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# Amino acid composition and protein quality of Tepary bean grains (*Phaseolus acutifolius* L.) as affected by sowing date and planting pattern under semi-arid condition

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## Abstract

Planting pattern and sowing date are the eminent factors affecting plant growth and development. The two-year experiment under field conditions was conducted as split plot design in three replications during 2017 and 2018. The main plot was planting pattern in two levels (PP1: one-row, and PP2: two-row plantation pattern) and subplot was sowing date in three levels (SD1: June 15, SD2: June 30 and SD3: July 15). The results revealed that the biological yield in SD2 had significant increase by 14% and 6% in comparison to SD1 and SD3 under two-row plantation pattern in the first year respectively. In PP2, the seed yield was improved by 19% and 15% compared to SD1 and SD3 at SD2 in 2017. Carbohydrate content varied in the two years. In mono row plantation method, the grain carbohydrate content was higher in SD1 significantly. In bean plants under PP2, protein content on SD2 increased by 13% compared to SD3 in the second year of experiments. The greatest amount of essential amino acids was found in leucine followed by lysine, arginine, phenylalanine, valine, isoleucine, histidine, and threonine. We found that the concentration of essential amino acid in tepary bean grains was significantly changed by sowing date. We suggest that in tepary bean cultivation the use of plantation pattern is very effectiveness and two-row plantation pattern was the most important when the sown date delayed by late of June under same experimental environment.

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## Introduction

In recent decades, the growing population needs higher food productions. This challenge has been fortified by climate change impacts in the world particularly in developing countries.[1] Farmers use different strategies to cope with climate change impacts such as adjusting the planting dates.[2,3] To predict how farmers might respond to climate change, it would help to know when they currently plant their crops in different regions of the world. This knowledge is important for determining sowing date and planting pattern. Sowing date and planting pattern can significantly affect plant habit, yield, and quality of plants, in which sowing date significantly influences temperature.[3,4,5] In late planting, the probability of occurring the warm temperatures is high, which results in greater plants, thereby larger leaf and stem area to maintain boll development later in the season.[6] Planting pattern modifies the interception of photosynthetically active radiation, the light rate and amount within the canopy, nutrients concentration, and moisture content, and finally influences the growth and yield of plant canopy.[6]

Common bean (Pharsalus vulgaris L; 2n=22) is the most important food legume rich in protein, minerals, and vitamins where its protein content is cheap and easily affordable for the famers of the country.[7] In arid and semiarid, the crops should be resistant to different stresses like diseases, pests, and drought. The tepary bean (*Phaseolus acutifolius* L.) is an annual plant and can be climbing, trailing, or erect with stems up to 4 m long. Tepary bean consists of different local races and shows in various colors sizes.[8] Germination of tepary bean requires wet soil, but plants can flourish in dry conditions once established.[9] However, a large amount of water prohibits bean production, which induces the common disease forepart bean due to high humidity. [9]

Most parts of Iran are located in arid and semiarid areas, where most of the annual precipitation events occur from October to April. In most areas of the country, the mean annual precipitation is 25 cm or less. Only 13% of the total land area is under cultivation, while less than one-third of the cultivated area is under irrigation treatments. Total field crop production in Iran is estimated at 75 million tons on 13,500,000 ha. Cereals (wheat, barley, maize, and rice) cover about 75% of the total field crop production.[1] The bean is an important grain crop that has been recently cultivated in Iran. The total grain bean production area is approximately 112000 ha with a total production of 210000 tones.[10]

The management of sowing date and planting pattern could help the farmers to obtain the optimum production. The response of both sowing date and planting pattern are different for getting an optimum bean production. For example, Molosiwa and Kgokong[11] suggested that the best date for tepary bean is the southern part of Botswana is December and January. In the wet season of Rwanda, east Africa, the planting date should be delayed two weeks to reach the optimum bean production.[12] In Pakistan, the faba bean can be planted up to October at 450,000 plants ha$^{-1}$ to obtain the maximum yield.[13]

To our knowledge, there is no published work on the effect of sowing date and planting pattern on tepary bean under semiarid and arid condition. Hence, the present work attempts: i) to find the effect of sowing date and planting pattern on the yield (fresh and dry weight yield of plant and seed), ii) to evaluate of sowing date and planting pattern on amino acid profile.

