

## Effect of Plant Parts and Harvest Period on Rutin, Quercetin, Total Phenol Contents and Antioxidant Activity of Buckwheat (*Fagopyrum esculentum* Mönch) Cultivated in Turkey

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The rutin, quercetin, total phenol contents and antioxidant activity of different parts of buckwheat (*Fagopyrum esculentum*) harvested at the different growing period in year 2009 were determined. Harvest period and plant parts had affected on these bioactive properties ( $p < 0.01$ ). Rutin values ranged from 1874.97 (stem) to 3329.99 (leaf) mg/g. Quercetin contents were found between 2.20 and 29.33 mg/g. The phenol contents ranged between 24261.82 mg (flower) to 2989.10 mg (stem). The DPPH inhibition of flower, leaf, root and stem were established as 89.71, 73.57, 56.70 and 50.85 %, respectively. While quercetin decreased toward the end of harvest period, it was increased in root.

**Key Words:** Buckwheat, Rutin, Total phenol, Antioxidant activity.

### INTRODUCTION

Buckwheat (*Fagopyrum esculentum* Moench), belong to the Polygonaceae family, is usually grouped with cereals because of its ways of cultivation and utilizations. But it is not a cereal grain. The dietary and health value of rutin in buckwheat seeds has received an increased attention in recent years<sup>1-6</sup>. At the same time, its seeds are used in many farms in foods particularly popular in Japan, Russia and Central and Eastern Europe<sup>7</sup>. Buckwheat is a native of Northern Europe and Asia<sup>8-10</sup>. Buckwheat is known as an origin plant of rutin known for its pharmacological effects.

Phenolic compounds in buckwheat have been shown to possess antioxidative activity<sup>11,12</sup>. Tian *et al.*<sup>13</sup> have identified rutin, quercetin, kaempferol-3-rutinoside and a trace quantity of a flavanol triglycoside. Rutin has been exhibited antioxidative, antihemorrhagic and blood vessel protecting properties<sup>3,14</sup>. Natural antioxidants from plant extracts have attracted increasing interest due to consumer concern about the safety of the synthetic antioxidants in food<sup>13</sup>. Buckwheat seed contains antioxidants such as rutin, tocopherols and phenolic acids<sup>15</sup>. Variation in antioxidant activity of buckwheat was mainly in antioxidant activity of buckwheat was mainly due to the cultivars and environment effects<sup>15</sup>. Among fruits, vegetables and grain crops, grapes and buckwheat are the most important rutin containing foods. Different cultivars of buck-

wheat may have different contents of rutin<sup>16,17</sup>. Most rutin is accumulated in the inflorescence (up to 12 %, d.w.b.-dry weight basis), in stalks (0.4-1.0 %, d.w.b.) and in upper leaves (8-10 %, d.w.b.)<sup>18</sup>. Ecological factors may also have a great influence on rutin content<sup>19</sup>. Several studies on rutin, quercetin, campherol contents and antioxidant properties of buckwheat grown at the different countries were carried out<sup>3-6,12,20</sup>.

The aim of this study is to compare the rutin, quercetin, total phenol contents and antioxidant properties of different parts (flower, root, leaf and stem) of buckwheat growing in Turkey.

### EXPERIMENTAL

The experiment was carried out in the field trials of Bahri Dagdas International Agricultural Research Institute. This area is 1028 m altitude from the sea level. The structure experimental land's soil was clayey, unsalted [1.27 EC (mmhos/cm)], slightly alkaline (7.50 pH) and organic matter content was good (4.29 % organic matter). Phosphorus (16.85 kg/da P<sub>2</sub>O<sub>5</sub>), potassium (203.03 kg/da K<sub>2</sub>O) and calcium (26.37 % CaCO<sub>3</sub>) level of soil was higher. In 2008, rainfall during the growing seasons of buckwheat (May, June, July) were 23.4, 7.5, 5.5 mm, respectively and average temperature were in the same order 15.7, 22.0, 24.6 °C. In the experiment, buckwheat seed [*Fagopyrum esculentum* Moench. (population)] was brought from Ukraine. Buckwheat seed was seeded to

20 cm row spacing with both hand and machine (200 seed/m<sup>2</sup>) and 1-1.5 cm deep of seed sowing. Sowing and harvesting dates were 16 May 2008 and 22 July 2008, respectively. 10 kg/da DAP fertilizer (18 % N and 46 % P<sub>2</sub>O<sub>5</sub>) was added to the soil at the sowing time. Irrigation were carried out in different time (14 May-17 June 2008). Green plant samples for laboratory analysis were taken four different time (16 June, 30 June, 15 July and 22 July 2008).

**Extraction:** The samples were dried in shade and grounded in mill. Then, ca. 10 g of the ground samples were extracted in 100 mL mixture of 90 % methanol + 10 % water at 24 °C for 24 h. After filtration, the filtrate was used as a sample for further analyses.

