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Protein Content and Seed Trait Analysis in a Subset of the USDA, ARS, PGRCU Cowpea [*Vigna unguiculata* (L.) Walp.] Core Collection

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ABSTRACT

Cowpea [*Vigna unguiculata* (L.) Walp] is used as a vegetable and feedcrop worldwide, but knowledge is limited regarding the variability and correlations for protein and seed traits within a cowpea core collection. We analyzed protein content and seed trait data from the cowpea core collection representing 111 accessions. The accession PI 354729 produced the significantly highest protein content of 25.527%. Eight Indian cowpea accessions produced significantly high protein content averaging 24.423%. Protein content was significantly correlated with seed pattern (r^2 = 0.274***), seed texture (r^2 = 0.346***), seed coat color (r^2 = 0.136*) and seed pattern significantly correlated with seed texture (r^2 = 0.197**). However, protein content showed a significant negative correlation (r^2 =-0.333***) with 100 seed weight. One hundred seed weight (gm) showed significantly negative correlations with seed pattern (r^2 = -0.191**) and seed texture (r^2 = -0.265***). Seed coat color showed a significant negative correlation with seed pattern (r^2 = -0.14**). Based on these results, potential cowpea cultivars could be developed with improved protein content and morphological seed traits.

Key words: Protein, Cowpea, Seed, Traits, *Vigna unguiculata.*

INTRODUCTION

Cowpea [*Vigna unguiculata* (L.) Walp.] is an annual legume and is also known as southern pea, blackeye pea and crowder pea. The plant tolerates hot, dry and flooding conditions, infertile soil and acid stress (Fery, 2002). Cowpea originated in Africa and is cultivated in Africa, Latin America, Southeast Asia and the southern United States for use as a vegetable and animal feed with additional markets for fresh, canned, frozen and dry-pack products (Fery, 2002). Africa is the major producer of cowpea and farmers there grow seeds, leaves and fruit for use as vegetables as well as plant material for livestock feed (Fery, 2002). Growth habits for cowpea range from erect, semi-erect, prostrate, or climbing with indeterminate and determinate types. Pea yields range from 1,000 to 2,000 lbs. per acre, however the green pods yield 2,500 to 4,000 lbs. per acre Brandenberger *et al.* (2007). Seed coat textures can be smooth or rough and seed coat colors can be white, tan, brown, black, grey, or red. Cowpea seeds also exhibit eye, solid, speckled, holstein, variable, saddle-eye and mottled patterns with pattern colors consisting of variable, black, red, brown and grey. The USDA, ARS, Plant Genetic Resources Conservation Unit (PGRCU) curates a cowpea (*V. unguiculata*) core collection representing 117 accessions. Cowpea protein consist of lysine Iqbal *et al.* (2006). Cowpea protein is digested easily by humans and has a good nitrogen balance Rangel *et al.* (2004). Water-soluble fraction proteins in cowpea seeds have been found to range from 31.2 to 35.5% Alghamdi *et al.* (2019). Several additional scientists including Weng *et al.*, (2019); Itatat *et al.*,(2013); and Ubini *et al.*, (2016) found cowpea protein content ranging from 22.8 to 28.9%, 20.57 to 24.95% and 15.06 to 38.5%, respectively.

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The average grain protein percent in cowpea has been found to be 22.8% in India Jukanti *et al.* (2017). Chavan *et al.* (2012) reported that adding 40 kg Zn/ha resulted in a protein content of 23.59% in cowpea. Mungbean [*Vigna radiata* (L.) Wilczek] which is a close relative of cowpea has been shown to contain 14.31 to 36.56% and 23.4 to 26.1% protein Das *et al.* (2015) and Singh, 2001, respectively. Cowpea protein has been shown to have antioxidant properties, lowers cholesterol and aids in reducing hypercholesterolemia from high-saturated fatty acid and cholesterol diets Frota *et al.* (2008). A core collection is a grouping of the most diverse accessions within a specific species. This core collection is an excellent source of cowpea plant material to study the variability of crude protein content and its correlation with other seed traits. Our objectives in this study were: 1) to identify variability for cowpea seed protein content and100 seed weight and 2) to evaluate seed coat color, seed pattern

color, seed pattern and seed texture from these cold stored accessions and 3) to determine correlations between these traits.