## Materials and Methods

### Plant material and growth condition

The seeds of tepary bean were obtained from PakanBazr© seed company (Iranian seed company, Isfahan, Iran - http://www.pakan-bazr.com/fa/ContactUs). They were sown in the research farm of Agriculture and natural resource faculty, Islamic Azad University (1710 m above sea level, 34°01´45"N, 49°08´30"E) of Arak, Iran at June 2017 and 2018. The mean annual temperature was 13.5°C with the lowest and highest to be 0.0 °C in January–February and 36.0 °C in July–August, respectively. The mean annual rainfall was 340 mm (Figure 1). EC and pH of the experimental soil (0-20 cm) were respectively 0.82 ds m$^{-1}$ 2 and 7.9 (Table 1 and 2).

### Treatment details

This work was done as a split plot design in three replications during 2017 and 2018. The main plot was planting pattern in two levels (PP1: one-row, and PP2: two-row plantation) and subplot was sowing date in three levels (SD1: June 15, SD2: June 30, and SD3: July 15). During the experiment, no pesticide and chemical fertilizers were used and we manually controlled the weeds. In both years, plants were harvested at the end-pod filling stage in October and also the occurrence of flower was on September 4, pod filling stage was on September 21 and physiological maturation stage was on October 4.

### Fresh weight yield and dry weight yield

Plants were cut from the bottom of stems to measure fresh weight, and they immediately were maintained in the envelopes and dried at 40º C in the dark condition till they got a constant weight to measure dry weight.[14]

### Preparation of protein isolates

Dried seeds were cleaned and ground using a seed blender (Moulinex, type AY46, Shenzhen, Guangdong, China) to obtain a fine powder. The powder was screened using a mesh (No. 35, ASTM E11, serial number 5666533, Fritsch GMBH, Idar-Oberstein, and Germany) with an aperture size of 500 µm.

Protein isolates were prepared according to the method of Kudre *et al.*[15] Seed powder was suspended in 10 volumes of NaOH (pH 12). The mixture was stirred for 120 min at 25°C, and centrifuged (8000×g) for 30 min at 25°C (Avanti J-E centrifuge, Beckman Coulter, Inc., Fullerton, CA, USA). HCl was used to adjust the supernatant pH at 4.5. The pellet was washed with 10 volumes of distilled water adjusted to pH 4.5, followed by centrifugation at 8000×g for 30 min. The obtaining pellet was freeze-dried using a freeze dryer.

### Harvest index

HI is the relationship between seed yield and biological yield. It was calculated by using the following equation:

$$HI(\%) = \frac{\text{Seed yield}}{\text{Biological yield}} \times 100$$

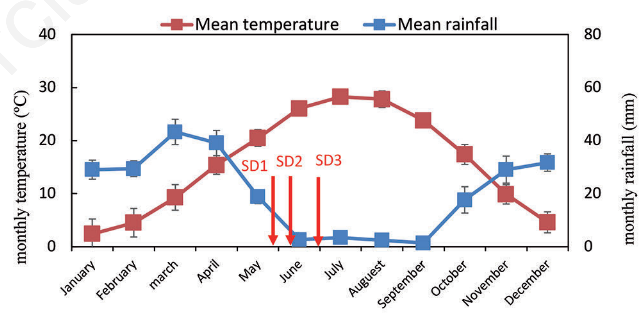


**Figure 1. Mean temperature and rainfall in the case study.**



**Table 1. Soil properties used in the case study in two soil depth (0-20 cm and 20-40 cm).**

| Year | Depth | pH | EC | OC (%) | N (%) | P (mg/kg) | K (mg/kg) | Sand (%) | Silt (%) | Clay (%) |
|---|---|---|---|---|---|---|---|---|---|---|
| 2017 | 0-20 cm | 8.1 | 0.8 | 1.1 | 0.97 | 18 | 270 | 21 | 46 | 33 |
| | 20-40 cm | 8.3 | 0.4 | 1.1 | 0.98 | 17 | 255 | 21 | 42 | 37 |
| 2018 | 0-20 cm | 8.1 | 0.9 | 1.3 | 0.96 | 21 | 273 | 20 | 47 | 33 |
| | 20-40 cm | 8.2 | 0.4 | 1.2 | 0.95 | 19 | 259 | 21 | 43 | 36 |

