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# Assessment on genotypic variability and character association using *Rhizobium* cultures on growth and nodulation of cowpea (*Vigna unguiculata* L. Walp.)

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#### Abstract

The present experiment was carried out with five genotypes of cowpea and three rhizobium cultures to assess the extent of genetic variability and character association for growth and nodulation. Field measurements and other studies reveal that nodulation and nitrogen fixation is a complex character being sensitive to environmental and genetic factors affecting the plant, bacteria and their interaction. Significant variability was also found for most of the characters when grown with rhizobium culture. Variable interaction was found with the genotypes e.g., groundnut culture showing positive interaction with cowpea genotypes. EC-101943 was found to be the best performing cowpea genotype in terms of grain yield spreading over the treatments. Grain protein content was not found to be correlated with the nodule characters directly though was found to be correlated with other non-nodulating characters. Shoot nitrogen content was found to be correlated with matured grain protein content indicating the possibility of assimilation of fixed nitrogen being utilized in a defined pathway. The enhanced effect of nitrogen fixation as evidenced from the results on grain yield might be due to indirect contribution of other correlated nodulating and non-nodulating characters, hence selection to be done accordingly.

Keywords: cowpea (Vigna unguiculata L. Walp.), Rhizobium, growth nodulation

#### Introduction

The rising population worldwide with time has necessitated the increased need for food and the food basket, if at all are made self-sufficient more and more emphasis be given on more production from the given area of land. But with declining soil fertility and associated small size of the holding intensification of agriculture seems to be difficult though more use of nitrogen fertilizers could not sustain the yield of the crop to the desired extent. The amount of nitrogen fertilizers needed for this is surely projected to increase and environmental degradation is also estimated to be on the higher side. Though the reliance on chemical fertilizer as the dominant source will continue with this persisting need, there is an urgent need to evolve to circumvent the problems of environmental hazards associated with the enhanced use of same and also to adopt such farming approaches so that economically viable and environment prudent friendly agriculture can be ensured (Woldeyohannes *et al.*, 2007; Zaman-Allah *et al.*, 2007) <sup>[11. 12]</sup>.

Legumes are widely accepted to have the unique properties of fixing atmospheric nitrogen through symbiotic association with *rhizobium*. The effect of nodulation on dry matter and grain yield of chickpea upon inoculation by *Rhizobium* has also been studied (Bhuiyan *et al.*, 2001)<sup>[1]</sup>.

The supplementary role of biofertilizers in this regard is well documented which is not only economical but also environment friendly. The nitrogen fixers like *Rhizobium* species are able to enter into a symbiotic relationship with the legumes which not only enhance the production of the crop but also leave behind a substantial amount of nitrogen in the soil benefiting the succeeding crop. Biological nitrogen fixation (BNF) as a source of nitrogen fertilizer has long been known as a potent source in the farming systems to minimise the use of chemical fertilizers (Shamseldin and Werner, 2004; Shamseldin, 2007; Vinuesa *et al.*, 2003) <sup>[8, 9]</sup>.

Rhizobium strain inoculation does not only favour nonrepeated application of nitrogen fertilizers but also has a direct effect on yield due to increased nodulation (Sanginga et al., 1994) <sup>[6]</sup>. The biological Nitrogen fixing ability for cowpea could be made more productive through intervention in the host plant system or in the nitrogen fixing system or their interaction. Hence efforts should be directed to increase the ability to fix atmospheric nitrogen inherent in such nitrogen fixing system to make agriculture more sustainable within its realm. Cowpea though adapted to the tropics is also grown nowadays in the terai region of Bengal. A good number of varieties are available for grain, fodder and vegetable purposes. But the use of crop specific rhizobium culture or unrelated culture in grain legumes has not been so popular in this region of West Bengal though rhizobium inoculation could enhance the yield and other yield related characters- both nodulating and non-nodulating characters. Against this background, the present study was taken up to investigate growth and nodulation characteristics of cowpea for genotypic variability and character association.

## **Material and Methods**

The present experiment was carried out with five genotypes of cowpea (*Vigna unguiculata* L. Walp) and three rhizobium cultures during the period 2013-14 and 2014-15 to assess the extent of genetic variability for growth and nodulation in the studied genotypes and to ascertain the extent of correlation existing between different nodulating and non-nodulating characters. The experiment was conducted in the instructional farm of Uttar Banga Krishi Viswavidalaya, Cooch Behar, West Bengal during the above stated period. The experimental site was situated at 26020/ N latitude and 89023/ E longitude and an altitude of 43 meter above the mean sea level. The soil of the experimental site used was sandy loam soil with optimum water holding capacity.

