

ANNEXURE

Annexure-I. List Of Abbreviations Used:

α = Alpha

β = Beta

- = minus

% = percentage

μ = micro

μg = microgram

μl = microlitre

μM = micromolar

$(\text{Ca}_3)_2\text{PO}_4$ = Tri Calcium Phosphate

$(\text{NH}_4)_2\text{SO}_4$ = Ammonium Sulphate

A. flavus = *Aspergillus flavus*

A. thaliana = *Arabidopsis thaliana*

ACC = 1-Aminoacyclopropane 1-Carboxylic acid

ANOVA = Analysis of Variance

Ar = Absorbance of reference

ARA = Acetylene Reduction Assay

As = Absorbance of sample

ATP = Adenine Triphosphate

B. siamensis = *Bacillus siamensis*

B. subtilis = *Bacillus subtilis*

BLAST = Basic Local Alignment Search Tool

BSL = Brine Shrimp Lethality

C_2H_4 = Acetylene

Ca = Calcium

CaCO_3 = Calcium Carbonate

CaOx = Calcium Oxalate

CAS = Chrome Azurol S

CaSO₄.2H₂O = Calcium Sulphate

CFU = Colony Forming Unit

cm = centimeter

Da = Dalton

DAI = Day after inoculation

DNA = Deoxyribo Nucleic Acid

DPPH = 1, 1- diphenyl-2-picrylhydrazyl

EEAA = Ethanol Extract of *Achyranthes aspera*

F =Forward

F. graminearum = *Fusarium graminearum*

Fe = Iron

FeCl₃ = Ferric Chloride

FeSO₄.7H₂O = Ferrous Sulphate

gm = gram

g = gravitation

G. diazotrophicus = *Gluconacetobacter diazotrophicus*

GA = Gibberellic Acid

GC-FID = Gas Chromatography Flame Induction Detector

GFP = Green Fluorescent Protein

gr = Glutathione Reductase

h = hour

ha = hacter

HDTMA = HexaDecylTrimethyl Ammonium

i.e. = that is

IAA = Indole Acetic Acid

IAM = Indole-3-acetamide

IC = Inhibition Concentration

IPyA = Indole 3-pyruvate

K_2HPO_4 = Di potassium hydrogen phosphate

KCl = Potassium Chloride

Kg = kilogram

KH_2PO_4 = Potassium dihydrogen phosphate

Km^r = Kanamycin Resistance

L = Linn

LB = Luria Bertani

LDH = Lactate Dehydrogenase

LDL = Low Density Lipid

Lux = Light Intensity

m = meter

MEGA = Molecular Evolutionary Genetic Analysis

mg = milligram

$MgSO_4 \cdot 7H_2O$ = Magnesium Phosphate

min = minute

mM = millimoles

$MnSO_4 \cdot 2H_2O$ = Manganese Sulphate

MR = Methyl Red

MS = Murashige and Skoog

N_2 = Nitrogen

N_2FA = Nitrogen Fixing Ability

NA = Nutrient Agar

NaCl = Sodium Chloride

NaOH = Sodium Hydroxide

NB = Nutrient Broth

NCBI = National Center for Biotechnology Information

NFb = Nitrogen Free Medium

nm = nanometer

nmol = nanomole

NPK = Nitrogen Phosphate Potassium

O. D. = Optical Density

O₂ = Oxygen

°C = Degree celcius

P = Phosphate

P. aeruginosa = *Pseudomonas aeruginosa*

P. indica = *Pseudomonas indica*

P. putida = *Pseudomonas putida*

PCR = Polymerase Chain Reaction

PGP = Plant Growth Promotion

PGPR = Plant Growth Promoting Bacteria

pH = Hydrogen Ion Concentration

PSB = Phosphate Solubilizing Bacteria

R = Reverse

rev = revolution

RH = Reducing Humidity

RNA = Ribonucleic Acid

rpm = revolution per minute

RT PCR = Real time Polymerase Chain Reaction

S = Second

S. aquamarine = *Sporosarcina aquamarine*

S. griseus = *Streptomyces griseus*

S. marcescens = *Serratia marcescens*

S. mutabilis = *Streptomyces mutabilis*

SEML = Soil and Environmental Molecular Laboratory

SM = Succinic medium

Sod = superoxide dismutase

sp. = species

SPSS = Statistical Package for Social Science

Tryp = Tryptophan

VP = Voges Proskauer

w/v = weight by volume

WHO = World Health organization

YEMA = Yeast Extract Mannitol Agar

YEMB = Yeast Extract Mannitol Broth

Annexure- II. Media And Reagents Used

(A) MEDIA

1. Dworkin and Foster minimal medium (g/l)

KH ₂ PO ₄	4.0
Na ₂ HPO ₄	6.0
MgSO ₄ .7H ₂ O	0.2
Glucose	2.0
Gluconic acid	2.0
Citric acid	2.0
FeSO ₄ .7H ₂ O	1 mg
H ₃ BO ₃	10 mg
MnSO ₄ .H ₂ O	11.19 mg
ZnSO ₄ .7H ₂ O	124.6 mg
CuSO ₄ .5H ₂ O	78.22 mg
MoO ₃	10 mg
pH- 7.2	

