Characterization of local sorghum (*Sorghum bicolor* L.) population grains in terms of nutritional properties and evaluation by GT Biplot approach

Mahmut Kaplan[a,*], Hasan Kale[b], Yusuf Murat Kardes[c], Kevser Karaman[d], Kevser Kahraman[e], Mehmet Fatih Yılmaz[f], Ridvan Temizgül[g], Taner Akar[h]

[a]Department of Field Crops, Faculty of Agriculture, University of Erciyes, Kayseri, Turkey

[b]Department of Crops and Animal Science, Bogazliyan Vocational School, University of Bozok, Yozgat, Turkey

[c]Department of Field Crops, Faculty of Agriculture, University of Bilecik Seyh Edebali, Bilecik, Turkey

[d]Department of Agricultural Biotechnology, Faculty of Agriculture, University of Erciyes, Kayseri, Turkey

[e]Department of Material Science and Nanotechnology Engineering, Faculty of Engineering, University of Abdullah Gul, Kayseri, Turkey

[f]East Mediterranean Transitional Zone Agricultural Research of Institute, Kahramanmaras, Turkey

[g]Department of Biology, Faculty of Science, University of Erciyes, Kayseri, Turkey

[h]Department of Field Crops, Faculty of Agriculture, University of Akdeniz, Antalya, Turkey

**Corresponding author: mahmutkaplan5@hotmail.com*

Abstract

This study was conducted to characterize some nutritional attributes and starch properties of 156 Turkish sorghum populations and 4 standard cultivars (*Sorghum bicolor* L.). Crude protein contents of the populations varied between 6.67 and 14.33%, digestible protein ratios between 6.67 and 82.24%, crude oil contents between 2.15 and 6.40%, phytic acid contents between 0.37 and 4.09%, resistant starch between 1.10 and 34.23%, nonresistant starch between 10.79 and 79.61%, total starch between 15.42 and 85.54%, amylose between 5.67 and 43.48%, amylopectin between 9.45 and 65.67%, total phenolic between 0.19 and 5.06 mg GAE/g and antiradical activity between 3.72-91.48%. Significant differences were obtained from starch based Rapid Visco Analyzer parameters of sorghum genotypes. As compared standard cultivars, several superior genotypes were identified in terms of nutritional characteristics. GT biplot analysis revealed ideal genotypes for investigated parameters. Present findings confirmed that there were many genotypes with superior nutritional properties in local sorghum genotypes.

Keywords: Sorghum, nutritive value, phenolic, pasting properties, GT biplot

1. Introduction

Sorghum, which is more resistant to different environmental stresses and whose production is generally more economical compared to other cereals, [1] is the fifth most important grain in the world [2]. Sorghum is an crucial source providing starch, protein, some vitamins and minerals for daily diets of poor people in the developing countries of the world [3]. For that reason, the richness of nutritional composition (protein, starch, vitamin contents, etc.) and highness of digestibility levels in sorghum grains consumed by the people have crucial importance. Due to the low protein digestibility of sorghum grains limits its use of food material, it should be characterized the sorghum genotypes having high nutritional value and digestibility potential [4]. Even though tannins and phenolic substances of sorghum provide resistance to the grains against the unfavorable conditions, they have an undesired effect on the nutritional compositions of the grains. But, in some locations in the world, there is cultivation for the sorghum grains having high tannin content which are stable for the adverse conditions to prevent the diseases [5]. Phenolic substances are active natural antioxidants because they inhibit free radicals and lipoxygenase enzyme and they are chelating agents [6]. Sorghum contains many phytochemicals having antioxidant activity but it is not very well known despite the fact that it helps to decrease cholesterol in the body like fruits and vegetables [1]. In the literature, it was reported that sorghum provided an important resistance against the cancer cells, [7] showed a positive effect on coronary heart diseases and decreased the cholesterol level of the body [8,9,10]. It was suggested that the consumption of sorghum as a whole grain is important for the health because many phytochemicals, dietary fibers, minerals present in the shell and embryo of the sorghum grain to decrease the negative aspects of the diseases [11].

Starch is an integral part of the sorghum grains, but it decreases the digestibility because of the interaction between starch and protein. [4,12] Low starch digestibility is not desired for human and animal nutrition in the case of energy absence. Sorghum grains having waxy endosperm contain almost 100% amylopectin,[13] and they have low protein and high starch content [14]. Sandstedt et al. [15] reported that the waxy sorghum starch digestibility was higher than that of regular sorghum starch. In general, germination and growing of the waxy hybrid genotypes were determined to be weaker compared to samples having typical starch composition [16]. Taking into account all of these reasons, determination of amylase and amylopectin levels of sorghum grains to guess both digestibility and germination ability and stability against deterioration have an important place in grain selection process.

Cereals could be used in the production of ethanol (biodiesel) because of their high starch level (70% starch for sorghum) [17]. The presence of starch in sorghum decreases the protein digestibility [18]. For that reason, the grains are used in ethanol production by the increase of their starch content instead of protein level [19] and the rest of the samples are valued as animal feed. For these purposes, chemical and nutrient content analyzes are conducted intensively in sorghum populations for breeding studies [20].

At present, different sorghum genotypes more than 7000 were identified. [21] The selection of varieties that meet specific local food and industrial requirements from this tremendous-high biodiversity is of great importance for food safety [22]. The researchers investigate different properties for many genotypes by performing breeding and characterization studies. Performing a selection of more than one feature makes the breeders busy too much when the number of investigated property and genotype was too high. To contribute to the selected studies in this kind of research, different statistical methods were developed. Biplot analysis, which is of these techniques provides excellent facilities for plant breeders and growers. Biplot analysis, which graphically displays bi-directional data is a multivariate analytical technique that allows to visualization of the interrelationships between the examined properties of genotypes or the properties of the genotypes which are the main improvement target [23,24]. Also, GT is an application of GGE biplot to evaluate genotype comparison and selection of different properties [25].

Although the sorghum was cultivated in large areas for a long time ago in Turkey, day by day, its agricultural activities decreased slowly. In recent years, sorghum is commonly used in human and animal nutrition because of its nutritional properties despite its decreased agriculture activity. Sorghum is a prominent plant because of its uses as food in temperate climates dominated by Turkey's coastal regions and areas with significant genetic diversity. In addition to all these, we could not find detailed research about the nutritional properties of local genotypes present too much in Turkey.

The main aims of the current research are 1) to determine the biochemical properties of local sorghum genotypes, 2) to characterize the different genotypes in terms of their nutritional characteristics, 3) to select the superior genotypes in terms of critical nutritional parameters by using GT biplot analysis approach to use in the breeding studies in the coming years.

