Evaluation of nutritional quality and antioxidant potential of pigeonpea genotypes

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Abstract Three released cultivars, forty four advance breeding lines and three wild species of pigeonpea (*Ca-janus cajan* L. Millsp) were evaluated for nutritional, antinutritional traits and antioxidant potential so as to identify promising genotypes. The average content of total soluble sugars, starch and total soluble proteins was found to be 43.66, 360.51 and 204.54 mg/g, respectively. Antioxidant potential in terms of free radical scavenging activity (DPPH), ferric reducing antioxidant power (FRAP), reducing power, hydroxyl radical scavenging activity and superoxide anion radical scavenging activity was estimated. The diversity was observed in genotypes with all the traits. AL 1960, AL 2000, AL 2009 and AL 2046 had high total soluble proteins, medium antinutritional factors and good antioxidant potential. AL 201, AL 1931, AL 1932, AL 1960, AL 2046, AL 2049 and AL 2060 had good nutritional value as protein and starch content ranged from 20 to 23 and 42 to 52%, respectively. Wild species *C. scarabaeoides* 1CP15683/W15 had lower carbohydrates, proteins, and antinutritional traits while high antioxidant potential due to high total phenols, DPPH, FRAP and reducing power. The diversity observed in genotypes with all the traits could be further used to develop nutritionally important genotypes.

Keywords Antinutritional factors · Antioxidant potential · Free radical scavenging activity · Nutritional quality · Pigeonpea

Introduction

Legume seeds occupy an important place in human diet all over the world as they are rich sources of proteins. Legumes are called the multi-purpose plants as apart from being used as food for humans, they are also used as animal feed, beneficial for farmers as they increase the overall fertility of the soil due to their ability to fix atmospheric nitrogen and thus reduces the need of expensive nitrogenous fertilizers (Blazos and Belski 2016). Legumes are named as poor man's meat as they are especially important as food in those regions where animal proteins are scarce or where poverty, religious or ethnic preferences preclude the consumption of meat (Mahala et al. 2013). The nutritional quality of legumes is negatively affected by the presence of antinutritional factors like protease inhibitors, tannins, saponins, raffinose series oligosaccharides as (RFOs) and phytic acid which limit digestion, absorption and proper metabolism of nutrients resulting in digestive losses (Wang et al. 2010).

Legumes consumption is important in the prevention of chronic diseases such as cancer and heart disease as they contain phytochemicals that combat oxidative stress in the body and helps to maintain a balance between oxidants and antioxidants (Khyade and Jagtap 2016). Free radicals produced in several biochemical reactions in human body are deleterious as they are mediators of many diseases and start chain reactions that cause structural and functional damage to protein, lipids, nucleic acid and cellular molecules. Antioxidants terminate these chain reactions by removing

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free radical intermediates and inhibit other oxidation reactions by being oxidized themselves (Aggarwal et al. 2015). There are two basic categories of antioxidants, namely synthetic and natural ones. Several synthetic antioxidants such as butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) are used in order to increase the storage of foods but these substances may be inappropriate for human consumption because of their carcinogenicity (Wu et al. 2009). Increasing demands of consumers on safety issues regarding the use of synthetic food additives, imposes the need to search for new sources of plant's antioxidants, as feasible and natural alternatives to synthetic antioxidants. Therefore, the development and utilization of more effective antioxidants of natural origin are favored and the present study explore the antioxidant potential of pigeonpea genotypes.

Pigeonpea (*Cajanus cajan* (L.) Millspaugh) is one of the valuable sources of proteins, vitamins and minerals among food legumes and occupy a very important place in human nutrition. Pigeonpea is fifth prominent grain legume crop in the world and it occupies second position among pulse crops in India (Pratibha et al. 2015). The Indian subcontinent alone contributes nearly 92% of total pigeonpea production in the world is popularly known as red gram, tur or arhar (Khanum et al. 2015; Reddy et al. 2016). Seeds of pigeon pea are known to be rich sources of proteins, carbohydrates and minerals with protein content generally varying from 18 to 25% and as high as 32% (Aggarwal et al. 2015). Pigeonpea is a good source of crude fibre, iron, sulphur, calcium, potassium, manganese and water soluble vitamins especially thiamine, riboflavin, niacin (Saxena et al. 2010). The need of hour is to identify promising pigeonpea genotypes with high nutritional quality, low amount of antinutritional factors and high antioxidant potential so that these legumes may prove beneficial for the poor people who depend solely on legumes for fulfillment of their nutritional requirements. The information about nutritional, antinutritional and antioxidant potential of pigeonpea is lacking so the present investigation was carried out to evaluate the nutritional and antioxidant potential of pigeonpea genotypes.

Materials and methods

Plant material

Fifty pigeonpea genotypes consisting of three recommended cultivars of PAU (AI 15, PAU 881 and AI 201), forty-four advance breeding lines (S. No. 4–47, Table 1) and three accessions of wild species *C. scarabaeoides* (1CP15683/W15, 1CP15701/W15 and 1CP15703/W15) obtained from Pulses Section, Department of Plant

Breeding and Genetics, Punjab Agricultural University, Ludhiana were taken for nutritional quality evaluation. Broken and damaged seeds and foreign material were handpicked from the sample before the analytical studies. The seeds of pigeonpea (in triplicates) were crushed to fine powder in pestle and mortar and the contents were passed through 80 µm sieve to have uniform powder which was stored for extraction and assay of various components in triplicates.

Chemicals

BAPNA (*N*- α -benzoyl-DL-arginine-p-nitroanilide), bovine trypsin, DPPH (2,2-diphenyl-1-picryl hydrazyl), 1,10-phenanthroline, sodium phytate and TPTZ (2,4,6-Tri-(2-pyridyl)-5-triazine) were obtained from Sigma Company, St Louis, MO, USA and all other chemicals used were of high analytical grade purchased from Sisco Research Laboratories, Mumbai, India.

Extraction and estimation of nutritional and antinutritional traits

Total soluble sugars in different pigeonpea cultivars/lines were estimated by using phenol sulphuric (Dubois et al. 1956). Bound fructose of sucrose and raffinose series oligosaccharides was determined after destroying the free fructose with 30% NaOH by resorcinol-HCl procedure (Williard and Slattery 1945). The dried sugar free residue was used for the estimation of starch by following the procedure of Yoshida et al. (1976). The powdered pigeonpea seeds (100 mg) were kept overnight in 25 ml of 0.1 N NaOH to extract total proteins from them. The supernatant after centrifugation at (5000×g) was used for the estimation of total proteins (Lowry et al. 1951).

Antinutritional factors such as tannins, saponins, phytic acid and trypsin inhibitors were estimated from pigeonpea seeds. Tannins were estimated using Folin-Denis reagent, the intensity of the colour developed was measured at 700 nm (Sadasivam and Manickam 1992). A standard curve of tannic acid (10–100 µg) was also prepared simultaneously. Saponins were extracted by homogenizing the seed powder (500 mg) with acetone and later with methanol (Fenwick and Oakenfull 1983). The methanolic extract (1 ml) was kept in boiling water bath at 100 °C in order to remove methanol and after cooling 2 ml of ethyl acetate was added followed by addition of 1 ml of anisaldehyde (0.5 ml of anisaldehyde mixed with 99.5 ml of ethyl acetate) and 1 ml of concentrated sulphuric acid. The test tubes were stirred and kept for 10 min at room temperature. The intensity of red colour developed was measured at 430 nm (Baccou et al. 1977). The amount of saponins was calculated from the standard curve using

Table 1 Distribution of total soluble sugars (mg/g), starch (mg/g) and total proteins (mg/g) in pigeonpea genotypes