2. Luria Bertani (LB) (g/l)

Casein enzymic hydrolysate	10.0
Yeast extract	5.0
NaCl	10.0

3. Murashige and Skoog medium (MS Medium) (mg/l)

Major salts

NH ₄ NO ₃	1,650
CaCl ₂ .2H ₂ O	440
MgSO ₄ .7H ₂ O	370
KH ₂ PO ₄	170
KNO ₃	1,900

Minor salts

H ₃ BO ₃	6.2
CoCl ₂ · 6H ₂ O)	0.025
CuSO ₄ · 5H ₂ O	0.025
FeSO ₄ · 7H ₂ O	27.8
MnSO ₄ · 4H ₂ O	22.3
KI	0.83
Na ₂ MoO ₄ · 2H ₂ O)	0.25
ZnSO ₄ ·7H ₂ O)	8.6
NaFe-EDTA	5
FeSO ₄ ·7H ₂ O	5.57 g
Na ₂ -EDTA	7.45 g .

Vitamins and organics

Myo-Inositol	100
Nicotinic Acid	0.5
Pyridoxine-HCl	0.5
Thiamine-HCl	0.1
Glycine	2
Indole Acetic Acid	1-30
Kinetin	0.04-10

4. Nitrogen free media (NFb) (g/l)

Mannitol	10
KH ₂ PO ₄	0.2
MgSO ₄ ·7H ₂ O	0.2
NaCl	0.2
CaSO ₄ ·2H ₂ O	0.1
CaCO ₃	5.0
pH-7	

5. Nutrient Agar (g/l)

Beef extract	10.0
Peptone	10.0
Sodium chloride	5.0
Agar	20.0
Distilled water	1000 ml
pH: 7.3±0.1 at 25°C	

6. Pikovskaya medium (g/l)

Glucose	10.0
(Ca ₃) ₂ PO ₄	5.0
(NH ₄) ₂ SO ₄	0.5
NaCl	0.2
MgSO ₄ ·7H ₂ O	0.1
KCl	0.2
FeSO ₄ ·7H ₂ O	0.002
Yeast extract	0.5
MnSO ₄ ·2H ₂ O	0.002
Agar	20
pH-7	

7. Succinic Medium (SM) (g/l)

K ₂ HPO ₄	6.0
KH ₂ PO ₄	3.0
MgSO ₄ ·7H ₂ O	0.2
(NH ₄) ₂ SO ₄	1.0
Succinic Acid	4.0
pH-7	

8. Yeast Extract Mannitol Agar (YEMA) (g/l)

Yeast extract	1.0
Mannitol	10.0
K ₂ HPO ₄	0.5
MgSO ₄ .7H ₂ O	0.2
NaCl	0.1
Agar	15.0
pH-7	

(B) REAGENTS**1. CAS reagent preparation:**

- Pour 6 ml of 10 mM strength HDTMA into 50 ml of sterile water
- Add one and half ml of Fe³ solution (1Mm FeCl₃.6H₂O + 10 mM HCl) and 7.5 ml 2mM aqueous CAS solution, turning the solution into dark blue.
- Make the final volume to 100 ml.

2. Peptone water (g/l)

Peptic digest of animal tissue	10.0
Sodium chloride	5.0
Distilled water	1000 ml
pH: 7.3±0.1 at 25°C	

3. Phosphate buffer saline (g/l)

Sodium chloride	8
Potassium chloride	0.20
Di-sodium hydrogen phosphate	1.15
Potassium di-hydrogen phosphate	0.20
Distilled water	1000 ml
pH: 7.2	

3. Salkowski reagent preparation:

- Preparation of 50 ml of 35% perchloric acid
- Preparation of 10 ml of 0.5M FeCl₃
- Add 1 ml of 0.5M FeCl₃ to 50 ml of 35% perchloric acid.

4. Tris-borate EDTA (TBE) buffer (10X stock solution; g/l)

Tris Base	108.0
Boric acid	55.0
EDTA (pH=8.0)	20 ml
Distilled water	1000 ml

5. Tris-acetate EDTA (TAE) buffer (10X stock solution; g/l)

Tris-HCl	48.4g
Glacial acetic acid	11.42 ml
EDTA (pH=8.0)	20 ml

6. 6X loading dye/buffer (g/l)

Sucrose	40
0.04% bromophenol blue solution	50 ml

The final volume of the solution was adjusted to 100 ml.

(C) STAINING SOLUTIONS**1. Crystal Violet stain (g/l)****Solution A**

Crystal Violet	2.5
Ethanol (95%)	25 ml

Solution B

Ammonium Oxalate	1.0
Distilled Water	100 ml

Solution A and solution B were mixed, filtered and stored at room temperature.

2. Gram's Iodine solution (g/l)

Stock solution:

Iodine	5.0
Potassium iodide	10.0
Distilled water	100 ml

Working solution:

The above solution was diluted in 1:5 ratio with distilled water for preparing working solution.

3. Safranin stain (g/l)

Stock solution:

Safranin	2.5 gm
Ethanol (95%)	100 ml

Working solution:

The above solution was diluted in 1:4 ratio with distilled water for preparing working solution.

4. Ethidium bromide solution (10 mg/ml)

Ethidium bromide	0.1 gm
Distilled water	10 ml

The above reagent was dissolved in distilled water and stored in room temperature.