JAP&T
TPA-Timişoara

Available online at http://journal-of-agroalimentary.ro

Journal of Agroalimentary Processes and Technologies 2017, 23 (1), 31-35

*Journal of Agroalimentary Processes and Technologies*

# The effect of drying on total phenol, antioxidant activity, and mineral contents of white and black mulberry fruits

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**Received: 17 January 2017; Accepted: 23 March 2017**

***Abstract***

Antioxidant activity of black mulberry extracts changed between 75.74% to and 81.95%. In addition, antioxidant activity of white mulberry ranged from 41.069% to 80.309% ($p<0.05$). Total phenol contents of both mulberry samples increased depending on drying time. While the total phenol contents of black mulberry increase from 97.88 mg/100g (control) to 373.23 mg/100g (20.h), total phenol contents of white mulberry fruits increased from 40.574 mg/100g (4.h) to 276.309 mg/100g (20.h) ($p<0.05$). Fatty acid composition of black mulberry fruits were found higher than those of white mulberry fruits. Also, palmitic, oleic, linoleic and stearic acids were established as the major fatty acids of mulberry fruit oils. Linoleic acid was determined as 58.89% and 63.83% in white and black mulberry fruits, respectively. All minerals increased depending on drying times. Potassium was the highest element in fresh and dried fruits of black and white mulberry.Cu, Fe, K, Mg and Mn contents of fresh and dried fruits of white mulberry were found higher compared to black mulberry fruits.

***Keywords***: mulberry fruit, drying, antioxidant, total phenol, fatty acid, mineral

## 1. Introduction

Mulberry (*Morus* spp.; Moraceae family) is widely distributed in Asia, Europe, North America, South America, and Africa [1,2]. Recently, the production and consumption of mulberry fruits increased rapidly depending on their good taste, nutritional value, and biological activities [3,5]. Anthocyanins have biological activities such as antioxidant, antimicrobial, neuroprotective, and anti-inflamatorty properties [6,7]. The phenolic compounds, including flavonoids, anthocyanins, and carotenoids are found more in the deep colored mulberry fruits [8].

Geographic location and soil affect to the total content of phenolic compounds of the mulberry tree growing [5]. Human, animals, microorganisms and plants are effected from heavy metals due to their significant toxicity [9,10]. Although black mulberry is mostly using for making processed foods [11], the white mulberry (*Morus alba*) is used as the primary food source for silk worms [12]. The aim of current study is to determine the total phenol, antioxidant activity of mulberry fruits dried at different times at 70 °C in oven, and to determine mineral contents of fresh and dried mulberry fruits.

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## 2.Material and methods

### 2.1.Materials

Black (*Morus nigra* L.) and white mulberry (*Morus alba* L.) fruits collected from Antalya (Kumluca) and Mersin (Tarsus) provinces in May 2015 were transferred to laboratory in cool bags, respectively. Moisture contents were determined soon. Then, fruits were washed with clear distilled water. They were kept in refrigerator by using. 1 kg fruit was used for each analyses. All reagents and solvents were analytical grade and purchased from Sigma-Aldrich Co. (St. Louis, MO, USA).

### 2.2.Methods

#### 2.2.1.Drying process

Mulberry fruits were dried in an oven (Nüve FN055 Ankara, Turkey, 55 l volume) at certain intervals (4, 8, 12, 16 and 20 h) at 70°C, and the moisture contents were decreased till 8.47 and 4.67%, respectively. Fresh and dried samples were analysed. The initial moisture contents of the samples were measured by drying in an oven at 105°C until a constant weight was obtained.

#### 2.2.2.Extraction of mulbery fruit for antioxidant and total phenol

The mulberry fruits which were taken from each varieties about 50-60g were immediately put on the stainless steel trays and transferred to the ovens setted at 70 °C. Phenolic compounds were extracted according to Natic et al. [13] with some modifications. 1 g of ground samples were added to 10 ml of methanol containing 1% HCl. Sample was placed ultrasonicator bath for 30 min. After sonication, centrifuged at 5000 rpm for 10 min and the supernatant was collected. The extraction procedure was repeated twice. The extract was concentrated at 40°C in a rotary evaporator under the vacuum. The dried extract was dissolved in 10 ml of methanol/water (60/40). The extract was used for analysis.

#### 2.2.3.Total phenolic content

Total phenol contents of obtained extracts were found by using the Folin-Ciacueltau (FC) reagent as applied by Yoo et al. [14] with some modifications. 1 ml of Folin–Ciacueltau was added and mixed for five minutes. Following the addition of 10 mL of $Na_2CO_3$ solution tubes were mixed and the final volume was completedto 25 ml with deionised water. At theend of 1 hours, total phenol content was determined as 750 nm wave length in spectrophotometer. Gallic acid was used (0-200 mg/ml) as the Standard for calibration curve. All determinations were performed in triplicate. The results were given as mg gallic acid equivalent (GAE)/100 grams of fresh weight.