In the present investigation, five (5) genotypes of cowpea (EC-101967, EC-101943, EC-121829, IC-549343 and Kashi Kanchan) were collected from different region of west Bengal and the *Rhizobium* cultures used in the experiment were collected from the Nodule Research Laboratory, Bidhan Chandra Krishi Viswavidyalaya, Nadia, West Bengal. The experiment was laid out in factorial randomized block design with genotypes and the cultures being the factors with three replications. The plots in each replication were measured 3m by 4m (area 12 square meters).

## Treatments of the experiment

Seven treatments were used in the experiment for each of the five varieties of cowpea in each replication in the investigation which have been listed below:

- a. Control without inoculation but with nitrogen fertilizer (WN)
- b. Cowpea culture (C1)
- c. Mungbean culture (M1)
- d. Groundnut culture (G1)
- e. Cowpea culture x mungbean culture (C1M1)
- f. Cowpea culture x groundnut culture (C1G1)
- g. Mungbean culture x groundnut culture (M1G1)

#### Method of inoculation

Seeds of cowpea uniform shape, size and weight were first surface sterilized to get rid of surface micro- organisms. And for this seeds were then rinsed with 70% ethyl alcohol and suspended for 10 minutes in 0.1% mercuric chloride (HgCl<sub>2</sub>) solution after which they were washed repeatedly with sterile distilled water. For inoculation, the seeds were moistened with a small amount of water and pelted with peat based *Rhizobium* inoculum containing per gram with concentrated sugar solution as an adhesive and were sown in the sterilized pots following drying. A control without inoculation along with nitrogen fertilizer was included for comparison. All agronomic practices were kept uniform and normal for all the treatments.

#### **Statistical Analysis**

The data observed on different parameters in both the crops were subjected to statistical analysis based on the sample means of various characters spread over three replications.

#### **Result and Discussion**

# Effects of rhizobium inoculation on different characters in cowpea genotypes

The analysis of variance of various characters studied in cowpea genotypes revealed significant differences among the genotypes, treatments and the interaction between genotype and treatment for all the except grain protein content and nitrogen content in soil with treatment (Table 1).

The genotype EC-121829 produced maximum number of lateral roots with a combination Cowpea-6 and M10, while in other two combinations; Kashi Kanchan had the maximum number for the same character at maturity stage (Table 2). Except EC-101943, other genotypes showed maximum values of total root length in either the sole culture treatment or in combination of cultures (Table 2). As far as the shoot fresh weight was concerned, EC-101967 showed maximum value for the character with all the treatment or their combination except Cowpea-6 and JCG-1, EC-121829 and EC -101943 had the same maximum value in response to Cowpea-6 and JCG-1 respectively (Table 3). When the effects of inoculation was studied with respect to dry weight of shoot in cowpea genotypes, variable response was noted with IC-549343 recording maximum values in three of the seven treatments (Table 3). The change in response was obvious with EC-101967 recording the maximum in M-10 and its combination with JCG-1 and also in the combination culture of Cowpea-6 and JCG-1. Kashi Kanchan, recorded maximum root fresh weight in response nitrogen application, and in culture JCG-1 and combination of cultures Cowpea-6 and M-10 (Table 4). The dry weight of root when studied in various treatments, variety Kashi Kanchan while recorded maximum values with nitrogen, it had the maximum root dry weight in in response to JCG-1 at maturity (Table 4). As far as the number of nodules per plant was concerned the response to treatments was also found to be variable while EC-101967 produced the maximum in culture combination of Cowpea-6 with M-10 and JCG-1, EC-121829 had the same trend in response to Cowpea-6 and M-10 as against Kashi Kanchan producing the maximum result for the character in response to nitrogen and in culture combination of M-10 and JCG-1 (Table 5). As far as the fresh weight of nodules was concerned, the genotype EC-121829 too had the same effect of having the maximum fresh weight of nodules with culture Cowpea-6 and JCG-1 (Table 5). Effects of rhizobium inoculation on dry weight of nodules revealed that the genotype EC-101967 had a similar effect in response to M-10, combination of Cowpea-6 and JCG-1. (Table 6) As far as nitrogen content in shoot was concerned EC-101967 gave the maximum value of nitrogen while EC-101943 in response to culture combination of M-10 with Cowpea-6 and JCG-1 respectively. In response to culture M-10 and JCG-1 along EC-121829 had the maximum nitrogen content in shoot as also in the culture combination of Cowpea-6 and JCG-1. In response to culture Cowpea-6 the shoots of IC-549343 were found to have maximum nitrogen content in shoot at maturity. Similar to shoot the nitrogen content in root was also measured which revealed that in all the three combination cultures and in culture M-10 the genotype EC-101943 had the maximum nitrogen content as against EC-121829 producing the maximum value in response to the sole culture of Cowpea-6 and JCG-1. The maximal effect was noticed in the genotype EC-101967 in their root in response to the application of nitrogen along (Table 7). The grain protein content was also affected by different treatments

