



Seed density (n m <sup>-2</sup> )	Forage yield (g m <sup>-2</sup> )	Rutin in the forage (g kg <sup>-1</sup> ) (kg ha <sup>-1</sup> )		Quercetin in the forage (mg kg <sup>-1</sup> ) (g ha <sup>-1</sup> )	
200	180.0 b	8.4 b	15.1 b	222.2 b	400.0 b
400	304.7 a	11.6 a	35.3 a	248.6 a	757.5 a

## Rutin and quercetin in the forage and grain of common buckwheat as affected by seeding density

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**Introduction.** Flavonoids, such as rutin and quercetin, are an important class of phytochemicals products found in most fruits, vegetables, herbs and forages. Rutin is the main flavonol glycoside in common buckwheat (*Fagopyrum esculentum* Moench) and is known for its antioxidant, antimicrobial, anti-inflammatory and anti-carcinogenic properties, whose activity in humans can reduce the fragility of blood vessels, thus preventing hemorrhagic diseases and hypertension. Quercetin is an aglycone formed after enzymatic degradation of rutin by rutinoidase and, such as rutin, exerts antioxidants and antimicrobials activity. In ruminant feeding, buckwheat whole plant can be used as a diet component for dairy cows with several positive effects. Many factors can affect rutin and quercetin concentration and content in buckwheat. There is some evidence that plants synthesize rutin to enhance the defence system against environmental or biological stress conditions. However, little information is available on the role of many agronomic factors on rutin and quercetin concentrations of common buckwheat in the Mediterranean environment. The aim of this research was to evaluate the effect of two level of intraspecific competition, via two seeding densities, on the two flavonoids concentrations and yield, harvesting the plants at the optimal stages for forage and grain production.

## Results

### Forage

- Increasing seed density, the forage yield of buckwheat increased by 70%.
- At the one time, rutin concentration and content increased by approximately 38% and 135%, respectively, and quercetin concentration and content by 12% and 89%.

### Grain

- The increase of the seeding density decreased the grain yield of buckwheat by 25%.
- Flavonoid concentration in the grain was markedly lower than in the forage (by about 15-25 times reduction).
- Increasing sowing density increased both the concentration (+ 61%) and the content (+ 21%) of the rutin in the grain.
- In contrast, it decreased both the grain concentration (-19%) and content (-39%) of the quercetin.

Seed density (n m <sup>-2</sup> )	Grain yield (g m <sup>-2</sup> )	Rutin in the grain (g kg <sup>-1</sup> )	Quercetin in the grain (mg kg <sup>-1</sup> )
200	71.5 a	0.33 b	14.7 a
400	53.8 b	0.53 a	11.9 b

## Conclusions

- Increase of flavonoids at increasing the stand density were likely due to an increased stress as a consequence of the increased intraspecific competition due to an augmented stand density.
- Probably, the increase of the intraspecific competition also hampered the translocation of flavonoids to the grain and this mostly occurred for the quercetin rather than for the rutin.
- These results have implications for the utilization of buckwheat as forage, grain or industrial crop.

**Materials and Methods.** The research was carried out at the Enrico Avanzi Interdepartmental Centre of Agro-Environmental Research (CIRAA) of the University of Pisa (43° 40' N, 10° 19' E). The main soil physical and chemical characteristics were: 45.6 % sand, 10.8 % clay, pH 8.3 (1:2.5), 28.5 g kg<sup>-1</sup> soil organic matter (Walkley-Black), 1.6 g kg<sup>-1</sup> total N (Kjeldahl), 6.6 g kg<sup>-1</sup> available P (Olsen), 128.1 g kg<sup>-1</sup> available K (BaCl<sub>2</sub>-TEA method). Treatments consisted of two seed densities (200 and 400 viable achenes/m<sup>2</sup>) laid out in a randomized block design with three replications. The crops were sown in April and N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O were applied at rates of 40, 100 and 100 kg/ha, as urea, triple superphosphate and potassium sulphate, respectively. Forage harvest was performed in June, at the appearance of green achenes (stage 70 of the Arduini et.al [2016] scale) and grain harvest was performed in July, at the end of fruit ripening (stage 88). After harvest, leaves, stems, inflorescences and achenes (when present) were separated, weighted, oven dried at 65°C to constant weight for the dry matter determination. All dried samples were milled, vacuum-sealed and stored at 4°C. The analysis for the rutin and quercetin concentration were carried out by HPLC UV/vis. Rutin or quercetin content were calculated multiplying the total yield by the concentration of rutin or quercetin. Analysis of variance was performed using the software CoStat 6.4 version.