#### 2.2.4.Antioxidant activity

The free radical scavenging activity of samples was determined using DPPH (1,1-diphenyl-2-picrylhydrazyl) according to Lee et al. [15]. The extract was mixed with 2 mL methanolic solution of DPPH. The mixture was shaken vigorously and allowed to stand at room temperature for 30 min. The absorbance was recorded at 517 nm by using a spectrophotometer. All determinations were performed in triplicate.

#### 2.2.5.Determination of minerals

Mulberry fruit samples were dried at 70 ºC in a drying cabinet with air-circulation until they reached constant weight. Later, about 0.5 g dried and ground sample was digested by using 5ml of 65% $HNO_3$ and 2 ml of 35% $H_2O_2$ in a closed microwave system (Cem-MARS Xpress) at 200 ˚C. The volumes of the digested samples were completed to 20 ml with ultra-deionized water and mineral concentrations were determined by inductively coupled plasma-optical emission spectroscopy (ICP-AES; (Varian-Vista, Australia). Measurement of mineral concentrations was checked using the certified values of the related minerals in the reference samples received from the National Institute of Standards and Technology (NIST; Gaithersburg, MD, USA). Distilled deionized water and ultrahigh-purity commercial acids were used to prepare all reagents, standards, and samples. After digestion treatment, samples were filtrated through whatman No 42. The filtrates were collected in 50 ml flasks and analysed by ICP-AES. The heavy metal contents of the samples were quantified against standard solutions of known concentrations which were analysed concurrently [16].

***Working conditions of ICP-AES:***

Instrument:ICP-AES (Varian-Vista)
RF Power: 0.7-1.5 kw (1.2-1.3 kw for Axial)
Plasma gas flow rate (Ar): 10.5-15 L/min. (radial) 15 “ (axial)
Auxilary gas flow rate (Ar):1.5 “
Viewing height: 5-12 mm

Copy and reading time:1-5 s (max.60 s)
Copy time: 3 s (max. 100 s)

### 2.3.Statistical analysis

A complete randomized split plot block design was used analysis of variance (ANOVA) was performed by using JMP version 9.0 (SAS Inst. Inc., Cary, N.C.U.S.A). The results are mean±standard deviation (MSTAT C) of independent mulberry samples [17].

## 3. Results and discussion

Moisture content, antioxidant activity and total phenol contents of black and white mulberry fruits dried at different drying times on 70 $^0$C in oven are given in Table 1. Moisture contents of both mulberry samples were decreased depending on drying times. While moisture contents are decreased from 86.42% to 8.47% in black mulberry fruit, it was decreased from 89.47% to 4.67% in white mulberry fruit in total drying time (Table 1) ($p<0.05$). While antioxidant activity of black mulberry increases from 75.739% to 81.950%, antioxidant activity of white mulberryfruits increased from 41.069% to 80.309% ($p<0.05$). The total phenol contents of black mulberry increased from 97.877 mg/100g (control) to 373.231 mg/100g (20.h). In addition, total phenol contents of white mulberry fruits dried at different times increased from 40.574 mg/100g (4.h) to 276.309 mg/100g (20.h) ($p<0.05$). Antioxidant activity values of black mulberry were found higher than those of white mulberry values depending on drying periods. At the end of drying, antioxidant values of both mulberry were found higher. This increasing is probably due to removing of moisture contents of mulberry samples. But antioxidant activity of white mulberry dried at 4 and 8.h was found lower compared with control sample. Total phenol contents of both mulberry samples increased depending on drying times. Generally, total phenol content of black mulberry fruit was found higher than those of white mulberry sample. Moisture and total phenol contents of eight mulberry cultivated in Chine were ranged from 70.0 to 87.4% and 189.67 to 246.00 GAE mg/kg, respectively [3]. In previous study in China, total phenol content of mulberry fruits in China was lower than the results presented here probably because of the different species, growth areas, and extraction method [3]. The total phenol contents of mulberry fruits were found at high levels (880-1650 mg/100g fresh weight) [2]. The contents of total phenolic of *Morus alba* L., *Morus nigra* L. and *Morus rubra* L. changed between 16.21 and 24.37 mg GAE/g [18

***Table 1***. Biochemical properties of two types mulberry fruits

| Samples | Drying Time | Moisture (%) | Antioxidant activity (%) | Total phenol (mgGAE/100g) |
|---|---|---|---|---|
| Black mulberry | Control | 86.42±1.17*a | 75.739±0.017c | 97.877±0.012d |
| | 4h | 81.83±1.28b** | 78.950±0.006b | 100.00±0.037d |
| | 8h | 70.98±1.09c | 79.606±0.014b | 118.634±0.033c |
| | 12h | 62.59±1.67d | 79.184±0.005b | 139.156±0.052c |
| | 16h | 38.17±0.81e | 79.466±0.001b | 223.691±0.028b |
| | 20h | 8.47±0.49f | 81.950±0.002a | 373.23±0.022a |
| White mulberry | Control | 89.47±1.52a | 41.069±0.008c | 56.529±0.012e |
| | 4h | 80.65±1.43b | 23.347±0.008d | 40.574±0.030e |
| | 8h | 65.14±0.97c | 26.582±0.030d | 73.184±0.019d |
| | 12h | 44.95±0.76d | 42.944±0.019c | 102.623±0.014c |
| | 16h | 37.52±0.38e | 52.602±0.005b | 155.803±0.021b |
| | 20h | 4.67±0.42f | 80.309±0.007a | 276.309±0.028a |