as presented in (Table 8). In addition to the maximum grain protein content produced by the mature seeds to Kashi Kanchan in response to the combination of Culture-6 and JCG-1. Grain yield calculated in kg on single plot basis revealed that the treatments used in the experiment all but two i.e. culture combination of Cowpea-6 and JCG-1 and that of M-10 JCG-1 had the effect of producing maximum grain yield in the genotype EC-1014943. While in culture combination of Cowpea-6 and JCG-1 produced maximum grain yield in the genotype EC-121829. The same of M-10 and JCG-1 produced maximum grain yield in the genotype EC-101943 (Table 8).

Sources	d.f.	NRL	TRL(c m)	SFW(g)	SDW(g)	RFW(g )	RDW (g)	NP	NS(mm )	NFW(g )	NDW(g )	NDW (g)	IGPC (%)	MGP C (%)	GY\P (kg)	SHT N (g\kg)	RN (g\kg)	NSC (kg\ha)
Replication	2	23.49*	23.98*	213.28*	20.64*	6.28X10 -2*	1.38	23.49 *	23.67*	0.038*	5.68x10 -4*	5.92X10 -4*	18.32 *	24.14*	5.85X10 -4*	0.122	0.121 *	6162.13 *
Genotype	4	171.22*	130.79*	5600.08*	3166.01 *	51.86*	6.97*	730.9 4*	3.09*	0.018*	8.46x10 -3*	7.42X10 -3*	0.245	0.311	3.18X10 -1*	0.031	0.915 *	344.08
Treatment	6	298.68*	447.01*	11687.02*	2514.40 *	115.8*	25.28 *	252.6 2*	3.16*	0.022*	9.54x10 -3*	7.99X10 -3*	0.379	0.478	6.56X10 -2*	0.134 *	1.259 *	508.99*
Genotype x Treatment	24	104.39*	91.57*	2182.80*	1221.50 *	27.47*	6.26*	137.9 9*	7.89*	0.022*	6.18x10 -3*	5.56X10 -3*	0.079	0.097	4.92X10 -2*	0.008 *	0.136 *	137.19
Error	68	0.32	0.31	2.84	0.316	8.04X10 -4	0.17	0.319	0.32	0.0005	8.59x10 -6	7.90X10 -6	0.243	0.312	8.02X10 -2	0.001	0.001	94.69

Table 1: Analysis of variance for	or different characters of five	cowpea genotypes at maturity
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\* Significant at 5%,

Table 2: Effects of rhizobium inoculation on number of lateral roots and total root length (cm) in cowpea genotypes

			Nu	nber	of lateral	roots			7	Fotal r	oot len	gth (cm	l)	
<b>Treatment Genotype</b>	with N	<b>C1</b>	<b>M1</b>	<b>G1</b>	C1M1	C1G1	M1G1	with N	C1	M1	G1	C1M1	C1G1	M1G1
EC-101967	23	32	40	27	29	31	22	15.50	17.64	28.50	37.81	15.61	19.21	36.13
EC-101943	26	28	35	49	29	28	38	11.56	25.71	20.94	32.74	19.62	13.23	24.16
EC-121829	20	23	39	47	35	25	22	15.06	36.70	27.91	28.55	28.11	24.41	26.21
IC-549343	25	28	30	27	31	33	36	15.52	18.71	29.54	23.23	16.31	16.78	23.51
Kashi Kanchan	31	34	43	37	35	39	33	14.91	16.10	19.68	29.67	18.87	30.21	33.11
S.E. (m)					0.123						0.122			
S. E. (d)					0.174						0.173			
C. V					1.8%						2.4%			