MATERIALS AND METHODS

This experiment consisted of a diverse core collection representing 111 cowpea accessions originating from Australia, Brazil, Burundi, Colombia, Ethiopia, India, Indonesia, Mexico, Mozambique, Nigeria, Turkey, Zaire and Zimbabwe during 2016 (Table 1). Six cowpea accessions in the core subset did not contain enough seeds for evaluations. These accessions were stored at 4°C located in the PGRCU cold storage.We analyzed cowpea seeds from each accession originating from cold storage. Seed coat color, seed pattern color, seed pattern and seed texture based on Crop Germplasm Committee derived descriptors were visually observed and recorded. Seed coat and seed pattern color descriptors used by the National Plant Germplasm System were assigned numerical codes: 1 = black, 2 = brown, 3 = grey, 4 = mixed (two or more colors), 5 = red, 6 = variable, 7 = white and 8 = tan. Seed patterns were assigned numerical codes: 1, 2, 3, 4, 5, 6, 7 and 8 representing eye, solid,speckled, mottled, holstein, saddle-eye, variable and mixed colors, respectively. Seed textures were assigned numerical codes: 1, 2, 3 and 4 representing rough, smooth, wrinkled and mixed, respectively (NPGS, 2019). The cowpea accessions originated from Mexico, Ethiopia, Australia, Brazil, India, Mozambique, Nigeria, Colombia, Indonesia, Turkey, Zimbabwe, Burundi, Zaire and the United States. One hundred seed weights (gm) were recorded prior to protein content analysis. Twenty-five to 50 cowpea seeds (depending on size) from each accession were ground to a powder in a coffee grinder and stored at -20°C until analysis. A small amount of powdered seed material (~225 mg) from each accession was wrapped in tin foil (50 x 50 mm, Art-No. 12.01-0033, Elementar). The wrapped seed powder was transferred into a small column and then pressed into a small pellet (10 mm diameter x 5 mm high). The seed protein content was measured using a Rapid N Exceed nitrogen analyzer (Elementar, Hanau, Germany/New Jersey, USA). We measured duplicate protein percentages for each accession. Sample pellets were combusted at 1000°C in a CO_2 carrier gas with an O_2 injection, resulting in a mixture of oxidized products and ash. We removed water, SO_2 and other combustion products using various absorbers. The Rapid N Exceed nitrogen analyzer reduced nitrogen oxides to molecular nitrogen and this was confirmed by a thermal conductivity detector. Total nitrogen content was expressed as a percentage of each samples seed weight and a protein factor of 5.6 was used to estimate crude protein percentages Mariotti *et al*. (2008).

Statistical analysis

We conducted an analysis of variance on the protein content and 100 seed weight (g) data. We used Tukey's multiple comparison procedure (SAS, 2012) to separate the protein content means. Protein content data was most important for this evaluation. All other seed trait data were not compared (using Tukey's multiple comparison procedure) because there were no within variation among accessions. However, correlations between protein content, 100 seed weight and morphological seed traits (using their assigned numerical codes) were determined using Pearson correlation coefficients.

