**Research Schemes under XI Five Year Plan**

**Tea Research Association: Projects sanctioned=11**

**Project No 1: Molecular basis of stress linked biochemical changes during processing of tea shoots and their relation to the quality of made tea**

**Name of Principle Investigator: - DR. Pradip Tamuly**

**Objectives:**
- To quantify stress related oxidized damage by the generation of toxic oxygen species and enzyme activity related to oxidative stress during processing.
- Chemical profiling of precursors and their product formation during processing.
- Monitoring the stress induced biochemical change and to link up the same to behaviour to our advantage.
- To understand the mechanism of biotic stress induced changes responsible for flavour generation with special reference to Assam Orthodox and CTC tea.

**Deliverables:**
- Technology developed from the study will help the Industry to improve the quality considerably and will reduce the processing time.
- CTC tea with flavour – a new type of tea will be developed
- Enhancement of flavour of Assam orthodox tea.

**Budget**
Rs. 52.00 lakhs

**Final Outcome/Technology Developed**

i. Regulation of moisture loss during initial stage of withering is one of the most important factors to improve the quality. Loss of moisture should not exceed 1 to 1.5% in the early part of the withering to improve brightness and briskness of liquor.

ii. Rise of leaf temperature beyond 30-32°C during withering is detrimental to quality specially the brightness and briskness. This is due to decline in polyphenol oxidase activity and degradation of catechin content. Restriction of catechin degradation could be achieved by restricting moisture loss at higher temperature.

iii. By regulating degree of maceration during CTC, the percentage of degradation of gallocatechins can be regulated. This will result in theaflavin digallate content along with the improvement of brightness and briskness of liquor.

iv. Optimum fermentation time reduces with rise of withering temperature, degree of withering and percentage of moisture loss in the initial stage of withering.

v. Optimum temperature during fermentation is found to cultivar specific i.e. for T₃E/₃, S₃A/3 having Assam character is 25-26°C while for TV-23, TV-26, TV-9 having cambod character is 30°C.

vi. Cultivar having more Assam character i.e. T₃E/₃, S₃A/3 produces better quality with moisture percentage slightly higher than 70% while cultivar like TV-23, TV-26, and TV-9 produces better cup character around 69% of moisture in the withered leaf.

vii. There is a reduction of fermentation time to the extent of 15-20
min with application of oxygen.

viii. Application of vacuum during withering improves the quality in terms of TF and volatile flavour constituent formation.

ix. Volatile flavour constituent formation is found to be higher at 30°C as compared to 25°C and 35°C during fermentation.

x. Optimisation of fermentation time of some commonly used tea cultivars at different temperature is completed.

2. Total no. of research papers published from the project with Impact Factors (if any): (Detailed list may be provided separately as annexure)

I. Published: 2

3. Seminars/workshops conducted for Technology transfer:

1. 8 (eight) nos. of workshops were conducted in commercial tea factory among factory workers in presence of factory assistants and Manager to discuss various aspects of tea processing and measures to be taken for improvement of quality.

2. 2(two) manufacturing workshops were organized for managers of commercial tea garden at Tocklai to discuss details about various biochemical aspects of tea processing.

Project No 2. Development of methodologies for the extraction food grade secondary metabolites from tea and up scaling the methods for commercial purpose

Name of Principle Investigator: DR. Pradip Tamuly

Objectives:

- Development of methods for the extraction of antioxidants, polysaccharides from tea using modern techniques
- Development of methodology for extraction of coloured pigments from black tea to use for food, cosmetic and health care products.
- Scaling up the already available technology for making tea tablets, tea soft drinks using different kinds of fruits, flavour, spices etc.
- To scale up Super Critical Extraction technology for the extraction of flavour of tea, yellow and red pigments from the various sources of tea to produce various cosmetics and functional food products.
- Conduct trials for the encapsulation of tea flavour for flavouring RTD and other products based on tea.
- Development of technology of flavoured instant tea and to scale ups the same to commercial level.

Deliverables:

- Tea tablet, soft drinks having different taste and flavour for commercial use.
- Technology for flavoured instant tea will be ready for commercial use.
- Product extracted by SCFE technology will be ready to use various cosmetics and health care product.

Budget

Rs. 63.10 lakhs
### Final Outcome/Technology Developed

| I. Different types of green and black tea soft drink are formulated. |
| II. Preparation of green soluble tea (can be used as antioxidant, beverage and food additive). |
| III. Preparation of black soluble tea (can be used as natural tea colour, antioxidant, beverage and food additive). |
| IV. Extraction of antioxidant from tea. |
| V. Extraction of natural tea colour. |
| VI. Different types tea tablet formulation. |

- Assignment deed between NRDC and TRA for following Title of technology on 31st December 2012:
  1. Tea tablet.
  2. Green soluble tea (can be used as antioxidant, beverage and food additive).
  3. Soluble black tea (can be used as natural tea colour, beverage, antioxidant and food additive).
  4. Different components of tea were utilized in developing diversified products.

#### 2. Total no. of research paper published from the project with Impact factor (if any):

- Research paper published: 2 (two) Nos.
- Research paper accepted: 4 (four) Nos.
- Published Article: 4 Nos.
- International presentation: 4 Nos.
- National presentation: 1 No.

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### Project No 3. Establishment of chain of quality testing laboratories and strengthening of existing analytical facilities at regional centres and Tocklai

**Name of Principle Investigator:** DR. Lakshin Prasad Bhuyan

<table>
<thead>
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<th>Objectives:</th>
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<tr>
<td>• Assessment of black (both CTC and Orthodox) and green tea quality including PFA/ISO parameters.</td>
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<tr>
<td>• Chemoprophiling of theaflavins, thearubigins, flavonolglycosides etc by liquid chromatographic technique couple with mass spectrophotometry.</td>
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<tr>
<td>• Feasibility study for predicting quality of tea sample using near infrared spectroscopy (NIR).</td>
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<tr>
<td>• Chemoprophiling of volatile flavoury constituent (VFC) using GC and GCMS of black tea collected from different regions.</td>
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<tr>
<td>• Metabolic profiling of some other biochemical constituents to find out potential indicators/marker of Geographical origin using HPLC and NIR technique</td>
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<tr>
<th>Deliverables:</th>
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<tr>
<td>• Chemical finger printing will be developed.</td>
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<td>• Chemical indicator of tea having geographical origin.</td>
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Study was conducted on black tea produced from 7 regions of Assam (Upper Assam, South Bank, North Bank and Barak Valley) and West Bengal (Dooars, Terai and Darjeeling). The observations/recommendations are:

I. Theaflavin (TF) and thearubigin (TR) content of CTC black tea from the regions of Assam except Barak valley are comparable and higher than other regions. Quality of Barak valley, Dooars and Terai regions are also comparable. Similar trend of total soluble solid (TSS) for the regions is also observed.

II. The variation in TF and TR content of CTC black tea from the regions is responsible for differences in tasters' quality.

III. Quality can be improved by improving plucking standard and processing system. Need further study in these areas.

IV. t-2-hexen-1-ol, linalool oxide, linalool, hotrienol, trans-geraniol, and tetratricontane of Darjeeling teas found higher than the other regions

V. Theanine as well amino acid content of samples from south bank is higher than the other regions in CTC tea.

VI. Distinct variation in total catechin, EGC, EGCG, ECG, TF, TR, TR1 and TR2 of Darjeeling tea with orthodox tea of other regions was observed. Hence, these chemical basis may be a criteria for fingerprint of Darjeeling tea

VII. TF and TR are the key components of black tea which not only influences the sensory evaluation of tasters' but also have high antioxidant properties.

VIII. Nitrite scavenging activity and lipid peroxidation scavenging activity of black tea are more than free radical (DPPF) scavenging activity and super oxide scavenging activity.

IX. Nitrite scavenging activity is correlated significantly with TF and TR content of black tea.

X. Lipid peroxidation scavenging activity is correlated significantly with TF content.

XI. 551 black tea samples were analyzed for iron filings.

2. Total no. of research papers published from the project with Impact Factors (if any): (Detailed list may be provided separately as annexure)

   I. Published: 2

3. Seminars/workshops conducted for Technology transfer:
   - Conducted two workshops at Tocklai Experimental Station in

Presented a paper entitled “CTC Black Tea – A Product of Taste and Health” in the 5th European conference on Sensory and Consumer Research held at Bern Expo Centre, Bern, Switzerland organized by Elsevier from 9th to 12th September, 2012.

## Project No 4. Sustaining soil productivity – some strategies

**Name of Principle Investigator:- DR. R. M. Bhagat**

### Objectives:

- To develop suitable technology for integrated nutrient supply and management system in tea and efficient recycling of residues within the estates (Vermicomposting) and to restore organic carbon status.
- To develop suitable technology for management of compact soil in mature tea.
- To study the effect of continuous addition of chemical fertilizers (NPK and S) on chemical, biological and physical properties and nutrient dynamics and yield and quality of tea.
- To develop a methodology for the quick organic bulk decomposition of the bio-waste to convert it to quality (including micro nutrients and heavy metals) and to study its impact on soil properties and growth of both young and mature tea.
- To explore the possibility of alternate methods of rehabilitation to shorten/eliminate the waiting period between uprooting and planting under textually different soils.
- To study the response of tea clones to graded doses of nitrogen for clone specific fertilizer application.
- To study the influence of various organic acids and P-solubilizing and mobilizing organisms on the transformation of native, applied, and insoluble phosphate and its availability in soil, uptake and yield of tea.