**Table 2. Temperature, relative humidity and rainfall changes in the case study.**

| Year | Daily mean temperature | | | | | RH | | | | | Rainfall | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | June | July | August | Sept. | October | June | July | August | Sept. | October | June | July | August | Sept. | October |
| 2017 | 25.33 | 27.72 | 26.06 | 23.07 | 16.53 | 28.5 | 42 | 27.5 | 33 | 45.5 | 0 | 0.2 | 0.01 | 0 | 0 |
| 2018 | 24.62 | 29.36 | 28.59 | 23.19 | 15.91 | 48.5 | 24.5 | 27.5 | 39 | 54.5 | 16.81 | 0 | 0 | 0.32 | 29.53 |

## Carbohydrate content

Carbohydrate was measured using the method of Opsah and Benner;[16] 0.02 g of dried plant tissue with 3 ml of 80% alcohol were centrifuged for 15 minutes, and the mixture was used to measure soluble sugars. So one ml of the extract containing the soluble carbohydrates was poured into the tube and one ml of 5% phenol solution was added and stirred well. In the final step, 5 mL of concentrated sulfuric acid was added to each tube. The tubes were incubated at room temperature for half an hour and the absorption was determined at 480 nm by a spectrophotometer. Using standard curve, the sugar content was calculated in grams per 100 gr dry plant tissue.

### Amino acid analysis

Amino acid profiles were determined with an HPLC system after samples were hydrolyzed with 6 M HCl according to the method described by Bidlingmeyer *et al*.[17]

### Statistical analysis

The data ($n$=3) were subjected to a one-way analysis of variance (ANOVA) using the SAS software package for Windows (SAS, version 9.3, SAS Institute, Cary, NC). Duncan's multiple range tests ($p<0.05$) were used for mean comparison.

## Results

### Plant yield

The biological yield was affected by the planting pattern and sowing date ($p\leq0.05$; Table 3). In the first year under PP2, biological yield in SD2 increased by 14% and 6% in comparison to SD1and SD3, respectively (Table 3). PP2 was slightly had higher biological yield relative to PP1. In the second year and SD1, PP2 increased by 5% compared to PP1 (Table 3).

The seed yield was influenced by planting pattern and sowing date ($p\leq0.05$, Table 3). Under PP2, the seed yield of plants sown on SD2 of 2017 was improved by 19% and 15% compared to SD1 and SD3 of 2017 (Table 3). Besides, the pod yield as an effective factor in bean growth was affected by year, sowing date, and the interaction of year and planting pattern ($p\leq0.05$, Table 3). A 10% increase of pod yield in plants under PP1 sown on SD1 in the first year was pronounced in comparison to the second year (Table 3). In terms of sowing date, pod yield was different in tow experimental years. For example, under PP1, the highest pod yield was found on SD1 for the first year, but on SD2 for the second year (Table 3).

### Harvest index

HI was not statistically influenced by the experimental factors ($p\leq0.05$, Table3). The highest HI (45.8±1) was obtained in a two-row plantation on SD2 2017. In contrast, the lowest HI (39.4±2.7) was found inPP1 on SD1 2018 (Table 3).

### Carbohydrate and protein content

Carbohydrate content had different varied in two years. In one row plantation, carbohydrate content on SD1 2018 was higher compared to SD1 2017. However, it was vice versa on SD2. In both first and second years under two-row plantation, SD2 was more effective than SD1 (Figure 2). Protein content was significantly changed under sowing date. In bean plants under PP2 in the second year, protein content onSD2 increased by 13% compared to SD3 (Figure 3).



