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SCREENING OF NATIVE ISOLATED RHIZOBIA FROM COWPEA GROUP OF PLANTS AND THEIR NODULATION EFFICIENCY IN GROUNDNUT (ARACHIS HYPOGAEA)

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The present study aimed to rapidly screen all the isolated fifty-two rhizobia (CPGR-1 to CPGR-52) for effective nodulation in groundnut using sterilized vermiculite as a medium for 40 days. Sterilized vermiculite was used for filling in polythene bags. The vermiculite surface was covered with sterile cotton to protect it from aerial contamination. The N-free nutrient solution was applied daily whereas the uninoculated control was supplemented with KNO $_3$.

ABSTRACT

Groundnut seeds were surface sterilized and sown aseptically in plastic polythene bags containing presterilized vermiculite. After seed germination two seedlings per pot were retained then one ml of the broth culture was poured around the seedling soon after germination of the seed with the help of a sterile pipette. Two replications were maintained for each treatment. The control plants were not inoculated. After 40 DAS, the plants were observed for a significantly increased number of effective nodules per plant, effective nodule dry weight (mg/plant), root length (cm), root weight (g/plant), total dry biomass (g/plant), shoot N content (%), root N content (%). Based on nodule number and total plant N content (at 40 DAS), the four best isolates (CPGR-8, CPGR-49, CPGR-33 and CPGR-52) were selected as efficient rhizobial strains which can help in overall plant growth and development sustainably also retaining soil fertility.

Key words: Screening, Cowpea group of plants, Nodulation, Groundnut.

Introduction

Groundnutis cultivated globally for their high oil content and goodseeds, groundnuts are a significant economic legume crop. It is a prized member of the Fabaceae family and has a variety of uses in both industrial and culinary settings. After China, India is the world's second-largest groundnut producer. Tropical and subtropical agroecological zones are among those where groundnut is grown since it grows well in warm climates. Though sandy loam soils with good drainage are ideal for its growth, this crop is adaptable and may thrive in a variety of soil types. Depending on the cultivar and environmental factors, groundnuts develop in both determinate and indeterminate ways, taking different amounts of time to mature (Nayak *et al.*, 2021).

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Groundnut seeds are so high in protein, unsaturated fats, vitamins and minerals, that they are important for

both agricultural economics and global food security. The importance of groundnuts as a staple food, especially for vegetarian and vegan diets is largely due to their high protein content(Arya *et al.*, 2016).

Like other legumes, groundnut forms symbiotic relationships with nitrogen-fixing bacteria called rhizobia, which inhabit specialized root structures known as nodules. This symbiosis allows groundnut plants to convert atmospheric nitrogen into a form that can be utilized for plant growth, thereby reducing the reliance on synthetic nitrogen fertilizers and promoting soil health.Rhizobia inoculation in groundnutis plant growth-promoting rhizobacteria (PGPR) offers numerous benefits for improving soil quality and crop nutrient uptake (Palai *et al.*, 2021).

Legumes are being introduced to previously uncultivated areas to enrich the soil with plant nutrients, particularly nitrogen, due to their well-established ability to boost soil fertility. Given this, the current study uses the effective rhizobia from the cowpea group of plants as microbial inoculants to increase legume production and reduce the need for chemical fertilizers by the plants. Additionally, these rhizobia were screened for effective nodulation in groundnuts.

Materials and Methods

The present study was conducted in the Department of Agricultural Microbiology at the University of Agricultural Sciences, Dharwad from 2020 to 2021. Isolated fifty-two native rhizobia from the cowpea group of plants (*viz.*, Groundnut, cowpea and pigeonpea) were taken and used in this study. So the present study aimed to rapidly screenall the fifty-two rhizobial isolates for effective nodulation in groundnut using sterilized vermiculite as a medium. Sterilized vermiculite was used for filling in polythene bags. This was a modification to that of Nambiar and Dart (1980) experiment, where they used sand and vermiculite in a ratio of 2:1.