RESULTS AND DISCUSSION

These accessions from a diverse cowpea core collection serves as a good source for the identification of protein content variation and correlations with important seed traits. Cowpea accessions showed variation for protein content and 100 seed weight (Table 1). Protein content ranged from 17.60 to 25.71% for all cowpea accessions. The ANOVA shows that there are significant differences (Probability< 0.0001) in most of the accessions evaluated for protein content (Table 1). The accession, PI 354729 originating from India with small seeds (100 seed weight of 6.15 gm), wrinkled white seeds consisting of a mixed pattern produced a significantly higher protein content of 25.527% [based on Tukey's minimum significant difference (MSD) = 0.5317] when compared to all other cowpea accessions. However, other researchers have reported similar protein content ranging from 20 to 27% (Henshaw, 2008), 21.3 to 26.9% Ajeigbe *et al.* (2008) and 23.7 to 27.4% Ravelombola *et al.* (2016). Henshaw, (2008) also reported an average seed protein content of 26.6% in three cowpea varieties and their seed coat colors ranged from cream to white with wrinkled seed textures, which were similar to our results. Higher protein content ranging from 27.9 to 38.5% have been reported Kochhar *et al.* (1988); Gupta *et al.* (2010); Jayathilake *et al.* (2018) and Ubini *et al.* (2016) in cowpea seeds. Several additional Indian cowpea accessions including PI 352979, PI 353040, PI 353041, PI 353129, PI 353289, PI 353362, PI 354518 and PI 354580 in this core collection produced the next significantly highest (MSD = 0.5317) protein content averaging 24.423% (Table 1). These accessions produced variable seed coat colors ranging from mixed to white. Seed pattern colors ranged from mixed, variable, brown or black. One hundred seed weights ranged from 7.08 to 15.57 gm and seed textures were primarily smooth. Of these, only PI 353041 and PI 353289 produced mixed seed textures.The accession, PI 527259 from Zimbabwe also produced a significantly high (MSD = 0.5317) mean protein content of 24.664%, medium size seeds (100 seed weight = 10.74 gm), mixed seed coat and pattern colors, variable seed pattern and smooth texture. The majority of the lower protein producing (<20% protein) cowpea accessions showed smooth seed textures and a range of seed coat and pattern colors (Table 1). Cowpea accessions originating from Indonesia, India, Australia, Mozambique and Mexico produced high mean protein content (23.19%, 22.45%, 21.94%, 21.92% and 21.76%, respectively) (Fig 1). A Tukey's multiple comparison procedure was not used for 100 seed weight, since there was no within variation among accessions. However, the cowpea accession producing the numerically

Table 1: Protein content, 100 seed weight and seed traits for the Cowpea accessions.