S. No.	Genotypes	Total soluble sugars	Starch	Total soluble proteins	S. No.	Genotypes	Total soluble sugars	Starch	Total soluble proteins
1	AL 15	47.02 ± 1.62 ^{abc}	315.04 ± 32.15 ^{mnopqrs}	213.66 ± 6.16 ^{abcdefghijk}	26	AL 2049	49.45 ± 4.05 ^{ab}	464.48 ± 14.40 ^{abcde}	220.37 ± 14.84 ^{abcdeghi}
2	PAU 881	43.78 ± 3.24 ^{abc}	375.43 ± 17.22 ^{defghijklmnopq}	194.76 ± 12.33 ^{ghijklm}	27	AL 2060	55.13 ± 8.11 ^{ab}	509.91 ± 12.18 ^{ab}	200.20 ± 3.73 ^{cdefghijklm}
3	AL 201	60.80 ± 8.91 ^a	476.62 ± 53.96 ^{abc}	209.30 ± 16.44 ^{bcdefghijkl}	28	AL 2061	47.02 ± 1.62 ^{abc}	419.05 ± 27.69 ^{bcdefghijk}	210.76 ± 1.41 ^{bcdefghijk}
4	AL 1758	47.09 ± 1.69 ^{abc}	390.55 ± 3.44 ^{cdefghijklmnop}	200.58 ± 4.11 ^{cdefghijklm}	29	AL 2062	42.16 ± 1.62 ^{abc}	268.26 ± 8.30 st	207.09 ± 5.09 ^{bcdefghijklm}
5	AL 1843	47.02 ± 0.86 ^{abc}	424.65 ± 42.48 ^{bcdefghi}	171.51 ± 4.11 ^{lm}	30	AL 2063	42.97 ± 0.81 ^{abc}	447.24 ± 12.18 ^{gabdef}	200.20 ± 3.78 ^{cdefghijklm}
6	AL 1849	44.59 ± 2.43 ^{abc}	466.06 ± 25.26 ^{abcd}	177.34 ± 5.23 ^{klm}	31	AL 2067	43.77 ± 4.86 ^{abc}	440.19 ± 31.0 ^{abcdefg}	197.53 ± 9.31 ^{defghijklm}
7	AL 1922	40.53 ± 1.62 ^{abc}	417.35 ± 9.18 ^{bcdefghijkl}	186.04 ± 1.59 ^{hijklm}	32	AL 2071	46.21 ± 2.43 ^{abc}	382.23 ± 13.29 ^{cdefghijklmnop}	220.57 ± 13.96 ^{bcdefghi}
8	AL 1931	46.21 ± 2.43 ^{abc}	521.28 ± 6.88 ^a	206.97 ± 3.28 ^{bcdefghijklm}	33	AL 2078	42.16 ± 5.42 ^{abc}	382.23 ± 8.86 ^{cdefghijklmnop}	216.87 ± 9.89 ^{bcdefghij}
9	AL 1932	50.26 ± 1.62 ^{abc}	421.20 ± 26.41 ^{bcdefghij}	210.85 ± 8.77 ^{bcdefghijk}	34	AL 2086	43.78 ± 3.24 ^{abc}	405.73 ± 2.21 ^{cdefghijklm}	213.99 ± 4.65 ^{bcdefghijk}
10	AL 1933	49.45 ± 0.81 ^{abc}	407.60 ± 11.58 ^{cdefghijklm}	207.75 ± 13.15 ^{bcdefghijklm}	35	AL 2087	42.97 ± 0.81 ^{abc}	426.10 ± 33.23 ^{abcdeghi}	203.70 ± 17.45 ^{cdefghijklm}
11	AL 1957	44.59 ± 4.05 ^{abc}	406.94 ± 22.02 ^{cdefghijklm}	213.56 ± 3.83 ^{abcdeefghijk}	36	AL 2088	46.21 ± 8.92 ^{abc}	390.06 ± 11.07 ^{cdefghijklmnop}	201.02 ± 13.67 ^{cdefghijklm}
12	AL 1960	52.69 ± 5.67 ^{abc}	407.60 ± 25.26 ^{cdefghijklm}	222.09 ± 1.64 ^{abcdeefgh}	37	AL 2090	39.72 ± 7.29 ^{bc}	310.33 ± 20.16 ^{nopqrs}	209.87 ± 8.73 ^{bcdefghijk}
13	AL 1965	34.86 ± 2.43 ^{bc}	398.14 ± 22.55 ^{cdefghijklmno}	188.37 ± 13.15 ^{hijklm}	38	AL 2091	41.34 ± 2.43 ^{abc}	282.15 ± 15.06 ^{grst}	233.82 ± 13.85 ^{abcd}
14	AL 1974	44.59 ± 2.43 ^{abc}	369.44 ± 17.22 ^{cdefghijklmnopqr}	189.34 ± 12.88 ^{ghijklm}	39	AL 2092	44.59 ± 0.81 ^{abc}	279.15 ± 19.49 ^{grst}	236.43 ± 1.09 ^{bc}
15	AL 1976	45.50 ± 4.86 ^{abc}	326.40 ± 34.44 ^{ijklmnopqrs}	200.58 ± 4.11 ^{cdefghijklm}	40	AL 2094	37.29 ± 0.39 ^{bc}	286.48 ± 17.44 ^{grst}	197.53 ± 9.31 ^{defghijklm}
16	AL 1992	33.24 ± 2.43 ^c	317.47 ± 5.74 ^{lmnopqrs}	183.13 ± 4.11 ^{ijklm}	41	AL 2095	42.97 ± 2.43 ^{abc}	272.70 ± 34.02 st	203.70 ± 1.59 ^{cdefghijklm}
17	AL 2000	34.05 ± 3.24 ^c	362.13 ± 18.37 ^{ghijklmnopqrs}	251.16 ± 4.38 ^a	42	AL 2096	40.53 ± 1.62 ^{abc}	295.29 ± 43.20 ^{pqrs}	209.87 ± 8.73 ^{bcdefghijk}
18	AL 2009	33.23 ± 4.05 ^c	303.67 ± 16.07 ^{opqrs}	243.60 ± 9.04 ^{ab}	43	AL 2098	36.48 ± 0.81 ^{bc}	300.46 ± 9.30 ^{pqrs}	227.65 ± 14.43 ^{abcdeef}
19	AL 2016	40.53 ± 4.86 ^{abc}	409.23 ± 20.66 ^{cdefghijklm}	195.34 ± 13.15 ^{cdefghijklm}	44	AL 2101	41.35 ± 0.81 ^{abc}	299.60 ± 9.41 ^{pqrs}	216.04 ± 1.59 ^{abcdeefghij}
20	AL 2019	43.78 ± 1.62 ^{abc}	338.58 ± 12.63 ^{hijklmnopqrs}	183.33 ± 12.60 ^{ijklm}	45	AL 2102	42.16 ± 1.62 ^{abc}	322.39 ± 40.32 ^{lmnopqrs}	177.46 ± 6.54 ^{klm}
21	AL 2021	47.83 ± 2.43 ^{abc}	345.89 ± 2.29 ^{ghijklmnopqrs}	180.23 ± 8.22 ^{ijklm}	46	AL 2108	41.35 ± 0.81 ^{abc}	335.82 ± 29.07 ^{ijklmnopqrs}	184.77 ± 0.58 ^{ghijklm}
22	AL 2024	46.21 ± 0.81 ^{abc}	362.13 ± 29.85 ^{ghijklmnopqrs}	226.74 ± 8.22 ^{abcdeefg}	47	AL 2122	38.91 ± 1.62 ^{bc}	319.57 ± 30.13 ^{lmnopqrs}	209.87 ± 8.73 ^{bcdefghijk}
23	AL 2025	48.64 ± 0.79 ^{abc}	323.97 ± 3.44 ^{klmnopqrs}	189.34 ± 12.88 ^{ghijklm}	48	<i>C. scarabaeoides</i> ICP15683/ W15	38.10 ± 2.43 ^{bc}	191.27 ± 7.31 ^{uv}	170.37 ± 2.40 ^m
24	AL 2028	43.78 ± 0.64 ^{abc}	433.58 ± 11.48 ^{abcdeefgh}	198.44 ± 1.59 ^{cdefghijklm}	49	<i>C. scarabaeoides</i> ICP15701/ W15	40.54 ± 1.25 ^{abc}	158.53 ± 14.62 ^{uv}	184.36 ± 0.90 ^{hijklm}
25	AL 2046	46.21 ± 0.81 ^{abc}	423.84 ± 6.88 ^{bcdeefghi}	233.13 ± 13.97 ^{abcde}	50	<i>C. scarabaeoides</i> ICP15703/ W15	39.72 ± 0.81 ^{bc}	90.46 ± 10.96 ^v	196.21 ± 3.90 ^{bcdefghijklm}

Values are mean ± SD of triplicates and values with different letters in the same column are significantly different ($p < 0.05$)

saponin (0–40 µg) as standard. The phytic acid was extracted from the powdered seeds with 1.2% HCl and precipitated with 0.4% ferric chloride (Zemel and Shelef 1982). Organic phosphorus was estimated by treating the sample with concentrated HCl and perchloric acid (Rouser et al. 1974). Sodium phytate (5–30 µg) was used to prepare standard curve. Trypsin inhibitor was estimated by using *N*-α-benzoyl-DL-arginine p-nitroanilide (BAPNA) as a substrate by the method of Hajela et al. (1999). The sample (100 mg) was homogenized with 1 ml of 0.01 M phosphate buffer (pH 7.5) containing 0.1 M NaCl and stirred for one hour at room temperature. The supernatant obtained after centrifugation at 10,000×g for 30 min was then kept at 80 °C for 20 min and then recentrifuged again at 10,000×g for 30 min. Trypsin inhibitor potential was determined in the supernatant. 20 µl of trypsin inhibitor extract was incubated with 50 µl of bovine trypsin in the presence of 180 µl 0.01 M Tris HCl buffer (pH 7.5) containing 0.02 M CaCl₂ at 37 °C for 10 min. Residual trypsin activity was measured by adding 1 ml of BAPNA and incubated for 10 min at 37 °C. The reaction was terminated by adding 200 µl of 30% acetic acid. In the control experiment, instead of trypsin inhibitor 20 µl of extraction buffer was added. The absorbance was measured at 410 nm. One inhibitor unit is defined as the quantity which inhibits 50% of trypsin activity.

Antioxidant potential

The powdered seeds (100 mg) were extracted with 2 ml of methanol. This mixture was vortexed for 2 h and then centrifuged at 10,000 rpm for 10 min. The supernatant was collected and the pellet was re-extracted twice with methanol and the supernatants were pooled. The pooled supernatants were used to measure antioxidant potential of pigeonpea genotype in terms of total phenols, flavonols, DPPH (2,2-diphenyl-1-picryl hydrazyl) free radical scavenging activity, ferric reducing antioxidant power, total reducing power, hydroxyl radical scavenging activity and superoxide anion radical scavenging activity in pigeonpea genotypes.

Total phenols from methanolic extract was estimated by the method given by Swain and Hillis (1959). Flavonol content was estimated using methanolic solution of aluminum chloride and yellow colour developed was measured at 420 nm (Balabaa et al. 1974). Rutin (40–200 µg) was used to prepare standard curve.

Free radical scavenging activity of methanolic extract of pigeonpea was measured based on the scavenging activity of stable DPPH (2,2-diphenyl-1-picryl hydrazyl) free radical (Blois 1958). To 1 ml of the methanolic extract, 3 ml of the 0.1 mM DPPH reagent was added. After vortexing, the reaction mixture was incubated in the dark for 30 min

at room temperature and discoloration of DPPH was measured against reagent blank at 515 nm. The reaction mixture containing methanol instead of sample extract was used as control.

