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Study on the diversity of Plant Growth Promoting Bacteria (PGPR) from Selected Medicinal Plants of Mokokchung, Nagaland, India

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ABSTRACT: Plant growth promoting bacteria (PGPR) have been found to be highly beneficial for plants as they help defend against fungal diseases commonly found in soil. They play an important role in plant growth, health and productivity. They increase seedling tolerance to drought, high temperatures, toxic heavy metals, high or low pH and even extreme soil acidity. The present study aim to isolate and characterization of beneficial PGPR for the growth of 17 PGPR isolates, except for isolate 1 and isolate 2 all of them observed a pink color thus indicating binding of iron. Thus, indicating production of *Siderophore.* All the 17 isolates observed a pink and reddish color thus indicating production of IAA. Most of the isolates showed positive result against ammonia production, phosphate solubilization test and ammonia production. This work highlights the future research works by the use of beneficial strains of PGPR to cultivate rare medicinal plants Mokokchung, Nagaland.

Keywords: PGPR, Ao Naga tribe, Soil acidity, medicinal plants, organic biofertilizer.

Introduction:

The Ao Naga tribe has been using medicinal plants for centuries, a custom that has been passed down from generation to generation. These methods served as the primary means of treating a wide range of illnesses long before civilization emerged (Petrovska, 2012, Huang, *et al.,* 2018). However, because humans use these plants so extensively for their own health, most of them have been overused, and precautions must be made to ensure that these therapeutic plants are used for a long time to come (Vejan, *et al*., 2016). This work uses plant growth to highlight the protection and application of some of the many medicinal plants found in Mokokchung, Nagaland. These plants are highly vulnerable to extinction because of the urbanisation of these wild areas brought about by population growth (Attitalla, *et al*., 2020; Vaghela, and Gohel,2023)

Bacteria that colonize plant roots and promote plant growth are referred to as plant growth-promoting rhizobacteria (PGPR). PGPR are quite diverse, with effects ranging from local antagonism to soil-borne pathogens to induction of systemic resistance against pathogens throughout the plant. When the inducing bacteria and the challenging pathogen are spatially separated, induced systemic resistance (ISR) in plants resembles pathogen-induced systemic acquired resistance (SAR) (Beneduzi, *et al*.,2012).

The rhizospheric bacteria established a complex mutualistic symbiotic network, which is favourable to the growth and development of medicinal plants. Host development stage and environmental conditions have a significant impact on the composition, diversity, community structure, and function of plant rhizosphere bacteria (Saeed, *et al*., 2021, Walusansa, *et al*.,2022). As a result, PGPR-mediated medicinal plant growth augmentation and bioactive phytochemical yield optimization opens up some new avenues in the development of low-cost herbal

pharmaceuticals, which may benefit the public in the face of health-care system failures (Rizvi, 2022).

