. Ferric Reducing Antioxidant Power

The antioxidant activity (AOA) was determined using the FRAP (Ferric Reducing Antioxidant Power) method according to the method described in [15]. The samples or standard solutions (15 µL) were mixed with the FRAP reagent (285 µL) and incubated in the dark at 37 °C. The FRAP reagent was prepared by mixing 0.3 M acetate buffer with 10 mM Fe^{3+}—TPTZ (2,4,6-tripyridyl-s-triazine) solution (in 40 mM HCl) and 20 mM $FeCl_3$ solution in a ratio of 10:1:1. After incubation, the absorbance was measured at 593 nm. Trolox was used as standard, and the calibration curve covered concentrations from 0 to 450 µM Trolox/mL. The results were expressed as millimoles of Trolox per gram of the of the quince pomace (S_QPE and S_QPW) and untreated quince pomace (QPU) substrates.

2.6.3. DPPH Radical Scavenging Assay

The DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay was performed based on the procedure outlined in [16] with some modifications. The reaction mixture consisted of 100 µL of the sample or standard solution and 100 µL of 0.3 M DPPH solution in ethanol. The absorbance was measured at 517 nm after a 30 min reaction using a UV-Vis spectrophotometer. Trolox was used as a standard, and the calibration curve ranged from 0.15 mM to 0.0125 mM, based on a 1 mM Trolox stock solution. The results were expressed as millimoles of Trolox per gram of the of the quince pomace (S_QPE and S_QPW) and untreated quince pomace (QPU) substrates.

2.7. Statistical Analysis

Statistical analysis was performed using IBM SPSS® Statistics, version 26 (IBM Corp., Armonk, NY, USA). Experiments were conducted in triplicate, and mean values ± standard deviations (SD) were reported. One-way ANOVA was used to assess significant differences between samples. Levene's test ensured homogeneity of variance. Tukey's HSD test was used to explore significant differences between means.

3. Results and Discussion

The phenolic acids and flavonoids extracted from quince pomace using ethanol (E_QPE) and water (E_QPW) were subjected to polyphenol identification and quantification via HPLC analysis. The HPLC chromatograms are presented in Supplementary Material Figures S1–S4. According to the findings from previous studies [3,17,18], the concentration of phenolic compounds is influenced by several factors, including species, cultivars, environmental conditions, storage, extraction methods, and analysis techniques.

As a result, the reported concentrations of phenolic compounds can vary across different scientific publications. Some authors [17] have reported the identification of epicatechin in quince pulp, and other authors [3] have reported the identification of rutin and quercetin.

The concentrations of polyphenols from the analyzed quince sample were significant, especially in the ethanolic extract (Table 1).

Table 1. The phenolic compounds of extracts determined via HPLC analysis.

	E_QPE µg/g	**E_QPW µg/g**
	Phenolic acids	
Galic acid	6.73	5.6
Chlorogenic acid	371.405	229.295
Ferulic acid	0.1725	1.585
Sinapic acid	204.03	185.125
Ellagic acid	405.32	206.15
	Flavonoids	
Epicatechin	198.8	-
Rutin	405.79	42.285
Myricetin	302.56	117.865
Quercetin	100.8	7.615
Apigenin	53.95	53.95
Luteolin	9.06	-

E_QPE extract of quince pomace after treatment with ethanol; E_QPW—extract of quince pomace after treatment with water.

Most identified polyphenols were found both in the E_QPE and E_QPW samples, except for two flavonoids, epicatechin and luteolin, which were detected only in the E_QPE extract. The E_QPE extract had higher chlorogenic acid, ellagic acid, quercetin, myricetin, and rutin content than the E_QPW extract. The E_QPW had higher content of ferulic acid than E_QPE, but the content was low in both extracts.

The hydration properties of fibers refer to their interaction with water, for which fibers can be either soluble or insoluble. Soluble fibers dissolve in water in the digestive system, forming a gel-like structure, slowing digestion, stabilizing blood sugar levels, and promoting a feeling of fullness. Insoluble fiber, such as that found in quince pomace, has excellent water-holding capacity, supporting the digestive system's health and regular bowel movements via preventing constipation [6,17].