Acc. no.	Name	Country of origin	100 Sd. Weight (gm.)	% Protein	Seed coat color (no. code)	Seed pattern color (no. code)	Seed pattern (no. code)	Seed texture (no. code)
190191	TVu 1557	Mexico	13.96	21.758	Mix (4)	Mix (4)	Eye (1)	Smooth (2)
347639	283	Ethiopia	7.06	19.934	Variable (6)	Variable (6)	Solid (2)	Smooth (2)
349674	ALOOMBA	Australia	9.62	22.402	Black (1)	Black (1)	Solid (2)	Smooth (2)
349852	PYRAMID	Australia	8.98	23.684	Red (5)	Red (5)	Solid (2)	Smooth (2)
352762	CURUGINHA	Brazil	12.71	23.447	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
352765	19	Brazil	23.94	19.821	Mix (4)	Mix (4)	Eye (1)	Smooth (2)
352872	TVu 2724	India	5.22	22.180	Variable (6)	Variable(6)	Mix(8)	Smooth (2)
352873	TVu 2725	India	11.86	21.520	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
352881	I.C. 2809	India	12.86	19.543	Mix (4)	Mix (4)	Speckled (3)	Smooth (2)
352887	TVu 2738	India	10.14	23.703	Mix (4)	Mix (4)	Holstein (5)	Smooth (2)
352905	UCR 1340	India	12.69	22.895	Mix (4)	Mix (4)	Speckled (3)	Smooth (2)
352907	TVu 2757	India	9.81	22.256	Mix (4)	Mix (4)	Speckled (3)	Smooth (2)
352935	TVu 2785	India	9.79	22.737	Red (5)	Red (5)	Solid (2)	Smooth (2)
352936	TVu 2786	India	9.53	21.205	Brown (2)	Black (1)	Solid (2)	Smooth (2)
352941	I.C. 2946	India	7.36	19.563	Brown (2)	Brown (2)	Holstein (5)	Smooth (2)
352948	TVu 2798	India	10.28	23.913	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
352956	TVu 2806	India	8.43	22.331	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
352979	TVu 2828	India	11.12	24.744	Mix (4)	Black (1)	Solid (2)	Smooth (2)
353001	TVu 2848	India	8.74	23.396	Mix (4)	Black (1)	Solid (2)	Smooth (2)
353017	TVu 2864	India	6.96	22.566	-	-	-	-
353040	TVu 2886	India	15.57	24.188	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
353041	TVu 2887	India	10.14	24.501	White (7)	Brown (2)	Variable (7)	Mix (4)
353050	TVu 2896	India	13.16	21.445	Mix (4)	Black (1)	Speckled (3)	Smooth (2)
353055	TVu 2901	India	8.44	21.311	Mix (4)	Brown (2)	Solid (2)	Smooth (2)
353059	TVu 2905	India	8.77	23.541	Mix (4)	Mix (4)	Variable (7)	Mix (4)
353062	TVu 2908	India	9.69	22.932	Mix (4)	Variable (6)	Variable (7)	Smooth (2)
353066	TVu 2912	India	10.9	19.377	White (7)	Brown (2)	Solid (2)	Smooth (2)
353081	TVu 2926	India	11.42	23.611	White (7)	Brown (2)	Variable (7)	Smooth (2)
353087	TVu 2932	India	7.27	23.948	White (7)	Brown (2)	Solid (2)	Mix (4)
353121	TVu 2965	India	13.45	23.630	White (7)	Brown (2)	Solid (2)	Rough (1)
353125	TVu 2969	India	8.22	21.324	Mix (4)	Brown (2)	Variable (7)	Smooth (2)
353127	TVu 2971	India	6.53	22.486	White (7)	Brown (2)	Solid (2)	Smooth (2)
353129	TVu 2973	India	12.94	24.387	White (7)	Brown (2)	Solid (2)	Smooth (2)
353166	TVu 3007	India	8.06	23.309	White (7)	Black (1)	Solid (2)	Smooth (2)
353190	TVu 3030	India	9.71	23.978	Variable (6)	Brown (2)	Variable (7)	Smooth (2)
353199	TVu 3039	India	6.95	23.830	Tan (8)	Brown (2)	Solid (2)	Smooth (2)
353210	TVu 3049	India	9.52	23.206	White (7)	Brown (2)	Speckled (3)	Smooth (2)
353215	TVu 3054	India	7.07	21.603	White (7)	Brown (2)	Solid (2)	Smooth (2)
353236	TVu 3073	India	11.44	23.190	Black (1)	Black (1)	Variable (7)	Mix (4)
353265	TVu 3099	India	7.13	23.676	White (7)	Brown (2)	Solid (2)	Smooth (2)
353289	PLL 282	India	13.22	24.030	White (7)	Brown (2)	Solid (2)	Mix (4)
353316	TVu 3146	India	7.36	22.000	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
353319	TVu 3149	India	9.58	23.926	Mix (4)	Brown (2)	Solid (2)	Smooth (2)
353345	TVu 3175	India	11.05	21.798	Tan (8)	Brown (2)	Speckled (3)	Smooth (2)
353352	TVu 3182	India	10.79	20.794	Brown (2)	Mix (4)	Speckled (3)	Smooth (2)
353362	PLL 363	India	8.88	24.634	Mix (4)	Black (1)	Variable (7)	Smooth (2)
354429	TVu 1433	India	9.82	22.653	Mix (4)	Brown (2)	Saddle-eye (6)	Smooth (2)
354464	TVu 3246	India	9.73	21.792	Brown (2)	Black (1)	Mottled (4)	Smooth (2)
354466	P 636	India	13	21.739	Mix (4)	Mix (4)	Solid (2)	Smooth (2)

Table 1 Continue.....

