

# Plant Tissue Culture and Biotechnological Research and Achievements at different Institutions of Bangladesh

## **Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur-1701, Bangladesh**

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Biotechnology Division, Bangladesh Agricultural  
Research Institute,  
Gazipur-1701.

### **Year of establishment**

Biotechnology research at BARI started with the establishment of a tissue culture lab for potato seed production in 1985. Another tissue culture lab for micropropagation of horticultural crops was established with FAO assistance in 1993. A separate Biotechnology division was established in 1998.

### **Objectives**

- i. To identify disease and pest resistant genes from various wild relatives or landraces and cloning, sequencing and expression of pest resistant genes.
- ii. To identify plant genes and genomes at molecular level, to characterize plants, diagnose plant disease and pests, assay of biologically active components and gene mapping.
- iii. To develop transgenic plants using the identified beneficial genes through genetic transformation of the cultivated varieties.
- iv. To develop new genotypes of different crops through somatic hybridization and gene pyramiding.
- v. To introduce transgenic or somatic hybrids from abroad.
- vi. To create facility for trial of transgenic crops and risk assessments and field release for commercial cultivations.
- vii. To create facilities for tissue culture, micro-propagation and multiplication of high value fruit, vegetable and ornamental crops.

### **Existing capacity**

Recently modern laboratory facilities including a state-of-the art laboratory and a Class II greenhouse have

been established with the help of government (GOB) funding at BARI. Now, facilities for research on Molecular biology and Genetic Engineering has been developed. Scientists of the Biotechnology Division have taken a pioneering role to start biotechnology research in the country.

### **Facility development**

- i. Facility development of identify plant genes and genomes at molecular level, to characterize plants, diagnose plant disease and pests, assay of biologically active components and gene mapping.
- ii. Development of transgenic plants using the identified beneficial genes through genetic transformation of the cultivated varieties.
- iii. Modernization of tissue laboratory for tissue culture, micropropagation and multiplication of high value fruit, vegetable and ornamental crops.

### **Human resource**

#### **Name, designation & specialization**

1. Dr. Md. Al-Amin, CSO  
Tissue culture and Transformation
2. Most. Dilafroza Khanam, PSO  
Tissue Culture
3. Dr. Md. Abdullah Yousuf Akhond, PSO  
Tissue culture and Genetic Engineering
4. Dr. Mosharraf Hossain Molla, SSO  
Tissue culture and Transformation
5. Dr. Mahmuda Khatun, SSO  
Tissue culture and Molecular Genetics
6. Mohammad Kamrul Hasan, SSO  
Molecular Genetics
7. Md. Ehsanul Haque, SO  
Molecular Cell Biology and Plant Proteomics
8. Md. Rezwan Kabir, SO  
Tissue Culture
9. Md. Saiful Islam, SO  
Tissue culture

## Foreign collaboration

Biotechnology Division has introduced bio-engineered crops with the collaboration of ABSP II, project funded by USAID and technical cooperation with Cornell University. Since 2005 development of nine Bt brinjal varieties and two LBR potato has been running at BARI.

## Research achievements

### A. Tissue culture

Development of different protocol for seeding production relating to tissue culture of different crops namely, banana, jackfruit, pineapple, papaya, coconut, grape, malta, brinjal, okra, sweetgourd, teasel gourd, ginger, chilli, Watermelon, Chrysanthemum, Rose, Tuberose, orchid, Gladiolas, Strawberry and potato.

### 1. Somatic embryogenesis of Papaya

Immature seeds were cultured in four different concentrations of 2,4-D for callus formation and embryo development. Maximum explants produced callus using 2,4-D at a concentration of 10 mg/l whereas maximum number of embryo per explant were produced at 12 mg/l 2,4-D concentrations. Highest per cent of embryo germination was observed in 0.08 and NAA including 3.5 mg/l GA<sub>3</sub>.

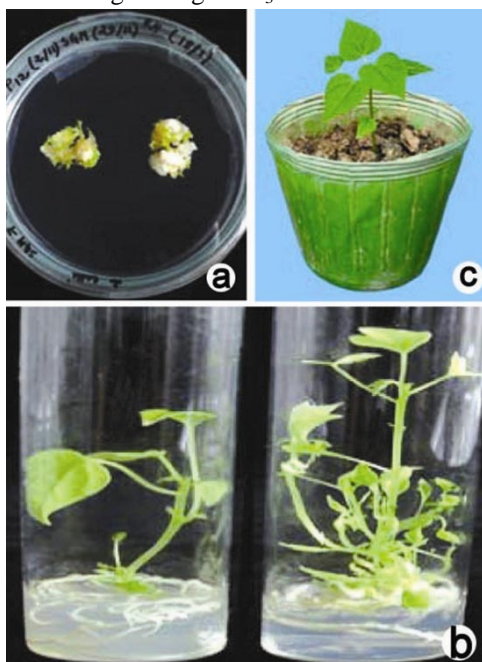


Fig. a-c. (a) somatic embryos (b) well developed plantlets (c) Established plantlet

### 2. *In vitro* regeneration of malta

Shoot tips of nucellar seedlings were cultured on MS medium supplemented with five different concentrations of BAP, Kn along with GA<sub>3</sub> 0.1 mg/l were tested for this study. Maximum shoot proliferation was recorded in T<sub>4</sub> - treatment. Well developed shoots were placed on half strength MS medium supplemented with four different concentrations of IBA. Maximum (95.13%) explants produced root in T<sub>3</sub> where 3.0 tap roots/explants were recorded at 28 days.

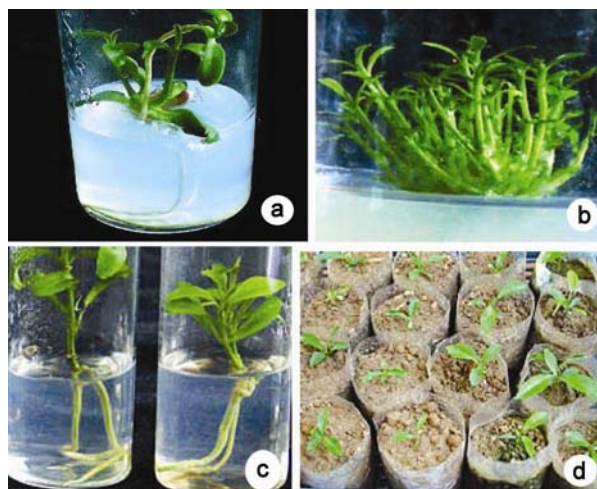


Fig. a-d. (a) *In vitro* seed germination (b) multiple shoot (c) rooting and (d) plantlet

### 3. *In vitro* regeneration of Chilli

Hypocotyls, shoot tip and cotyledonary nodes were inoculated on instant MS basal medium supplemented with different concentration and combination of BAP, TDZ, ZR and 2ip. Excellent growth and development of shoot tip was found on MS medium supplemented 1.0 and 1.5 mg/l ZR. Sufficient adventitious shoots were not yet found from shoot tips.

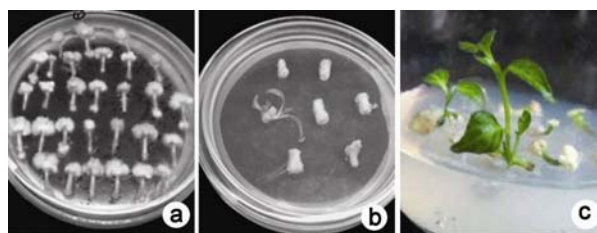


Fig. a-c. (a) Callus. (b) shoot from hypocotyle and (c) shoot from cotyledonary nodes.

### 4. *In vitro* regeneration of okra

Cotyledons and hypocotyls of BARI dharash1 were cultured on MS medium supplemented with different

concentrations of BAP, thidiazuron (TDZ), 2,4-D, BAP with NAA, BAP with IAA and zeatin with IAA. TDZ showed better compare to all other treatments where the highest percentage (75) and number of shoots (3.07) per explants were found in T<sub>1</sub> media.

