



## PHYTOCHEMICAL AND NUTRITIONAL PROFILING OF INDIGENOUS SOYBEAN VARIETIES; A POTENTIAL PLANT-BASED PROTEIN SOURCE TO ALLEVIATE MALNOURISHMENT

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### Abstract

Soybean (*Glycine max*) is an excellent source of plant-based protein with a substantial amount of minerals, fatty acids, and some therapeutic substances such as isoflavones, flavonoids, etc. Many people in developing countries suffer from protein-energy malnourishment due to the unavailability of a balanced diet or lack of protein in food. Several Pakistani soybean varieties have been introduced, but their use as food is not very common as people are concerned about their characteristics and quality. Hence, in this study, native soybean varieties were studied for nutritional quality to assess their potential as food sources. Eight different Pakistani Soybean varieties were assessed for their nutritional aspects including chemical composition, mineral content, free radical activity, amino acid assay, and fatty acid profile. Statistical analysis was performed to analyze the level of significance. The protein content for all the varieties was seen within the range of 36% to 43%. Amino acid analysis showed an appreciable profile with a good amount of all the essential amino acids. Moreover, the fatty acid profile showed the highest value of oleic acid and linoleic acid in V<sub>5</sub> (Malakand-96) and V<sub>8</sub> (Swat-84) at 15.590% and 59.863% respectively. However, for linolenic, palmitic, and stearic acid, the highest value was seen for V<sub>1</sub> (Ajmeri) at 7.066%, 14.677%, and 5.190% respectively. Overall, findings suggested that all the varieties have a satisfactory nutritional profile with the potential to be used as an effective source of plant-based protein to combat the increasing protein demand and meet the nutritional needs of people.

**Keywords:** Soybean; protein energy malnourishment; nutritional profile; amino acid assay; fatty acid profile

### 1. INTRODUCTION

Soybean (*Glycine max Merr.*) is a globally important oilseed crop, that belongs to the *Leguminosae* family and originated in China around five thousand years ago (Dixit et al., 2011). It is one of the oldest Far East crops with annual worldwide production of 1.266 MT (Misal et al., 2018). A huge amount of soybean produced in the world is used for the production of feed for animals. Moreover, it is also utilized as food for humans as its products are considered one of the most effective sources of protein and certain bioactive compounds like flavonoids, saponins, peptides, hemagglutinins, and

lectins. The proximate composition of soybean constitutes about 16% moisture, 5% ash, 40% protein, 36% carbohydrates, and 20% oil and 8% fiber respectively (Liu et al., 2020). Furthermore, it is said to be an excellent source of minerals, dietary proteins, mono and polyunsaturated fatty acids, and some therapeutic substances such as isoflavones and flavonoids, etc. (Maythem et al., 2019).

Plant protein is gaining a lot of popularity among people and is being encouraged as an essential constituent in the daily diet in place of animal protein due to its potential health benefits and cheap sources (Saeed et al., 2022). Due to the presence of complete protein with all the essential amino acids, soybean is known to be the best source of plant protein and is highly recommended against protein deficiency diseases such as kwashiorkor, marasmus, protein energy malnourishment, etc. (Messina et al., 2017). Protein-energy malnutrition (PEM) is one of the most significant types of malnutrition that arises from the lack of dietary protein or energy. It is one of the most prevalent nutritional complexities in developing countries as it contributes to nearly half of the total deaths of children, under 5 years of age in South Asia and Africa (Javed et al., 2021). To overcome this globally prevalent issue, one should focus on the management of balanced and sufficient nutrients with the right kind of protein in the diet (Aadil et al., 2023)..

In general, soybean is considered the cheapest source of plant-based protein along with all the other essential nutrients. However, in developing countries, its use is far less due to a lack of awareness regarding its benefits and usage. There are several studies conducted on the nutritional quality of soybeans around the world but Pakistani varieties have not been reported for their nutritional profile and characteristics. Therefore, this study was designed to determine the nutritional profile of the Pakistan soybean and the estimation of its potential for product development because its consumption is a dire need in today's time, especially in developing countries, where many people suffer from protein deficiency issues, leading towards malnourishment.

## **2. MATERIALS AND METHODS**

### **2.1. Material**

Soybean varieties including NARC-1, William-82, and Ajmeri were procured from the National Agriculture Research Centre (NARC) in Islamabad, Pakistan. Faisal soybean was procured from Ayyub Agriculture Research Institute (AARI), Faisalabad, Pakistan. Swat-84, Swat-18, Malakand-96, and Rawal were acquired from Swat Research Institute, Mingora Swat, Pakistan. All the Chemicals and reagents were acquired from Merck (Germany), Sigma-Aldrich, and Cayman Chemicals Europe, Estonia.

