African Journal of Biotechnology Vol. 10(16), pp. 3077-3086, 18 April, 2011
Available online at http://www.academicjournals.org/AJB
DOI: 10.5897/AJB10.2469
ISSN 1684–5315 © 2011 Academic Journals

Full Length Research Paper

Genetic analysis of seed proteins contents in cowpea (*Vigna unguiculata* L. Walp.)

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Accepted 18 March, 2011

In order to select cowpea genotypes with high food value, 10 varieties were genetically screened in Ngaoundéré (Cameroon) for seed crude protein and its soluble fractions contents. Five divergent lines were studied through a 5 x 5 half diallel cross mating. The genotypes presented a significant genetic variability for these parameters ($p < 0.05$). The globulins constituted the major seed protein fraction, followed by albumins. Diallel analysis demonstrated that, both additive and non-additive gene effects were responsible for the genetic variation of these traits. However, dominance variance was more important than additive variance for all traits. The model of over-dominance was most widespread, suggesting delayed selection to fairly good improvement. All these parameters were found highly inheritable ($h^2 = 0.68$ to 0.83). The parents differed significantly for their general combining ability (GCA) and the F_1 progenies showed specific combining ability (SCA). Dominant genes have positive effects for high levels of albumins, globulins and prolamins, while high percentage of seed protein and high glutenins content appeared to be associated with recessive genes. In the Guinea savannah zone, these results would help breeders to improve these biochemical traits in terms of initial parent selection and subsequent crossbred selection and breeding procedures.

Key words: *Vigna unguiculata*, seed crude protein content, soluble protein fractions, diallel analysis, genetic improvement, Guinea savannah zone.

INTRODUCTION

Proteins are major components of grain legumes and their nutritional and functional properties depend on the nature of soluble fractions (Mandal and Mandal, 2000; Vasconcelos et al., 2010). Cowpea (*Vigna unguiculata* L. Walp.), a widely adapted and nutritious grain legume, constitutes one of the main source of plant protein in northern Cameroon (Fotso et al., 1994; Noubissié et al., 2007). Dry seeds for human consumption are the principal product of the plant, but leaves, fresh peas, and fresh green pods are consumed by many poor people who do not have access to broadly based diet (Hall et al., 2003; Langyintuo et al., 2003). Cowpea has potential of becoming an industrial crop and widespread consumption of convenience foods containing significant amounts of cowpea substantially increased the demand of cowpea grain (Prinyawiwatkul et al., 1996; Ajeigbé et al., 2008). In west and central Africa, cowpea is processed into an array of traditional foods such as "*akara*", "*moin-moin*" and "*koki*" (Phillips et al., 1988; Singh et al., 2003). According to Hall et al. (2003), a significant market may be present for some new value-added-foods based on cowpea because some people have an allergic reaction to foods containing soybean proteins. Protein improvement in legumes was approached very little because breeding programs have produced cultivars primarily for high yield and correlations between yield and protein have generally been negative (Griffiths and Lawes, 1978; Mandal and Mandal, 2000; Giami, 2005). Variations in content among varieties were found for protein and other

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Table 1. Origin and description of cowpea genotypes.

Genotypes	Origin	Growth	Flower	Seed	Testa
BA (Bafia)	Local	Erect	Cream	Violet	Smooth
BR1	IRAD	Erect	White	White	Rough
CRSP (CRSP niébé)	IRAD/CRSP	Prostrate	White	White	Rough
HM (Hadiam Marva)	Local	Prostrate	White	White	Rough
HMO (Hadiam Mouchiche)	Local	Erect	White	White	Rough
HALG (Halagare Gongourdem)	Local	Prostrate	White	White	Rough
HALM (Halagare Memedem)	Local	Prostrate	White	White	Rough
Hoyo (Hoyo Hoyo)	Local	Prostrate	White	White	Rough
573 (IT97K-573-1-1)	IITA	Erect	White	White	Rough
NH (Niébé Hosseré)	Local	Prostrate	White	White	Rough

CRSP: Collaborative research support program; IITA: international institute of tropical agriculture; IRAD: institute of agricultural research for development (Maroua regional center).

physical, functional and chemical seed characteristics by Nielsen et al. (1993), Oluwatosin (1998), Giami (2005), Ajeigbé et al. (2008) and Vasconcelos et al. (2010). According to Fotso et al. (1994) and Ragab et al. (2004), knowledge on the genetics of storage proteins becomes a prerequisite in order to fully utilize the potential in improving the nutritional quality of the crop. Legume proteins are known to differ in digestibility and technological properties (Ajeigbé et al., 2008; Park et al., 2010; Vasconcelos et al., 2010). In cowpea, protein types comprise globulins (at least 16 protein bands), albumins (at least 20 protein bands), glutelins (21 protein bands) and prolamins (one protein band) (Ragab et al., 2004; Vasconcelos et al., 2010). Albumins play an essential role in seeds as enzymatic and metabolic proteins, such as lipoxygenase, protease inhibitors and lectins (Shutov et al., 2003; Park et al., 2010). Globulins composed of two major groups of protein on the basis of sedimentation coefficient - 11S fraction (legumin) and 7S fraction - play an important role as storage proteins and were mostly digested by proteases (Shutov et al., 2003; Coelho and Benedito, 2008; Park et al., 2010). Prolamins are storage protein found mainly in seeds of cereal grain with high proline and glutamine content (Shewry and Halford, 2002). Glutelins have poor lysine content in cowpea (Vasconcelos et al., 2010).

Proper understanding of genetic mechanisms involving the expression of these characters would help in planning effective breeding strategies. The purpose of this study was to evaluate in *V. unguiculata* the varietal differences and assess through diallel analysis the genetic control of seed protein content and its four soluble fractions so that the quality of the seed can be improved scientifically under the Sudano-Guinea conditions.