**HPLC conditions for phenolic detection:** Instrument: Shimadzu 10Avp; Software: Shimadzu, Class-VP; Injection volume: 10 µL (10 mg oleoresin/mL methanol); Column: Nucleodur 100-5 C18 (250 × 4.6 mm, 5 µ); Mobile phase: A) % 0.5 HCOOH, B) CH<sub>3</sub>CN; Flow rate: 1 mL/min; Detector: Shimadzu SPD-M10Avp; wavelengths: 330 nm; oven temperature: 40 °C

**Total phenol content:** Folin-Ciocalteu colorimetric method were applied and the results were expressed as µg GAE/g dry sample<sup>21</sup>.

**Free radical scavenging activity:** It was determined by DPPH method<sup>22</sup> and the results were expressed as per cent inhibition of 1,1-diphenyl-2-picrylhydrazyl.

**Statistical analysis:** Results of the research were analyzed for statistical significance by analysis of variance<sup>23</sup>. The data from experiment were subjected to ANOVA using randomized complete block design with statistical analysis system-ANOVA procedure<sup>24</sup>.

## RESULTS AND DISCUSSION

The total phenol and inhibition rates of DPPH pertaining to the flower, root, stem and leaves of Buckwheat were presented in Fig. 1. Rutin contents of samples ranged from 1874.97 (Stem) to 3329.99 (leaf) mg/g. Quercetin values were found between 2.20 mg/g and 29.33 mg/g. The phenol contents ranged between 24261.82 mg (flower) to 2989.10 mg (stem). The DPPH inhibition of flower, leaf, root and stem were established as 89.71, 73.57, 56.70 and 50.85 %, respectively (Table-1).

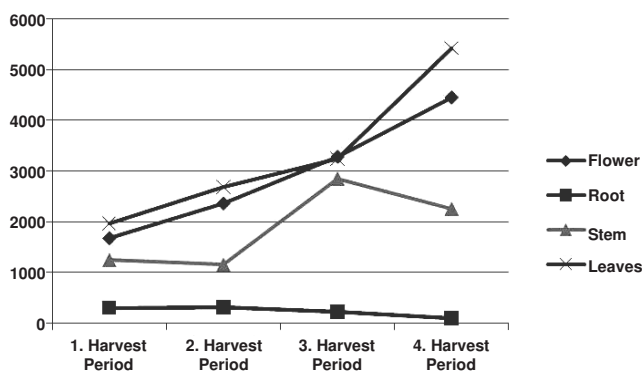


Fig. 1. Harvest period × plant parts interactions effected on rutin

The contents of rutin, quercetin, total phenol and the percentage of DPPH inhibition based on the harvest periods

TABLE-1  
CHEMICAL PROPERTIES OF DIFFERENT PARTS

Plant parts	Rutin (µg/g dry sample)	Quercetin (µg/g dry sample)	Total phenol (µg GAE/g dry sample)	% Inhibition of DPPH
Flower	2945.26a	29.33a	24261.82a	89.71a
Root	2358.80c	2.35c	4373.90c	56.70c
Stem	1874.97b	2.20c	2989.10d	50.85d
Leave	3329.99a	18.72b	13512.56b	73.57b

of Buckwheat parts are given in Fig. 2. According to this, rutin contents increased until the end of harvest period. Whereas other parameters were found high in the 1st and 4th harvest period. It was found low at the other two harvesting period. Rutin contents ranged from 1298.42 mg/g (1st harvest period) to 3056.17 Mg/g (4th harvest period) (Table-2). The values of rutin, quercetin, total phenol contents and the percentage of DPPH inhibition differed based on harvest periods.

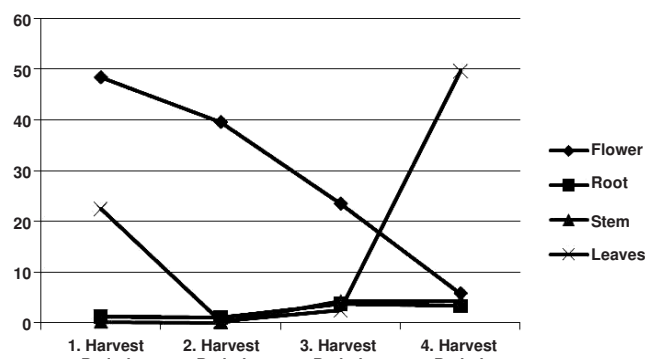


Fig. 2. Harvest period × plant parts interactions effected on quercetin

TABLE-2  
EFFECT OF HARVEST PERIOD ON CHEMICAL PROPERTIES

Harvest period	Rutin (µg/g dry sample)	Quercetin (µg/g dry sample)	Total Phenol (µg GAE/g dry sample)	Inhibition of DPPH (%)
1. Harvest period	1298.42b	18.10a	10923.89b	68.69a
2. Harvest period	1630.45b	10.26b	9780.74c	65.88b
3. Harvest period	2401.06a	8.47b	10813.33b	66.44b
4. Harvest period	3056.17a	15.76a	13619.39a	69.82a

While there is an increase rutin content of leaf and flower depending on harvest period, there was a decrease after a partial increase (Fig. 1). Whereas quercetin content decreased toward the end of harvest, it increased in root (aside from the 2nd period). While the component of the leaf is high in the 1st and 4th harvest period, it turned out to be low in the 2nd and 3rd harvest period (Fig. 2).