#### **2.1.1. Preparation of raw material**

All the procured varieties were grounded into fine powder and then subjected to different analyses to test them for their particular characteristics to select the best varieties for further use.

### **2.2. Chemical analysis**

All the procured varieties were analyzed for their chemical composition according to the methods described by AACC (2016).

### **2.3. Titratable acidity**

The titratable Acidity of all the varieties was determined by the titrimetric method as described in Paez et al. (2016). 10 ml sample from each treatment was taken and diluted with 100 ml ethanol. 2-3 drops of phenolphthalein indicator were added and then titrated against 0.1N NaOH until the light pink color appeared which persisted for 30 seconds.

## 2.4. Mineral analysis

The mineral content of soybean varieties was estimated for calcium (Ca), magnesium (Mg), and iron (Fe) according to the method demonstrated by Szostak et al. (2020). Minerals were analyzed by atomic absorption spectrophotometer (Varian AA240, Victoria, Australia).

## 2.5. Antioxidant assay

The antioxidant activity of all the soybean varieties was assessed based on their ability to scavenge DPPH (1,1-diphenyl-2-picrylhydrazyl) through the method reported by Chen et al. (2014). For this purpose, 0.5 ml of the extract of each variety was mixed with 0.25 ml of 0.5 mmol DPPH sol. and 0.5 ml of 100 mmol acetate buffer at pH 5.5. After that, the mixture was left for almost 30 minutes and the first absorbance was taken at 515 nm against a blank sample through a spectrophotometer containing only 0.5ml ethanol, right after the reaction was initiated. This process was done in triplicates to obtain the accurate results. The free radical (DPPH scavenging) activity is expressed as % inhibition.

## 2.6. Amino acid analysis

The amino acid composition of all the varieties was analyzed through an amino acid analyzer (Biochrom 30+) by the method given in Moldes et al. (2017). For amino acid assay, soybean seeds were finely ground into powder form. To break the peptide linkages, hydrolysis was done by the addition of a sample into a 6M HCl solution. and was heated at 100°C for 24 hours. After hydrolysis, the separation of amino acids was done through ion exchange chromatography. Following that, quantification of amino acids was performed through the addition of ninhydrin into all the separated amino acids. However, for cysteine and methionine, performic acid was added as a reagent. After that, a 1.5 ml solution of each mixture was poured into cuvettes to test for light absorption at the wavelength of 260 to 280 nanometers in the amino acid analyzer.

## 2.7. Fatty acid profile

All the soybean varieties were assessed for their fatty acid profile. The fat was extracted from all the varieties through the Soxhlet apparatus with hexane as a solvent, followed by rotary evaporation. The fatty acid methyl esters were prepared and held at 4°C before the analysis. 100µL ±5µL of oil sample was taken and added into a test tube, preloaded with n-hexane (5mL), and briefly shaken to dissolve the mixture. After that sodium methoxide solution (250µL) was added to the mixture, followed by vigorous vortex shaking for 1min and then set for 10 secs for vortex to break. After that, 5mL of saturated NaCl soln was added to the mixture and shaken for 15 mins for complete esterification of fatty acids. The mixture was then allowed to be set for 10 mins and three layers appeared after that, the top layer was then separated into a clean vial with a small amount of sodium sulfate. The vials containing esterified fatty acids were then assayed for the five essential fatty acids (oleic, linoleic, linolenic, palmitic, stearic) through GC (Agilent Technologies 6890N Network GC System) with the flame ionized detector. The chromatographic separation was performed on an RTX-WAX column (Restek, Germany, 30 m length × 0.25 mm internal diameter × 0.25 mm thickness). The oven temperature was as follows: after the sample injection, the temp was set at 60°C for 2 to 3 mins and then it was raised to 185°C for 10 °C m<sup>-1</sup> for 1min and then further raised to 200 °C with 5°C m<sup>-1</sup> for 10 mins and then finally increased to 220°C with 5°C m<sup>-1</sup> heating ramp for 20 mins. Injector temperature: 250°C, detector temperature: 275°C, carrier gas: Nitrogen; inlet pressure, 40.65 psi; linear gas velocity, 39cm/s; column flow rate, 2.7mL/m in; split ratio, 40:1; injected volume, 1 µL.