Table 3: Effects of rhizobium inoculation on fresh weight (g) of shoot and dry weight (g) of shoot in cowpea genotypes

		ŀ	resh w	eight (g	) of shoo	t			D	ry wei	ight (g	) of sho	ot	
<b>Treatment Genotype</b>	with N	C1	M1	G1	C1M1	C1G1	M1G1	with N	C1	M1	<b>G1</b>	C1M1	C1G1	M1G1
EC-101967	68.08	141.32	147.80	97.54	168.88	147.11	167.44	16.53	32.57	39.6	26.61	23.38	59.45	46.61
EC-101943	62.20	84.41	109.41	175.67	88.32	114.62	148.72	18.57	27.90	76.03	62.44	31.04	34.20	59.45
EC-121829	44.84	159.50	73.36	94.42	86.58	84.87	88.16	16.76	35.51	12.81	18.10	31.61	25.07	21.30
IC-549343	36.09	129.58	84.22	132.20	157.8	91.88	120.52	10.97	71.35	22.41	84.60	88.54	31.20	76.25
Kashi Kanchan	35.84	134.39	96.52	115.58	126.279	103.73	88.99	5.93	76.84	30.24	66.58	62.77	59.34	26.95
S.E. (m)				0.368							0.123	3		
S. E. (d)				0.520							0.174	Ļ		
C. V				1.5%							1.4%			

Table 4: Effects of rhizobium inoculation on fresh weight (g) of root and dry weight (g) of root in cowpea genotypes

		F	resh w	eight (	g) of ro	ot			Ι	Dry w	eight	(g) of re	oot	
Treatment Genotype	with N	C1	M1	G1	C1M1	C1G1	M1G1	with N	<b>C1</b>	<b>M1</b>	<b>G1</b>	C1M1	C1G1	M1G1
EC-101967	3.89	11.68	20.29	11.76	10.76	14.55	17.40	0.78	4.68	6.90	3.71	3.36	5.40	8.50
EC-101943	6.67	8.31	13.05	13.38	14.37	10.13	17.05	1.64	2.96	5.88	4.57	6.32	5.32	7.68
EC-121829	2.70	13.26	8.46	9.94	9.84	9.14	8.19	0.98	5.11	2.74	3.74	3.69	4.85	3.68
IC-549343	6.13	14.52	8.68	15.22	15.86	9.57	11.58	2.58	6.32	2.98	6.32	5.97	3.59	5.29
Kashi Kanchan	7.59	13.99	9.65	15.32	16.20	12.62	9.69	3.51	6.01	3.12	6.85	6.20	5.21	3.67
S.E. (m)				0.006	i						0.0	)9		
S. E. (d)				0.009							0.1	27		
C. V				0.2%							8.8	3%		

Table 5: Effects of rhizobium inoculation on nodules per plant and size (mm) of the nodule in cowpea genotypes

			No	dule	s per pla	nt			S	ize (1	mm)	of the no	odule	
<b>Treatment Genotype</b>	with N	<b>C1</b>	<b>M1</b>	<b>G1</b>	C1M1	C1G1	M1G1	with N	<b>C1</b>	<b>M1</b>	<b>G1</b>	C1M1	C1G1	M1G1
EC-101967	11	15	19	28	19	22	15	5.3	5.1	4.3	4.9	5.1	5.6	6.1
EC-101943	18	17	35	22	18	19	24	6.1	2.3	3.2	6.3	3.3	5.2	4.9
EC-121829	13	21	47	19	11	14	21	4.3	9.3	7.1	3.1	4.2	2.3	2.3
IC-549343	14	10	20	21	25	18	17	6.2	3.9	5.1	5.1	3.9	5.3	4.2
Kashi Kanchan	26	37	26	36	38	30	34	6.3	4.6	5.3	5.3	4.6	4.3	7.1
S.E. (m)				0	.123						0	.123		
S. E. (d)				0	.174						0	.173		
C. V				2	2.5%						1	1.4%		

Table 6: Effects of rhizobium inoculation on fresh weight (g) of nodules and dry weight (g) of nodules in cowpea genotypes