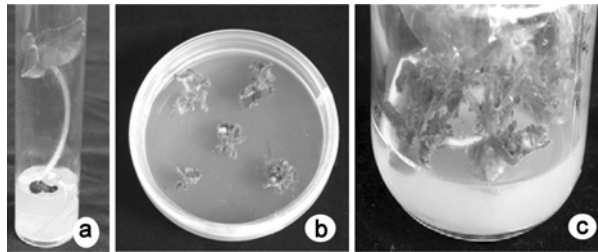


Fig. a-c. (a) Germinated seedling, (b) shooting from hypocotyle and (c) multiple shoot

### 5. *In vitro* regeneration of brinjal

Hypocotyl explants of BARI Begun-1 and BARI Begun-4 varieties were cultured on MS medium having

various concentrations of 2,4-D along with a control. BARI Begun-4 produced the highest (80.07) number of embryos was observed on T<sub>2</sub> medium. Success was noticed by both the varieties regarding *ex vitro* establishment of plantlets. The regenerated plantlets were successfully established in soil after proper hardening.

### 6. *In vitro* regeneration of sweetgourd

Cotyledons of BARI Mistikumra-2 were cultured on MS medium supplemented with different concentrations of BAP, NAA, TDZ and 2,4-D. the highest frequency (60.09%) of shoot induction and number of shoots per explants (4.74) were observed in T<sub>4</sub> treatment. The results obtained that both the highest percentage (83.33) of rooting and the lowest days (8.40) for root induction were found in T<sub>3</sub> combination.

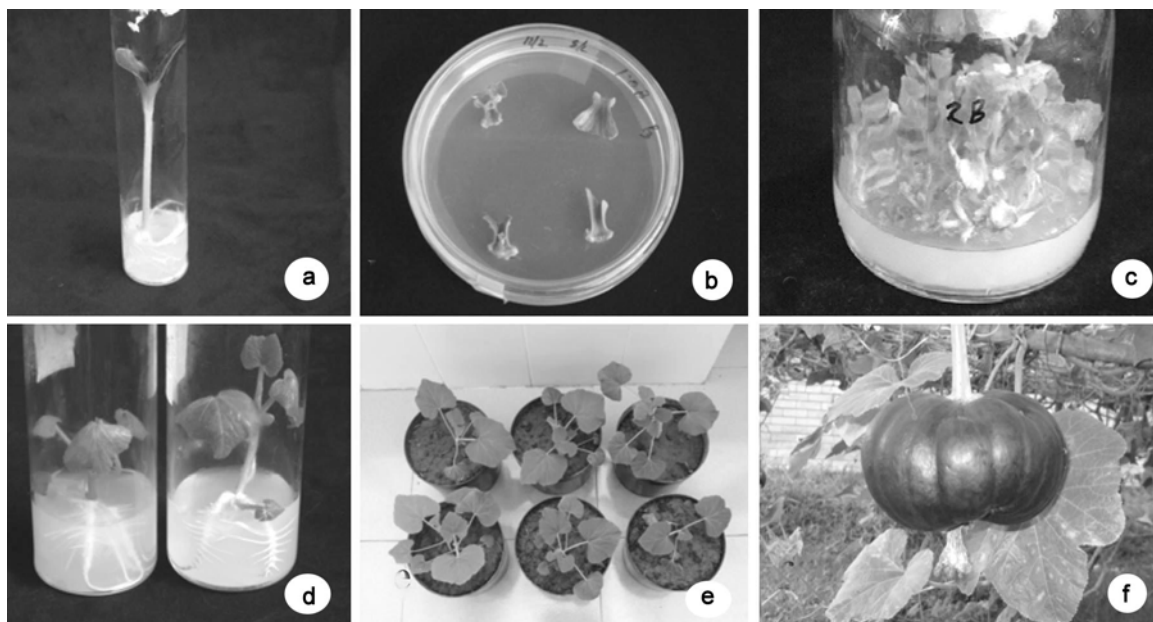


Fig. Regeneration of sweetgourd. a) 8-10 days old germinated seedling, b) shoots, c) multiple shoots, d) rooting, e) hardening of plantlets in greenhouse and f) fruit on established plant.

### 7. Standardization of protocol for *in vitro* production of bari kola- and BARI kola-4 and their validation in hilly areas

MS medium supplemented with 3 and 5 mg/l BAP were found suitable for *in vitro* production of multiple shoot from the shoot tip of sword sucker of BARI kola-3 and

BARI kola-4, respectively. Thirty four validation trials were established at farmer's field of Khagrachari and Rangamati Hill Districts along with two on-stations Performance was very good.



Fig. *In vitro* production of BARI Kola-3 and BARI Kola-4 and their validation in hilly areas.

## B. Genetic engineering

### 1. PCR-based detection and characterization of tomato leaf curl and other related geminiviruses in Bangladesh

To characterise geminivirus strains in different crops beginning with tomato, samples were collected from 8 different locations of the country and DNA from the infected leaf samples along with the virus particles was isolated. PCR was performed using degenerate and gene-specific primers following a standardized protocol. Most of the samples produced the diagnostic band of Tomato Leaf Curl Virus. Further analysis with PCR-RFLP and PCR-RCA methods revealed the specific virus genotypes. Sequencing of the complete 'A' genome of 32 isolates has been done to facilitate their phylogenetic analysis.

### 2. Development of tomato transformation protocol

Transformation was carried out using a binary plasmid containing the kanamycin resistant selectable marker gene and the GUS reporter gene. The assembled binary vector was transformed into the electrocompetent *Agrobacterium* cells using electroporation. Hypocotyls from a promising BARI variety were co-cultivated with *Agrobacterium* cells and cultured on selection medium containing kanamycin and carbenicillin. The regenerated shoots were transferred on rooting media and insertion of the transgenes was confirmed in 12 transformed tomato plants by PCR using transgene-specific primers for *nptII* and *gus*, respectively. GUS

testing was also performed for confirmation of the transgenic status.



Fig. Confirmation of transgenic tomato plants by GUS staining.

### 3. *Agrobacterium*-mediated genetic transformation of potato for salinity tolerance

*Agrobacterium tumefaciens* strain LBA4404 harboring Ti plasmid *PsCIPK* having one reporter gene (*GUS*) and selectable marker gene (*nptII*) was considered for this study. Some progress has been made and material was under processing. Presence of *nptII* gene in the construct was detected through PCR amplification. Transformation protocol is developed using this construct.



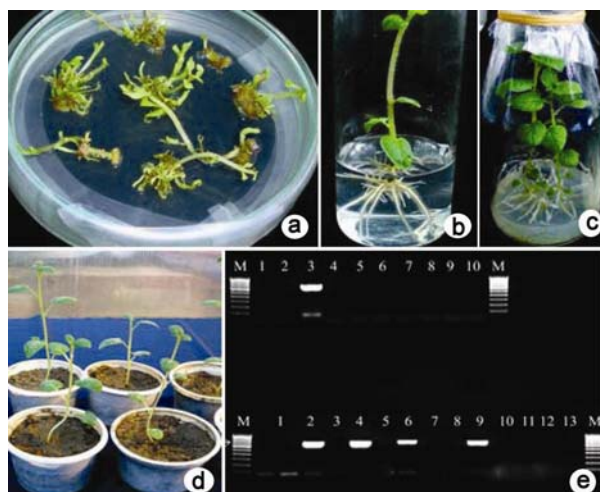


Fig. a-e. (a) Direct shoot regeneration, (b) Rooting, (c) *In vitro* plantlets, (d) *Ex vitro* establishment and (e) PCR amplification for inserted gene.

Biotechnology Division is also introducing bio-engineered crops with the collaboration of ABSP II, a USAID project. Since 2005 development of nine Bt brinjal varieties and two LBR potato has been running at BARI under collaborative research program with Maharashtra seed Company (Bt brinjal) and Cornell University (LBR potato).

### Bt brinjal

Brinjal mostly affected by brinjal shoot and fruit borer (*Leucinodes orbonalis* Gune) and sometimes 70% of crop yield damaged by it. Most of the cases farmers use pesticides superfluous way in field each day to get more yields. As a result, production cost of farmers is increasing due to excessive use of pesticide. That's why environment pollution and human health are becoming endangered. On the contrary, insects are developing resistance to pesticides.

BARI has developed transgenic Bt brinjal varieties against brinjal shoot and fruit borer by introgressing of Cry1Ac genes in nine popular varieties with the help of MAHYCO. Among the nine, four varieties were released as BARI Bt brinjal 1, 2, 3 and 4. To investigate the presence of Bt protein, ELISA test was performed for nine Bt varieties.