### Deliverables:

- Soil management practices during different stages of growth of tea.
- An effective INM package with inclusion of biofertiliser will be released to tea estates of NE India.
- Quality and eco-friendly organic manure (vermicompost) for use in tea and that can be prepared in the garden itself.
- Enriched vermicompost, phosphate solubilizer/mobilizer suitable for reducing P-fixation and increasing active phosphate available.
- Technology for management of compact soil under mature tea.
- Clone specific fertilizer application and fertilizer use efficiency.
- Balance application of N,P,K and S.
- An alternate method of rehabilitation.
- To establish standard as regard to chemical, physical, physicochemical and biological parameters for soil that will be ideal for tea plantation.
### Budget

<table>
<thead>
<tr>
<th>Final Outcome/Technology Developed</th>
<th>Technology Developed:</th>
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<td></td>
<td>- Technology for the preparation of quality organic manure (vermicompost) was developed at Tocklai and transferred to many tea estates.</td>
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### Recommendations:

- Application of vermicompost @250-300 g/pit at the time of planting was found to be a suitable alternative to conventional cattle manure 4 kg/pit. Most of the tea estates have started using vermicompost in lieu of cattle manure.

- Application of P-enriched vermicompost @250-300 g/pit at the time of planting is a viable alternative to conventional cattle manure + 30 g SSP + 30 g Rock phosphate and cost-effective.

- For improving compaction under heavy soil, application of semi-decomposed vermicompost at trenching in alternate rows during LP is an alternative and more effective than the present practice of forking and hoeing.

- Phosphate solubilizing bacteria/fungus when applied in combination with rock phosphate at reduced dose than the recommended, could maintain the available phosphate at par and produced higher tea biomass over RDF. A. niger performed better than PSB and application of 25 kg P₂O₅ along with 20 L A. niger can be considered as optimum fertilization strategy for tea cultivation in P deficient acid soil in Assam, India. Field studies are in progress to generate more data on long term basis.

- Foliar application of vermiwash (5-10%) has been found to be effective on the growth and productivity of young and unpruned mature tea. It is easily producible, eco-friendly and can be one of the best liquid organic manure for foliar application in tea.

- To reduce the chemical load and maintain soil fertility, 25% of recommended dose of inorganic fertilizer can be replaced by 4 to 6 t/ha of vermicompost or mixed biofertiliser without affecting the yield of tea.

2. Bulletins published for Technology transfer:

   I. Vermicomposting in Tea (sent to many tea estates including the small tea growers).

   II. Enriched Vermicompost – the new planting pit mixture for tea

3. Total no. of research papers published from the project with Impact Factors (if any): (Detailed list may be provided separately as annexure)
Project No 5. Development of alternative strategies for management of tea mosquito bug and blister blight disease in tea plantation of north east India

Name of Principle Investigator:- DR. B. K Borthakur

Objectives:
- Collection and evaluation of sex pheromone (developed by UPASI) for their effective utilization in IPM packages.
- Survey, collection, identification of indigenous microbial bio-control agents and their utilization in controlling tea mosquito bug and blister blight disease.
- Evaluation and utilization of natural enemies in terms of feeding rate, stage preference, rate of parasitization etc. in controlling tea mosquito bug.
- Survey, collection, identification, evaluation of plant species having pesticidal properties grown in an around tea plantations and their utilization as IPM components for controlling tea mosquito bug and blister blight disease.
- To develop cost effective and eco-friendly IPM packages for the tea growers which may have direct impact on reducing the load of chemical pesticides.
- To demonstrate the IPM technologies among the tea growers for their quick implementation.

Deliverables:
- Development of pheromone traps for periodical monitoring of tea mosquito bug population in the field, which will help in scheduling the IPM packages.
- Reduction of pest population through implementation of pheromone traps.
- Conservation, documentation and evaluation of potent native microbial biocontrol agents, entomopathogens, parasites, predators and plant products of tea ecosystem.
- Development of appropriated technology for mass production and implementation of microbial bio-agents, entomopathogens and natural enemies as components of IPM for adoption of sustainable pest and disease control strategies.
- Patenting and commercialization of mass production technologies for the potent microbial entomopathogens, phytochemicals, parasites, predators and pheromone traps. Conservation of biodiversity and reduction of chemical pesticides inputs by exploring the above mentioned alternative strategies.
- Awareness generation and a technology demonstration to
minimize the load of toxic chemicals through adopting IPM packages.

**Budget**  
R. 81.40 lakhs

**Final Outcome/Technology Developed**

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<tr>
<th><strong>Phytochemicals:</strong></th>
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<td>Few commonly grown herbs in and around tea plantations, having pesticidal property, were shortlisted and inducted as one of the effective components of IPM. Methodologies on aqueous and solvent extraction have been standardized and recommended the effective doses for field application for controlling Tea Mosquito Bug and Blister Blight disease.</td>
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<td>Extracts of <em>Cassia alata</em>, <em>Amphineuron opulentum</em>, <em>Leonurus sibiricus</em>, <em>Polanisia icosandra</em>, <em>Polygonum viviparum</em>, <em>Polygonum hamiltonii</em>, <em>Ipomoea convolvula</em>, <em>Polygonum chinense</em>, <em>Eupatorium cannabinum</em>, <em>Urtica dioica</em> are found effective for controlling Tea Mosquito Bug and <em>Cassia alata</em>, <em>Polanisia icosandra</em>, <em>Ipomoea convolvula</em>, <em>Argemon Mexicana</em>, <em>Polygonum hamiltonii</em>, <em>Polygonum chinense</em>, <em>Amphineuron opulentum</em>, <em>Leonurus sibiricus</em> and <em>Helianthus species</em> were found to be effective in controlling Blister Blight disease.</td>
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**Natural enemies:**

- Natural enemies like *Mallada*, *Chrysoperla*, *Oxyopes* spp. the predators of Tea Mosquito Bug were studied and found to have potentiality as component of IPM in controlling Tea Mosquito Bug. Mass rearing technique has been developed based on specific diet.

**Entomopathogen:**

- Successful attempts were made to isolate antagonistic and entomopathogenic microbes from tea phylloplane and Tea Mosquito Bug respectively. Laboratory and field evaluation have shown promise in utilizing three strains of entomopathogenic fungi and six strains of antagonistic/hyperparasitic microbes as components of IPM for controlling Tea Mosquito Bug and Blister Blight respectively. Three strains for controlling Tea Mosquito Bug are *Aspergillus niger*, *Aspergillus flavus* and *Cephalosporium* sp. Six microbes found to be effective in controlling Blister Blight disease are *Aspergillus niger*, *Bacillus subtilis*, White yeast, *Curvularia*, yellow bacteria and *Cephalosporium* sp.

- IPM packages have been formulated by incorporating different tools for controlling Tea Mosquito Bug and Blister Blight.

2. **Bulletins published for Technology transfer:**
2 numbers of bulletins have been published for technology transfer.

1. Native Plant Species for Management of Tea Mosquito Bug and Blister Blight (Bulletin No. PP/01/2012)

2. Native Species of Natural Enemies for Management of Tea Mosquito Bug (Bulletin No. PP/02/2012)

3. Total no. of research papers published from the project with Impact Factors (if any): (Detailed list may be provided separately as annexure)

IV. Published: 4 (Conference papers)

4. Seminars/workshops conducted for Technology transfer:

A series of demonstration programmes has been organized in different member gardens of TRA. In these demonstration programmes the technologies developed under the project have been demonstrated to the field staffs with an aim to educate them to take up appropriate measures for minimization of the use of chemical pesticides for controlling pests and diseases of tea.

5 demonstration programmes were organized at 5 different tea estates (Teok, Greenwood, Hoolunguri, Gatoonga and Oating) of Upper Assam and South bank involving both grass root level workers and field executives. 161 participants took part in the programme. The participants showed their interest in using the various alternative IPM components demonstrated.

Project No 6. Studies on heavy metals – Phase II (chromium and arsenic)

Name of Principle Investigator: DR. A. K. Barooah

| Objectives: | To generate data on chromium and arsenic contents in tea in Northeast India
|             | To study the accumulation of these elements in tea soils of Northeast India
|             | To study the source of contamination by chromium and arsenic. |

| Deliverables: | The, heavy metals have already been identified as non-tariff trade barrier under WTO. Therefore, the information generated in the project will help formulate realistic limits which will aid in marketing of Indian Teas abroad and will also help importing of only quality teas by the country. This will help to assure food safety and enhance consumer acceptability globally.
|               | The database on chromium, arsenic etc, in tea will providing a basis for establishing their realistic maximum permissible limits so that not only the health aspect but also the potential threat in the near future to tea export could be effectively taken care of by ensuring the limits which are safe and practically achievable.
|               | The data on metal contents in tea soils will indicate the extent of contamination and the status of accumulation in tea garden soils |
of the Northeast India. The information generated will also provide means of identifying sources of contamination and help to devise prevention measures for limiting contamination.

