BNF Technology for Sustainable Agriculture in the Semi-Arid Tropics

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Abstract. Biological nitrogen fixation plays an important role in maintaining soil fertility. However, as BNF is dependent upon physical, environmental, nutritional and biological factors, mere inclusion of any N_2-fixing system does not guarantee contributions to soil N pool. In the SAT in situations where plant stover is also removed to feed animals, most legumes deplete soil N. Beneficial effects of legumes in terms of increased yields of succeeding cereal crops have been reported. Such benefits are due to N contribution from legumes through BNF and soil N saving effect. In addition, other non-N rotational benefits for eg. improved nutrient availability, improved soil structure, reduced pests and diseases, hormonal effects are also responsible. For exploiting BNF technology for developing sustainable cropping systems in the SAT, we need to take holistic approach involving plant host, bacterium, and environment. Selection of the appropriate host types and genotypes which fix larger proportion of their N requirement under adverse conditions through BNF is needed. Along with this optimum management practices must be provided to ensure maximum contribution from the BNF. For success of inoculation technology in the SAT, concerted efforts right from production, demonstration to distribution will be needed.

Introduction. Sustainable agriculture involves the successful management of resources for agriculture to satisfy changing human needs while maintaining or enhancing the quality of the environment and conserving natural resources [59]. Intensive agricultural systems are characteristically expanded nutrient cycles involving export of crops from a farm and require continued import of nutrients to the farm. Nitrogen is the most limiting nutrient for increasing crop productivity. The continued and unabated use of N fertilizers would further deplete stocks of nonrenewable fossil fuels used in fertilizer production.

Sustainable agriculture relies greatly on renewable resources and on-farm nitrogen contributions are achieved through biological nitrogen fixation (BNF). Biological nitrogen fixation helps in maintaining and or improving soil fertility by using N_2 which is in abundance in the atmosphere. Annually, BNF is estimated to be around 175 million tones of which close to 79% is accounted for by terrestrial fixation (Fig. 1) which is indicative of the importance of BNF in the context of the global N cycle. The BNF offer an economically attractive and ecologically sound means of reducing external inputs. In this paper we deal with the BNF systems involving the upland legume crops grown in the semi-arid tropics (SAT).

Nitrogen contributions through legumes. Symbiotic N_2-fixation by *Rhizobium* with legumes contribute substantially to total BNF [47, 64]. Nitrogen fixation is dependent upon physical environmental, nutritional and biological factors [39, 47] and it can not be assumed that any N_2-fixing system will automatically make large contributions to the N cycle. In most of the studies while estimating BNF, plant roots and fallen leaf material is not taken into account which results in underestimation of quantity of N_2 fixed. It is essential that BNF in roots and fallen leaves should be considered to accurately estimate contribution of N_2 fixed by legumes.

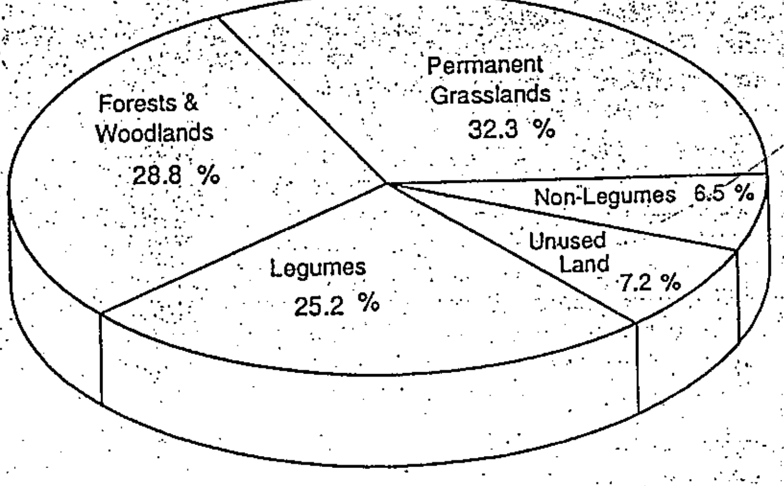

Figure 1. Distribution of 139 million tonnes of N_2 estimated to be biologically fixed in various terrestrial systems. Source: Burns and Hardy (1975).