Ferric reducing antioxidant power was determined by the method of Benzie and Strain (1996) which is based on the ability of the sample to reduce Fe³⁺ to Fe²⁺ ions in the presence of TPTZ (2,4,6-Tri-(2-pyridyl)-5-triazine). Freshly prepared 1.9 ml of FRAP reagent (0.2 M Sodium acetate buffer (pH 3.6), 10 mM TPTZ in 40 mM HCl and 20 mM FeCl₃·6H₂O mixed in the ratio of 10:1:1, respectively) was incubated at 37 °C for 10 min and then 100 µl of sample was added to it. The absorbance of reaction mixture was measured at 593 nm after 30 min incubation at 37 °C. The FRAP activity was calculated from FeSO₄·7H₂O standards (5–30 µg) run simultaneously.

Total reducing power was determined by the method of Sreeramulu et al. (2009). To 250 µl of the appropriately diluted methanolic extract, 0.2 M sodium phosphate buffer (pH 6.6) was added to it to make its volume to 1 ml. The mixture was then incubated at 50 °C for 20 min after adding 500 µl potassium ferricyanide (10 mg/ml). The reaction was terminated with 500 µl TCA (100 mg/ml). The reaction mixture after vortexing was centrifuged at 1000 rpm for 10 min. The upper layer was mixed with equal volume of the distilled water and 0.1 ml of ferric chloride solution (1 mg/ml) was added to it. The absorbance of reaction mixture was measured spectrophotometrically at 700 nm against reagent blank. Ascorbic acid (5–40 µg) was used to prepare standard curve simultaneously.

Hydroxyl radical scavenging activity was determined by the modified method of Li et al. (2008) and is based on the Fenton-type reaction system generating hydroxyl radical. The reaction mixture contained 1 ml of 1,10-phenanthroline (0.75 mM) and 1 ml of ferrous sulphate (0.75 mM), 1.5 ml of 0.2 M phosphate buffer (pH 7.4) and 0.5 ml of sample extract. The reaction mixture without sample was termed as control. The reaction was started by adding 1 ml of hydrogen peroxide (0.01%) to each tube. The absorbance was measured at 536 nm after incubating the reaction mixture at 37 °C for 60 min. Percentage scavenging activity was calculated. The superoxide radical scavenging activity was estimated by spectrophotometric monitoring the inhibition of pyrogallol autoxidation as described by Marklund and Marklund (1974). Pyrogallol solution (3 mM) was added into a tube containing sample mixture previously dissolved in 0.1 M Tris-HCl EDTA buffer (pH 8.0). The optical density was measured at 320 nm and the antioxidant activity was determined as the percentage of inhibiting pyrogallol autoxidation, which was calculated from the optical density in the presence or absence of pyrogallol and sample mixture.

Categorization of pigeonpea genotypes into equal classes

The maximum value (max) of each parameter (nutritional, antinutritional and antioxidant activity) was subtracted from its minimum value (min) and the obtained value ($d = \text{max} - \text{min}$) was divided by 3 so as three equal classes were prepared having a class interval of d . The pigeonpea genotypes were categorized into three groups; low, medium and high for all the parameters on the basis of these equal classes.

Statistical analysis

Mean and standard deviation were calculated. The results of seed data were analyzed by Tukey's test to identify the significant relation between the genotypes and the biochemical parameters. Cluster analysis and principle component analysis (PCA) was carried out using the software Minitab. The correlation between the nutritional, antinutritional and antioxidant potential was also estimated using the software SPSS pc version 20.0.

Results and discussion

Carbohydrates

Total soluble sugars, starch and total soluble protein content in pigeonpea genotypes are represented in Table 1. The total soluble sugars content varied from 33.23 mg/g (AL 2009) to 60.80 mg/g (AL 201) with mean value of 43.66 mg/g. The genotypes were divided into three equal classes as high, medium and low groups based on equal class distribution method. The pigeonpea genotypes with total soluble sugar content higher than 51.61 mg/g I were categorized under higher total soluble sugar group, value between 42.42 and 51.61 mg/g placed under medium total soluble sugar group and between 33.23 and 42.42 mg/g placed under low total soluble sugar group ($H \geq 51.61$ $L \leq 42.42$). One cultivar (AL 201) and two advance breeding lines, AL 1960 and AL 2060 possessed higher total sugars ranging between 51.61 and 60.80 mg/g. Twenty seven genotypes possessed medium content of total soluble sugars and the remaining twenty genotypes had lower content of total soluble sugars. The soluble sugar content in wild pigeonpea species ranged from 38.10 to 40.54 mg/g. Sharma et al. (2011) reported 31 mg/g soluble sugars in mature seeds of pigeonpea. Total soluble sugar content in lentil varied from 34.5 to 70.0 mg/g (Ahuja et al. 2015).

Starch content in the pigeonpea cultivars and advance breeding lines varied from 272.70 mg/g (AL 2095) to

521.28 mg/g (AL 1931) and in wild species it varied from 90.46 to 191.27 mg/g (Table 1). The wild genotypes had the lowest starch content. The range for high, medium and low starch content in all the pigeonpea genotypes ranged from: 377.66–521.28, 234.06–377.66 and 90.46–234.06 mg/g, respectively ($H \geq 377.66$ $L \leq 234.06$). Twenty four genotypes had higher starch content and eleven genotypes had medium starch content and rest of genotypes had lower starch content. Six genotypes namely AL 201, AL 1849, AL 1931, AL 2049, AL 2060 and AL 2063 had starch content more than 44% (440.89–521.28 mg/g). Starch content in pigeonpea seeds had been reported to be 41 and 53% by Torres et al. (2006) and Sharma et al. (2011), respectively.

Grain legumes are important sources of food proteins. Proteins in legume seeds represent from about 20% (dry weight) in pea and beans, up to 38–40% in soybean and lupin. The young pods and dried seeds of legumes contain 4.5% protein. Plant seeds are a major source of dietary proteins. The total soluble protein content in pigeonpea genotypes varied from 170.37 to 251.16 mg/g (Table 1). Lowest total soluble protein content was found in *C. scarabaeoides* 1CP15683/W15 and highest content was found in AL 2000. Eight genotypes (AL 1960, AL 2000, AL 2009, AL 2024, AL 2046, AL 2091, AL 2092, AL 2098) had higher protein content (222.4–251.16 mg/g), twenty genotypes had medium protein content (197.27–222.4 mg/g) and the remaining sixteen genotypes had lower protein content (170.37–197.27 mg/g). Protein content in pigeonpea genotypes has been reported to be 25.97% (Fasoryio et al. 2005), 18.8% (Sharma et al. 2011) and 22% (Sabahelkhier et al. 2014). Protein content of HTC (hard-to-cook) grains from different kidney bean accessions ranged from 19.06 to 28.89% and for ETC (easy-to-cook) grains, it ranged from 21.01 to 29.24% (Parmar et al. 2017).

Bound fructose of sucrose and raffinose series oligosaccharides

Raffinose series oligosaccharides (RFO's) are known to cause flatulence and the negative impacts of RFO's are mainly due to the absence of α -galactosidase enzyme in small intestine, which is necessary for the hydrolysis of α -1, 6 linkages present in RFOs (Ramadoss and Shunmugam 2014). The data of bound fructose of sucrose and raffinose series oligosaccharide has been depicted (Table 2). The highest content of bound fructose was found to be 14.27 mg/g (AL 2019) and lowest was found to be 4.83 mg/g (*C. scarabaeoides* 1CP15683/W15) with mean value of 10.42 mg/g. Three genotypes namely *C. scarabaeoides* 1CP15683/W15, AL 20678 and AL 2102 had lower content of bound fructose (7.11–9.53 mg/g). Twenty one genotypes had higher content of bound fructose

(11.95–14.39 mg/g) and twenty seven genotypes had medium level of bound fructose (9.53–11.95 mg/g). Bound fructose in chickpea genotypes had been reported from 7.64 to 13.39 mg/g (Kaur et al. 2016).

Tannins

Tannin content in pigeonpea genotypes was found to be in the range of 14.61 mg/g (AL 1974) to 29.40 mg/g (*C. scarabaeoides* ICP15703/W15) with an average value of 20.02 mg/g (Table 2). Two advance breeding lines (AL 2078, AL 2088) and two wild species (*C. scarabaeoides* ICP15701/W15, *C. scarabaeoides* ICP15703/W15) had higher tannin content (24.47–29.40 mg/g). Nineteen genotypes had medium tannin content (19.54–24.47 mg/g) and the twenty seven genotypes had medium content of tannin (14.61–19.54 mg/g). Singh and Jambunathan (1981) reported the range of tannins in pigeonpea to be 4.3–11.4 mg/g. Tannin-protein complexes are reported to be responsible for low protein digestibility, decreased amino acid availability and increased fecal nitrogen. These complexes may not be dissociated and may thus be excreted with the feces (Jain et al. 2009).

Trypsin inhibitor activity

Protease inhibitors are present in significant amount in storage organs of plants, especially seeds (Mittal et al. 2014). Trypsin inhibitor activity (TIA) in pigeonpea genotypes varied significantly from 23.53 to 103.20 IU/g with an average value of 79.87 IU/g (Table 3). TIA in wild species was found to be lie between 23.53 and 65.88 IU/g. In other genotypes the activity ranged from 68.20 to 103.20 IU/g. TIA in pigeonpea genotypes has been reported to be 24.72 TIU/mg (Torres et al. 2006), 9.9 TIU/mg (Sharma et al. 2011) and 30.27 TIU/g. (Fasoyiro et al. 2005).

