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Inoculation of *Rhizobiums sp* Strains to Improve Soil Fertility: A Peanut Trial in Covè and Ouessè (Benin)

Alida Adjile[1*], Oscar Didagbe[2,3], Carrel Ayelo[1], Frechno Boko[2], Pascal Houngnandan[2], Roch Mongbo[1]

[1]Laboratoire d'Analyse des Dynamiques Sociales et des Etudes du Développement (LADYD), Faculté des Sciences Agronomiques, Université d'Abomey-Calavi, Abomey-Calavi, Bénin
[2]Laboratoire de Microbiologie des Sols et d'Ecologie Microbienne, Faculté des Sciences Agronomiques, Université d'Abomey-Calavi, Abomey-Calavi, Bénin
[3]Laboratoire de Biologie et de Typage Moléculaire en Microbiologie, Faculté des Sciences et Techniques, Université d'Abomey-Calavi, Abomey-Calavi, Bénin
Email: *adjilealida01@gmail.com

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Abstract

Thought the increasing demand *Arachis hypogaea* L. (groundnut), its yields remain low with increasingly using chemical fertilizers. To reduce the costs for chemical fertilizers inquisition and their long-term toxic effects on soils, microbial bio-fertilizers could be an accessible alternative to peanut farms. Thus, the aim of this study was to assess the performance of rhizobia strains on peanut varieties production. The experiments were conducted in two agro-ecological zones of Benin, in a peasant environment peasant-researcher control or under peasant and researcher control. The experimental device used was a complete random block with nine repetitions and two factors namely inoculation (with *Rhizobium* sp and without *Rhizobium* sp) and mineral fertilizer (with $N_{15}P_{15}K_{15}$ and without $N_{15}P_{15}K_{15}$). The effects of these factors divided into four treatments were evaluated on the plants vegetative, symbiotic and production parameters. In addition, an evaluation of each treatments' comparative advantages was carried out. The results showed that the association *Rhizobium* sp and $N_{15}P_{15}K_{15}$ induced groundnut plants best vegetative and productive parameters. The best comparative advantages in economic terms were also recorded with the same combination (*Rhizobium* sp + $N_{15}P_{15}K_{15}$). Considering the technical performance, the recorded treatments effects can be classified as follows: Control < *Rhizobium* sp < $N_{15}P_{15}K_{15}$ < *Rhizobium* sp + $N_{15}P_{15}K_{15}$. Thus, the association *Rhizobium* sp + $N_{15}P_{15}K_{15}$ induced both the best plants vegetative and productive parameters and the best comparative advantage from an economic point of view. The results also showed that the plants' response to inoculation, the application of manure and their combination was more marked in the bar soil zone (Covè) than in the cotton zone (Ouessè). Considering the negative effects linked to the use of chemical fertilizers, the use of *Rhizobium* sp could be an interesting path to increase the groundnut production.

Keywords

Arachis hypogeae L., *Rhizobium* sp, Bio Fertilizer, Soil Fertility, Benin

1. Introduction

Peanuts are annual oilseed legume often produced in rotation or in association with cereal crops in Benin. However, in last years, the yield of peanut has decreased drastically. According to UDA [1], Benin recorded a peanut yield of 0.93 t/ha against 1.2 t/ha in Nigeria, 1.10 t/ha in Ghana and 1.13 t/ha in Côte d'Ivoire. The world average is 1.5 t/ha and more than 3 t in the United States and China [2]. Although this peanuts production yield decrease is imputed to several factors, the lowering soil fertility appears to be non-negligible. According to Igué *et al.* [3], 68% of the soils in Benin have suffered a severe decrease in its agricultural potential specially the southern and central regions. This is, we according to these authors, to the soil's low nitrogen, phosphorus and potassium content and its cation exchange capacity.

To improve their peanuts production yields then, producers often make use of chemical fertilizers. However, due to their high cost, the use of chemical fertilizers is restricted to a small number of peanuts producers. Although chemical fertilizer seems to improve their production yield, their impact on the environmental is disastrous at long term.

Actually, the success of groundnut cultivation is mainly due to its ability to fix atmospheric nitrogen thanks to the presence of root nodules hosting colonies of *Rhyzobium* sp strains, which allow a slight improvement on the production yield on poor soils, and this, with minimal intervention [4]. The success of this crop therefore requires the establishment of an efficient association between the host plant and the bacteria of the genus *Rhizobium*. Inoculating the peanut with effective rhizobia has been proven a beneficial practice for improving the peanuts productivity [5] [6] in Benin. This practice would also constitute a prospect for promoting a sustainable and environment-friendly farming system.

