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Full Length Research Paper

Effects of cropping system and Cowpea variety (*Vigna unguiculata* L. Walp) on the diversity of native cowpea bradyrhizobia and millet yield in the Sudano Sahelian zone of Mali

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Abstract

Cowpea is an important food grain legume in the Sudano Sahelian zone of Mali. So far, an attention has not been addressed to the diversity of bradyrhizobia nodulating cowpea in this area. In order to fill this gap, effects of 3 cowpea varieties (IT89KD-374, CZ1-94-23-1 and CZ11-94-5C) on the diversity of native bradyrhizobia has been studied in 3 cropping systems from 2007 to 2008 at Cinzana Agronomic Research Station. The objective of the study was to identify effective bradyrhizobia strains nodulating cowpea and their impact on the intercropped or subsequent millet yield in the main cropping systems. Experimental design was a Randomized Complete Block Design using a factorial combination of 3 cowpea varieties and 3 cropping systems in 4 replications. PCR-RFLP and sequencing of the 16S – 23S of rDNA IGS were used to identify bradyrhizobia strains associated with cowpea varieties. Six IGS types have been identified. IGS type I (strain *Bradyrhizobium* sp.ORS 3642) was the most competitive regardless of cropping system and cowpea variety. The greater diversity averaged bradyrhizobia was found in variety IT89KD-374 (H' means = 0.39). Best millet grain yield in intercropped system (1520 kg ha^{-1}) was obtained with cowpea variety IT89KD-374. The best above ground dry biomass production of subsequent millet (6 210 kg ha^{-1}) has been obtained under CZ11-94-5C – millet rotation. Types IGS IV and VI (ORS 3637 and STM 3378 strains respectively) appeared to be responsible for the performance of the Cowpea-millet rotation and cowpea variety CZ11-94-5C.

Keywords: Sudano sahelian zone, Mali, cropping system, millet, cowpea, diversity, Bradyrhizobia.

INTRODUCTION

Cowpea (*Vigna unguiculata* L. Walp) is one of the main pulses crops contributing to nitrogen economy in low input cropping systems (Sanginga et al., 2000). It has been shown that cowpea nodulation in the field could be induced by different rhizobia strains (Gueye, personal communication). Pule-Meulenberg et al. (2010) observed a greater biodiversity of cowpea bradyrhizobia in South Africa in comparison to Botswana and Ghana. Few studies have yet considered the biodiversity of rhizobia and bradyrhizobia of cowpea in Africa (Belane and Dakora, 2009; Steenkamp et al., 2008 Krasova Wade et al., 2014). Law et al. (2007) showed that cowpea rhizobia are more diversified in low rainfall areas. According to Mpepereki and Wollum (1991), cultural practices and plant host can determine the distribution of the rhizobia in a given site. Symbiotic nitrogen fixation resulting from cowpea x native rhizobium symbiosis can reduce the rate of nutrient depletion of cultivated soils where legume-cereal rotation is practiced (Bado, 2002). According to Mpepereki (2002), legumes biological nitrogen fixation can potentially improve the production and food security in Africa. Many authors have shown a diversity of rhizobia in tropical soils (Diouf et al., 2000; Ba et al., 2002). Very little information is known about cowpea rhizobia diversity in the cropping systems of the Sudano Sahelian zone of Mali. Therefore, there was a need to have a thorough knowledge on the diversity of rhizobia nodulating cowpea varieties in the main cropping systems and identify the best cowpea/rhizobium associations to optimize productivity in this area. The objective of this research was to study both cropping system and cowpea variety effect on the diversity of native bradyrhizobia population's nodulating cowpea and on millet yield in Sudano Sahelian zone of Mali.

MATERIAL AND METHODS

Site description and soil characteristics

The experiment was established in 2007 at the Cinzana Agronomic Research Station (SRA/Cinzana) in Mali (13° 15' N 5° 57' W, altitude 281 m). The Cinzana Agronomic Research Station is located between the isohyets 600 mm and 800 mm of rain per year in the Rural Commune of Cinzana in Segou Region, near the Bani River. The climate is Sudano-Sahelian type. Mean annual rainfall over a 30-year period (1972 to 2001) was 670 mm. The rainy season usually begins in June - July and ends in September-October. More than half of the rain usually falls in July-August (Figure 1). The experimentation period (2007, 2008) was characterized by a high inter-annual variability of rainfall.

The average minimum and maximum temperatures are respectively 18° C and 40° C under sun and 18° C and 36° C under shade. Maximum temperatures are recorded from April to May and the minimum from December to February.