Phytic acid

Phytic acid is known as an antinutritional factor as it chelates micronutrient and decreases their bioavailability for monogastric animals, including humans because they lack enzyme phytase in their digestive tract (Gupta et al. 2015). Phytic acid content in different pigeonpea genotypes varied from 1.57 to 6.54 mg/g with mean value of 4.07 mg/g (Table 3). Eleven genotypes namely AL 1758, AL 1849, AL 1922, AL 1957, AL 1960, 1976, AL 1992, AL 2046, AL 2049, AL 2086 and wild species *C. scarabaeoides* ICP15701/W15 had higher phytic acid content ranging from 4.87 mg/g to 6.54 mg/g (Table 3). Twenty six genotypes had medium content of phytic acid (3.22–4.87 mg/g) and thirteen genotypes had lower content

of phytic acid (1.57–3.22 mg/g). Phytic acid content in pigeonpea was reported to be 4.80 g/100 g (Fasoyiro et al. 2005). The average phytic acid content in ETC (easy to cook) and HTC (hard to cook) grains of kidney accessions was found to be 2.09 and 1.89 mg/g, respectively (Parmar et al. 2017).

Saponins

Saponin content in pigeonpea genotypes varied from 4.73 mg/g (AL 2087) to 17.98 mg/g (AL 2060) with an average value of 10.01 mg/g (Table 3). Thirteen genotypes had low content of saponins ranging from 4.73 to 7.37 mg/g. Nine genotypes (AL 15, AL 881, AL 1758, AL 1931, AL 2025, AL 2028, AL 2046, AL 2049 and AL 2060) had high content of saponin ranging from 13.55 to 17.98 mg/g. Twenty two pigeonpea genotypes had lower content of and nineteen genotypes had medium content of saponins. Jain et al. (2009) reported the saponins content in pigeonpea to be 0.5 to 2.3 mg/g whereas Duhan et al. (2001) observed that saponin content of unprocessed pigeonpea cultivars ranged from 21.64 to 34.94 mg/g. Kaur et al. (2016) reported that saponin content in chickpea cultivars ranged from 9.29 mg/g to 35.14 mg/g. Saponins are a heterogeneous group of naturally occurring triterpene or steroidal glycosides that occur in a wide range of plants, including pulses and oil seeds. It affects animal performance and metabolism in a number of ways like erythrocyte haemolysis, reduction of blood and liver cholesterol, depression of growth rate, inhibition of smooth muscle activity, enzyme inhibition and reduction in nutrient absorption. Saponins produce some toxic effects when ingested as they alter cell wall permeability. Saponin bind to the cells of the small intestine affecting the absorption of nutrients across the intestinal wall (Akande et al. 2010).

Total phenol and flavonols in pigeonpea genotypes

The highest phenolic content was found to be 1.29 mg/g (*C. scarabaeoides* ICP15683/W15) and the lowest was found to be 0.41 mg/g (AL 2086) with an average value of 0.74 mg/g (Table 2). Ten pigeonpea genotypes namely AL 15, AL 201, AL 1843, AL 1931, AL 1932, AL 1933, AL 1957, AL 1960, AL 2046 and *C. scarabaeoides* ICP15683/W15 had higher content of total phenols (0.99–1.29 mg/g). Saxena et al. (2010) reported that total phenol content varied from 3 to 18.3 mg/g in pigeonpea genotypes. Marathe et al. (2011) reported that phenolic content in different legumes lablab bean, chickpea, lentils, cowpea, green gram, pigeonpea and horsegram ranged from 0.325 to 6.378 mg/g and in pigeonpea it was found to be 1.054 mg/g. They reported that there was no correlation of phenolics with seed coat colour but it was negatively correlated with

Table 2 Distribution of bound fructose (mg/g), total phenols (mg/g), tannins (mg/g) and flavonol (mg/100 g) in pigeonpea genotypes

S. No.	Genotypes	Bound Fructose	Total phenols	Tannins	Flavonol
1	AL 15	8.45 ± 0.21 ^{ijk}	1.15 ± 0.02 ^{abc}	21.53 ± 0.61 ^{defgh}	16.02 ± 0.80 ^a
2	PAU 881	13.52 ± 0.49 ^{ab}	0.61 ± 0.07 ^{ghijklm}	19.19 ± 0.73 ^{ghijklm}	8.03 ± 0.19 ^{ijklm}
3	AL 201	13.04 ± 0.09 ^{abc}	1.19 ± 0.18 ^{bcd}	23.65 ± 0.65 ^{cdef}	7.10 ± 0.15 ^{klmn}
4	AL 1758	9.10 ± 0.05 ^{hij}	0.63 ± 0.02 ^{ghijkl}	19.61 ± 0.38 ^{ghijkl}	8.66 ± 0.51 ^{hijkl}
5	AL 1843	11.59 ± 0.04 ^{bcde}	0.99 ± 0.01 ^{bcd}	19.03 ± 0.03 ^{ghijklm}	7.21 ± 0.08 ^{klmn}
6	AL 1849	11.63 ± 0.08 ^{bcde}	0.72 ± 0.01 ^{ghijk}	19.26 ± 0.65 ^{ghijkl}	7.00 ± 0.12 ^{klmn}
7	AL 1922	10.95 ± 0.13 ^{defg}	0.63 ± 0.02 ^{ghijkl}	16.73 ± 0.5 ^{klmn}	8.17 ± 0.17 ^{ijklm}
8	AL 1931	13.35 ± 0.15 ^{ab}	1.04 ± 0.01 ^{bc}	19.30 ± 0.46 ^{ghijkl}	7.57 ± 0.34 ^{ijklmn}
9	AL 1932	12.96 ± 0.39 ^{abc}	1.03 ± 0.01 ^{bc}	17.61 ± 0.61 ^{ijklmn}	6.73 ± 0.08 ^{klmn}
10	AL 1933	13.09 ± 0.04 ^{abc}	1.06 ± 0.05 ^{bc}	18.57 ± 1.19 ^{ghijklm}	7.69 ± 0.28 ^{ijklmn}
11	AL 1957	13.60 ± 0.16 ^{ab}	1.02 ± 0.01 ^{bc}	19.92 ± 0.07 ^{fg hijk}	7.39 ± 0.34 ^{ijklmn}
12	AL 1960	13.15 ± 0.06 ^{abc}	1.20 ± 0.03 ^{ab}	23.76 ± 1.70 ^{cdef}	8.14 ± 0.27 ^{ijklm}
13	AL 1965	12.61 ± 0.02 ^{abc}	0.97 ± 0.03 ^{cde}	19.03 ± 0.42 ^{ghijklm}	7.63 ± 0.17 ^{ijklm}
14	AL 1974	12.33 ± 0.22 ^{abcd}	0.65 ± 0.02 ^{ghijkl}	14.61 ± 0.61 ⁿ	6.61 ± 0.17 ^{lmn}
15	AL 1976	10.69 ± 0.38 ^{efgh}	0.97 ± 0.12 ^{cde}	19.73 ± 0.03 ^{fg hijk}	7.04 ± 0.23 ^{klmn}
16	AL 1992	13.54 ± 0.13 ^{ab}	0.52 ± 0.01 ^{klm}	16.15 ± 2.31 ^{lmn}	7.78 ± 0.63 ^{ijklmn}
17	AL 2000	10.38 ± 0.16 ^{efgh}	0.71 ± 0.03 ^{ghijk}	18.30 ± 0.15 ^{hijklmn}	6.67 ± 0.42 ^{lmn}
18	AL 2009	11.57 ± 0.31 ^{cdef}	0.96 ± 0.06 ^{cdef}	19.26 ± 0.57 ^{ghijkl}	7.03 ± 0.34 ^{klmn}
19	AL 2016	13.10 ± 0.05 ^{abc}	0.76 ± 0.05 ^{efghi}	18.49 ± 0.49 ^{ghijklm}	6.46 ± 0.04 ^{mn}
20	AL 2019	14.27 ± 0.05 ^a	0.55 ± 0.02 ^{ijklm}	16.76 ± 0.30 ^{klmn}	6.67 ± 0.08 ^{klmn}
21	AL 2021	13.18 ± 0.51 ^{ab}	0.65 ± 0.13 ^{ghijkl}	17.07 ± 0.38 ^{ijklmn}	5.32 ± 0.63 ⁿ
22	AL 2024	12.29 ± 0.10 ^{bcde}	0.80 ± 0.03 ^{defg}	21.07 ± 0.61 ^{defghi}	8.57 ± 0.04 ^{ghijk}
23	AL 2025	13.15 ± 0.10 ^{abc}	0.76 ± 0.16 ^{efgh}	20.26 ± 0.96 ^{fg hijk}	6.22 ± 0.29 ^{mn}
24	AL 2028	12.03 ± .34 ^{bcde}	0.72 ± 0.01 ^{ghijk}	18.15 ± 0.07 ^{hijklmn}	7.66 ± 0.07 ^{ijklm}
25	AL 2046	11.65 ± 0.17 ^{cdef}	1.00 ± 0.01 ^{bcd}	20.23 ± 0.69 ^{efghij}	6.67 ± 0.49 ^{lmn}
26	AL 2049	8.06 ± 0.20 ^{jk}	0.74 ± 0.01 ^{fg hij}	21.57 ± 0.65 ^{defgh}	11.46 ± 0.78 ^{cdef}
27	AL 2060	9.66 ± 0.21 ^{ghij}	0.96 ± 0.01 ^{cde}	20.34 ± 0.76 ^{efghij}	9.84 ± 0.14 ^{efghi}
28	AL 2061	8.39 ± 0.19 ^{ijk}	0.58 ± 0.01 ^{ghijklm}	19.48 ± 0.68 ^{ghijkl}	12.80 ± 0.51 ^{bcd}
29	AL 2062	9.24 ± 0.10 ^{hij}	0.62 ± 0.02 ^{ghijklm}	18.58 ± 0.55 ^{ghijklm}	9.22 ± 0.58 ^{fg hij}
30	AL 2063	8.83 ± 0.22 ^{hij}	0.66 ± 0.01 ^{ghijkl}	18.84 ± 0.47 ^{ghijklm}	8.96 ± 0.07 ^{ghijk}
31	AL 2067	7.82 ± 0.14 ^{jk}	0.63 ± 0.03 ^{ghijkl}	19.57 ± 0.51 ^{ghijkl}	11.50 ± 0.29 ^{cdef}
32	AL 2071	8.05 ± 0.17 ^{jk}	0.54 ± 0.01 ^{ijklm}	16.62 ± 0.12 ^{klmn}	9.07 ± 0.13 ^{ghijk}
33	AL 2078	9.17 ± 0.29 ^{hij}	0.64 ± 0.01 ^{ghijkl}	25.90 ± 0.12 ^{abc}	11.41 ± 0.13 ^{cdef}
34	AL 2086	9.31 ± 0.06 ^{hij}	0.41 ± 0.01 ^m	15.89 ± 0.46 ^{mn}	8.97 ± 0.13 ^{ghijk}
35	AL 2087	8.37 ± 0.23 ^{ijk}	0.61 ± 0.02 ^{ghijklm}	18.29 ± 0.25 ^{ghijklm}	10.38 ± 0.20 ^{defgh}
36	AL 2088	7.99 ± 0.23 ^{jk}	0.67 ± 0.02 ^{ghijkl}	24.57 ± .012 ^{bcd}	9.32 ± 0.14 ^{ghijk}
37	AL 2090	9.38 ± 0.12 ^{fg hij}	0.54 ± 0.01 ^{ijklm}	17.26 ± 0.51 ^{ijklmn}	11.70 ± 0.13 ^{cde}
38	AL 2091	8.05 ± 0.04 ^{jk}	0.46 ± 0.03 ^{lm}	19.78 ± 0.29 ^{ghijkl}	9.09 ± 0.06 ^{ghijk}
39	AL 2092	8.74 ± 0.31 ^{ijk}	0.62 ± 0.01 ^{ghijklm}	18.93 ± 0.12 ^{ghijklm}	11.65 ± 0.06 ^{cde}
40	AL 2094	9.26 ± 0.35 ^{ghij}	0.46 ± 0.01 ^{lm}	24.18 ± 0.25 ^{bcde}	14.25 ± 0.36 ^{ab}
41	AL 2095	11.45 ± 0.20 ^{cdef}	0.54 ± 0.01 ^{ijklm}	21.70 ± 0.08 ^{defg}	11.99 ± 0.13 ^{cde}
42	AL 2096	10.03 ± 0.14 ^{fg hij}	0.60 ± 0.02 ^{ghijklm}	18.80 ± 0.51 ^{ghijklm}	14.63 ± 0.41 ^{ab}
43	AL 2098	10.48 ± 0.31 ^{efgh}	0.54 ± 0.03 ^{ijklm}	21.62 ± 0.94 ^{defg}	10.29 ± 0.20 ^{efghi}
44	AL 2101	8.28 ± 0.24 ^{ijk}	0.52 ± 0.02 ^{klm}	18.88 ± 0.76 ^{ghijklm}	10.70 ± 0.24 ^{cdefg}
45	AL 2102	6.87 ± 0.36 ^k	0.59 ± 0.02 ^{ghijklm}	21.79 ± 0.68 ^{defgh}	10.43 ± 0.13 ^{defgh}
46	AL 2108	8.01 ± 0.26 ^{jk}	0.55 ± 0.01 ^{hijklm}	18.97 ± 0.51 ^{ghijklm}	12.07 ± 0.38 ^{cde}