Preparation of assembly

Plastic polythene bags (30×16 cm) used for the test were punched to provide proper drainage. These bags were washed with 2 percent formaldehyde and filled aseptically with 750 g of sterilized vermiculite. The vermiculite surface was covered with sterile cotton to protect it from aerial contamination. The N-free nutrient solution (Jensen, 1942) was applied every day whereas, the uninoculated control was supplemented with KNO₃.

Sowing of seeds

Undamaged clean uniform sized seeds of groundnut were surface sterilized. Two surface sterilized seeds were sown aseptically in plastic polythene bags containing presterilized vermiculite, watered with sterile distilled water and N-free plant nutrient solution alternately from a wash bottle. While uninoculated control was watered with plant nutrient solution with KNO₃. After seed germination two seedlings per pot were retained. The vermiculite surface was covered with a 1-2 cm deep layer of sterile cotton to prevent fall of dust particles from the air.

Inoculation

Log phase culture of the *Rhizobium* isolates grown on YEMA slopes was inoculated into sterile YEM broth and incubated for 2-3 days. One ml of the broth culture was poured around the seedling soon after germination of the seed with the help of a sterile pipette. Two replications were maintained for each treatment. The control plants were not inoculated.

Effective nodulation

After 40 DAS, the plants were observed for number of effective nodules per plant, effective nodule dry weight (mg/plant), root length (cm), root weight (g/plant), total dry biomass (g/plant), shoot N content (%), root N content (%). Based on nodule number and total plant N content (at 40 DAS) four efficient isolates were selected as efficient rhizobial strains.

Results

All fifty-two rhizobia were rapidly screened for nodulation of groundnut in polythene bag using vermiculite-based carrier material under greenhouse conditions for 40 days and the results are presented in Table 1 and Plate 1.

Inoculation of plants with different rhizobial isolates improved the root length of groundnut. A significant difference was observed in root length by inoculation of rhizobial isolates. The highest root length was recorded with isolate CPGR-8 and CPGR-49 which recorded 47.00 and 46.50 cm respectively, which were statistically on par with each other. This was followed by the isolate CPGR-33 (45.00 cm) and CPGR-52 (43.50 cm), which were on par with each other. However, the reference strain (NC-92) which showed a root length of 37.50 cm was on par with isolates CPGR-41 (39.50 cm) and CPGR-36 (37.50 cm). Significantly the lowest root length was recorded in the uninoculated control (19.00 cm).

Root dry weight of groundnut inoculated with different rhizobial isolates ranged from 0.28 to 0.69 g/plant. A significant difference was observed in root dry weight due to inoculation of rhizobial isolates. The highest root dry weight was recorded with isolate CPGR-8 (0.69 g/plant) which was followed by CPGR-49 (0.65 g/plant), CPGR-33 (0.62 g/plant), CPGR-52 (0.60 g/plant) and reference strain NC-92 (0.51 g/plant) all of these were

Table 1 : Screening of native rhizobiafor effective nodulation in groundnut (40 DAS).