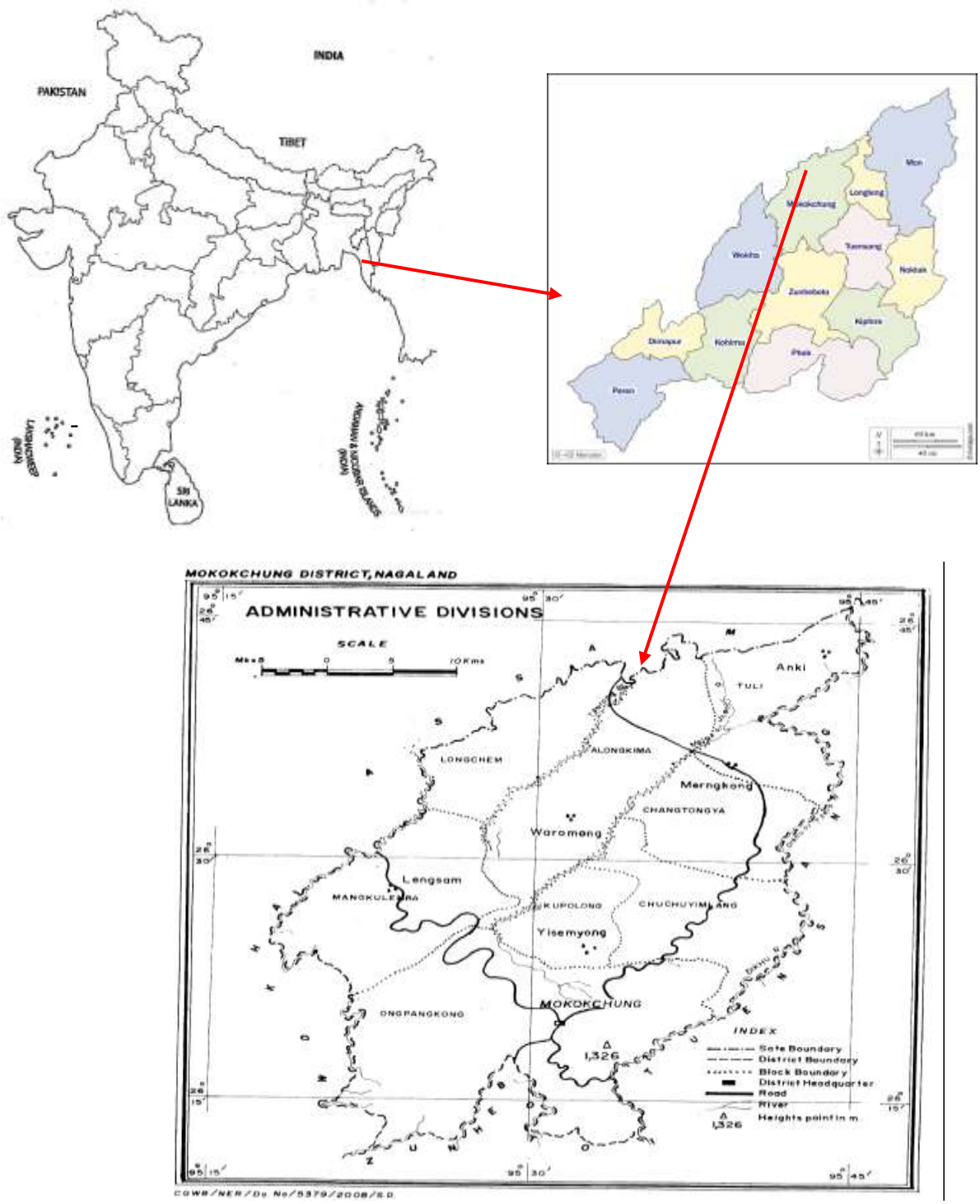
Materials and Methods:

1. Location of the study site.

The region of Ao Naga, Mokokchung District, is found in the northwest of Nagaland State. It covers an area of 1,615 sq. km and is bordered by 26.12° and 26.45° North Latitudes and 94.18° and 94.50° East Longitudes, respectively. The district's elevation ranges from 155 to 2,000 meters above mean sea level (AMSL). The district's administrative center, Mokokchung town, is located at a height of roughly 1,325 m AMSL. The district has been divided into 6 R. D. Blocks and 8 Circles. (Ground Water Information Booklet Mokokchung District, Nagaland 2013)

2. Climate of the study site.

The climate in the district is humid subtropical. The district's yearly average rainfall is 2,500 mm. The months of June and July have the most rainfall. Rainfall often begins in April and lasts until the end of September. The climate in the area is frigid in the winter and moderate in the summer. The coldest months are January and February, when the nighttime temperature drops to 2℃. Even in the summer, it is not hot; rather, it is cool in comparison to the Assam plains. The temperature does not get above 32℃ throughout the summer, and the average summer temperature is 27℃.The average summer temperature is 27℃, and it seldom rises above 32℃.



Photoplate 1. Map showing India map and the study site (Photo Courtesy: Google Source)

3. Vegetation of the study area.

In the district of Mokokchung, along the Assam-Nagaland border**,** Northern Tropical Semi Evergreen Forest (Champion and Seth 1968) kinds of forests can be found. These woodlands' plant life is comparable to that of the Northern Tropical Wet Evergreen woodlands. The only difference is that while there are deciduous species like Bhelu (*Tetrameles nudiflora*), Paroli (*Stereospermum chelonoides*), Jutuli (*Altingia excelsa*), etc., the evergreen species predominate in the former case while there are more deciduous species in the latter.

4. Selection of the medicinal plants:

Three rare medicinal plants are taken for the study of the diversity of PGPR which are (i).Asogshi (Asong),(ii).Wara Komo (Local name), and (iii) Pi Pi Waa (*Drymaria cordata Willd*). The first two plants are under the process of taxonomical classification to get the accession from BSI. Shillong (**Photoplates 1 and 2**).