Legumes are important component of agriculture since ancient times. It is widely believed that legumes improve soil fertility because of their N_2-fixing ability. In support of this argument always amounts of N_2 fixed by legumes are cited. However, in order to assess the role of BNF in sustainability of different cropping systems in the SAT, not only the amount of nitrogen fixed by the legume component crop in the system is important but overall nitrogen balance of the system need to be considered. The SAT is characterized by a harsh environment with erratic seasonal rainfall and dense human and animal population and it has unique problems in agriculture also. It is a common practice to remove plant material also from the field for feeding the animals. In such a case only nodulated roots and fallen leaves go back to the soil.

Different maturity groups of pigeonpea cultivars grown at Patancheru, India fixed 4-53 kg N ha^{-1} $season^{-1}$ [28] and also depleted 20-49 kg N ha^{-1} from soil. In case of chickpea, different cultivars fixed 23-40 kg N ha^{-1} $season^{-1}$ (O.P. Rupela, personal communication) and removed 63-77 kg N ha^{-1} $season^{-1}$ from soil (Table 1). Groundnut fixed 190 kg N ha^{-1} $season^{-1}$ when pod yields are around 3.5 t ha^{-1} at Patancheru [38], however, groundnut relied for its 20-40% (47-127 kg N ha^{-1} $season^{-1}$) of the N requirement on soil or from fertilizer [17], obviously resulting into negative N balance. Positive net N balances up to 136 kg ha^{-1} for several legume crops following seed harvest have been shown [47]. However, if crop residues are removed from the field then net N balances for groundnut workout to be -27 to -95 for soybean -28 to -104, common bean -28, greengram -24 to -65 and cowpea -25 to -69 kg ha^{-1}. Similarly, for soybean, N balances with seed and stover removed ranged from -12 to -35 kg ha^{-1} in northern Thailand [22]. For different cropping systems where pigeonpea and groundnut were grown as intercrops nitrogen balances worked out negative [32]. In case of sole pigeonpea grown in rotation with sole castor a positive balance of 18 kg N ha^{-1} during two years crop rotation was observed at Patancheru. At Patancheru, without any N fertilizer application, sorghum and millet cultivars removed 22-34 kg N ha^{-1} y^{-1}. These results show that legumes also mine soil N as cereals do. However, total plant N yields from legumes are far higher than the cereal plant N yields. We reach to the conclusion that in general legumes slow the decline of, rather than enhance, the N fertility of the soil in comparison with cereal systems.

Table 1. Net nitrogen balances for pigeonpea and chickpea cultivars grown at Patancheru, India.

Cultivar	Total plant N uptake (kg ha^{-1})	Plant N derived from fixation (kg ha^{-1})	Net N balance (kg ha^{-1})[a]
Pigeonpea[b]			
Prabhat	69	4	-49
UPAS 120	92	27	-39
T 21	108	43	-39
BDN 1	118	53	-32
Bhedaghat	101	36	-20
JA 275	78	13	-33
Bhandara	108	43	-22
NP (WR) 15	114	50	-27
Chickpea[c]			
Annigeri	110	31	-77
G 130	104	26	-75
ICC 435	102	29	-72
ICCC 42	88	23	-64
ICCV 6	107	30	-76
K 850	104	40	-63

Source: Derived from Kumar Rao and Dart [28], Rupela, O.P. (personal communication).

a. Net N balance calculated as Total plant N uptake - (N derived from BNF + N derived from fertilizer + N added to soil through plant roots and fallen plant parts).

b. N derived from fixation calculated for roots also.

c. N derived from fixation calculated for above ground plant parts only.

Beneficial effects of legumes. Despite the negative N balances for the legume crops grown in rotation or as intercrops reported benefits of legumes to succeeding non-legume crops have been observed consistently (Table 2). Improvement in cereal yield following monocropped legumes lie mainly in the 0.5 to 3 t ha^{-1} range, representing around 30 to 350% increase over yields in cereal-cereal cropping sequences [47]. Such increased cereal yields following legume crops attributed to the N contribution from legumes in crop rotation [13, 39]. This opinion is not held by all [10, 11, 54, 66].

Table 2. Residual effect of preceding legume on cereal yield in terms of fertilizer N equivalents.