## 2.8. Statistical analysis

All the analyses were done in triplicates and the results obtained were then statistically analyzed through a completely randomized design (CRD). Analysis of variance (ANOVA) was applied to check the level of significance among all the values. Beyond ANOVA was performed through

Tuckey HSD and the results were interpreted at a 5% probability level according to the method described by Montgomery (2019). Results are presented as means  $\pm$  standard errors, which were calculated through Microsoft Excel (Microsoft, Redmond, WA, USA), and the software used for statistical analysis was Statistics 8.1.

### 3. RESULTS AND DISCUSSION

#### 3.1. Chemical composition

The results for the chemical composition of all the soybean varieties are presented in Table 1. A significant difference was seen among all the varieties. The highest amount of moisture and ash was seen in V<sub>7</sub> (Swat-18) and V<sub>1</sub> (Ajmeri) respectively while the highest amount of protein was observed in V<sub>6</sub> (Rawal) i-e  $42.67 \pm 0.02\%$ . The fat content of soybean varieties ranged from  $19.06 \pm 0.02\%$  to  $24.06 \pm 0.01\%$  where V<sub>2</sub> showed the highest amount of fat while for fiber content the highest value was observed in V<sub>1</sub> ( $7.17 \pm 0.01\%$ ), while the lowest in V<sub>7</sub> ( $5.34 \pm 0.01\%$ ).

Although, all the cultivars showed a good chemical composition still there was a significant difference observed in their values about their region and the physiological conditions of their growth. A previous study done by Zhu et al. (2018a) reported the change in structure, function, and composition of different soybean cultivars due to genetic factors. Similarly, McClure et al. (2017) determined the effect of different climatic conditions on 10 different cultivars of soybean and the results suggested that the nutritional composition varies greatly with the change in environment and showed a significant difference among the nutritional contents (especially oleic and linoleic acid) of all the cultivars with different climatic conditions. Wijewardana et al. (2019) also revealed that the chemical composition of soybeans varies significantly with the change in cultivars due to certain factors including soil type, moisture stress, and other climatic factors. (Zhu et al., 2018b) reported the quality parameters of soybeans through near-infrared spectroscopy, which indicated the protein content to be somewhat close to the current study.

#### 3.2. Mineral Analysis

All the varieties were investigated for minerals including iron, calcium, and magnesium and the results are given in Table 1. A significant difference was observed in the mineral content of all the varieties. The highest amount of iron content ( $0.38 \pm 0.02$  mg/g) was found in V<sub>6</sub> (Rawal) while the calcium content was observed within the range of  $3.13 \pm 0.02$  mg/g to  $4.12 \pm 0.02$  mg/g. The highest value of calcium content was seen for V<sub>5</sub> (Malakand-96) while the lowest one was for V<sub>2</sub> (Faisal soybean). In the case of magnesium, the mean values were found within the range of  $1.56 \pm 0.02$  mg/g to  $2.52 \pm 0.02$  mg/g with the highest value for V<sub>6</sub> (Rawal) and the lowest for V<sub>2</sub> (Faisal soybean).

According to the results, almost all the varieties showed a good amount of all three minerals but V<sub>6</sub> was seen to be the highest for iron and magnesium, which might be due to its genetic features and growing conditions as Wijewardana et al. (2019) reported the mineral content of soybean to be affected by environmental conditions including moisture deficiency, heat cold stress and many other like these. Soares et al. (2019) studied the change in mineral composition of soybean in response to elevated CO<sub>2</sub> and reported that calcium content increases significantly under elevated CO<sub>2</sub> while iron and magnesium remain unaffected.

#### 3.3. Antioxidant activity

The results for the radical scavenging activity of all the soybean varieties are given in Table 1. The highest value was seen to be for V<sub>5</sub> (Malakand-96) at  $78.06 \pm 0.04\%$  and the lowest for V<sub>1</sub> (Ajmeri) at  $65.02 \pm 0.02\%$ . Statistical analysis revealed a significant difference among all the varieties. As per the results, all the cultivars showed a great antioxidant potential but V<sub>5</sub> (Malakand-96) showed the highest antioxidant activity among all the varieties, which mainly refers to the origin difference as reported by Sakthivelu et al. (2008) who studied the antioxidant capacity of soybean seeds from

different area including India and Bulgaria and indicated a considerable difference among all those varieties, based on their origin.