**Table 3. The plant yield under planting patter and sowing date during 2018 and 2019.**

| Year (Y) | Planting pattern (PP) | Sowing date (SD) | Biological yield (kg/ha) | Seed yield (kg/ha) | Pod yield (kg/ha) | Harvest index (%) |
|---|---|---|---|---|---|---|
| 2017 | one-row | June 15 | 1673.3±27g | 716.7±33ef | 951.7±65a | 42.8±1.8ab |
| | | June 30 | 2030.0±57ab | 860.0±21b-d | 919.0±67a-c | 42.4±0.5ab |
| | | July 15 | 1873.3±61c-e | 773.3±30de | 878.7±11b-e | 41.3±2.1ab |
| | two-row | June 15 | 1846.7±40d-f | 810.3±15cd | 878.0±9b-e | 43.9±1.5ab |
| | | June 30 | 2116.7±62a | 970.0±35a | 892.3±2b-d | 45.8±1.2a |
| | | July 15 | 1990.0±15a-c | 843.3±26b-d | 865.7±5c-e | 42.4±1.4ab |
| 2018 | one-row | June 15 | 1726.7±55fg | 680.0±45f | 863.3±12de | 39.4±±2.7b |
| | | June 30 | 1976.7±71b-d | 873.0±17bc | 923.3±25ab | 44.3±2.4ab |
| | | July 15 | 1880.0±88c-e | 830.3±37cd | 830.0±10e | 44.2±1.1ab |
| | two-row | June 15 | 1826.7±52ef | 799.0±59c-e | 862.7±11de | 43.8±2.9ab |
| | | June 30 | 2066.7±47ab | 925.0±26ab | 890.3±4b-d | 44.8±0.8ab |
| | | July 15 | 1963.3±26b-d | 828.7±39cd | 871.3±12b-e | 42.2±2.1ab |
| Significance levels | | | | | | |
| Y | | | ns | ns | ** | ns |
| PP | | | ** | ** | ns | ns |
| Y*PP | | | ns | ns | * | ns |
| SD | | | ** | ** | ** | ns |
| Y*SD | | | ns | ns | ns | ns |
| PP*SD | | | ns | ns | ns | ns |
| Y*PP*SD | | | ns | ns | ns | ns |

Means in same columns followed by the same letter were not significantly different at the 5% level; *P=0.05; **P=0.01; ns, not significant.

## Correlations among seed yield, carbohydrate, and amino acid composition

Biological yield was positively correlated with protein (r=0.496). We observed a significant correlation between seed yield and protein (r=0.615).[18] HI was positively correlated with protein content (r=0.499). Close correlation was also found for carbohydrate content with EAA (r=0.442), NAA (r=0.444), and TAA (r=0.448) (Table 4). Similar slopes and correlation coefficients between seed protein supported the view that higher Essential Amino Acids (EAA), Non-Essential Amino Acids (NAA), and Total Amino Acids (TAA) amino acid contents are generally achieved under higher protein content (Figure 4).

## Amino acid profile

The highest concentration of essential amino acids was found in leucine followed by lysine, arginine, Phenylalanine, valine, isoleucine, histidine, and threonine (Figure 5). The essential amino acids *viz.*, leucine, isoleucine, lysine, histidine, threonine, Phenylalanine, valine, arginine, and tyrosine were identified in tepary bean (Table 5 and 6). Histidine ($C_6H_9N_3O_2$) was significantly affected by sowing date. Histidine in PP2 on SD2 of 2018 (13.72 g/kg) was greater than other experimental treatments. Isoleucine ($C_6H_{13}NO_2$) was influenced by triple interaction of year, planting pattern, and sowing date. In the first year under PP1, the plants sown on SD2 had greater amount of isoleucine than other sowing dates.