Preceding legume	Following cereal	Fertilizer N equivalent (kg ha^{-1})
Berseem	Maize	123
Sweet clover	Maize	83
Winged bean	Maize	70
Blackgram	Sorghum	68
Greengram	Sorghum	68
Greengram (monocrop)	Wheat	68
Chickpea	Maize	60-70
Cowpea	Maize	60
Groundnut	Pearl millet	60
Cowpea	Pearl millet	60
Chickpea	Pearl millet	40
Lentil	Pearl millet	40
Peas	Pearl millet	40
Pigeonpea	Wheat	40
Cowpea	Monocrop	38
Latyrus	Maize	36-48
Lablab bean	Maize	33
Pigeonpea	Pearl millet	30
Greengram	Pearl millet	30
Groundnut (monocrop)	Wheat	28
Pigeonpea	Maize	20-67
Peas	Maize	20-32
Lentil	Maize	18-30
Greengram (intercrop)	Wheat	16
Cowpea (intecrop)	Wheat	13
Groundnut (intercrop)	Wheat	12
Groundnut	Maize	9-60
Soybean	Maize	7

Source: Wani et al. [68].

Nitrogen effect. Terms like "N residual effect" [13] and "Fertilizer N replacement value (FRV)" or N equivalent [20] are used to describe the role of legumes in crop rotations. This concept does not distinguish between biological N_2 fixation and the "N-conserving effect" which results from substitution by legumes of biologically fixed N for soil N. The N contribution estimated by FRV method from hairy vetch and big flower vetch was almost doubled (135 vs. 75 kg N ha^{-1}) using grain sorghum as test crop instead of maize [3]. Recently, ^{15}N methodology has been used to measure the residual effects of legumes to circumvent problems with non-isotopic methods [11, 55, 66]. Based on estimates obtained via ^{15}N methodology, Hesterman et al. [20] argued that the amount of N credited to legumes in a crop rotation in the north-central U.S. may be inflated by as much as 123% due to the use of fertilizer replacement value method. Based on ^{15}N methodology it is reported that only 7.3 to 28% of the ^{15}N in legume crops is taken up by a following grain crop [29, 63]

Growing legumes in rotation does improve mineral N content in soil as that of the non-legume crops. At ICRISAT Center, Patancheru, near Hyderabad, a long-term rotation experiment is being conducted since 1983 using two-year crop rotation treatments. The surface soil (0-20 cm) samples collected after harvest of 9th season crop, showed higher amounts of mineral N contents in soil under legume-based cropping system than the non-legume based cropping system (Fig. 2). Inclusion of greengram in cropping sequence increased the available nitrogen in soil to the extent of 12.6% in non-fertilized control plot [49].

In addition to mineral N content in soil from the long-term rotation experiment, N mineralization potential (N_0) of the soils under pigeonpea(pp)-based cropping systems was almost two times higher than that of the fallow-sorghum treatment. Such increased N_0 values at Patancheru were not associated with chickpea(cp) which is grown during postrainy season on residual soil moisture. The "active N fraction" the quotient of N_0 and N_{total} and expressed as percentage varied between 9-17 % with higher values reported for the soil under pigeonpea-based cropping systems (Wani et al. unpublished data). Using N_0 and K (N mineralization rate constant) values derived through exponential model, time required to mineralize 25 mg N kg^{-1} soil was less in case of pigeonpea-based systems (1.5-13.8 wk) than that of cropping systems which contained chickpea or no legume or which was left fallow during rainy season (19.6-21.4 wk). Analysis of field soil samples collected prior to start of the experiment in 1983 and later in 1993 showed that in case of Fallow+Sorghum (F+S) system total soil N content was decreased by 72 µg g^{-1} soil during ten years period. Similarly, S+CP-S+SF and S+SF-S+SF plots also recorded decrease in total soil N. The continuous greengram + sorghum maintained the soil N while substantial increase in total N was observed in S/PP-S+SF (70 µg g^{-1}) and cowpea(COP)/PP-S+SF (102 µg g^{-1}) (Wani et al. 1994a). These results demonstrated that pigeonpea-based cropping systems increased total soil N substantially during ten years.

