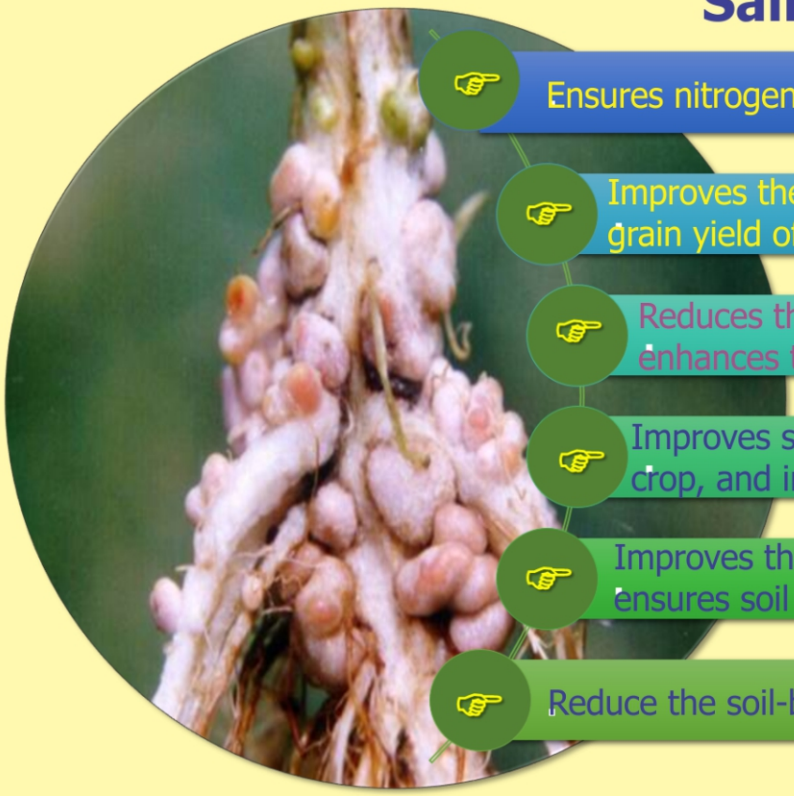
Technical Bulletin

Rhizobium Biofertilizer

for Tamil Nadu Pulses

Salient Features



- Ensures nitrogen requirement of the pulses.
- Improves the root and shoot length, biomass, and grain yield of legumes.
- Reduces the chemical fertilizer usage and thereby enhances the farm profit.
- Improves soil nitrogen through green manure, cover crop, and intercrop and legume crop rotation.
- Improves the soil microbial activity and thereby ensures soil health.
- Reduce the soil-borne pathogens of legume crops.

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Introduction

Rhizobium is a symbiotic bacterium that fixes atmospheric nitrogen in root nodules of plants and has a mutually beneficial relationship with their host plant. The Rhizobia comprises five genera: Rhizobium, Bradyrhizobium, Mesorhizobium, Sinorhizobium (now renamed as Ensifer), and Azorhizobium. Neorhizobium and Pararhizobium are two more genera recently added to the rhizobial group. Nearly 125 different species of rhizobia form a symbiotic association with specific legume plants and fix the nitrogen. Due to high-efficient nitrogen fixation, the legumes rely less on the inorganic nitrogen fertilizer. Further, the legume adds nitrogen to the soil through their biological nitrogen fixation. The biological nitrogen fixation by legumes is now being explored for soil fertility and nitrogen management for other non-legume crops through green manures, legume-based crop rotation, legume as intercrop.

Rhizobia Biofertilizer

Though Indian soils are sufficient with rhizobia to nodulate diverse legume crops, the native rhizobia often fail to make high N turn over through biological nitrogen fixation due to less efficiency. This might be due to sensitive and inefficient nitrogenase present in these strains. Hence, high-efficient rhizobia as biofertilizer should be inoculated to the legume crops through seed to enhance the BNF efficiency.

TNAU Rhizobia

Legume	Organism	Strain
Redgram	Rhizobium sp.	CC1, CoC1
Groundnut	Rhizobium sp.	TNAU14
Blackgram	Rhizobium sp.	CRU7
Greengram	Rhizobium sp.	BMBS47, CoC10
Bengalgram	Rhizobium sp.	CoB1
Tree legumes	Rhizobium sp.	ALM10
Soybean	Bradyrhizobium japonicum	CoS1

Through extensive research, TNAU center under ICAR's AINP SBB developed legume-specific Rhizobial strains as biofertilizer mother cultures.

TNAU Rhizobium Biofertilizers

Rhizobium biofertilizer is commercially produced as the carrier (lignite) based and liquid formulations at TNAU.

Carrier-based biofertilizer: This lignite-based carrier formulation will have 10^7 cells per g with three months shelf-life.

Liquid biofertilizer: The liquid formulation of Rhizobia is with 10^8 cells per ml without any contaminants and can be stored for one year.

Liquid biofertilizer:

- Seed treatment: 250 ml/ha of seeds
- Soil application: 500 ml/ha

Carrier-based biofertilizer

- Seed treatment: 1 kg/ha of seeds
- Soil application: 2 kg/ha

Technology adoption and impact analysis

Adoptive research trials, milti-location trials and front-line demonstration trials have been conducted to identify the potential rhizobial strains and to popularize the rhizobial biofertilizer for the pulses.