The present study was initiated, in the same framework, and, conducted in a peasant environment to assess the response of peanut seeds (*Arachis hypogaea*) grown in Ouèsse- and on the Agonli uplands in the south and center of Benin to inoculation with a *rhizobium* and to compare its advantage to other fertilization options in these environments.

2. Material and Methods

2.1. Effects of the Factors Studied and Their Interactions on the Different Parameters

The study was carried out from August 2015 to January 2016 in peasant farms in the sub-districts of Houéhounta (district of Covè) and Gbanlin (district of Ouèssè) (**Figure 1**).

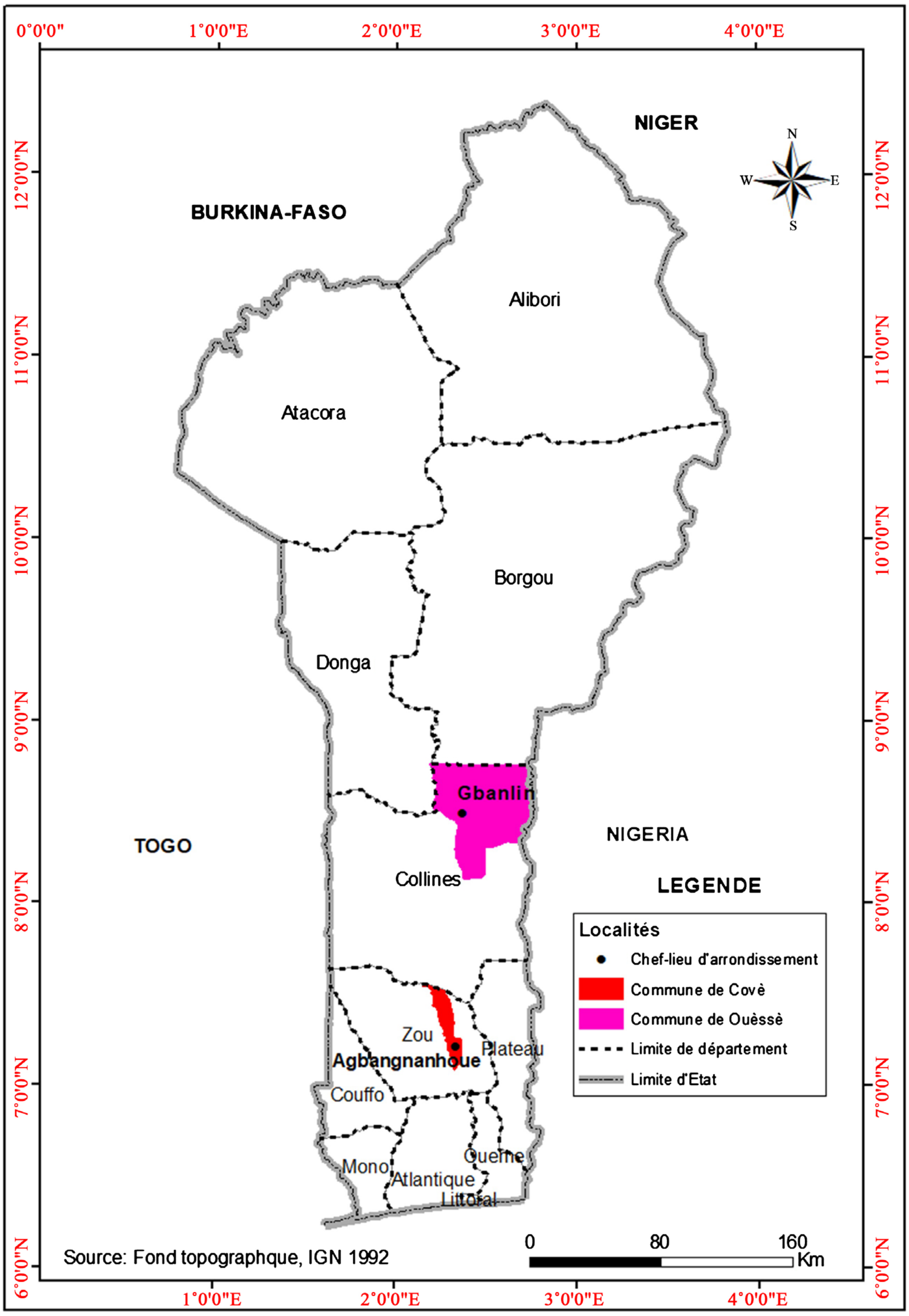

Figure 1. Map indicating the study area.