Sorghum was grown in the greenhouse using surface soil samples collected from field plots which are under different cropping systems since last 9 years. Sorghum grown in soil from COP/PP-S+SF plots yielded 63% and in other pigeonpea-based systems yielded 36-56% higher as that of sorghum grown in soil from S+SF-S+SF plots. In case of chickpea-based cropping systems, sorghum yields were lowered by 18-24.5% over S+SF-S+SF plot yields (Wani et al. unpublished results). Using ^{15}N methodology and sorghum grown in S+SF-S+SF soil as check

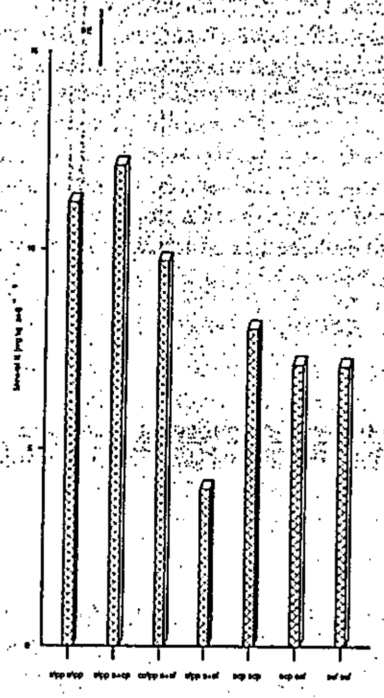

Figure 2. Mineral N content in surface soil samples (0-20 cm) from plots under different cropping systems since last nine years.

it was estimated that 8.4 to 20% of total sorghum plant N in case of pigeonpea-based cropping systems was derived from N that was either fixed previously and had accumulated, or soil N that was made more available due to the presence of pigeonpea in the rotation. The 'A' values for soil from pigeonpea-based cropping system plots were higher by 25.6 to 76.3 mg pot^{-1} (4.5-13.3 kg N ha^{-1} equivalent) than that of the S+SF-S+SF treatment. The fertilizer N replacement values calculated for these treatments using soil from S+SF-S+SF treatment ranged from 65-161 mg N pot^{-1} (24-28 kg N ha^{-1} equivalent). All these results indicate that increased sorghum yields from pigeonpea-based cropping systems are partly due to increased soil N availability and all the benefits can not be explained in terms of N effects (Wani et al. unpublished results). Similarly, non-N rotational benefits of legumes towards yield of subsequent crop have been observed by many researchers [10, 11, 47, 22, 66, 67].

Non-N rotational effects. If the benefits of crop legumes in rotations cannot be solely explained in terms of residual fixed N, then what are the sources of the benefits demonstrated in Table 2? Several factors can be involved, the relative importance of each dictated by site, season, and crop sequences. Crop rotations increased availability of nutrients other than N through increased soil microbial activity [30, 66, 67]. Further, through positive relationships between levels of mycorrhizal colonization and K, Ca, Mg, Zn, S, and Fe accumulations and barley yields it was inferred that increased mycorrhizae acted as agents to mediate enhanced soil fertility in the legume-based rotations over that of continuous barley [67]. Improvements in soil structure following legumes mainly improved soil aggregate formation after three years of alfalfa, clover and hairy vetch mixture [31] or with numerous years of a Sod pasture, or hay crop [46] have been observed. Incorporation of legume residues improved soil water-holding [70] and buffering capacity [6]. Ries et al. [50] suggested that growth promoting substances in legume residues are responsible for the rotation effect. Crop rotations break cycles of cereal pests and diseases, and phytotoxic and allelopathic effects of different crop residues [14]. Crop rotation is an effective tool against certain pests and diseases, and that efficacy may contribute to the rotation effect [10], but rotation does not control all diseases [67].

Ways for exploiting BNF technology in the SAT

Host variability for nodulation and nitrogen fixation. Presence of a large genotypic variability for BNF traits like nodule number, nodule mass and acetylene reduction activity (ARA) has been known since early eighties for chickpea, groundnut and pigeonpea [39]. Genotypes with high N_2-fixing ability for various legumes have been identified, however, efforts to use this variability in breeding for improved BNF has been limited or non existent in all these legumes.