		Fre	sh we	eight (	(g) of no	dules			D	ry weiş	ght (g)	of nodu	les	
Treatment Genotype	with N	C1	<b>M1</b>	<b>G1</b>	C1M1	C1G1	M1G1	with N	C1	M1	G1	C1M1	C1G1	M1G1
EC-101967	0.18	0.11	0.20	0.12	0.15	0.16	0.21	0.011	0.036	0.061	0.058	0.065	0.086	0.101
EC-101943	0.04	0.08	0.13	0.48	0.10	0.16	0.14	0.029	0.021	0.050	0.209	0.038	0.074	0.033
EC-121829	0.12	0.22	0.14	0.09	0.13	0.56	0.03	0.012	0.056	0.059	0.016	0.060	0.029	0.009
IC-549343	0.12	0.17	0.06	0.14	0.15	0.08	0.20	0.075	0.033	0.009	0.047	0.077	0.034	0.053
Kashi Kanchan	0.25	0.06	0.11	0.30	0.30	0.10	0.12	0.081	0.013	0.039	0.132	0.194	0.058	0.072
S.E. (m)				0.0	05						0.001			
S. E. (d)				0.0	07						0.001			
C. V				15.4	4%						4.8%			

Table 7: Effects of rhizobium inoculation on nitrogen content in shoot (%) and root (%) in cowpea genotypes

		Nitro	ogen o	conter	nt in sho	ot (%)			Nitr	ogen	conte	nt in roo	ot (%)	
Treatment Genotype	with N	C1	M1	G1	C1M1	C1G1	M1G1	with N	C1	<b>M1</b>	<b>G1</b>	C1M1	C1G1	M1G1
EC-101967	15.2	18.8	17.7	13.1	35.5	23	20.7	7.2	8.9	13.2	8.8	10.5	9.3	8.3
EC-101943	15.6	13.8	18.3	27.3	20	23.4	24.2	7.1	11.3	8.3	11.3	9.1	9.4	9.3
EC-121829	16.2	21.4	24.7	15.7	12.1	21.1	22.9	6.9	10.7	8.9	11.7	9.9	11.5	8.2
IC-549343	12.5	10.3	13.9	16	19.1	12.9	15.9	7.5	8.8	8.4	9.5	11.7	8.2	8.7
Kashi Kanchan	12.7	22.6	10.4	11.9	16.3	15.8	15.6	7.1	8.3	7.3	8.7	9.2	8.8	8.4
S.E. (m)				0.0	08						0.0	08		
S. E. (d)				0.0	11						0.0	12		
C. V				6.5	%						3.2	%		

Table 8: Effects of rhizobium inoculation on grain protein content (%) and grain yield (kg plot<sup>-1</sup>)in cowpea genotypes

		Gı	ain pr	otein c	ontent (	%)			6	Frain	yield	(kg plot	-1))	
<b>Treatment Genotype</b>	with N	C1	M1	G1	C1M1	C1G1	M1G1	with N	C1	M1	<b>G1</b>	C1M1	C1G1	M1G1
EC-101967	23.89	24.44	24.95	24.56	24.84	24.42	24.56	0.27	0.48	0.47	0.44	0.10	0.36	0.37
EC-101943	24.00	24.87	24.58	24.63	24.32	24.50	24.37	0.55	0.59	0.57	1.12	0.77	0.53	0.52
EC-121829	24.31	24.56	24.14	24.23	24.32	24.18	24.38	0.39	0.48	0.26	0.34	0.20	0.55	0.38
IC-549343	24.23	24.74	24.61	24.62	24.58	24.53	24.49	0.31	0.46	0.45	0.52	0.47	0.47	0.43
Kashi Kanchan	24.15	24.71	24.82	24.57	24.69	24.83	24.63	0.47	0.46	0.51	0.53	0.47	0.35	0.42
S.E. (m)				0.122							0.0	61		
S. E. (d)				0.172							0.0	86		
C. V				2.3%							6.1	%		

C1 = Cowpea-6, M1 = M-10, G1 = JCG-1

# Studies on phenotypic correlation among different characters in cowpea in response to various treatments

The phenotypic correlation among the different characters when studied at maturity in cowpea genotypes with seven treatments showed that grain yield was not correlated with any of the other characters under study. Interestingly, grain protein content in both immature and mature seeds was positively and significantly correlated with nitrogen content in soil at flowering stage and also at maturity. The fresh and dry weight of shoot were positively and significantly correlated with the same of root and root nitrogen content was positively and significantly correlated with shoot nitrogen content though root nitrogen content was negatively correlated with shoot dry weigh (Table 9).