Leucine ($C_6H_{13}NO_2$) was significantly changed under sowing date, and the interaction of years and sowing date. It ranged from PP1 on SD1 2017 as 35.91 g kg$^{-1}$ protein to PP1 on SD2 2017 to be 37.69 g kg$^{-1}$ protein. Lysine ($C_6H_{14}N_2O_2$) was significantly affected by planting pattern. Its concentration in SD2 of 2017 was higher compared to other experimental treatments. Threonine ($C_4H_9NO_3$) had no significant change under sowing date and planting pattern (Table 5). Triple interaction of year, planting pattern, and sowing date was significant on phenylalanine ($C_9H_{11}NO_2$). In first year under PP2, SD3 increased phenylalanine amount by 5% compared to SD1. Valine ($_5H_{11}NO_2$) varied from 23.78 to 24.48 gkg$^{-1}$ protein. Arginine ($C_6H_{14}N_4O_2$) was influenced by triple interaction of year, planting pattern, and sowing date. We found the arginine concentration from the first year under two-row plantation on SD1to the first year under PP2 on SD3, ranging from 28.5 to 29.75 g kg$^{-1}$ protein. The interaction of year and sowing date was significant on tyrosine ($C_9H_{11}NO_3$). It differed from PP2 on SD3 of 2018 toPP1 on SD2 of 2017 (Table 6).

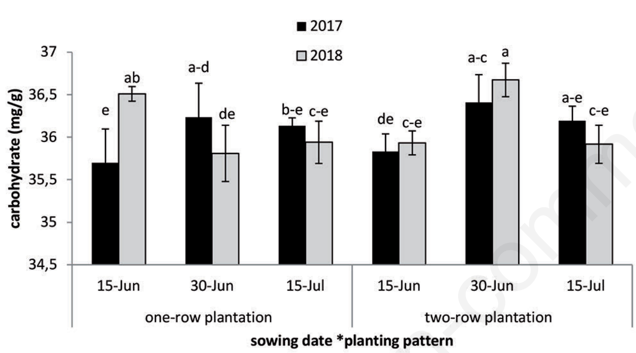


**Figure 2. Carbohydrate content of tepary bean under planting pattern and sowing date during 2017 and 2018.**

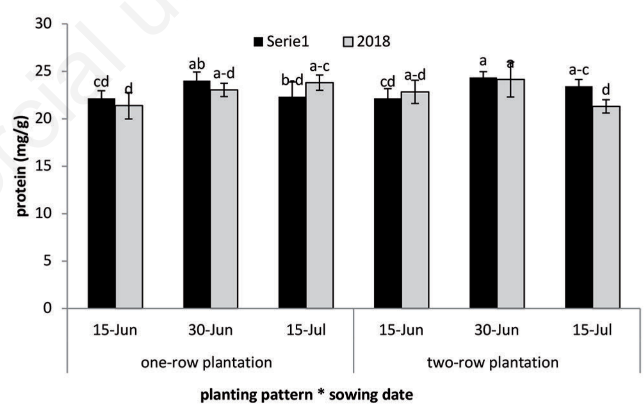


**Figure 3. Protein content of tepary bean under planting pattern and sowing date during 2017 and 2018.**

**Table 4. Correlations among experimental traits under planting patter and sowing date.**

| | Seed weight | Biological yield | Seed yield | HI | Pod yield | Protein | Carbohydrate | EAA | NAA | TAA |
|---|---|---|---|---|---|---|---|---|---|---|
| Seed weight | 1 | | | | | | | | | |
| Biological yield | -0.226 | 1 | | | | | | | | |
| Seed yield | -0.203 | 0.827** | 1 | | | | | | | |
| HI | -0.083 | 0.194 | 0.710** | 1 | | | | | | |
| Pod yield | 0.157 | -0.055 | 0.29 | 0.126 | 1 | | | | | |
| Protein | -0.077 | 0.496** | 0.615** | 0.499** | 0.041 | 1 | | | | |
| Carbohydrate | 0.016 | 0.294 | 0.247 | 0.040 | 0.80 | 0.316 | 1 | | | |
| EAA | -0.172 | 0.293 | 0.151 | -0.117 | 0.241 | 0.332* | 0.442** | 1 | | |
| NAA | -0.179 | 0.277 | 0.226 | 0.045 | 0.030 | 0.389* | 0.444** | 0.479** | 1 | |
| TAA | -0.191 | 0.319 | 0.185 | -0.090 | 0.234 | 0.379* | 0.488** | 0.978** | 0.653* | 1 |