2.2. Physical Characteristics of the Experiments Sites (Ouèssè and Covè)

Ouèssè is located in the fifth agro-ecological zone of Benin in the heart of the humid tropical zone. It is beneficiary of a tropical climate intermediate between the Guinean climate and the Sudanese climate [7]. The annual rainfall varies between 1100 mm and 1200 mm. It has lands and lowlands appropriate to the good development of agricultural crops and an agricultural diversity estimated at around 1500 km^2 of cultivable land. On the soil level, almost all of Ouèssè is covered with ferruginous soils [8].

Located in the sixth Agro-Ecological Zone of Benin, Covè covers an area of 525 km^2 in the Southern-East of the Department of Zou. It has a Sudan-Guinean or subequatorial type climate with two rainy seasons alternated by two dry seasons. The average rainfall recorded in the area varies between 900 mm to 1100 mm. More than 60% of the district is covered with ferralitic soils characterized by a dominance of iron oxides due to a still incomplete alteration of the primary minerals [9].

The first rainy season which extends from March to July, experienced the rainiest month in March in the two municipalities with a height of rain of 116mm in Covè and 162.3 mm in Ouèssè. A drop in precipitation, more pronounced in Ouèssè than in Covè, immediately followed this moderately wet month. A gradual recovery was observed in May, June and July.

As for the second rainy season, mid-August to November, the period during which the tests were conducted, we note that the month of August was very wet due to the height of the rains recorded in Ouèssè unlike Covè. However, there has been a gradual fall in precipitation during the months of September, October and November in Ouèssè. In Covè, October was an extremely humid month with precipitation ranging to 174.5 mm before declining in November with a transition to the dry season. In sum, for this growing season, a total of 162.6 mm and 218.3 mm of rain was obtained in 3 months (September to November) of rain respectively in Ouessè and Covè.

2.3. Biological and Chemical Materials

The main materials used are: 1) farmer seeds obtained from producers' harvests. 2) Inoculum prepared from strain *Rhizobium* sp (WSM 4412) 3) Phosphorus and nitrogen used as triple superphosphate (46% P_2O_5), urea (46% N) and potassium as potassium chloride (60%). The Laboratory of Soil Microbiology and Microbial Ecology (LMSEM) of the Faculty of Agronomic Sciences of the University of Abomey-Calavi (FSA/UAC) provided the biological material.

2.4. Method of Preparing the Seed Inoculant

2.4.1. Isolation of Bacteria Nodulating Peanut Roots

The collected nodules from the underground part of the peanut plants were sterilized with 8% bleach for 10 min. These nodules were crushed using the sterilized glass rod to obtain a milky suspension of bacteroids. This was streaked on the Yeast Extract Mannitol Agar (YEMA) medium + 0.0025% (w/v) of Red Congo and incubated at 30˚C ± 1˚C for 24 - 48 h [10]. The strains obtained after spreading on culture medium were purified after successive subculturing on YEMA medium [11].

2.4.2. Preparation of the Bacterial Suspension

Bacterial culture was carried out according to the modified method described by Montage and Beunard [12]. Each 250 ml Erlenmeyer flask containing approximately 100 ml of sterile YEM is inoculated using a loop under a host, a bacterial strain stored on YEMA medium contained in a sealed petri dish by Para film. The obtained culture was stirred (200 rpm) at 28˚C for 7 days. The control of the bacterial growth was ensured by samples of an aliquot in the liquid inoculum in preparation according to the dilution method and counting [13], making the gram staining and pH control (the pH is adjusted by aseptically adding 5N KOH).

2.4.3. Inoculation Support and Seed Sterilization

The peat used as a carrier for the inoculant underwent several stages (dried, crushed, sieve and neutralized) of preparation. This support was packaged (30 g per bag) in heat-resistant polypropylene bags (10 cm × 15 cm). The latter underwent three successive sterilizations each 24 hours [14].

The methodology adopted for seed sterilization is an adaptation to that of Ngo Nkot *et al.* [15]. Thus, the peanut seeds have been surface sterilized in 3.3% calcium hypochlorite solution for 3 minutes and washed with sterile distilled water. After sterilization, some seeds were incubated to germinate in petri dishes containing 0.9% (w/v) TYA for 2 to 3 days at 28˚C for viability tests. The appearance of the radicle shows that the seeds selected are of good quality.

2.5. Experimental Design

The system set up in each farmer field is a two factor Complete Random Block with nine (09) repetitions of four (04) treatments on experimental units of 20 m^2 each.

These treatments are distributed randomly as follows: Seeds not inoculated and without fertilizer (T1), seeds with only $N_{15}P_{15}K_{15}$ (T2), seeds only inoculated by *Rhizobium* sp (T3) and seeds inoculated *Rhizobium* sp + $N_{15}P_{15}K_{15}$ (T4).