Table 2 continued

S. No.	Genotypes	Bound Fructose	Total phenols	Tannins	Flavonol
47	AL 2122	8.45 ± 0.17 ^{ijk}	0.65 ± 0.01 ^{ghijkl}	21.58 ± 0.98 ^{defg}	13.03 ± 0.06 ^{bc}
48	<i>C. scarabaeoides</i> 1CP15683/W15	4.83 ± 0.09 ^l	1.29 ± 0.02 ^a	23.88 ± 0.38 ^{bcde}	6.76 ± 0.25 ^{klmn}
49	<i>C. scarabaeoides</i> 1CP15701/W15	8.95 ± 0.20 ^{hij}	0.69 ± 0.02 ^{ghijk}	27.30 ± 0.72 ^{ab}	8.68 ± 0.55 ^{hijkl}
50	<i>C. scarabaeoides</i> 1CP15703/W15	8.41 ± 0.23 ^{ijk}	0.70 ± 0.05 ^{ghijk}	29.40 ± 0.68 ^a	10.87 ± 0.20 ^{cdefg}

Values are mean ± SD of triplicates and values with different letters in the same column are significantly different ($p < 0.05$)

seed size. Our results showed consistency with this as the wildtype species (*C. scarabaeoides* 1CP15683/W15) with smallest seed size had the highest phenolic content. However, Parmar et al. (2014) reported that total phenol content varied with the color of kidney bean. Nwosu et al. (2013) reported total phenol content in pigeonpea to be 1.6 mg/g.

Flavonol content in pigeonpea lied in the range between 5.32 mg/100 g and 16.02 mg/100 g with mean value of 9.18 mg/100 g (Table 2). The values for characterizing genotypes w.r.t flavonols was found to be $H \geq 12.44$ $L \leq 8.88$. AL 15, AL 2094 and AL 2096 had the higher phenol content. Total phenolics are naturally produced during the growth and development of plants to protect themselves from biotic stresses such as diseases, insects and environmental stresses (Khang et al. 2016). They can delay or inhibit oxidation process of lipids by inhibiting the initiation or propagation of oxidative chain reactions (Khanum et al. 2015). Flavonoids also act as natural antioxidants (Bouaziz et al. 2005). They are widespread plant secondary metabolites including flavones, flavanols, and condensed tannins. Epidemiological studies suggest that the consumption of flavonoid-rich foods protects against human diseases which are associated with oxidative stress. In vitro, flavonoids from several plant sources have shown free-radical scavenging activity and protection against oxidative stress.

Free radical scavenging activity

The principle of DPPH radical scavenging assay is based on the reduction of 2,2-diphenyl-1-picrylhydrazyl (DPPH). Due to the presence of an odd electron it gives a strong absorption maximum at 517 nm. As this electron becomes paired of in the presence of a hydrogen donor, i.e. a free radical scavenging antioxidant, the absorption strength is decreased. The DPPH free radical scavenging activity in pigeonpea genotypes ranged from 18.28 to 93.76% (Table 4). Five pigeonpea genotypes namely AL 1960, AL 1965, AL 1976, AL 1992 and *C. scarabaeoides* 1CP15683/W15 had the higher scavenging activity ranging from 68.60 to 93.76%. Twenty four genotypes were found to have the lower rate of scavenging from 18.28 to 43.444%. The

remaining twenty one genotypes had medium range of scavenging activity ranging from 43.44 to 68.60%. Higher the DPPH scavenging activity, higher is the antioxidant power i.e. the capacity of the sample to inhibit the free radicals. Marathe et al. (2011) compared the DPPH free radical scavenging activities in different commonly consumed legumes and reported that horse gram, common bean, cowpea and fenugreek had higher free radical scavenging activities; lablab bean, chickpea, butter bean and pea had lower scavenging activities and green gram, black gram, pigeon pea, lentils, cowpea and common bean had intermediate scavenging activities.

FRAP (ferric reducing antioxidant power)

Ferric reducing antioxidant power (FRAP) assay is based on the reduction of ferric tripyridyl triazine (Fe^{3+} -TPTZ) complex into ferric tripyridyl triazine (Fe^{2+} -TPTZ) form in the presence of antioxidants (Thaipong et al. 2006). FRAP activity in pigeonpea genotypes ranged from 3.11 mg/g (AL 1758) to 5.36 mg/g with an average value of 3.74 mg/g (AL 1932) (Table 4). Twelve genotypes possessed medium FRAP activity ranging from 3.86 to 4.61 mg/g and three genotypes namely AL 1932, *C. scarabaeoides* 1CP1515683/W15 and *C. scarabaeoides* 1CP1515703/W15 possessed higher FRAP activity ranging from 4.61 to 5.36 mg/g. Other genotypes had FRAP activity in lower range (3.11–3.86 mg/g). Marathe et al. (2011) reported that FRAP activity in pigeonpea, lentil, pea, blackgram and cowpea was found to lie in the medium range from 2.38 to 4.58 mg/g.

Total reducing power

The reducing properties are generally associated with the presence of reductones which break the free radical chain by donating a hydrogen atom. They also react with precursors of peroxides, thus prevent the peroxide formation (Kumaran and Karunakaran 2007). In this assay, the yellow color of the test solution changes to various shades of green and blue based upon the reducing power of the extract. The presence of reductants causes the reduction of the

Table 3 Distribution of trypsin inhibitor activity (IU/g), phytic acid (mg/g) and saponins (mg/g) in pigeonpea genotypes

