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Effect of Drying Temperature of Ambar Pumpkin on Proximate Composition and Content of Bioactive Ingredients

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Featured Application: The results of this work can determine the optimal drying temperature for pumpkin (var. Ambar) depending on the further use of the raw material.

Abstract: Pumpkins are often used as a fodder component and food due to their high nutritional value and share of bioactive components (e.g., carotenoids, polyunsaturated fatty acid (PUFAs)). Due to their high moisture content, they must be preserved; drying is still the most popular method. Our work aimed to assess the optimal drying temperature to keep the best possible nutritional value of the raw material. For this purpose, pumpkin was dried at 40 °C, 60 °C and 80 °C. Then, the proximate composition, carotenoid content, fatty acids, and antioxidant properties were determined. The results indicate that the carotenoids were relatively stable up to 60 °C and then decreased sharply. Furthermore, antioxidant activity was the highest at 40 °C and 60 °C. However, in the case of PUFA content, drying at 80 °C was the most effective, probably due to the shorter exposure time to the stimulus.

Keywords: pumpkin; drying temperature; bioactive substances; *Cucurbita maxima*; Ambar

Citation: Koziorzębska, A.; Łozicki, A.; Rygało-Galewska, A.; Zglińska, K. Effect of Drying Temperature of Ambar Pumpkin on Proximate Composition and Content of Bioactive Ingredients. *Appl. Sci.* **2023**, *13*, 8302. https://doi.org/10.3390/app13148302

Academic Editors: Anthony William Coleman and Leonel Pereira

Received: 4 June 2023
Revised: 27 June 2023
Accepted: 17 July 2023
Published: 18 July 2023

1. Introduction

Pumpkin is the most popular plant of the gourd family (*Cucurbitaceae Juss*), which is divided into *Cucurbita pepo*, *Cucurbita moschata*, *Cucurbita maxima* and *Cucurbita mixta*, depending on the texture and shape of the stems [1,2].

Thanks to its high bioactive compound content, pumpkin has many health-promoting properties. Carotenoids are the red and yellow pigments in plants, fish meat and bird feathers, known for their antioxidant properties. Epidemiological studies show that consuming them in natural products lowers the risk of degenerative diseases and protects the macula from light-induced damage [3]. They are also an important ingredient in animal feed. A carotenoid diet has been shown to improve health, production indices and meat quality [4]. Pumpkin contains a high amount of β-carotene but is also a source of lutein, violaxanthin, lycopene, cryptoxanthin and zeaxanthin [5]. Unfortunately, carotenoids have pro-oxidative properties in high doses, but this problem mainly concerns dietary supplements with very high doses compared to their content in vegetables and fruits [3]. In addition, pumpkin is a source of vitamins C, B1 (thiamine), B6 (pyridoxine), fat-soluble vitamins [5] and minerals, such as magnesium, iron, selenium, potassium and phosphorus [6].

Recent research shows that pumpkin flowers may become a functional food due to their high antioxidant content (mainly carotenoids and phenolic compounds) and iron [7].

The dietary fiber in pumpkin positively affects the digestive tract, making it a valuable ingredient in human and animal nutrition and dietetics [8].

Pumpkin seeds are enjoyed as a culinary addition and a source of polyunsaturated fatty acid (PUFA)-rich fat [9,10]; in particular, omega-3 fatty acids are known for their

health-promoting properties, which include anti-inflammatory effects and homeostasis maintenance—prophylactic effects and supporting the treatment of cardiovascular diseases and others [11]. Studies on the impact of pumpkin seed oil have shown that it soothes prostate hypertrophy, inhibits the development of hypertension, reduces hypercholesterolemia and relieves diabetes symptoms [12].

Thanks to the content of these ingredients, pumpkins can be an important component of the human diet and the diets of animals accompanying humans and farm animals. Pumpkin seeds, oil or flowers can be eaten as a dietary ingredient and a base material for creating innovative, functional food [13].