2. Materials and methods

2.1 Field experiments and samples

In the present study, 190 local sorghum genotypes and 4 standard sorghum varieties (Öğretmenoğlu, Akdarı, Beydarı and Rox) were used as experimental material. Sorghum genotypes were provided from different province as follows: 106 items from National Plant Germplasm System (USDA), 52 items from International Crop Research Institute for the Semi-Arid Tropics (ICRISAT), 10 items from Ege Agricultural Research Institute (ETAE) and 22 items from different cities where sorghum agricultural activity was performed in Turkey (Antalya, Hatay, Muğla, Gaziantep, Kilis, Şanlıurfa, Burdur, Diyarbakır and Mardin). The characterization measurements were conducted for only 156 genotypes because the rest of the genotypes could not complete their vegetation term due to their adaptation problem to the ecological conditions of the research region. The field experiment was conducted on the areas of Agricultural Faculty at Erciyes University (Turkey). The seeds were planted according to the augmented experimental design. The experiment was established as 10 replicates (in each block, 4 standard varieties were replicated), and 230 plots (10 block and 19 genotypes and 4 varieties in each block) were prepared. The plants were sowed in 5 m long, 70x15 cm wide, and 4 rows. 120 kg/ha P_2O_5 and 200 kg/ha N were incorporated in to the soil according to the quality analysis results of the soil. All of the phosphorus fertilizer and the half of the nitrogenous fertilizer were given with sowing, while the other half of the nitrogenous fertilizer was given when the plants reach a length of 30-40 cm. Middle breaking and weeding were performed after the plants reached a length of 30-40 cm. Two times weeding and one-time agricultural spraying were performed during the plant developing. The sprinkler systems irrigated the plants in the germination in the first development period, and drip irrigation was applied after the plants reached a length of 30-40 cm. Irrigation per week was performed to eliminate the absence of useful water after the determination of field capacity. During flowering, the clusters were isolated by using paper-bag for each genotype daily, and seed amplification was conducted by appropriation. The seeds were harvested separately during the hard ripening period. The temperature in the year in which the experiment was conducted was generally similar to the temperature during many years, and the amount of precipitation was higher than the precipitation occurred for long years. It was observed that the relative humidity was generally lower than that of the long years. Unexpected short-term low temperatures were observed during harvesting process. Soil samples were taken from 0-30 cm and 30-60 cm depth in the soil of the experimental area. The soil samples taken were classified as sandy loam, lime and salt ratio was low and it was rich in potassium and phosphorus. The pH of the soil was slightly alkaline and organic matter was a little low (Table 1).

2.2. *Biochemical Assays*

The sorghum samples harvested at the stage of the hard-ripening term were ground (IKA MF 10.1, Staufen, Germany) and stored at +4 ºC for biochemical analysis. All analyses were duplicated with three replications.

2.2.1. *Crude protein*

To determine the crude protein content of the samples, the Kjeldahl method was applied. The calculated N level was multiplied with 6.25 to calculate the crude protein content of the grain samples. [26]

2.2.2. *Crude oil (Ether extract)*

To determine the ether extract level of the samples, the Soxhlet extraction system was used. For this purpose, 3 g milled grain sample was placed in a cellulosic Soxhlet cartridge and petroleum ether was used as an extraction solvent. At the end of the oil extraction, the oil samples were kept at 95 ºC for 1 h to evaporate the residual solvent and the ether extract level was calculated using the mass balance system. [26]

2.2.3. *Pepsin protein digestibility*

The in vitro pepsin digestibility level of the sample proteins were conducted according to the methodology described by some researchers [27,28]. For this aim, 200 mg of milled samples was weighed into the Erlenmeyer and mixed with 35 ml of porcine pepsin solution (1.5 g/L pepsin content in 0.1 M KH_2PO_4 buffer, pH 2.0, Sigma P-7000, 890 U/mg protein activity, Sigma Chemical Co., St. Louis, MO). After that, the samples were incubated for 2 h in a shaking water bath at 37 ºC to complete the digestion. At the end of the incubation, the digestion was stopped by adding 2 ml of 2N NaOH solution. Then the samples were centrifuged at 4000 *g,* and +4 ºC for 20 min, and the supernatant was removed. The pellets were washed with buffer (0.1 M KH_2PO_4, pH 7.0) two times and again centrifuged. The undigested N was analyzed by using a Kjeldahl nitrogen analyzer.

2.2.4. *Total phenolic content*

To determine the total phenolic content, the samples (0.5 g) were extracted with 25 ml of 1% HCl/methanol (v/v) for 2 h. Then a filtration was applied and 0.1 ml of filtrated extract was mixed with 1.1 ml distilled water. Finally, 0.4 ml Folin Ciocalteu reagent (diluted 1/10 with distilled water) and 0.9 ml of 0.5 M ethanolamine were added to the tubes, and all tubes were incubated at room temperature for 20 min. At the end of the incubation, the absorbances were measured at 600 nm, and total phenolic contents of the samples were calculated using a calibration curve as mg GAE/g sample [29].

2.2.5. *Antiradical activity*

The antiradical activity of the samples was determined using DPPH (2,2-diphenyl-1-picrylhydrazyl) method [30]. For this purpose, the samples were extracted using 70% acetone for 2 h in a shaking water bath. Then the samples were centrifuged at 2790 *g* for 15 min, and the supernatant was collected. To prevent the oxidation, the samples were kept in dark conditions at -20 ºC. Similarly, DPPH solution was prepared using methanol and stored at dark conditions. In the analysis, 150 µl of sample extract was mixed with 2850 µl of DPPH solution and final mix was vortexed and incubated for 30 min. At the end of the incubation, the absorbance values were determined at 517 nm. % inhibition value showing the antiradical activity was calculated using the following formula:

$$\%\text{Inhibition} = ((\text{Abs}_{\text{control}} - \text{Abs}_{\text{sample}})/\text{Abs}_{\text{control}})*100$$

2.2.6. *Total, resistant and non-resistant starch content*

Resistant starch contents were determined with the aid of Megazyme Resistant Starch Assay (K-RSTAR, Megazyme International Ireland Ltd, Co. Wicklow, Ireland) kit developed based on AOAC 2002. 02 Method and AACC 32-40 Method. Resistant starch and total starch values were separately obtained through determining glucose spectrophotometrically.

2.2.7. *Amylose/Amylopectin ratio*

To determine the amylose and amylopectin levels of the starch part in sorghum grains, Megazyme Amylose/Amylopectin Kit (K-AMYL) was used. The samples were treated with dimethyl sulfoxide to disperse the starch content of the samples in a boiling water bath and then ethanol was incorporated to precipitate the starches. The precipitated starch was dissolved in acetate and conA solution was added. Then the mix was centrifuged to remove amylopectin part. Finally, amylose and total starch were hydrolyzed to D-glucose, and the total glucose level was determined using glucose oxidase/peroxidase addition to calculate the amylose content.

2.2.8. *Phytic acid analysis*

Phytic acid content of the samples was determined using Megazyme Phytic acid Kit (K-PHYT). By using this kit, no purification process by anion changing was not needed. In this analysis, total phosphorus obtained with phytase and alkaline phosphates and Myo-inositol phosphate forms as IP were determined and the results were given as g/100 g.

2.2.9. *Pasting properties*

Pasting properties of the samples were analyzed by Rapid Visco-Analyzer (RVA 4, Perten Inst., Australia) using STD1 pasting profile. In this assay, 3.5 g sample (14% moisture basis) was mixed with distilled water to make a total mixture weight of 28.0 g. The sample was equilibrated at 50°C for approximately 1 min, heated to 95°C for 3 min 42 sec, held at 95°C for 2.5 min, cooled down to 50°C for 3 min 48 sec, kept at 50°C for 2 min. The constant rotating speed of the paddle was 160 rpm. Pasting data were acquired using Thermocline Software (Perten Inst., Australia). Pasting tests were repeated twice for each sample, and RVA parameters ((peak 1 (cP); trough 1 (cP); breakdown (cP); final viscosity (cP); setback (cP); peak time (min); pasting temperature (°C)) were determined.