2.6. Evaluation of Growth and Production Parameters

2.6.1. Vegetative Parameters: Dry Weight Aerial, Root and Total Biomass

At 42 days after sowing, 12 plants were removed from approximately 1 m^2 to assess the effect of the different treatments on aerial and underground growth of plants. They were estimated by weighing after drying in an oven at 65˚C for 72 hours [16].

2.6.2. Symbiotic Parameters: Number and Dry Weight of Nodules

The nodules are removed from the peanut roots at 42 days after sowing. After counting, the dry weight of the nodules was measured after drying in an oven at 65˚C for 72 hours.

2.6.3. Production Parameters: Yield in Full Pods

The yield was evaluated by treatment for each trial on a 9 m^2 unit of production area (kg/ha). The peanut plants were harvested at 90 days after sowing.

2.7. Economic Performance Evaluation

The analysis of the economic performance of the treatments was made from the yield of pods and expenses of production.

2.8. Statistical Analysis

The data were processed with the Excel spreadsheet 2010 version. Statistical Analysis System software version 9.1 was then used for the statistical analysis. One-way analysis of variance has been used to assess the effect of the treatments on the various parameters.

3. Results

In general, the highest values for all the parameters observed are recorded on the plants having received the combined contribution of *Rhizobium* sp and $N_{15}P_{15}K_{15}$. A higher mean value significantly different to plants having benefited from all the other types of treatment regardless of the experimental site has been recorded.

3.1. Effects of the Factors Studied and Their Interactions on the Different Parameters

Table 1 presents the results of the analysis of variance (ANOVA) of the effects of the two variables (treatments and experiment site) and their interaction on the growth and nodulation parameters. These results show very highly significant differences ($p < 0.001$) between the experiment site for the dry weights (DW), the aboveground biomass (AB) and total biomass (TB). It was observed non-significant differences ($p > 0.05$) for the dry weights of the root biomass (RB) and the dry weights of the nodules. On nodulation the difference in effects is highly significant ($p < 0.01$) when considering the actual values of nodules per plant.

Independently to the experiment site, there is a very highly significant difference ($p < 0.001$) in the effects of the treatments on all the growth and nodulation parameters apart. The data on the dry weight of the root biomass (BR) shows a rather significant difference at the 0.05 threshold. The interaction experiment site and treatments reveals non-significant differences effects on the dry weight of root biomass but high different was recorded on the total biomass per plant and very high on the other measured parameters.

3.2. Effects of Treatments on Growth Parameters by Study Area

Table 2 presents the effects of the treatments on the growth parameters by study area. At Covè, the highest dry weight values of aboveground biomass, root biomass and total biomass are recorded with the plants treated with the combination of *Rhizobium* sp and $N_{15}P_{15}K_{15}$. The recorded value were significantly higher with the treatment *Rhizobium* sp and $N_{15}P_{15}K_{15}$ than in comparison with the

Table 1. Analysis of variance of the recorded parameters in the experiment site according to the treatments.

Variant source	Fisher Value						
	Degree of Freedom	DW AB (g/plant)	DW RB (g/plant)	DW TB (g/plant)	Nodules	Nod_tr	DW Nodules
Communes	1	61.35*** <0.0001	2.41 ns 0.13	26.58*** <0.0001	9.37** 0.0048	4.66* 0.0395	0.02 ns 0.8960
treatments	3	37.31*** <0.0001	3.68* 0.02	29.18*** <0.0001	72.09*** <0.0001	94.34*** <0.0001	10.87** <0.0001
experiment site * treatments	3	8.95*** 0.0003	0.16 ns 0.91	5.52** 0.0042	13.63*** <0.0001	14.36*** <0.0001	8.90*** 0.0003

ns: not significant (p > 0.05) *: significant at the threshold of 0.05 (p < 0.05) **: highly significant at the threshold of 0.01 (p < 0.01) ***: very highly significant at the threshold of 0.001 (p < 0.001). DW AB: Dry Weights Aerial Biomass; DW RB: Dry Weights Root Biomass; DW Nodules: Nodules Dry Weights.

Table 2. Effects of the different treatments on growth parameters pf groundnut by study area.