The soil is classified as a leached tropical ferruginous soil with spots and concretions (French classification, CPCS, 1969), corresponding to a plinthaquic Haplustalf according to the American classification (Keita et al., 1982). Composite soil samples were collected in May 2007 on the depths 0-20 cm, 20-40 and 40-60 cm before the establishment of the experiment. Soil organic carbon (SOC) content was measured by the Anne method. Mineralization was done using concentrated sulfuric acid (H_2SO_4) with dichromate of potassium ($K_2Cr_2O_7$) and the solution was titrated with iron sulfate ($FeSO_4$). The pH was determined by the electrometric method in a soil solution with a soil/water ratio of 1/2.5. Phosphorus was determined by Bray2 method (solution of HCl + NH 4 F).

The texture of the soil is predominantly sandy in the first 40 cm layer of the soil (Table 1). The upper horizon (0-20 cm) contains only 6% clay. It was chemically a very poor soil. The pH was very strongly acid (4.5) in the first 20 cm layer, extremely acid between 20 and 60 cm deep (3.8 and 4.0). The soil was very poor in organic matter (OM < 0.6%) and deficient in available phosphorus (P < 13 ppm) with a low reserve of total phosphorus.

Plant

Plant material consisted of three (3) improved cowpea (*Vigna unguiculata* L. Walp.) varieties and one pearl millet (*Pennisetum glaucum* L.) variety. The three cowpea varieties were IT89KD-374 (65-70 days of cycle) from IITA (International Institute of Tropical Agriculture), CZ1-94-23-1 (75-80 days), and CZ11-94-5C (65-70 days) from IER (Institut d'Economie Rurale), which is the Natonal Agronomic Research Institute of Mali. These cowpea varieties are semi prostrate and semi determinate. Millet variety used was NKO x Toroniou C1(110 days) also from IER.

Experimental design and treatments

The experimental design was a randomized complete block in a factorial combination of 3 cropping systems and 3 cowpea varieties (9 treatments) with 4 replications. The 3 cropping systems consisted of the millet/cowpea inter cropping in alternate rows (1 row millet/1 row cowpea), the millet/cowpea intercropping in inter-hills (2 hills millet/1 hill cowpea) and cowpea-millet rotation. Treatments were: (T1) millet / IT89KD-374 intercropping (1 row/1 row), (T2) millet / CZ1-94-23-1 intercropping (1 row/1 row), (T3) millet / CZ11-94-5C intercropping (1 row/1 row), (T4) millet / IT89KD-374 intercropping (2 hills/1 hill), (T5) millet / CZ1-94-23-1 intercropping (2 hills/1 hill), (T6) millet / CZ11-94-5C intercropping (2 hills/1 hill), (T7) IT89KD-374–millet rotation, (T8) CZ1-94-23-1- millet rotation and (T9) CZ11-94-5C – millet rotation. Plot size was 7.5 m wide x 8 m long = 60 m^2.

Practical farming operations

Experiment was conducted after previous sesame (*Sesamum indicum* L) established on a three-year fallow history. Soil was prepared by ridging with a tractor at 0.75 m spacing between ridges (10 ridges of 8 m long). Seedlings were made manually on the ridges between 12 and 23 of July. Cowpea in monocropping system was sown on top of ridges by hand at 0.40 m between the hills, or 0.75 m × 0.40 m, then thinned at two plants per hill after crop emergence (66 667 plants ha^{-1}). In the millet/Cowpea intercropping alternating rows (1millet/1cowpea), spacing of the cowpea seedlings was 0.40 m between the hills, or 1.50 m x 0.40 m and thinned at two plants per hill after crop emergence (33 333 plants