Table 9: Phenotypic correlation among different characters of cowpea genotypes at maturity

$\rightarrow$	GY	IGPC	MGPC	NDW	NFW	NLR	NP	NS	RDW	RFW	RN	SDW	SFW	SHTN	NS F	NSC	TRL
GY																	
IGPC	0.066																
MGPC	0.063	0.886*															
NDW	0.371	0.057	0.153														
NFW	0.413	0.265	0.383	0.832*													
NLR	0.281	0.288	0.191	0.365	0.349												
NP	0.043	0.163	0.146	0.339	0.204	0.422											

NS	-0.029	0.358	0.437	0.324	0.504	0.037	0.234										
RDW	0.203	0.181	0.218	0.342	0.282	0.181	0.188	-0.048									
RFW	0.259	0.111	0.176	0.413	0.336	0.335	0.243	-0.015	0.859*								
RN	-0.215	0.073	-0.017	-0.327	-0.061	-0.184	-0.135	0.150	-0.430	-0.433							
SDW	0.291	0.111	0.173	0.321	0.255	0.259	0.246	-0.115	0.674*	0.709*	-0.518*						
SFW	0.207	0.135	0.229	0.392	0.399	0.257	0.067	0.130	0.701*	0.790*	-0.390	0.648*					
SHTN	-0.201	0.387	0.296	-0.184	0.041	-0.014	-0.052	0.240	-0.409	-0.425	0.720*	-0.352	-0.414				
NS F	0.340	0.602*	0.730*	0.147	0.387	0.137	0.244	0.491	0.050	0.113	0.181	0.097	0.152	0.261			
NSC	-0.049	0.756*	0.848*	0.135	0.310	0.012	0.045	0.348	0.249	0.141	0.031	0.034	0.269	0.174	0.507		
TRL	0.108	0.169	0.185	0.217	0.241	0.224	0.203	0.204	0.232	0.317	-0.181	0.041	0.276	-0.170	0.250	0.220	

NLR= Number of lateral roots, TRL = Total root length, SFW= Shoot fresh weight, SDW= Shoot dry weight, RFW= Root fresh weight, RDW= Root dry weight, NP= Nodules per plant, NS= Nodule size, NFW= Nodule fresh weight, NDW = Nodule dry weight,

IGPC=Immature grain protein content, MGPC=Mature grain protein content, GY=Grain yield per plot, SHT N=Shoot nitrogen, RN=Root nitrogen, NSC=Nitrogen content in soil with culture, NS F =Nitrogen content in soil at flowering stage

### Discussion

Culture combination of Cowpea-6 and JCG-1 and that of M-10, JCG-1 had the effect of producing maximum grain yield in the genotype EC-1014943. Prabakaran and Rangarajan (1992) while studying the effect of Rhizobium inoculation (two strains) on nodulation, nitrogen fixation and yield of four cultivars of lablab (Phaseolus vulgaris L.) similarly found the highest seed yield in cultivar Co-7 by 45.8 to 56.9% by inoculation with both the strains. The level of nitrogen accumulation and nodulation were also increased by the inoculums. Cowpea is generally recognized as being nodulated by a large range of soil rhizobia (Ulzen, 2013) <sup>[10]</sup>. This condition makes the establishment of an effective inoculant strain very difficult and as a result, cowpea inoculation is rarely performed under field conditions. However, there are reports of increased grain yield of cowpea due to inoculation (Martins *et al.*, 2003; Nyoki and Ndakidemi, 2013; Sarker *et al.*, 2001) <sup>[3, 4, 7]</sup>. Results obtained from this study showed that in all cases, a combined application of inoculants increased yields more than the sole inoculant application (Table 8). The increase in yield may be attributed to the effectiveness of the inoculant in fixing nitrogen thereby meeting the nutrient requirement of the plant (Nyoki and Ndakidemi, 2013)<sup>[4]</sup>. The observed low grain yield from the sole inoculation application may be due to restriction of nodulation, reduced functioning of nodules and subsequently low yields as asserted by Sanginga et al. (2003) <sup>[5]</sup>. The inability of cowpea to show positive yield response to inoculation could be attributed to the variety, environmental conditions and type of inoculant used. Inoculation success/failure is highly site specific (Date, 2000) and makes generalizations difficult. Comparing the yield obtained from inoculated plants and uninoculated plants dependent on mineral fertilizer, there was no significant difference which further confirmed that the indigenous rhizobia did not out compete the introduced strain. The results indicated that N content significantly increased in the rhizobia inoculated treatments above the control. Perhaps this could be attributed to increased biological nitrogen fixation following inoculation as asserted by Nyoki and Ndakidemi (2014). It was also noted that rhizobial inoculation increased biomass N content indicating that most of the biomass N content was derived from BNF. Nitrogen uptake in cowpea biomass and grain may also be attributed to the nodulation enhanced by phosphorus which increased biological nitrogen fixation.