<table>
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<tr>
<th>Budget</th>
<th>Rs. 47.00 lakhs</th>
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| **Final Outcome/Technology Developed** | 1. Data on chromium in tea was generated on the basis of a total of 309 samples of made tea collected from different tea gardens and analyzed for total chromium contents.  
2. Data on arsenic content in tea was generated based on 145 samples of made tea collected from different tea gardens and analyzed for total arsenic contents and another 56 samples processed.  

The above findings will provide a basis for fixing permissible limits in tea under FSSAI.  

3. Extractable Chromium contents in tea soils of about 102 samples were determined and testing for arsenic will be completed by the end of this month to indicate the extent of accumulation of these elements in tea soils of NE India.  
4. Different inputs used in the tea gardens were analyzed for identifying possible sources of contamination. 136 water samples collected from 52 tea gardens of Assam and Dooars and used both in field and factory showed low levels of arsenic in general. Limited speciation studies indicated that up to 82% of the arsenic present in the water was due to the non-toxic pentavalent arsenic only. The levels of the toxic trivalent arsenic was below the WHO permissible levels (10 mg/L) while the total arsenic content exceeded this limit in a few samples.  
5. Organic manure and fertilizer samples have also showed the presence of chromium and arsenic. These inputs are potentials sources of contamination. Tea manufacturing process also contributed to heavy metal contents in tea.  

It is therefore recommended that all inputs used in tea plantations should be tested for heavy metal contents before use. Sewage sludge based organic manures should be avoided.  

2. Bulletins published for Technology transfer:  
1. Preventing contamination of teas with heavy metals (under preparation).  

3. **Total no. of research papers published from the project with Impact Factors (if any):** (Detailed list may be provided separately as annexure)  

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**Project No 7. Biotic and abiotic stress analysis for development of stable quality genotypes**

**Name of Principle Investigator:** DR. S. Das
**Objectives:**

- To study and analyse stability of quality genotypes under abiotic (moisture, temperature, radiation and CO$_2$) and biotic (blister blight and Helopeltis) stresses.
- Transport analysis and gene expression studies using stress responsive promoter driven receptors to genetically dissect different types of abiotic stress signaling pathways.
- Development of trait specific molecular markers and establishment of marker trait association, studies on QTL analysis.
- Basis of Hypersensitive Reaction (HR) and Systemic Acquired Resistance (SAR) due to Biotic stress in plants.
- Development/Selection of germplasms adaptive to specific climatic conditions and resistant to various abiotic and biotic stresses.

**Deliverables:**

- Stress tolerant cultivars for commercial exploration
- Exploration of chemical signals between tea plants and the biotic and abiotic stresses.
- Understanding the variation in quality within the same genotypes under different environmental conditions using gene expression studies (transcriptome analysis)
- Marker assisted selection (MAS) in order to expedite conventional breeding.
- The knowledge base developed through this integrated approach would pave the way for location specific selection of germplasms.

**Budget**

Rs. 66.49 lakhs
### Final Outcome/Technology Developed

1. Transcript identification and analysis related to water stress and *Helopeltis* infestation was completed.

2. Candidate genes related to waterlog stress and *Helopeltis* infestation has been analyzed.

3. Expression profiles of genes generated suggesting correlation to the traits of interest.

4. Selection from existing germplasm with the help of EST-SSR markers is in progress and would be continued.

5. Identification of germplasms for utilization as parental combination in future marker assisted selection; in order to expedite conventional breeding can now be achieved by the breeding group. Breeding can be expedited using the information and molecular tools developed out of the project, both for biotic and for abiotic stress.

6. Identification of transcription factors and miRNAs which may help to understand the regulatory mechanism associated with the two traits.

7. Sequences obtained out of the project have been submitted in public domain (NCBI) and would be made available once the papers are published.

**1. Total no. of research papers published from the project with Impact Factors (if any):** (Detailed list may be provided separately as annexure)

**II. Accepted: 3**

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### Project No 8 Establishing a pesticides residues testing laboratory

**Name of Principle Investigator:** DR. A. K. Barooah

**Objectives:**
- To establish a pesticide testing laboratory

**Deliverables:**
- Pesticide residue has already been identified as non-tariff trade barrier under WTO. Therefore, the benefit of setting up the pesticide testing laboratory will help the tea industry in producing quality teas by ensuring MRL conformations.
- Results from such regular testing of tea will also help the industry to go for corrective measures should there be some non-conformations. This will help to assure food safety and enhance consumer acceptability of Indian teas globally.

**Budget**
Rs. 184.00 lakhs

**Final Outcome/Technology Developed**

I. The analytical service of the laboratory was opened to the Tea Industry in November 2011.

II. The laboratory was recommended for accreditation under ISO 17025:2005 standard by NABL for the period 17.10.2012 to 16.10.2014.
III. The scope of NABL accreditation includes a total of 77 pesticides of different classes.

IV. Analysed more than 370 tea samples received from the industry for multi residue analysis till date. Tested 34 tea samples collected from gardens for Nicotine to generate a data for supporting fixing of MRL in tea in EU.

V. Generated data on new molecules from supervised field trials (Fenpyroximate, thiacloprid, flubendiamide) for fixing MRLs.

VI. Successfully participated in international proficiency testing organized by FAPAS, UK in 2013 enhancing scope of international recognition of the laboratory.

2. Bulletins published for Technology transfer:


TRA, NBRRDDC, Nagarkata

Project No 9. Study the biochemical aspects of tea processing in respect of ctc as well as green tea manufacture.

Name of Principle Investigator:- DR. Pradip Tamuly

| Objectives: | • To standardize the process parameter bases on cultivar characteristics under North Bengal condition.
• To understand the biochemical mechanism for quality improvement under North Bengal condition.
• To study the variation of quality parameters under different conditions of green tea manufacture, i.e., steaming and panning.
• To study the role of plucking standard and different combination of planting material on biochemical constituents of made tea in relation to quality. |
| Deliverables: | • Data generated from the project will help in providing proper manufacturing advisory service to industry.
• From the information industry will be benefited to improve the quality and to promote the export of Indian teas. |
| Budget | Rs. 68.00 lakhs |
| Final Outcome/Technology Developed | Black tea |
| i. | By regulating moisture loss of withered leaf in different troughs to follow a definite sequence systematically to mesh with the capacity of the CTC as well as dryer, a considerable improvement could be achieved i.e. brightness, briskness, strength of liquor along with appearance. |
| ii. | Degree of withering and moisture percentage of withered leaves should be maintained certain level based on fine percentage. Percentage of moisture in withered leaf as well as duration of withering should increase with decrease of fine percentage of |
leaf.

iii. In order to follow certain sequence, withering should be regulated (by restricting moisture loss in the initial stage of withering) in such a way so that leaf should not be ready more than 30 minutes before the processing to retain the quality of made tea.

iv. Feeding at the rotovane should not exceed the capacity of CTC to maintain the quality.

v. In order to retain best cup character, leaf temperature should not exceed beyond 35°C at any point of time during CTC processing.

vi. Sharpening interval to be reduced with the decline in fine percentage of leaf.

vii. There should be proper aeration of the CTC room to dissipate heat from the room.

viii. Rise of temperature beyond 30°C in the later part of fermentation is detrimental to quality.

ix. Degree and duration of withering along with sharpening of CTC in time is significantly contributing towards the make style and appearance.

**Green tea**

i. Around 6 min steaming time is found to be the most suitable for green tea manufacture.

ii. Pan fried tea for 5-7 min at 300°C produces better quality green tea as compared to steaming.

1. **Seminars/workshops conducted for Technology transfer:**

   Two workshops were conducted in commercial factory to discuss and demonstrate various aspects of tea processing and measure to be taken to improve quality.

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**Project No 10. Studies on drought in tea areas of Dooars and Terai in respect of soil properties, physiology and yield with a view to schedule irrigation in a cost effective way**

**Name of Principle Investigator:- DR. S. K. Pathak**

| **Objectives:** | • To classify drought of different intensity from long term rainfall and evaporation data.  
• Establishment of crop response to irrigation and identification of limiting value of soil moisture stress or climatological parameters like saturation vapour pressure deficit.  
• To compare the ongoing irrigation practices and its effect at estate level under similar agro climatic condition of the experimental locations.  
• To study certain physiological parameters of tea, viz., leaf water potential, diffusivity resistance etc. and to correlate it with the soil moisture stress.  
• Cost-benefit aspect of irrigation under different schedules. |
| **Deliverables:** | • The project envisages proper irrigation schedule in tea areas of |
North Bengal.

- Identification of plants which has better consumptive use of water and is suitable for this region will be conducted.
- Assessment of crucial period of irrigation and critical meteorological factors during drought will help in overall planning and management of tea estates.
- Subsequent to this study improvement in designing of irrigation system or in comparison of material whereby water holding capacity of soils improvement can be pursued in a cost effective way.