Unfortunately, despite its advantages, pumpkin is a seasonal plant with low microbiological resistance [14]. In addition, fresh pumpkins stored after harvest slowly lose their color and become soft [15]. Preservation is necessary to ensure access to the raw material throughout the year. It is essential to choose a method that preserves as many valuable bioactive ingredients as possible. Drying is one of the most popular methods due to the immediate effect and lack of need for expensive equipment. Removing water from fruits and vegetables reduces the reactions caused by moisture, which ultimately affects the product's usability. However, it is a very diverse process. Traditionally, agricultural products are dried in the sun. However, this process is slow, dependent on environmental conditions and difficult to control. Drying with warm air extends the product's life to a year but decreases health-benefiting compounds [15–18]. Choosing the right conditions for the drying process is advisable to maximize the shelf life and bioactive ingredient content, especially the temperature and drying time [15,17,19]. Previous research has shown the influence of temperature and drying time on the content of carotenoids, flavonoids and others [15,17,20,21].

The available studies analyzed the effect of pumpkin drying temperature on the basic composition of products or individual parameters. However, the Polish variety, Ambar, is still poorly researched. Only two teams investigated the effect of drying on pumpkin quality, but only the technological parameters and the content of three carotenoids (lycopene, β-carotene and lutein) were assessed. The Polish variety is good for industry (e.g., lack of spaces between seeds) and has high dry matter and carotenoid content [22]. This study aimed to determine the effect of different temperatures on the proximate composition and content of valuable bioactive compounds (characteristic carotenoids: α- and β-carotene, lutein, violaxanthin, zeaxanthin and fatty acid profile) in dried Ambar pumpkins (*Cucurbita maxima*).

2. Materials and Methods

2.1. Samples

The study was conducted on *Cucurbita maxima,* var. Ambar pumpkin samples were obtained from the Warsaw University of Life Science's crops.

After delivery of the pumpkin samples to the laboratory, they were washed, shredded and homogenized. Whole pumpkins, including the seeds, were used. Then, the samples were divided into three groups and dried at three different temperatures (40 °C, 60 °C and 80 °C) until a constant weight was obtained. At 40 °C, it took 98.6 h to dry; at 60 °C, 81 h and at 80 °C, 33.5 h.

Prepared samples were protected from sunlight and stored vacuum-sealed in low-density polyethylene bags.

2.2. Analyses

2.2.1. Proximate Analysis

The chemical composition was determined according to The International Association of Official Analytical Chemists (AOAC) (2005): dry matter (DM) by drying at 104 °C for 24 h, crude protein (N × 6.25) using the micro-Kjeldahl technique (Kjeltec System 1026 Distilling Unit. Foss Tecator, FOSS, Stockholm, Sweden), ash by incineration at 550 °C for six h and crude fat after extraction with petroleum ether using the Soxhlet method [23]. Neutral

detergent fiber (NDF) and acid detergent fiber (ADF) were determined according to Van Soest et al. (1991) [24]. NDF was expressed as the ash-free residue after extraction with boiling neutral solutions of sodium lauryl sulfate and EDTA in a Tecator apparatus. ADF was determined as the ash-free residue after extraction with boiling in an acid detergent solution of sulfuric acid and cetyltrimethylammonium bromide (CTAB).

2.2.2. Gas Chromatography

Fatty acid (FA) profile analysis was carried out by extracting lipids according to Folch's method [25], and Fas was esterified in accordance with PN-EN ISO 5509:2001. The analysis was conducted using a TRACE GC ULTRA gas chromatograph (Thermo Electron Corporation, Waltham, MA, USA) equipped with a flame ionization detector (FID) on a SUPELCOWAXTM 10 Capillary GC Column (Merck KGaA, Darmstadt, Germany) (30 m $\times$ 0.25 mm $\times$ 0.25 µm). The injector temperature was 225 °C, and the detector temperature was 250 °C. Nitrogen was used as the carrier gas. The fatty acids were identified by comparing the retention times with a standard (Supelco 37 Component FAME Mix). For the quantitative analysis, the peak area in the chromatogram was determined, and the percentage of a given acid was calculated.