*mean±standard deviation (n:3)
**Values within each column followed by different letters are significantly different ($p<0.05$)

***Table 2.*** Mineral and heavy metal contents of fresh and dry mulberry fruits

| Samples | Process | Mo | Ca | B | Cu | Fe | K | Mg |
|---|---|---|---|---|---|---|---|---|
| Black mulberry | Fresh | 0.88 ± 0.03*b | 881.770 ± 10.40c | 11.751 ± 4.09c | 8.953 ± 1.61d | 39.921± 0.31d | 3371.282±390.18c | 501.328±123.97c |
| | Dried | 1.342±0.28a** | 2448.656 ± 239.97b | 14.357 ± 0.97b | 26.045 ± 22.89a | 63.981 ± 14.03b | 14852.139 ± 45.64b | 1767.078 ± 238.89b |
| White mulberry | Fresh | 0.655 ± 0.09b | 880.941 ± 98.52c | 9.380 ± 2.13d | 12.538 ± 11.14c | 43.733 ± 14.23c | 3481.523 ± 110.c | 541.755 ± 162.25c |
| | Dried | 1.360 ± 0.10a | 5137.203 ± 312.28a | 22.139 ± 1.62a | 14.523 ± 0.39b | 81.396 ± 12.38a | 24425.925 ± 386.53a | 2985.318 ± 211.66a |

| Samples | Process | Mn | Na | Ni | P | S | Zn |
|---|---|---|---|---|---|---|---|
| Black mulberry | Fresh | 2.200±0.419c | 125.821±59.531b | 1.229±0.18c | 642.002±87.17c | 1847.035±118.37c | 10.129±2.495c |
| | Dried | 9.229 ± 2.437b | 128.453 ± 22.975a | 2.836 ± 0.17b | 2103.109 ± 177.76b | 2622.892 ± 105.62b | 21.735 ± 0.128a |
| White mulberry | Fresh | 2.203 ± 0.323c | 110.175 ± 59.773d | 0.892 ± 0.199d | 534.183 ± 16.39d | 1765.819 ± 585.80c | 13.757 ± 11.303b |
| | Dried | 17.373 ± 1.103a | 113.450 ± 13.090c | 5.946 ± 0.25a | 3195.955 ± 254.66a | 3006.871 ± 31.35a | 22.084 ± 1.344a |

*mean±standard deviation (n:3)
**Values within each column followed by different letters are significantly different (p<0.05)

The antioxidant activity of the mulberry cultivars were found between 50% and 96% [3]. Dimitrijevic et al. [5] reported that total phenol in the extracts of fresh mulberry fruit in acetone-water (1:1, v/v) and in ethanolic extractvaried from 629.7 mg/kg to 4326.0 mg/kg (GAE), respectively [5]. The highest antioxidant activity (33.1%) was observed from methanol extract of black mulberry leaves (Yiğit et al. 2008).

Mineral contents of fresh and dried black and white mulberry fruits are shown in Table 2. K contents of fresh and dried black mulberry were determined as 3371.28 mg/kg and 14852.14 mg/kg, respectively. The K contents in fresh and dried white mulberry were found as 3481.52 mg/kg and 24425.93 mg/kg, respectively ($p<0.5$). Ca contents of fresh and dried black mulberry was determinated as 881.77 mg/kg and 2448.66 mg/kg, respectively. In fresh and dried white mulberry, Ca content was established as 880.94 mg/kg and 5137.20 mg/kg, respectively ($p<0.05$). While Fe contents of fresh and dried black mulberry are determinated as 39.92 mg/kg, and 63.98 mg/kg, respectively, Fe contents of fresh end dried white mulberry was found as 43.73 mg/kg and 81.40 mg/kg, respectively. Mn and Zn contents of white mulberry were found higher than those of black mulberry samples. Na contents of black mulberry were found high compared with results of white mulberry. K contents of *Morus alba*, *Morus nigra* and *Morus laevigata* ranged from 1270 to 1731 mg/100 g, while Ca, Na and Mg contents were found between 440 to 576 mg/Kg, 260 to 280 mg/Kg, and 240 to 360 mg/Kg, respectively [2].

## 4. Conclusion

In the current work, antioxidant activity and total phenol contents of both mulberry samples increased depending on drying. The radical scavenging activity of extracts of mulberry fruits showed a correlation with total phenolic constituents of the mulberry fruits. The fruits of mulberry contained the lowest amount of heavy metals. According to results, both mulberry fruits serve as a potential nutrition and natural antioxidant sources, and may be useful in a balanced nutrition.

**Compliance with Ethics Requirements.** Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human / or animal subjects (if exist) respect the specific regulation and standards.

**Acknowledgements**

The authors would like to extend their sincere appreciation to the Deanship of Scientific Research at King Saud University for its funding the Research group NO (RG-1435-049.

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