S. No.	Genotypes	Trypsin inhibitor activity	Phytic acid	Saponins	S. No.	Genotypes	Trypsin inhibitor activity	Phytic acid	Saponins
1	AL 15	90.30 ± 5.42 ^{abcdef}	3.74 ± 0.97 ^{abcde}	15.42 ± 0.28 ^{bc}	26	AL 2049	68.44 ± 7.42 ^{fgh}	6.54 ± 0.56 ^a	16.51 ± 0.25 ^{ab}
2	PAU 881	73.53 ± 0.16 ^{cdefgh}	4.43 ± 0.28 ^{abcde}	14.41 ± 0.17 ^{bcd}	27	AL 2060	85.32 ± 8.79 ^{abcdefgh}	4.15 ± 0.75 ^{abcde}	17.98 ± 0.38 ^a
3	AL 201	86.91 ± 4.45 ^{abcdefgh}	4.87 ± 0.34 ^{abcd}	12.92 ± 0.32 ^{defg}	28	AL 2061	77.14 ± 0.61 ^{bcd}	3.05 ± 0.47 ^{abcde}	7.56 ± 0.33 ^{mnopq}
4	AL 1758	86.97 ± 4.24 ^{abcdefgh}	5.59 ± 0.25 ^{abc}	13.61 ± 0.24 ^{cdef}	29	AL 2062	85.73 ± 6.38 ^{abcdefgh}	2.79 ± 0.34 ^{cde}	8.92 ± 0.19 ^{klmn}
5	AL 1843	83.85 ± 3.67 ^{abcdefgh}	4.40 ± 0.62 ^{abcde}	12.33 ± 0.01 ^{efgh}	30	AL 2063	86.71 ± 1.58 ^{abcdefgh}	1.57 ± 0.06 ^e	8.59 ± 0.24 ^{klmno}
6	AL 1849	85.52 ± 3.40 ^{abcdefgh}	5.59 ± 0.06 ^{ab}	10.76 ± 0.17 ^{ghij}	31	AL 2067	90.50 ± 1.05 ^{abcdef}	3.39 ± 0.37 ^{abcde}	5.20 ± 0.24 ^f
7	AL 1922	76.99 ± 6.99 ^{bcdefgh}	6.19 ± 0.28 ^{ab}	12.11 ± 0.30 ^{efgh}	32	AL 2071	85.48 ± 1.07 ^{abcdefgh}	4.11 ± 0.66 ^{abcde}	6.01 ± 0.19 ^{pqr}
8	AL 1931	80.11 ± 4.60 ^{bcdefgh}	4.37 ± 0.09 ^{abcde}	14.28 ± 0.46 ^{bcd}	33	AL 2078	89.48 ± 2.40 ^{abcdefg}	3.96 ± 0.75 ^{abcde}	6.97 ± 0.22 ^{nopqr}
9	AL 1932	103.20 ± 4.25 ^a	4.90 ± 1.00 ^{abcd}	8.67 ± 0.35 ^{klmno}	34	AL 2086	88.29 ± 3.37 ^{abcdefg}	5.15 ± 0.31 ^{abcd}	7.23 ± 0.06 ^{nopqr}
10	AL 1933	84.62 ± 1.52 ^{abcdefgh}	2.70 ± 0.37 ^{cde}	6.46 ± 0.67 ^{pqr}	35	AL 2087	85.98 ± 6.35 ^{abcdefgh}	3.01 ± 0.18 ^{bcd}	4.73 ± 0.12 ^f
11	AL 1957	74.68 ± 0.98 ^{cdefgh}	5.44 ± 0.34 ^{abc}	7.40 ± 0.24 ^{mnopq}	36	AL 2088	90.72 ± 0.38 ^{abcdef}	2.48 ± 0.47 ^{cde}	11.79 ± 0.01 ^{efgh}
12	AL 1960	75.40 ± 2.41 ^{cdefgh}	4.90 ± 0.18 ^{abcde}	9.53 ± 0.38 ^{ijklm}	37	AL 2090	80.29 ± 3.19 ^{bcdefgh}	3.39 ± 0.37 ^{abcde}	5.58 ± 0.11 ^{qr}
13	AL 1965	70.29 ± 0.74 ^{efgh}	4.59 ± 0.75 ^{abcde}	9.61 ± 0.33 ^{ijklm}	38	AL 2091	91.11 ± 0.33 ^{abcde}	4.43 ± 0.09 ^{abcde}	9.80 ± 0.04 ^{hijkl}
14	AL 1974	80.25 ± 7.90 ^{bcdefgh}	2.61 ± 0.09 ^{bcde}	9.44 ± 0.41 ^{ijklm}	39	AL 2092	98.41 ± 1.01 ^{ab}	3.67 ± 0.09 ^{abcde}	10.57 ± 0.11 ^{hijk}
15	AL 1976	67.44 ± 1.49 ^{gh}	5.06 ± 0.91 ^{abcd}	8.33 ± 0.33 ^{klmno}	40	AL 2094	91.83 ± 1.38 ^{abcde}	4.24 ± 0.40 ^{abcde}	4.84 ± 0.30 ^f
16	AL 1992	74.30 ± 0.15 ^{cdefgh}	6.06 ± 0.84 ^{ab}	10.72 ± 0.16 ^{ghij}	41	AL 2095	82.02 ± 3.71 ^{abcdefgh}	3.58 ± 0.25 ^{abcde}	5.74 ± 0.17 ^{pqr}
17	AL 2000	70.75 ± 5.25 ^{efgh}	4.15 ± 1.00 ^{abcde}	11.44 ± 0.14 ^{ghi}	42	AL 2096	91.02 ± 0.55 ^{abcde}	3.86 ± 0.59 ^{abcde}	6.67 ± 0.24 ^{opqr}
18	AL 2009	84.50 ± 4.12 ^{abcdefgh}	3.30 ± 0.47 ^{abcde}	12.83 ± 0.28 ^{defg}	43	AL 2098	77.90 ± 0.18 ^{bcdefgh}	3.61 ± 0.09 ^{abcde}	8.12 ± 0.38 ^{klmno}
19	AL 2016	72.91 ± 4.57 ^{cdefgh}	3.93 ± 0.34 ^{abcde}	11.37 ± 0.40 ^{ghi}	44	AL 2101	86.36 ± 1.74 ^{abcdefgh}	3.14 ± 0.31 ^{bcd}	8.08 ± 0.24 ^{lmnop}
20	AL 2019	69.69 ± 1.19 ^{efgh}	4.02 ± 0.88 ^{abcde}	10.57 ± 0.33 ^{hijk}	45	AL 2102	85.75 ± 1.24 ^{abcdefgh}	4.62 ± 0.34 ^{abcde}	7.61 ± 0.19 ^{mnopq}
21	AL 2021	72.39 ± 0.17 ^{cdefgh}	3.67 ± 0.84 ^{abcde}	10.83 ± 0.30 ^{ghij}	46	AL 2108	93.55 ± 0.66 ^{abcd}	4.15 ± 0.50 ^{abcde}	7.13 ± 0.03 ^{nopqr}
22	AL 2024	72.02 ± 6.94 ^{defgh}	4.15 ± 0.50 ^{abcde}	13.45 ± 0.27 ^{cdef}	47	AL 2122	94.54 ± 3.21 ^{abc}	1.66 ± 0.34 ^e	11.28 ± 0.24 ^{fghi}
23	AL 2025	68.20 ± 3.50 ^{fgh}	3.11 ± 0.09 ^{bcde}	14.22 ± 0.30 ^{bcd}	48	C. scarabaeoides ICP15683/W15	23.53 ± 3.13 ^j	2.76 ± 0.12 ^{bcd}	9.77 ± 0.40 ^{ijklm}
24	AL 2028	73.09 ± 2.49 ^{cdefgh}	4.62 ± 0.40 ^{abcde}	14.02 ± 0.33 ^{cde}	49	C. scarabaeoides ICP15701/W15	64.88 ± 1.29 ^{hi}	5.18 ± 0.22 ^{abcd}	5.85 ± 0.19 ^{pqr}
25	AL 2046	76.52 ± 0.94 ^{bcdefgh}	4.96 ± 0.25 ^{abcde}	16.41 ± 0.25 ^{ab}	50	C. scarabaeoides ICP15703/W15	44.32 ± 0.23 ^{ij}	3.64 ± 0.44 ^{abcde}	6.72 ± 0.57 ^{opqr}

Values are mean ± SD of triplicates and values with different letters in the same column are significantly different ($p < 0.05$)

Table 4 Distribution of FRAP(mg/g), reducing power(mg/g), scavenging activity(%) of DPPH, hyrdoxal radical and superoxide anion radical in pigeonpea genotypes