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354469	P 640	India	11.72	22.989	Mix (4)	Brown (2)	Mottled (4)	Smooth (2)
354500	TVu 3279	India	8.78	23.571	Brown (2)	Variable (6)	Speckled (3)	Smooth (2)
354501	P 679	India	10.55	23.874	Mix (4)	Black (1)	Variable (7)	Smooth (2)
354518	TVu 3296	India	7.08	24.401	Mix (4)	Variable (6)	Variable (7)	Smooth (2)
354524	P 703	India	7.69	21.679	Variable (6)	Brown (2)	Variable (7)	Smooth (2)
354553	P 734	India	15.05	20.053	Brown (2)	Variable (6)	Variable (7)	Smooth (2)
354580	P 769	India	10.39	24.499	Mix (4)	Brown (2)	Solid (2)	Smooth (2)
354673	P 1127	India	11.35	22.988	Mix (4)	Black (1)	Variable (7)	Smooth (2)
354680	P 1139	India	10.22	21.881	Brown (2)	Brown (2)	Solid (2)	Smooth (2)
354687	P 1150	India	7.71	21.506	Brown (2)	Brown (2)	Mottled (4)	Smooth (2)
354708	P 1179	India	5.38	23.844	Brown (2)	-	Solid (2)	-
354715	TVu 3444	India	7.77	23.150	Mix (4)	Black (1)	Variable (7)	Smooth (2)
354729	P 1208	India	6.15	25.527	White (7)	Mix (4)	Mix (8)	Wrinkle (3)
354743	P 1224	India	13.67	21.526	Black (1)	Black (1)	Solid (2)	Smooth (2)
354766	P 1264	India	11.07	19.755	Brown (2)	Brown (2)	Solid (2)	Smooth (2)
354778	P 1279	India	11.06	21.811	Variable (6)	Mix (4)	Variable (7)	Smooth (2)
354782	P 1285	India	11.58	20.812	Mix (4)	Variable (6)	Variable (7)	Smooth (2)
354801	P 1308	India	10.35	23.896	Mix (4)	Brown (2)	Solid (2)	Mix (4)
354823	P 1337	India	7.19	21.486	Brown (2)	Black (1)	Speckled (3)	Smooth (2)
354827	P 1343	India	11.66	22.843	Variable (6)	Brown (2)	Variable (7)	Smooth (2)
354833	P 1351	India	16.26	18.688	Brown (2)	Variable (6)	Mottled (4)	Smooth (2)
354837	P 1356	India	12.44	19.984	Grey (3)	Black (1)	Speckled (3)	Smooth (2)
354838	P 1357	India	11.04	20.280	Brown (2)	Mix (4)	Variable (7)	Smooth (2)
354840	P 1359	India	9.46	20.938	Mix (4)	Variable (6)	Solid (2)	Smooth (2)
354845	P 1364	India	6.04	19.713	White (7)	Mix (4)	Mix (8)	Smooth (2)
354857	TVu 3552	India	6.01	22.844	Black (1)	-	Variable (7)	-
354864	P 1392	India	11.22	22.178	Black (1)	Black (1)	Solid (2)	Smooth (2)
354881	P 1421	India	13.23	22.286	Black (1)	Black (1)	Variable (7)	Smooth (2)
358307	38 a	Ethiopia	8.2	22.219	Variable (6)	Brown (2)	Solid (2)	Smooth (2)
358715	22786	Ethiopia	4.54	20.642	-	-	-	-
358716	22905	Ethiopia	5.97	23.914	Brown (2)	-	Solid (2)	-
367863	V. 67-05	India	11.42	22.061	Brown (2)	Mix (4)	Speckled (3)	Smooth (2)
367918	CALOONA	Australia	4.76	19.727	-	-	-	-
367919	IIAM 111	Brazil	13.14	20.166	Grey (3)	Variable (6)	Speckled (3)	Smooth (2)
367921	NAVAJA	Mozambique	7.58	20.914	Tan (8)	Brown (2)	Solid (2)	Smooth (2)
367927	DR. SAUNDERS	Mozambique	7.39	22.924	Red (5)	Black (1)	Solid (2)	Smooth (2)
382107	588/2	Nigeria	17.51	23.070	White (7)	Mix (4)	Solid (2)	Rough (1)
382121	H51-1	Nigeria	-	23.351	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
382124	H64-8	Nigeria	12.49	21.604	White (7)	Mix (4)	Solid (2)	Rough (1)
382128	H113-2-1	Nigeria	11.4	18.770	Variable (6)	Grey (3)	Solid (2)	Smooth (2)
382135	H144-4	Nigeria	11.5	18.110	Mix (4)	Mix (4)	Speckled (3)	Rough (1)
390421	W-C 966	Colombia	9	21.740	Mix (4)	Black (1)	Variable (7)	Smooth (2)
406290	IFH 27-8	Nigeria	17.57	18.478	White (7)	Mix (4)	Solid (2)	Rough (1)
487518	Bogor 1	Indonesia	10.84	23.187	Brown (2)	Variable (6)	Solid (2)	Smooth (2)
491193	Borulce	Turkey	12.04	19.449	White (7)	Variable (6)	Solid (2)	Smooth (2)
503326	280785-11	Turkey	-	19.692	White (7)	Black (1)	Solid (2)	Rough (1)
517910	ILCA 6783	Ethiopia	9.68	18.812	White (7)	Brown (2)	Solid (2)	Smooth (2)
527259	AMM 338	Zimbabwe	10.74	24.664	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
527263	AMM 563	Zimbabwe	12.07	21.372	Mix (4)	Brown (2)	Solid (2)	Smooth (2)
527267	AMM 617	Zimbabwe	12.2	21.805	Mix (4)	Mix (4)	Variable (7)	Smooth (2)
527270	AMM 665	Zimbabwe	20.02	19.716	Red (5)	Black (1)	Solid (2)	Smooth (2)
527272	AMM 682	Zimbabwe	13.84	17.701	Mix (4)	Brown (2)	Solid (2)	Smooth (2)