MATERIALS AND METHODS

Study site, biological material and experimental plots

The research was carried out during the year 2008 to 2009 at the University of Ngaoundéré campus (1113 m altitude, 7.28°N latitude and 13.34°E longitude), which is located at Dang, a village of Ngaoundéré in the Adamawa region, Cameroon. This region belongs to the Guinea savannah ecological zone. The climate is characterized by two seasons: a rainy season (April to October) and a dry season (November to March). The annual rainfall is about 1500 mm. The mean annual temperature is 22°C, while the annual humidity is about 70%. The soil is ferruginous type, developed on basalt, with 9.4 mg kg^{-1} organic matter, ratio C/N = 0.33 and pH 5.2. The texture of the brown reddish soil is predominantly made of clay.

Ten cowpea homozygous varieties including seven local landraces and three improved lines were used for the studies (Table 1). Seeds were obtained from the Institute of Agricultural Research for Development (IRAD Maroua and Foumbot stations). A preliminary field trial was conducted during the 2008 growing season to evaluate the genetic variability for seed protein content and its soluble fractions. The seeds of 10 entries were sown in a randomized complete block design (RCBD) with three replications. Sowing took place on May 06, 2008, on an experimental surface of 75 m^2 (10.5 m length x 7 m broad). Each plot unit consisted on one row of 3 m length x 0.35 m broad, spaced 30 cm apart. Three seeds of each variety were sown at an intra-row spacing of 25 cm and thinned to one per hill, 20 days after sowing (DAS). The plots were manually weeded at 20, 40 and at 60 DAS. At flowering stage, plots were sprayed with a standard insecticide formulation, cypermethrin + dimethoate at the rate of 30 g + 250 g a.i/L, to control pod borers and flower midges. A mineral fertiliser (7% N; 14% P_20_5; 7% K_20) was applied to the seedlings three weeks after planting at rate of 60 kg per ha. Strings were tied to the genotype to provide support. At maturity, harvesting was done at four-day intervals, when the pods were ready for picking and seeds were separated from dry pods.

Five genotypes (BA, BR1, HM, NH and 573) which were chosen based on their genetic variation for these traits were planted in pots from September to December 2008 for crossings. The five parental lines along with the 10 F_1 hybrids obtained were planted in field in RCBD with three replications during the rainy season 2009. Sowing took place on April 15, 2009, at the beginning of the rainy season on an experimental surface of 105 m^2 (10.5 m x 10 m). Plot unit size, spacing, weeding, treatments and harvesting were as previous described for variability study.

Production of cowpea flour and biochemical analysis

To determine the biochemical content of seeds, a random sample of 200 seeds per genotype was taken from a bulk sample of seeds

Table 2. Biochemical composition of cowpea genotypes in the previous study.

Genotype	Protein (% DM)	Albumin (% SP)	Globulin (% SP)	Prolamin (% SP)	Glutelin (% SP)
BA	27.52±1.87^{b}	30.02±5.05^{b}	55.44±9.72^{a}	2.70±0.91^{a}	11.84±3.26^{g}
BR1	23.62±1.79^{cd}	29.00±2.99^{b}	47.07±6.13^{cde}	1.58±0.22^{cd}	22.38±3.16^{d}
CRSP	29.91±1.53^{a}	23.61±4.96^{cde}	45.91±5.07^{def}	2.23±0.66^{b}	29.28±1.97^{b}
HM	31.78±1.26^{a}	29.68±2.68^{b}	43.70±1.96^{f}	1.02±0.22^{fg}	25.60±7.60^{c}
HMO	20.93±2.54^{e}	25.84±2,20^{c}	48.76±1.88^{bcd}	0,81±0,17^{g}	24.58±3.49^{c}
HALG	20.79±1.05^{e}	35.86±3.12^{a}	49.40±7.98^{bc}	0.73±0,13^{g}	14.00±2.14^{fg}
HALM	21.67±1.37^{de}	24.41±4.44^{cd}	54.32±5.12^{a}	1.45±0.17^{de}	19.82±3.49^{de}
Hoyo	21.39±0.94^{e}	30.23±1.43^{b}	51.34±2.52^{b}	1.17±0,43^{ef}	17.26±2.68^{ef}
NH	23.90±1.39^{c}	22.61±1.44^{de}	46.33±2.48^{de}	0.93±0.26^{fg}	30.13±2.18^{ab}
573	25.43±2.23^{c}	20.65±1,83^{e}	45.32±3,68^{ef}	1.82±0.48^{c}	32.21±3.68^{a}
Mean	22.15±5.28	27.20±3.12	48.76±2.71	1.44±0,72	22.71±3.51
LSD (0.05)	2.04	3.37	2.62	0.33	2.83

Means with the same subscript within the same column do not differ ($p > 0.05$); LSD (0.05): least significant difference at 5% level; % DM: percentage of dry matter basis; % SP: percentage on total soluble proteins basis.

from each replication. These samples were subdivided into four groups of 50 seeds that were used for the production of flour according to the method described by Phillips et al. (1988). The selected seeds were soaked in a ratio (1/3) (weight / volume) during 12 h and dried at 60°C for 24 h in a hot-air fan drier (Riviera and Bar, France). They were decoated and crushed in a hammer mill (Culatti, Polymix, Germany) through a 1500 µm sieve. The crude seed protein content was estimated by Lowry et al. (1951) procedures, after extraction of 0.5 g flour finely crushed to the SDS 1% in 0.1% NaOH under agitation for 24 h. Protein fractions were sequentially extracted based on their solubility in various solvents, as described by Osborne (1988). The proportion of each soluble fraction obtained was expressed on the basis of total soluble protein.

Genetic analysis

For the variability study, data of different parameters of the 10 pure lines were subjected to ANOVA using computer program STATGRAPHICS PLUS version 3.0 (Manugistics, 1997). The genotypic means were compared using least significant difference at 5% level of probability (LSD 5%).