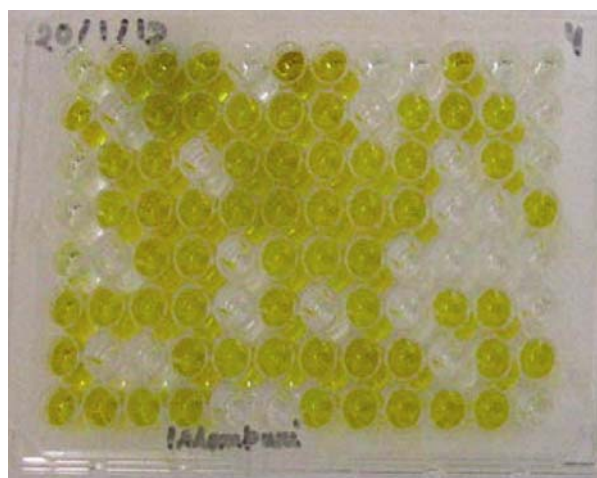


Fig. An exemplary view of ELISA plate for Bt protein.



Fig. Bt brinjal varieties

### LBR potato

The major challenge of potato is Late blight of potato which is the most devastating disease of potato. To protect this disease ABSP II Project with the financial aid by USAID and Scientist of Wisconsin University inserted RB gene from wild potato variety in our popular varieties cardinal and Diamonds to develop Late blight resistant potato variety. The confined field trial is being conducted at the different locations of the country. It is expected to release the Late blight resistant potato varieties very short time.



Transgenic Diamond lines



Transgenic cardinal lines

#### RESEARCH PROGRAM (2013-2014)

Sl. no.	Title
<b>Project 01: Protocol Development and micropropagation</b>	
1.	Standardization of protocol for advanced lines of strawberry and their large scale multiplication
2.	<i>In vitro</i> regeneration of okra ( <i>Abelmoschus esculentus</i> L. Moench)
3.	Development of an efficient regeneration system of banana
4.	Standardization of protocol, <i>in vitro</i> production of BARI Kola-3 and BARI Kola-4 plantlets and their validation trial at hilly areas
5.	Development of <i>in vitro</i> regeneration system for chilli
6.	Rescue of amritsagar banana from extinction through biotechnological approaches
7.	Development of <i>in vitro</i> propagation technique for heliconia
8.	<i>In vitro</i> regeneration of chickpea
9.	Study of comparative regeneration efficiency of different potato varieties
<b>Project 02: Molecular genetics and genetic engineering</b>	
10.	PCR-based detection and characterization of tomato leaf curl and other related geminiviruses in Bangladesh
11.	Transformation of tomato for broad spectrum resistance against leaf curl viruses
12.	Development of an efficient genetic transformation system for brinjal
13.	Marker-assisted transfer of salt tolerance <i>nax</i> genes in Bangladeshi wheat varieties
14.	Molecular and biochemical marker assisted assessment of bread-making quality in wheat
15.	Assessment of stress-tolerance attributes in wheat using gene-specific molecular markers
<b>Project 03: Validation/on-farm trial</b>	
16.	Validation trial of tissue cultured saplings of malta under field conditions
17.	Validation trial of tissue cultured jackfruit plantlets under field conditions

18. Observational trial of tissue cultured pineapple plantlets under field condition

**Project 04: ABSP-II research activities**

19. Confined field production trial of Bt brinjal
20. Multilocation trial of transgenic late blight resistant clones under natural field condition
21. Pathological tests of RB gene contained potato clones under natural field condition

**Future thrust area**

Potential utilization of the biotechnology tools in different BARI mandate crops are among the priorities in view of socio-economic improvement of the country.

Serial No.	Name of the crop	Characters to be improved
<b>A. Vegetables</b>		
1.	Brinjal	Shoot and Fruit borer resistance
2.	Potato	Late blight and Virus resistance
3.	Tomato	Virus resistance
4.	Okra	Virus resistance
5.	Cucurbits	Virus resistance
<b>B. Fruits</b>		
6.	Banana	Bunchy top virus, Panama and Sigatoga
7.	Papaya	Mosaic and Ring Spot virus resistance

**Tissue Culture Section,  
Biological Research Division,  
BCSIR Laboratories Dhaka, Bangladesh**

Bangladesh Council of Scientific and Industrial Research (BCSIR) was established in 1955 as a multidisciplinary research organization involved in scientific and industrial research for achieving self-reliance in industrial development.

The council has eight research laboratories/Institutes that has been pursuing research and development (R&D) activities in various fields of scientific and industrial interest and has contributed noteworthy services to the national causes covering research activities of both fundamental and fields in food and nutrition, pulp and paper, fiber and polymer, glass and ceramics, renewable and conventional energy aspects, mining, mineralogy and metallurgy, leather technology, aromatic and edible oils, physical instrumentation, drugs and medicines, biological science, biotechnology

and tissue culture and other major areas of research according to the need of the country.

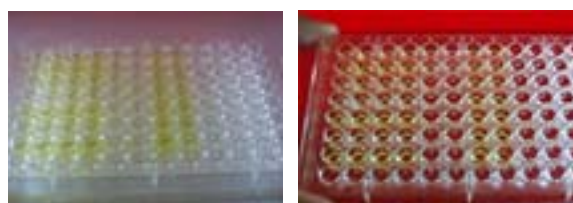


Fig. 1. Detection of plant viruses by DAS-ELISA method.

A programme on plant tissue culture was first initiated in the plant tissue culture section of BCSIR, Dhaka in 1993. Later on the programme was extended to a wide range. The research laboratory has gradually developed into a well-established biotechnology laboratory of Bangladesh with all lab facilities of molecular biology. In addition, the laboratory being used most modern instruments including virus-testing kit ELISA, PCR, gel



electrophoresis etc. that are headed by highly qualified and experienced scientists for generating the technology.



Fig. 2. *In vitro* regenerated Stevia, apple and Thankoni plants

Research on medicinal, fruit, fuel wood and ornamental plants is constantly underway to meet the requirements of the nation. Many of the findings can be applied commercially. Meanwhile some process has been developed for the large-scale production of economically important plants. A process has been developed on “Development of a cultivation technology of *Bixa orellana* L., a natural food color producing plant in the year 2007. Beside this, large-scale production of potato seedlings is one of the important achievements of this section.

Scientists in BCSIR tissue culture laboratory have been successful in developing the important plants namely *Stevia rebaudiana*, *Feronia limonia*, *Agle marmelos*, *Centella asiatica*, *Rauvolfia serpentina*, amlaki, apple, orchid, potato, tomato etc.



Fig. 3. PCR machine and ELISA-Reader.

Molecular biotechnology and genetic engineering is being paid special attention at BCSIR considering its potential for improving quality and quantity of important plants production. The thrust is to introduce and develop transgenic plants with a variety of traits of interest and relevant technologies. A number of Graduate, Post graduate and Ph.D. students from different universities are also engaged in the above research programmes.

#### Scientists engaged in tissue culture section:

Dr. Md. Salim Khan, Senior Scientific Officer.

Md. Ahashan Habib, Senior Scientific Officer.

Mrs. Shahina Akter, Senior Scientific Officer.

Mrs. Tanzina Akhter Banu, Senior Scientific Officer.

Mrs. Mousona Islam, Scientific Officer

### Bangladesh Tea Research Institute (BTRI) Botany Division Srimangal-3210, Moulvibazar

**Crop working with:** Tea (*Camellia sinensis* (L.) O. Kuntze]

**Objectives:** Plant Tissue Culture and Biotechnology Laboratory of this institute was initiated with following objectives.

1. Development of *in vitro* protocol for micropropagation of elite tea clones.



2. Establishment of *in vitro* propagation system for tea through organogenesis and somatic embryogenesis.
3. Establishment of anther culture to obtain homozygous diploid.

#### Existing capacity

- Laboratory facilities:** The institute has a small tissue culture laboratory with inadequate facilities.
- Human resources:** Three scientists are working in the institute who have technical know how about tissue culture.
  - Md Abdul Aziz, SSO
  - Shefali Boonerjee, SO
  - Md Abul Kashem, SO
- Local/foreign collaboration:** None at the moment.

#### Progress/accomplishment so far made

Shoot tip explants of different elite clones of tea (*Camellia sinensis*) were cultured *in vitro*. Regeneration of plantlets was observed. Plantlets thus obtained from regenerating media were subsequently transferred to rooting media but no root initiation was observed.

Stem segments of different elite clones were cultured *in vitro*. Callus initiation was observed. The calli thus obtained were transferred to regenerating media but no organogenesis was observed.

Anther of several tea clones were cultured aseptically *in vitro*. Profuse callusing was observed. The calli thus obtained were transferred to regenerating media but no organogenesis was recorded.

#### Main constraints

1. The laboratory is not properly equipped.
2. A non-availability of journal/literature regarding tissue culture/biotechnology in the library of the institute.
3. Collaboration with the other research institute especially which are working on tissue culture of woody plant needed.