**Budget**

Rs. 57.00 kakhs

**Final Outcome/Technology Developed**

- During light prune year (2010) with respect to control yield increased by 21.86% in 50 mm irrigation at 40 days interval.
- 50 mm irrigation at 30 days interval (T3) has shown 5% increase in yield during un-pruned year (2011).
- During 2012 (dip skiff year) yield increased by 22.45% with respect to control in 50 mm irrigation at 30 days interval (T3).
- Water use efficiency increased in 50 mm irrigation at 30 days interval (T3) with respect to control in both un-pruned and dip skiff year.

**Project No 11. Current pest problems in tea of North Bengal and their possible management strategies**

**Name of Principle Investigator:- DR. S. K. Pathak**

**Objectives:**

- To review the present pest scenario against known earlier.
- To assess the migratory behaviour of the pests as influenced by ecological factors.
- To assess the host range of pests.
- To study the life history module of the pest against the host range.
- To assess the natural enemies of the current pests.
- To assess the presently recommended pesticide molecules and/or Newer and/or bio-pesticides against the pest.

**Deliverables:**

- Information on bio-ecology of different pest-insects and their naturally occurring bio-control agents is a prerequisite to develop non-polluting, safe methods of pest control in tea.
- A complete data base on different aspects of bio-ecology of important pests under Dooars tea plantations will be prepared.
- Knowledge on bio-ecology of different pests will enable planters to assess the status of different pests and help them in deciding the proper and timely management strategies.
- Evaluation of currently recommended pesticides against different pests under North Bengal condition will redefine their usage (including right time and dose of application).
- The efficacy of newer and safer molecules under tea ecosystems and potentiality of different bio-pesticides/botanical pesticides once established will widen the scope of their use in the control programme.
- The proposed study will also provide details on effective natural enemies associated with specific pest, their efficacy as bio-control agents under field conditions.
**Recommendations may lead to minimize the use of hard pesticides broadly resulting in a safer and better ecosystem.**

**Sustain Bio-diversity in tea areas through minimum use of pesticide.**

<table>
<thead>
<tr>
<th>Budget</th>
<th>Rs. 39.68 lakhs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Final Outcome/Technology Developed</td>
<td>IPM package for looper caterpillars</td>
</tr>
<tr>
<td></td>
<td>An IPM package of looper caterpillars, presently the major and new pest on tea in North Bengal, has been developed and suggested to manage the pest. It includes the following:</td>
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<tr>
<td></td>
<td>1. Collection of chrysalids mainly during winter months from November to April.</td>
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<tr>
<td></td>
<td>2. Light trapping of moths and manual/physical (fire) destruction of eggs on shade trees up to around 6 m during November-April. Few monitoring lights should be continued throughout the year to know the activity of the pests.</td>
</tr>
<tr>
<td></td>
<td>3. A new set of insecticides other than synthetic pyrethroid, like Emamectin benzoate, Flubediamide, Drona and Naturalis were found very effective in laboratory as well as field reducing the use of synthetic pyrethroid drastically, which is a mass killer and thus damage the eco system. These are under further trial at Tocklai to generate required data for CIB approval.</td>
</tr>
<tr>
<td></td>
<td>4. A new commercial light trap has been tried and found more effective than crude method suggested earlier against all the lepidopteran pest</td>
</tr>
</tbody>
</table>

**Bulletins published for Technology transfer:**

- A special bulletin was published in February, 2010 entitled-“Looper caterpillars, a menace to tea industry of Dooars and Terai and their management”.
- It was further revised and published as **Tea looper caterpillar.** 2011. *Tea Research Association.* Bulletin No. PP/02/2011

**Total no. of research papers published from the project with Impact Factors (if any):** (Detailed list may be provided separately as annexure)

- **V. Published:** 12

**Seminars/workshops conducted for Technology transfer:** 6

**B. C. Guha Centre for Genetic Engineering & Biotechnology, University of Calcutta: Project sanctioned= 1**
Project No 12. Evaluation of the health beneficiary effects of tea products and modulatory role of tea flavonoids on emphysematous lung damage for the formulation of a high potency tea tablet

Name of Principle Investigators:- Dr. Kaustubh Panda

<table>
<thead>
<tr>
<th>Objectives:</th>
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<tbody>
<tr>
<td>• To study the modulatory role of tea tablets on cigarette smoke-induced protein damage, apoptosis and pulmonary emphysema in a guinea pig model.</td>
</tr>
<tr>
<td>• To study the modulatory role of tea polyphenols, namely catechins, theaflavins, thearubigons on cigarette smoke-induced protein damage, apoptosis and pulmonary emphysema in a guinea pig model and thereby evaluate the health beneficiary effect of different tea by-products.</td>
</tr>
<tr>
<td>• To study the modulatory role of tea tablets on the major harmful cigarette smoke component p-benzosemiquinone/p-banzoquinone-induced protein damage, apoptosis and pulmonary emphysema in a guinea pig model.</td>
</tr>
<tr>
<td>• To study the modulatory role of tea polyphenols, namely, catechins, theaflavins, thearubigins on p-benzosemiquinone/p-banzoquinone-induced protein damage, apoptosis and pulmonary emphysema in a guinea pig model.</td>
</tr>
<tr>
<td>• To study the modulatory role of tea tablet and the tea polyphenols (cathchins, theaflavins, thearubigins) and their mechanism of action on cigarette smoke and p-banzoquinone/p-banzoquinone-induced protein damage and apoptosis on relevant human lung epithelial cells.</td>
</tr>
<tr>
<td>• To study the additive effect of the antioxidant vitamin C on the shelf life and modulatory effect of tea tablet in order to order prevent auto-oxidation and loss of biological activity of polyphenols. This particular objectives is designed to prepare a high potency tea tablet and tea byproducts with increased shelf life.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Deliverables:</th>
</tr>
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<tbody>
<tr>
<td>• A commercially viable and marketable high potency tea tablet which is expected to receive substantial social acceptance for its potential to prevent emphysematous lung damage encountered by cigarette smokers as well as a versatile antioxidant for control of aging symptoms.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Budget</th>
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<tbody>
<tr>
<td>Rs. 35.44 lakhs</td>
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</table>

Final Outcome/Technology Developed

The modulatory role of concentrated tea tablets developed by TRA, Tocklai, on cigarette smoke and PBQ induced protein damage, apoptosis and pulmonary emphysema in a guinea pig model was evaluated.

I. Results from our studies on a guinea pig model established the protective role of TRA black tea tablets on cigarette smoke and PBQ -induced protein damage, apoptosis and pulmonary emphysema.

II. However, as evident from the experimental data, the protective effect of the TRA black tea tablets was partial (imparting around 40-50 % protection) in that it could not substantially prevent tissue damage implicated with increased oxidative stress.
III. Our data indicate a necessity as well as scope of increasing the antioxidant potency of the tea tablet developed by TRA through a component-based evaluation of the different tea polyphenols and formulate a high potency tea tablet against cigarette smoke-induced emphysematous lung damage and increased oxidative damage as is associated with senile decay or aging.

Formulation of a high potency tea tablet by evaluating the relative antioxidant potency and bioavailability of different tea polyphenols in a physiological setting.

I. An extensive comparative bioavailability study revealed the actual differential bioavailability and the metabolic fate of the constituent tea polyphenols in guinea pig blood plasma and different organs

II. The comparative efficacies of the constituent polyphenols were evaluated through both in vitro and cell culture based studies.

III. A high potency tea tablet formulation was developed by combining the polyphenols with the highest bioavailability and bioefficacy indices.

IV. The proposed formulation is ready for validation studies through clinical trials for the required development of a high potency antioxidant tablet from black tea for human use.

The modulatory role of the major black tea polyphenol, thearubigin on cigarette smoke induced protein oxidation and damage was elucidated

I. The modulatory role of thearubigins, the major black tea polyphenol was studied separately, after isolation of crude extract of the same from Assam variety of black tea (since the polyphenol could not be commercially procured)

II. The thearubigin fraction showed acceptable protection against cigarette smoke-induced oxidative damage.

Differential polyphenolic constitution of different varieties of tea cultivated in India was assessed.

I. It was seen that whilst the Assam variety is rich in the oxidized, typical black tea polyphenols like theaflavins and thearubigins, the Darjeeling variety has comparatively higher amounts of the green tea catechins.

UPASI-TRF: Projects sanctioned ≥4
## Project No. 13. Studies on the residues of pesticides and heavy metals in tea

**Name of Principle Investigator:** DR. V.S. Sanmukhaselvam

### Objectives:
- Quantification of residues of certain new generation pesticides and their possible.
- Dissipation of the above pesticides during processing and storage.
- Quantification of different oxidation species of heavy metals (specification).
- Phyto – remediation studies using certain hyper – accumulating plants.
- Influence of heavy metals on enzyme activity of soils.
- Aflatoxin in tea.