Table 1 Continue.....

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527277	AMM 777	Zimbabwe	22.11	17.631	Brown (2)	Brown (2)	Holstein (5)	Smooth (2)
527286	AMM 915	Zimbabwe	11.68	22.679	Red (5)	Black (1)	Solid (2)	Smooth (2)
527289	AMM 923	Zimbabwe	12.44	23.552	Variable (6)	Brown (2)	Solid (2)	Smooth (2)
527299	AMM 1083	Zimbabwe	12.66	23.153	Mix (4)	Variable (6)	Solid (2)	Smooth (2)
527302	AMM 1208	Zimbabwe	11.23	21.576	Mix (4)	Variable (6)	Solid (2)	Smooth (2)
527561	IZ 29	Burundi	11.72	19.968	Mix (4)	Variable (6)	Solid (2)	Smooth (2)
527563	IZ 37	Burundi	11.19	19.791	Mix (4)	Mix (4)	Solid (2)	Smooth (2)
527675	IZ 316	Zaire	11.77	20.878	Variable (6)	Variable (6)	Solid (2)	Smooth (2)
548784	CALIFORNIA BLACKEYE 46	United States	20.73	19.670	White (7)	Black (1)	Solid (2)	Rough (1)
599213	CALIFORNIA BLACKEYE 3	United States	18.73	21.415	White (7)	Black (1)	Solid (2)	Rough (1)
		Mean		21.98				
		Std. Dev.		1.76				
		ANOVA MS	24.33	6.23				
		ANOVA Pr > F		<0.0001				
		Tukey's Minimum Significance Difference		0.5317				

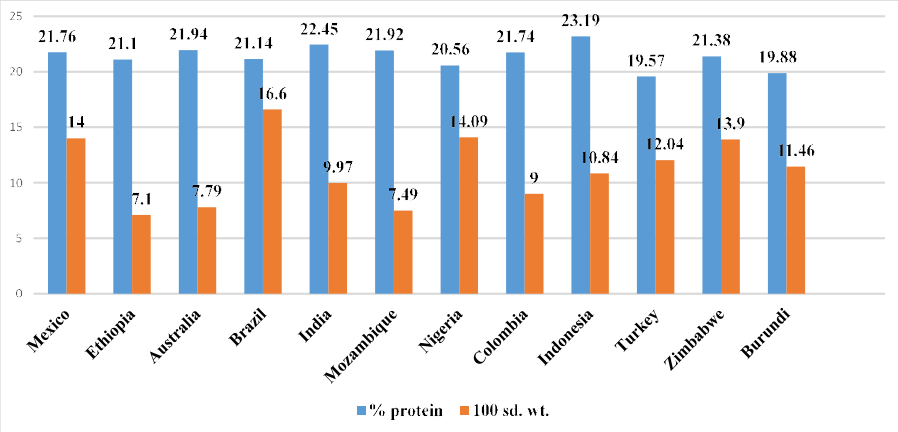


Fig 1: Protein content and 100 seed weight (gm) based on country of origin.

largest seeds was observed in PI 352765 from Brazil with a 100 seed weight of 23.94 gm (Table 1). Cowpea accessions producing numerically large seeds include PI 527270 (100 seed weight = 20.02 gm) and PI 527277 (100 seed weight = 22.11 gm) from Zimbabwe and the California Blackey 46 (100 seed weight = 20.73 gm) accession from the United States (Table 1). The remaining cowpea accessions produced numerically high 100 seed weights ranging from 4.54 gm to 18.73 gm. Accessions from Brazil and Nigeria produced the numerically largest mean 100 seed weights (16.6 gm and 14.09 gm, respectively) (Fig 1).