5. Isolation and characterization of Bacteria

The bacteria was isolated on Nutrient Agar medium (NA) using serial dilution method (Ben-David and Davidson, 2014) followed by spread plate method (Wise, 2006). Number of valuable bacteria in soil will be expressed as colony forming units (CFU) in gram of dry weight soil. Plates of bacteria will be incubated for 24-48 hours. A total of 17 Pure Culture of PGPR isolates was carried out by Katz (2008).

6. Cultural and morphological features of the bacteria isolates.

The cultural and morphological features fall under the phenotypic characterization, which were studied by adopting standard methods. (Goodfellow, 1989). Bacterial colonies were screened for siderophore production (Hettiarachchi, *et. al.,* 2017), IAA production (Gang, *et. al.,* 2019),

Ammonia production (Cappuccino and Sherman, 1992), Phosphate solubilization test (Sousa, *et. al*., 2016), Hydrogen cyanide (HCN) production (Neerincx, *et. al*.,2016) and Nitrogen Fixation Assay (Burris, 1972).

Plant Specimen 1

Plant Specimen 2

Plant Specimen 3

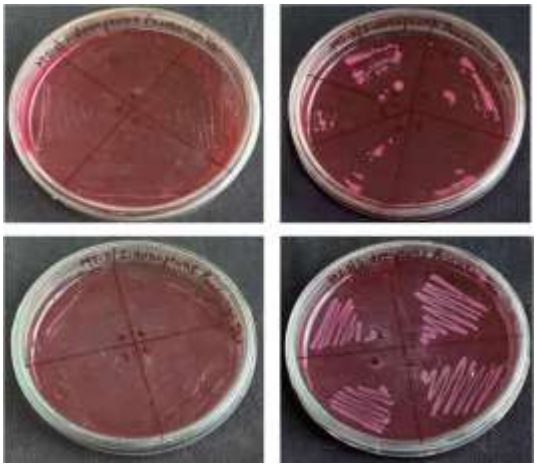
Photoplate 2: Selected rare medicinal Plants of Mokokchung, Nagaland

Results and Discussion:

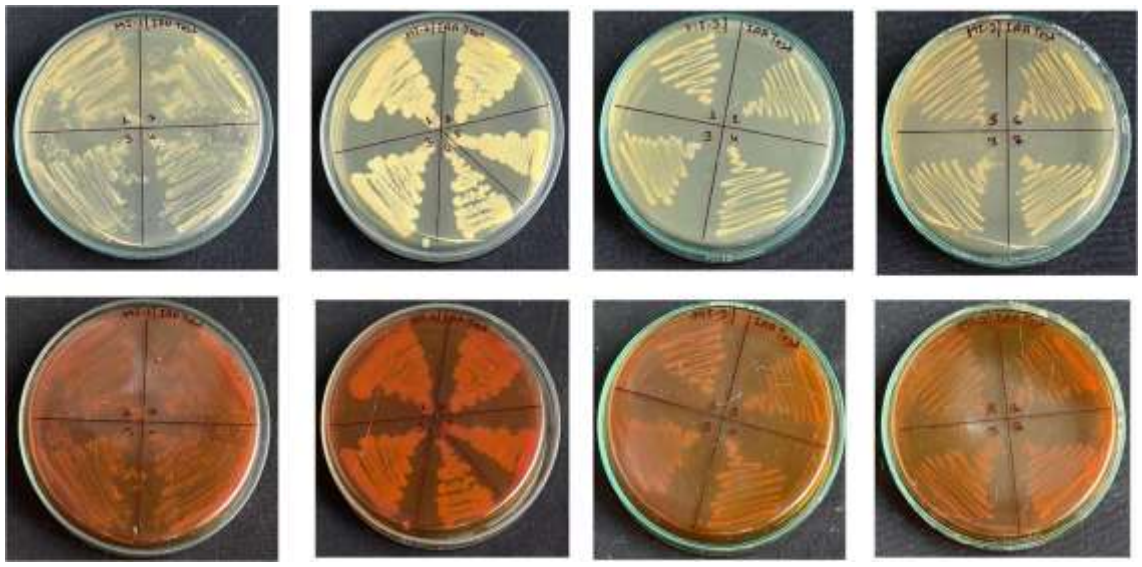
Table 1: Biochemical characteristics of selected resistant PGPR isolates.