The hydration properties of the three quince pomace samples were determined (Table 2), and it was observed that the S_QPE sample, obtained from the treatment with ethanol at 80 °C, exhibited the highest hydration value, while the lowest hydration value was observed for the untreated quince pomace (QPU).

Table 2. The hydration properties of the samples.

Samples	**SC, mL/g**	**WRC (g/g)**
QPU	6.3 ± 0.1^{a}	5.30 ± 0.26^{a}
S_QPE	8.02 ± 0.02^{b}	7.55 ± 0.36^{b}
S_QPW	6.9 ± 0.4^{c}	6.78 ± 0.11^{c}

QPU—untreated quince pomace, S_QPE—substrate of quince pomace after treatment with ethanol, and S_QPW—substrate of quince pomace after treatment with water. Different letters ^{a}, ^{b}, ^{c} show statistically different differences (±error bars, $\sigma < 0.05$, $n = 3$).

Table 3 presents the results obtained from the acid hydrolysis of the substrates. Our study reveals a higher lignin content (ranging from 28.7 g/100 to 33.3 g/100 g of dry mass) compared to that in the data reported in [6,18,19]. This difference could be attributed to the different processing method [6], different quince species used [19], and the growth conditions during the experiments.

Table 3. Results regarding lignin content, cellulose content, total carbohydrate content (TCC), total non-cellulose polysaccharide (NCP) content, and total polyphenol content (TPC).

	QPU	S_QPE	S_QPW
Cellulose, g/100 g	10.16 ± 0.91 [b]	6.8 ± 0.56 [a]	9.55 ± 0.5 [a]
Lignin, g/100 g	30.1 ± 0.93 [a]	33.3 ± 1.67 [b]	28.7 ± 0.92 [a]
TCC, mg/100 g	27.53 ±2.46 [a]	51.24 ±0.59 [b]	34.73 ±3.35 [c]
NCP, mg/100 g	23.43 ± 1.36 [a]	32.93 ± 0.69 [b]	25.56 ± 0.26 [a]
TPC, mg/100 g	59.9 ± 0. 3 [a]	20.84 ± 0.9 [b]	38.13± 2.31 [c]

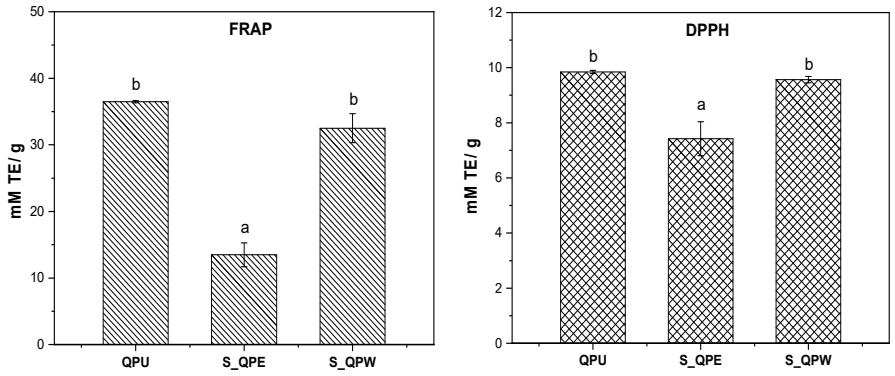
QPU—untreated quince pomace, S_QPE- substrate of quince pomace after treatment with ethanol, and S_QPW—substrate of quince pomace after treatment with water. Different letters [a], [b], [c] show statistically different differences (±error bars, $\sigma < 0.05$, $n = 3$).