Raw black beans, red beans, white beans, lima beans, fava beans and chickpeas seeds have been reported to contain 21.6%, 22.53%, 23.36%, 6.84%, 7.92%, 20.47% protein, respectively (USDA, ARS, 2019). However, we found higher or similar protein content in our cowpea seeds.

We observed highly significant correlations among protein content and seed morphological traits (Table 2). We assigned numerical values to each morphological trait so that correlations could be determined. Protein content was significantly correlated with seed pattern (r^2 = 0.228**) and seed texture (r^2 = 0.344***), however protein content showed a significant negative correlation (r^2 = -0.333***) with 100 seed weight. The positive correlations between protein content, seed pattern and texture suggested that protein increases as the seed pattern and texture become solid and smooth, respectively. However, the negative correlation between protein content and 100 seed weight indicates that as 100 seed weight declines, protein content increases. One hundred seed weight showed significantly negative correlations with seed pattern (r^2 = -0.198**) and seed texture (r^2 = -0.261***). This is indicative of 100 seed weight increasing as seed pattern becomes non-solid and smooth seed texture. Seed coat color showed a significant negative correlation with seed pattern (r^2 = -0.137**) and seed pattern significantly correlated with seed texture (r^2 = 0.192**). This suggests that as seed coat color becomes darker, seed patterns become more non-solid. The positive correlation between seed pattern and seed texture suggests that as seed pattern becomes more solid, seed texture becomes smoother.

Table 2: Pearson correlation coefficients for protein content, 100 seed weight (g) and seed traits in the cowpea core collection.

	100 sd. wt.	Seed coat color	Seed pattern color	Seed pattern	Seed texture
% Protein	-0.333***	0.072	-0.131	0.228**	0.344***
100 sd. wt.		-0.021	0.021	-0.198**	-.261***
Seed coat color			-0.053	-0.137**	-0.078
Seed pattern color				0.113	-0.058
Seed pattern					0.192**

CONCLUSION

Protein content and seed morphological trait analysis of cowpea can identify and quantify accessions useful for human nutrition. We observed several significant correlations for protein content, 100 seed weight, seed coat color, seed pattern and seed texture in this study. Our results show that potential cowpea cultivars could be developed with improved protein content and morphological seed traits. However, these cowpea accessions require further field evaluations to verify if protein content in the seed remained at similar levels observed in this study. There is a need for field evaluations, since significant genotype, environment and genotype by environment interactions occurred among cowpea lines for protein.

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REFERENCES

Ajeigbe, H.A., Ihedioha, D., Chikoye, D. (2008). Variation in physico-chemical properties of seedof selected improved varieties of cowpea as it relates to industrial utilization of the crop. African Journal of Biotechnology. 7: 3642-3647.

Alghamdi, S.S., Khan, M.A., Migdadi, H.M., El-Harty, E.H., Afzal, M., Farooq, M. (2019). Biochemical and molecular characterization of cowpea landraces using seed storage proteins and SRAP marker patterns. Saudi Journal of Biological Sciences. 26: 74-82.

Brandenberger, L., Shrefler, J., Damicone, J., Rebek, E. (2007). Southern pea production. Oklahoma State University Cooperative Extension Service Fact Sheet HLA-6029.

Chavan, A.S., Khafi, M.R., Raj, A.D., Parmar, R.M. (2012). Effect of potassium and zinc on yield, protein content and uptake of micronutrients on cowpea [*Vigna unguiculata* (L.) Walp.]. Agricultural Science Digest. 32: 175-177.

Das, S., Dewanjee, A.M., Sarkar, K.K. (2015). Breeding mungbean [*Vigna radiate* (L.) Wilczek] for development of high yielding protein rich lines with low trypsin inhibitor. Legume Research-An International Journal. 3: 447-451.

