Plant Growth Promoting Rhizobacteria For Abiotic Stress Tolerance And Agricultural Sustainability

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Outlines

- Introduction
- PGPR and their mechanism of action
- Rice roots-\textit{Bacillus amyloliquifaciens} interaction
- \textit{Pseudomonas putida}-responsive small RNA profiling in chickpea
- Summary
- Acknowledgements
Abiotic stresses affect both plants and animals, but being sessile, plants are especially affected by environmental factors.

Abiotic stress results in yield losses ranging from 10% to 50% or more, depending on the crop.

Drought, salinity and temperature extremes are among the important abiotic stresses.
66% of people will live in cities by 2050, compared to 54% in 2014.

50% increase in food demand by 2050.

31.8% drop in agricultural workers in developed countries since 1950.

32.8% drop in agricultural workers in developing countries.

3 billion more mouths to feed by 2050.

We will need major innovations in how we eat and farm.

Source: FAO
What are PGPR?

- A group of soil born bacteria (Rhizobacteria)
- Actively colonize plant roots/rhizosphere
PGPR enhance plant growth and yield, either by:

- Direct:
  - By production of growth hormones, phosphate solubilization, nitrogen fixation etc.,
- Indirect:
  - By suppression of deleterious microorganisms or secretion of antifungal metabolites.

**Mechanism of action of PGPR**

- **Biofertilizer**
  - Biological nitrogen fixation
  - Solubilization of insoluble potassium
  - Phosphate solubilization
  - Production of auxin (mainly indole-3-acetic acid)
  - Zeatin production (cytokinin)
  - Production of GA3 (giberellin)
  - Production of abscisic acid (ABA)
  - Production of ethylene

- **Biologic control**
  - Production of compounds (antibiotics and antifungal)
  - Volatile organic compounds (VOC) synthesis and liberation
  - Competitive exclusion and predation
  - Acquired and induced systemic resistance (ASR and ISR)
  - Production of lytic enzymes (protease, cellulase and others)
  - ACC deaminase activity (low ethylene levels)
  - Antifreeze proteins
  - Antioxidant enzymes (mainly SOD, CAT and POX)

- **Tolerance to stress**
  - Proline and quaternary amine biosynthesis
Based on functional activities PGPR are classified in:

- Biofertilizers
- Phytostimulators

The principle rhizobacterial genera known to act as PGPR include Agrobacterium, Arthrobacter, Azotobacter, Azospirillum, Bacillus, Caulobacter, Chromobacterium, Flavobacterium, Micrococcous, Pseudomonas and Serratia etc.
How PGPR affects plants?

Tiwari et al. 2016; Plant Physiology and Biochemistry
**Bacillus amyloliquefaciens (NBRI-SN13):**

A PGPR, isolated from alkaline soil of Banthara, Lucknow at CSIR-NBRI.

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>GenBank accession</td>
<td>Bacillus amyloliquefaciens KC293995</td>
</tr>
<tr>
<td>Motility</td>
<td>75 mm ± 3.0</td>
</tr>
<tr>
<td>Auxin production</td>
<td>35.6 ± 0.55 µg/ml</td>
</tr>
<tr>
<td>Phosphate solubilization</td>
<td>18.5 ± 0.09 µg/ml</td>
</tr>
<tr>
<td>ACC deaminase activity</td>
<td>168.5 ± 6.97 nmol alpha ketobutyrate/mg protein/h</td>
</tr>
<tr>
<td>NaCl tolerance</td>
<td>Up to 2500 mM</td>
</tr>
<tr>
<td>Temperature</td>
<td>Up to 45 °C</td>
</tr>
<tr>
<td>Drought tolerance</td>
<td>60%</td>
</tr>
<tr>
<td>pH tolerance</td>
<td>5.0–9.0</td>
</tr>
</tbody>
</table>

**Proline accumulation:**

<table>
<thead>
<tr>
<th>NaCl Concentration</th>
<th>Proline Accumulation (µM ± SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 mM NaCl</td>
<td>61.05 ± 5.61 µM</td>
</tr>
<tr>
<td>250 mM NaCl</td>
<td>80.46 ± 4.3 µM</td>
</tr>
<tr>
<td>500 mM NaCl</td>
<td>91.52 ± 0.88 µM</td>
</tr>
<tr>
<td>1000 mM NaCl</td>
<td>101.25 ± 0.82 µM</td>
</tr>
</tbody>
</table>

*Nautiyal et al. 2013; Plant Physiology and Biochemistry*
Investigations on the effects of NBRI-SN13 (*Bacillus amyloliquefaciens*) inoculation on various abiotic stresses and phytohormone treatments in rice.

**LIPID PEROXIDATION**

**GENE EXPRESSION**
All abiotic stresses and phytohormone treatments significantly affected various physiological and biochemical parameters and also positively modulated stress-responsive gene expressions suggesting its multifaceted role in cross-talk among stresses and phytohormones.