Communes	Treatment	DW AB (g/plant)	DW RB (g/plant)	DW TB (g/plant)
Covè	Control	8.42 ± 0.31b	1.01 ± 0.25b	9.42 ± 0.38b
	$N_{15}P_{15}K_{15}$	9.38 ± 1.36b	1.20 ± 0.25b	10.58 ± 1.30b
	Rhizobium sp	8.93 ± 0.56b	1.27 ± 0.08b	10.21 ± 0.49b
	Rhizobium sp + $N_{15}P_{15}K_{15}$	16.03 ± 0.66a	2.11 ± 0.23a	18.14 ± 0.88a
	Average	**10.69 ± 1.00A**	**1.40 ± 0.14A**	**12.09 ± 1.12A**
Ouessè	Control	6.20 ± 0.21c	1.29 ± 0.25a	7.49 ± 0.16b
	$N_{15}P_{15}K_{15}$	8.13 ± 0.41b	1.86 ± 0.25a	9.99 ± 0.60a
	Rhizobium sp	6.49 ± 0.35c	1.57 ± 0.36a	8.04 ± 0.56b
	Rhizobium sp + $N_{15}P_{15}K_{15}$	9.35 ± 0.58a	2.36 ± 0.41a	11.70 ± 0.95a
	Average	**7.54 ± 0.33B**	**1.77 ± 0.17A**	**9.31 ± 0.46B**

The means followed by the same alphabetical letter and in the same column are not significantly different from each other at the 5% threshold according to the Student Newman-Keuls test.

all other types of treatments.

The same trends were observed in the commune of Ouèssè. However, there was no significant difference between the average dry weight values in root biomass of the four treatments including the control treatment. The average values obtained in Covè are globally higher and significantly different from those obtained in Ouèssè for the dry weights (DW) of the aboveground (AB), root (RB) and total (TB) biomass per peanut plant.

3.3. Evaluation of the Treatments Effects on Symbiotic Parameters

Table 3 presents the effect of the different treatments on the number and dry weight of nodules per plant in Covè and Ouèssè. In the two-experiment site, the highest number of nodules per plant is observed on the plants that received the Combined *Rhizobium* sp and $N_{15}P_{15}K_{15}$ with an average value of Covè higher than that of Ouèssè.

It was observed homogeneity of the treatments effects on the nodules dry weight independently to the experiment site ($p > 0.05$). However, plants treated with $N_{15}P_{15}K_{15}$ than those with the combination *Rhizobium* sp and $N_{15}P_{15}K_{15}$, which present the higher values of the other treatments, in Covè and Ouessè respectively.

Table 3. Effects of treatments on nodulation in each study area.

Experiment site	Treatments	Nodules (number/plant)	Nodules tr	DW Nodules (g/nodule)
Covè	Control	57.33 ± 1.87c	1.76 ± 0.01d	0.23 ± 0.01b
	$N_{15}P_{15}K_{15}$	103.00 ± 1.89b	2.01 ± 0.01b	0.37 ± 0.01a
	Rhizobium sp	71.00 ± 2.65c	1.85 ± 0.01c	0.24 ± 0.01b
	Rhizobium sp + $N_{15}P_{15}K_{15}$	141.33 ± 8.95a	2.15 ± 0.03a	0.27 ± 0.01b
	Average	**93.167 ± 10.10A**	**1.94 ± 0.05A**	**0.28 ± 0.02A**
Ouèssè	Control	63.83 ± 1.87c	1.80 ± 0.01c	0.23 ± 0.01b
	$N_{15}P_{15}K_{15}$	83.67 ± 1.89b	1.92 ± 0.01b	0.27 ± 0.01b
	Rhizobium sp	83.00 ± 1.18b	1.92 ± 0.01b	0.27 ± 0.01b
	Rhizobium sp + $N_{15}P_{15}K_{15}$	104.00 ± 5.87a	2.01 ± 0.02a	0.35 ± 0.03a
	Average	**83.63 ± 3.33B**	**1.91 ± 0.02B**	**0.28 ± 0.01A**

The means followed by the same alphabetical letter and in the same column are not significantly different from each other at the 5% threshold according to the Student-Newman-Keuls test. The actual values of the parameters (in bold) have been transformed on log10 (n).

3.4. Effects of Treatments on Production Parameters

The results of the production parameters from the different treatments are recorded in Table 4. The application of the different treatments generally led to an improvement in the yield of aerial and ground peanuts compared to the controls.

Table 4. Effects of treatments on average haulm and pod yields in the two study areas.