The genetic analysis was done from a 5 x 5 half-diallel mating using DIALL microcomputer package (Ukai, 1989). The Griffing's (1956) method 2 (excluding reciprocal F_1 crosses), model 1 (fixed effects) was used to analyze the GCA of lines and the SCA of crosses. The analysis of variance by Walters and Morton (1978) was used for the study of genotypic effects. The genetic parameters were estimated as per Hayman (1954). With this approach, the components of variation were partitioned into the additive effects (*a*) and the dominance effect (*b* which is further sub-divided into *b1*, *b2* and *b3*). The simple additive-dominance model was tested by plotting the covariance values between the parents and their offspring in the rth array (Wr) against variance values of the rth array (Vr). Heritability in broad sense (h^2) was measured as the proportion of genetic variance of homozygous parents (σ^2_g) in the phenotypic variance between parents (σ^2_p), while heritability in narrow sense (h^2_n) was calculated as the proportion of additive variance (σ^2_A) in the phenotypic variance(σ^2_p) (Mather and Jinks, 1982).

RESULTS

Genotypic variation for seed protein and soluble fractions contents

Analysis of variance indicated significant differences between the parents for all the investigated traits (Table 2). The values of seed crude protein ranged between 20.79 to 31.78% (mean = 22.15%) and genotypes HM and CRSP were the best parents. Upon protein fractionation, it was verified that the salt-soluble fraction (globulins), which ranged from 43.70 to 55.44% of total soluble proteins (mean = 48.76%), was the major protein constituent, with BA and HALG as best parents. The second most abundant seed protein for the studied cowpea cultivars was the water-soluble fraction (albumins), which varied from 20.65 to 35.86% of total soluble proteins (mean = 27.20%). The albumins contents of landraces HALG and Hoyo were 50% higher than that of line 573. The rates of glutelins were relatively high particularly for lines 573 and NH, and varied from 11.84 to 32.21% (mean = 22.71%). Prolamins appeared as the minor soluble fractions in cowpea seed (range of 0.73 to 2.70%) but varieties BA and CRSP showed the highest percentages. Among the F_1 progenies, protein content varied from 22.23 to 32.67% (mean = 26.25%). The values of different protein soluble fractions in F_1 were 19.43 to 36.23% (mean = 28.27%) for albumins, 45.00 to 55.80% (mean = 48.09%) for globulins, 15.64 to 27.27% (mean = 21.72%) for glutelins and 0.84 to 3.23% (mean = 1.91%) for prolamins.

Genetic analysis of proteins contents

The ANOVA of the 5 x 5 half-diallel mating with Walters

Table 3. Computation of mean squares for ANOVA of 5 x 5 half diallel tables for cowpea seed protein and soluble fractions contents.

Source	Df	Mean square for biochemical parameter				
		Crude protein	Albumin	Globulin	Prolamin	Glutelin
Block	2	1.08	28.99	35.27	0.11	29.08
a	4	57.68**	259.31**	219.40**	0.52**	727.84**
b	10	59.91**	127.39**	355.51**	4.29**	302.16**
b_1	1	36.02**	225.28*	456.80**	3.40**	2419.89**
b_2	4	81.63**	72.81*	166.47**	3.80**	283.79**
b_3	5	47.31**	151.48**	486.49**	4.86**	-106.69**
Error	28	1.29	32.91	15.95	0.11	29.94

a = Additive effects of genes; b = dominant effects of genes; b_1 = mean dominance effects; b_2 = additional dominance deviation due to the parents, b_3 = residual dominance effects, * indicates significance at 5%, ** indicates significance at 1%.

Table 4. Genetic components estimates and heritability value for cowpea seed protein and soluble fractions based on a 5 x 5 half-diallel.

Genetic parameter	Crude protein	Albumin	Globulin	Prolamin	Glutelin
Average degree of dominance $(H_1/D)^{1/2}$	2.196	1.357	1.708	5.401	1.285
Proportion of dominant genes (kd)	0.65	0.63	0.49	0.60	0.69
Direction of dominance (h)	-2.68	+7.52	+10.80	+0.93	-24.86
Broad sense heritability (h^2)	0.74	0.76	0.83	0.71	0.68
Narrow sense heritability (h^2_n)	0.28	0.31	0.43	0.29	0.22
r (Pr, Wr+Vr)	$+0.77^{ns}$	-0.80*	-0.57^{ns}	-0.52^{ns}	$+0.70^{ns}$

r (Pr, Wr+Vr): Correlation between the degree of dominance of the parents (Wr+Vr) and the parental value (Pr); ns : not significant; * Significant at the 5% level.

and Morton's (1978) method showed the significance of genotypic effects and their components (Table 3). Both additive (*a*) and dominant effects (*b*) were all significant ($p < 0.05$). Within (*b*), the mean dominance effects (b_1), the additional dominance effects due to the parents (b_2) and the residual dominance effects were also significant ($p < 0.05$).

