APPENDIX I

INSTRUMENTS	MAKE/ MANUFACTURER
Autoclave	Adhir Dutt and Co., India
Laminar flow hood	Toshiba, India
Micropipettes and tips	Tarson,
Inoculating needle	Tarson
L- Spreader	Tarson,
ELISA plate reader	MRX Revelation, DYNEX
Spectrophotometer	Rayleigh UV-2601
Incubator	Remi,
Phase contrast microscope	Olympus
Camera	MC Camera, Motic, Olympus, Cannon
Cooling centrifuge	Eppendorf
Table top centrifuge	Remi,
Electronic balance	Satorius
PH meter	Hanna
Vortex mixture	Biorad
-20°C freezer	Bluestar,
Refrigerator	Godrej, India
Magnetic stirrer	Remi,
Horizontal electrophoresis unit	Genei, India
Vertical electrophoresis unit	Biotech,
Electrophoresis power supply	Genei, India

UV trans-illuminator	Gibco BRL, Life technologies
Gel documentation system	Spectronics corporation
PCR machine	Eppendorf,
Water bath	Labman scientific instrument
Water distillation plant	Riviera,
Deionised water system	Millipore corporation, USA

APPENDIX II

Composition of different agars

Anaerobic Agar

Ingredients	Amount/1000ml
Trypticase	20g
Glucose	10g
Sodium chloride	5g
Sodium thioglycolate	2g
Sodium formaldehyde sulfoxylate	1g
Agar	15g
Distilled water	1000ml

Adjust the pH to 7.2 distribute into 15 mm test tubes in amounts sufficient to give a 75 mm depth of medium and sterilize by autoclaving at 121°C for 20min.

Nutrient Agar

Ingredients	Amount/1000ml
Nutrient agar	28g
Distilled water	1000 ml

Adjust pH 7.4 and autoclave at 121°C for 15 min.

Nutrient Broth

Ingredients	Amount/1000ml
Nutrient Broth	13g
Distilled water	1000 ml

Adjust pH 7.4 and autoclave at 121°C for 15 min.

Milk Agar

(For Casein hydrolysis)

Ingredients	Amount/1000ml
Casein/skimmed milk powder	100g
Peptone	5g
Agar	15g
Distilled water	1000 ml

Adjust pH 7.2 and autoclave at 121°C for 15 min.

Starch Agar

(For startch hydrolysis)

Ingredients	Amount/1000ml
Peptone	5g
Beef extract	3g
Starch soluble	2g
Agar	15g
Distilled water	1000 ml

Adjust pH 7.0 and autoclave at 121°C for 15 min.

Catalase Agar

(For catalase test)

Ingredients	Amount/1000ml
Tripticase	15g
Phyton/Teptron	5g
NaCl	5g
Agar	15g
Distilled water	1000 ml

Adjust pH 7.0 and autoclave at 121°C for 15 min.

Bacillus thuringiensis maintaining media

Ingredients	Amount/1000ml
Glucose	3g
Ammonium sulphate	2g
Yeast extract	2g
Potassium phosphate dibasic	0.5g
Magnesium sulphate hepta hydrate	0.2g
Calcium chloride dihydrate	0.08g
Manganese sulphate	0.05g
Agar	15g
Distilled water	1000 ml

Adjust pH 7.3 and autoclave at 121°C for 15 min.

Composition of different reagents

Preparation of the various stains used

Crystal violet stain

Solution A: 2g of crystal violet (90%) dye content) was dissolved in 20 ml of ethanol.
Solution B: 0.8g of ammonium oxalate was dissolved in 80ml of distilled water
Solution C: Solutions A and B were mixed together and filtered through tissue paper to remove the particulate matter, if any and stored in a dark bottle at room temperature. Fresh staining solutions were prepared once in two weeks.

Malachite stain

5.8 g of Malachite green was dissolved in 100ml of distilled water and mixed thoroughly. Strained and filtered and stored at room temperature.

Safranin stain

2.5 g of Safranin was dissolved in 100 ml of 95 per cent ethanol and from this 5% aqueous solution was prepared and stored at room temperature.

Gram's stain

Gram's iodine: Dissolve 0.33 g of iodine and 0.66 g of potassium iodide in 100 mL of distilled water; alternately, dilute 0.1 (N) iodine in 1:4.

Counter stain stock solution: Dissolve 2.5 g of certified safranin in 100 mL of 95% ethyl alcohol.

Composition of different buffers and dye for Electrophoresis

Agarose gel electrophoresis

TAE buffer (50 X)

Tris base	242 g
Glacial acetic acid	57.1 ml
EDTA 0.5 M (pH 8.0)	100 ml

Tris-Glycine running buffer (10X)

Tris base	30.0g
glycine	144.0g
SDS	10.0g
Distilled water	1000ml

The pH of the buffer should be 8.3.

Loading dye (5 X)

Bomophenol blue	0.25%
Xylene cyanol	0.25%
Glycerol	30%

Sample Loading Buffer (5X)

Tris·HCl (pH 6.8)	250 mM
SDS	10%
Glycerol	30% (v/v)
DTT	10 mM
Bromophenol Blue	0.05% (w/v)

APPENDIX III

LIST OF PUBLICATIONS

Ananda Mukhopadhyay, Soma Das, Ritesh Biswa, **Sangita Khewa (Subba),** Anjali Km Prasad, Kumar Basnet, Jayashree Saren and Mrinal Ray. 2015. Tea pests of Terai-Dooars, Implication of their conventional control and sustainable management options-an overview. ***Global Journal of Environmental Science and Research***. 2(3): 93–106.