#### Future planning and needs

**Planning:** The institute has taken up a plan to equip the laboratory in order to restart the work on tissue culture

and biotechnology. Initially we would like to start the work with micropropagation and anther culture.

**Needs:** The institute needs assistance in capacity development as well as technical support for human resource development. Collaboration with the other research institute especially which are working on tissue culture of woody plant.

### Supreme Seed Company Ltd. Leading Seed Enterprise in Bangladesh

**Founder :** Agriculturist Mohammed Masum

**Year of establishment** : 1978

**Registered as Private Ltd Co.** : 2001

**Registered as Public Ltd Co.** : 2010

**Brand** : HEERA

**Manpower** : 318

**Activities:** Breeding, production, processing, importing and marketing

**Crops** : Vegetable, cotton, maize, potato, and rice

#### Marketing Network:

Distributors: 270

Dealers : 1,400

Marketing Force: 55 (Veg.-18, Rice - 37)

Marketing Zone : 6 (Rangpur, Bogra, Jessore, Bhairab, Noakhali & Khagrachari)

#### Seed production (2012-13)

Sl. No.	Crop	Land area (acre)	Location
1	Hybrid Rice	2,200	Muktagacha and Madhupur
2	Hybrid Maize	80	Sherpur, Bogra
3	Potato	350	Birgonj, Dinajpur
4	Hybrid Cotton	70	Dimla, Nilphamari
5	Hybrid Vegetable	25	Birgonj, Dinajpur

**Storage Facilities :**

Sl. No.	Crop	Capacity (MT)	Location	Condition
1	Rice	2,000	Trishal, Mymensingh	Dehumidified & Cold
2	Maize	500	Trishal, Mymensingh	Dehumidified & Cold
3	Potato	4,500	Chandina, Comilla	Humidified & Cold
4	Cotton	20	Trishal, Mymensingh,	Dehumidified & Cold
5	Vegetable	25	Uttara, Dhaka	Dehumidified & Cold

**NB.** We have also 3 ambient stores with a capacity of 400 MT at Rangpur

**Research & development (R&D) facilities :**

Sl. No.	R&D Station	Location	Purpose	Area (acre)
1	R&D, Hybrid Rice	Trishal, Mymensingh	Breed Hybrid Variety	12
2	R&D, Hybrid Rice	Muktagacha, Mymensingh	Trial & evaluation	14
3	R&D, Vegetable	Bhaluka, Mymensingh	Breed Hybrid Variety	12

**Laboratory facilities :**

Sl. No.	Location	Purpose	Remarks
1	Uttara, Dhaka	Seed Testing with Seed Health	Central Lab
2	Trishal, Mymensingh	Seed Testing	
3	Dinajpur Sadar, Dinajpur	<b>Tissue Culture</b>	

**Prime products :**

Sl. No.	Crop	Popular Hybrid Varieties	Origin/ Developed
1	Rice	Heera-1, Heera-2, Heera 5, Heera 10	Imported & own developed
2	Maize	Heera 101	Developed by Supreme Seed
3	Maize	Heera 109	Imported from China
4	Potato	Diamant, Cardinal, Granola	Imported from Holland
5	Cotton	Rupali-1	Imported from China
6	Watermelon	Glory Jumbo	Imported from Japan
7	Cabbage	K-K Cross	Imported from Japan
8	Radish	Everest	Imported from Japan
9	Cauliflower	Summer Diamond	Imported from Taiwan
10	Bitter gourd	Heera 27, Dil, Dipto	Developed by Supreme Seed
11	Ridge gourd	Asim, Satabdi, Summer Short	Developed by Supreme Seed
12	Tomato	Red Angel, Red Heart	Developed by Supreme Seed

**"Quality is Our Commitment and Farmers' Faith is Our Pride"**

**Kbd. M. Abu Sayeed**  
Deputy Project Director  
R&D (Vegetable), SSCL  
Mob: 01713-145213

## Bangladesh Sugarcane Research Institute (BSRI), Ishurdi-6620, Pabna, Bangladesh

Bangladesh Sugarcane Research Institute (BSRI) is one of the oldest research institute of Bangladesh conducting research on sugarcane – the raw material for sugar, goor and cane juice. Recently, new dimension is added in its research by adding other sweetener crops such as sugar beet, date palm, palmyra palm, golpata and stevia.

BSRI is proud to serve the nation attaining self reliance in the sugar and gur sector with its limited resource and manpower. Two basic functions are performed by this institute: a) Development of sugarcane varieties as well as improved production technologies and b) Dissemination of varieties and technologies to the farming community.

Biotechnological research activities of BSRI started establishing its own Laboratory in 1997. The Biotechnology Laboratory of BSRI established as Biotechnology Division on 27 February, 2011. Since 1997 to-date protocol development and optimization for micropropagation of sugarcane and stevia, somaclonal technique for sugarcane variety development, genetic transformation for stress tolerant variety development are achieved. Molecular markers technique for Marker Assisted Selection (MAS) of sugarcane varieties is in progress. To gear-up biotechnological research activities “Biotechnological Research Strengthening Project” of Bangladesh Sugarcane Research Institute under the Ministry of Agriculture is being running smoothly for sugarcane and ancillary sweetening crops development in Bangladesh.

### Research activities

- Studies with regeneration and micropropagation protocols development of sugarcane and stevia using leaf segments via callus culture, shoot tip and meristem culture.
- Working on to develop improved varieties of sugarcane for sugar, gur and chewing using biotechnological tools such as DNA Fingerprinting, Molecular Marker Assisted Selection (MAS) and Quantitative Trait Loci (QTLs) determination.

- Genetic transformation for development of stress (salt and drought) tolerant transgenic sugarcane.
- Trying to develop protocols for regeneration and micropropagation of date palm, palmyra palm, palm leaves (Gol pata) and Sugar beet.

### Research achievements

- Protocol of plant regeneration using leaf segments via callus culture for sugarcane somaclones development has been developed and optimized. Callus derived somaclones are being tested in the field conditions.
- Protocol of microropagation using shoot tip, leaf segments, tip and meristem for high quality setts (HQS) production and rapid multiplication of sugarcane has been optimized. Yield performances of micropropagated plants are being evaluated in the field conditions.
- Tissue culture techniques for development of salt and drought tolerant sugarcane have been optimized. Somaclones developed from salt and drought tolerant callus are being tested in the field conditions.
- Protocols for development and rapid multiplication via callus culture and shoot tip culture of stevia-an elite sweetening herb have been developed. Developed stevia plants cultivated in the field and stevia tea developed from harvested leaves.
- Protocols for Mushroom tissue culture without Lamiar-flow-hood with and without Aseptic Box and production using sugarcane bagasse have been optimized.

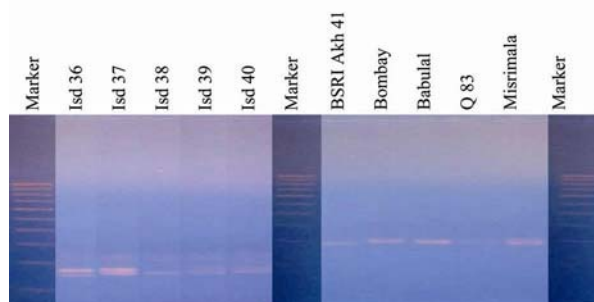


Fig. DNA fingerprinting of five bred variety one bred chewing variety and four chewing germplasm using SSR marker.