2.2.3. High-Performance Liquid Chromatography (HPLC)

The carotenoid separation and contents were analyzed using the HPLC (Dionex Corporation, Sunnyvale, CA, USA) equipped with a CoulArray electrochemical detector (ESA Inc., Chelmsford, MA, USA). The separation was conducted on a Hypersil BDS 150 4.6 mm, 5 µm column (Sigma-Aldrich, Saint Louis, MO, USA) at a mobile phase flow rate of 1.2 mL/min. The mobile phase consisted of a methanol/isopropanol mixture (98:2). The conditions of electrochemical detection were as follows: four electrodes with potentials 400, 500, 600 and 750 mV. The chromatograms were processed by identifying the pigments based on standards and areas of chromatographic peaks, considering their retention times and the peak area ratio for the dominating electrode to that of neighboring electrodes.

2.2.4. Antioxidant Activity

Antioxidant potential was measured using the 2,2 Diphenyl 1 picrylhydrazyl (DPPH) radical reduction test. Then, 0.290 mL of 0.1 M DPPH was vortexed with 0.01 mL of the dried pumpkin samples at 0.100 mg/mL concentration. After 20 min in the dark, the results were read at a wavelength of 570 nm in a plate spectrophotometer (Tecan, Männedorf, Switzerland). Meanwhile, a standard curve for TROLOX was prepared. On this basis, the antioxidant potential of samples from each group was calculated.

2.2.5. Statistics

Statistical analysis was performed using one-way analysis of variance (ANOVA) with Tukey's post hoc test by Statistica 13.3 software (TIBCO Software Inc., Palo Alto, CA, USA). Differences with a p-value ≤ 0.05 were defined as significant.

3. Results

The proximate composition analysis demonstrated a significant effect of the drying temperature on each chemical component of pumpkin (Table 1). For all parameters, the highest value was observed in the samples dried at 60 °C. Apart from dry matter, all values were lowest for samples obtained at 80 °C. The dried material obtained at the temperature of 60 °C was the most distinguished because it retained the most nutrients.

Table 2 shows the content of different carotenoids in pumpkin *C. maxima* (var. Ambar) dried at three different temperatures (40 °C, 60 °C and 80 °C). The main carotenoids in pumpkin were α- and β-carotene.

Statistical analysis showed no significant differences between the dried materials prepared at 40 °C and 60 °C. However, the differences between drying at 40 °C and 60 °C over 80 °C were significant for all carotenoids except α-carotene.

Table 1. Proximate content of *Cucurbita maxima* pumpkin (var. Ambar) dried at different temperatures. Letters A, B, and C mean the values are statistically different for $p < 0.05$.

Parameter	40 °C	60 °C	80 °C	*p*-Value	SE
Dry matter (%)	90.36 A	92.27 B	91.16 A	0.0003	0.2718
Ash (g/kg DM)	8.37 A	10.53 B	7.90 A	0.0000	0.1232
NDF (g/kg DM)	24.50 A	27.12 B	23.83 A	0.0028	0.6191
ADF (g/kg DM)	26.75 A	27.29 A	21.60 B	0.0000	0.5511
ADL (g/kg DM)	5.68 A	7.80 B	5.48 A	0.0000	0.2806
Protein (g/kg DM)	19.15 A	21.88 B	16.50 C	0.0000	0.2767
Fat (g/kg DM)	13.01 A	13.02 A	11.10 B	0.0000	0.2603
Fiber (g/kg DM)	18.04 A	19.86 B	14.49 C	0.0000	0.346

Table 2. Content (mg/100 g d.m.) of carotenoids in pumpkin *C. maxima* (var. Ambar) dried at different temperatures. Letters A and B mean the values are statistically different for $p < 0.05$.