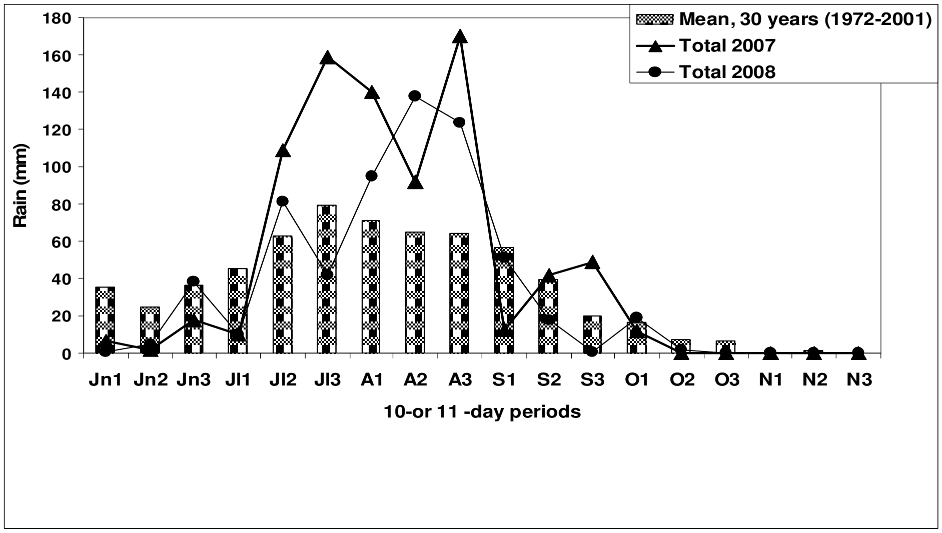


Figure 1. Rainfall (mm) in 10-or 11-day periods at SRA/Cinzana, (1972-2001), 2007 and 2008.

Table 1. Soil characteristics at the beginning of the study at SRA/Cinzana, 2007.

	Depth (cm)		
	0-20	20-40	40-60
Sand	92	81	70
Loam	02	02	11
Clay	06	17	19
Texture	S	SL	LS
pH(H_2O)	4.5	3.8	4.0
Org. carb. (%)	0.04	0.16	0.12
Total N%	0.03	0.01	0.03
Total P (ppm)	64.70	64.70	79.12
P ass. (ppm)	09	02	01

S = sandy; SL = Loamy sand; LS = Sandy loam

ha^{-1}). In inter hills system (2 hills millet/1hill cowpea), cowpea was sown at 1.60 m between the hills of cowpea, or 0.75 m x 1.60 m and thinned at two plants per hill after crop emergence (16 667 plants ha^{-1}). Cowpea was sown at millet stand in the inter hills cropping system (2/1), while it was sown simultaneously with millet in the alternating rows cropping system (1/1). Weedings were done manually as needed using hoe and mounded at the millet growth stage. No fertilizer was applied.

Diversity of natives Bradyrhizobia populations nodulating cowpea

Cowpea nodules collection

Study of diversity focused on native rhizobia in nodules collected on cowpea plants in the different cropping systems. Nodules harvesting was carefully done at cowpea flowering stage by digging up 10 hills of 2 plants (20 plants) in different basic plot points in three replications. All the nodules collected were washed, then dried in shade, and kept in bottles for the characterization of bradyrhizobia strains in the laboratory.

Evaluation of native bradyrhizobia population diversity

Diversity of 225 native nodulating bradyrhizobia was studied by PCR-RFLP of 16S-23S rDNA IGS (Inter Genic spacer Rehion) technic as described by Krasova-Wade and Neyra (2007) using DNA extracted from the collected nodules. Amplification products were digested with two restriction enzymes (*Hae* III and *Msp* I).

Rhizobia strains diversity index calculation

The dominance of the IGS types was divided into four levels (adapted from Zhnag et al. 2004) according to nodulation frequency (NF), i.e. the dominant types (NF >50%), the most common types (30% < NF ≤ 50%), the common types (10% < NF ≤ 30%), and the rare types (NF ≤ 10%).

Specific diversity was estimated using Shannon-Wienner statistical non-parametric index (*H'*) according to Borcard and Buttler (2001).

$H' = 1/N \sum - (\log N_i - \log N)\, N_i$

Where *N* is the total number of individuals of the collection and *Ni* the number of individuals representing the specie *i*. Mixtures of types were not taken into account in the index calculations.

rDNA 16S - 23S sequences analysis

Sequencing was done by COGENICS, the Genomics Services Company Meylan - France (600-900 bases sequence, single-stranded). Sequences alignments obtained from representants of IGS types using a BLAST program with sequences of international data GenBank, allowed identifying closed rhizobia strains.

Evaluation of agronomic parameters of millet

Spikes, grains and straws were harvested, sun dried and weighted, then extrapolated to obtain yield per hectare. Total dry biomass was assessed at harvest by summing spikes weight and sun dried straw weight in the plot.

Statistical analyses

Analyses of variance of all collected data and linear regressions calculations were made using the software MSTATC (Nissen, 1989). Effects of treatments and correlations between parameters were considered significant at the probability threshold of p < 0, 05. Duncan test was used for means separation.

RESULTS

Rainfall distribution graph (Figure1) indicated more rain (909 mm) in 2007 than 2008 (654 mm). In the two years, monthly rainfall was above the 30 years' average. In 2008, the last 10 day period of July and the two last 10 day periods of September were dry, while they were wet in 2007. This rainfall distribution would have some influence on crops growth and yields.

Cowpea native rhizobia diversity

Restriction analysis of 16S-23S rDNA IGS for 225 nodules DNA highlighted six (6) different genetic types of native rhizobia (Table 2) confirmed by sequencing.