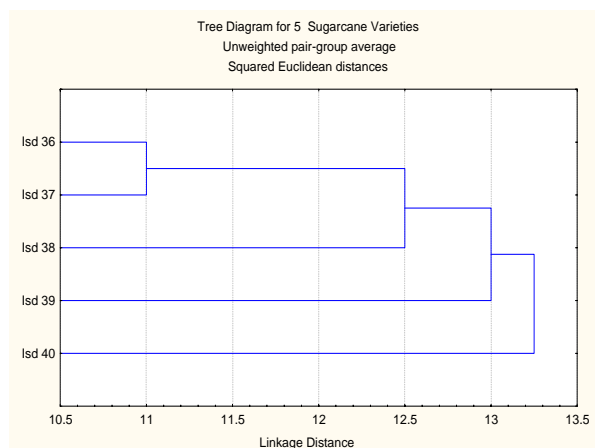


Fig. Tree diagram of five sugarcane varieties based on SSR markers

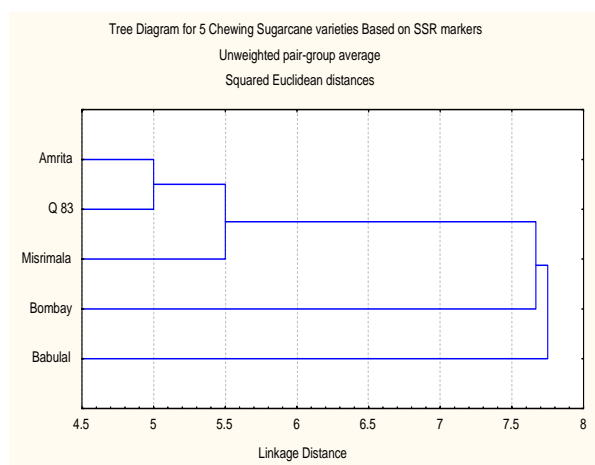


Fig. Tree diagram of one bred chewing variety and four chewing germplasm

- ☐ DNA fingerprinting of all released sugarcane varieties and five chewing varieties have been completed. Fingerprinting of all germplasms has to be completed to generate data for genetic linkage mapping, quantitative trait loci (QTLs) determination and Molecular Marker Assisted Selection (MAS) of sugarcane.
- ☐ Genetic Position of Sugarcane Breeding Programme of Bangladesh with Philippines, Indonesia, Thailand and Malaysia has been determined.
- ☐ Transformed sugarcane using *Agrobacterium*-mediated method is being maintained under *in vitro* contained conditions in the laboratory.

### Academic activities

- ☐ Six Ph. D. research students (including 3 university teachers) are being carried out their Thesis works at BSRI Biotechnology Division. So far two Ph. D. and 27 Master students have successfully been completed their research works.

### Future thrust

- ☐ Development of Marker Assisted Selection (MAS) for Sugarcane.
- ☐ QTL determination for Sugarcane improvement.
- ☐ Gene Construction for Our Own Use.
- ☐ Genome Analysis of Our Sugarcane Varieties for Specific Use.

### Present manpower (Scientists)

- ☐ Dr. Md. Amzad Hossain, CSO and Head
- ☐ Kuasha Mahmud, Principal Scientific Officer
- ☐ Nadira Islam, Senior Scientific Officer
- ☐ Asish Kumar Ghose, Scientific Officer
- ☐ Md. Abu Sayem Jiku, Scientific Officer

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### Department of Biotechnology Bangladesh Agricultural University, Mymensingh

The Department Biotechnology at Bangladesh Agricultural University (BAU) started functioning since 2002 with mandate of education, research and extension on biotechnology and molecular biology to meet the global need of researches and education on latest



developments in biotechnology. The Department has been offering courses at Undergraduate, MS and Ph.D. levels. The major activities of the Department include teaching Undergraduate, MS and Ph.D. students and conducting research on *in vitro* propagation of agronomic and horticultural crops, flower and medicinal plants, molecular characterization and diversity analysis of plants and fishes, and molecular breeding of plants, gene transfer techniques including *Agrobacterium*-mediated genetic transformation, etc. The mission of the department is to teach students about application of technologies using living organisms or its products to manufacture industrially valuable products, to improve plants or animals with expected characteristics, to develop microorganisms for specific uses. At the undergraduate level, Biotechnology department is offering elective courses: Basic Biotechnology (Theory & Practical) in Level 1, Semester 2, and Genetic Engineering and Biotechnology (Theory & Practical) in Level 4, Semester 1 & 2. By this time, three hundred and one (301) students have successfully completed their MS degree from this Department. From the very beginning, the Departmental Laboratory is well versed with the sophisticated equipments and systems. Nine Ph. D. Fellows obtained Ph. D. and 14 are enrolled and pursuing their researches for Doctor of Philosophy on genetic transformation of potato, garlic, chilli and other crops with a view to produce transgenic plants resistant to abiotic and biotic stresses. Some of the fellows are doing researches on molecular markers and molecular breeding. The department has successfully completed research on screening and transgenic potato production for salinity and drought resistance funded by USDA. The department has completed some other projects funded by BAURES, MOSICT, BARC, UGC and BCSIR. At present two projects are running under BAS-USDA fund. Transgenic researches are going on with potato, garlic, cucumber, chili papaya and brinjal for virus and insect resistance. Different tissue culture practices are going on with orchid, banana, rice, lentil, sugarcane, tomato, soybean, onion, gerbera anthurium and mushroom. Moreover, marker aided selection and genetic diversity analysis using RAPD and SSR markers are being conducted on many field and horticultural crops and fishes. The department is

hosting BABGE which has started publishing the Molecular Biology and Biotechnology Journal since 2003. Training facilities are also available on various aspects of Biotechnology and Genetic Engineering. Our vision is to develop tools and technique for the development of new crop varieties appropriate for the welfare of humankind and friendly for environment.

The Department of Biotechnology is therefore, playing an important role in both academic and research activities. However, it is essential to expand both academic and research activities of the Department with modern laboratory and field research facilities. Since major fields of biotechnology include plants, animals, fisheries and microbes and BAU has renowned experts, it is time to establish an “Institute of Genetic Engineering and Biotechnology” in the BAU campus where experts from different field of biotechnology can work together and can make BAU as a Centre of excellence for research on molecular biology and biotechnology and thus can contribute substantially to increase agricultural production.

#### **Completed and on-going research projects from the Department of Biotechnology:**

##### **01. Professor Dr. K. M. Nasiruddin**

- i. **BARC funded project title:** “Fungal disease resistance of local potato through gene transfer.” (completed)
- ii. **UGC funded project title:** “*Agrobacterium tumefaciens* mediated genetic engineering of potato for late blight resistance.” (completed)
- iii. **BAS-USDA funded project title:** “Genetic Engineering approaches for development of blight and streak resistant rice varieties.” (on-going)

##### **02. Professor Dr. Md. Shahidul Haque**

- i) **BAURES funded project title:** “*Agrobacterium*-mediated genetic transformation of garlic. (completed)
- ii) **BCSIR funded project title:** “*In vitro* plant regeneration and *Agrobacterium*-mediated genetic transformation of chilli” (completed)
- iii) **MOSICT funded project title:** Improvement of cucumber (*Cucumis sativus* L.) through *in vitro* regeneration and genetic transformation” (completed)

- iv) **UGC funded project title:** “Production of virus free propagules of garlic by in vitro meristems culture.” (completed)
- v) **BAS-USDA funded project title:** “Selection for Anthracnose Resistance in Chilli Germplasms and Development of Resistant Lines.” (completed)

#### **Teaching and research team:**

- 01. Professor Dr. K. M. Nasiruddin
- 02. Professor Dr. Md. Shahidul Haque
- 03. Dr. Md. Shahidul Islam, Associate Professor (Head)
- 04. Dr. Sabina Yasmin, Associate Professor
- 05. Dr. Fahmida Khatun, Associate Professor
- 06. Md. Jakir Hasan, Assistant Professor (Study leave)
- 07. Sumitra Saha, Lecturer

### **Seed and Biotechnology Centre Rural Development Academy (RDA), Bogra**

Rural Development Academy (RDA), Bogra is implementing diversified programs in the field of rural development in Bangladesh as a national autonomous institute. It is located in the North Bengal and is about 16 km away from the Bogra city as well as 200 km from capital city. Total area of the campus is 120 acres. The academy along with its stakeholders has achieved many national and international recognition as an efficient and leading organization for dissemination of seed and different aspects of Biotechnology up to village level. A seed and Biotechnology centre has already been established at RDA. Main objectives of this centre are to produce high quality disease free seed, conduct training, research and action research. Under this centre RDA has a seed production farm of 80 acres of cultivable land with seed processing facilities, seed health laboratory and tissue culture laboratory. Ministry of local government, Rural Development and Cooperatives, Seed wing of Ministry of Agriculture, Planning Commission and some other public agricultural universities are providing assistance to develop this centre as an effective one.

RDA tissue culture laboratory started its journey in 2006. At the beginning this laboratory was only involved in production of disease free potato seed of two varieties Diamant and Cardinal but at present it is working with seven varieties of potato, strawberry, stevia, grape, orchid, banana along with mushroom and trichoderma. Seven persons including 4 scientists are working in this center presently.



#### **Aims and objectives**

Specific objectives of the center are as follows

- Conduct research and demonstration activities on different aspects of Biotechnology for rural development
- Provide training for Human Resource Development on tissue culture
- Technology transfer to the stakeholders and beneficiaries
- Initiation of collaborative research program with different relevant organization and
- Conduct awareness building programs on different biotechnological issues.