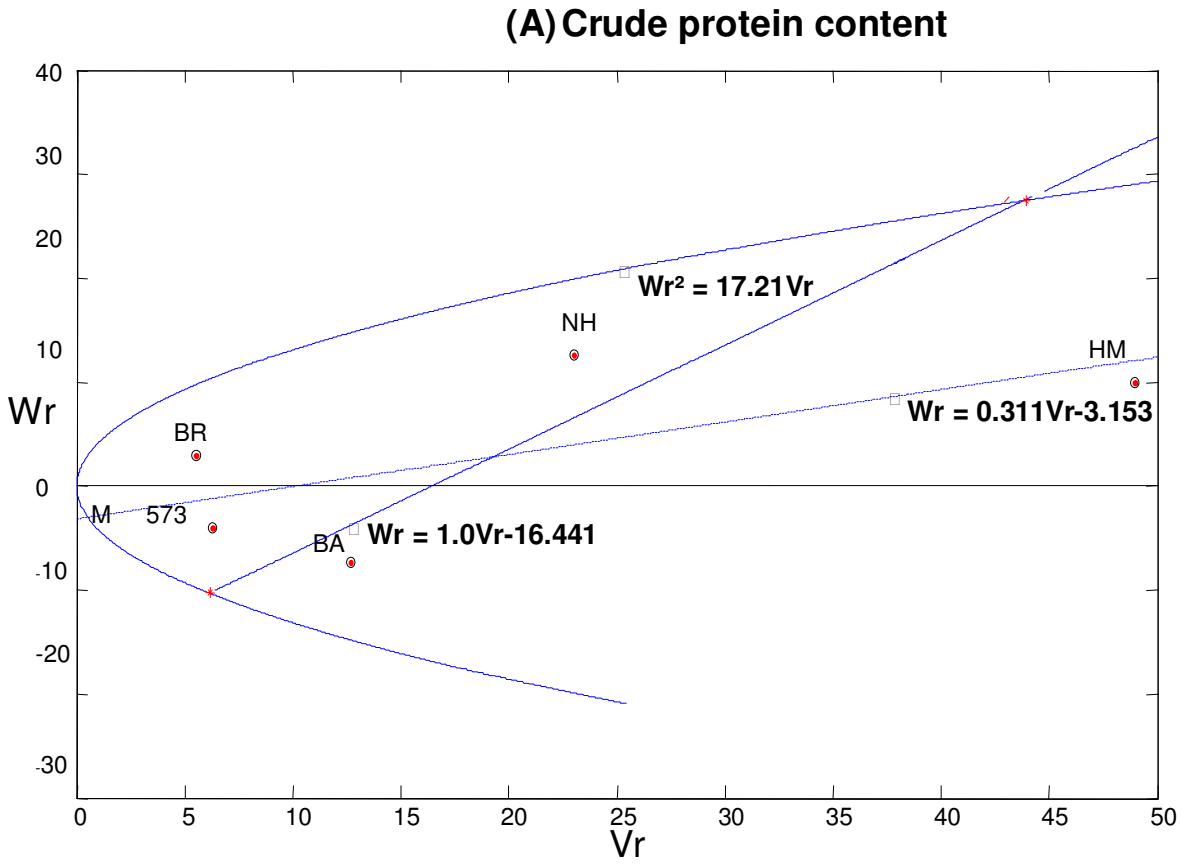
Broad and narrow sense heritability ranged from 0.68 to 0.76 and from 0.22 to 0.43, respectively (Table 4). Moderate to high proportion of dominant genes was noted for various traits (0.49 to 0.69) (Table 4). The direction of dominance was negative only for crude protein and glutelins contents (Table 4). The average degree of dominance was greater than one (1.28 to 1.40) and the component due to the dominance effects (H_1) was greater than that of additive effects (D) (Table 4).

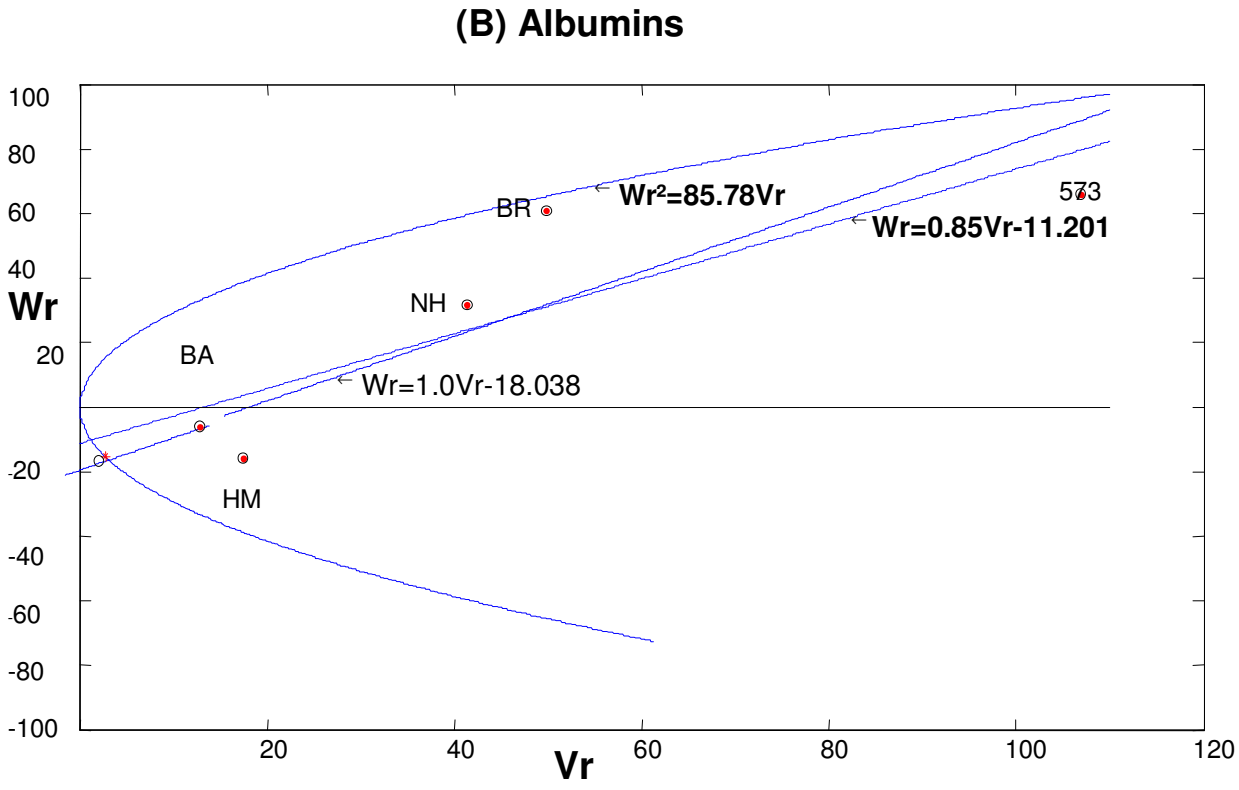
The Wr/Vr graphs (Figure 1) showed that, the scatter points were within the limiting parabola for all the traits. The regression line passed below the origin and the regression coefficient of Wr against Vr varied from 0.311 for protein content to 0.890 for prolamins. The array points for different traits showed that, some varieties were nearest to the origin (maximum dominant genes), while others were farthest from the origin (most recessive genes).