2.3. *Data analysis*

Experimental data were subjected to variance analysis with Statistical Analysis System Software 9.0 (SAS Institute). Analyses of variance of the biochemical composition of sorghum landraces were achieved using a Least Significant Difference (LSD) multiple comparison procedure with a 1% significance level. To determine which genotype or genotypes are the best in terms of which nutrient content, and to determine the genotypes to be used in breeding studies to be developed in order to increase the nutrient content and quality in sorghum, genotypes biplot graphics were generated. [24]

3. Results

Minimum, maximum and mean values of biochemical analysis results for genotypes were given in Table 2. The effect of genotypes showed significant effect on biochemical parameters. Crude oil (ether extract), crude protein, pepsin protein digestibility, phytic acid, resistant starch, non-resistant starch, total starch, amylose level, amylopectin level, total phenolic content and antiradical activity values were determined as in the ranges of 2.15-6.40%, 6.67- 14.33%, 6.71-82.24, 0.37-4.09%, 1.10-34.23%, 10.79-79.61%, 15.42-85.54%, 5.67-43.48%, 9.45-65.67%, 0.9-5.06 mg GAE/g and 3.72-91.48%, respectively.

Pasting properties of the samples were shown in Table 2. The peak, trough and final viscosities, breakdown, and setback values of the genotypes varied between 657-4162, 566-2422 and 756-6473 cP, 92-2344 and 190-4051, respectively. In addition to that, peak time and pasting temperature of the samples varied between 3.90-6.41 min and 50.28-93.30 °C, respectively.

Biplot analysis was used to identify the best genotype groups in terms of genotypes or chemical properties by comparing them based on the biochemical properties of local sorghum genotypes. The interactions of the properties and their interactions with each other can be obtained from the vector image of GT-biplot using the interrelationships of the biochemical properties examined in sorghums. The property having a long vector can separate the genotypes accurately and quickly. Besides, the relationship between the angle present among the vectors and properties examined were investigated (Figure 1). According to this, crude oil and amylose levels showed the lowest separating performance among the studied properties. Based on the biplot graphics, three groups were created according to the relationship among the studied properties. Non-resistant starch, amylose, amylopectin and, starch parameters like trough 1, peak 1, final viscosity, breakdown and setback parameters placed on first group and total phenolic content, antiradical activity and resistant starch placed on second group and crude oil, phytic acid, peak time, pepsin protein digestibility, crude protein and pasting temperature placed on third group. A positive and significant relationship with pepsin protein digestibility and non-resistant starch was observed in the Biplot chart as an exceptional case (Figure 1) Genotype treatment (GT) biplot analysis could be used to determine the relationships among the examined properties of local sorghum genotypes and which genotypes are superior in terms of the studied properties compared to others (Figure 2). On the GT biplot created according to the examined properties, 7 sections were comprised. The genotype coded as 5 with crude protein and genotype 69 with amylose, amylopectin, total starch, trough 1, peak 1, final viscosity and setback placed on same section (section 1). Genotype 125 with resistant starch, total phenolic content and antiradical activity placed on section 3 while genotype 3 placed in section 4 with phytic acid and pasting temperature.

Genotype 5 with crude protein in section 4 and genotype 50 with pepsin protein digestibility and non-resistant starch level placed in same section (section 7). These genotypes placed in the highest group in terms of these properties (Figure 2).

The ideal genotype or genotypes can be determined by using the GT biplot analysis method to increase the selection success of the researchers. In this evaluation, genotypes are evaluated with circles formed based on the center determined according to the average of the properties examined. As they move away from the innermost circle to the outermost circle, the genotypes move away from the ideal genotype (Figure 3). None of the local sorghum genotypes used in the study placed on the core of the center. However, the closest genotype to the ideal genotype was genotype 30 (Figure 3).

GT biplot method with the analysis of the data set according to the mean of the measurements, the average feature axis, or proximity to the genotypes according to the proximity allows the evaluation of changes (Figure 4). The genotype, which is the closest to the average feature axis of the sorghum genotypes compared to the other genotypes, was the genotype with the most balanced biocompatible properties.

4. Discussion

The crude protein content of the nutrients is one of the most important criteria for quality evaluation. [31] Singh et al.[32] reported that the crude protein level of sorghum grain was in the range of 9.90-19.80% while it was reported as in the range of 7-15% by FAO[33] and Beta et al. [34] The main reason for the differences in dry matter and crude protein content of the varieties could have resulted from the genetic structure of the plant, and it was reported that it was changed depending on the ripening period, temperature, and fertilization process. [35] Chemical composition and nutritional value of sorghum grains are affected by some different factors such as genotype, climate, soil structure and fertilization [36]. A special protein called kafirin, which formed a part of the proteins present (kafirin proteins formed 50-70% of total protein in sorghum,[4] in sorghum structure was reported as it decreased the digestion and suppressed the energy usage. Disulfide cross bond in sorghum kafirin is bigger than that of the corn protein, and this event decreases the digestible protein level [37,38]. Some scientists reported that the cell walls which surrounded the protein and starch granules in cereal grains restrict the enzymes to reach the endosperm and this event changed depending on the grain structure [39,40] . With the increase of testa components in sorghum grain, the digestible protein content decreases [40,41]. The interactions of non-protein compounds like starch, nonstarch polysaccharides, lipids, phytates with sorghum proteins have also an effect on digestible protein ration in sorghum grain [4]. The digestible protein content of the sorghum genotypes having different level of tannins was determined as similar to the results reported by some researchers [40,42,43,44] while the digestible protein contents of sorghum grains changed depending on phenolic content and tannins level [4].

Sorghum contains many phenolic and other chemical compounds having positive effect on human health [45]. These phenolic compounds present in the different tissues of the plants (leaf, stem, grain etc.). The purple/red colored plants having high phenolic content and antioxidant activity are a kind of foods which are quite an important source for healthy compounds and so, this event gives useful information to manufacture sorghum grains due to some genotypes have purple/red color intensity [29]. The grain colors of the genotypes used in the study varied from white to purple. At the same time, phenolics in plant tissues vary according to plant species, environmental factors (precipitation, temperature, and humidity, wind, etc.), soil structure, death of plant tissues, bacterial and viral infection [46]. Phytic acid (myo-inositol hexaphosphoric acid) is usually found in grains as mixtures of potassium, magnesium and calcium salts (phytin or phytate). [47] The phytic acid content of sorghum grain is quite variable according to the varieties. Phytic acid content of sorghum grains was reported as quite variable like 0.27% [48], 0.3% [49], 0.886% [50] and 1% [51]. The highest phytic acid level is in embryo and shell of the sorghum grain, respectively [52]. The phytic acid level is high in the grains consumed as whole meal [51]. The phytate molecule is highly charged with its six phosphate groups and it is an excellent chelator that forms insoluble complexes with mineral cations and proteins [47]. This property causes to decrease in bioavailability of minerals (Ca, Zn, Fe, Mn) and protein digestibility [53].

As is known, vegetable oils are important, and high-quality energy source for human and animal foods for providing essential nutrients and they contain some important bioactive compounds [54]. Some varieties were monitored in terms of oil content and fatty acid composition among the sorghum genotypes [54,55]. The crude oil content and fatty acid composition are not constant parameters for the plants, and some studies showed that the synthesis of fatty acids changed depending on genetic, ecologic, morphologic, physiologic and cultural applications. [56] The effects of genotype, cultural treatments, and environment and stress conditions are important on starch content of sorghum grains. It was reported that the high temperature decreased the starch content of grain during ripening period of sorghum. [57] The role of matrix protein in sorghum endosperm and components of cell wall are important factors on limiting the starch extraction, and these create some differences in red sorghum (purity is 93.3%) and white sorghum (purity is 94.1%) genotypes. Liu et al. reported that the starch granules are destroyed, and the level of starch changed during the milling of the grains [58]. This event not only caused a variety among the genotypes but also a difference in starch extraction of the same samples. Liu et al. reported the starch contents were in the range of 55.6-75.2% in a study including 160 sorghum genotypes as a material and it was also reported that it ranged from 66 to 73% by Buffo et al. [59] and Fernholz [60] and 60-80% by Boudries et al. [61] and Zhang et al.[62]. The starch content of the samples in the current study was found as similar to the results of some researches [37,58,63,64].