### Available facilities

- Well-equipped laboratory for plant tissue culture of various species
- Automated greenhouse, hardening shed
- A demonstration farm of about 80 acre of land with all kinds of modern facilities
- Skilled and efficient manpower.
- Modern laboratory for trichoderma and mushroom production

### Research Programs already conducted

- Production of disease free potato seeds through meristem culture for commercial use
- *In vitro* propagation of grape (*Vitis vinifera*)
- Protocol development for in vitro regeneration of some commercially important varieties such as banana, orchid, stevia etc.
- Regeneration of strawberry through shoot tip culture.



### Achievements

- More than 300 beneficiaries are given skill development training on plant tissue culture in which most of them are self-employed at present;
- Each year around 3500 man-days work opportunity has been created by this center
- Seed potato of seven different varieties e.g.

Granola, Diamant, Cardinal, Ultra, Courage, Lady Rosetta and Asterix are regularly produced

- From the beginning more than 500 MTs of disease free seed potatoes of different level e.g. prebreeder/mini-tuber, breeder and foundation are produced
- Around 0.5 million of disease free potato plantlets are produced
- Every year farmers' field day is arranged due to demonstrate practical experience
- Each year thousands of farmers are getting advisory services.

### Future Plan

- Extension of biotechnology laboratory in terms of working area and capacity;
- Conduct more technology specific national and international training on plant tissue culture;
- Apply biotechnological tools for the wellbeing of rural poor.

### Scientific manpower of Seed & Biotechnology Centre

1. Md. Feroz Hossain, Director (Project Planning and Monitoring division)
2. Md. Mizanur Rahman, Deputy Director
3. Md. Asaduzzaman, Assistant Director
4. Suvagata Bagchi, Assistant Director



**National Institute of Biotechnology  
Ganakbari, Asulia, Savar, Dhaka-1349,  
Bangladesh**

National Institute of Biotechnology (NIB) is functioning as an autonomous organization under the Ministry of Science & Technology, Govt. of Bangladesh. It is expected that the establishment of this Institute is essential for ensuring socioeconomic development for ever increasing population of the country through the benefits of biotechnology. Recent gazette notification of National Institute of Biotechnology Act 2010; National Institute of Biotechnology service regulations, 2011 and approval of organization structure of the Institute will facilitate administrative and research management through appointing board of governors, researchers and employees and to formulate biotechnology policy, policy guidelines and action plan.

The Institute is operating eight separate division's viz., plant biotechnology, animal biotechnology, fisheries biotechnology, environmental biotechnology, microbial biotechnology, molecular biotechnology, Gene bank and Human resource development.

NIB has been organizing Human Resource Development (HRD) program since 2009 to develop skilled manpower on biotechnology. Presently, the institute provides HRD programs with recent advancement in the field of biotechnology for graduate and post graduate students, researchers, academicians and professionals. Moreover, awareness programs are also being organized on the potentials of biotechnology and genetic engineering among the policy makers, researchers, stakeholders, farmers and consumers.

NIB is working as an affiliated center of International Centre for Genetic Engineering and Biotechnology (ICGEB), Trieste, Italy. NIB is the nodal agency in Bangladesh for the Biotechnological issues of the SAARC countries and focal point of National Task Force on Biotechnology of Bangladesh (NTBB) and

National Executive Committee on Biotechnology (NECB).

#### **Major facilities of NIB**

NIB is facilitated with modern equipment and other physical and research infrastructures. Genetic engineering and molecular research facilities like genome analysis (PCR, qPCR, DNA sequencing), DNA fingerprinting and cloning, fluorescent microscopy, HPLC, IR, Biolog are available at NIB. Moreover, Plant biotechnology division has a well equipped laboratory with tissue culture room, media preparation room, growth room, automated green house, hardening house and experimental plots. Other research divisions have laboratories with amenities like animal cell culture, immunological study, cryopreservation, environmental samples analysis and fermentation technology, experimental animal house and shed, experimental and brood ponds for biotechnology and genetic engineering research.

#### **Plant biotechnology division**

Research activities of plant biotechnology division are focused on two broad aspects; plant tissue culture and plant genetic engineering. At present, the division is working on micropropagation of economically valuable plants and to develop abiotic stress tolerant transgenic crop.

#### **Objectives of the division**

- Development of pathogen free, insect-pest resistant plant through tissue culture and genetic transformation.
- Development of stress (salinity, draught, flood) tolerant crop varieties through biotechnology and genetic engineering
- Micropropagation of rare, endangered, ornamental, medicinal and commercially important plants through tissue culture

#### **On-going research program**

- Development of stress (drought) tolerant rice varieties through genetic engineering
- Micropropagation of *Aloe vera* on a commercial scale



- Development of transgenic eggplant (*Solanum melongena* L.) with enhanced abiotic stress tolerance.

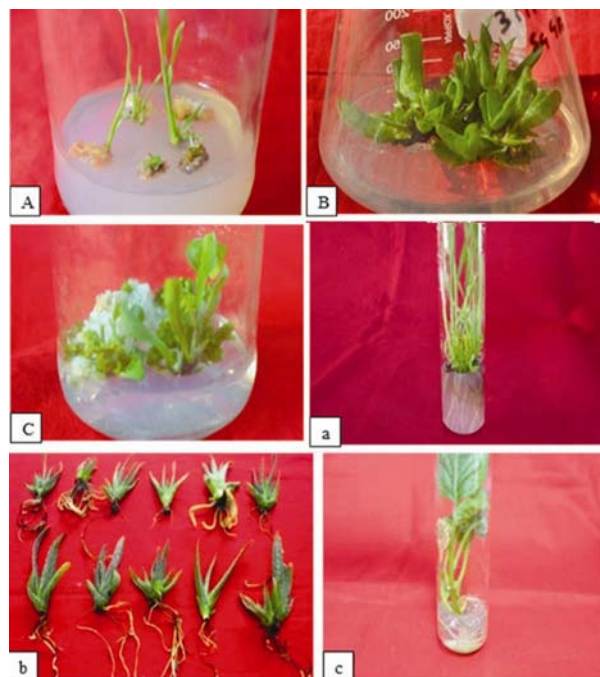


Fig 1. Regeneration of rice (A, a), *Aloe vera* (B, b), eggplant (C, c).

#### Achievement of the division

- Development of *in vitro* regeneration and gene transformation protocols for local high-yielding varieties of rice
- Micropropagation of various fruits (banana, strawberry, apple and jackfruit), medicinal (*Vitex*, *Aloe vera*, *Eclipta*, *Abroma*, *Stevia*, *Achyranthus*), ornamental (orchids, *Gerbera*, *Chrysanthemum*), vegetable (potato and tomato) and spice (zinger) plants have been developed and acclimatized in green house and field conditions

#### Future plan

- Detection of stress tolerance traits and genes and construction of vector for crop genetic transformation
- Marker aided selection to accelerate plant breeding.



Fig 2. Micropropagation of different plants at Plant Biotechnology Division of NIB.

D) Banana, E) Potato, F) *Vitex*, G) *Chrysanthemum*, H) Orchid, I) Apple, J) Zinger, K) *Stevia* and L) *Gerbera*.

#### Needs

- Manpower
- National and International collaborations

#### Collaboration

Bangladesh Institute of Nuclear Agriculture (BINA), Mymensingh (proposed).

### Tissue Culture Laboratory Silviculture Genetics Division Bangladesh Forest Research Institute, Soloshohor, Chittagong, Bangladesh

#### Background

The Bangladesh Forest Research Institute (BFRI) is the only national research organization for the forestry research which was established in 1955. BFRI has 17 research divisions that have been pursuing research on various activities of proper management of forest resources and efficient utilization of its production. BFRI has been conducting research on various aspects of forestry for the last fifty years and generated on related noteworthy technologies for end users. Among the 17 divisions, Silviculture Genetics Division has

been conducting research on development of tissue culture technique of forest species under the breeding and tree improvement programme since 1988.

### Laboratory facilities

Tissue Culture Laboratory of Silviculture Genetics Division of BFRI is well and modern equipped such as Top Pan Balance (3 digit), Analytical balance (4 digit), temperature and light controlled incubator, cool centrifuge, laminar flow bench, water distillation plant, water purification system, orbital shaker, fluorescent microscope, PCR machine, Gel Electrophoresis to conduct research on various aspects of tissue culture smoothly. Besides, green house and mist house is available.