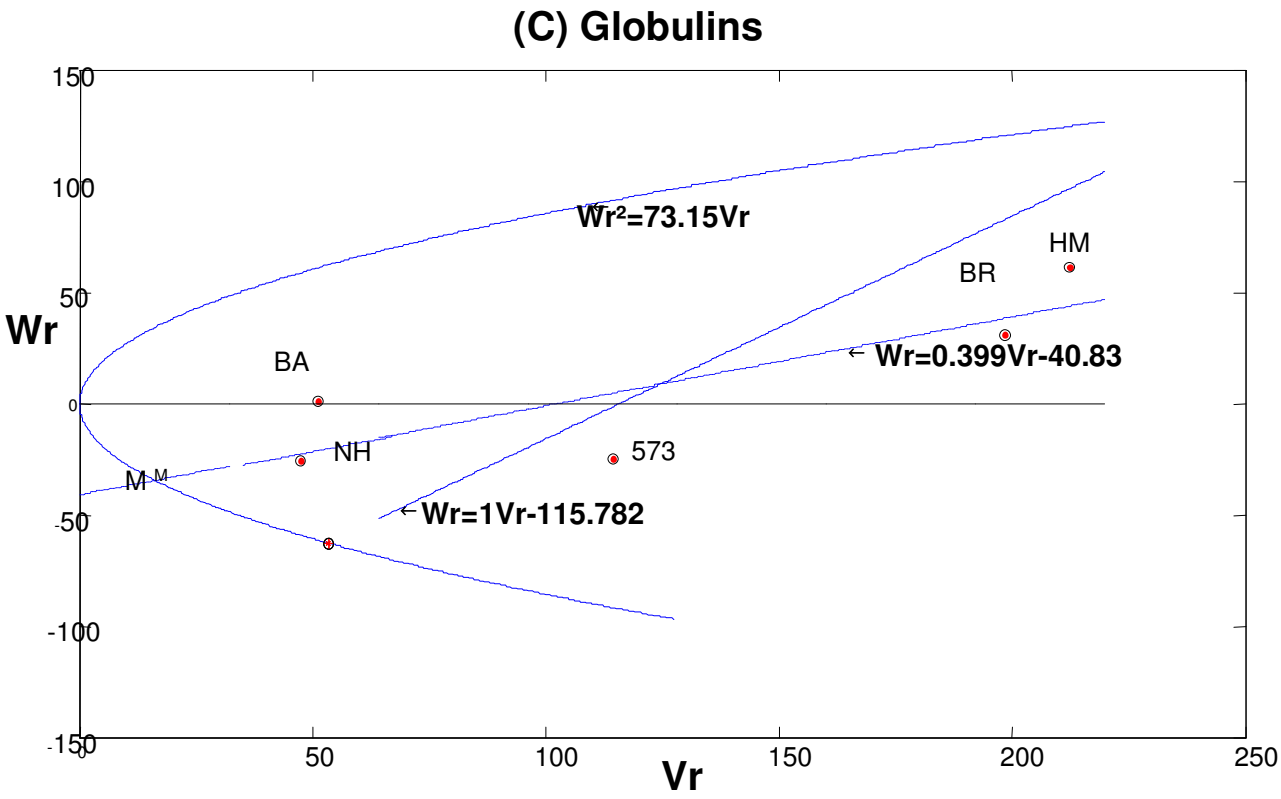
The correlation between parental values (Pr) and recessive factor (Wr+Vr) was positive for crude protein content (r = 0.77) and gutelins (r = 0.70) but negative for albumins (r = -0.80), globulins (r = -0.57) and prolamins (r = -0.52) (Table 4).

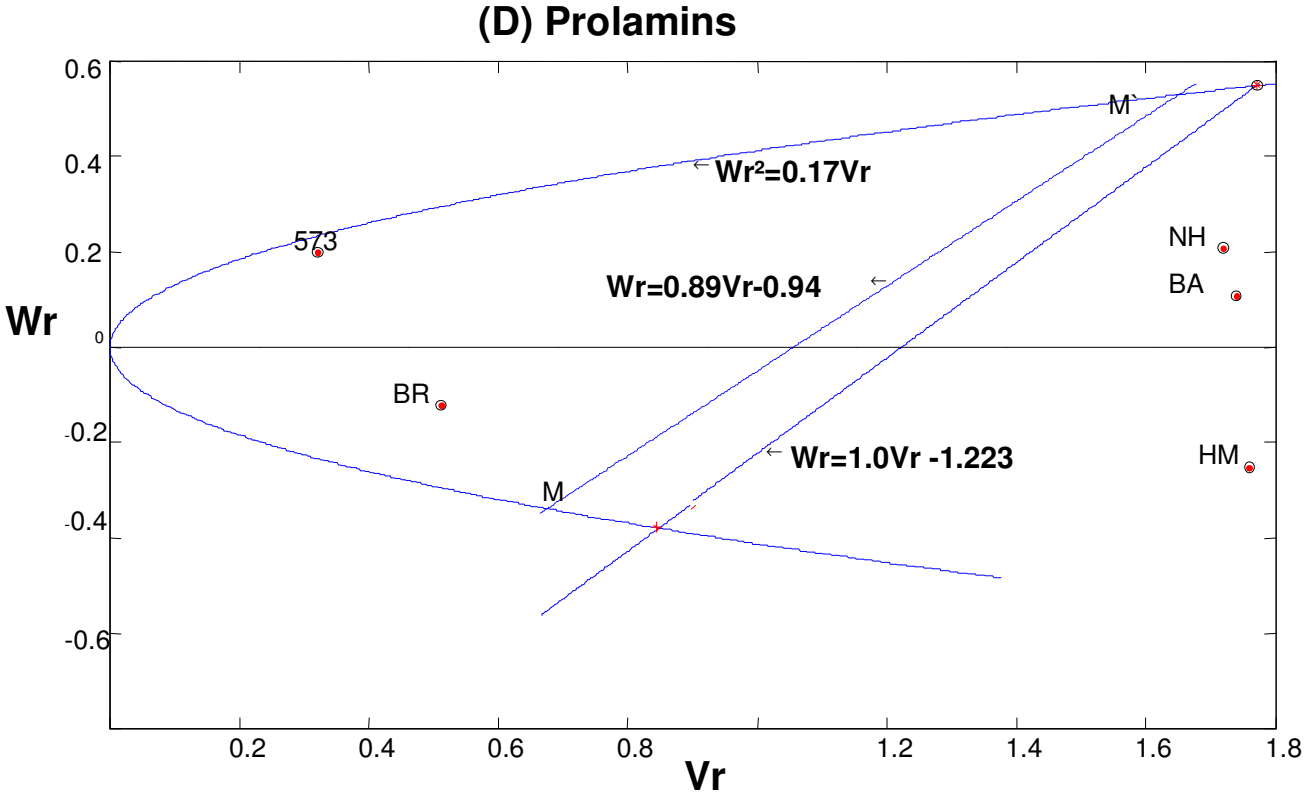
The analysis of variance based on Griffing's (1956) method showed that, the mean squares of GCA and SCA were significant ($p < 0.05$) for all traits (Table 5). The value of σ^2GCA /σ^2 SCA ratios showed that, SCA variance was higher than GCA variance component (0.33 to 0.60) except for globulin fraction (2.99).