From the technological point of view, amylose complexes formed by emulsifiers (monoglycerides, sucrose esters, etc.) are essential in terms of being used as anti-staling and dough-improving agents in the bakery industry. Amylose content of sorghum grains is quite different according to the genotype and environment [66]. The grain color showing the phenolic content of the sorghum grains and the moisture during the growing period affects the amylose level of the sorghum [61]. In the literature, amylose level of sorghum was in the range of 21-34% [66], 19.2-22.4% [67], 20.9-30.2% [66], 22.06-28.46% [58], 12.96-18.72% [68]. In a study performed by FAO, averaged amylose content of 160 genotypes was reported as 26.9% [33]. Our results were found similar to the results stated above. It was reported that the amylopectin level of the grain changed depending on botanical origin, climate, soil conditions and development of grain [69]. Boudries et al. reported that the amylopectin level of white sorghum was 72.9% while the red sorghum contained 75.2% [61]. Amylopectin levels were reported as in the range of 81.28-89.20% and 25.28-28.26% by some researchers, respectively [37,68]. The genotypes used in the current study contained lower amylopectin compared to the levels in literature reports. It is thought that the differences caused by the genotypical properties. The amylopectin levels of the samples in the present study were in accordance by the results reported by some researchers [22,61,70].

Cereal grains are rich in many bioactive components such as dietary fibre, lignans, phenolic acids, phytosterols, minerals, tocopherols, and tocotrienols [11]. These types of bioactive compounds are effective in the reduction of diabetes, [71] cardiovascular diseases, and some cancer types [72]. Because these compounds present in the germ and seed of the grain in high quantities, [73] it is suggested that the grains could be consumed as wholegrain [11]. For that reason, RVA parameters and other analyses were conducted on the whole grains of sorghum. Pasting properties of the sorghum genotypes showed quite differences depending on environmental conditions and sample variance [35,61]. The big differences in the stickness properties of sorghum dough change depending on amylose, lipid, fatty acid composition, protein content and storage conditions [62,74]. The starch granules having high swelling capacity result in higher peak viscosity and the peak viscosity affects the quality of final product [11]. The presence of lipids (sorghum flour) that can form insoluble complexes with amylose both during swelling and during gelatinization acts as both diluent and a swelling inhibitor [74]. Amylase activity level in grains affect the viscosity of starch during processing of flours and pasting properties of sample are affected by this event, [11] while there was no relationship between amylose level of starch and setback viscosity [58]. A positive correlation was observed between amylose level and trough 1, peak 1, final viscosity, breakdown and setback values in the current study (Figure 1). Sorghum starch viscosity was found to be similar to viscosity of potato but higher compared to that of wheat starch. Breakdown viscosity is higher like as in potato and millet starch [75]. Compared to sorghum starch, sorghum flour had higher stickiness temperature which attributes to the protein of the sample [74]. On the biplot chart, the correlation between the cosine value of the two property vectors and the correlation coefficient between these two properties are closely related. Genotypes close to each other on GT biplot chart are similar in terms of studied properties. Therefore, in this method, polygons formed by combining genotypes that are farthest from the center of the biplot and genotypes which are diagonalized in sections formed with lines drawn perpendicular to these polygon lines are evaluated as different from other genotype groups [24]. Properties between genotypes with close angle positively correlated with one [76]. According to this event, three different property groups were created according to the studied parameters. Additionally, the parameters having the short vector length on the biplot graphic are the ones having low separation performance [24]. Crude oil and amylose levels were the parameters having the lowest separation performance among the studied parameters of sorghum genotypes in the current study. In other words, most of the genotypes showed high similarity in terms of crude oil and amylose level. Genotypes positioned as close to each other on GT biplot graphics are similar in terms of examined properties. Therefore, in this technique, polygons formed by combining genotypes that are farthest from the center of the biplot and genotypes, which are diagonalized in sections formed with lines drawn perpendicular to these polygon lines are evaluated as different from other genotype groups.

In our study, genotypes found in diagonal diagrams were observed to be in the group with the highest values in terms of the features in the same region. GGE biplot is a developed technique for the interaction of genotype and environment. But, in recent years, it is widely used to compare the relationship between genotypes and properties. It is created to examine the relationship between the properties. Especially in the biplot (GT) graph, the relationship between the properties of the parameters on the graph according to the correlation coefficients can be seen in the relationship between the properties according to the correlation, and the evaluation of the best genotype can be misleading in some cases. For example, despite the genotype coded as 30 was in same group with pepsin protein digestibility, it is not the best genotype in terms of this property. The reason for this event is that the graph is drawn according to the properties with very different values. So, when the commenting the GT biplot graphics, comments on the relationships among the studied properties give more accurate results instead of evaluation on genotypes. To evaluate the genotypes to be evaluated by taking into account the average values are healthier [29]. In biplot analysis, the properties placed in the farthest from the origin are the most important ones in terms of the population performances [24]. From this point of view, the genotypes in the first circle are evaluated as ideal genotypes [24,77]. An ideal genotype(s) is a genotype having both highest values in terms of the desired characteristics and high stability [29]. According to this, the most ideal genotype in the current study was the genotype coded as 30 (PI 177159 03).

Conclusion

According to the research results, a lot of genotypes having superior properties in terms of nutritional characteristics compared to standard varieties were identified. These varieties showing superior properties could be included into the filed crop cultivation according to the usage aim (nutrition, health, ethanol etc.). In addition, these plants will be used as rootstocks in terms of targeted features in future breeding activities. Among the local, the sorghum genotypes having a wide variation in terms of biochemical properties, the ones coded as 30 and 69 for first property group, 3 and 125 for second property group and 5 and 6 for third property group were selected easily as the superior genotype by GT biplot technique.

Acknowledgement

This research was supported by The Scientific and Technological Research Council of Turkey (TUBITAK) (Project number: 113O914).

References

[1] J. M. Awika, L. W. Rooney, Phytochemistry. **2004**, 65, 1199.

[2] R. Li, H. Zhang, X. Zhou, Y. Guan, F. Yao, G. Song, J. Wang, C. Zhang, Genet. Resour. Crop Evol. **2010**, 57, 1.

[3] H. Hill, L. S. Lee, R. J. Henry, Food Chem. **2012**, 131, 175.

[4] K. G. Duodu, J. R. N. Taylor, P. S. Belton, B. R. Hamaker, J. Cereal Sci. **2003**, 38(2), 117.

[5] P. C. Anunciação, L. de Morais Cardoso, J. V. P. Gomes, C. M. Della Lucia, C. W. P. Carvalho, M. C. Galdeano, H. M. Pinheiro-Sant'Ana, Food Chem. **2017**, 221, 984.

[6] A. Serteser, V. Gök, Third Food Engineering Congress, Ankara. 83-98, **2003**.

[7] F. Chen, P. Cole, Z. Mi, L. Y. Xing, Int. J. Cancer. **1993**, 53(6), 902.

[8] B. B. Lin, H. L. Chen, P. C. Huang, Nutr. Rep. Int. **1986**, 34, 821.

[9] K. Tebib, J.M. Rouanet, P. Besancon, Food Chem. **1997**, 59, 135.