**Table 1. Chemical composition, mineral content, and antioxidant activity of soybean cultivars**

| Varieties        | V <sub>1</sub>          | V <sub>2</sub>          | V <sub>3</sub>           | V <sub>4</sub>           | V <sub>5</sub>          | V <sub>6</sub>          | V <sub>7</sub>          | V <sub>8</sub>          |
|------------------|-------------------------|-------------------------|--------------------------|--------------------------|-------------------------|-------------------------|-------------------------|-------------------------|
| Moisture (%)     | 7.76±0.01 <sup>d</sup>  | 8.40±0.03 <sup>c</sup>  | 8.26±0.02 <sup>c</sup>   | 7.93±0.02 <sup>d</sup>   | 9.07±0.01 <sup>b</sup>  | 10.34±0.01 <sup>a</sup> | 10.44±0.01 <sup>a</sup> | 9.19±0.01 <sup>b</sup>  |
| Ash (%)          | 5.14±0.02 <sup>a</sup>  | 4.87±0.02 <sup>bc</sup> | 4.64±0.01 <sup>d</sup>   | 4.96±0.01 <sup>ab</sup>  | 4.73±0.02 <sup>cd</sup> | 4.36±0.02 <sup>e</sup>  | 4.24±0.02 <sup>e</sup>  | 5.02±0.01 <sup>ab</sup> |
| Protein (%)      | 40.6±0.02 <sup>b</sup>  | 36.27±0.04 <sup>f</sup> | 39.84±0.03 <sup>cd</sup> | 40.23±0.01 <sup>bc</sup> | 40.5±0.02 <sup>c</sup>  | 42.67±0.02 <sup>a</sup> | 38.56±0.03 <sup>c</sup> | 39.57±0.01 <sup>d</sup> |
| Crude Fat (%)    | 19.06±0.02 <sup>b</sup> | 24.06±0.01 <sup>a</sup> | 22.30±0.02 <sup>d</sup>  | 23.19±0.01 <sup>e</sup>  | 22.70±0.02 <sup>c</sup> | 20.71±0.01 <sup>f</sup> | 19.43±0.01 <sup>g</sup> | 21.46±0.01 <sup>e</sup> |
| Fiber (%)        | 7.17±0.01 <sup>a</sup>  | 5.63±0.01 <sup>c</sup>  | 5.49±0.02 <sup>ef</sup>  | 5.48±0.01 <sup>f</sup>   | 5.57±0.02 <sup>cd</sup> | 6.68±0.01 <sup>b</sup>  | 5.34±0.01 <sup>g</sup>  | 5.56±0.03 <sup>de</sup> |
| NFE (%)          | 28.05±0.02 <sup>c</sup> | 29.15±0.04 <sup>b</sup> | 27.73±0.03 <sup>c</sup>  | 26.13±0.04 <sup>e</sup>  | 26.88±0.05 <sup>d</sup> | 25.58±0.05 <sup>e</sup> | 32.42±0.03 <sup>a</sup> | 28.39±0.04 <sup>c</sup> |
| Iron (mg/g)      | 0.31±0.02 <sup>bc</sup> | 0.29±0.01 <sup>c</sup>  | 0.26±0.01 <sup>ab</sup>  | 0.28±0.01 <sup>c</sup>   | 0.37±0.01 <sup>a</sup>  | 0.38±0.02 <sup>a</sup>  | 0.32±0.02 <sup>bc</sup> | 0.29±0.03 <sup>c</sup>  |
| Calcium(mg/g)    | 3.32±0.01 <sup>f</sup>  | 3.13±0.02 <sup>g</sup>  | 3.94±0.02 <sup>c</sup>   | 3.68±0.01 <sup>d</sup>   | 4.12±0.02 <sup>a</sup>  | 4.04±0.03 <sup>b</sup>  | 3.16±0.01 <sup>g</sup>  | 3.51±0.01 <sup>e</sup>  |
| Magnesium (mg/g) | 2.05±0.02 <sup>d</sup>  | 1.56±0.02 <sup>g</sup>  | 2.33±0.01 <sup>b</sup>   | 1.92±0.02 <sup>e</sup>   | 2.24±0.02 <sup>c</sup>  | 2.52±0.02 <sup>a</sup>  | 2.00±0.01 <sup>d</sup>  | 1.67±0.02 <sup>f</sup>  |
| DPPH (%)         | 65.02±0.02 <sup>b</sup> | 73.05±0.05 <sup>c</sup> | 75.04±0.04 <sup>b</sup>  | 68.07±0.05 <sup>f</sup>  | 78.06±0.04 <sup>a</sup> | 71.02±0.04 <sup>e</sup> | 72.36±0.02 <sup>d</sup> | 67.75±0.02 <sup>g</sup> |

V<sub>1</sub>: Ajmeri, V<sub>2</sub>: Faisal soybean, V<sub>3</sub>: NARC-2, V<sub>4</sub>: Wiliam-82, V<sub>5</sub>: Malakand-96, V<sub>6</sub>: Rawal, V<sub>7</sub>: Swat-18, V<sub>8</sub>: Swat-84

### 3.4. Amino acid analysis

All the varieties of soybean were assessed for their amino acid composition through an amino acid analyzer and the results are presented in Table 2,3.