**P≤0.01; *P≤0.05; EAA, Essential Amino Acids; NAA, Non-essential Amino Acids; TAA, Total Amino Acids.

## Discussion

Sowing date is an important factor for tepary bean production. In our study, the SD2 crops significantly produced higher yield than early and late sown crops, which can be explained by the fact that SD2 crops could take advantage of soil moisture and nutrients for the longer growing season and produced more biomass.[19] In contrast, SD2 crops experienced a short reproductive period due to increased air temperatures and reduced canopy photosynthesis due to less radiation interception.[5,20] SD2 was an effective time for obtaining the higher protein and carbohydrate contents. Recently, the effect of sowing date has been reported on yield and protein content of sunflower (sowing date in sunflower was on November 10, November 20, November 30, December 10, December 20) and quinoa had winter sowing and mid-spring, and also soybean had different sowing date (early, mid, late).[21-23] In the present study, the response of amino acid was different from sowing date (in this study sowing date was on June 15, June 30 and July 15). In this regard, histidine, isoleucine, leucine, valine, and tyrosine were higher in SD2, but phenylalanine and arginine were greater in SD3. Hence based on our purposes, we can manage the amino acid profile to meet the optimum concentration. Sowing date changes the environmental conditions (Table 2) for grain, which may affect the corresponded protein content and composition.[24] Mlakar *et al.*[25] declared the change of grain protein content and the sum of essential amino acids under the growing season and sowing date.

We found that panting pattern can change the plant yield and amino acid profile. The dry matter of plants at the same planting density but in different patterns varies at the time of ripening.[26] We explained that the important factor corresponding to improved yield in PP2 plants was a consequence of the better light-intercepting characteristics of the canopy in plants of this planting pattern. The photosynthetic features of individual leaves are eminent determinants of plant production at canopies. These results revealed that the capacity of leaf photosynthesis was influenced by planting pattern, which results in a change of plant growth. We did not observe a determined trend under planting pattern for the amino acid profile. For example, Lysine, which is used for preventing and treating cold sores in PP1, was greater than PP2. The negative response of some amino acids to more desirable environmental conditions, such as greater water content and solar radiation, could be associated with the decline of protein concentration, which is a result of the accumulation of relatively greater oil concentration in seeds. Highly negative correlate between oil and protein content support this approach, and also, we can mention the negative correlation between yield and protein content.[27,28] A positive correlation between high solar radiation and yield during the seed filling period is probably because of an increase in seed weight.[29] This was supported by the previous results, in which there was a negative correlation between seed yield and protein amount.[27]

The fine-scale correlation between bean seed yield and the seed protein content can provide valuable information on how to optimize cultivation management. Wilcox and Shibles[27] showed a negative