Large plant to plant variability for nodulation and ARA observed in different legume cultivars was investigated in chickpea and pigeonpea. It was observed that not only consistent low and high nodulating plants were present within chickpea cultivars [52], even nonnodulating (Nod$^-$) plants occurred in normal cultivars or landraces [51]. Consistent variability for nodulation extent was subsequently detected within pigeonpea cultivars also. Unlike in chickpea, however, nonnodulating plants in pigeonpea were found in segregating populations at F_2 [52]. Using appropriate screening procedures, low- and high-nodulation types under low- and high-soil N conditions have been identified within several chickpea cultivars since 1985 at ICRISAT. Preliminary studies of Venkateswarlu and Katyal [64] also indicate plant to plant variability within cultivars of groundnut. Intracultivaral variability for nodulation may be present in other legumes also. The natural occurrence of Nod$^-$ and the low-nodulating plants in different cultivars is undesirable. Appropriate checks should be built in legume breeding programs to ensure that such plants are eliminated in early stages of development of cultivars. High nodulating selections are expected to improve yield under low N conditions. In our screening studies the high-nodulating selection generally grew better than non-nodulating and low nodulating selections of a given cultivar. In large plot yield trials with low- and high-nodulating selections of chickpea cv. ICC 4948 and cv. ICC 5003, the high nodulating (HN) selection of cultivar ICC 4948 produced 31% higher grains than its low nodulating (LN) selection at low N fertility. The HN selection of ICC 4948 yielded superior even at high N fertility. But the LN and HN selections of another cultivar ICC 5003 yielded similarly. In a previous pot trial the root length density of low-nodulating ICC 5003 was 32 m plant^{-1} which was 2-times greater than that of the low-nodulating ICC 4948. Perhaps the cultivar ICC 5003 could scavenge the soil N more efficiently than that of ICC 4948 due to its high root length density and as a result both the high and low nodulating lines of ICC 5003 yielded similarly.

Similarly, sufficient variation for BNF in presence of high mineral N in soil between different legume types [74] and also amongst genotypes of a given legume have been observed [19].

Management practices. Legumes are generally grown as intercrops with cereals or other nonlegumes in the SAT [73] and application of N to cereal crop reduced N_2 fixation by the component legume crop [36, 43]. To improve BNF contribution from legumes under such circumstances soil N must be managed through inclusion of appropriate nitrate tolerant legume crop or genotype of a given crop (as mentioned earlier) and/or appropriate cropping and management practices.

In intercropping, shading by associated cereals reduced BNF by component legumes [36]. Strip cropping of cereals and legumes can overcome both these problems and improve the systems productivity without reducing BNF contributions in the system from associated legumes. Indeterminate legumes fix more N than determinate types in intercropping [16]. BNF in climbing bean [18], cowpea [44] and Serratro [45] was unaffected by intercropping with cereals. In cases where strip cropping is not possible, climbing type legumes can be used to overcome shading effect of component cereal crop.

Nodulation and N_2 fixation in soybean grown in sub-tropical Australia were substantially improved under no tillage with N balance of 80 kg N ha^{-1}, compared with the cultivated system with 30 kg N ha^{-1} N balance. Increased N_2 fixation resulted mainly from the higher proportion of plant N derived from fixation since yields were unaffected by tillage practice [47]. Clean cultivation accelerates the oxidation of organic matter in soils and generally results in higher nitrate-N in the profile [62] which would affect BNF in legumes.

It should be realized however, that poor N_2 fixation can be due to poor plant growth resulting from pests, diseases, and nutrient deficiencies. In Karnataka, India, trials on farmer's fields with pigeonpea showed dramatic increase in nodulation due to application of diammonium phosphate (DAP) alone than to inoculation with *Rhizobium* alone. The plots receiving DAP and *Rhizobium* yielded 100% more than the control plots [9]. Field-grown soybean had a higher P requirement when it was dependent on BNF for its N supply as compared to mineral N dependency [8]. Based on the results from 140 on-farm demonstration plots with soybean in Uganda it was observed that on an average 300 kg ha^{-1} yield increase was obtained with 40 kg P_2O_5 ha^{-1} application and further increase of 300 kg ha^{-1} was obtained through inoculation with *Rhizobium* [25].

In groundnut, fertilization with B, Co, Mo and Zn in a medium calcareous soil, with and without *Rhizobium* inoculation significantly increased nodulation, percentage of effective nodules and plant dry matter [23]. It has been reported that Fe deficiency specifically limits nodule development in groundnut grown in the calcareous soils of Thailand [42]. Soil acidity along with Mn and aluminium (Al) toxicities can also restrict N_2 fixation in groundnut. Excess Mn was detrimental to plant growth per se rather than to nodulation, but nitrogenase activity was more affected by Al than plant growth [34]. Application of Co and Mo significantly increased nodulation and grain yield of pigeonpea [26, 48]. Similarly, soil application of 1 kg cobalt chloride ha^{-1}, 1 kg ha^{-1} sodium molybdate and 25 kg $ZnSO_4$ ha^{-1} increased chickpea grain yield by 10, 7 and 4% respectively over control. Inoculation with *Rhizobium* increased chickpea yield by 26% over uninoculated control however, inoculation along with Co, Mo and Zn application increased yield by 41, 39 and 28% respectively over control [41].