Sequences alignments with international data Genbank showed that all the sequenced types belonged to the genus *Bradyrhizobium* (Table 3). Seven (07) *Bradyrhizobia* strains have been identified as similar to *Bradyrhizobium* sp. ORS 3642, *Bradyrhizobium* sp. ORS 3257, *Bradyrhizobium* sp. ORS 3637, *B. liaoningense* SEMIA5062, *Bradyrhizobium* sp. ORS 3636, *Bradyrhizobium* sp. STM 3378, *Bradyrhizobium* sp. STM 3347. The IGS type I and the IGS type III sequences were closely related to those of the ORS 3642 strain of collection from tropical trees plantations in Senegal (Sene et al., 2012) and of the ORS 3257, *Bradyrhizobium sp.* cowpea strain, isolated in Senegal (Krasova-Wade et al., 2003). The IGS types II and IV were similar (93% identity on 97% query sequence) to those of the strains of ORS 3637 strain of tropical trees collection in Senegal and *B. liaoningense* SEMIA5062 of *Glycine* max. Sequence of the IGS type V showed high similarities with those of strains ORS 3642 and ORS 3636 of tropical trees collection and the IGS type VI, to bradyrhizobia STM 3378 and STM3347 isolated from *Zornia* nodules in Senegal.

Cropping systems effects on the native bradyrhizobia populations

According to dominance scale (Table 4), IGS type I was dominant, whatever is the cropping system or cowpea variety. 80% of total analyzed nodules were occupied by the IGS type I, 10% for common IGS type III. The other IGS types were rare because they occupied less than 5% of total analyzed nodules. Proportions of IGS type I were quite variable in cropping systems with varieties IT89KD-374 (58% in intercropping, 1 row millet/1 row cowpea to 96% in intercropping 2 hills millet/1 hill of cowpea or 84% in sole cowpea) and CZ1-94-23-1 (100% in intercropping, 1 row of millet/1 row of cowpea to 75% in intercropping 2 hills millet/1 hill of cowpea and 84% in sole cropping), while they were relatively equilibrated in variety CZ11-94-5C (75% in alternate rows intercropping, 74% in intercropping 2 hills millet/1 hill of cowpea, and 72% in sole cowpea).

In terms of Shannon-Wienner diversity index (H'), diversity of nodulating rhizobia was more important in S1 (millet/cowpea IT89KD-374 alternate rows intercropping; H'=0.39) than in the others treatments (Table 2). In contrast, the lowest diversity (H'= 0 with a single genetic group) was observed (Table 2) in the CZ1-94-23-1 variety grown in S1 cropping system (1 row millet/1 row cowpea).

The six IGS type's profiles identified by 16S-23S rDNA IGS restriction analysis are represented in Figure 2.

Linear regression analyses didn't show any significant correlation between yields of cowpea, of millet and diversity index (H') of rhizobia nodulating the cowpea. Cowpea grain yield (kg ha^{-1}) was y = - 51.5 x + 587; where x = H', r^2 = - 0.02, p = 0.96; millet grain yield (kg ha^{-1}) was y = - 338 x + 1235; where x = H', r^2 = - 0.17 and p = 0.66). The correlation coefficient was r^2 while p was

Table 2. Diversity and nodulation frequency of Bradyrhizobia IGS types strains according to cropping systems and cowpea varieties at SRA/Cinzana, 2007.

Cowpea varieties	Cropping systems								
	S1			S2			S3		
	IGS types	NF (%)	H'	IGS types	NF (%)	H'	IGS types	NF (%)	H'
IT89KD-374	I	58		I	96		I	84	
	II	8		III	4		II	8	
	III	33	0.39	-	-	0.07	III	4	0.26
	-	-		-	-		VI	4	
CZ1-94-23-1	I	100		I	75		I	84	
	-	-		II	5		II	4	
	-	-	0	III	10	0.36	III	8	0.26
	-	-		V	10		IV	4	
CZ11-94-5C	I	75		I	74		I	72	
	III	25		II	4		II	12	
	-	-	0.26	III	13	0.36	IV	16	0.34
	-	-		IV	9			-	

Cropping systems: S1 (Millet/cowpea alternate rows intercropping, 1 row millet/1row cowpea), S2 (Millet/cowpea intercropping, 2 hills millet/1 hill cowpea), S3 (Cowpea sole crop) ;
NF: nodulation frequency; H': Shannon-Wienner diversity index.