Individual Carotenoids	40 °C	60 °C	80 °C	*p*-Value	SE
β-carotene	33.92 A	31.27 A	27.20 B	0.0005	1.012
α-carotene	38.56	38.54	33.62	0.0901	0.0577
violaxanthin	3.72 A	3.16 A	1.29 B	0.0001	0.2591
zeaxanthin	0.27 A	0.25 A	0.18 B	0.0394	0.0231
lutein	8.74 A	8.14 A	6.58 B	0.0470	0.5279

For β-carotene, there was a decrease of 13.06% between 60 °C and 80 °C. Violaxanthin experienced a substantial decrease of 59.41%. This indicates a significant reduction in violaxanthin content as the drying temperature increased. Zeaxanthin also showed a decrease in content, with a reduction of 28.00%. Similarly, lutein exhibited a decrease of 19.57%. These percentage changes highlight the impact of drying temperature on the content of specific carotenoids. Higher drying temperatures generally decreased β-carotene, violaxanthin, zeaxanthin and lutein, suggesting that the drying process significantly affects the carotenoid level in pumpkin.

Antioxidative Potential

Statistical analysis of the antioxidant potential of DPPH showed highly significant differences between the pumpkin dried at 40 °C, 60 °C and 80 °C (Table 3). It was observed that the ability to inhibit the DPPH radical decreased with increasing temperature. The differences, although statistically significant, were very small.

Table 3. Antioxidant activity of pumpkin *C. maxima* (var. Ambar) dried at different temperatures. Different letters mean that the values are statistically different for $p < 0.05$.

Antioxidant Power	40 °C	60 °C	80 °C	*p*-Value	SE
DPPH mmol TROLOX/l	0.51 A	0.50 B	0.49 C	0.0026	0.001

Table 4 shows the fatty acid composition of pumpkin dried at different temperatures (40 °C, 60 °C and 80 °C), listing the percentage of different fatty acid types, including saturated, monounsaturated, polyunsaturated, and individual fatty acids. The analyses showed significantly fewer monounsaturated fatty acids (MUFAs) in the dried material obtained at 80 °C. No differences were found for the sum of saturated and polyunsaturated acids (PUFAs), but they were noticeable for some individual fatty acids from this group: C20:4, C20:3, C20:2 and C18:3. Of the saturated fatty acids, only 24:0 was significantly affected by temperature. The highest value of this acid was observed for samples dried at the lowest temperature; drying at 60 °C and 80 °C reduced its content. The results indicate that raising the temperature to 80 °C keeps more of the most valuable fatty acids from the omega-3 group (C18:3).

Table 4. Fatty acid content (g/100 g) of pumpkin dried at different temperatures. Different letters mean that the values are statistically different for $p < 0.05$.

Fatty Acids (Groups and Individuals)	40 °C	60 °C	80 °C	*p*-Value	SE
Saturated fatty acids (SFAs)	22.26	20.14	21.04	0.1111	0.6797
Monounsaturated fatty acids (MUFAs)	18.97 A	19.16 A	18.31 B	0.0010	0.1414
Polyunsaturated fatty acids (PUFAs)	58.59	60.49	60.45	0.0877	0.6566
C10:0	0.0062	0.0059	0.0066	0.6471	0.00047
C12:0	0.044	0.04	0.046	0.1284	0.0019
C14:0	0.394	0.367	0.405	0.0575	0.0106
C14:1	0.0285 A	0.0087 B	0.0117 B	0.0069	0.0042
C15:0	0.0511	0.0425	0.0448	0.1141	0.0028
C16:0	15.97	14.36	15.2	0.0694	0.4597
C16:1n-9	0.028 A	0.046 B	0.043 B	0.0049	0.0035
C16:1n-7	0.2373 A	0.1751 B	0.1983 C	0.0000	0.0041
C17:0	0.0856	0.0796	0.0826	0.4659	0.0033
C17:1	0.0204	0.026	0.0253	0.3676	0.003
C18:0	5.047	4.692	4.698	0.3318	0.1883
C18:1n-9	16.279 A	16.954 B	16.206 C	0.0012	0.1338
C18:1n-7	2.273 A	1.867 B	1.728 C	0.0000	0.0428
C18:2n-6	54.45	56.19	55.01	0.1260	0.5873
C18:3n-3	3.91 B	4.22 B	5.2 A	0.0000	0.1517
C20:0	0.319	0.314	0.32	0.9599	0.0164
C20:1	0.107	0.086	0.107	0.3169	0.0108
C20:2	0.215 A	0.07 B	0.21 A	0.0089	0.0336
C20:3n-6	0.016 A	0.014 A	0.026 B	0.0184	0.0029
C20:4n-6	0.0048 A	0.0057 A	0.0102 B	0.03	0.0014
C22:0	0.1962	0.175	0.1593	0.152	0.0129
C24:0	0.1518 A	0.0631 B	0.0771 B	0.004	0.139