Extensive nodule damage to pigeonpea by a Dipteran larva, *Rivellia angulata* in farmers' fields reduced yields significantly. The extent of nodule damage was greater in pigeonpea grown in Vertisols (up to 86%) as compared to 20% in Alfisols [39]. Possible solution is to select pigeonpea genotypes that can resist or tolerate attack by nodule damaging insects.

Deep sowing of groundnut results in the development of an elongated hypocotyl, poor rooting, poor nodulation and nitrogen fixation, notably in spanish types. Virginia types have considerable nitrogenase activity even when sown deep because of their ability to nodulate on the hypocotyl [39]. Farmers tend to sow chickpea at a sufficient depth to ensure good crop stand as it is generally grown on residual moisture. Deep sown chickpea crop in heavy black soils suffer a substantial reduction in nodulation and N_2 fixation [53]. In lighter soils chickpea have been found to nodulate at depth. Greengram, pigeonpea and soybean grown on broad bed and furrows (BBF) on Vertisol improved nodulation than when grown on flat surface. However, improved nitrogenase activity on BBF was recorded with greengram and pigeonpea only [68].

Use of Inoculants. Much of the applied research efforts in studying BNF have gone into identifying efficient strains of bacteria as inoculants. Before inoculation with appropriate strains to be used, it needs to be determined whether inoculation is needed?

Need for Inoculation. The need to inoculate the legumes grown on cultivated soils must be assessed by considering the interacting factors between the soil, the host plant and *Rhizobium*. Most cultivated tropical soils are assumed to have relatively large populations (> 100 g^{-1} dry soil) of rhizobia capable of nodulating the legumes grown in such soils [39]. The results of surveys of farmers' grain and fodder legume crops however, revealed poor nodulation in large areas and good nodulation only in a few pockets [21, 24, 27]. In a similar survey conducted for 43-47 villages from three districts of Madhya Pradesh, India for nodulation of pigeonpea, black gram, green gram and lentil showed poor nodulation (0-10 nodules $plant^{-1}$) in 64 to 100% of the surveyed area [41].

Presence of nodules on plant roots does not necessarily mean that sufficient N_2 is being fixed for maximum benefit to the host plant. In a survey of groundnut grown on farmers' fields in southern India, 52 out of 95 fields showed inadequate nodulation with less than 10 per cent N_2-fixing (acetylene reducing) activity of what can be obtained under reasonable field conditions [35]. Although adequate nodulation was observed in some parts, ineffective nodules exceeded the number of effective nodules. Out of 87 groundnut rhizobial strains isolated from different parts of India, only 5 were found to be effective [27]. However, the ability to fix high amounts of N (efficiency) is governed by the symbiotic capability between *Rhizobium* and the host plant. Hence, it may be necessary to introduce superior (more competitive and efficient) strains of *Rhizobium* to ensure adequate N_2 fixation for maximum growth and yield of the host plant.

Thies et al. [61] developed mathematical model using native rhizobia numbers (estimated by most probable number method) and soil mineral N data as inputs to predict the inoculation responses at different sites. This approach accounted 83% of the variation observed in inoculation. These models have been incorporated into interactive computer program called "RESPONSE" which reduces the need for costly, site-specific field inoculation trials to determine the need for inoculation with *Rhizobium*. However, Nambiar [33] reported significant yield increases from Cameroon, India, and China in case of groundnut due to inoculation with NC 92 strain from the soils having large populations of native rhizobia. These results indicate that simulation model using MPN data and mineral N data cannot provide reliable answers in all the cases and this approach need to be followed cautiously.