### Limitations

A well equipped tissue culture laboratory of BFRI having shortage of trained manpower is a serious setback.

### Research activities

Research activities of Silviculture Genetics Division of BFRI are mainly on two aspects: i) macro propagation

through vegetative propagation technique ii) micro propagation through tissue culture technique. Research is being conducted on bamboos and forest trees including ornamental, fruits and medicinal plants.

### Objectives of the research of tissue culture technique

- i) To develop easy micro-propagation techniques for the forest species
- ii) To produce a homogenous plant population.
- iii) *In vitro* conservation of plants.

### Progress/accomplishment so far made

Successful protocols for micro-propagation of 12 bamboo species, six tree species and four medicinal plant species have been developed. Demonstration plots from the tissue culture raised plantlets of bamboo species have been established at Sugar Cane Research Institute, Ishwardi, Chittagong University, Jahangir Nagar University, Rajshahi University campuses and farmer's field at Bandarban.

The lists of established protocols of bamboo, forest tree, ornamental, fruits and medicinal plant species are given below:

### List of different bamboo species:

Sl. No.	Name of species	Explants used	Success and present status	Importance
1	Muli <i>Melocanna baccifera</i>	Nodal bud	Multiple shoots, rooted plantlets.	Dominant and natural hilly bamboo
2	Kanta <i>Bambusa bambos</i>	Nodal bud	Multiple shoots and rooted plantlets. Full grown clumps at bambusetum of BFRI	Thick walled village bamboo
3	Brandisii <i>Dendrocalamus brandisii</i>	Nodal bud	Multiple shoots and rooted plantlets. Full grown clumps in the demonstration plots	Large bamboo
4	Thai <i>Thyrsostachys siamensis</i>	Nodal bud/Seed	Multiple shoots and rooted plantlets. Full grown clumps at bambusetum of BFRI	Ornamental/Important bamboo
5	Budum <i>Dendrocalamus giganteus</i>	Nodal bud/Seed	Multiple shoots and rooted plantlets. Full grown clumps in the demonstration plots	Largest bamboo
6	Borak <i>Bambusa balcooa</i>	Nodal bud	Multiple shoots and rooted plantlets. Full grown clumps in the demonstration plots	Thick walled village bamboo
7	Rangoon <i>Thyrsostachys oliveri</i>	Nodal bud	Multiple shoots and rooted plantlets. Full grown clumps in the demonstration plots	Ornamental bamboo
8	Baizza	Nodal bud	Multiple shoots and rooted plantlets. Full	Important village bamboo

	<i>Bambusa vulgaris</i>		grown clumps in the demonstration plots	
9	Swarno <i>Bambusa vulgaris-var-striata</i>	Nodal bud	Multiple shoots and rooted plantlets. Full grown clumps in the demonstration plots.	Attractive coloured thick walled bamboo
10	Jaotha <i>B. salarkhanii</i>	Nodal bud	Multiple shoots and rooted plantlets. Full grown clumps in the bamboo	Important village bamboo
11	Makal/Talla <i>B. nutans</i>	Nodal bud	Multiple shoots and rooted plantlets. Full grown clumps in the demonstration plots	Important village bamboo
12	Bethua <i>B. cacharensis</i>	Nodal bud	Multiple shoots and rooted plantlets. Full grown clumps in the demonstration plots	Important village bamboo

#### List of different tree species:

Sl. No.	Name of species	Explants used	Success and present status	Importance of species
1	Nil gul mohor <i>Jacaranda ovalifolia</i>	Seed	Multiple shoots and rooted plantlets.	Very beautiful small tree with attractive blue flowers for ornamental, roadside or garden plantation
2	Hybrid <i>acacia</i>	Shoots tip	Multiple shoots and rooted plantlets.	Fast growing timber/fuel wood species. Free from heart rot.
3	Kanthal <i>Artocarpus heterophyllus</i>	Shoot tip	Multiple shoots and rooted plantlets.	Multipurpose timber and fruit tree species
4	Neem <i>Azadirachta indica</i>	Shoot tip	Multiple shoots and rooted plantlets.	Medicinal and multipurpose tree species
5	Segun <i>Tectona grandis</i>	Seed	Multiple shoots and rooted plantlets.	Best timber tree species
6	Haldu <i>Adina cordifolia</i>	Shoot tip	Multiple shoots and rooted plantlets. Seedlings are being maintained in the nursery	Multipurpose threatened forest species.

#### List of medicinal plant species:

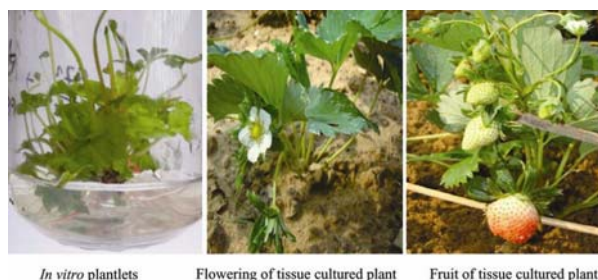
SL. No.	Name of species	Explants used	Success and present status	Importance of species
1	Kurchii <i>Holarrhena antidysenterica</i>	Node/ Shoot tip	Multiple shoots and rooted plantlets. Seedlings are being maintained in the nursery	Medicinal plant
2	Keshoraz <i>Wedelia chinensis</i>	Node/ Shoot tip	Multiple shoots and rooted plantlets. Seedlings are maintained in the nursery	Medicinal plant
3	Daaiful <i>Woodfordia fruticosa</i>	Node/ Shoot tip	Multiple shoots and rooted plantlets. Seedlings are being maintained in the nursery	Medicinal plant
4.	stevia ( <i>Stevia rebaudiana</i> )	Shoot tip	Multiple shoots and rooted plantlets. Seedlings are being maintained in the nursery	Non-caloric sweetening plant

#### Marketing status of tissue culture products

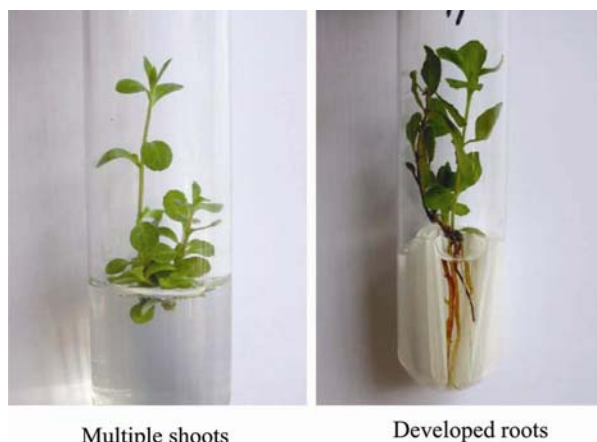
Tissue culture raised seedlings are being distributed among Government, Non-government Organizations

and Private Planters with the minimum revenue to the government fund.

### ***In vitro* regeneration of strawberry**



### ***In vitro* regeneration of Stevia**



### **Main constraints**

Lack of trained manpower and supporting staff.

### **Linkage with national and international organizations**

BFRI has very strong linkage with the NARS institutes of the country. On the other hand, it maintains co-operation/linkage through seminar; workshop and training programme at district level with the different NGO's, public and private planters throughout the country to disseminate the developed technologies and information.

### **Future plan**

In future, we may develop the tissue culture laboratory gradually into a tissue culture and biotechnology laboratory with all lab facilities, modern equipments and expert manpower of related work.

### **Need**

- i) Trained/expert manpower
- ii) Local / foreign training/degree/study visit for manpower development.

### **Researchers of Plant Tissue Culture:**

1. Md. Mahbubur Rahman  
Senior Research Officer
2. Nusrat Sultana  
Senior Research Officer
3. Shirin Akhter  
Research Officer
4. Saiful Alam Md. Tareq  
Field Investigator

### **Progress of Biotechnological Research and Development Activities at Lal Teer Seed Limited**

P. Banerjee, S. Mitra, M. Zaman, M E Haque and  
G.M. Mohsin  
Biotech Lab., LTSL

Lal Teer Seed Limited (LTSL) is the first private sector research based company which has been in operation since 1996. Since inception we have been involved in development and production of new breeds of high yielding, open pollinated and hybrid seeds as well as their processing and marketing. LTSL is now working as a development partner with public sector in supplying seed to the farmers; specially hybrid seeds of major vegetables along with pesticide products as crop protection measure. The company is now engaged in developing high yielding year round nutritious and ecologically sustainable crop varieties particularly sub mergence, salt and drought tolerance to contribute in enhancing productivity and income of rural agricultural households to achieve over all food security and alleviate nutritional deficiency of the population of the country.

