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# Crop Legumes Grown in *Jhum* Fields and Home Gardens in Nagaland and Study of the Root Nodule Morphology

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#### ABSTRACT

Surveys were conducted in five districts of Nagaland to collect crop legumes grown in Jhum fields and home gardens. Ten crop legumes, namely Canavalia gladiata (Jacq.) DC, Glycine max (L.) Merr, Lablab purpureus L., Mucuna prurita (L.) DC, Phaseolus lunatus L., Phaseolus vulgaris L., Psophocarpus tetragonolobus L., Vicia faba (L.) D.C, Vigna umbellata (Thunb.) Ohwi and H. Ohashi and Vigna unguiculata (L.) Walp were commonly cultivated. Vigna unguiculata and Vigna umbellata were the most popularly grown beans. Root nodule was observed in all nine crop legumes except for Vicia faba. The nodules were of the determinate type. Number of root nodules and nodule size varies with the species. Large nodules were found in Canavalia gladiata, Psophocarpus tetragonolobus, and Mucuna prurita with the mean size of  $5.15 \pm 0.617$  mm,  $4.93 \pm 0.115$ 

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mm, and  $4.83 \pm 0.289$  mm. The smallest was in *Vigna umbellata*, with  $1.45 \pm 0.610$  mm. Root nodule endophytes belonging to *Rhizobium* sp. were isolated from three popular crop legumes, namely, *V. umbellata, V. unguiculata,* and *P. vulgaris.* The study underscores the importance of further research on the crop legume symbiotic association with native root nodulating bacteria (rhizobia) for crop productivity.

**Keywords** Crop legumes, Home garden, *Jhum* field, Nagaland, Rhizobia, Root nodule.

## **INTRODUCTION**

Leguminosae represents the third largest plant family next to Orchidaceae and Asteraceae (Lewis et al. 2005). Leguminosae comprises of 800 genera and 20,000 species which are separated into three subfamilies, namely Caesalpinioideae, Mimosoideae, and Papilionoideae (Lewis et al. 2005, Semba et al. 2021). Leguminosae are considered economically important because of their potential use in food, fuel, feed, and fiber and their biological nitrogen-fixing abilities (Lewis et al. 2005, Semba et al. 2021). Legumes (Leguminosae family with edible seeds) constitute the second largest group of food crops after cereals, comprising 1,041,345 accessions and accounting for 15% of all crop accessions (FAO 2010, Semba et al. 2021). Soybean, mungbean, faba bean, lentil, chickpeas, peas, common bean or lupin, and lima beans are the

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most common crop legumes worldwide (Smykal et al. 2015). Concomitantly, legume symbiosis with nitrogen-fixing bacteria not only plays an essential role in natural ecosystems but also adds value to agriculture by reducing the use of synthetic fertilizers in crop production (Smykal et al. 2015, Goyal et al. 2021). The symbiotic association between the legume and root-nodulating bacteria (rhizobia) resulted in nodule formation, where the bacteria convert atmospheric nitrogen into ammonia, improving plant growth and yield (Dénarié et al. 1992, Oldroyd et al. 2011). Morphologically, the root nodules are divided into determinate and indeterminate types (Hirsch 1992). Determinate nodules are spherical, lacking persistent meristem. Meanwhile, indeterminate nodules possess an apical meristem, cylindrical and irregularly shaped (Hirsch 1992).

Nagaland is a small, hilly state located in the biodiversity-rich North-Eastern part of India, surrounded by Arunachal Pradesh to the north, Manipur to the south, Assam to the west, and the country of Myanmar (Burma) to the east. Several ethnic tribes inhabit the state. Agriculture is the primary occupation, with more than 70% of the state population depending on agriculture (Kehie et al. 2017). As important sources of plant protein, essential nutrients and its contribution to sustainable diets (Tharanathan and Mahadevamma 2003, Semba et al. 2021), legumes are a significant crop grown along with other edible plants to meet the dietary requirements of the ethnic population. The region has a rich diversity of legumes represented by numerous landraces (Pradheep et al. 2017). Despite the rich crop legume diversity, information on the root nodulation status of the cultivated legumes from the region is lacking.