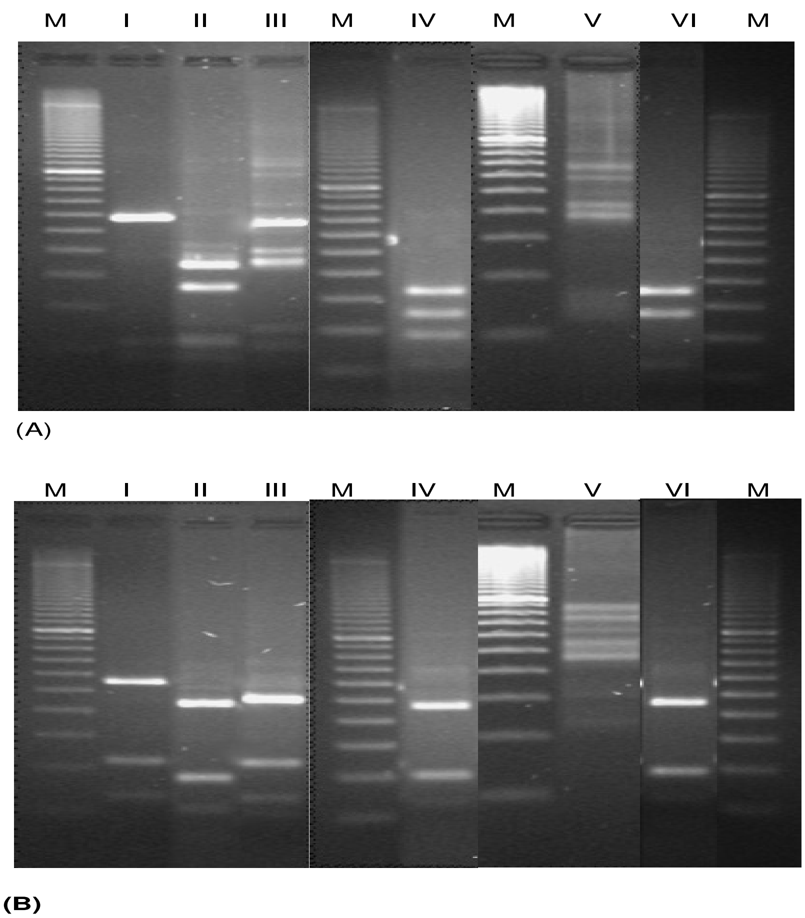


Figure 2: Diversity of 16S - 23s rDNA IGS genetic profiles obtained after PCR-RFLP DNA extracted from cowpea nodules by digestion with Hae III (A) and MspI (B); M, 100 pb, Amersham Biosciences, molecular weight marker.

Table 3. BLAST sequences similarity of the 16S-23S rDNA IGS types of cowpea rhizobia

IGS types	Sequences producing most significant alignments	Length of alignment (bp)	Percentage of frequency in alignment (%)	Identity to closest match (%)	Host plant of rhizobia matched sequence (strain origin and reference)
I	*Bradyrhizobium* sp.ORS 3642	870	100	99	Tropical trees plantations (Sene et al., 2012)
	Bradyrhizobium sp.ORS 3257		100	99	*Vigna unguiculata* (Krasova –Wade et al., 2003
II	*Bradyrhizobium* sp.ORS 3637	964	97	93	Tropical trees plantations (Sene et al., 2012)
	B.liaoningense SEMIA 5062		86	95	*Glycine max* (Menna et al., 2009)
III	*Bradyrhizobium* sp.ORS 3642	870	100	99	Tropical trees plantations (Sene et al., 2012)
	Bradyrhizobium sp.ORS 3257		100	99	*Vigna unguiculata* (Krasova –Wade et al., 2003)
IV	*Bradyrhizobium* sp.ORS 3637	939	97	93	Tropical trees plantations (Sene et al., 2012)
	B.liaoningense SEMIA 5062		89	94	*Glycine max* (Menna et al., 2009)
V	*Bradyrhizobium* sp.ORS 3642	818	100	99	Tropical trees plantations (Sene et al., 2012)
	Bradyrhizobium sp.ORS 3636		100	99	Tropical trees plantations (Sene et al., 2012)
VI	*Bradyrhizobium* sp.STM 3378	912	72	99	*Zornia glochidiata* (Gueye et al., 2009)
	Bradyrhizobium sp.STM 3347		72	99	*Zornia glochidiata* (Gueye et al., 2009)

Table 4. Dominance level of IGS types according to nodulation frequencies

IGS types	NF (%)	Dominance level
I	80.09	D
II	4.63	R
III	10.65	C
IV	3.24	R
V	0.92	R
VI	0.46	R

NF: nodulation frequencies; D: dominant types (NF >50%); MC - most common types (30% < NF ≤ 50%); C: Common types (10% < NF ≤ 30%); R: rare types (NF ≤ 10%).

the probability and H' the Shannon-Wienner diversity index. Any significant correlation was observed between crops (cowpea and millet) yields and nodulation frequencies.