PGPR isolates.	Siderophore Production	IAA Production	Ammonia Production	HCN Production	Phosphate solubilization test	Nitrogen Fixation
1	-	+	+	-	-	+
2	-	+	+	-	-	+
3	+	+	+	-	-	+
4	+	+	+	-	-	+
5	+	+	+	-	+	-
6	+	+	+	-	-	+
7	+	+	+	-	+	+
8	+	+	+	-	+	+
9	+	+	+	-	+	+
10	+	+	+	-	-	+
11	+	+	+	-	-	-
12	+	+	+	-	-	+
13	+	+	+	-	-	+
14	+	+	+	-	-	-
15	+	+	+	-	-	-
16	+	+	+	-	-	+
17	+	+	+	-	-	-

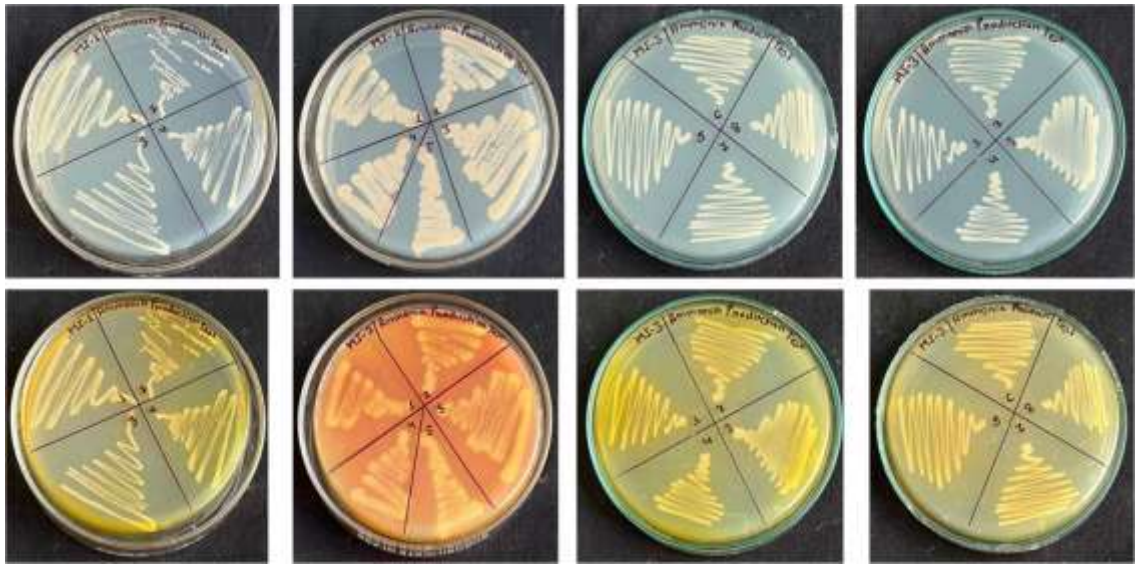
+ve =‘+’=Positive;’–‘ = Negative; IAA=Indole Acetic acid, HCN=hydrogen cyanide



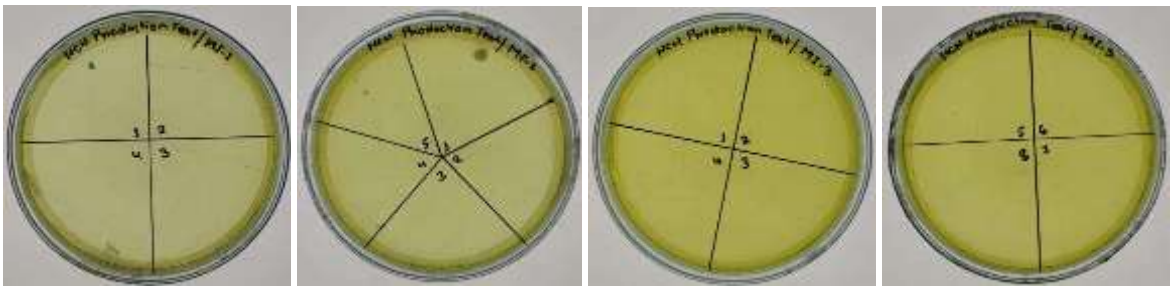
Photoplate 3: Observation of s i d e r o p h o r e production from PGPR colonies