Experiment site	Treatments	Prod Biomass (kg·ha^{-1})	Full Pods (kg·ha^{-1})
Covè	Control	1030.6 ± 68.21b	726.7 ± 36.25c
	$N_{15}P_{15}K_{15}$	1141.2 ± 148.50b	1207.3 ± 91.47b
	Rhizobium sp	1251.7 ± 77.15b	1138.6 ± 39.94b
	Rhizobium sp + $N_{15}P_{15}K_{15}$	1693.5 ± 115.73a	2147.8 ± 120.56a
	Average	**1279.3 ± 85.43A**	**1305.11 ± 160.08A**
Ouèssè	Control	1021.0 ± 68.21c	735.6 ± 36.25c
	$N_{15}P_{15}K_{15}$	1261.4 ± 148.50bc	1155.7 ± 91.47b
	Rhizobium sp	1530.3 ± 168.88b	937.6 ± 38.13bc
	Rhizobium sp + $N_{15}P_{15}K_{15}$	2063.1 ± 137.23a	1388.5 ± 108.13a
	Average	**1468.9 ± 102.72A**	**1054.33 ± 61.79B**

The means followed by the same alphabetical letter and in the same column are not significantly different from each other at the 5% threshold according to the Student Newman-Keuls test.

The highest yields for the two parameters were obtained with the plants treated with the combination of *Rhizobium* sp + $N_{15}P_{15}K_{15}$. The highest average recorded at Covè (2147.8 ± 120.56a kg/ha) compared to Ouèssè (1388.5 ± 108.13 kg/ha) for the yield of solid peanuts.

In the experiment site of Covè, there are no significant differences between the average biomass values of the control plants, those treated with only $N_{15}P_{15}K_{15}$ and inoculated with *Rhizobium* sp. For pod yield full, we note an increase between the average yield from the control plots and those of the plots having received the $N_{15}P_{15}K_{15}$ fertilizer combination, the *Rhizobium* sp strain and the *Rhizobium* sp + $N_{15}P_{15}K_{15}$ combination. There are, however, no significant differences between the average yields obtained with plants amended in $N_{15}P_{15}K_{15}$ (1207.3 ± 91.47 Kg/ha) and those inoculated with *Rhizobium* sp (1138.6 ± 39.94 Kg/ha).

In the experiment site of Ouèssè, the highest yield of withered biomass wars recorded with the treatment *Rhizobium* sp + $N_{15}P_{15}K_{15}$ (2063.1 ± 137. 23 Kg/ha) followed by treatment receiving only *Rhizobium* sp (1530.3 ± 168.88 Kg). The lowest average values was recorded with the control (1021.0 ± 68.21 Kg/ha). In terms of yields of solid peanut pods for this experiment site, there is a positive increase in yields between the control plots and all the treatments with mean values statistically different from each other. The optimal yield is obtained with the combination *Rhizobium* sp + $N_{15}P_{15}K_{15}$ (1388.5 ± 108.13 Kg/ha). Furthermore, the average yield of full pods from inoculated plots, although higher than that of control plots, remains lower than that of plots amended only with $N_{15}P_{15}K_{15}$ fertilizer (Figure 2).

3.5. Economic Evaluation of Treatments

Table 5 below presents the capital gains in kind (Kg) and in money associated with the use of each type of treatment and the balance of capital gains with

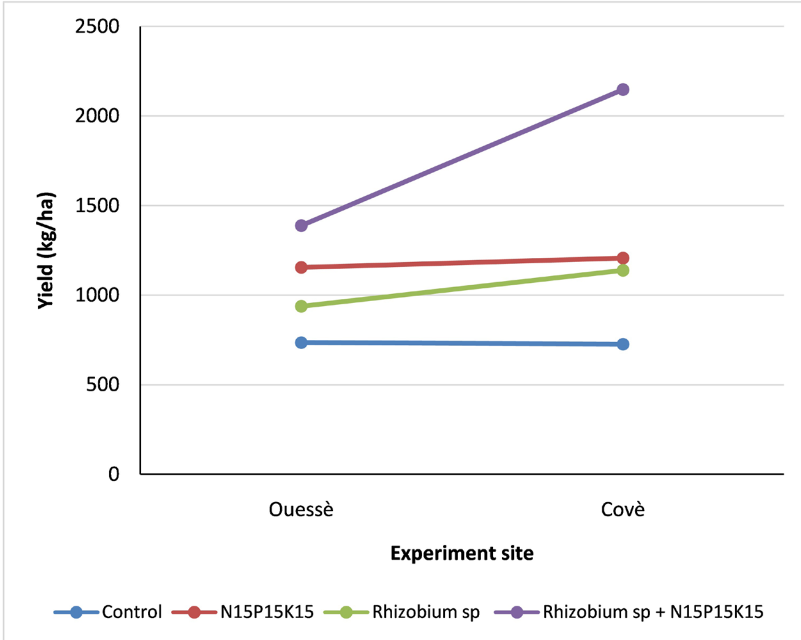

Figure 2. Peanut response on pod yield per treatment at Ouessè and Covè.

Table 5. Comparative advantages of the effect of various treatments on peanut production.