### Deliverables:
- The project is expected to provide detailed information on residues of new generation acaricides/fungicides/weedicides at different harvest interval, degradation of pesticides in made tea and brew and aflatoxins contamination in tea. Further data generated on parent compound and metabolite will be helpful in fixing realistic MRL and in monitoring the chemical residues in the end product to maintain the European Unions standards, thereby improving south Indian tea quality.
- Since this lab is accredited by NABL, Govt. of India for pesticide residues and heavy metals, the state of art equipments facility established in this project will be utilized for extending the analytical services to the tea members in south India for analyzing their tea samples in sun ppb level as per International requirements.
- The advantage of phytoremediation technology over other approaches of decontamination are that they are relatively low cost, less disruptive of the remediation site, creates a beneficial habitat for soil and fauna, plant roots and shoots can take up heavy metals this effecting the physical removal of toxicity. Thus it is clear the utilization of the remarkable potential of green plants to accumulate elements and compounds from the environment is going to serve as the green cure technology for the agro – eco system in future.

### Budget
Rs. 144.00 lakhs

### Final Outcome/Technology Developed

| I. | Developed and validated the methodologies for the quantification of residues of abamectin, bifenazate, dimethoate, thiacloprid, thiamethoxam, clothianidin, carbendazim, spiromesifen, mancozeb and oxyfluorfen. |
| II. | Dry & wet season trials were completed for determining the residues of abamectin, bifenazate, dimethoate, thiacloprid, thiamethoxam, clothianidin, carbendazim, spiromesifen, mancozeb and oxyfluorfen. |
| III. | Quantification of residues both in black tea and tea infusion was completed for the pesticides abamectin, bifenazate, dimethoate, thiacloprid, thiamethoxam, clothianidin, carbendazim, spiromesifen, mancozeb and oxyfluorfen. |
| IV. | Dissipation study during the different stages of tea processing was completed for the pesticides abamectin, bifenazate, dimethoate, thiacloprid, thiamethoxam, clothianidin, carbendazim, |
spiromesifen, mancozeb and oxyfluorfen.

V. Degradation of residues during storage was completed for the pesticides abamectin, bifenazate, dimethoate, thiamethoxam, clothianidin, carbendazim, mancozeb and spiromesifen.

VI. Studies on the Impact of Lead (Pb), Cadmium (Cd) and Arsenic (As) on the activity of the enzymes Protease, Urease, Nitrate reductase & Acid Phosphotase in soil were completed.

VII. Method development and Quantification of Cr6+ & Ni2+ species in black tea were completed.

VIII. Data on residues for the pesticides abamectin, bifenazate, dimethoate, thiacloprid, thiamethoxam, clothianidin, carbendazim, spiromesifen, mancozeb and oxyfluorfen were generated and ready for submission for setting up of MRL.

IX. Safe harvest interval was arrived for the pesticides abamectin, bifenazate, dimethoate, thiacloprid, thiamethoxam, clothianidin, carbendazim, spiromesifen, mancozeb and oxyfluorfen.

2. Bulletins published for Technology transfer:

Safe harvest interval of Different pesticides to be published (in progress).

3. Total number of research papers published from the project with Impact factor (if any):

Published: 2
Accepted: 7

4. Seminars/workshops conducted for Technology transfer:
A one day “National workshop on Pesticide Residue” was organized at Coimbatore on 12th April 2012.

Project No 14. Development of Integrated Pest and Disease Management (IPDM) strategies for tea with special reference to non-chemical control methods

Name of Principle Investigator:- DR. R. Premkumar

Objectives: Pest Management:
- Evaluation of predatory potential of major predators of tea pests such as red spider mites and thrips, and the impact of the commonly used (recommended) acaricides/insecticides on these natural enemies.
- Increasing the efficiency of entomopathogens like *Paceilomyces fumosoroseus*, *Beauveria bassiana* and *Verticillium lecanii*
- Monitoring insecticide resistance in major pests like red spider mite, thrips and tea mosquito.
- Studies on the Kairomones/pheromones for important tea pests like *Helopeltis*, Shot hole borer and red spider mite and their possible utility in pest management.
- Micro-plot/multi-location field trials on the evaluation of predatory potential of the major predators and assessing their establishment in
different agro-climatic conditions under normal estate practice as well as in organic tea gardens.

- Developing a suitable pest and disease management strategy, especially for organic tea garden.
- Training on familiarizing integrated pest and disease management techniques for incorporation in the IPDM strategies for efficient management of tea pests and diseases.

**Disease Management:**

- Development of germplasm which consists of the following biocontrol agents, a) *Trichoderma*, b) *Pseudomonas* and c) *Bacillus*
- Sensitivity of the biocontrol agents to agrochemicals used in tea.
- *In vitro* studies on the antagonistic potential of the biocontrol agents against tea pathogens such as *Macrophoma*, *Hypoxylon* and *Pestalotiopsis*.
- Mass multiplication of biocontrol agents.
- Field studies using the biocontrol agents against Branch canker, Wood rot and Grey blight diseases of tea.
- Evolving integrated control strategies for the control of the three diseases.

**Deliverables:**

Effort will be made to come with an integrated pest and disease management strategy for organic tea gardens by adopting biological means. It will definitely form a component of IPM in the conventional fields by way of reducing the use of synthetic pesticides.

**Budget**

Rs. 99.00 lakhs

**Final Outcome/Technology Developed**

**PLANT PATHOLOGY PART**

I. The indigenous wood rot and branch canker causal organisms were collected and identified as *Hypoxylon serpens* and *Macrophoma* sp. respectively through molecular tools (5.8S ribosomal RNA gene), their sequences were submitted to NCBI and published through EMBL.

II. *In vitro* studies on the antagonistic potential of the biocontrol agents against tea pathogens (*Pestalotiopsis theae*, *Hypoxylon serpens* and *Macrophoma* sp.) were completed.

III. Among the 645 bacterial isolates and 40 *Trichoderma* isolates, five each of *Pseudomonas* sp. and *Trichoderma* sp. showed higher inhibition against grey blight pathogen compared to *Bacillus* sp. under *in vitro* level.

IV. Among the isolates of biocontrol agents, 5 *Bacillus* strains showed higher antagonism against wood rot pathogen. Three each of *Bacillus* sp. and *Pseudomonas* sp. had higher antagonism against branch canker pathogen in *vitro*.

V. *Bacillus* sp. and *Pseudomonas* were compatible with selected pesticides (Propargite, Fenpyroximate, Hexythiazox, Thiamethoxam and Deltamethrin) and fungicides (Hexaconazole, Carbendazim, Mancozeb, Copper oxychloride, Nativo, Tridemorph, Benomyl and Tebuconazole).

VI. Proven *Trichoderma* sp. isolates were studied for their compatibility/tolerance with the chemical pesticides. They were
compatible with Propargite 57 EC, Fenpyroximate 5 EC, Hexythiazox 5.45 EC, Deltamethrin 2.8 EC, Thiamethoxam 25 WG except Dicofol 18.5 EC and quinophos 25 EC, but not with any recommended fungicides in tea.

VII. Chemical fertilizers at recommended doses used in tea highly supported the growth of both *Trichoderma* and bacterial biocontrol agents.

VIII. Bioefficacy of certain plant aqueous extracts (Neem kernel, Pongam kernel, *Cinnamom*, and *Artemisia*), acetone extracts (*Lantana camera, Hibiscus rosasinensis, Ageratum conyzoides*) and bryophyte (*Heteroscyphus argutus*) were also evaluated against three pathogens under *in vitro* condition. Results revealed that *Artemisia* followed by *H. argutus* and Neem kernel extract controlled the wood rot pathogen effectively.

IX. Mass production techniques of biocontrol agents with suitable carrier materials have been standardized for the preparation of both solid and liquid bioformulations.

X. A systemic fungicide Benomyl 50% WP at the concentrations of 0.02, 0.05, 0.10, 0.50 and 1.0 % was tested to check its bioefficacy against the pathogens under *in vitro* and was found effective at 0.5%.

XI. The consortium of *Bacillus*, *Pseudomonas* and *Trichoderma* bioformulations were prepared using talcum powder. The final concentration of the organism was made as $10^8$cfu/g for bacteria and $10^7$cfu/g for *Trichoderma*. Experiments were conducted in BSS-1 seedling area in randomized block design. There were eleven treatments each replicated three times and each replicate consisted of 54 bushes. Formulations were mixed with required quantity of water (175 L/ha) and sprayed using hand operated knapsack sprayer at 10 days interval for chemical fungicides and at seven days for biocontrol agents. In the integrated schedule, two rounds of chemical fungicide were given alternately with one round of bioformulation. This study indicated that the antagonistic ability of *Pseudomonas* sp. and *Trichoderma* sp. when combined with carbendazim for satisfactory grey blight control. Apart from carbendazim and mancozeb, a new formulation, Nativo 75 WG provided effective control.

XII. Evaluation of biocontrol agents against three diseases under field level indicated that straight application of *Pseudomonas* sp., *Trichodema* sp. and consortium of *Bacillus* sp and *Pseudomonas* sp. provided good control of grey blight disease. Outcome of this study, antagonistic ability and compatibility properties of *Pseudomonas* sp. (5 kg /ha.) when combined with carbendazim (5g/10L) and *Bacillus* sp. (5 kg per ha.) combined with mancozeb (30g/10L) strengthened the integrated approach of grey blight disease management in tea.