The estimates of GCA effects of the parents (Table 6) revealed that, HM and 573 had positive significant values for protein (1.07 and 2.32) and for glutelins (2.08 and 8.11); BA had positive significant values for albumins (4.65), globulins (3.07) and prolamins (0.31), while BR showed positive significant values for prolamins (0.61) and globulins (0.63). Among ten cross combinations (Table 7), the hybrids HM x NH (good x moderate general combiners) for crude protein content; HM x 573 (moderate x poor general combiners) and HM x BR1 (moderate x moderate general combiners) for albumins; BR1 x 573 (moderate x moderate general combiners) and BA x HM (good x poor general combiners) for globulins; BR1 x HM (good x poor general combiners) for prolamins; BA x 573


(A) Crude protein content
Wr² = 17.21Vr
Wr = 0.311Vr-3.153
Wr = 1.0Vr-16.441
NH
HM
BR
M
573
BA
Wr
Vr
40
30
20
10
0
-10
-20
-30
0
5
10
15
20
25
30
35
40
45
50


(B) Albumins
Wr²=85.78Vr
Wr=0.85Vr-11.201
Wr=1.0Vr-18.038
BR
573
NH
BA
HM
Wr
Vr
100
80
60
40
20
-20
-40
-60
-80
-100
20
40
60
80
100
120


(C) Globulins
Wr²=73.15Vr
HM
BR
Wr=0.399Vr-40.83
BA
NH
573
M
M
Wr=1Vr-115.782
Wr
Vr
150
100
50
0
-50
-100
-150
0
50
100
150
200
250


(D) Prolamins
M`
Wr²=0.17Vr
573
NH
BA
Wr=0.89Vr-0.94
BR
Wr=1.0Vr -1.223
HM
M
Wr
Vr
0.6
0.4
0.2
0
-0.2
-0.4
-0.6
0.2
0.4
0.6
0.8
1.2
1.4
1.6
1.8

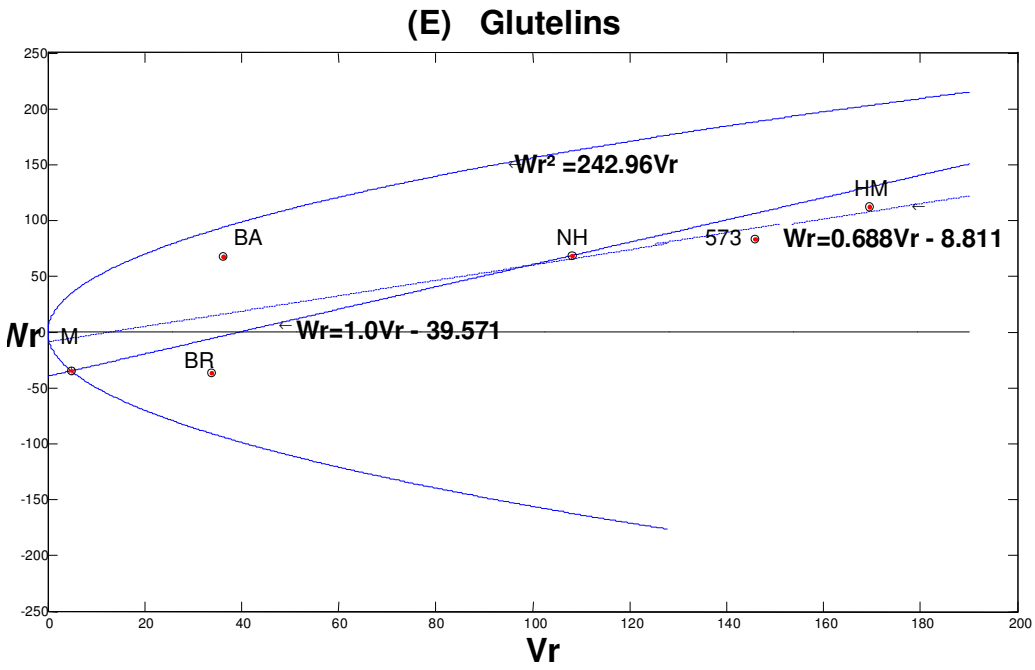


Figure 1. Wr/Vr graphs for seed crude protein (A), albumins (B), globulins(C), prolamins (D) and glutelins (E) contents in cowpea. Wr^2 = VrVp: Limiting parabola where Vp is the variance of the parents; Vr the variance of the rth array and Wr the covariance between the parents and their offspring in the rth array. Solid line: tangent to the limiting parabola (Wr = 1vr + b); dotted line: regression of Wr on Vr. BA: Bafia; BR: BR1; HM: hadiam marva; NH: Niébé Hosseré; 573: IT97K-573-1-1.