S. No	Genotypes	DPPH	FRAP	Reducing power	Hydroxyl radical scavenging activity	Superoxide anion radical scavenging activity
1	AL 15	50.2 ± 6.3 ^{cdefghijklm}	3.49 ± 0.03 ^{ab}	41.9 ± 0.8 ^{def}	7.8 ± 0.5 ^{op}	27.5 ± 0.8 ^{ijklmnop}
2	PAU 881	35.9 ± 5.0 ^{ijklmnop}	3.30 ± 0.19 ^b	31.5 ± 2.0 ^{mnopq}	24.4 ± 0.4 ^{fghij}	32.5 ± 0.8 ^{fghijklmn}
3	AL 201	41.6 ± 0.5 ^{fghijklmnop}	3.25 ± 0.04 ^b	34.8 ± 1.1 ^{hijklm}	30.2 ± 1.8 ^{gbcdefgh}	33.3 ± 3.3 ^{fghijklm}
4	AL 1758	35.3 ± 1.5 ^{ijklmnop}	3.11 ± 0.02 ^b	24.4 ± 0.2 ^{uvw}	22.0 ± 1.0 ^{hijk}	32.5 ± 2.5 ^{fghijklmn}
5	AL 1843	48.5 ± 4.4 ^{cdefghijklmn}	3.44 ± 0.04 ^b	31.7 ± 1.7 ^{lmnop}	10.5 ± 1.2 ^{lmnop}	30.0 ± 0.9 ^{ghijklmno}
6	AL 1849	36.6 ± 6.6 ^{hijklmnop}	3.58 ± 0.18 ^{ab}	24.3 ± 0.8 ^{uvw}	34.9 ± 0.8 ^{abcd}	41.6 ± 0.8 ^{abcdef}
7	AL 1922	42.2 ± 1.0 ^{fghijklmno}	3.11 ± 0.05 ^b	27.2 ± 0.5 ^{rstu}	31.4 ± 0.3 ^{bcdef}	25.8 ± 0.8 ^{klmnopq}
8	AL 1931	57.7 ± 0.9 ^{cdefghi}	4.51 ± 0.09 ^{ab}	28.1 ± 0.6 ^{pqrst}	28.4 ± 1.0 ^{bcdefgh}	33.3 ± 3.3 ^{fghijklm}
9	AL 1932	61.6 ± 1.6 ^{cdef}	5.36 ± 0.05 ^a	45.3 ± 1.5 ^d	22.9 ± 1.17 ^{ghijk}	22.5 ± 2.5 ^{mnopqrs}
10	AL 1933	50.1 ± 6.0 ^{cdefghijklm}	3.45 ± 0.15 ^{ab}	32.4 ± 0.7 ^{klmno}	31.7 ± 0.4 ^{bcdef}	19.1 ± 0.8 ^{opqrs}
11	AL 1957	53.9 ± 2.2 ^{cdefghij}	3.33 ± 0.04 ^b	28.1 ± 0.1 ^{qrstu}	35.2 ± 1.1 ^{abcd}	49.1 ± 2.5 ^{ab}
12	AL 1960	70.1 ± 4.2 ^{bcd}	3.68 ± 0.08 ^{ab}	32.6 ± 0.9 ^{klmno}	28.3 ± 0.6 ^{bcdefgh}	28.3 ± 3.3 ^{ijklmnop}
13	AL 1965	71.2 ± 7.0 ^{abcd}	3.63 ± 0.23 ^{ab}	35.4 ± 0.3 ^{ijkl}	27.7 ± 1.3 ^{cdefgh}	46.6 ± 1.6 ^{abcd}
14	AL 1974	62.1 ± 1.1 ^{cdef}	3.54 ± 0.05 ^{ab}	21.0 ± 1.1 ^{wx}	24.5 ± 0.1 ^{fghij}	47.5 ± 2.5 ^{abcd}
15	AL 1976	91.1 ± 1.3 ^{ab}	4.11 ± 0.13 ^{ab}	34.8 ± 0.1 ^{hijklm}	34.9 ± 0.5 ^{abcd}	39.1 ± 2.5 ^{bcdefghi}
16	AL 1992	72.3 ± 4.9 ^{abc}	3.58 ± 0.10 ^{ab}	20.3 ± 0.3 ^x	41.1 ± 0.9 ^a	47.5 ± 0.8 ^{abcd}
17	AL 2000	39.4 ± 4.4 ^{fghijklmnop}	3.35 ± 0.14 ^b	34.5 ± 0.4 ^{ijklmn}	30.7 ± 2.3 ^{bcdefg}	42.5 ± 0.8 ^{abcdef}
18	AL 2009	49.7 ± 0.5 ^{cdefghijklm}	3.30 ± 0.19 ^b	34.5 ± 0.7 ^{ijklmn}	35.7 ± 0.6 ^{abc}	14.1 ± 0.8 ^{rs}
19	AL 2016	29.6 ± 3.2 ^{lmnop}	3.35 ± 0.05 ^b	28.8 ± 0.5 ^{pqrst}	35.4 ± 0.3 ^{abc}	18.3 ± 1.6 ^{pqrs}
20	AL 2019	18.2 ± 1.0 ^p	3.16 ± 0.05 ^b	22.7 ± 0.4 ^{vw}	36.45 ± 1.00 ^{ab}	20.8 ± 0.8 ^{opqrs}
21	AL 2021	24.6 ± 2.2 ^{op}	3.39 ± 0.09 ^b	25.3 ± 1.7 ^{tuv}	22.74 ± 2.00 ^{ghijk}	25.8 ± 0.8 ^{klmnopq}
22	AL 2024	46.1 ± 0.3 ^{efghijklmno}	3.54 ± 0.55 ^{ab}	41.1 ± 0.4 ^{efg}	27.59 ± 0.83 ^{cdefgh}	25.8 ± 0.8 ^{klmnopq}
23	AL 2025	39.8 ± 1.7 ^{fghijklmnop}	3.93 ± 0.29 ^{ab}	27.0 ± 0.3 ^{stu}	33.6 ± 1.8 ^{abcde}	25.0 ± 1.6 ^{lmnopqr}
24	AL 2028	53.6 ± 8.0 ^{cdefghij}	3.89 ± 0.10 ^{ab}	28.4 ± 0.7 ^{pqrst}	28.2 ± 0.1 ^{bcdefgh}	32.5 ± 2.5 ^{fghijklmn}
25	AL 2046	65.9 ± 0.6 ^{cde}	4.28 ± 0.03 ^{ab}	36.0 ± 0.2 ^{hijk}	28.5 ± 1.5 ^{bcdefgh}	13.3 ± 1.6 ^s
26	AL 2049	48.4 ± 3.9 ^{defghijklmno}	4.01 ± 0.10 ^{ab}	42.4 ± 0.9 ^{de}	28.9 ± 0.8 ^{gbcdefgh}	27.5 ± 2.5 ^{ijklmnop}
27	AL 2060	52.5 ± 7.3 ^{cdefghijkl}	3.92 ± 0.05 ^{ab}	37.2 ± 0.9 ^{gh}	27.0 ± 1.3 ^{defgh}	34.1 ± 2.5 ^{efghijkl}
28	AL 2061	41.5 ± 0.5 ^{fghijklmnop}	3.75 ± 0.04 ^{ab}	34.6 ± 0.8 ^{ijklmn}	29.2 ± 0.8 ^{gbcdefgh}	25.0 ± 1.6 ^{lmnopqr}
29	AL 2062	53.4 ± 3.1 ^{cdefghijk}	3.51 ± 0.16 ^{ab}	29.5 ± 0.5 ^{opqrs}	29.9 ± 1.5 ^{bcdefgh}	36.8 ± 0.8 ^{defghijk}
30	AL 2063	39.0 ± 1.7 ^{fghijklmnop}	3.86 ± 0.10 ^{ab}	35.8 ± 0.6 ^{ijkl}	26.0 ± 1.3 ^{efghi}	34.4 ± 0.2 ^{efghijkl}
31	AL 2067	58.2 ± 6.2 ^{cdefghi}	3.85 ± 0.09 ^{ab}	38.4 ± 0.6 ^{fghi}	35.6 ± 0.8 ^{abc}	39.3 ± 3.2 ^{bcdefghi}
32	AL 2071	37.8 ± 0.5 ^{ghijklmnop}	4.42 ± 0.72 ^{ab}	30.4 ± 1.3 ^{opqrs}	29.7 ± 0.6 ^{bcdefgh}	48.3 ± 2.4 ^{abc}
33	AL 2078	50.0 ± 9.4 ^{cdefghijklm}	3.56 ± 0.19 ^{ab}	39.7 ± 0.6 ^{efgh}	32.2 ± 0.8 ^{gbcdef}	40.1 ± 2.4 ^{bcdefgh}
34	AL 2086	48.9 ± 2.3 ^{cdefghijklm}	3.79 ± 0.10 ^{ab}	37.6 ± 1.2 ^{gh}	28.7 ± 1.3 ^{bcdefgh}	42.6 ± 1.6 ^{abcdef}
35	AL 2087	54.0 ± 5.9 ^{cdefghij}	3.63 ± 0.16 ^{ab}	34.7 ± 0.7 ^{ijklmn}	17.2 ± 2.3 ^{ijklm}	41.8 ± 2.4 ^{abcdef}
36	AL 2088	59.8 ± 6.1 ^{cdefgh}	4.11 ± 0.11 ^{ab}	30.9 ± 0.5 ^{opqrs}	17.6 ± 0.7 ^{ijklm}	52.4 ± 1.6 ^a
37	AL 2090	29.4 ± 2.6 ^{lmnop}	3.77 ± 0.10 ^{ab}	27.4 ± 0.5 ^{rstu}	21.9 ± 2.0 ^{hijk}	45.0 ± 0.8 ^{abcde}
38	AL 2091	25.1 ± 0.9 ^{nop}	3.76 ± 0.01 ^{ab}	22.0 ± 0.1 ^{vw}	27.7 ± 1.2 ^{cdefgh}	37.7 ± 1.6 ^{cdefghij}
39	AL 2092	29.8 ± 3.1 ^{klmnop}	3.70 ± 0.17 ^{ab}	31.0 ± 1.0 ^{nopqr}	18.0 ± 2.1 ^{ijkl}	40.9 ± 0.3 ^{bcdefg}
40	AL 2094	36.5 ± 0.7 ^{hijklmnop}	3.74 ± 0.12 ^{ab}	35.1 ± 0.1 ^{hijklm}	22.1 ± 0.9 ^{hijk}	28.6 ± 0.8 ^{ijklmnop}
41	AL 2095	35.0 ± 1.9 ^{ijklmnop}	3.48 ± 0.02 ^{ab}	32.5 ± 0.7 ^{klmno}	35.9 ± 2.0 ^{ab}	32.7 ± 1.6 ^{fghijklmn}
42	AL 2096	31.5 ± 1.8 ^{ijklmnop}	3.85 ± 0.30 ^{ab}	37.4 ± 0.7 ^{gh}	16.8 ± 2.4 ^{ijklmn}	27.8 ± 1.6 ^{ijklmnop}
43	AL 2098	43.0 ± 5.3 ^{efghijklmno}	3.79 ± 0.02 ^{ab}	32.5 ± 0.7 ^{klmno}	15.8 ± 1.2 ^{klmn}	35.2 ± 0.8 ^{efghijkl}
44	AL 2101	24.3 ± 1.3 ^{op}	3.54 ± 0.11 ^{ab}	28.4 ± 1.3 ^{qrst}	31.9 ± 2.3 ^{bcdef}	29.5 ± 1.6 ^{hijklmnop}
45	AL 2102	27.0 ± 4.6 ^{mnop}	3.41 ± 0.04 ^b	32.4 ± 0.1 ^{lmnop}	16.9 ± 2.6 ^{ijklm}	28.6 ± 2.4 ^{ijklmnop}