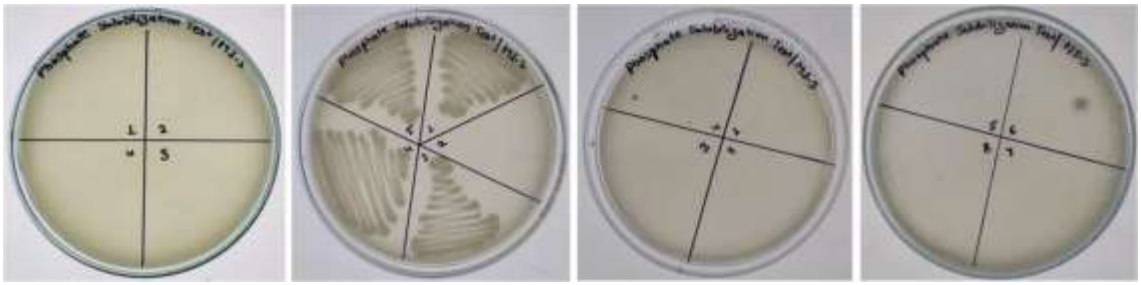
Photoplate 4: Observation of I A A production from PGPR colonies



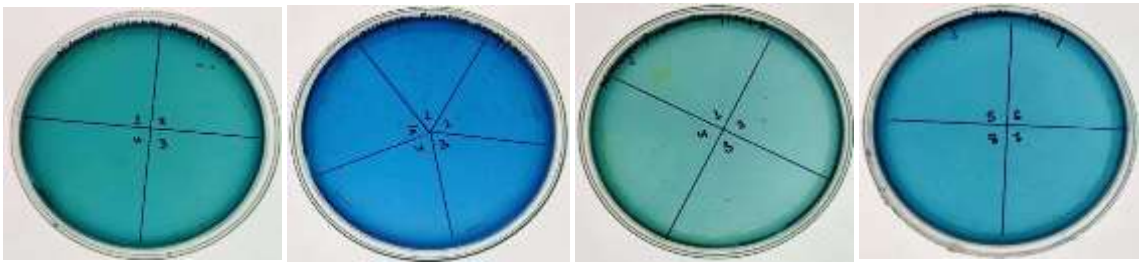
Photoplate 5: Observation of A m m o n i a production from selected PGPR isolates



Photoplate 6: Observation of Hydrogen Cyanide (HCN) production from selected PGPR isolates



Photoplate 7: Observation of Phosphate Solubilization test from selected PGPR isolates



Photoplate 8: Observation of Nitrogen fixation assay from selected PGPR isolates

Out of 17 PGPR isolates, Except for isolate 1 and isolate 2 all of them observed a pink color thus indicating binding of iron. Thus, indicating production of *Siderophore.* All the 17 isolates observed a pink and reddish color thus indicating production of IAA. Numerous studies also showed the positive result of beneficial PGPR strains against siderophore and IAA production for the cultivation of economically important crops (Deb and Tatung,2024, Ganesh, *et al*., 2024, Olagunju, *et al*., 2025) (**Table 1**) (**Photoplates 3 and 4**)

All the 17 isolates observed produced a yellow color thus indication of Ammonia production. No HCN production was observed for all the 17 isolates (Cristescu, 2015). Among all the samples, isolate no 5,7,8,9, showed Phosphate Solubilization whereas the rest shows negative result. Most of the isolates showed positive result against Nitrogen fixation except isolate no 5,11,14,15 and 17 (Aasfar, *et al*., 2024). (**Photoplates 5-8**)

Conclusion:

Plant growth promoting rhizobacteria (PGPR) play a significant part in the industry of sustainable agriculture. Increasing crop output demand while reducing the usage of synthetic chemical fertilizers and pesticides is a major challenge today. The usage of PGPR has been shown to be an environmentally friendly method of enhancing agricultural yields by encouraging plant development via a direct or indirect process. Furthermore, PGPR exhibits both synergistic and antagonistic interactions with microorganisms in the rhizosphere and in bulk soil, which indirectly increases plant growth rate. The isolated PGPR strains will be helpful for possible application as an organic biofetilizer for the cultivation of rare and endangered medicinal plants of Nagaland, India.

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