The effect of harvest period on the percentage of DPPH inhibition was given in Fig 3. While the percentage of DPPH inhibition rates of flower, root, stem and leaf were high at the 1st harvest period, there was fluctuation in other harvest periods. The effect of harvest period on total phenolic substance was given in Fig. 4. The highest total phenolic contents were established in flower. It was followed by leaf, root and stem in a decreasing order.

Buckwheat leaf flour contains about 2700 mg/Kg (d.w.b.) rutin and is a suitable material for enriching functional foods, giving it the potential for preventive nutrition<sup>17</sup>. Most rutin is

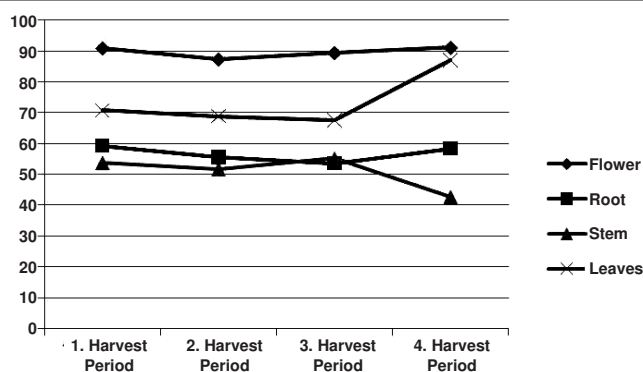


Fig. 3. Harvest period x plant parts interactions effected on inhibition % of DPPH

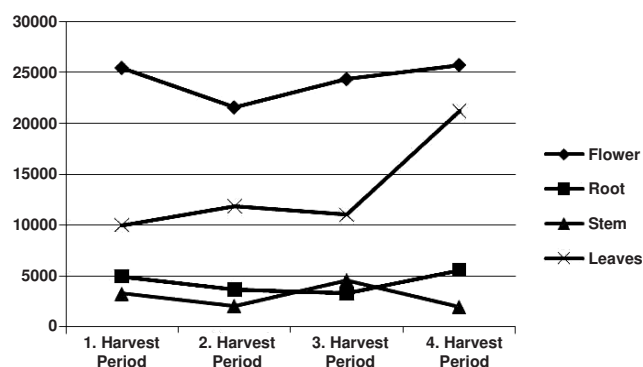


Fig. 4. Harvest period x plant parts interactions effected on total phenol

accumulated in the inflorescence (up to 12 %, d.w.b.), in stalks (0.4-1.0 %, d.w.b.) and in upper leaves (8-10 %) <sup>18</sup>. Buckwheat materials have potential, at least in regard to the rutin content, as a functional food. However, attention should be paid, during processing, to the factors which may lower the rutin content <sup>17</sup>. The rutin contents of three buckwheat species (*Fagopyrum esculentum*, *Fagopyrum tataricum* and *Fagopyrum homotropicum*) were investigated. The contents of rutin were significantly different depending on species, 0.02 % in *F. esculentum*, 0.10 % in *F. homotropicum* and 1.67 % *F. tataricum* <sup>4</sup>.

Alvarez-Jubete et al. <sup>20</sup> studied on the polyphenol composition and antioxidant properties of methanolic extracts from amaranth, quinoa, buckwheat and wheat. The total phenol content amongst the seed extracts were significantly higher in buckwheat (323.4 mg GAE/100 g). Also, buckwheat sprouted seeds showed <sup>20</sup> the highest antioxidant capacity of all sprouted seeds tested ( $p < 0.01$ ).

Total phenolics of methanol extract of buckwheat seeds <sup>12</sup> were 2.1 g catechin equivalent/100 g. Fig. 4 shows the percentage inhibition of free radical by buckwheat extracts due to hydrogen donation from the antioxidant. With the DPPH method, it seems that the inhibition percentage and total phenolics correlated significantly.

The antioxidant activity of buckwheat extract was affected by the extraction solvent and the analysis method <sup>25</sup>. The antioxidant activity of buckwheat showed promise as a food additive to replace artificial antioxidants <sup>26</sup>.

As a conclusion, the flower and leaves of buckwheat are rich in rutin, quercetin and total phenolic substance and DPPH inhibition percentage but it decreases toward the end of harvest period. Rutin and quercitrin have the same aglycone, quercetin. The edible parts of buckwheat were analyzed for their free radical-scavenging activity by DPPH assay. The radical-scavenging activities on the edible parts of green seed sprouts showed very similar values. Given their differences in phenolic levels, this suggests that overall antioxidative activity might be affected by the combination of both minor compounds and identified compounds <sup>27</sup>. In general, tissues with high flavonoid contents have high antioxidant activities <sup>28</sup>. Rutin content in buckwheat flower and leaves significantly correlated to the antioxidant activity.

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