#### ➤ Essential amino acids

The results of the amino acid analysis showed that soybean consists of all the essential amino acids in considerable amounts with the highest amount of lysine and methionine in V<sub>1</sub>(Ajmeri) and V<sub>6</sub> (Rawal) respectively. The highest value of histidine was shown by V<sub>1</sub> (1.34±0.01g/100g) while the lowest value was observed for V<sub>7</sub> (0.78±0.02 g/100g). Isoleucine, leucine, and valine, termed branched-chain amino acids were found in greater amounts in V<sub>6</sub>, V<sub>5</sub>, and V<sub>3</sub> respectively. The results showed the amount of phenylalanine within the range of 2.06±0.02g/100g to 2.4±0.01g/100g with the highest amount for V<sub>6</sub> and the lowest one for V<sub>4</sub>. Threonine and tryptophan are two other essential amino acids and the results for these two amino acids showed the highest value for V<sub>6</sub> (Rawal).

According to the results, a significant difference was observed among all the varieties and V<sub>6</sub> (Rawal) was seen with the highest amount of almost all the essential amino acids, which might refer to its environmental conditions. Carrera et al. (2011) reported that environmental conditions as one of the prime factors in the variation of physicochemical traits among different cultivars corresponding to the interaction between their genotype and environment. Protein concentration also plays a significant role in the variation of amino acids among different varieties (Pfarr et al., 2018) A previous study conducted on the quality screening of soybean and amino acid content of different imported cultivars (Aykas et al., 2020) showed the amino acid composition to be around our study for yellow soybean, which makes these Pakistani varieties equally competitive and useful in terms of protein quality and amino acid composition to the already existing imported cultivars.

#### ➤ Non-essential amino acids

The results of this study showed all the varieties to have a considerable amount of non-essential amino acids as shown in Table 3. Aspartic acid and glutamic acid are important non-essential amino acids and V<sub>1</sub> (Ajmeri) had the highest value for these two amino acids. Another two significant non-essential amino acids are alanine and arginine. Both of these are synthesized by glucose in the body and the results revealed that V<sub>6</sub> (Rawal) and V<sub>7</sub> (Swat-18) showed the highest amount of alanine and arginine respectively. In the case of glycine, V<sub>4</sub> (William-82) showed the highest results while V<sub>7</sub> (swat-18) had the lowest amount of glycine. proline and serine, on the other hand, were found highest in V<sub>6</sub> (Rawal) and V<sub>8</sub> (Swat-84) respectively. Tyrosine is the only non-essential amino acid that is not synthesized by glucose but by phenylalanine, which is an essential amino acid and is taken from the diet as not produced in the body. The amount of tyrosine ranged from

2.07±0.02g/100g to 2.57±0.02g/100g with the highest value for V<sub>6</sub> (Rawal) while for cysteine, V<sub>1</sub> (Ajmeri) and V<sub>8</sub> (Swat-84) showed the best results.

Statistical results of this study indicated that the difference in cultivars poses a significant impact on the number of amino acids and protein quality. A study done by Pfarr et al. (2018) on the determination of the effect of increasing protein concentration on amino acids revealed that with the increase in the protein concentration in seed, glutamic acid increases in the soybean, which mainly depends upon the change in the cultivar. Li et al. (2021) studied the amino acid composition of different foodstuffs including soybean and showed soybean to be one of the best sources of plant protein with almost all the non-essential amino acids, as described in the current study.