S.	Isolate	Root	Root dry	No. of	Nodule dry	Total dry	Root N	Shoot N
no.	code	length	weight	nodules	weight (mg/plant)	Biomass (g/plant)	content	content
		(cm)	(g/plant)	per plant	(mg/plant)	(g/plant)	(%)	(%)
1	CPGR-1	28.00st	0.47 ^{a-e}	9.50 [⊩] n	12.00 ^t	0.98^{j-1}	1.16 ^{n-q}	2.15 ^{jk}
2	CPGR-2	31.50о-г	0.53 ^{a-e}	10.50 ^{j-1}	13.50 ^r	1.09 ^{f-h}	1.23 ^{k-n}	2.23 ^{g-i}
3	CPGR-3	33.00 ^{m-p}	0.55ª-e	13.50 ^{fg}	18.00 ^{hi}	1.12 ^{e-g}	1.30 ^{f-k}	2.36 ^{b-е}
4	CPGR-4	26.50 ^t	0.43ª-e	8.00 ^{op}	12.00 ^t	0.81°-r	1.22 ^{Lp}	2.25 ^{f-h}
5	CPGR-5	33.00 ^{m-p}	0.55 ^{a-e}	12.00 ^{hi}	19.50°	1.12 ^{e-g}	1.33 ^{e-i}	2.32 ^{ef}
6	CPGR-6	27.00 ^t	0.47 ^{a-e}	7.00 ^p	12.00 ^t	0.99 ⁱ⁻¹	1.05st	2.02 ^{n-q}
7	CPGR-7	32.50 ^{n-q}	0.53 ^{a-e}	12.00 ^{hi}	17.50 ^{h-j}	1.04 ^{h-k}	1.29 ^{f-l}	2.34 ^{de}
8	CPGR-8	47.00ª	0.69ª	19.50 ^a	24.00 ^a	1.35a	1.52ª	2.46a
9	CPGR-9	27.00 ^t	0.41 ^{a-e}	11.00 ^{i-k}	16.00 ^{k-m}	0.74 st	1.23 ^{k-o}	2.35с-е
10	CPGR-10	29.50 ^{rs}	0.50^{ae}	10.50 ^{jkl}	19.00 ^f	0.84 ^{n-r}	1.31 ^{f-j}	2.25 ^{f-h}
11	CPGR-11	34.50 ^{k-n}	0.45 ^{a-e}	14.00 ^{ef}	18.5 ^g	0.11 ^u	1.36 ^{c-f}	2.40 ^{a-d}
12	CPGR-12	33.50 ¹⁻⁰	0.47 ^{a-e}	10.50^{jkl}	18.00 ^h	$1.07^{\text{f-h}}$	1.30 ^{f-k}	2.42a-c
13	CPGR-13	32.50 ^{n-q}	0.50 ^{a-e}	12.50gh	14.00 ^q	0.99^{i-1}	1.23 ^{k-n}	2.26 ^{f-h}
14	CPGR-14	35.00 ^{j-m}	0.57^{a-d}	12.50gh	21.00°	$1.09^{\mathrm{f-h}}$	1.40 ^{с-е}	2.40 ^{a-d}
15	CPGR-15	31.50о-г	0.58 ^{a-d}	10.50 ^{jkl}	14.00 ^q	1.04 ^{h-k}	1.12 ^{qr}	2.09 ^{k-n}
16	CPGR-16	34.50 ^{k-n}	0.54 ^{a-e}	11.00 ^{ijk}	14.50°	0.97 ^{kl}	1.16 ^{n-q}	2.26 ^{f-h}
17	CPGR-17	35.00 ^{j-m}	0.55 a-e	12.50gh	10.00 ^w	1.28 ^b	0.94 ^u	1.98 ^{pq}
18	CPGR-18	34.50 ^{k-n}	0.52 a-e	15.00 ^{de}	15.00°	1.17 ^{de}	1.17 ^{m-q}	2.06 ^{l-o}
19	CPGR-19	31.00 ^{p-r}	0.51 a-e	11.50 ^{h-j}	19.00 ^f	1.07 ^{f-h}	1.26 ⁱ⁻¹	2.30 ^{e-g}
20	CPGR-20	33.00 ^{m-p}	0.35 ^{с-е}	11.50 ^{h-j}	17.50 ^{hj}	0.88 ^{no}	1.24 ^{j-m}	2.19 ^{h-j}
21	CPGR-21	30.50 ^{qr}	0.49 ^{a-e}	13.50 ^{fg}	18.00hi	0.90 ^{mn}	1.30 ^{f-k}	2.35с-е