# Conclusion

Significant variability was also found for most of the characters in both the crops even when grown with rhizobium culture. Variable interaction was found with the genotypes e.g., groundnut culture showing positive interaction with

cowpea and even with mungbean genotypes. Thus a cross combination of cultures in unrelated crops could be used to have the desired result as far as nitrogen fixing ability of the crop and final yield and other desirable attributes were concerned.

EC-101943 similarly was found to be the best performing cowpea genotype in terms of grain yield spreading over the treatments. Grain protein content was not found to be correlated with the nodule characters directly though was found to be correlated with other non-nodulating characters. In cowpea shoot nitrogen content was found to be correlated with matured grain protein content indicating the possibility of assimilation of fixed nitrogen being utilized in a defined pathway. The enhanced effect of nitrogen fixation as evidenced from the results on grain yield might be due to indirect contribution of other correlated nodulating and nonnodulating characters, hence selection to be done accordingly.

## References

- Bhuiyan MAH, Khanam D, Khatun A, Hossain AKM, Islam MS. Nodulation, dry matter weight and grain yield of chickpea as influenced by *Rhizobium* inoculation. Bangladesh Journal Agricultural Research. 2001; 26(3):463-466.
- 2. Date RA. Inoculated legumes in cropping systems of the tropics. Field Crops Research. 2000; 65:123-136.
- Martins LMV, Xavier GR, Rangel FW, Ribeiro JRA, Neves MCP, Morgado LB, Rumjanek NG. Contribution of biological nitrogen fixation to cowpea: a strategy for improving grain yield in the semi-arid region of Brazil. Biology and Fertility Soils. 2003; 38:333-339.
- 4. Nyoki D, Ndakidemi PA. Effects of *Bradyrhizobium japonicum* inoculation and supplementation with phosphorus on macronutrients uptake in cowpea (*Vigna unguiculata* [L.] Walp). American Journal of Plant Sciences. 2014; 5:442-451.
- 5. Sanginga N, Dashiell K, Diels J, Vanlauwe B, Lyasse O, Carsky RJ, Tarawali S, *et al.* Sustainable resource management coupled to resilient germplasm to provide new intensive cereal-grain legume livestock systems in the dry savanna. Agriculture, Ecosystems and Environment. 2003; 100:305-314.
- Sanginga N, Mulongoy K, Ojeifo AA. Persistence and recovery of induced Rhizobium ten years after inoculation on *Leucaena leucocephala* grown on an Alfisil in Southwestern Nigeria. Plant Soil. 1994; 159:199-204.
- Sarker PC, Rahman AFMF, Shiblee SMA., Muquit A, Uddin ASMM, Anwar MN. Effect of rhizobium inoculants on cowpea under rainfed condition. Parkistan Journal of Biological Sciences. 2001; 4(6):654-656.

- 8. Shamseldin A, Werner D. Selection of competitive strains of *Rhizobium* nodulating *Phaseolus vulgaris* and adapted to environmental conditions in Egypt, using the gus-reporter gene technique. World Journal of Microbiology and Biotechnology. 2004; 20:377-382.
- 9. Shamseldin A. Use of DNA marker to select welladapted Phaseolus - symbionts strains under acid conditions and high temperature. Biotechnology Letter. 2007; 29:37-44.
- 10. Ulzen J. Assessing the need for inoculation of soybean and cowpea at Tono in the Kassena Nankana district of the upper east region of Ghana. MSc thesis Faculty of Agriculture, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana, 2013.
- 11. Woldeyohannes WH, Dasilva MC, Gueye M. Nodulation and nitrogen fixation of *Stylosanthes hamata* in response to induced drought stress. Arid Land Research and Management. 2007; 21:157-163.
- 12. Zaman-Allah M, Sifi B, L'Taief, B, El-Aouni MH, Drevon JJ. Rhizobial inoculation and P fertilization response in common bean (*Phaseolus vulgaris*) under glasshouse and field conditions. Experimental Agriculture. 2007; 43: 67-77.