| Amino acids   | V <sub>1</sub>          | V <sub>2</sub>          | V <sub>3</sub>          | V <sub>4</sub>          | V <sub>5</sub>          | V <sub>6</sub>           | V <sub>7</sub>         | V <sub>8</sub>          |
|---------------|-------------------------|-------------------------|-------------------------|-------------------------|-------------------------|--------------------------|------------------------|-------------------------|
| Lysine        | 2.38±0.01 <sup>a</sup>  | 2.14±0.01 <sup>d</sup>  | 1.85±0.01 <sup>cd</sup> | 2.17±0.01 <sup>cd</sup> | 2.23±0.02 <sup>bc</sup> | 2.2±0.01 <sup>c</sup>    | 2.06±0.01 <sup>e</sup> | 2.28±0.01 <sup>b</sup>  |
| Methionine    | 0.66±0.02 <sup>ab</sup> | 0.65±0.01 <sup>b</sup>  | 0.67±0.01 <sup>ab</sup> | 0.66±0.01 <sup>ab</sup> | 0.66±0.02 <sup>ab</sup> | 0.73±0.02 <sup>a</sup>   | 0.47±0.02 <sup>c</sup> | 0.67±0.01 <sup>ab</sup> |
| Isoleucine    | 1.74±0.01 <sup>d</sup>  | 1.64±0.02 <sup>e</sup>  | 1.79±0.02 <sup>cd</sup> | 1.88±0.01 <sup>b</sup>  | 1.84±0.02 <sup>bc</sup> | 1.96±0.02 <sup>a</sup>   | 1.75±0.01 <sup>d</sup> | 1.75±0.02 <sup>d</sup>  |
| Leucine       | 2.58±0.02 <sup>a</sup>  | 2.26±0.01 <sup>d</sup>  | 2.54±0.02 <sup>b</sup>  | 2.65±0.01 <sup>a</sup>  | 2.60±0.01 <sup>ab</sup> | 2.66±0.01 <sup>a</sup>   | 2.43±0.01 <sup>c</sup> | 2.47±0.01 <sup>c</sup>  |
| Histidine     | 1.34±0.01 <sup>a</sup>  | 1.14±0.01 <sup>b</sup>  | 1.07±0.01 <sup>c</sup>  | 0.98±0.01 <sup>d</sup>  | 0.94±0.01 <sup>d</sup>  | 0.87±0.01 <sup>e</sup>   | 0.78±0.02 <sup>f</sup> | 1.20±0.01 <sup>b</sup>  |
| Valine        | 0.48±0.02 <sup>a</sup>  | 0.48±0.01 <sup>ab</sup> | 0.5±0.02 <sup>ab</sup>  | 0.47±0.02 <sup>ab</sup> | 0.47±0.01 <sup>ab</sup> | 0.46±0.02 <sup>abc</sup> | 0.41±0.01 <sup>c</sup> | 0.44±0.01 <sup>bc</sup> |
| phenylalanine | 2.13±0.02 <sup>d</sup>  | 2.13±0.02 <sup>d</sup>  | 2.28±0.01 <sup>bc</sup> | 2.06±0.02 <sup>e</sup>  | 2.3±0.01 <sup>b</sup>   | 2.4±0.01 <sup>a</sup>    | 2.14±0.01 <sup>d</sup> | 2.23±0.01 <sup>c</sup>  |
| Threonine     | 0.44±0.02 <sup>b</sup>  | 0.43±0.02 <sup>b</sup>  | 0.46±0.02 <sup>b</sup>  | 0.46±0.02 <sup>b</sup>  | 0.47±0.01 <sup>b</sup>  | 0.57±0.02 <sup>a</sup>   | 0.42±0.01 <sup>b</sup> | 0.44±0.01 <sup>b</sup>  |
| Tryptophan    | 1.59±0.01 <sup>d</sup>  | 1.53±0.02 <sup>e</sup>  | 1.63±0.01 <sup>c</sup>  | 1.74±0.01 <sup>b</sup>  | 1.69±0.01 <sup>bc</sup> | 1.85±0.01 <sup>a</sup>   | 1.48±0.01 <sup>e</sup> | 1.66±0.02 <sup>c</sup>  |

V<sub>1</sub>: Ajmeri, V<sub>2</sub>: Faisal soybean, V<sub>3</sub>: NARC-2, V<sub>4</sub>: Wiliam-82, V<sub>5</sub>: Malakand-96, V<sub>6</sub>: Rawal, V<sub>7</sub>: Swat- 18, V<sub>8</sub>: Swat-84