This study documents the diversity of crop legumes grown in different parts of Nagaland and records each species' root nodulation status and nodule morphology. The work has significance to improve pulse production for food and nutritional security in Nagaland.

### MATERIALS AND METHODS

Field surveys were conducted in five districts of Nagaland, namely Mokokchung, Zunheboto, Wokha,

Mon, and Kiphire. Crop legumes and other non-legume crops found in the study sites consisting of *Jhum* field and home garden were recorded. Plants were carefully uprooted and thoroughly washed with water to study nodule morphology. The root was then further examined for the presence of nodules, and the nodules were classified into determinate or indeterminate types, as described by Howieson *et al.* (2016). The size, number, and color of nodules were recorded from each crop legume. The root nodules were cut to determine the presence of leghaemoglobin. Root nodule bacteria were isolated, following the standard protocols of Howieson *et al.* (2016). *Recombinase A* (*recA*) gene amplification and sequencing were performed as described by Sankhla *et al.* (2017).

### **RESULTS AND DISCUSSION**

A total of 10 crop legumes including Canavalia gladiata (Jacq.) DC, Glycine max (L.) Merr, Lablab purpureus L., Mucuna prurita (L.) DC, Phaseolus lunatus L., Phaseolus vulgaris L., Psophocarpus tetragonolobus L., Vicia faba (L.) D.C, Vigna umbellata (Thunb.) Ohwi and H. Ohashi and Vigna unguiculata (L.) Walp were grown in 26 sites, including Jhum fields and home gardens in various Nagaland districts (Table 1). Vigna unguiculata was the most cultivated legume, grown in 19 sites, followed by Vigna umbellata in 16 sites (Fig. 1). The farmer's preference might be the reason for the prevalence of Vigna unguiculata, and V. umbellata. The legumes were cultivated alongside numerous crop plants in the same plot (Table 1). The mix-cropping system is more resilient to biotic and abiotic stress, providing

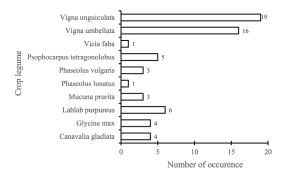


Fig. 1. Bar graph showing the occurrence of each crop legume cultivated on the 26 sites surveyed.

 Table 1. List of crop legumes and other crops cultivated in Jhum fields and home gardens.