Tiwari et al. 2017; Frontiers in Plant Science
Transcript profiling of rice under salt stress in the presence and absence of *Bacillus amyloliquefaciens* (NBRI-SN13)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Shoot length (cm.)</th>
<th>Root length (cm.)</th>
<th>Fresh weight (mg.)</th>
<th>Dry weight (mg.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>16.25±1.7</td>
<td>6.475±1.25</td>
<td>116.35±14.9</td>
<td>5.55±1.2</td>
</tr>
<tr>
<td>salt</td>
<td>8.575±0.85</td>
<td>7.65±0.73</td>
<td>57.225±5</td>
<td>5.725±0.59</td>
</tr>
<tr>
<td>SN13</td>
<td>17.45±0.98</td>
<td>8±0.67</td>
<td>223±13.8</td>
<td>10.525±1.28</td>
</tr>
<tr>
<td>Salt+SN13</td>
<td>9.25±0.47</td>
<td>6.425±0.72</td>
<td>75.975±9.7</td>
<td>7.275±1.4</td>
</tr>
</tbody>
</table>

Chauhan et al. 2019; Scientific Reports
Our results indicate qualitative and quantitative differences between rice roots responses to SN13 under salt stressed and unstressed conditions, based on both morpho-physiological and transcriptomic data.
The mechanism of rice-SN13 interaction
Our results indicated that yeast cells expressing both genes showed enhanced tolerance to osmotic stresses and better survival at higher temperatures as compared to the strain carrying empty vector.
Genome-wide investigation and expression analysis of the GRAM-domain family in rice and *Arabidopsis*

**Physical map of GRAM-domain containing genes**

*Oryza sativa*

Physical mapping on chromosomes revealed uneven distribution of *OsGRAM* genes in rice genome

Tiwari et al. 2019; Int. J. Biol. Macromol. (Accepted)
Expression profile of candidate \textit{GRAM} genes in response to abiotic stress and phytohormone treatments in the presence of SN13

\textit{Oryza sativa}

Variance observed in gene expression patterns suggested the multifaceted role of \textit{OsGRAM} gene in regulating stress-responsive pathways for tolerance and adaptation towards multiple abiotic stresses.
**Pseudomonas putida (NBRI-RA):**
A PGPR, isolated from desert regions of Rajasthan and submitted to IMTECH, Chandigarh with depository no. MTCC5279

- MTCC5279 shows growth in the presence of up to 60% PEG and 500 mM NaCl.
- It produces Auxin and solubilized insoluble tri-calcium phosphate (TCP)

**Pseudomonas putida (MTCC5279)**

<table>
<thead>
<tr>
<th>Trait</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>IAA</td>
<td>48.69±1.66µg/ml</td>
</tr>
<tr>
<td>Phosphate solubilisation</td>
<td>117.68±2.61µg/ml</td>
</tr>
<tr>
<td>Siderophore</td>
<td>+</td>
</tr>
<tr>
<td>ACC deaminase</td>
<td>+</td>
</tr>
</tbody>
</table>

Srivastava et al. 2012, Plant Signaling & Behavior
Positive modulation of stress responsive gene expression in Kabuli type chickpea cultivar, BG-1003 which is generally more sensitive to drought stress as compared to desi BG-362, suggested its greater potential in enhancing agricultural yield of this economically important legume.

Tiwari et al. 2016; Plant Physiology and Biochemistry
Genome-wide profiling reveals extensive alterations in *Pseudomonas putida* mediated miRNAs expression during drought stress in chickpea

- 923 conserved and 216 novel miRNAs were identified
- Conserved miRNAs were categorized into 24 miRNA families

![Bar chart showing the number of miRNA members per family](chart.png)
GO analysis of the predicted targets of chickpea miRNAs showed its diverse function in biological processes, cellular component and molecular functions.

Jatan et al. 2019; Environmental & Experimental Botany
Inverse correlation in the expression patterns of miRNAs with their respective targets indicating the involvement of RA in amelioration of drought stress in chickpea.

Jatan et al. 2019; Environmental & Experimental Botany
PGPR are economically and environmentally beneficial for plant growth promotion.

PGPR alter physio-biochemical as well as molecular mechanism of plants to withstand adverse environmental conditions.

Transcriptome analysis and functional characterization of selected genes indicated rice roots-SN13 cooperation under salt stress conditions.

Genome wide study suggested the multifaceted role of OsGRAM gene in regulating stress-responsive pathways for tolerance and adaptation towards multiple abiotic stresses.

Gene expression patterns of miRNAs and their respective targets indicate the involvement of RA in amelioration of drought stress in chickpea.

Therefore, it can be concluded that SN13 and RA can be formulated and used as biofertilizers and abiotic stress busters in rice and chickpea, respectively.
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Thank You