Table 4 continued

S. No	Genotypes	DPPH	FRAP	Reducing power	Hydroxyl radical scavenging activity	Superoxide anion radical scavenging activity
46	AL 2108	32.9 ± 0.2 ^{ijklmnop}	3.25 ± 0.10 ^b	34.7 ± 1.0 ^{hijklm}	15.1 ± 1.4 ^{klmno}	22.1 ± 0.8 ^{nopqrs}
47	AL 2122	30.7 ± 2.4 ^{ijklmnop}	4.26 ± 0.07 ^{ab}	37.1 ± 1.2 ^{hij}	8.7 ± 0.6 ^{nop}	21.3 ± 1.6 ^{nopqrs}
48	<i>C. scarabaeoides</i> ICP15683/W15	93.7 ± 0.1 ^a	4.98 ± 0.04 ^a	98.6 ± 0.6 ^a	9.3 ± 2.1 ^{mnop}	15.5 ± 0.8 ^{qrs}
49	<i>C. scarabaeoides</i> ICP15701/W15	57.0 ± 6.8 ^{cdefghi}	4.07 ± 0.05 ^{ab}	69.3 ± 0.2 ^c	4.8 ± 0.4 ^p	24.5 ± 1.6 ^{lmnopqrs}
50	<i>C. scarabaeoides</i> ICP15703/W15	60.9 ± 1.0 ^{cdefg}	4.73 ± 0.02 ^a	76.1 ± 0.2 ^b	3.2 ± 0.1 ^p	22.9 ± 1.6 ^{lmnopqrs}

Values are mean ± SD of triplicates and values with different letters in the same column are significantly different ($p < 0.05$)

Fe³⁺/ferricyanide complex to the ferrous form which is monitored by measuring the formation of Perl's Prussian blue colour at a wavelength of 700 nm. Higher the reducing power, higher is the antioxidant activity. The reducing power in pigeonpea genotypes ranged from 20.38 mg/100 g (AL 1992) to 98.69 mg/100 g (*C. scarabaeoides* ICP15683/W15) as depicted in Table 4. The genotypes with content from 20.38 mg/100 g to 46.48 mg/100 g were considered to have the low range of reducing power and from 72.58 mg/100 g to 98.69 mg/100 g have the high range of reducing power. Only the three wild type species possessed the high range of reducing power and the other genotypes have the lowest and the medium reducing power. In pigeonpea, the reducing power was reported to be 1.63 mg/g by Sreeramulu et al. (2009).

Hydroxyl radical scavenging activity

Scavenging of hydroxyl radical is an important antioxidant activity because hydroxyl radical is the most potent radical among the oxygen radicals as it severely damages adjacent biomolecules such as proteins, DNA, nucleic acid and almost every biological molecule it touches. This damage results in aging, cancer and several other diseases. It also causes initiation of lipid oxidation process. Therefore, the removal of hydroxyl radical is one of the most effective defences of a living body against various diseases (Ilesanmi et al. 2014). Hydroxyl radical is a strong oxidant and can abstract a hydrogen atom from any carbon-hydrogen bond and oxidize the compound. For example- linoleic acids are mainly located in glycerolipids and phospholipids of cell membranes; therefore, cell membranes are easily oxidized and lose their functionality during the aging process.

Hydroxyl radical scavenging rate of pigeonpea genotypes ranged from 3.27 to 41.12% (Table 4). Seven genotypes had lower rate of scavenging activity ranging from 3.27 to 15.88%; twenty one genotypes possessed medium

rate of scavenging activity ranging from 15.88 to 28.48% and twenty two genotypes possessed higher rate of scavenging activity ranging from 28.48 to 41.12%. Overall the wild type cultivars had the lowest scavenging activity.

Superoxide anion radical scavenging activity

Superoxide anions are a precursor to active free radicals that have the potential to react with biological macromolecules and induce tissue damage. It initiates lipid peroxidation and plays an important role in the formation of other reactive oxygen species such as hydroxyl radical, which induce oxidative damage in lipids, proteins and DNA. The scavenging activity of pigeonpea extract to inhibit superoxide anion radical ranged from 13.33 to 52.45% with an average value of 32.21% (Table 4). Sixteen genotypes had lower superoxide radical scavenging activity ranging from 13.33 to 26.37% and thirteen genotypes showed high scavenging activity ranging from 39.41 to 52.45%. Lowest activity was observed in AL 2046 and highest in AL 2088. Other genotypes possessed medium scavenging activity (22.77–42.33%).

Principal component analysis (PCA) is a useful statistical technique which is used to find out interrelationships between the different variables. Cluster analysis was done to find out the similarity between different pigeonpea genotypes on the basis of their nutritional, antinutritional and antioxidant potential by using the software Minitab. The genotypes were grouped into eight clusters Fig. 1a. Genotypes in the same cluster showed similarity among themselves and they were distinct from the genotypes in other clusters. Among the eight clusters, cluster 6 was largest with 15 genotypes followed by cluster 2 with 13 genotypes. The wild species formed a different major cluster with two sub clusters. One sub cluster contained only *C. scarabaeoides* ICP15683/W15 and the other sub-cluster contained *C. scarabaeoides* ICP1701/W15 and *C. scarabaeoides* ICP15703/W15. This showed that among

the three wild species two are similar and one is distinct. Cluster 1 had six genotypes namely AL 15, AL 2094, AL 2096, AL 2102, AL 2108 and AL 2122. And cluster 3 had seven genotypes namely AL 201, AL 1931, AL 1932, AL 1960, AL 2046, AL 2049, AL 2060.

The loading plot and biplot of nutritional, antinutritional and antioxidant potential of pigeonpea genotypes is shown Fig. 1b. The first and the second component described 26.9 and 19.1% variance respectively. Together the first two components represent 46% of total variability. The parameters V9 (Trypsin inhibitor activity), V16 (Superoxide anion radical scavenging activity), V4 (Total soluble Proteins), V2 (Starch), V5 (Bound fructose), V10 (Phytic acid), V1 (Total soluble sugars), V11 (Saponin) and V15 (Hydroxyl radical scavenging activity) were found to be occupied on the right side of the plot whereas the parameters V8 (Flavonol), V7 (Tannin), V14 (Reducing power), V13 (FRAP), V12 (DPPH) and V6 (Total phenols) were found to be occupied on the left side of the plot. This suggested that trypsin inhibitor

activity, superoxide anion radical scavenging activity, total soluble proteins, starch, bound fructose, phytic acid, total soluble sugars, saponin and hydroxyl radical scavenging activity had correlation among themselves. Flavonol was on left side of the corner. Total phenols were found to be in the middle portion of the plot. Based on the mathematical rule, uncorrelated variables occur at right angles to one another because the cosine of angles between them is zero that means not correlated. Similarly the variables at zero degree are correlated as the cosine of angle between them is 1. Flavonol showed negative correlation with hydroxyl radical scavenging activity, starch, bound fructose, phytic acid, total soluble sugars and saponin. Tannin, reducing power, FRAP, DPPH, total phenols were found to have negative correlation with total soluble proteins, superoxide anion radical scavenging activity, trypsin inhibitor activity and hydroxyl radical scavenging activity.

Total soluble sugars (V1) showed a positive correlation with starch (V2) ($r = 0.494$). DPPH has positive

Fig. 1 a Dendrogram of pigeonpea genotypes based on nutritional, antinutritional and antioxidant potential, b loading plot and biplot of nutritional, antinutritional and antioxidant potential of pigeonpea genotypes

correlation with FRAP ($r = 0.478$). Total phenols showed positive correlation with DPPH (0.578), FRAP (0.244) and reducing power (0.343). Sowndhararajan and Kang (2013) observed that content of total phenolics showed a good correlation with most of the antioxidant assays such as FRAP ($R^2 = 0.828$), DPPH radical scavenging activity ($R^2 = 0.900$) and hydroxyl radical scavenging activity ($R^2 = 0.970$). Some authors have reported excellent linear correlations between antioxidant activity tests and total phenolic contents (Sultana et al. 2007). Xu and Chang (2007) reported that correlation analysis between antioxidant activities of all legumes showed significant linear correlation between DPPH and FRAP ($r = 0.96$), total phenolic content (TPC) and oxygen radical absorbing capacity (ORAC) ($r = 0.75$) and FRAP and TPC ($r = 0.72$) and TPC and DPPH ($r = 0.65$). The information about quality traits in pigeonpea genotypes can be utilized in breeding programme for the incorporation of desirable traits into elite breeding lines to develop high yielding cultivars coupled with better nutritional quality traits after evaluation at multilocal trials for 2–3 years.

Conclusion

Fifty pigeonpea genotypes consisting of three recommended cultivars, forty-four advance breeding and three accessions of wild species were characterized for nutritional, antinutritional and antioxidant potential by using equal class distribution method into low, medium and high groups. AL 201 had high carbohydrate content, medium protein, high total phenol, hydroxyl radical and superoxide anion radical scavenging activity. AL 2000, AL 2009 and AL 2046 had high amount of total soluble proteins, medium starch, medium content of antinutritional factors and good antioxidant potential. AL 1960 had high carbohydrate and protein content; medium level of tannin, trypsin inhibitor and saponin; high level of total phenols, DPPH, hydroxyl and superoxide anion radical scavenging activity. AL 2087 had high starch content, medium protein, lower antinutritional factors except trypsin inhibitor. AL 1931, AL 1932, AL 1960, AL 2046, AL 2049, AL 2060 had good nutritional value as protein content in them ranged from 20 to 23%, starch content 42–52%. Wild species *C. scarabaeoides* ICP15683/W15 had lower amount of carbohydrates and proteins, lower antinutritional traits and high antioxidant potential due to high total phenols, DPPH, FRAP and reducing power. The diversity was observed in genotypes with all the traits.

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