Sl. No.	Collection site and location (district, GPS coordinates, and altitude)	Crop legume	Other crops	
1	Longkhum (JF), Mkg E 94°25'40.95", N 26°16'44.38", 1119 msl	V. unguiculata	Tomato, chilli, ladyfinger, maize, ginger, pumpkin, sponge gourd	
2	Longkhum (JF), Mkg E 94°25'23.14", N 26°16'42.52", 1117 msl	V. unguiculata	Maize, rice, Job's tears millet, bottle gourd, sponge gourd, colacass (Taro), perilla	
3	Mangmetong (HG), Mkg E 94°23'49.47", N 26°11'07.63", 1115 msl	L. purpureus, V. umbellata	Chilli, maize, ginger, chives, Elsholtzia griffithii	
4	Mangmetong (HG), Mkg E 94°24'13.78'', N 26°18'03.34'', 1135 msl	P. vulgaris, V. unguiculata	Ginger, chilli, colacassia.	
5	Mangmetong (HG), Mkg E 94°24'01.44", N 26°17'30.50", 1110 msl	P. vulgaris, L. purpureus, M. prurita	Chilli, ginger, chives, colacassia	
6	Mangmetong (HG), Mkg E 94°23'49.66", N 26°17'23.86", 1125 msl	M. prurita, V. umbellata, V. unguiculata	Ginger, chilli, colacassia, maize, Elsholtzia griffithii	
7	Mangmetong (HG), Mkg E 94°24'20.78'', N 26°16'42.16'', 1096 msl	V. unguiculata	Ginger, chilli, colacassia	
8	Minkong (HG), Mkg E 94°33.792', N 26°21.756', 1302 msl	P. tetragonolobus, V. unguiculata	Maize, ginger, chilli, chives, colacassia	
9	Mongchen (JF), Mkg E 94°29'7.82", N 26°29'7.86", 392 msl	L. purpureus, M. prurita, P. tetragonolobus, V. umbellata	Colacassia, tapioca, ginger, bitter gourd, chives, chilli	
10	Impur (HG), Mkg E 94°32'52.87", N 26°23'07.85", 1096 msl	C. gladiate, V. umbellata, V. unguiculata	Chilli, chives, ginger, <i>Elsholtzia</i> blanda, maize	
11	Lumami (HG), Zbt E 94°26.944', N 26°22.028', 958 msl	C. gladiata, P. lunatus, P. tetragonolobus, V. faba, V. umbellata, V. unguiculata	Maize, ginger, chilli, Clerodendrum colebrookianum	
12	Lumami (JF), Zbt E 94°28'27.186'', N 26°12'13.482'', 940 msl	C. gladiata, G. max, P. tetragonolobus, V. umbellata, V. unguiculata	Ginger, Clerodendrum colebrookianum, colacassia, chilli, Solanum indicum	
13	Alaphumi (HG), Zbt E 94°29'06.27", N 26°11'31.92", 1067 msl	L. purpureus, V. umbellata V. unguiculata	Maize, ginger, chives	
14	Akuluto (HG), Zbt E 94°29'46.55", N 26°10'14.22", 1125 msl	C. gladiata, P. tetragonolobus,	Ginger, chilli	
15	Sumi Settsu (HG), Zbt E 94°28'24.24", N 26°12'47.52", 1081msl	L. purpureus, V. umbellata, V. unguiculata	Ginger, tomato, chilli, maize, cucumber	
16	Sumi Settsu (JF), Zbt E 94°28'38.38", N 26°15'18.56", 1044 msl	V. umbellata, V. unguiculata	Rice, maize, colacassia, cucumber, chilli, tomato, ginger	
17	V.K (JF), Zbt E 94°24'56.78", N 26°10'02.64", 878 msl	V.umbellata, V. unguiculata	<i>Clerodendrum colebrookianum,</i> rice, colacassia, maize	
18	Awotsakilimi (JF), Zbt E 94°31'27.38'', N 26°08'36.42'', 1514 msl	L. purpureus, V. umbellata	Maize, rosella, colacassia, ginger, amaranthus, job's tear millet, chives, pumpkin	
19	Pangti (JF), Wka E 94°18'50.40'', N 26°14'23.85'', 470 msl	G. max, V. unguiculata	Tapioca, <i>Clerodendrum</i> <i>colebrookianum</i> , maize, rice, colacassia.	
20	Okotso (JF), Wka E 94°21'11.84'', N 26°16'47.33'', 654 msl	V. unguiculata	Tapioca, <i>Clerodendrum</i> colebrookianum, maize, rice, colacassia	
21	Anatongre (JF), Kphr E 94°49.973', N 25°57.973', 1220 msl	V. umbellata, V. unguiculata	Maize, rice, colacassia.	
22	Phelungre (JF), Kphr E 94°48.821', N 25°55.298', 1079 msl	G. max, P. vulgaris, V. unguiculata, V. umbellata	Maize, sweet potato, colacassia, tapioca	
23	Keor (JF), Kphr E 94°44.054', E 25°55.326',1664 msl	V. umbellata V. unguiculata	Rice, maize, colacassia, Job's tears millet, Pearl millet	
24	Singrep (HG), Kphr E 94°45.887', E 25°55.195', 1955 msl	V. umbellata	Potato, maize, colacassia	

#### Table 1. Continued.

Sl. No.	Collection site and location (district, GPS coordinates, and altitude)	Crop legume	Other crops
25	Tangnyu (JF), Mon E 95°4.987', E 26°42.361', 793 msl	V. unguiculata	Rice, colacassia, tapioca, maize.
26	Kenchenshu (JF), Mon E 94°57.823', E 26°26.430', 1017 msl	G. max, V. umbellata	Maize, tapioca, colacassia.

Abbreviations: JF-Jhum field; HG-Home garden; Mkg (Mokokchung), Zbt (Zunheboto), Wka (Wokha), Kphr (Kiphire) and Mon district; msl-mean sea level.

sustainable crop yield and preserving genetic diversity (Singh and Singh 2017). These data supported the prevalence of high crop diversity in traditional Jhum and home garden agricultural practicing regions, including Nagaland.