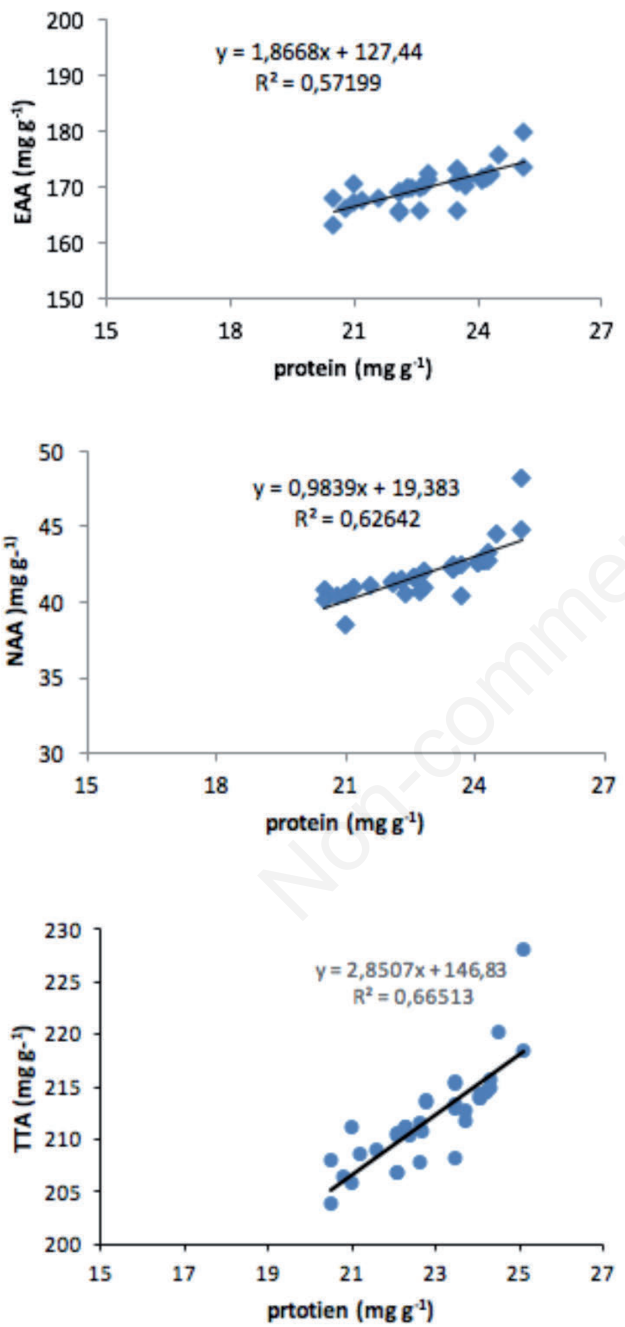


**Figure 4. Relationships between seed protein content and Essential Amino Acids (EAA), Non-essential Amino Acids (NAA) and Total Amino Acids (TAA) content.**

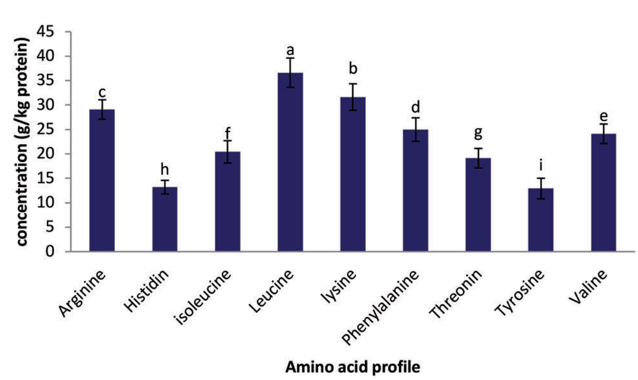


**Figure 5. The concentration of amino acid composition of tepary bean.**

correlation between wheat grain yield and protein content (r=-0.117) under different tillage and residue management in a three-year field experiment. However, our results revealed that in two rows cultivation, the highest protein content, carbohydrate content and grain yield were obtained in both years from sowing date June 30. In addition, a positive correlation was obtained between grain yield and protein content, carbohydrate content, EAA, NEAA and total amino acids (Table 4). Since sowing date is a crucial factor in bean production particularly in semiarid areas, it is strongly important in management of yield and protein content compare.[29] Tosti and Guiducci[30]

**Table 5. The amino acid profile under planting patter and sowing date during 2018 and 2019.**

| Year (Y) | Planting pattern (PP) | Sowing date (SD) | Histidine ($g\ kg^{-1}$ protein) | Isoleucine ($g\ kg^{-1}$ protein) | Leucine ($g\ kg^{-1}$ protein) | Lysine ($g\ kg^{-1}$ protein) | Threonine ($g\ kg^{-1}$ protein) |
|---|---|---|---|---|---|---|---|
| 2017 | one-row | June 15 | 13.01b | 20.45a-d | 35.91c | 31.87ab | 19.51a |
| | | June 30 | 13.52ab | 20.86a | 37.69a | 32.34a | 19.39ab |
| | | July 15 | 12.92b | 20.35b-d | 36.66a-c | 31.65ab | 19.38ab |
| | two-row | June 15 | 13.11ab | 20.12cd | 36.00bc | 31.08b | 19.03ab |
| | | June 30 | 13.44ab | 20.54ab | 37.04ab | 31.35ab | 19.76a |
| | | July 15 | 13.41ab | 20.67ab | 36.70a-c | 31.61ab | 19.13ab |
| 2018 | one-row | June 15 | 12.91b | 20.31b-d | 36.58bc | 31.88ab | 19.48a |
| | | June 30 | 13.43ab | 20.34b-d | 36.54bc | 31.57ab | 19.12ab |
| | | July 15 | 12.96b | 20.20cd | 36.70a-c | 31.70ab | 17.24b |
| | two-row | June 15 | 13.23ab | 20.24b-d | 36.72a-c | 31.53ab | 19.11ab |
| | | June 30 | 13.72a | 20.87a | 36.70a-c | 31.73ab | 19.25ab |
| | | July 15 | 13.14ab | 20.08d | 36.14bc | 31.41ab | 19.09ab |
| **Significance levels** | | | | | | | |
| Y | | | ns | ns | ns | ns | ns |
| PP | | | ns | ns | ns | * | ns |
| Y*PP | | | ns | ns | ns | ns | ns |
| SD | | | ** | ** | ** | ns | ns |
| Y*SD | | | ns | ns | ** | ns | ns |
| PP*SD | | | ns | ns | ns | ns | ns |
| Y*PP*SD | | | ns | * | ns | ns | ns |