Millet yield

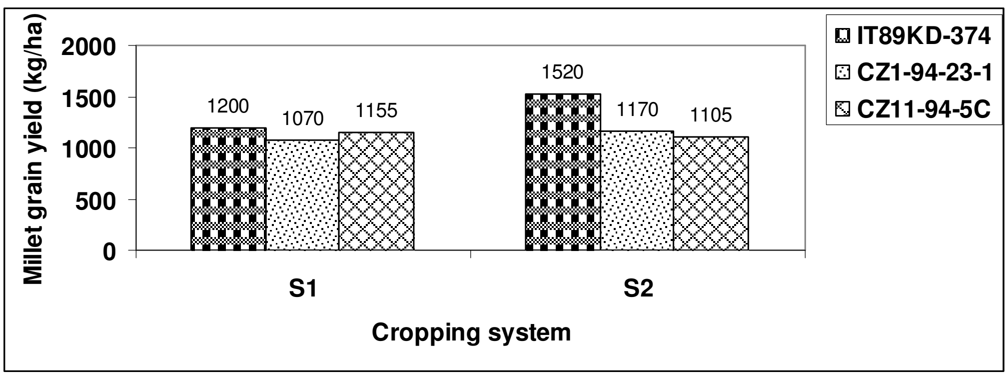
In intercropping system, multiyear analysis of millet grain and above ground total biomass dry yields showed high significant effects of year (p<0. 01). The best yields were observed in 2007 (1400 and 4560 kg ha^{-1}) compared to 2008 (1015 and 3015 kg ha^{-1}). Significant (p = 0.031) and high significant (p = 0.006) interactions were observed between cropping system and cowpea variety, respectively for millet grain and above ground total biomass dry yields. The best yields were obtained under intercropping 2 hills millet / 1 hill of cowpea IT89KD-374 (Figures 3a, 3b). All other treatments were equivalent.

Cropping system and cowpea variety had highly significant effects (p<0. 01) on the performance of the subsequent millet grain yield in favor of cowpea - millet rotation (Table 5).

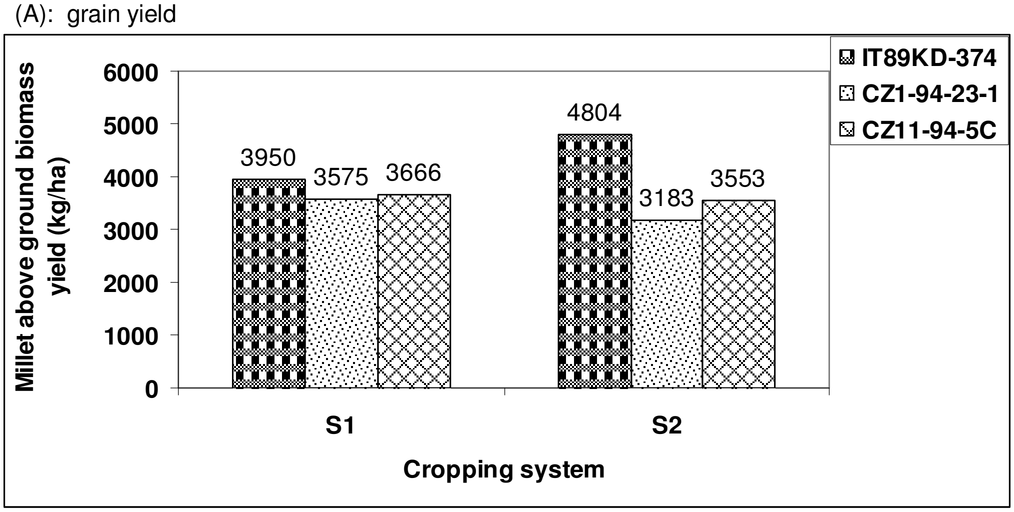
Table 5. Subsequent millet grain and above-ground dry biomass yields (kg ha^{-1}) as affected by cropping system and cowpea variety at Cinzana Agronomic Research Station, 2008.