Experiment site	Treatments	Kg/ha of pods	Gain in kg	Gain in FCFA	Average costs of fertilizer /Bio fertilizer	Balance of capital gains	Trend
Covè	Control	726.7(c)	-	-	-	-	-
	$N_{15}P_{15}K_{15}$	1207.3(b)	480.6	149,947.2	78,304.35	71,642.85	Positive
	Rhizobium sp	1138.6(b)	411.9	128,512.8	21,600	106,912.8	Very positive
	Rhizobium sp + $N_{15}P_{15}K_{15}$	2147.8(a)	1421.1	443,383.2	79,904.35	343,478.85	Best
	Average	1305.11A					
Ouessè	Control	735.6(c)	-	-	-	-	-
	$N_{15}P_{15}K_{15}$	1155.7(b)	420.1	131,071.2	78,304.35	52,766.85	Very positive
	Rhizobium sp	937.6(c)	202	63,024	21,600	41,424	Positive
	Rhizobium sp + $N_{15}P_{15}K_{15}$	1388.5	652.9	203,704.8	99,904.35	103,800.45	Best
	Average	1054.33B					

deduction of the charges linked to the acquisition of the different kinds of manures and the spreading of mineral ones.

From the comparative analysis of the values of surplus value in monetary unit (FCFA) and independently of the experiment site, it appears that the use of fertilizer alone is more profitable than the use of *Rhizobium* sp. In addition, the highest values were obtained at the level of the association of the two factors.

However, by integrating the costs of fertilization and the manpower or labour cost for spreading the fertilizer, the use of mineral fertilizer is more costly and therefore presents an economic gain lower than the balance of the application of the inoculum. Therefore, treatment with *Rhizobium* sp would be more cost-effective than the single supply of mineral fertilizer.

In the experiment site of Ouèssè, we observe a trend opposite to that of Covè. The capital gain in FCA and the capital gain balance of the plots amended with mineral fertilizer are higher than those inoculated are. However, the combined contribution of the two treatments (*Rhizobium* sp + $N_{15}P_{15}K_{15}$) would be better regardless of the experiment site.

4. Discussion

The results showed that the nutrients provided by the various treatments have a variable and significant influence on the different parameters evaluated.

From the results of our investigation on the growth parameters (dry weights of aerial biomass, root biomass and total biomass), a homogeneity of effects was observed overall on all the plots except for the plots sowed with seeds inoculated and then amended with NPK fertilizer. This would be due to the growing conditions (rainfall) which, overall, were not favorable to a good plant productivity. The non-significant responses thus obtained especially between the three treatments (T1, T2, T3) are similar to the observations made by Ndiaye [16] on the growth of the plant on which the biomass yield also depends. Thus, "the drought occurred in full flowering period probably caused a slowing down of the vegetative development and mostly on the inoculated plots; which must have reduced the differences in growth between treatments (particularly at Ouèssè)". Indeed, for the flowering-formation of pods phase observed between 30 to 70 days after sowing, Covè seems to have been favored compared to Ouèssè by the drop in rainfall.

The highest values observed for the treatment combining *Rhizobium* sp + $N_{15}P_{15}K_{15}$ are in accordance with the observations made by Didagbé *et al.* [17]. According to their investigation carried in the Center and North Benin, overall, the treatments by *Rhizobium* strains supplemented with phosphorus produced the highest aerial biomass. This would imply the positive response ($p < 0.05$) to the addition of NPK at inoculation observed on the peanut plants growth parameters. These same trends were observed for the symbiotic parameters and the production parameters. In reality, in addition to the phosphorus demand from the host plant, nodules require greater amounts of P and energy than other plant tissues [18]. Phosphorus contributes to root growth [19]; nitrogen being a quality of and growth factor [20] [21] and potassium not only improves the plant's water regime but also increases its tolerance to drought, frost and salinity [20].

Furthermore, the number of nodules observed on controls revealed the presence of *Rhizobium* specific for peanuts in the soil at each site. Indeed this parcel has been used for peanuts crop cultivation. However, the small quantities of spontaneous nodules obtained could be due to the low strains population density of these soils [22].