22	CPGR-22	38.00 ^{e-h}	0.53 a-e	8.50 ^{no}	21.50 ^b	0.88 ^{n-p}	1.36 ^{d-h}	2.25 ^{f-h}
23	CPGR-23	39.00 ^{e-g}	0.43 ^{a-e}	9.50 ^{l-n}	16.00 ^{k-m}	0.87 ^{n-q}	1.22 ^{l-p}	2.32 ^{ef}
24	CPGR-24	31.50о-г	0.46 ^{a-e}	9.00 ^{m-o}	10.50°	1.05 ^{g-j}	1.03 ^t	2.01°-q
25	CPGR-25	34.50 ^{k-n}	0.45 ^{a-e}	13.50 ^{fg}	16.50 ^k	1.07 ^{f-h}	1.26 ⁱ⁻¹	2.12 ^{j-m}
26	CPGR-26	37.00 ^{g-j}	0.48 ^{a-e}	12.50gh	18.00 ^{hi}	1.05 ^{g-j}	1.26 ⁱ⁻¹	2.25 ^{f-h}
27	CPGR-27	20.50 ^u	0.31 ^{de}	10.00 ^{k-m}	14.00 ^q	1.13 ^{d-f}	1.15 ^{pq}	2.26 ^{f-h}
28	CPGR-28	31.50о-г	0.49 ^{a-e}	12.50gh	16.00 ^{k-m}	0.88 ^{no}	1.16 ^{n-q}	2.17 ^{ij}
29	CPGR-29	28.00st	0.47 ^{a-e}	8.00 ^{op}	13.50 ^r	0.98^{j-1}	1.10 ^{q-t}	2.09 ^{k-n}
30	CPGR-30	34.50 ^{k-n}	0.51 ^{a-e}	9.00 ^{m-o}	11.50 ^u	1.03 ^{h-1}	1.13 ^{qr}	2.16 ^{i-k}
31	CPGR-31	31.50о-г	0.43 ^{a-e}	11.00 ^{ijk}	15.50 ⁿ	1.09 ^{f-h}	1.15 ^{pq}	2.14 ^{jk}
32	CPGR-32	26.00 ^t	0.44 ^{a-e}	9.50 ^{l-n}	11.50 ^u	1.08 ^{f-h}	1.16 ^{n-q}	2.26 ^{f-h}
33	CPGR-33	45.00 ^{ab}	0.62 ^{a-c}	16.50 ^{bc}	20.50 ^d	1.27 ^b	1.43 ^{bc}	2.44a
34	CPGR-34	36.50 ^{h-k}	0.43 a-e	12.50gh	15.00°	0.80rs	1.17 ^{m-q}	2.16 ^{i-k}
35	CPGR-35	27.50 st	0.40 ^{b-e}	10.50 ^{j-1}	13.50 ^r	0.98 ^{j-1}	1.15 ^{pq}	2.15 ^{jk}
36	CPGR-36	37.50 ^{f-i}	0.52 ^{a-e}	13.50 ^{fg}	17.50 ^{h-j}	1.18 ^{de}	1.26 ⁱ⁻¹	2.23 ^{g-i}
37	CPGR-37	35.50 ^{i-l}	0.53 ^{a-e}	10.00 ^{k-m}	16.50 ^{kl}	1.03 ^{h-l}	1.29 ^{f-l}	2.31 ^{ef}
38	CPGR-38	37.00 ^{g-j}	0.54 ^{a-e}	10.50 ^{j-1}	15.20 ^{no}	1.07 ^{f-h}	1.12 ^{q-s}	2.06 ^{l-o}
39	CPGR-39	41.50 ^{cd}	0.52 ^{a-e}	11.00 ^{i-k}	14.50°	1.25 ^{bc}	1.13 ^{qr}	1.98 ^{pq}
40	CPGR-40	42.50°	0.56ª-e	10.00 ^{k-m}	16.00 ^{k-m}	1.17 ^{de}	1.11 ^{q-s}	2.05 ^{m-p}
41	CPGR-41	39.50 ^{d-f}	0.50 ^{a-e}	12.00 ^{hi}	19.00 ^f	1.07 ^{f-h}	1.26 ⁱ⁻¹	2.31 ^{ef}
42	CPGR-42	26.00 ^t	0.40 ^{b-e}	9.50 [⊩] n	14.00 ^q	1.12 ^{d-g}	1.15 ^{pq}	2.19 ^{h-j}
43	CPGR-43	40.00 ^{de}	0.42 ^{a-e}	11.50 ^{h-j}	17.75 ^{h-j}	1.06 ^{f-i}	1.29 ^{f-1}	2.26 ^{f-h}