	Millet yields (kg ha^{-1})	
Treatments	Grain	Above ground dry biomass
Cropping systems		
Millet/cowpea intercropping (1 row/1 row)	995 b	3180
Millet/cowpea intercropping (2 hills/1 hill)	1035 b	2850
Cowpea –millet rotation*	1470 a	5045
Cowpea varieties		
V1: IT89KD-374	1340 a	4050
V2: CZ1-94-23-1	1005 b	3070
V3: CZ11-94-5C	1150 b	3960
Effect	P-values	P-values
Cropping system (S)	< 0.01	< 0.01
Cowpea variety (V)	< 0.01	< 0.01
Cropping system x cowpea variety (S x V)	0.08	< 0.01
Coefficient of variation (%)	19.43	14.301

Means followed by the same letters in the same column for each parameter are not significantly different (P ≤ 0.05), according to Duncan test.
*: Millet yields in the rotation



(A): grain yield



(B): above ground biomass yield

Figure 3. Intercropping system and cowpea variety effects on millet grain (A) and above ground biomass (B) yields (kg/ha) at SRA/Cinzana, 2007 and 2008.
Legend:
S1: Alternate rows intercropping (1row millet/1 row cowpea)
S2: Inter hills inter cropping (2 hills millet/1 hill cowpea)
V1, V2, V3: cowpea varieties (IT89KD-374, CZ1-94-23-1 and CZ11-94-5C)

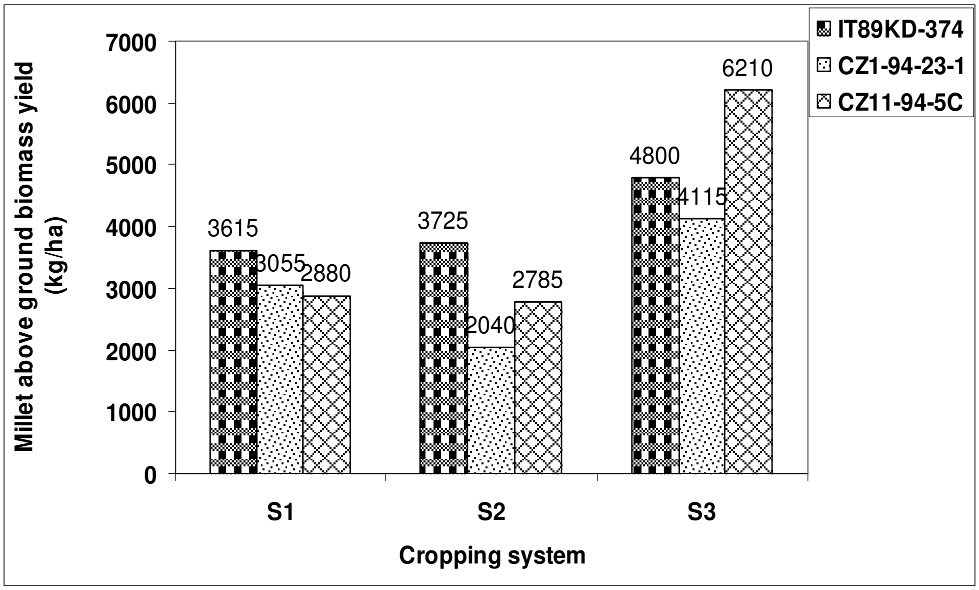


Figure 4. Cropping system and cowpea variety effects on subsequent millet above ground biomass yield (kg/ha) at SRA/Cinzana, 2008.
Legend:
S1: Alternate rows intercropping (1row millet/1 row cowpea)
S2: Inter hills inter cropping (2 hills millet/1 hill cowpea)
S3: Millet in cowpea – millet rotation
V1, V2, V3: cowpea varieties (IT89KD-374, CZ1-94-23-1 and CZ11-94-5C)

Concerning cowpea varieties, the best millet grain yield was obtained with IT89KD-374 (Table 5). The two-way interaction between cropping system x cowpea variety was highly significant (p<0. 01) on the aboveground dry biomass yield of millet. The best yield was obtained under CZ11-94-5C – millet rotation (Figure 4).