Cellulose and lignin are significant components of dietary fibers present in quince pomace and other foods. Cellulose, as an insoluble fiber, supports digestive health, while the water-holding capacity of lignin aids in bowel regularity and prevents constipation. These components play essential roles in managing blood sugar levels, promoting heart health, and supporting a healthy gut microbiota. A diet with cellulose- and lignin-rich foods is essential for overall health and disease prevention [19,20]. The elevated lignin content in the ethanol-treated sample (QPE) can be attributed to the efficient ethanol extraction process and the solubilization of lignin, especially at higher temperatures. Conversely, the untreated sample (QPU) had a higher cellulose content compared to the water- (S_QPW) and ethanol (S_QPE)-treated samples, likely due to its partial removal during the extraction processes. The water and ethanol treatments might have caused the partial degradation or solubilization of cellulose, resulting in reduced cellulose content in the treated samples [21].

Total carbohydrates (TCC) include all types of carbohydrates present in a sample, including non-cellulose polysaccharides (NCP). Non-cellulose polysaccharides refer to the carbohydrates in a sample other than cellulose, including digestible sugars, starches, and other soluble and insoluble carbohydrates. Our results showed that the levels of TCC were slightly lower than the values reported in [7] from quince peels, which were determined using an enzymatic-gravimetric method. The highest levels of TCC and NCP were observed in the S_QPE- substrate of the quince pomace after treatment with ethanol.

The total polyphenol content was higher in the untreated sample (QPU) compared to the samples treated with water (S_QPW) or ethanol (S_QPE), as shown in Table 3. This difference can be attributed to the extraction of water-soluble and ethanol-soluble polyphenols during the water and ethanol treatments, respectively.

Figure 1 depicts the antioxidant activity of the fiber-rich substrates.



(**a**) (**b**)

Figure 1. The antioxidant activity of the untreated quince pomace (QPU), S_QPE—substrate of quince pomace after treatment with ethanol, S_QPW—substrate of quince pomace after treatment with water via the FRAP method (**a**) and DPPH assay (**b**). Different letters indicate statistically different differences (± error bars, $\alpha < 0.05$, $n = 3$).

The lowest values in the ethanol-treated sample (S_QPE) were compared to those of the untreated and water-treated samples. The trend in the antioxidant activity (AOA) mirrored that of the total polyphenol content (TPC). The ethanol treatment led to a higher quantity of phenolic compounds, as evidenced via the HPLC analysis, than the extraction of water. This significant difference in extraction greatly influenced the TPC and the antioxidant activity of the fiber-rich substrate resulting from ethanol extraction.

4. Conclusions

The results of this study, which assessed the chemical composition of quince pomace and the extracted compounds in water and ethanol, as well as the fiber-rich products obtained after extraction, demonstrate the potential of all fractions for various applications, particularly in the food industry. Quince pomace can be used to produce fiber-enriched foods and foods enriched with polyphenols, which will probably become more bioavailable. Moreover, these valuable compounds can be obtained through a cascade process and utilized independently or together for different purposes.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/chemproc2023013006/s1: Figure S1: HPLC chromatogram of phenolic acids in the aqueous extract of quince pomace (E_QPW); Figure S2: HPLC chromatogram of phenolic acids in the alcoholic extract of quince pomace (E_QPE); Figure S3: HPLC chromatogram of flavonoids in the aqueous extract of quince pomace (E_QPW); Figure S4: HPLC chromatogram of flavonoids in the alcoholic extract of quince pomace (E_QPE).

Author Contributions: Conceptualization, F.O.; methodology, L.D. and D.P.; validation, L.D., D.P. and D.C.-A.; formal analysis, L.D. and D.C.-A.; investigation, L.D. and D.P.; resources, D.C.-A., and F.O.; data curation, L.D.; writing—original draft preparation, L.D.; writing—review and editing, D.C.-A. and F.O.; visualization, D.C.-A.; supervision, F.O.; project administration, D.C.-A. and F.O.; funding acquisition, F.O. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the project POC-A1-A1.2.3-G-2015-P_40_352-SECVENT, Sequential processes to close the bioeconomy side stream and innovative bioproducts resulted from these processes—SECVENT, contract 81/2016, SMIS 105684, funded by Cohesion Funds of the European Union, subsidiary project 2609/2020 and 1393/2022-FructiRan.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: All the data are contained within the article.

Acknowledgments: We thank Bogdan Trică for his assistance with the statistical analysis and Qubio Cert for the quince pomace samples.

Conflicts of Interest: The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

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