|          |                         |                          |                          |                         |                          |                         |                        |                         |
|----------|-------------------------|--------------------------|--------------------------|-------------------------|--------------------------|-------------------------|------------------------|-------------------------|
| Aspartic | 4.42±0.01 <sup>ab</sup> | 4.02±0.03 <sup>d</sup>   | 4.27±0.01 <sup>c</sup>   | 4.32±0.02 <sup>c</sup>  | 4.30±0.01 <sup>c</sup>   | 4.35±0.02 <sup>a</sup>  | 3.64±0.01 <sup>e</sup> | 4.33±0.02 <sup>bc</sup> |
| Glutamic | 5.60±0.01 <sup>a</sup>  | 4.48±0.01 <sup>f</sup>   | 5.30±0.02 <sup>c</sup>   | 4.97±0.02 <sup>e</sup>  | 5.06±0.01 <sup>d</sup>   | 5.53±0.01 <sup>b</sup>  | 4.24±0.01 <sup>g</sup> | 5.27±0.01 <sup>c</sup>  |
| Alanine  | 2.00±0.01 <sup>de</sup> | 1.96±0.01 <sup>e</sup>   | 2.06±0.01 <sup>ed</sup>  | 2.12±0.02 <sup>b</sup>  | 2.08±0.01 <sup>bc</sup>  | 2.23±0.02 <sup>a</sup>  | 1.62±0.01 <sup>f</sup> | 2.02±0.01 <sup>de</sup> |
| Arginine | 2.93±0.02 <sup>b</sup>  | 2.63±0.01 <sup>c</sup>   | 2.95±0.02 <sup>b</sup>   | 2.95±0.01 <sup>b</sup>  | 2.95±0.02 <sup>b</sup>   | 2.96±0.02 <sup>a</sup>  | 3.15±0.01 <sup>b</sup> | 2.91±0.01 <sup>b</sup>  |
| Glycine  | 2.45±0.01 <sup>e</sup>  | 2.66±0.02 <sup>d</sup>   | 2.78±0.03 <sup>c</sup>   | 2.97±0.02 <sup>a</sup>  | 2.88±0.01 <sup>b</sup>   | 2.96±0.02 <sup>a</sup>  | 2.35±0.01 <sup>f</sup> | 2.66±0.01 <sup>d</sup>  |
| Proline  | 2.38±0.01 <sup>cd</sup> | 2.46±0.01 <sup>e</sup>   | 2.44±0.02 <sup>bc</sup>  | 2.50±0.01 <sup>b</sup>  | 2.40±0.01 <sup>cd</sup>  | 2.73±0.02 <sup>a</sup>  | 2.21±0.02 <sup>e</sup> | 2.70±0.01 <sup>d</sup>  |
| Serine   | 1.78±0.01 <sup>de</sup> | 1.80±0.01 <sup>cde</sup> | 1.96±0.02 <sup>abc</sup> | 1.98±0.01 <sup>ab</sup> | 1.95±0.02 <sup>bcd</sup> | 1.05±0.02 <sup>ab</sup> | 1.65±0.02 <sup>e</sup> | 2.13±0.01 <sup>a</sup>  |
| Tyrosine | 2.27±0.01 <sup>d</sup>  | 2.26±0.02 <sup>d</sup>   | 2.38±0.03 <sup>c</sup>   | 2.07±0.02 <sup>f</sup>  | 2.47±0.01 <sup>b</sup>   | 2.57±0.02 <sup>a</sup>  | 2.16±0.02 <sup>e</sup> | 2.37±0.02 <sup>a</sup>  |
| Cysteine | 0.53±0.02 <sup>a</sup>  | 0.46±0.01 <sup>bc</sup>  | 0.46±0.01 <sup>bc</sup>  | 0.38±0.02 <sup>d</sup>  | 0.44±0.02 <sup>cd</sup>  | 0.52±0.01 <sup>ab</sup> | 0.26±0.01 <sup>e</sup> | 0.53±0.02 <sup>c</sup>  |

V<sub>1</sub>: Ajmeri, V<sub>2</sub>: Faisal soybean, V<sub>3</sub>: NARC-2, V<sub>4</sub>: Wiliam-82, V<sub>5</sub>: Malakand-96, V<sub>6</sub>: Rawal, V<sub>7</sub>: Swat- 18, V<sub>8</sub>: Swat-84

### 3.5. Fatty acid profile

All the soybean varieties were assessed for five essential fatty acids including oleic, linoleic, linolenic, palmitic, and stearic, and the results are presented in Table 4. The findings suggested that all the varieties showed a good amount of all these fatty acids with the values ranging between 11.770% to 15.590% for oleic acid. The highest value of oleic acid was observed in V<sub>5</sub> (Malakand-96) and the lowest value was seen in V<sub>4</sub> (William-84). For Linoleic acid, the highest value was observed for V<sub>8</sub> (Swat-84); 59.863%, and the lowest one for V<sub>1</sub>(Ajmeri); 55.760%. the values for linolenic were observed between 6.033% and to7.066% with the highest one for V<sub>1</sub> (Ajmeri). In the case of palmitic and stearic acid, the highest values were seen for V<sub>1</sub> (Ajmeri) as 14.677% and 5.190% respectively. Statistical analysis revealed all the varieties showed a significant difference for oleic, linoleic, and palmitic acid with a p-value less than 0.05 while in the case of linolenic and stearic acid, no significant was observed among the values of all the varieties.