DISCUSSION

Diversity of native Bradyrhizobia populations

The results showed a relationship between cowpea variety and diversity of modulating bradyrhizobia strains, and also, between cropping system and bradyrhizobia diversity. These results were consistent with those obtained respectively by Thiès et al. (1992), Palmer and Young (2000). The abundance of nodulation by the IGS type I could be explained by its specificity to nodulate under favorable water conditions, as previously shown in Senegal by Krasova Wade et al. (2014). The nodulation frequency of this dominant IGS type I and the diversity index varied relatively little with the cropping system under cowpea variety CZ11-94-5C; while they varied a lot with the other two varieties (IT89KD-374 and CZ1-94-23-1) depending on the cropping system. These results were in agreement with those given by Palmer and Young (2000), according to which, crop management can affect the competition of indigenous bradyrhizobia for nodule occupancy. The relative constancy of IGS type I nodulation frequency in CZ11-94-5C, and the variability observed in other varieties were consistent with the conclusions of Mpepereki and Wollum (1991). These authors have shown that soil conditions, cultural practices and host can only determine the distribution of rhizobia in a given site and their performance. Results showed that nodulation frequency and diversity index of rhizobia nodulating cowpea were not relevant indicators determining yields of cowpea and the subsequent millet crop. The performance of a cowpea variety or cropping system could be attributed to one or more specific strains associated with a concerned cowpea variety. Pule-Meulenberg et al. (2010) reported that N_2-fixing efficiency varied between bradyrhizobia IGS type's nodulating cowpea in soils of Ghana, Botswana and South Africa. This symbiotic performance was also depending on variety and location. Although based on the cropping systems, the dominance of IGS type I at Cinzana Agronomic Research Station was consistent with results obtained by Krasova-Wade et al. (2003), Kanté (2004) and Kouyaté (2006) and Krasova Wade et al. (2014). It was a very competitive strain in the Sudano-Sahelian zone of Mali, Niger and Senegal under 600-700 mm of rain falls.

Millet yields

Soil low clay content was not favorable to the development and the maintenance of a good structure. Its unfavorable chemical properties such as pH acidity, available low phosphorus content and low organic matter level may restrict crop growth and yields.

The best millet grain and total biomass dry yields observed under cowpea – millet rotation were consistent with the results obtained by different authors (Wani et al., 1995; Bationo and Ntare, 2000; Bado, 2002). These results can be explained by the improvement of millet water and nutrient supply in the cowpea-millet rotation due to a greater section of rooting (2605 cm^2) than those of intercropping millet/cowpea (1298 cm^2 in alternate rows intercropping and 1768 cm^2 in inter-hills millet/cowpea intercropping). These results were in agreement with those obtained by Bertrand and Gigou (2000), who reported that, crop water supply and mineral nutrition will be even better assured as the volume of soil operated by its roots will be more important. In the Sudano-Sahelian region of Mali, with low soil fertility and irregular rainfall, the degree of colonization of the soil by the roots influence the supply of water and nutrients to the plant. Best nodulation observed in 2007 in cowpea sole crop and the CZ11-94- 5C (data not presented) suggested that these treatments fixed more nitrogen, thereby improving subsequent millet yields. It should be recalled that nitrogen is the second nutrient limiting crops performance in the Sudano Sahelian zone of Mali (Doumbia et al., 2009). However, the superiority of CZ11-94-5C – millet rotation compared to CZ1-94-23-1 – millet rotation was not consistent with findings of some authors (Carsky et al., 2003; Stoop and Van Staveren, 1982; Ennin et al., 2002) on the effects of previous pulse cycle (early or late maturity) in the rotation on the subsequent cereal yield performance. In the present study, rhizobia associated with cowpea in different cropping systems might explain the results obtained. One may think that strains associated with genetic groups, IGS types IV and VI were the source of cowpea-millet rotation and CZ11-94-5C performance in the Sudano Sahelian zone of Cinzana. The results confirmed previous work (Kouyaté, 2006). Although the nodulation frequencies of these genetic groups were low (4 to 16% of infected nodules), they seem to be effective in soil improvement in cowpea cropping for subsequent millet crop in the experimental context of the Sudano Sahelian zone of Mali. Differences in subsequent millet yields, between treatments, in favour of cowpea –millet rotation, mainly the previous cowpea variety CZ11-94-5C, can be explained by the presence of specific genetic bradyrhizobia strains whose compatibility with this variety can be more efficient, due to its early abundant and efficient nodulation that depends on the specific strains and cowpea variety as reported by N'diaye et al. (2000), Fall et al. (2003).

CONCLUSION

Results showed that diversity of bradyrhizobium strains depended on cowpea variety and cropping system in the context of this experiment. Strain ORS 3257 was the most representative whatever were the cropping system and cowpea variety. The diversity of bradyrhizobia is more important in variety IT89KD-374 in the intercropping (1 row of millet / 1 row of cowpea) with a diversity index H'= 0.39. The best grains and above ground biomass yields of millet were observed in the intercropping 2 hills of millet/1 hill of cowpea (variety IT89KD-374). Those of subsequent crops millet were obtained under CZ11-94-5C - millet rotation. Some genetic groups IGS (types IV and VI), seemed to be the source of rotation performance. The integration of IT89KD-374 and CZ11-94-5C cowpea varieties, respectively in millet/cowpea intercropping and cowpea-millet rotation may offer an opportunity of production diversification and security under the conditions of this experiment.

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