[10] C. Santos-Buelga, A. Scalbert, J. Sci Food Agric. **2000**, 80, 1097.

[11] S. Ragaee, E. S. M. Abdel-Aal, Food Chem. **2006**, 95(1), 9.

[12] J. H. Wong, T. Lau, N. Cai, J. Singh, J. F. Pedersen, W. H. Vensel, B. B. Buchanan, J. Cereal Sci. **2009**, 49(1), 73.

[13] L.W. Rooney, F.R. Miller, Variation in the structure and kernel characteristics of sorghum'' Page 143 *in* Proceedings of the International Symposium on Sorghum Grain Quality, **1981**,

[14] R.D. Sullins, L.W. Rooney, Cereal Chem. **1975**, 52, 361.

[15] R. M. Sandstedt, D. Strahan, S. Veda, R. C. Abbot, Cereal Chem. **1962**, 39, 123.

[16] E. B. Ellis, Ph.D. Dissertation, Texas A&M University, College Station, **1975**.

[17] T. C. Smith, D. R. Kindred, J. M. Brosnan, R. M. Weightman, M. Shepherd, R. Sylvester-Bradley, Wheat as a feedstock for alcohol production Research review no.61. Home grown cereals authority, Caledonia house, 223 Pentoville Rd., London, **2006**.

[18] L. I. Ezeogu, K. G. Duodu, M. N. Emmambux, J. R. N. Taylor, Cereal Chem. **2008**, 85(3), 397.

[19] I. Burešová, L. Hrivna, Effect of wheat gluten proteins on bioethanol yield from grain. Appl. Energy. **2011**, 88, 1205.

[20] T. Hooks, J. F. Pedersen, D. B. Marx, Crop Sci. **2006**, 46, 751.

[21] C. O. Kangama, X. Rumei, Afr. J. Biotechnol. **2005,** 4(7).

[22] M. H. Dicko, H. Gruppen, A. S. Traoré, A. G. J. Voragen, W. J. H. Van Berkel, Afr. J. Biotechnol, **2006**, 5(5), 384.

[23] M. Akcura, Turk. J. Agric. For. **2011**, 35(2), 115.

[24] W. Yan, Crop Variety Trials: Data Management and Analysis. John Wiley Sons. **2014**.

[25] W. Yan, M. S. Kang, GGE Biplot Analysis: A Graphical Tool for Geneticists, Breeders, and Agronomists, CRC Press, **2003**.

[26] AOAC. Official method of analysis. Association of official analytical chemists 15th. edition, pp. 66-88, **1990**.

[27] M. Aboubacar, M. F. Webster, J. Non-Newtonian Fluid Mech. **2001**, 98(2), 383.

[28] A. Nunes, I. Correia, A. Barros, I. Delgadill, J. Agric. Food Chem. **2004**, 52(7), 2052.

[29] L. Dykes, L. W. Rooney, R. D. Waniska, W. L. Rooney, J. Agric. Food Chem. **2005**, 53(17), 6813.

[30] J.M. Awika, L.W. Rooney, X. Wu, R.L. Prior, L. Cisneros-Zevallos, J. Agric. Food Chem. **2003**, 51, 6657.

[31] G. Assefa, I. Ledin, Anim. Feed Sci. Technol. **2001**, 92(1-2), 95.

[32] P. Singh, U. Singh, B. O. Eggum, K. A. Kumar, D. J. Andrews, J. Sci. Food Agric. **1987**, 38, 41.

[33] FAO. http://faostat.fao.org/site/567/DesktopDefault.aspx?PageID=567#ancor, **1995**.

[34] T. Beta, L.W. Rooney, R.D. Waniska, Cereal Chem. **1995**, 72, 533.

[35] D. M. Ball, M. Collins, G. D. Lacefield, N. P. Martin, D. A. Mertens, K. E. Olson, D. H. Putnam, D. J. Undersander, M. W. Wolf, Understanding forage quality. American Farm Bureau Federation Publication 1-01, Park Ridge, IL, **2001**.

[36] M. R. Ebadi, J. Pourreza, J. Jamalian, M. A. Edriss, A. H. Samie, S. A. Mirhadi, Int. J. Poult. Sci. **2005**, 1, 27.

[37] I. Salinas, A. Pro, Y. Salinas, E. Sosa, C. M. Becerril, M. Cuca, M. Cervantes, J. Gallegos, J. Cereal Sci. **2006**, 44, 342.

[38] P. H. Selle, D. J. Cadogan, X. Li, W. L. Bryden, Anim. Feed Sci. Technol. **2010**, 156(3-4) 57.

[39] T. Evers, S. Millar, J. Cereal Sci. **2002**, 36(3), 261.

[40] K. G. Duodu, A. Nunes, I. Delgadillo, L. M. Parker, E. N. C. Mills, P. S. Belton, R. N. Taylor, J. Cereal Sci. **2002**, 35, 161.

[41] B. A. K. Chibber, E.T. Mertz, J. D. Axtell, J. Agric. Food Chem. **1980**, 28, 160.

[42] M. P. Oria, B.R. Hamaker, J. M. Shull, J. Cereal Sci. **1995a**, 22, 85.

[43] M. P. Oria, B. R. Hamaker, J. M. Shull, J. Agric. Food Chem. **1995b**, 43, 2148.

[44] M. E. Arbab, A. H. El Tinay, Food Chem. **1997**, 59, 339–343.

[45] L. Dykes, L. W. Rooney, J. Cereal Sci. **2006**, 44(3), 236.

[46] P. J. Van Soest, Nutritional ecology of the ruminant. Cornell University Press, Ithaca, NY, USA, **1994**.

[47] P. Ryden, R. R. Selvendran, Phytic acid: Properties and determination. In: Macrae, R., Robinson, R. K. Sadler, M. J. (Eds.) Encyclopaedia of food science, food technology and nutrition, Academic Press, London pp. 3582-3587, **1993.**

[48] E. A. I. Elkhalil, A. H. El Tinay, B. E. Mohamed, E. A. E. Elsheikh, Food Chem. **2001**, 72(1), 29-32.

[49] S.O. Mahgoub, S.A. Elhag, Cereal Chem. **1998**, 61, 77.

[50] E. K. Marfo, B. K, Simpson, J. S. Idowu, O.L. Oke, J. Agric. Food Chem. **1990**, 38, 1580.

[51] R. M. García-Estepa, E. Guerra-Hernández, B. García-Villanova, Food Res. Int. **1999**, 32, 217.

[52] H. I. Ali, B. F. Harland, Cereal Chem. USA, **1991**.

[53] B. E. Knuckles, D. D. Kuzmicky, A. A. Betschart, J. Food Sci. **1985**, 50, 1080.

[54] S. Mehmood, I. Orhan, Z. Ahsan, S. Aslan, M. Gulfraz, Food Chem. **2008**, 109, 855.

[55] L. W. Rooney, R. D. Waniska, Sorghum food and industrial utilization. Sorghum: Origin, history, technology, and production, 689-729, **2000**

[56] H. Baydar, Ekin J. **2000**, 11, 50-57.

[57] J. Matsuki, T. Yasui, K. Kohyama, T. Sasaki, Cereal Chem. **2003**, 80(4), 476.

[58] L. Liu, T. J. Herald, D. Wang, J. D. Wilson, S. R. Bean, F. M. Aramouni, J. Cereal Sci. **2012**, 55(1), 31.

[59] R. A. Buffo, C. L. Weller, A. M. Parkhurst, Cereal Chem. **1998**, 75, 100.