Table 1 continued....

Table 1 continued....

S. no.	Isolate code	Root length (cm)	Root dry weight (g/plant)	No. of nodules per plant	Nodule dry weight (mg/plant)	Total dry Biomass (g/plant)	Root N content (%)	Shoot N content (%)
44	CPGR-44	38.75 ^{e-h}	0.37 ^{b-e}	8.00 ^{op}	13.25 ^{rs}	1.05 ^{f-j}	1.16 ^{n-q}	2.12 ^{j-1}
45	CPGR-45	32.50 ^{n-q}	0.46 ^{a-e}	9.50 ^{l-n}	15.50 ⁿ	1.06 ^{f-i}	1.10 ^{q-t}	2.04 ^{n-q}
46	CPGR-46	36.50 ^{h-k}	0.53 ^{a-e}	10.00 ^{k-m}	13.00°	0.99 ⁱ⁻¹	1.26 ^{i-l}	2.15 ^{jk}
47	CPGR-47	29.50 ^{rs}	0.43 ^{a-e}	7.00 ^p	15.50 ⁿ	1.07 ^{f-h}	1.10 ^{q-s}	2.25 ^{f-h}
48	CPGR-48	30.50 ^{qr}	0.45 ^{a-e}	11.00 ^{i-k}	13.00°	0.96 ^{k-m}	1.05st	1.98 ^{pq}
49	CPGR-49	46.50 ^a	0.65 ^{ab}	17.50 ^b	24.00a	1.29 ^{ab}	1.47 ^{ab}	2.44a
50	CPGR-50	38.50 ^{e-h}	0.44 ^{a-e}	15.50 ^{cd}	16.50 ^{kl}	0.96 ^{lm}	1.06 ^{r-t}	2.15 ^{jk}
51	CPGR-51	29.50 ^{rs}	0.47 ^{a-e}	11.50 ^{h-j}	9.50 ^x	0.88 ^{no}	0.96 ^u	1.97 ^q
52	CPGR-52	43.50 ^{bc}	0.60 ^{a-c}	15.50 ^{cd}	21.5 ^b	1.26 ^{bc}	1.40 ^{cd}	2.42 ^{ab}
53	NC-92	37.50 ^{f-i}	0.51 ^{a-e}	13.50 ^{fg}	16.50 ^{kl}	1.19 ^{cd}	1.36 ^{с-д}	2.23 ^{g-i}
54	UIC	19.00 ^u	0.28 ^e	$O_{\rm d}$	Oa	0.70 ^t	0.86 ^v	1.70 ^r
	S. Em. ±	0.720	0.010	0.385	0.484	0.025	0.016	0.017
	C.D.(p=0.01)	2.040	0.020	1.094	1.375	0.072	0.046	0.047

Note: Mean values followed by the same letter are not significantly different based on DMRT (P<0.01), a>b>c.



Plate 1: Rapid screening of native rhizobia on root length, nodulation and nitrogen content in groundnut (40 DAS).

on par with each other. The lowest root dry weight was recorded in the uninoculated control with a value of 0.28 g/plant.

The nodule number of plants inoculated with selected rhizobial isolates was found to vary significantly. Significantly the highest nodule number per plant was observed in the plants inoculated with the isolate CPGR-8 with 19.50 which was followed by CPGR-49 with 17.50 nodules per plant. However, the isolates CPGR-33, CPGR-52 and reference strain (NC-92) showed nodule numbers of 16.50, 15.50 and 13.50 respectively and were statistically on par with each other. Significantly the lowest nodule number was noticed with the isolate CPGR-47 (7

nodules per plant) and the control was devoid of nodules.

A significant difference was observed in nodule dry weight due to inoculation of rhizobia. The highest nodule dry weight was recorded with isolates CPGR-8 and CPGR-49 which showed values of 24.00 mg/plant each. This was followed by CPGR-52 (21.50 mg/plant), CPGR-33 (20.50 mg/plant) and reference strain NC-92 (16.50 mg/plant) which differed significantly with each other. The lowest nodule dry weight was recorded in plants inoculated with isolate CPGR-51 (9.50 mg/plant).