Fery Richard L. (2002). New Opportunities in *Vigna.* In: *Trends in New Crops and New Uses.* [Janick J and Whipkey A (eds)] Alexandria,VA, USA: ASHS Press, pp. 424-428.

Frota, K.M.G., Mendonca, S., Saldiva, P.H.N., Cruz, R.J., Areas, J.A.G. (2008). Cholesterol-lowering properties of whole cowpea seed and its protein isolate in hamsters. Journal of Food Science. 73: H235-H240.

Gupta, P., Singh, R., Malhotra, S., Boora, K.S., Singal, H. R. (2010). Characterization of seed storage proteins in high protein genotypes of cowpea [*Vigna unguiculata* (L.) Walp.]. Physiology and Molecular Biology of Plants. 16: 53-58.

Henshaw,F.O.(2008). Varietal differences in physical characteristics and proximate composition of cowpea (*Vigna unguiculata*). World Journal of Agricultural Sciences. 4: 302-306.

Iqbal, A., Khalil, I.A., Ateeq, N., Khan, M.S. (2006). Nutritional quality of important food-legumes. Food Chemistry. 97: 331-335.

Itatat, S.L., Nworgu, E.C., Ikpe, E.N., Osakwe, J.A. (2013). Evaluation of the protein contents of selected cowpea [*Vigna unguiculata* (L.) Walp.] varieties for production in Port Harcourt. Acta Agronomica Nigeriana. 13: 71-75.

Jayathilake, C., Visvanathan, R., Deen, A., Bangamuwage, R., Jayawardana, B.C., Nammi, S., Jukanti, A.K., Dagla, H.R., Kalwani, D., Goswami, J.M., Upendra, R.K.K.,Bhatt, R.K. (2017). Grain protein estimation and SDS-PAGE profiling of six important arid legumes. Legume Research-An International Journal. 40: 485-490.

Liyanage. R. (2018). Cowpea: an overview on its nutritional facts and health benefits. Journal of the Science of Food and Agriculture. 98: 4793-4806.

Kochhar, N., Walker, A.F., Pike, D.J. (1988). Effect of variety on protein content, amino acid composition and trypsin inhibitor activity of cowpeas. Food Chemistry. 29: 65-78.

Mariotti, F., Tome, D., Mirand, P.P. (2008). Converting nitrogen into protein – beyond 6.25 and Jones' factors. Critical Reviews in Food Science and Nutrition. 48: 177-184.

National Plant Germplasm System. (2019). Germplasm Resources Information Network (GRIN). Database Management Unit (DBMU). National Plant Germplasm System, U.S. Department of Agriculture, Beltsville.

Rangel, A., Saraiva, K., Schwengber, P., Narciso, M.S., Domont, G.B., Ferreira, S.T., Pedrosa, C. (2004).Biological evaluation of a protein isolate from cowpea (*Vigna unguiculata*) seeds. Food Chemistry. 87: 491-499.

Ravelombola, W.S., Shi, A., Weng, Y., Motes, D., Chen, P., Srivastava, V., Wingfield, C. (2016). Evaluation of total seed protein content in eleven Arkansas cowpea [*Vigna unguiculata* (L.) Walp.] lines. American Journal of Plant Sciences. 7: 2288-2296.

SAS. (2012). Cary, NC: SAS Institute.

Singh, A.B. (2001). Influence of phenolic compounds on yield and protein quality of green gram [Vigna radiata (L.) Wilczek]. Legume Research-An International Journal. 24(4).

Ubini, A. Richard, Idegba, C.M., Isaac, I.E., Ileleji, Alfred, Bozeinghien, H. (2016). Seed protein content variation in cowpea genotypes. African Journal of Plant Breeding. 3: 143-147.

US Department of Agriculture, Agricultural Research Service. (2019). Food Data Central. fdc.nal.usda.gov.

Weng, Y., Qin, J., Eaton, S., Yang, Y., Ravelombola, W.S., Shi, A. (2019). Evaluation of seed protein content in USDA cowpea germplasm. Hort. Science. 54: 814-817.