[60] M.C. Fernholz, Master thesis, Available on line: ‘‘http://krex.k-state.edu/dspace/bitstream/2097/729/1/MaryFernholz2008.pdf, **2008**.

[61] N. Boudries, N. Belhaneche, B. Nadjemi, C. Deroanne, M. Mathlouthi, B. Roger, M. Sindic, Carbohydr. Polym. **2009**, 78, 475.

[62] G. Zhang, B. R. Hamaker, Cereal Chem. **2005**, 82(5), 534.

[63] FAO, FAO Corporate Document Repository. http://www.fao.org/docrep/t0818e/T0818E0b.htm, **2017**.

[64] D.H. Rhodes, Jr. Hoffmann, L. Rooney, W.L. Herald, T.J. Bean, S. Boyles, R. Brenton, Z.W. S. Kresovich, BMC Genomics, **2017**, 18, 15

[65] T. Beta, H. Corke, J. Cereal Sci. **2001**, 34(3), 261.

[66] T. Beta, H. Coke, L. Rooney, J. R. N. Taylor, J. Sci. Food Agric. **2001**, 81, 245.

[67] M. Benmoussa, B. Suhendra, A. Aboubacar, B. R. Hamaker, Starch- Stärke. **2006**, 58(2), 92.

[68] I. S. Udachan, A. K. Sahoo G. M. Hend, Int. Food Res. J. **2012**, 19(1), 315.

[69] N. Singh, L. Kaur, K. S. Sandhu, J. Kaur, K. Nishinari, Food Hydrocol. **2006**, 20(4), 532.

[70] A. Shegro, N. G. Shargie, A. van Biljon, M. T. Labuschagne, J. Crop Sci. Biotechnol. **2012**, 15(4), 275.

[71] M. A. Pereira, D. R. Jacobs, J. J. Pins, S. K. Raatz, M. D. Gross, J. L. Slavin, E. R. Seaquist, Am. J. Clin. Nutr. **2002**, 75, 848.

[72] D. R. Jacobs, L. Marquart, J. Slavin, L. H. Kushi, Nutr. Cancer. **1998**, 30, 85.

[73] L. V. Glitsø, K. E. Bach Knudsen, J. Cereal Sci. **1999**, 29, 89.

[74] Q. Sun, Z. Han, L. Wang, L. Xiong, Food Chem. **2014**, 145, 756.

[75] A. C. Roudot, Rhéologie et analyse de texture des aliments, Ed. tec & doc. Lavoisier, Paris. p. 196, **2002**.

[76] W. Yan, I. Rajcan, Crop Sci. **2002**, 42, 11.

[77] M. Akcura, K. Kokten, Qual. Assur. Saf. Crops Foods. **2017**, 9(2), 153.

Conflict of interest statement

We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work, there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the position presented in, or the review of, the manuscript entitled "Characterization of local sorghum (*Sorghum bicolor* L.) population grains in terms of nutritional properties and evaluation by GT Biplot approach".

Figure captions

Figure 1. GGT-biplot based on nutritional composition-focused scaling.

Figure 2. Polygon views of the GT-biplot based on symmetrical scaling for the which-won-what pattern for genotypes and chemical composition. Details of genotypes are presented in Supplementary Data 1

Figure 3. GT-biplot based on genotype-focused scaling for comparison the genotypes with the ideal genotype.

Figure 4. Genotype ranking based on biochemical properties

Table 1. Some physicochemical properties of the field experiment soils

Properties	Deep	
	0-30 cm	30-60 cm
Clay (%)	13.1	8.94
Silt (%)	4.16	10.4
Sand (%)	82.74	80.66
Class	Sandy-Loamy	Sandy-Loamy
pH	7.94	7.75
Organic Matter (%)	1.05	1.27
$CaCO_3$ (%)	0.28	0.27
K_2O (kg ha^{-1})	1092.2	755.14
P_2O_5 (kg ha^{-1})	89.63	11.56
EC (mmhos/cm^{-1})	0.96	0.23

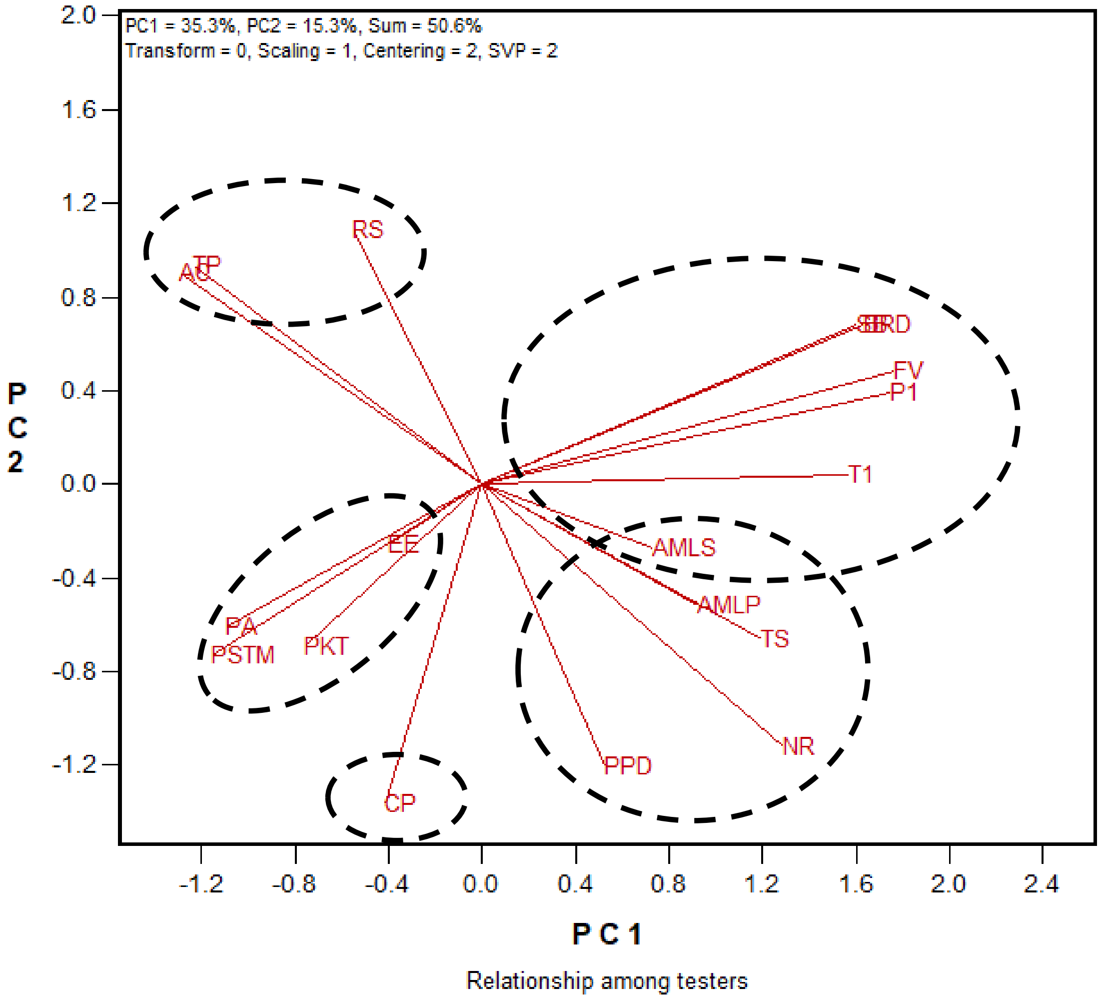
Table 2. Minimum, maximum and mean values for nutritional compositions and pasting properties of Turkey local sorghum varieties*