XIII. *Bacillus* sp. (WR46-2 and HBCWR-3) with chemical fungicides
(Benomyl 50%WP or Copper oxychloride) at 0.5 % were much effective against wood rot and branch canker pathogens.

XIV. Another field experiment reported that, *Pseudomonas* sp. (5 kg per ha.) in combination with carbandazim (2.5g /10 L) and *Bacillus* sp. (5 kg per ha.) combined with mancozeb (15g / 10 L) strengthened the integrated approach of grey blight disease management in tea.

XV. Discussions on MOU with TARI Organic plus for commercial production of bioinoculants are in progress. Recommendation to the industry will be brought once the data compilation is completed.

**ENTOMOLOGY PART**

I. A mass rearing technique using an artificial diet was standardized for green lace wing, *Mallada boninensis* and field release of the eggs has also been attempted.

II. Entomopathogenic bacterium, *Pseudomonas* fluorescence and fungus *Lecanicillium lecanii* were isolated from the infested mites and thrips respectively and their efficacy was evaluated in laboratory and field against the respective pests.

III. Enhancement of efficacy of entomopathogens namely *Paecilomyces fumosoroseus* and *Lecanicillium lecanii* were studied with addition of jaggery.

IV. Kairomones and pheromone were tested in large scale for the management of shot hole borer and tea mosquito bug.

2. **Bulletins published for Technology transfer:**
   - Organic tea crop guide: A manual for practitioners of Organic tea
   - Bulletin of UPASI tea research foundation
   - Guidelines of reducing pesticide residues in tea
   - Blister blight and grey blight disease of tea

3. **Total no. of research papers published from the project with Impact Factors (if any) (Detailed list may be provided separately as annexure):**
   - I. Published: 17
   - II. Accepted: 6

4. **Seminars / workshops/Trainings conducted for Technology transfer:**
   1. Training on disease management to official and executives of Stanmore, Sheikalmudi and Iyerpadi estates in Anamallais.
   2. Demonstration on mass multiplication of *Trichoderma* for field application to the officials of HML and Woodbriyar group.
   3. Training on pest management in tea to official and executives of HML and TATA Coffee group.
   4. Training programme on tea culture for the students of TNAU, Periyalkulam.
5. Training programme on integrated pest management to Asst. Field Officers of Woodbriar group, Parry Agro group, TATA Coffee group and to development officers of Tea board.

Project No 15. Construction of Hi-tech Tea factory (HTF) at UPASI-TRF, Coonoor

Name of Principle Investigator:- DR. P Mohan Kumar

Objectives:
- Construction of HTF at UPASI-TRF, Coonoor.
- Purchase and commissioning of machines and utilities.
- Development of networked data acquisition and on-line control systems.
- Commissioning of advanced research outcomes in the HTF.
- Experimental trails for fine-tuning the various processes.
- In-process monitoring of physical parameters.
- Virtual instrumentation technologies for on-line presentation and monitoring over internet.
- Technology dissemination.

Deliverables:
- Enhancing the productivity.
- Improvement in efficiency of manufacture.
- Improving quality of tea.
- Energy saving in tea manufacturing processes.
- Reducing cost of production.
- Achieving consistency in the end product.
- Enhancing efficiency of operation from field to market.

Budget
Rs. 301.00 lakhs

Final Outcome/Technology Developed
I. Determination of Optimum Fermentation Time (OFT) using E-Nose
II. Changes in enzyme activities and their substrates during black tea processing

3. Total no. of research papers published from the project with Impact Factors (if any) (Detailed list may be provided separately as annexure):

I. Published: 2

Project No 16. Analysis of gene expression during phyto-pathogenic stress in tea using transcriptomic approach

Name of Principle Investigators:- Dr. R. Rajkumar

Objectives:
- Construction of cDNA library from disease included plants, sequencing of ESTs and analysis.
- Differential display (DD) analysis of control and disease induced
plants.  
- Isolation and sequencing of differentially expressed genes.  
- Full length sequencing of identified candidate genes

### Deliverables:

- Different types of libraries being developed will be important resources for searching agronomically important genes. A number of genes are expected to be identified from these libraries.  
- The cDNA libraries and the accompanying EST database will be valuable resources for tea research to understand the molecular mechanism of disease resistance and other physiological process.  
- The sequencing of cDNA library clones and analysis of ESTs will provide both a mRNA expressions profile and a rapid, low cost and efficient way to identify new functional genes.  
- Differential display (DD) analysis will identify up or down regulated genes during disease infection in tea.

### Budget

Rs. 54.64 lakhs

### Final Outcome/Technology Developed

The data generated in this project will serve as a base line data/information for the future researchers. UPASI TRF, TRI has submitted several sequences to NCBI data base which can be used by research institutes within the nation and across the tea growing regions.  
- Blister blight \((Exobasidium vexans)\) and grey blight \((Pestalotiopsis theae)\) diseases related gene sequences have been submitted to NCBI database 542 EST’s, 4 partial CDS and 2 full length sequences.  
- The role of antioxidative enzyme activity, the ROS is scavenged before it gives signal to the defense related genes. This study provided fundamental information about the role of antioxidative enzymes in the grey blight and blister blight disease development in tea.  
- Differential expression of biochemical constituents and antioxidative enzymes in response to blister blight & grey blight pathogens have been established along with molecular tools during fungal infection.  
- Basic information about the expression of PR-proteins in the resistant and susceptible cultivars during grey blight and blister blight disease development.  
- The purified PR proteins were further used for lab studies to control the insect and disease infection, in future these enzymes can be introduced in controlling pest and disease using these biomolecules.  
- Metallothioneine like protein is an important gene in imparting stress tolerance in plants. The full length sequence was characterized by bioinformatics tools. This gene can be used in future breeding programme for plants with stress tolerance.  
- Differential gene expression studies through microarray revealed 129 genes differential expressed during blister infected and 182 genes from grey blight infected which was found to be defense
relegated genes which play important role providing resistant tea clones to respective diseases in SA-6 & UPASI-10.

- Some important defence related genes involved in grey blight infestation - **AT1G08720.1** - LRR and NB-ARC domains-containing disease resistance protein, **AT1G12280.1** - Disease resistance protein (TIR-NBS-LRR class), **AT1G29715.1** - Component of R gene-mediated disease resistance in Arabidopsis thaliana with homology to eukaryotic lipases, **AT5G44900.1** - encode a protein involved in negatively regulating salicylic acid-related defense responses and cell death programs

- Some important defence related genes encoded for blister blight infestation **AT2G01040.1** - Disease resistance/zinc finger/chromosome condensation-like region, **AT5G45210.1** - Encodes a NBS-LRR disease resistance protein that possesses N-terminal kinase subdomains, **AT4G09420.1** - Disease resistance-responsive (dirigent-like protein) family protein

- Totally 546 common genes are involved form metabolic pathways, glutathione metabolism, protein metabolism, biosynthesis of alkaloids, terpenoids ubiquitin and other pathways in grey blight resistant clone UPASI – 10 and 511 common genes are encoded in blister blight resistant clone SA-6.

- Data generated in the present study have revealed fundamental information about the molecular and biochemical interactions between the pathogen and plant. The genes identified from this project can be used for future plant breeding program in tea.

2. BULLETINS PUBLISHED / TO BE PUBLISHED (MENTION TIME FRAME) FOR TECHNOLOGY TRANSFER:


- Microarray data has been submitted to Gene Expression Omnibus (GEO) a public functional genomics data repository supporting MIAME-compliant data submissions. Public release date - August 15, 2013

3. RESEARCH PAPERS (MENTION TOTAL IMPACT FACTORS):

I. Published: 2,
II. Accepted: 1

C-DAC: Project sanctioned= 1

Project No 17 Corpus creations of measurable physical parameters of Indian tea
Name of Principle Investigator: Dr. N. Bhattacharyya

**Objectives:**
- With the prior approval by the Tea Board, to establish a few suitable data acquisition infrastructure using E-Nose and E-Vision systems at each agro-climatic tea producing zones of India (say, in six Tea Auction Centres where most of Arrival takes place)
- To develop requisite Hardware/Software Infrastructure for such dispersed data acquisition, meta-data creation, data preservation, data Scrutiny and eventually, data analysis/processing.
- To create an image database with E-Vision system and aroma database with E-Nose system for teas arrival at those infrastructures.
- To pursue research in determining aging of tea by instrumental methods.

**Deliverables:**
- Aroma and appearance corpus of Indian Teas will be built-up following a reliable and repeatable manner.
- New objective physical standards of tea quality will be evolved.
- New marketing paradings like issuance of commercially tradable Warehouse, receipts may be possible backed up by such new methods.