Nodulation status has been shown to be important for the productivity of legume crops Goyal *et al.* (2021). Root nodules were found in all 9 crop legumes except for *Vicia faba* (Table 2), where root nodules were absent. Although the crop legumes are

well nodulated, the number of nodules per rootlet varies widely depending on the species (Table 2). The root nodule number and size are influenced by environmental, climatic, soil parameters, and genetic factors (Atieno and Lesueur 2018). The root nodules were of determinate type with a spherical to globose shape in all the nine crop legumes (Table 2). The nodule size ranges from  $5.15 \pm 0.617$  mm in *Canavalia gladiata* to  $1.45 \pm 0.610$  mm in *Vigna umbellata* (Table 2). Nodules, when cut open, were red, indicating the presence of leghaemoglobin. The symbiotic

Table 2. Nodule morphology and characteristics.

Crop legume	Nodule type	Mean nodule size $(mm) \pm SD$	Nodule color	Lb	Nodules /rootlet
Canavalia	Determinate, less spherical				
gladiata	nodules with lenticels on secondary roots	$5.15\pm0.617$	Brown	Positive	9-20
Glycine max	Determinate, globose nodules on secondary and tertiary roots	$3.50\pm0.500$	Brown	Positive	5-11
Lablab	Determinate, less spherical				
purpureus	nodules with lenticels on secondary roots	$3.40\pm0.528$	Brown	Positive	8-15
Mucuna	Determinate, less spherical				
prurita	nodules with lenticels on secondary and tertiary roots	$4.83\pm0.289$	Reddish to brown	Positive	10-15
Phaseolus	Determinate, less spherical				
lunatus	nodules with lenticels on secondary and tertiary roots	$3.30\pm0.167$	Brown	Positive	5-14
Phaseolus vulgaris	Determinate, globose nodules on secondary roots	$3.33\pm0.577$	Brown	Positive	6-18
Psophocarpus tetragonolobus	Determinate, globose nodules on secondary and tertiary roots	$4.93\pm0.115$	Whitish to brown	Positive	6-12
Vigna umbellata	Determinate, globose nodules on secondary and tertiary roots	$1.45\pm0.610$	Whitish to brown	Positive	6-10
Vigna	Determinate, less spherical				
unguiculata	nodules with lenticels on secondary and tertiary roots	$4.27\pm0.649$	Whitish to brown	Positive	10-20
Vicia faba	absent	-	-	-	-

Note: Nodule size is the mean of 10 plants, with a minimum of 10 nodules taken for measurement from each plant. While the number of nodules is counted from 10 plants for each species. The presence of reddish pigment inside the root nodule is taken as positive for leghaemoglobin (Lb). SD: Standard deviation.

relationship between the legume and rhizobia occurs due to the signaling exchange, regulated explicitly by the Rhizobial nod genes, which determine host specificity, infection, and nodulation Dénarié et al. (1992). Since rhizobia genetics determined host range variation in nodulation, the lack of root nodules on Vicia faba might be due to the absence of compatible rhizobia in the sampling sites. To further characterize the root nodule endophytes, six bacterial isolates were obtained from the root nodules of V. unguiculata (4 isolates), V. umbellata (1 isolate), and P. vulgaris (1 isolate), the three popularly cultivated species. Sequence analysis of the Recombinase A (recA) gene confirmed that the isolates belonged to Rhizobium sp. The sequences were submitted to the National Center for Biotechnology Information (NCBI) GenBank with accession numbers MT796838, PP379910, MT796839, PP379911, MT796836, and MT796837.

### CONCLUSION

The present study underscores the prevalence and importance of crop legumes cultivated by the ethnic tribes of Nagaland. Traditional practices of Jhuming and home gardens help maintain crop diversity. Further, root nodules in the plants suggested successful symbiosis between the crop legume and native rhizobia. The absence of root nodules on *Vicia faba* suggested more studies on the nodulation status of other introduced crop legumes. The Rhizobium and legume symbiosis represent the best source for fertilizers and, therefore, can be a great opportunity for future research in developing potential inoculants for crop productivity. Increasing pulse production can expand food and nutritional security in Nagaland.

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