Nutritional Parameters	**Min**	**Mean**	**Max**	**Sig. Dg.**	**LSD**
Crude oil (%)	2.15	3.78	6.40	**	0.48
Crude protein (%)	6.67	10.47	14.33	**	1.20
Pepsin protein digestibility (%)	6.71	55.63	82.24	**	0.60
Phytic acid (%)	0.37	1.83	4.09	**	0.04
Resistant starch (%)	1.10	7.27	34.23	**	0.51
Non-resistant starch (%)	10.79	47.67	79.61	**	0.46
Total starch (%)	15.42	54.94	85.54	**	0.59
Amylose (%)	5.67	17.84	43.48	**	0.50
Amylopectin (%)	9.45	37.27	65.67	**	0.46
Total phenolic content (mg GAE/g)	0.19	2.62	5.06	**	0.72
Antiradical activity (%)	3.72	42.02	91.48	**	2.09
Pasting Properties					
Peak 1 (cP)	657	2022	4162	**	120.64
Trough 1 (cP)	566	1435	2422	**	114.71
Breakdown (cP)	92	600	2344	**	124.53
Final Viscosity (cP)	756	2544	6473	**	167.77
Setback (cP)	190	1109	4051	**	184.42
Peak Time (min)	3.90	5.69	6.41	**	0.13
Pasting Temperature (°C)	50.28	85.58	93.30	**	3.80

**Detailed nutritional compositions of all sorghum genotypes were given in Supplementary Data 1-2.*

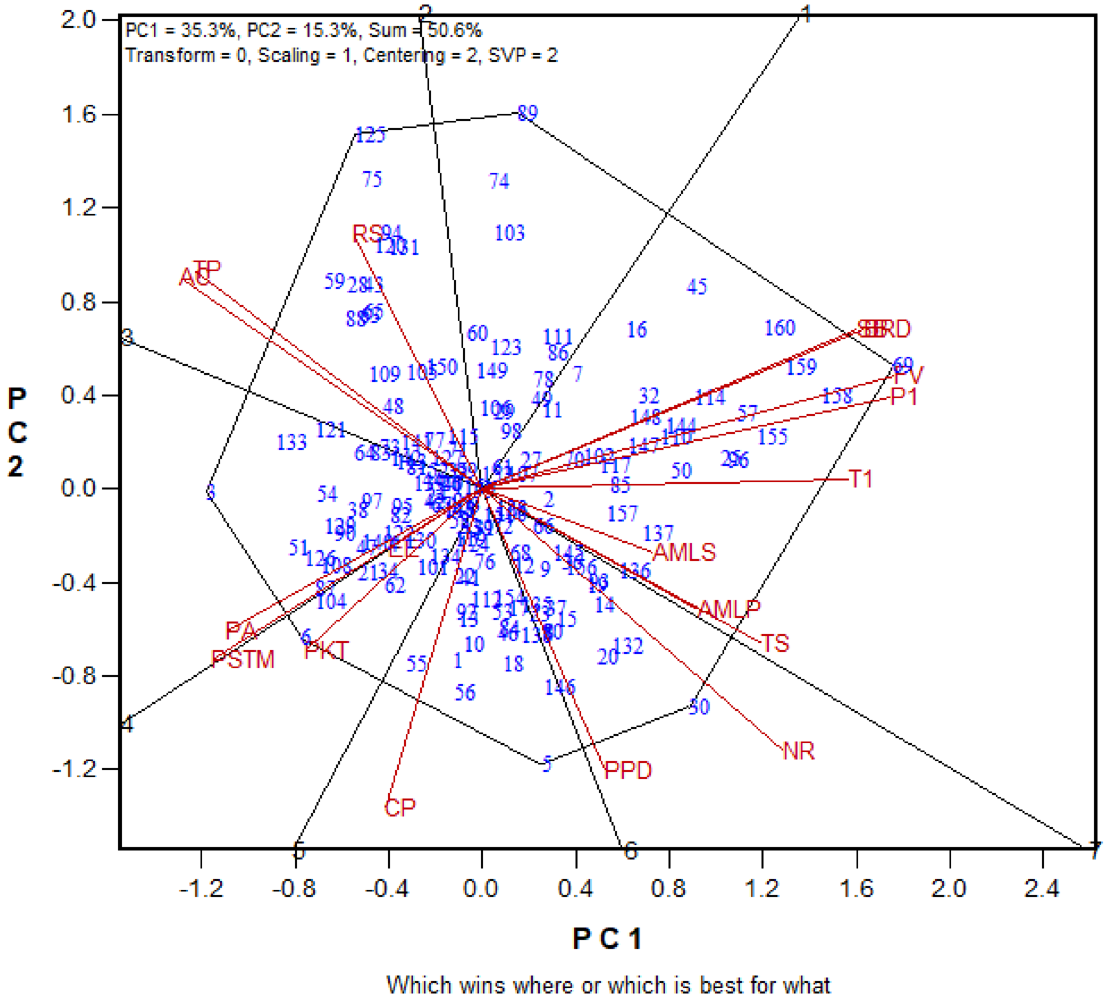
Sig. Dg.: *significant difference;* *****:*** *$P \leq 0.01$;* ***LSD:*** *Least Significant Difference*

Figure1.



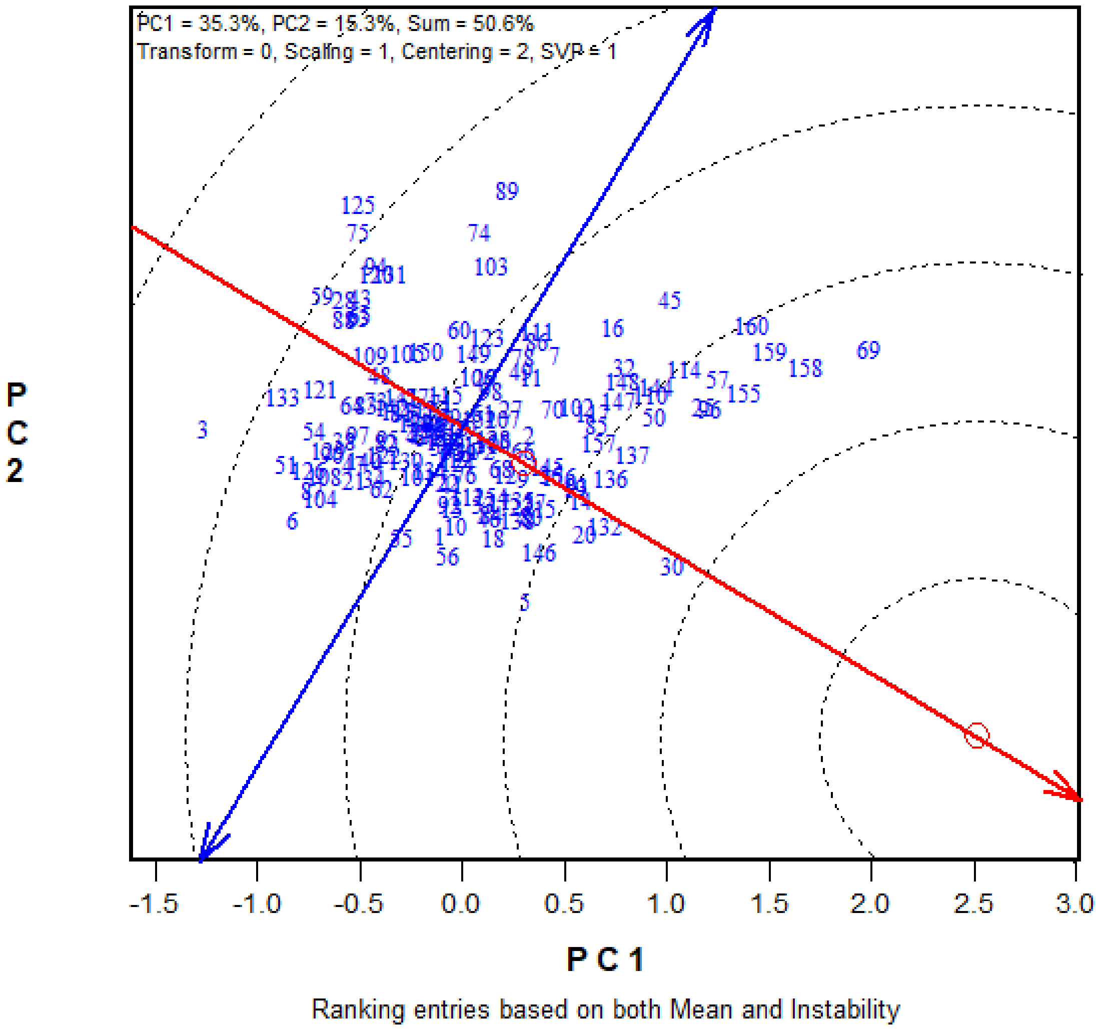
***EE:** crude oil; **CP:** crude protein; **PPD:** pepsin protein digestibility; **PA:** phytic acid; **RS:** resistant starch; **NR:** non-resistant starch; **TS:** total starch; **AMLS:** amylose; **AMLP:** amylopectin; **TP:** total phenolic; **AC:** antiradical capacity; **P1:** peak 1; **T1:** trough 1; **BRD:** breakdown; **FV:** final viscosity; **SB:** setback; **PKT:** peak time; **PSTM:** pasting temperature*