**Budget**
Rs. 74.98 lakhs

**Final Outcome/Technology developed**
- Web enabled tea corpus database for measurable physical parameters (aroma, colour, texture, taste etc) of Indian Tea using indigenously developed olfaction and vision technology.
- An Image database for different liquor, infusion, appearance, tea grades etc.
- Flavour and aroma database for all the teas produced in various geographic regions of India.
- A study reports on evaluation of tea quality due to aging by instrumental analysis.
- Indigenous Electronic Nose, Vision and Tongue Technology and customized Integrated Electronic Nose and Vision system tuned as needed by the Tea industries in different geographical locations in India.
- Broadening the applicability of newly developed ENTV Technology (Electronic Nose, Vision & Tongue) in the Indian Tea Industries.
- Research output has proved that ENV technology has potential to estimate the % of Theaflavins (TF) and Thearubigins (TR) in black tea.

**Recommendation made to the Tea Industries** (Through Information Brochures, Seminars / Workshops and Publications):
- Electronic Nose can be used to detect the optimum time of fermentation (based on smell intensity profile) in Black tea processing and quantify aroma intensity in finished tea.
- Electronic Vision solution can be used to detect end pint of...
fermentation (based on colour changing) and indicate the speed of fermentation by tracking the colour profile. It is also used to evaluate tea quality based on colour, brightness attributes of tea liquor with out and after adding milk. This system is also useful to evaluate granular domination of different tea graded and estimate the presence of different sized tea particle (grades) in a bulk tea, which can be used as a measure of consistency tea production or bleeding process. Electronic Vision System is also useful to detect the degree of blackness in CTC tea, measure the percentage of fiber in finished tea and finally provide tea taster like score, which determines the ultimate quality of tea.

- Electronic Tongue can be used to detect taste attribute (like astringency, briskness) of finished tea.

2. Bulletins published for Technology transfer:

- Information Brochures, Leaflets
- Expression of Interest (EoI) for transfer of technology
- Proceedings of seminars / workshops.
- Journal and conference publications.
- Book Chapters.

3. Total no. of research papers published from the project with Impact Factors (if any):

I. Published:
   A. Journal: 12
   B. Conference/ Seminar: 32

II. Book Chapter: 3

4. Seminars/workshops conducted for Technology transfer:

I. A one day workshop on “Tea Quality – Enhancement and Rapid Measurement”, held on 3rd September’ 2010 at Tea Research Association, Tocklai, organized by TRA-Tocklai, C-DAC (K), Jadavpur University jointly sponsored by C-DAC, Kolkata & Department of Science and Technology (DST), Govt. Of India.

II. A one day “Workshop and industry meet on Electronic Nose and Tongue was organized by C-DAC, Kolkata at Rotundra Hall, Manikanchan SEZ, Kolkata on 30th October’ 2009 jointly sponsored by Tea Board, Kolkata & Department of Science and Technology.

DTR & DC & UBKV: Project sanctioned= 2

Project No 18 Development of phosphate solubilizing biofertilizer for tea (Camellia sinensis l.) In acids soils of North Bengal
Name of Principle Investigators:- DR. A. Choudhury

Objectives:
- Isolation of various groups of phosphate solubilizing microorganisms from rhizosphere and rhizoplane of tea in Darjeeling Hills, Dooars and Terai Zone of West Bengal.
- Screening of the efficient strains on the basis of their phosphate solubilizing capacity acid tolerance and their ability to produce growth promoting substance.
- Evaluation of the performance of some efficient phosphate solubilizing microorganisms in tea rhizosphere.
- Characterization of some tested promising phosphate solubilizing microorganisms.

Deliverables:
- It is expected to get some promising phosphate solubilizing microorganisms of diverse group isolated from tea rhizophere which can thrive well in acidic tea soils and can solubilize soluble soil-P and applied rock phosphate.

Budget
Rs. 37.00 lakhs

Final Outcome/Technology Developed

A. Isolation and screening of potential phosphate solubilizing bacteria:
- Total 41 phosphate solubilizing bacterial isolates were screened on the basis of solubilization of tri-Calcium phosphate, Rock phosphate, Aluminium Phosphate and Ferric Phosphate in liquid culture.
- On the basis of the insoluble phosphate solubilizing capacity eleven (11) bacterial isolates (T-1, T-5, T-8, T-20, T-30, T-33, T-38, T-40, T-52, T-56 and T-57) have been identified as potential solubilizing bacteria.

B. Screening of phosphate solubilizing bacteria on stress tolerance and organic-P mineralization capacity:
- The growth performance of phosphate solubilizing microorganisms was assessed at low temperature, low pH (4.0), at different levels of Al concentration and at different levels of desiccation (with polyethylene glycol 600) with a view to study their survivability under adverse soil condition.
- Tea soils of Darjeeling hills contain high amount of organic matter which contain a sizable amount of organic phosphate. Hence, profile for organic-P mineralization potential of various phosphate solubilizing bacteria (PSB) under laboratory condition has been developed.

C. Identification of potential phosphate solubilizing bacteria isolated from Darjeeling tea soils:
- Potential PSB isolates have been identified to its nearest species based on 16s rDNA sequence data. The ~1.4kb rDNA fragment was amplified using high –fidelity PCR polymerase. The PCR product was sequenced bi-directionally using the forward, reverse and internal primer. The sequence data was aligned and analyzed to identify the bacterium and its closest neighbors. Neighbor joining protocol was adopted for identification.
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- *Bacillus* and *Burkholderia* have been identified as dominant bacterial genera present in Darjeeling tea soil rhizosphere. *Bacillus* is gram positive bacteria which can generally tolerate adverse environmental condition due to its spore bearing nature. Whereas, *Burkholderia* has been reported as phosphate solubilizer which can perform under low pH.

**D. Performance of phosphate solubilizing bacteria in tea rhizosphere:**

- Inoculation experiment was conducted with TPB1, TPB33, TPB38, TPB40, TPB52 and TPB57 isolates. All the six isolates showed significantly higher P-uptake over un-inoculated control. Inoculation along with application of Rock Phosphate (RP) increased the P-uptake over inoculation alone. Carrier based (with charcoal powder) and liquid *inocula* were used for inoculation, but no significant difference was observed.

**E. Development of phosphate solubilizing bacterial biofertilizer formulation for Darjeeling tea plantation:**

- Compatibility test of isolated *Bacillus* and *Burkholderia* showed that none of the isolates inhibited other strains.
- On the basis of types of isolates, compatibility test and inoculation experiment two consortia have been developed with *Bacillus* and *Burkholderia* species.

**Consortium-1:** Bacillus firmus; HNS012 + Burkholderia sp. J62  
**Consortium-2:** Bacillus firmus; UST00620-011 + Burkholderia cepacia; BAM 12

2. **Total no. of research papers published from the project with Impact Factors (if any):** (Detailed list may be provided separately as annexure)

VI. **Published: 1** (Conference paper)

**Project No 19. Nitrogen mineralization of organic matter in acid soils of tea (*Camellia sinensis* l.) in northern districts of West Bengal**

**Name of Principle Investigators:- DR. A. Choudhury**

| **Objectives:** | • To study the carbon and nitrogen mineralization of various organic matter in acids soils of tea.  
• Development of nitrogen mineralization indices predicting net nitrogen mineralization.  
• To study the performance of carious organic matter under field condition.  |
| **Deliverables:** | • It is expected to get information about the mineralization potential as well as the pattern of N-mineralization of various organic matter |
used in tea soil.

- Some nitrogen availability indices can be generated which may be used for quick assessment of the quality of organic matters with respect to N-mineralization.
- Moreover, some formulated organic products will be screened and selected for better performance in hill and dooars tea garden.

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<tr>
<th>Budget</th>
<th>Rs. 31.00 lakhs</th>
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<tbody>
<tr>
<td>Final Outcome/Technology Developed</td>
<td>A. Development of quality parameter standards for different organics used in Darjeeling tea plantation:</td>
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<td></td>
<td>1. Assessment of various quality parameters of organic matters has been done and a data base have been developed for nutrients (N, P, K, micronutrients) and heavy metals content of a range of organic matters collected from different sources. Wide variation in nutrient content was observed in organics collected from different sources. This data base will be used to prevent adulteration during the procurement of various organic inputs for organic tea cultivation in Darjeeling Hills.</td>
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<td>B. Selection of organics as nutrients source for Darjeeling tea industry with respect to C and N mineralization:</td>
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<td>The C-mineralization and nitrogen release pattern was measured up to 91 days and it was observed that nitrogen mineralization was medium to high from organic matters like Fish meal, Meat and bone meal, Soybean cake, Mustered deoiled cake, Groundnut cake, Til cake, Karanja cake and Neem cake. Nitrogen mineralization was lower in soils treated with Mahua cake, Copra cake and as expected the stabilized organic matters like Farm Yard Manure, Vermi compost and Poultry manure also exhibited lower N-release pattern.</td>
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<td></td>
<td>C. Development of carbon and nitrogen mineralization indices useful in acid soils of Darjeeling tea: Parameters like water soluble organic carbon (WSOC), total organic nitrogen (TON), C/N, short term N-mineralization at high temperature (40°C) with the objective to develop N-mineralization indices were studied in depth. It was observed that anaerobic nitrogen mineralization at high temperature for short period can be a useful tool for predicting carbon and nitrogen mineralizing capacity of various organic matters. Hence, this parameter can be used during the preparation of nitrogen fertilization schedule with organics for organic tea cultivation and for quick assessment of the quality of organic matters with respect to N-mineralization.</td>
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<td>D. Development of Liquid Organic Fertilizer for Darjeeling tea industry:</td>
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| | The concentrated organic matters may be used as liquid organic
fertilizer after fermentation. During fermentation the proteins are converted to amino acids and mineral nitrogen. Both aerobic and anaerobic submerged fermentation techniques were adopted for decomposition. It was observed that fish meal and karanja cake has potential to be used as liquid organic fertilizer after 7-14 days of fermentation under anaerobic condition.