Table 5. ANOVA for combining ability of crude protein and soluble fractions in 5 x 5 half-diallel cross of cowpea.

Source	Df	Mean square of biochemical parameter				
		Crude protein	Albumin	Globulin	Prolamin	Glutelin
GCA	4	153.6**	14442.60**	5676.46**	1096.86**	1437.18**
SCA	5	11.05*	359.27**	85.62**	59.84**	126.96*
Error	18	1.19	16.42	5.84	4.4	13.14
σ^2GCA /σ^2 SCA		0.33	0.34	2.99	0.60	0.40

GCA: Variation due to general combining ability; SCA: variation due to specific combining ability; Error: error variation or interaction between the replication and genotypes; σ^2GCA: variance of general combining ability; σ^2SCA: variance of specific combining ability; *and ** indicates significance at $P \leq 0.05$ and $P \leq 0.01$, respectively.

(good x poor general combiners) for glutelins were identified as good specific combiners.

DISCUSSION

Significant differences amongst the ten pure lines for percentage of seed protein and its soluble fractions indicated the presence of diversity in the material. Variability for cowpea seed protein content was also reported by Senanayake and Wijerathne (1998), Emebiri (1991), Nielsen et al. (1993), Oluwatosin (1998), Singh et al. (2003), Giami (2005), Noubissié et al. (2007) and Ajeigbé et al. (2008). As reported by Tabe et al. (2000), seed composition is genetically controlled but the implementation of the program is affected by the environmental factors in particular nitrogen and suphur availability. The genotypic variability of cowpea seed for soluble proteins content was in agreement with other studies (Fosto et al., 1994; Ragab et al., 2004; Vasconcelos et al., 2010). Analyzing seven high protein content cowpea genotypes, Gupta et al. (2010) discovered that globulins were the major fraction (55.6 to 58.8%) followed conversely by glutelins (14.4 to 158.6%),

Table 6. Predicted general combining ability effects (GCA) for protein and soluble fractions of five cowpea cultivars on a 5 x 5 half-diallel.

Parent	GCA effect of biochemical parameter				
	Crude protein	Albumin	Globulin	Prolamin	Glutelin
BA	-1.25**	+4.65**	+3.07**	+0.31**	-8.22**
BR1	-1.97**	-0.51*	+0.63*	+0.61**	-2.31**
HM	+1.07**	+0.41	-3.17**	-0.26*	+2.08**
NH	-0.18	-1.36**	-0.22	-0.29*	+0.36
573	+2.32**	-3.19**	-0.29	-0.36**	+8.11**
SE	0.36	0.75	0.48	0.19	1.46

*Significant at p = 0.05 and **significant at p = 0.01; SE: standard error.

Table 7. Estimation of specific combining ability (SCA) of ten cowpea crosses in 5 x 5 half-diallel.

Hybrid combination	Crude protein	Albumin	Globulin	Prolamin	Glutelin
BA x BR1	0.50	4.46*	0.14	0.41*	1.30
BA x HM	0.52	-4.99*	9.39**	-0.45*	-5.06**
BA x 573	-5.76**	-0.60	-8.87**	-0.23	9.64**
BA x NH	-1.52	5.67*	-2.84	0.11	-4.22*
BR1 x HM	-2.94*	8.01**	-14.54**	0.77**	3.43*
BR1 x 573	-2.08	-7.46**	12.38**	0.32*	-5.22**
BR1 x NH	2.43	1.63	4.28 *	0.05	-3.8
HM x 573	-7.41**	10.69**	4.53*	-0.45**	-3.02
HM x NH	5.53**	-4.21*	5.71*	0.51**	-2.12
573 x NH	-4.56**	3.24*	-1.85	-0.25	-2.67
SE	1.49	1.06	1.23	0.23	1.12

SE: Standard error; *significant at p = 0.05 and **significant at p = 0.01.

albumins (8.2 to 11.9%) and prolamins (2.3 to 5.0%). According to Vasconcelos et al. (2010), the discrepancies among the contents of different protein types within cowpea genotypes depend on extracting method employed, the cultivars and also on genetic and environmental variability. Albumins and globulins, major stored protein in cowpea, had high rates of sulfur amino-acid (Ragab et al., 2004). Varieties with great amounts of these protein fractions like BA and HALG could be used extensively to fortify cereal-based weaning foods (Giami, 2005). Ceyhan (2006) and Ajeigbé et al. (2008) reported high positive correlations between the content of crude protein and ash (0.78), but negative correlations were observed between the content of crude protein and carbohydrate (-0.98), viscosity of cowpea flour (-0.76) and cellulose content (-0.81).

The significance of the mean dominance deviation (b_1) for traits indicated that, there is a non-directional dominance effects (Walters and Morton, 1978). The significant b_2 item illustrated an uneven distribution of dominant genes among the parents, reflecting that some parents harbored considerably dominant genes than others. Dominant and recessive loci are not harmoniously distributed among the parents. The residual dominance (b_3) which tests the part of the dominance unique to each F_1, was significant for all characteristics confirming the presence of specific dominance or combining ability in some crosses. Similar observations were noticed for protein content in pea by Gupta et al. (1984).