Means in same columns followed by the same letter were not significantly different at the 5% level; *P=0.05; **P=0.0; ns, not significant.

**Table 6. The amino acid profile under planting patter and sowing date during 2018 and 2019.**

| Year (Y) | Planting pattern (PP) | Sowing date (SD) | Phenylalanine ($g\ kg^{-1}$ protein) | Valine ($g\ kg^{-1}$ protein) | Arginine ($g\ kg^{-1}$ protein) | Tyrosine ($g\ kg^{-1}$ protein) |
|---|---|---|---|---|---|---|
| 2017 | one-row | June 15 | 25.04cd | 24.39b | 28.60d | 13.01ab |
| | | June 30 | 25.22b | 24.48a | 29.63ab | 13.10a |
| | | July 15 | 25.70a | 24.22a-d | 29.07a-d | 12.92a-c |
| | two-row | June 15 | 24.42e | 23.79d | 28.5d | 12.69bc |
| | | June 30 | 25.17b-d | 24.30a-c | 29.18a-d | 13.00a-c |
| | | July 15 | 25.62ab | 24.05a-d | 29.75a | 13.07a |
| 2018 | one-row | June 15 | 24.91c-e | 24.26a-d | 28.94b-d | 13.05a |
| | | June 30 | 24.70de | 23.78d | 28.81cd | 12.84a-c |
| | | July 15 | 25.08cd | 24.14a-d | 28.80cd | 12.91a-c |
| | two-row | June 15 | 24.87c-e | 23.92b-d | 29.62ab | 12.90a-c |
| | | June 30 | 24.67de | 24.30a-c | 29.39a-c | 12.92a-c |
| | | July 15 | 24.43e | 23.82cd | 28.59d | 12.66c |
| **Significance levels** | | | | | | |
| Y | | | ns | ns | ns | ns |
| PP | | | ** | * | ns | ns |
| Y*PP | | | ns | ns | ns | ns |
| SD | | | ** | ns | ns | ns |
| Y*SD | | | ** | ns | ** | * |
| PP*SD | | | ns | * | ns | ns |
| Y*PP*SD | | | * | ns | * | ns |

Means in same columns followed by the same letter were not significantly different at the 5% level; *P=0.05; **P=0.01; ns, not significant.

also observed a favorite impact on grain yield and protein after merging teabag bean into the soil (improved N availability for the cereal component). Therefore, to a certain extent, late June and two-row plantation are an efficient way of increasing the protein content without causing yield reductions.

## Conclusions

The present study attempted to find the improvement use of planting pattern and sowing date on yield, carbohydrate content and amino acid profiles of tepary bean during 2017 and 2018. We found that the seeds sown on June 30 have higher yield and concentration of essential amino acid compared to other dates. In terms of planting pattern, there are no significant differences in amino acid profile, but the plant yield in a two-row plantation was slightly higher than a one-row plantation. Hence, we can prefer the use of two-row plantation, when the tepary bean seeds are sown on the late of June in semiarid conditions.

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