E. Development of Organic Fertilizer Formulation for Darjeeling tea plantation:

Two organic fertilizer formulations for Darjeeling Tea have been developed considering the nutrient content (N, P and K), carbon and nitrogen mineralization pattern in acid soils of tea.

Formulation-1: Without fish meal, contains 3.60 %N, 1.25% P2O5 and 1.88% K2O
Formulation-1: With fish meal, contains 3.98 %N, 1.46% P2O5 and 1.89% K2O

IIT, Kharagpur: Project sanctioned= 1

Project No 20. Standardization of process parameters for machinery development in withering, maceration, rolling, fermentation and drying of tea

Name of Principle Investigator:- Prof. B. C. Ghosh

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<thead>
<tr>
<th>Objectives:</th>
<th>Withering</th>
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<tr>
<td></td>
<td>Improvement of energy efficiency and reduction of energy cost in withering.</td>
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<td></td>
<td>Achievement of uniform withering in the trough.</td>
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<td></td>
<td>Development of customized withering trough for reducing withering time in orthodox and CTC tea.</td>
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<tr>
<th>Maceration</th>
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<tr>
<td>Development of bench scale single cut devices for maceration of tea leaves.</td>
</tr>
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<td>Determination of the effect of process variables on maceration and physical characteristics of tea leaves.</td>
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<tr>
<td>Determination of the effect of process variables on chemical characteristics of macerated leaves and quality of made tea.</td>
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<tr>
<td>Determination of the effect cryo-cooling before and during maceration on physical and chemical characteristics of macerated leaves.</td>
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<th>Rolling</th>
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<tr>
<td>Determination of the effect of feed rate, size of leaf, speed of rotation, rolling pressure, path locus, duration of rolling and operating temperature on the degree of rolling achieved in existing machinery.</td>
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<tr>
<td>Finding the effects of alternate operating media on the quality of made tea.</td>
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<tr>
<td>Design, development, fabrication and testing of pilot plant batch type rolling device based on the results of the first two steps.</td>
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<tr>
<td>Design, development, fabrication and testing of pilot plant continuous rolling machine.</td>
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<tr>
<th>Fermentation</th>
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<tr>
<td>• Development of bench scale set up for continuous fermentation of macerated tea leaf.</td>
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<td>• Determination of physical and chemical changes in macerated tea leaf as affected by the design and operating variables of the fermenter.</td>
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<td>• Optimization of independent variables affecting the fermentation for maximum desirable characteristics in fermented leaf.</td>
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<th>Drying</th>
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<tr>
<td>• Thin layer drying study on orthodox and CTC tea in recirculatory, vacuum and fluidized bed dryers with the specific aim of establishing drying requirement of moisture reduction and aroma retention.</td>
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<tr>
<td>• Design, development and fabrication of pilot plant model of vibro-fluid bed dryer for drying of orthodox and CTC tea leaves.</td>
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<tr>
<td>• Drying study on orthodox and CTC tea using vibro-fluid bed dryer.</td>
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<td>• Statistical and computational analysis of drying data to identify relevant design parameters necessary for scale up of pilot plant models.</td>
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<th>Deliverables:</th>
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<td>• To help the tea industry for the improvement of tea machineries and process parameters in a new and innovative way.</td>
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<th>Budget</th>
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<td>Rs. 366.96 Lakhs</td>
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<tr>
<th>Final Outcome/technology Developed</th>
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<tr>
<td>The following novel tea processing machineries are developed on tea leaf withering, maceration of tea leaves, rolling of tea leaves, environment control fermentation chamber and modification/alternative tea leaf dryer.</td>
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1. Withering
As against conventional horizontal trough presently used in tea industry, a novel circular withering trough is designed and developed. The merit of newly designed trough is as follows:

i) Attaining uniformity in withering of tea leaf

ii) Less space requirement (by 2/3rd or 66% than the conventional one)

iii) Less energy requirement

A prototype model of 80 kg capacity of circular trough per batch was developed initially followed by increasing capacity to 200 kg and to 500 kg per trough. After the concept of withering of tea leaves using circular trough is proved, a 200kg capacity trough is installed at Nagrakata (Sub-station of TRA) for the validation and demonstration.

**Technology deliverable:** circular withering trough of 200 kg capacity

**Scope of improvement:** After demonstration the machine may be modified and further improved after receiving feedback from tea scientists / experts / industry.

2. Maceration
Compared to conventional three step CTC machine for attaining desired degree of maceration, a one step maceration unit is designed and developed. The merit of newly designed maceration device is as follows:

i) Withered leaves get macerated in a fraction of second in a single step compared to three steps of cutting required in conventional CTC machines. This reduces the chances of leaf damage due to localized temperature rise.
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ii) Less energy requirement
iii) Less space requirement
iv) Re-sharpening process is absent
v) Less chances of metal contamination
vi) Low rise of temperature during cutting process
vii) Scope of achieving desired grain size
viii) Output of the macerator can be connected directly to a new granulating machine avoiding the use of “Googhi” in the conventional system. The new granulating machine is very fast.
A prototype of single cut horizontal maceration unit having an output of 200 kg / hr is developed for demonstration. The machine will be demonstrated at Nagrakata for validation.

Technology deliverable: Design of a single cut horizontal tea maceration unit of 200 kg/hr capacity, scalable to higher capacities.

Scope of improvement: After demonstration the design would be modified with regard to mechanical performance and further improved after receiving feedback from tea scientists / experts / industry.

3. Rolling

Flat Belt Tea Roller: As compared to existing tea rolling machine which follows elliptical path of movement, a novel continuous tea rolling machine of linear path of movement is designed and developed. The merit of the machine is as follows: i) Roll the leaf continuous basis than batch process
A prototype of continuous flat belt tea rolling machine is developed of capacity 20-25 kg/ hr. The machine will be demonstrated in DTRDC, Kurshiong, Darjeeling for validation.

Technology deliverable: A continuous flat belt tea rolling device.
Scope of improvement: After demonstration the machine may be modified and further improved after receiving feedback from tea scientists / experts / industry.

4. Fermentation

In conventional fermentation bed perfect control device of temperature and humidity is not generally maintained during processing. An environmental control chamber is designed and developed for maintaining a standard temperature and humidity in the bed during fermentation process, resulting uniformity of oxidation process for achieving desired character of made tea. The merit of the machine is as follows:
i) Maintaining uniform temperature and humidity irrespective of change of ambient temp and humidity due to seasonal variation and climatic variation of tea growing area.
ii) This will help for getting uniform quality of tea year round.
iii) Cost of manufacturing is less.

Environment control Machine (ECM)
A pilot plant, Environment Control Machine at laboratory scale is designed and developed. The machine can be used as a research tool in R&D activity of tea related research.
Humidification unit:
A scaleup model of humidification unit is designed for the control fermentation of tea during processing. The machine will be demonstrated in Nagrakata (Sub-station TRA) for validation.

Technology deliverable:
i) Laboratory model of Environment control Machine (ECM)
ii) Design of scale up model of Humidification unit

Scope of improvement: After demonstration the machine may be modified and further improved after receiving feedback from tea scientists/experts/industry.

5. Drying

ECP dryer:
In conventional ECP dryer, after drying of tea, the exhaust hot air is generally released in the open environment. For reuse of hot air a recirculatory duct is designed and developed in ECP dryer. The advantage of the dryer is as follows:
i) Increase in thermal efficiency from 11-24%.
ii) Reduces steam requirement from 9.75 to 5 kg steam per kg water removed. The concept of hot air re-circulation will be demonstrated in tea industry in Darjeeling using ECP dryer.

Vacuum dryer:
As compared to conventional ECP or VFBD dryer mostly used in tea industry, the tea drying using vacuum dryer is experimented. The merit of this dryer is as follows:
i) Retaining more aroma in tea when dried in vacuum dryer in a temperature ranging from 50 to 80°C depending on imposition of vacuum pressure varying from 25 to 29” Hg.
The capacity of prototype vacuum dryer is 6-9 kg fermented leaf per batch. A design of vacuum dryer for 30 kg fermented leaf per batch is made.

Technology deliverable:
i) Hot air re-circulation system for ECP dryer
ii) Vacuum drying for achieving high level of aroma in made tea

Scope of improvement: After demonstration the dryer may be modified and further improved after receiving feedback from tea scientists/experts/industry.

Patent Applied:
i. Circular withering trough
ii. Horizontal maceration device of Research

2. Total no. of research papers published from the project with Impact Factors (if any): (Detailed list may be provided separately as annexure)
I. Published: 10
II. Under review :2

3. Seminars/workshops conducted for Technology transfer: Two