Figure2.



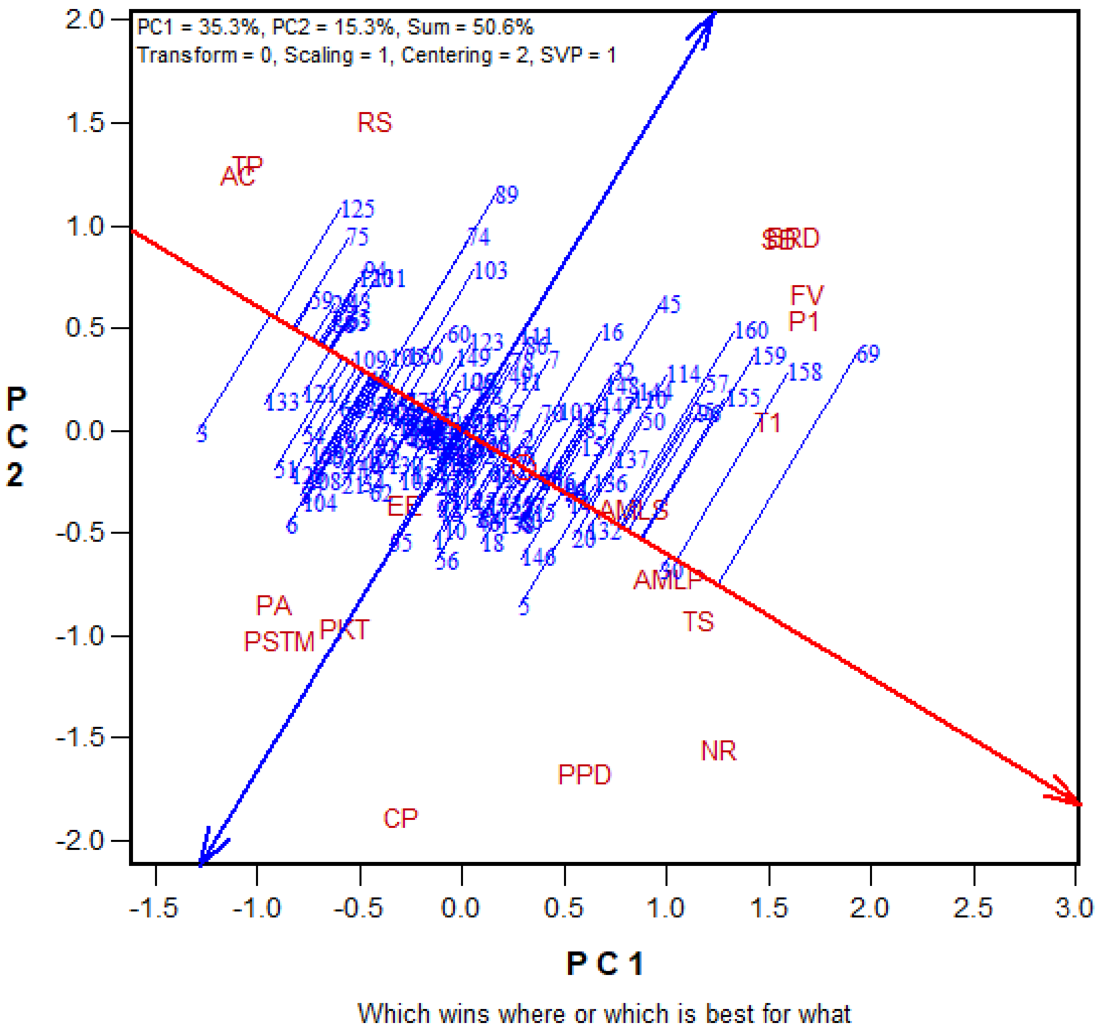
EE: *crude oil;* ***CP:*** *crude protein;* ***PPD:*** *pepsin protein digestibility;* ***PA:*** *phytic acid;* ***RS:*** *resistant starch;* ***NR:*** *non-resistant starch;* ***TS:*** *total starch;* ***AMLS:*** *amylose;* ***AMLP:*** *amylopectin;* ***TP:*** *total phenolic;* ***AC:*** *antiradical capacity;* ***P1:*** *peak 1;* ***T1:*** *trough 1;* ***BRD:*** *breakdown;* ***FV:*** *final viscosity;* ***SB:*** *setback;* ***PKT:*** *peak time;* ***PSTM:*** *pasting temperature*

Figure 3.



EE: *crude oil;* ***CP:*** *crude protein;* ***PPD:*** *pepsin protein digestibility;* ***PA:*** *phytic acid;* ***RS:*** *resistant starch;* ***NR:*** *non-resistant starch;* ***TS:*** *total starch;* ***AMLS:*** *amylose;* ***AMLP:*** *amylopectin;* ***TP:*** *total phenolic;* ***AC:*** *antiradical capacity;* ***P1:*** *peak 1;* ***T1:*** *trough 1;* ***BRD:*** *breakdown;* ***FV:*** *final viscosity;* ***SB:*** *setback;* ***PKT:*** *peak time;* ***PSTM:*** *pasting temperature*

Figure 4.



__EE:__ crude oil; __CP:__ crude protein; __PPD:__ pepsin protein digestibility; __PA:__ phytic acid__; RS:__ resistant starch; __NR:__ non-resistant starch; __TS:__ total starch; __AMLS:__ amylose; __AMLP:__ amylopectin; __TP:__ total phenolic; __AC:__ antiradical capacity; __P1:__ peak 1; __T1:__ trough 1; __BRD:__ breakdown; __FV:__ final viscosity; __SB:__ setback; __PKT:__ peak time; __PSTM:__ pasting temperature