Inoculation of plants with different rhizobial isolates on total dry biomass of groundnut ranged from 0.7 g/plant to 1.35 g/plant. A significant difference was

observed in total dry biomass due to inoculation of rhizobia. The highest total dry biomass was recorded with isolates CPGR-8 (1.35 g/plant). This was followed by the isolate CPGR-49 (1.29 g/plant) and CPGR-33 (1.27 g/pant). However, the treatment CPGR-49, CPGR-33, CPGR-52 and reference strain (NC-92) showed 1.29, 1.27, 1.26 and 1.19 g/plant which were on par with each other. The lowest total dry biomass was recorded in uninoculated control (0.70 g/plant).

The nitrogen content in the root ranged between 0.86 to 1.52% as influenced by inoculation of native rhizobial isolates. The maximum nitrogen content of the root was recorded with the isolate CPGR-8 at 1.52%, which was

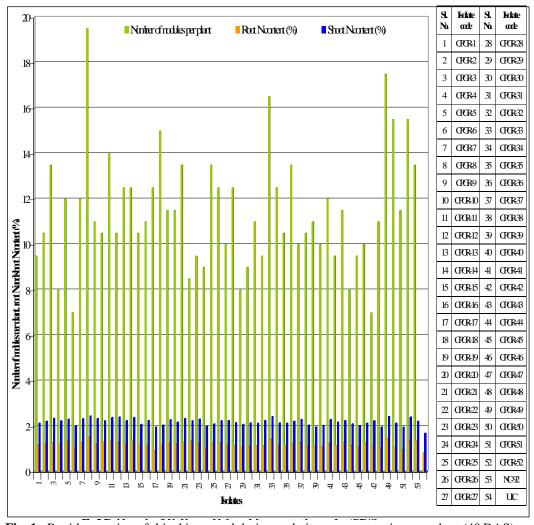


Fig. 1: Rapid screening of rhizobia on nodulation and nitrogen content in groundnut (40 DAS).

followed by the isolates CPGR-49, CPGR-33, CPGR-52 and reference strain (NC-92) with 1.47, 1.43, 1.40 and 1.36% respectively, which were statistically on par with each other. Significantly the lowest root nitrogen content was recorded in uninoculated control (0.86%).

The shoot N content of groundnut inoculated with different rhizobial isolates ranged from 1.70 to 2.46%. A significant difference was observed in shoot N content by inoculation of rhizobia. The highest shoot N content was recorded with isolate CPGR-8, CPGR-33 and CPGR-49 which showed 2.46, 2.44 and 2.44%, followed by the isolate CPGR-52 and reference isolate NC-92 of 2.42 and 2.23% respectively, which were on par to each other. Significantly the lowest shoot N content was recorded in uninoculated control of 1.70%.

Discussion

Rhizobia are Gram-negative soil bacteria that fix nitrogen from the atmosphere. To exploit the benefit of rhizobia for increased agriculture production, the current work focused on screening native rhizobia after isolation and characterization of rhizobial isolates from root nodules of the cowpea group of plants (*viz.*, Groundnut cowpea and pigeonpea), which can enhance nodulation in groundnut and promote plant growth, N content and ultimately yield. The discussion is dealt with as follows.

All the fifty-two rhizobia were rapidly screened for nodulation in groundnut under greenhouse conditions for 40 days. The isolates CPGR-8, CPGR-33, CPGR-49, CPGR-52 and reference strain were able to produce maximum root length, total dry biomass, shoot and root N content, root nodules and nodule dry weight as compared to uninoculated plants (40 DAS). The results revealed that, isolates CPGR-8, CPGR-33, CPGR-49 and CPGR-52 were significantly superior over other isolates including reference strain (NC-92) and uninoculated control. The efficiency rating was done based on nodulation, plant biomass and N content in shoot and root.