The findings of this study are somewhat similar to some previous investigations made on the fatty acid profile of different soybean varieties from various origins. The results of a study conducted by Abdelghany et al. (2020) on the fatty acid profile of different varieties of Chinese origin are relevant to the findings of the current study with values ranging from 3.9%, 7.4%, 12.4%, 23.4%, and 52.8% for stearic, linolenic, palmitic, oleic and linoleic acid respectively.

**Table 4. Fatty acid profile of soybean varieties**

| Treatments     | oleic                   | Linoleic                | linolenic              | palmitic                | Stearic                |
|----------------|-------------------------|-------------------------|------------------------|-------------------------|------------------------|
| V <sub>1</sub> | 14.14±0.01 <sup>c</sup> | 55.76±0.02 <sup>h</sup> | 7.46±0.02 <sup>a</sup> | 14.67±0.02 <sup>a</sup> | 5.19±0.01 <sup>a</sup> |
| V <sub>2</sub> | 15.10±0.01 <sup>b</sup> | 56.36±0.01 <sup>g</sup> | 6.03±0.02 <sup>e</sup> | 14.36±0.02 <sup>b</sup> | 5.17±0.01 <sup>a</sup> |
| V <sub>3</sub> | 12.92±0.03 <sup>e</sup> | 57.94±0.02 <sup>c</sup> | 7.12±0.03 <sup>b</sup> | 14.03±0.01 <sup>c</sup> | 5.02±0.02 <sup>b</sup> |
| V <sub>4</sub> | 15.59±0.04 <sup>a</sup> | 57.47±0.01 <sup>d</sup> | 6.14±0.02 <sup>d</sup> | 13.05±0.01 <sup>f</sup> | 4.76±0.02 <sup>d</sup> |
| V <sub>5</sub> | 11.77±0.01 <sup>g</sup> | 58.83±0.02 <sup>b</sup> | 7.18±0.01 <sup>b</sup> | 14.07±0.01 <sup>c</sup> | 5.16±0.01 <sup>a</sup> |
| V <sub>6</sub> | 11.57±0.01 <sup>h</sup> | 56.64±0.02 <sup>f</sup> | 7.16±0.01 <sup>b</sup> | 13.90±0.02 <sup>d</sup> | 4.90±0.02 <sup>c</sup> |
| V <sub>7</sub> | 12.25±0.02 <sup>f</sup> | 57.24±0.01 <sup>e</sup> | 6.45±0.01 <sup>c</sup> | 13.24±0.01 <sup>e</sup> | 5.14±0.01 <sup>a</sup> |
| V <sub>8</sub> | 13.39±0.01 <sup>d</sup> | 59.86±0.02 <sup>a</sup> | 7.15±0.01 <sup>b</sup> | 12.89±0.01 <sup>g</sup> | 4.87±0.01 <sup>c</sup> |

V1: Ajmeri, V2: Faisal soybean, V3: NARC-2, V4: Wiliam-82, V5: Malakand-96, V6: Rawal, V7: Swat-18, V8: Swat-84

#### 4. CONCLUSION

Soybean is one of the most significant oilseed crops with excellent nutritional profile. However, its use as a food commodity is not very common in developing countries like Pakistan. There is very little work done on the native Pakistani soybean varieties in terms of their nutritional significance and quality. In the current study, eight local Pakistani varieties including Ajmeri, Faisal soybean, NARC-2, Wiliam-82, Malakand-96, Rawal, Swat- 18, and Swat-84 were studied to check their potential as a food source to meet the increasing protein demand in developing countries. Characterization of varieties was done through various analyses including chemical, mineral, amino acid assay, and fatty acid profile. All the varieties showed a great chemical and mineral composition along with an excellent amino acid profile and a good amount of unsaturated fatty acids. Four varieties (Ajmeri, NARC-2, Malakand-96, Rawal) were termed as the best varieties based on the overall results but the rest of the cultivars also showed satisfactory results with very little difference. Overall, all the varieties showed an excellent nutritional profile with great potential to serve as one of the best alternatives to animal protein and one of the most beneficial and healthiest food options.

#### Declaration

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