4. Discussion

Drying is one of the most used methods for preserving fruits and vegetables. However, the parameters of this process significantly affect the chemical composition. The key is to choose such drying parameters to obtain a product with low moisture content and, at the same time, minimize the loss of nutrients and bioactive ingredients.

Drying at 40 °C, 60 °C and 80 °C was performed in this study. At the highest temperature, the lowest content of protein, fat, ash, crude fiber, NDF and ADF fractions was observed. Looking at the basic composition of pumpkins, the most optimal temperature seems to be 60 °C. It maintains a high nutrient content, and at the same time, the drying time is shorter than in the case of lower temperatures. A similar conclusion was obtained from a study by Guine et al. (2011), which assessed that drying pumpkin at 70 °C allows the process to be completed much faster than in the case of 30 °C, and changes in the basic composition are insignificant [14].

However, pumpkin is also a source of bioactive ingredients sensitive to high temperatures. That is especially true for double chemical bonds in the chemical structure, e.g., carotenoids and unsaturated fatty acids [26]. Inadequate raw pumpkin-based material processing can lose significant amounts of biologically active ingredients.

The degradation and structural modification of carotenoids occurs during thermal treatments such as cooking [27,28] and hot drying [29]. Lago-Vanzela et al. (2013) found that drying pumpkin at 70 °C for 8–10 h caused a maximum loss of 9–13% for α- and β-carotene [29]. On the other hand, Dutta et al. (2006) found that the total β-carotene in pumpkin puree was reduced by 26% and 62% after heating at 70 °C and 100 °C, respectively, for 2 h compared to fresh samples. This study showed that the β-carotene content of pumpkin dried at 80 °C was 13% lower than at 60 °C and 20% lower than at 40 °C. For α-carotene, it was 13% in both cases [30]. Ouyang et al. (2022) obtained similar results. Drying at 80 °C led to a 14% decrease in the content of β-carotene and 16% of α-carotene

compared to 60 °C. However, they observed a decrease in lutein by 68%, compared to 19% in the reported study. They also found complete degradation of violaxanthin upon drying at 25 °C and above [31].

Provesi et al. (2011) showed that by cooking pumpkin for 20 min at 100 °C, the content of lutein decreased by 15.5%, α-carotene by 17.9% and β-carotene by 16.9% compared to the fresh raw material and complete disintegration of violaxanthin. In our study, the carotenoid content retained the same level when drying at 40 °C and 60 °C, and a significant decrease was observed only at 80 °C [27].

Despite the observed differences in individual studies, there is a tendency for the increase in temperature to cause a reduction of carotenoids and even their complete decomposition. The differences are most likely due to the research methodology being different. It is known that not only temperature affects the degradation of carotenoids. The duration of the drying process is also important, as well as the type of thermal treatment (e.g., convection, sun, vacuum drying or cooking) and pre-treatment (e.g., blanching, dipping in sulfite) [32]. Our study focused on the simplest and most popular method because we wanted the results to be widely applicable in practice.

The content of carotenoids is also associated with the antioxidant properties of pumpkins. β-carotene and lutein, in particular, have very well-documented capabilities. The antioxidant potential of dried pumpkin is no less affected by ingredients such as vitamins C, D, E and polyphenols. The ability to inhibit the DPPH radical was compared for samples dried at 40 °C, 60 °C and 80 °C. Even though the values differed only slightly, similar to the content of carotenoids, statistically significantly lower antioxidant activity was observed for pumpkin samples processed at 80 °C compared to lower temperatures. The results of studies by other authors also confirm this. Köprüalan et al. (2021) observed a decreasing ability to inhibit the DPPH radical with increasing temperature in the 50 °C –70 °C range, and they obtained the best results for freeze-dried samples [33]. Similarly, the antioxidant power of tomato skins dried at 50 °C and freeze-dried did not differ, but the samples processed at 80 °C had negligible antioxidant value [34].