Competitiveness and Effectiveness of Strains. For effective nodulation of legumes by rhizobia the introduced strains should be competitive and efficient. The degree of establishment and persistence of an inoculant strain generally decreased with increase in population density of the native rhizobia [33]. However, some inoculant strains have succeeded in forming more nodules even in the presence of active indigenous competing rhizobia eg. NC 92 on groundnut [37]. Little is known of the factors controlling competitiveness but host cultivar, soil properties, soil microflora, environmental factors and the nature of the competing strains influence the success of inoculant strains in nodule formation [2]. The success of introduced strains in terms of nodule formation increased with repeated inoculation [71], and with higher inoculum rate [37]. Competition between *Rhizobium* strains and inoculation response was less pronounced in the presence of soil mineral N than under conditions where such N was immobilized and unavailable [56].

Factors Affecting Performance of Inoculant Strains. Crop responses to inoculation with biofertilisers are not as dramatic as those with fertilizer N. Being biological agents, these are subjected to range of hostile environments and their survival and efficiency is governed by several factors [65]. Generally, there is a decline in the rhizobial population on seeds but conventional wisdom is that multiplication should occur as the rhizosphere forms, so that accelerated germination can also assist in ensuring an adequate population. In case of crops grown on residual moisture, such as chickpea, the inoculated rhizobia cannot move downwards with the growing root from the top soil where inoculated resulting in poor nodulation. Secondly, deep sowing results in good crop stand but affects nodulation adversely [39].

Carrier-based inoculants are usually coated on seeds for the introduction of bacterial strains into the soil. However, alternative inoculation methods are necessary where seed treatment with fungicides and insecticides is needed or where seed of crops such as groundnut and soybean can be damaged when inoculated with an adhesive. Increased groundnut yields were obtained when inoculation was done by applying a slurry of peat-based inoculation in the seed furrow (Table 3). The normal carrier-based inocula can be successfully applied separately from the seed [4, 7]. While all methods of inoculation were successful under favorable conditions, "liquid" and "solid" methods were superior to seed inoculation under adverse conditions [5].

Soil properties can also affect the survival of inoculated rhizobia. For example, out of 11 locations tested for response of groundnut cv Robut 33-1 inoculation with strain NC 92 failed to increase yields at two locations, namely Tirupathi and Kadiri, India [1]. Subsequent analysis of soil samples from Tirupathi revealed a high (150 ppm) available manganese content [33]. Manganese and aluminum can be toxic to symbiotic N_2 fixation even if they are not at a level high enough to affect plant growth [15]. Soil acidity and alkalinity can also pose problems for symbiotic N_2 fixation. For such problem areas, specific strains with the ability to overcome such adverse conditions need to be selected as inoculants. Significant differences were observed among pigeonpea rhizobial strains for their ability to nodule and fix N_2 under saline conditions

[59]. Nambiar et al. [40] reported reduced nodule damage by 50% due to inoculation of pigeonpea with engineered *Bradyrhizobium* carrying insecticide gene (*Bacillus thuringiensis* subsp. israelensis) in the presence of *Rivellia angulata* larvae under greenhouse conditions.

Table 3. Persistence of inoculum strain NC92 over two seasons on groundnut

Season		% nodules formed on groundnut plants	
1st	2nd	72 days after sowing	116 days after sowing
Uninoculated	Uninoculated	9 (5)*	11 (8)
Uninoculated	Inoculated	31 (27)	27 (25)
Inoculated	Uninoculated	28 (25)	42 (32)
Inoculated	Inoculated	39 (41)	75 (54)
SE		± 2.5	± 5.4

* Data analysed after arsine transformation: original means in parenthesis.

Source: Nambiar [33].

Yield Response to Inoculation. The field performance of inoculation is variable. For example, with chickpea significant improvement in grain yield was reported from 7 out of 16 [57] and 6 out of 12 locations [58], predominantly in central and northern India with yield increases varying from -14 to 30% over the control plots. In pigeonpea significant increases in early nodulation due to inoculation were not always well correlated with the final grain yields. Increase in grain yield of the pigeonpea inoculated with effective *Rhizobium* ranged from 19 to 68% over uninoculated controls [39]. In groundnut, inoculation responses varied from decreased yields to significant increased yields over uninoculated controls [27, 39, 57]. Using network approach Niftal initiated Worldwide Rhizobial Ecology Network (WREN) and conducted standardized inoculation trials with extensive environmental data. Over 228 inoculation trials were conducted under the International Network of Legumes Inoculation Trials (INLIT) by cooperating scientists in 28 countries over the years. In approximately 52% of the cases, inoculation resulted in significant yield increases [12]. In summary, yield responses to inoculation were site specific, depending on location, species, fertility, and other factors.