The inoculation of rhizobia to groundnut plants significantly increased the nodule number at 40 DAS. The significantly highest number of nodules was recorded with isolate CPGR-8 (19.50 nodules per plant), which

was followed by CPGR-49 (17.50 nodules per plant) and isolates CPGR-33, CPGR-52 and reference strain NC-92 (16.50, 15.50 and 13.50 nodules per plant, respectively) were statistically on par to each other (Fig. 3). These results are in agreement with findings of Hadad and Loynachan (1985), who carried out nodulation test for six *Rhizobium* strains isolated from groundnut (*Arachis hypogaea*), mung bean (*Vigna radiata*), lubia (*Dolichos lablab*), cowpea (*Vigna unguiculata*), pigeonpea (*Cajanus cajana*) and bambara groundnut (*Voandzeia subterranean*). All the isolates were able to nodulate each of the six legumes when grown in sterile vermiculite.

A significant difference was observed in nodule dry weight by inoculation of rhizobia. The highest nodule dry weight was recorded with isolate CPGR-8 and CPGR-49 (24 mg per plant), which was followed by the isolate CPGR-52, CPGR-33 and reference strain NC-92 (21.50, 20.50 and 16.50 mg per plant), which differed significantly with each other. These results are in line with the findings of Tsegaye *et al.* (2015), who screened *Rhizobium* isolates for infectivity and efficiency in fenugreek. Plants were uprooted after 45 days of planting and observed for increased nodule number, nodule dry weight and the symbiotic effectiveness of inoculated rhizobial isolates.

Inoculation of plants with different rhizobial isolates on total dry biomass of groundnut ranged from 0.70 to 1.35 g per plant. The significantly highest total dry biomass was recorded with isolates CPGR-8 and CPGR-49 (1.35 and 1.29 g per plant), which were on par with each other, followed by the isolate CPGR-33 (1.27 g per plant). However, the isolates CPGR-49, CPGR-52 and reference strain NC-92 (1.29, 1.26 and 1.19g per plant) were on par with each other. The results followedLaurette *et al.* (2015), who reported that rhizobial inoculation in bambra groundnut increased the total dry biomass of the plant.

The root N content of groundnut inoculated with different rhizobial isolates differed significantly which ranged from 0.86 to 1.52%. The significantly highest root N content was recorded with isolate CPGR-8 (1.52%), which was followed by CPGR-49, CPGR-33, CPGR-52 and reference strain NC-92 (1.47, 1.43, 1.4 and 1.36% respectively), which were on par to each other (Fig. 3). These results are well followingNambiar and Dart (1980) who screened different rhizobial isolates of groundnut by using modified sterile sand culture method and found that inoculation of rhizobia increased root N content.

The shoot N content of groundnut inoculated with different rhizobia significantly differed which ranged from 1.70 to 2.46%. Significantly highest shoot N content was recorded with isolate CPGR-8, CPGR-33 and CPGR-49 (2.46, 2.44 and 2.44%, respectively), which was followed by isolate CPGR-52 and reference isolate NC-92 (2.42

and 2.23% respectively) which were on par to each other (Fig. 1). The results are in agreement with the findings of Fentahun *et al.* (2013), who carried out nodulation test for 8 rhizobial isolates obtained from Haricot bean, where inoculation of rhizobia increased shoot and root nitrogen content compared to uninoculated control.

The findings of the rapid screening studies showed that native isolates *viz.*, CPGR-8, CPGR-33, CPGR-49 and CPGR-52 performed well in terms of their efficiency in fixing atmospheric nitrogen as revealed by nodulation, nitrogen content of shoot and root and plant biomass.

Conclusion

The repeated and injudicious application of chemical fertilizers has led to the loss of soil fertility and productivity by disturbing microbial diversity. Nitrogen is one of the essential macro-nutrients required for the growth of groundnut. Generally, groundnut requires a huge amount of nitrogenous fertilizers and it can enter into a symbiotic relationship with nitrogen-fixing rhizobia. Improving the nodulation and nitrogen fixation processes are seen as pivotal steps towards enhancing agricultural sustainability, soil biodiversity, nutrient recycling, ecosystem and even food security. Therefore, rhizobia could be used as low-cost eco-friendly input for growth promotion in legumes. In this perspective, the study was carried screen isolated native rhizobia from the root nodules of groundnut, cowpea and pigeonpea plants.

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