In contrast, Fei Que et al. (2007) obtained a higher antioxidant activity from pumpkin flour dried at 70 °C than from freeze-dried. At the same time, they observed a 4.6× higher content of polyphenols when the pumpkin was dried compared to freeze-dried. It is known that polyphenols have strong antioxidant activity and are relatively heat resistant [35]. Soong and Barlow (2004) even observed an increase in the content of these compounds with increasing drying temperature, up to 200 °C [36]. In our study, we did not assess polyphenol content. Perhaps the variety of *Cucurbita moschata* evaluated in the Chinese study is characterized by a higher content of polyphenols than the variety of *C. maxima* produced in Poland.

Another health-promoting ingredient of pumpkin is PUFA. Pumpkin oil is abundant in C18:2 omega-6 fatty acids; the 18:3 acid represents the omega-3 family. The total polyunsaturated fatty acids in the assessed oils from pumpkin varieties *Cucurbita maxima* from Poland ranged from 47.5% to 57.5% [37]. PUFAs, due to the presence of unsaturated bonds, are highly sensitive to temperature, which increases lipid peroxidation. As a result of their reaction with oxygen, secondary oxidation products are formed, which can be toxic. Susceptibility to oxidation is proportional to the number of unsaturated bonds, specifically fatty acids [38].

Unexpectedly, the content of individual polyunsaturated acids was the highest for dried samples obtained at 80 °C. A similar result was obtained for other raw materials. Han et al. (2019) showed that roasting walnuts at 105 °C compared to 60 °C resulted in a higher omega-3 α-linolenic acid and omega-6 linoleic acid content. However, the content of this compound decreased at 140 °C [39]. Furthermore, in the case of acorns, higher values of alpha-linolenic acid were noted in proportion to the increase in drying temperature at 40 °C–120 °C [40]. That is most likely because the drying time was longer when processing at lower temperatures. Shorter exposure to the peroxidative agent was

found to be protective in this case. Perhaps this is also because high temperature inhibits the action of lipoxidase enzymes that break down polyunsaturated fatty acids.

Inferring from the results obtained in this study, it can be determined that in the case of pressing pumpkin oil, it is worth using the raw material after processing at 80 °C. However, it should be noted that the total amount of fat decreased after exceeding 60 °C.

5. Conclusions

Pumpkin is a valuable component of human and animal diets, but due to high moisture, it is unstable. One of the most common preservation methods is drying. Comparing the drying temperatures of 40 °C, 60 °C and 80 °C, we have shown that in terms of the content of carotenoids and antioxidant properties, the temperature of 60 °C is the most optimal because it allows for a shorter drying time compared with 40 °C, while simultaneously maintaining the optimal level of carotenoids. In contrast, raising the temperature to 80 °C caused a significant decrease in the content of carotenoids and antioxidant activity; yet, for the content of polyunsaturated fatty acids, drying at the highest temperature was the most effective.

The results of this study highlight the effect of temperature on the quality of pumpkin products. The drying temperature can be adjusted to the expected outcomes based on the obtained data. If the goal is to obtain a product with high PUFA content, it is best to use a temperature of 80 °C. However, considering the basic composition, carotenoid content and antioxidant activity, drying at 60 °C is optimal.

Author Contributions: Conceptualization, A.K.; Methodology, A.Ł.; Formal analysis, A.K. and A.R.-G.; Investigation, A.K. and K.Z.; Writing—original draft, A.K.; Writing—review & editing, A.Ł. and K.Z.; Visualization, A.R.-G.; Supervision, A.Ł. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Conflicts of Interest: The authors declare no conflict of interest.

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