Legume	Strain	Period	Inoculated (kg/ha)	Control (kg/ha)	Percent increase
Groundnut	TNAU14	1991-2010	1977	1651	18.50
Soybean	CoS1	1991-2000	1680	1424	17.53
Blackgram	BMBS47	2018-2021	1044	820	27.31
Greengram	CoG15	2018-2021	954	741	28.75

Rhizobial inoculation through seed treatment ensures 15-30% higher yield than uninoculated control.





Economic Analysis

Crop	Total area	Benefit (Rs)	Benefit per ac
Groundnut	100 acres	53,095	Rs. 5,310
Pulses	100 acres	59,800	Rs. 5,980

The yield increase and fertilizer savings were accounted for yield increase and marginal benefit of legumes due to Rhizobium biofertilizer application. By using Rhizobium biofertilizer alone, nearly Rs. 5000-6000 could be the monetary benefit being obtained by the farmers.

New Technologies with Rhizobium

Co-inoculation with other inoculants

Phosphorus is the major nutrient-limiting factor for effective nodulation and nitrogen fixation. To improve the BNF-efficiency of Rhizobia in legume, Phosphobacteria and mycorrhizal biofertilizers were recommended as co-inoculant with Rhizobium. The co-inoculant ensures sufficient phosphorus requirement for the legume – rhizobial symbiosis.

Blackgram

Biofertilizer	Strain	Grain yield (kg/ha)	Percent increase
Rhizobium	CRU7	429	11.3
Phosphobacteria	Pb1	304	2.8
Rhizobium + Phosphobacteria	CRU7 + Pb1	570	92.9
Control		297	

Horsegram

Biofertilizer	Strain	Grain yield (kg/ha)	Percent increase
Rhizobium	BMBS14	690	25.4
Phosphobacteria	Pb1	630	14.5
Rhizobium + Phosphobacteria	BMBS14 + Pb1	715	30.0
Control		550	

Biofertilizer	Blackgram		Redgram	
	Grain yield (kg/ha)	Percent increase	Grain yield (kg/ha)	Percent increase
Rhizobium	730	15.32	792	4.92
Rhizobium + AMF	795	25.60	1011	34.26
Control	633		753	

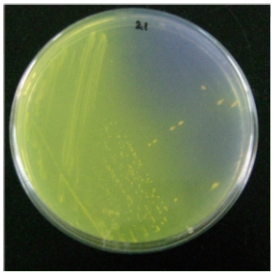
Co-inoculation with antagonistic bacteria

An antagonistic bacteria compatible with Rhizobium and Phosphobacteria was developed as an inoculant to improve Rhizobium's colonization efficiency and nitrogen-fixation in redgram.

Biofertilizer	Strain	Grain yield (kg/ha)	Percent increase
Rhizobium	CoC10	655	38.7
Phosphobacteria	Pb1	483	2.3
Rhizobium + Phosphobacteria	CoC10 + Pb1	612	29.6
Rhizobium + Phosphobacteria + Antagonistic bacteria	CoC10 + Pb1 + KB133	760	48.7
Control		472	

Rhizobium sp. (CoC10); *Bacillus megaterium var. phosphaticum* (Pb1); *Pseudomonas sp.* (KB133)

Co-inoculation with Sulphur bacteria



Sulfur nutrition limits the oil content of groundnut like oil crops. Sulfur-oxidizing bacteria, compatible with Rhizobium as co-inoculant, was developed at TNAU to ensure enhanced yield and oil content of groundnut.

Biofertilizer	Strain	Yield(Kg/ha)	Oil (%)
Rhizobium sp.	TNAU14	1760.0	45.13
Thiobacillus sp.	LCH	1940.0	52.16
Rhizobium + Thiobacillus sp.	TNAU14 + LCH	2006.7	52.58
Control		1700.1	44.11

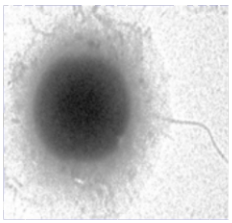
Non-rhizobial endophytes

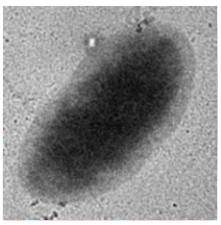
Two non-rhizobial endophytes developed as co-inoculants of Rhizobium.

Bacillus tequilensis (NBB13)

Pantoea dispersa (YBB19B)

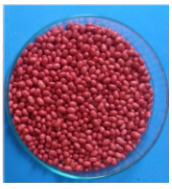
- Co-inoculation showed significant improvement in Rhizobial nodulation and nitrogen fixation in groundnut.
- These endophytes improve the nodule occupancy of inoculated rhizobial strain and enhance its efficiency.





Seed coat formulation of Rhizobium

Crop	FLDs	Yield (kg/ha)			B:C ratio	
		Seed-coat	Control	% increase	Regular	Seed-coat
Red gram	2	245	205	20	1.20	1.30
Groundnut	24	1225	892	37	1.81	2.17



Seed coat formulation using lyophilized powered Rhizobia along with biopolymer can coat the seed surface uniformly. The cell load will be 1010 per g, and the shelf-life is one year.

Seed coating: Legume seed (per ha), 25 g of high-concentrated rhizobia and 20 ml of binder (biopolymer) per kg of seed should be coated on the surface of the seed through a seed coating machine.

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