On the inoculated plots, the formation of nodules and the symbiotic activities in the different peanut populations were therefore carried out with the native strains of *Rhizobium* present in the soil as well as the *Rhizobium* sp strains. The number of nodules formed (transformed value) differ significant to ($p < 0.05$) from the inoculated plants to the control plants. However, the confident interval of these differences is narrower compared to the number of nodules of the plants fertilized with $N_{15}P_{15}K_{15}$ only. This could be due to the competitiveness capacity of the strains. The native competitive but efficient less strain [23] can affect the inoculation of a given legume by a selected rhizobium with high efficiency. Nevertheless, the inoculation were important because the native *Rhizobium* strains would have been infectious but not efficient for the most part. The difference on the number of nodules counted in the control plots and in the inoculated plots is small was statistically different on the two experimental sites and no depressive effect of the inoculation on the nodulation was observed. This would indicate that inoculation has a favorable effect on the nodulation of legumes [16] [24]. Didagbé *et al.* [17] have shown that *Rhizobium* strain is one of the best performing exogenous strains of *Bradyrhiobium* used in their investigation. However, our results on plant nodulation are relatively lower than those previously obtained [17]. The competition between native and introduced strains could be low. As Ndiaye [16] observed on the CB 756 strain, the increases in yield following inoculation seem to indicate that the strain *Rhizobium* sp has a fixing potential greater than the native soil strains, but the competitiveness of the native strains and the poor survival conditions make it (very) difficult to increase the yields.

According to Tombozara [25], the possibility that the introduced rhizobia have not competed with the microorganisms present in the tested parcels must be considered. Other factors such as the quality of the plant material could also have had an impact on the low performance observed. Notwithstanding the probable competitiveness effect between the strains, the analysis of the dry weights of the nodules indicates that the difference between the average values are insignificant between the controls, the $N_{15}P_{15}K_{15}$ and *Rhizobium* sp + $N_{15}P_{15}K_{15}$ at both Covè and Ouèssè. It is therefore possible that the rainfall regime mentioned above contributed to this reduction in the differences of the effects between the treatments. A water deficiency would induce a significant decrease in the number and yield of nodules [26]. This water deficit is the consequence of pockets of drought occurring at the beginning of the plant development cycle and in full bloom. This could partly explain the fact that the results obtained at Covè are better than at Ouessè.

In addition, the physicochemical properties including among others the phosphorus deficiency affect the multiplication of rhizobia in the soil. It has been established that the effectiveness of endomychorizial symbiosis depends on the level of assimilable phosphorus in the soil [19]. For Mandimba and Djondo [27] phosphorus could increase nodulation in peanuts from 57% to 176% and, by extension, the yield. Phosphorus deficit would result in a reduction in the growth of nodules. Thus, the addition of N-P-K fertilizer has considerably improved the efficiency of the Peanut/*Rhizobium* symbiosis. N-P-K fertilization alone had more influence on yields than inoculation alone despite the non-significant difference observed. In accordance with our results and those obtained by other researchers [28] [29] [30], like all crops, legumes have nutrient requirements that, depending on their availability, influence their yields. According to these same authors, even nitrogen fertilizer is necessary to improve the yields of legumes. Therefore, the increases in the yield of full pods obtained indicate that the low dose of nitrogen provided by the mineral fertilizer made it possible to increase the groundnut yields from 26% to 36% [30].

The pod yield is one of the first parameters on which all the changes affecting the plant are reflected [31]. Analysis of the average yields in full pods revealed that the local variety "Monto" responded favorably to inoculation *Rhizobium* sp. This positive effect of inoculation with exogenous *Rhizobium* on the yields of legumes has been demonstrated by several studies on peanuts, soybeans, cowpeas and other legumes [6] [18] and [26].

However, by including production costs, the use of mineral manure costs more than the inoculum. Therefore, the treatment with *Rhizobium* sp would be more profitable than the single contribution of the mineral fertilizer especially at Covè. In addition, the use of this type of fertilization also helps to protect the environment by limiting the pollution by nitrites.

5. Conclusion

The experiment conducted in a peasant environment made it possible to evaluate and compare the response of peasant seed to inoculation with the *Rhizobium* strain. Compared to Ouessè, the results obtained at Covè were significantly better. It was observed that the nitrogen nutrition of the peanut could be ensured by the inoculation of the seeds and mineral fertilizers could provide the nutritive elements in particular urea, triple superphosphate (TSP) and potassium chloride. However, these two simple factors separately induced a significantly slight improvement in the vegetative parameters (growth, nodulation) compared to the control. The effect of the fertilizer factor was more favorable on the symbiotic activity of the peanut than the growth of the plant thanks to the moderate supply of both N, P and K. As for inoculation alone, despite the increase in nodulation, it had beneficial but not very remarkable effects on the production parameters, certainly due to the poor survival conditions of the bacteria introduced. Regarding the performance of the treatments, the vegetative and productive behaviors of the plants were more influenced by the combined effects of inoculum and mineral fertilizer. The best results in terms of comparative advantages and from the economic point of view were obtained by combining the

Rhizobium strain with the manure $N_{15}P_{15}K_{15}$.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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