Sangita Khewa (Subba), Ananda Mukhopadhyay and Damayanti De. 2014. Isolation and characterization of two *Bacillus* strains toxic to *Hyposidra talaca* (Walker) (Lepidoptera: Geometridae) from Darjeeling foothill region. ***Journal of Applied Zoological Researches***. 25(1):39-44.

Sangita Khewa (Subba) and Ananada Mukhopadhyay. 2014. Characterization and evaluation of entomopathogenicity of two *Bacillus* strains isolated from the sporadic tea pest, *Orgyia postica* (Walker) (Lepidoptera: Lymantriidae) of Darjeeling hills. ***NBU Journal of Animal Sciences***. 8:23-31.

Sangita Khewa (Subba) and Ananada Mukhopadhyay. 2012. Characterizing and testing biocontrol potential of two *Bacilllus* strains isolated from the tea defoliating pest, *Arctornis submarginata* (Walker) (Lepidoptera: Lymantriidae). ***Advances in Life Sciences***. 1(2): 114-117.

Ananda Mukhopadhyay, **Sangita Khewa (Subba)** and Damayanti De. Characteristics and virulence of nucleopolyhedrovirus isolated from *Hyposidra talaca* (Walker) (Lepidoptera: Geometridae), a pest of tea in Darjeeling Terai, India. ***International Journal of Tropical Insect Science***. Vol 31, No 1-2, pp 13-19, 2011. Cambridge University Press, UK.

Ananda Mukhopadhyay, Damayanti De and **Sangita Khewa (Subba)**. 2011. Occurrence of baculovirus in black inch worm, *Hyposidra talaca* (Walker) (Lepidoptera: Geometridae) from Darjeeling. ***Insect Environment***. 16(4):175-176

Sangita Khewa Subba and Ananda Mukhopadhyay 2010. Biocontrol potential of a newly isolated bacterial agent against *Arctornis submarginata* (Walker) (Lepidoptera:Lymantriidae) occurring in Darjeeling Terai region ***Journal of Biopesticides*** 3(special issue) 114-116.

Ananda Mukhopadhyay, Damayanti De and **Sangita Khewa (Subba)** 2010 Exploring the biocontrol potential of naturally occurring bacterial and viral entomopathogens of defoliating lepidopteran pests of tea plantation. ***Journal of Biopesticides*** 3(special issue) 117-120.

Ananda Mukhopadhyay, **Sangita Khewa (Subba)** and Damayanti De. 2007. A report on occurrence of a new defoliator of tea, *Arctornis submarginata* (Walker) (Lepidoptera: Lymentriidae) from Darjeeling terai with notes on its life history performance. ***Insect Environment***. **13**(2): 53-54.

APPENDIX IV

LIST OF ABSTRACT PUBLISHED IN PROCEEDINGS OF CONFERENCES/ SEMINAR/ WORKSHOPS

Sangita Khewa (Subba), Pali Dhar and Ananda Mukhopadhyay. 2008. A glimpse of the campus birds of North Bengal University. Silver Jubilee Symposium on **Dimension of Research Application in Animal Sciences**, Dept of Zoology University of North Bengal. **(Oral presentation)**

Sangita Khewa (Subba) and Ananda Mukhopadhyay. 2009. Biocontrol potential of a newly isolated bacterial agent against *Arctornis submarginata* (Walker) (Lepidoptera:Lymentriidae) occurring in Darjeeling Terai region. **2nd International BIOCICON 2009**, Crop Protection Research Centre (CPRC), Dept of Advance Zoology and Biotechnology, St. Xavier's College (Autonomous), Palayamkottai, Chennai. **(Poster presentation)**

Sangita Khewa (Subba) and Ananda Mukhopadhyay. 2010. Comparison of entomopathogenic bacterial strains isolated from white grub, *Phyllognathus dionisius* (Fabricius) (Coleoptera: Dynastiidae) occurring in tea plantations of Darjeeling foothills. **Evaluation of Biodiversity of Eastern Himalaya and Adjoining Plains**, Department of Zoology, University of North Bengal. **(Oral presentation)**

Sangita Khewa (Subba) and Ananda Mukhopadhyay. 2011. Characterizing and testing biocontrol potential of two *Bacilllus* strains isolated from the tea defoliating pest, Arctornis submarginata (Walker) (Lepidoptera: Lymantriidae). **Biodiversity & Food security- Challenges & Devising strategies** organized by Dheerpura society for advancement of science and rural development and Indian institution of pulses research, Kanpur (UP), India. **(Poster presentation)**

Sangita Khewa (Subba) and Ananda Mukhopadhyay. 2012. Isolation and characterization of two *Bacillus* strains toxic to *Hyposidra talaca* (Walker) (Lepidoptera:

Geometridae) from Darjeeling foothill region**. PLACROSYM XX**, UPASI Tea Research Foundation, Tea Research Institute, Valparai, Coimbatore, Tamil Nadu. **(Poster presentation)**

Sangita Khewa (Subba) and Ananda Mukhopadhyay. 2014. Isolation of Bacillus strains from Bunch caterpillars of Darjeeling Tea Andraca bipunctata (Walker) (Lepidoptera: Bombycidae) with bioassay of their host pathogenicity. AZRA Silver Jubilee International Conference on **Probing biosciences for food security and environmental safety**, Central Rice Research Institution, Cuttack, Odisha. **(Oral presentation)**

Sangita Khewa (Subba) and Ananda Mukhopadhyay. 2014. Characterization and evaluation of entomopathogenicity of two *Bacillus* strains isolated from the sporadic tea pest, *Orgyia postica* (Walker) (Lepidoptera: Lymantriidae) of Darjeeling hills. National Symposium on **Plant Protection in Tea: Recent Advances**, Tea Research Association, Toklai Tea Research Institute, Assam. **(Poster presentation)**