The significance of GCA and SCA for all traits shows the importance of both additive and dominance effects (Hayman, 1954). The values of σ^2GCA /σ^2SCA ratios and the variance components showing the preponderance of SCA for all characteristics except for the globulins, demonstrated the higher influence of non-additive gene effects (Mather and Jinks, 1982). The preponderance of non-additive genes was confirmed by the high proportion of dominant genes. Emibiri (1991) and Noubissié et al. (2007) reported both additive and dominant types of gene action in *V. unguiculata* for seed protein percentage. Ashokkumar and Ravikesavan (2008) reported similar results for seed protein content in cotton. In pigeonpea (*Cajanus cajan*), a legume crop, Beekham and Umaharan (2010) obtained a GCA / SCA ratio of 0.56 for pod crude protein content suggesting that, this character was also governed by a preponderance of non additive effects. Conversely, in field beans (*Vicia faba*), Griffiths and Laves (1978) noted that additive genes were preponderant

for seed protein content.

Broad sense heritability values were high (68 to 76%) indicating that, these characters are controlled mainly by genetic factors. High heritability values were also suggested for cowpea seed crude protein content by Senanayake and Wijerathne (1988), Nielsen et al. (1993), Emebiri (1991) and Ajeigbé et al. (2008). Ranges of moderate to high heritability for grain protein have been found in many crops like *V. faba* (Griffiths and Lawes, 1978), *Brassica napus* (Jieu, 1990), *Vigna sesquipedalis* (Rahman and Saad, 1999), *Glycine max* (Tajuddin et al., 2003), *Phaseolus vulgaris* (Ceyhan, 2006), upland cotton and *Gossypium hirsutum* (Ashokkumar and Ravikesavan, 2008). The low values of narrow sense heritability (0.22 to 0.43) showed remarkably large non-additive effects except for globulins which might be controlled mainly by additive genes. Hence, it may not be possible to improve these traits by adopting pedigree method (Mather and Jinks, 1982). For all traits, the average of degree of dominance $(H_1/D)^{1/2}$ was greater than one suggesting over dominance. Globally, the parents had a high proportion of dominant genes.

The Wr / Vr graph showed that, an additive-dominance model was verified for all traits. The coefficients of regression of Wr on Vr were not significantly different for unity in albumins (0.85) and prolamins (0.89) indicating also the adequacy of the simple additive-dominance genetic model (Hayman, 1954). However, the coefficients were significantly different from unity in globulins (0.39), proteins (0.31) and glutelins (0.68), suggesting the possibility of non-allelic interaction. Significant epistasis has been reported for protein content in *V. sesquipedalis* (Rahman and Saad, 1999) and *Triticum durum* (Bnejdi and El Grazzah, 2010). Emibiri (1991) noticed by comparing reciprocal segregating generations of two crosses of cowpea that cytoplasmic factors could influenced the inheritance pattern of seed protein content. The regression analysis showed over-dominance type of gene action for all characters (Figure 1). As over-dominance type of gene action was present, selection based on these traits would be difficult in early generations (Shi et al., 1999). It means that, the genotypes are more efficient for producing increased seed proteins contents in hybrid condition.

The positive correlation between parental values (Pr) and recessive factor (Wr+Vr) indicated that, dominance was in favor of low crude protein content and low percentage of glutelins. Similar findings were reported in pea by Gupta et al. (1984) for seed protein content. In *B. napus,* Jieu (1990) also reported that, dominance decreesed the protein rate. The association of high protein or glutelins content with recessive genes might present some difficulties for selection during the early generations. For albumins, globulins and prolamins content, correlation analysis of the genotypes showed positive dominant gene control.

The relative position of the arrays points on the regression line depicted that NH and HM had the most recessive genes for seed protein content, while BR had the most dominant genes. For albumins, HM and BA had the most favorable dominant genes, whereas 573 had the least. BA and HM having a positive and significant GCA would be good combiners to increase the albumins rates. BA would have dominant genes with positive effect and NH would have recessive genes with negative effect on globulin rates. BA with high good AGC would be a good combiner to increase the globulin rates. Genotypes 573 and HM possessing most recessive genes and having high AGC would be good combiners for high glutelins content. For prolamins, the parental lines possessed both positive and negative alleles. BA harbored more recessive genes with positive effect for high prolamin content. Line 573, a good combiner, located near the lower end of the regression line would have dominant genes with positive effect for this trait.

SCA performance might be considered as a criterion for selecting the best crosses. The low x low or low x moderate general combiners exhibiting high SCA effects suggested gene dispersion and genetic interaction between favourable alleles contributed by both parents (Fonsecca and Patterson, 1968; Angenon et al.1999). Inclusion of F_1 hybrids showing high SCA and having parents with good GCA, into multiple crosses, could be a worthwhile approach for tangible improvement of cowpea grain protein or its soluble fractions for specific uses in different recipes (Griffiths and Leaves, 1978). Pedigree, bulk or single seed/pod descent methods are suggested to developing elite populations (Emebiri, 1991).

Improvement of nutritional quality might include both conventional and biotechnological means (Tajuddin et al., 2003). For example, the classical breeding efforts to increase the concentration of limiting amino acids in most grain legumes have so far not been successful (Osborne, 1988; Vasconcelos et al., 2010). In legumes, gene engineering can contribute to the improvement of seed amino acids content and the reduction of antinutritional factors like phytate and raffinose-family-oligosaccharides (Angenon et al., 1999; Mandal and Mandal, 2000). Genome mapping has allowed development of DNA markers linked to quantitative trait loci (QTL) which contribute to the phenotypic expression of quantitatively inherited characters (Tabe et al., 2000; Tajuddin et al., 2003). In soybeans, Shi et al. (2010) identified simple sequence repeat (SSR) markers associated with protein content.

Conclusion

Cowpea genotypes are highly variable for seed protein and its soluble fractions contents. These characteristics were controlled predominantly by non-additive genes. Understanding of the inheritance of these characteristics made a clear view-point of the selection of parental lines for improvement of seed quality. Improved methods to

predict genetic gain and evaluate these quantitative traits without the environmental influence are also needed.

ACKNOWLEDGEMENTS

The authors are grateful to Professor Ukai Yasuo of the University of Tokyo for providing the microcomputer program DIALL. We are also thankful to Mr Guidana of the Department of Computers Science, University of Ngaoundéré for his guidance on data analysis.

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