Sometimes, legumes yields are not increased by inoculation but N concentration in grains or plant parts is increased over the uninoculated control. In cases where both types of responses are not observed, it might simply result in a saving of soil N which might be useful for the succeeding crop.

Strategy for improving BNF contribution in the SAT. Biological nitrogen fixation plays an important role in sustaining productivity of the soils in the SAT. Although, legumes fix substantial amounts of nitrogen through BNF, net N balances for legumes or legume-based cropping systems in the SAT indicated that such balances are negative and legumes also deplete soil N in the situations where plant residues are removed from the field. However, total plant N yields are far higher than the cereal yields and legumes slow the decline of, rather than enhance, the N fertility of the soil in comparison with cereal systems. The important issue is how best we can exploit BNF technology for developing sustainable cropping systems in the SAT?

There is need to understand BNF system by taking holistic approach which includes host, bacterium and environment and ensure that all the partners involved work in harmony to deliver maximum benefit. Accurate quantification of N_2 fixed by legumes will help us to identify the systems which really maintain or improve the soil N status. Host controlled factors play an important role in regulating BNF but have not received its due share by researchers. We need to identify type of legume and also genotype of a given legume which yields more and also derive larger part of its N requirement from fixation in a particular cropping system. For example, we need to identify crops and genotypes of legumes which can fix more N_2 under sole cropping and intercropping situations without being affected by high mineral N contents in soil. There is a need to identify host genotypes which can fix well under adverse soil conditions like soil acidity, Al & Mn toxicity, alkalinity, water logging, etc.

At ICRISAT non-nodulating lines of chickpea, pigeonpea and groundnut have been developed from the existing cultivars and/or segregating populations. Natural occurrence of nonnodulating plants ranged from 120 to 490 per million plants and efforts are required to see that occurrence of such plants do not increase. Most of the breeding and testing work is done at the research stations where mineral N contents are far higher than observed on the farmers fields. There is every likely hood that low- or non-nodulating plants may not be identified as they will grow normally using soil N. To avoid this, appropriate checks during breeding and testing for discarding low-nodulating plants must be built in the breeding programs.

Along with the appropriate host types and genotypes, optimum management practices must be provided to ensure maximum contribution from the BNF. In intercropping situations where application of fertilizer N is essential to obtain higher cereal yields, appropriate form of fertilizer eg. slow releasing formulations, organic N and suitable method of application eg. placement between cereal rows than broadcasting and mixing in soil must be worked out. Appropriate amendments with nutrients other than N which might limit the plant growth should be done.

Application of farm yard manure and other organic amendments improved BNF in legumes. Suitable land management practices which can improve water storage capacity of soils or which can drain excess water away from the plant depending on the situation need to be used to harness maximum benefits from BNF. For eg. pigeonpea grown on Vertisols fixed more nitrogen when grown on BBF than the one grown on flat surface, whereas it was reverse for chickpea.

Efforts for selection of efficient strains of bacteria for using as inoculants and identification of specific host-bacteria combinations must go on. Situations which need inoculation should be identified and efforts for success to inoculation in such areas must be concentrated. For increasing crop yields through biofertilizers, the following strategy is suggested. Most important constraints to effective exploitation of BNF technology in the SAT are:

- the quality of the inoculants
- lack of knowledge about inoculation technology for the extension personnel and the farmers.
- effective inoculant delivery system
- formulation of the policy dictating the desire to exploit BNF successfully.

For success of biofertilizers in the SAT, concerted efforts right from production, demonstration to distribution will be required. Biofertilizers should be used or considered as an insurance for harnessing BNF to its maximum potential taking systems approach. As discussed earlier in chickpea, the non-nodulating or low nodulated plants look similar in appearance to well nodulated plants but this is at the cost of soil or fertilizer N. We must take the view that in the end we may derive benefit in terms of maintaining or improving the productivity of our soils. We should not be disappointed by not seeing the direct benefits in some cases. A holistic approach to improve production of legumes is needed and we must ensure that all the constraints for good plant growth other than N nutrition are alleviated and suitable management practices are provided for better performance of BNF technology.

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