At present, LTSL offers 161 varieties of 33 vegetable crops where 65 are hybrid and 99 are HYV. Besides, it has also 9 hybrid and 5 op rice, 3 hybrid maize , 2 hybrid cotton, 3 hybrid flowers, 2 varieties of jute seeds, one variety of each for potato, mustard and mung bean.

The company is operating its research activities through three R&D farms including the Central Research Station at Gazipur. Besides, it has 14 production zones across the country involving 6200 contract growers for vegetable seed production. In addition the company is



producing hybrid rice seed in five locations composed of both own management and contract growers system along with a Central Rice Breeding Station at Valuka, Mymensingh. Adaptation trials are regularly conducted in 30 agro-ecological zones of the country. It has also established its own facilities for processing, preservation and quality control, fully automated dehumidified air conditioned storage facility, seed grading, seed coating machineries and cereal and vegetable storage Go downs.

To ensure easy access of farmers to quality seeds it has established 30 marketing offices, 1000 dealers and 25000 retailers /mobile vendors across the country through a marketing net work. The company's marketing network has also been expanded in world seed export market revolving around 12 countries of the world at the moment. It has also a joint venture Seed Production Unit in Nepal.

The company is working with many national and international organizations including IRRI, IJSG, USAID-PRICE, AVRDC, IFC, IDE,BGI, Katalyst, Winrock International etc. Besides research collaboration, the company is keeping liaison with them in innovating new ideas to serve the humanity in terms of income generation, food and nutritional security. The company's recent introduction of low cost mini seed pack and seven colored mini seed pack through homestead vegetables gardening will go a long way in alleviating nutritional deficiency. However, the company through its activities contributing towards the economic development of the country in the form of raising development of climate resilient varieties, crop productivity, income and employment generation ,achieving food and nutritional security.

### **Policy of LTSL**

Lal Teer Seed Limited has been providing the environmentally adaptable and best quality seed to farmers. Considering customer need and preference we breed & develop new varieties that are high yielding, pests/diseases tolerant, heat tolerant and adapted to wide seasonal variation and changed climate. We enhance research consciousness in discovering new dimensions for development and enrichment with modern biotechnology and method. We are confident to

minimize seed import and build up capacity to export seed. We also develop and disseminate cultivation technology appropriate for the growers/producers at home and abroad. We establish strong networking programs by sharing resources with local and international institutes/organizations.

### **Aims and objectives**

- To ensure the availability of quality seeds for the farmers of Bangladesh and other tropical nations.
- To innovate and produce hybrid and high yielding vegetables of seeds. To focus on pest resistant saline and drought tolerance varieties
- To transform Bangladesh into a seed exporting nation
- To establish strong networking programs by sharing resources and expertise with local and international institute and organizations
- To achieve 65% share of the hybrid vegetable seeds and 20% share of the hybrid field crops to be marketed in the country by 2015
- To perform research programs for hybrid and high yielding varieties of rice, wheat, maize, pulses and other crops adaptable to the climate. To come up with at least one variety of each crop by the year 2015

The population of Bangladesh is growing and our demand for food is increasing day by day in an alarming rate. Thus, the major challenges we face in the country in the 21<sup>st</sup> century are to achieve food and nutritional security for the fast growing population of the world. Climate change is impeding crop production, distribution, and yields directly through changes in temperature, rainfall and precipitation, and indirectly by increasing pest and disease outbreaks. There is no other alternative but to introduce biotechnological interventions in conjunction with conventional plant breeding to increase cropping yields dramatically. In this situation Biotechnologists are capable of developing new plant varieties having resistance to pests and environmental stress, in lowering the cost of inputs and improving nutritional value of food crops. The biotechnology thus can offer opportunities for

increasing productivity, conservation of biodiversity and alleviation of poverty. In realizing to meet up the challenges of new millennium, LTSL has established a Biotech Lab to battle against environmental stress viz. heat, drought, salinity, diseases and insect pests.



Fig. Lab Assistant working in Laminar Airflow Hood.. Fig. Genomic DNA extraction in the Lab.

Biotechnology is one of the important Division of Lal Teer. The Division has established lab facilities for doing research on plant Tissue Culture and molecular biology. The lab is equipped with sophisticated equipment for doing world class research in the area of modern technology. The department has highly qualified, team oriented, skilled, and motivated man power as well as expert consultant for conducting quality research in breeding and biotechnology. At present, the Department is working on *in vitro* propagation and rapid multiplication of potato to ensure virus free healthy seed tuber production. Every year the lab is producing more than 50,000 (fifty thousand) plantlet under *in vitro* condition. Production of nuclear stock, pre-foundation and certified seed tuber is routine work of this Department. The tissue culture group is not only confined to potato seed tuber production, but it has also programs to regenerate plantlets on other crop like banana, orchid, gerbera, strawberry and ornamental plants. Molecular Biology Group of Biotech lab has been working on DNA fingerprinting of major hybrid varieties as well as hybridity test of produced hybrids seeds to ensure quality seed. The PCR base hybridity test is doing by the use of SSR marker to confirm the hybrid at DNA level. Germplasm characterization and molecular diversity analysis of available genetic materials are also in progress. Genetic transformation and transgenic variety development is one of the major objectives of this Division. Gene mapping and sequencing is also another objective of the research

area. It has started the activities with resistant gene hunting from wild relative of eggplant to develop bacterial wild resistant variety in Brinjal, late blight resistant variety for potato etc. Evaluation of drought resistant wheat, saline tolerant maize and subsequent marker assisted selection work is under process to fulfill the above objectives.



Fig. Scientist working in gel eletrophoresis. Fig. PCR amplification by SSR markers in Bitter gourd

It is noted here that a collaborative research program is going on with AVRDC (The World Vegetable Centre) and the formal agreement has made between Molecular Genetics Division, AVRDC and Lab Division, LTSL to give technical support to develop protocol regarding advanced molecular research.

Lal Teer has taken an ambitious task on Whole Genome Sequencing of Revering Buffalo (*Bubalus bubalis*) and it has been successfully completed by 2013 with the collaboration of Beijing Genome Institute (BGI), China. It is the first time in the world where the full genome sequencing is done in Revering Buffalo. In very brief, the whole genome sequencing works include sample collection, quality genomic DNA extraction, DNA library construction and sequencing, filtering of sequence data, genome assembly, annotation and bioinformatics analysis. The estimated genome size of Revering Buffalo is 2.946 Gbp. SOAP *de novo* was used for the assembly of the genome. It observed that, the contig size was 25036 bp, contig number was 31357 and a total of 21,550 genes were identified.

Genome analysis allow not only to understand the species but also know the molecular mechanism of economically important traits like Milk, Meat and disease resistance. Now, the genome skeleton of Water

Bufallo is available which can be the common platform for scientist all over the world.



Fig: The announcement ceremony of genome sequencing of the revering buffalo

Lal Teer believes it is very high time for Bangladesh to adopt strong biotechnology research program to face the challenge of food security and nutrition as well, otherwise country has to face a serious food and malnutrition problem in near future due to high rate of population growth with the effect of global atmospheric changes.

Lal Teer also will keep strengthening its own research infrastructure especially molecular characterization, sequencing and bioinformatics on variety development of agricultural crops.

### **Research at the Plant Biotechnology Laboratory of Dr. Zeba I. Seraj, Department of Biochemistry and Molecular Biology, University of Dhaka.**

#### **Collaborators**

International Rice Research Institute (IRRI), Los Banos, Philippines

Bangladesh Rice Research Institute (BRRI), Gazipur, Bangladesh

International Center for Genetic Engineering and Biotechnology, ICGEB

Section of Integrative Biology, University of Texas at Austin

National Institute of Biotechnology (NIB), Savar, Bangladesh

Dept. of Genetic Engineering and Biotechnology (GEB), DU

#### **Associate investigators:**

##### **Ph.D. students**

Dr. Mahbub Hasan, Assist Prof. BMBDU

Sabrina M. Elias: MBBISP fellow

Md. Sazzadur Rahman, SSO, BRRI

Richard Malo

Sajib Chakraborty, Lecturer, BMBDU

Habibul Bari Shozib

Kawsar Khan, Lecturer, BMBSUST

Shafiul Azam, ACME

Sumaiya F. Khan, Assist Prof, Jagannath U

Rumana Sultana Tammi, Asst Prof. JNU

##### **3 Completed PhD**

Noorain Rasul, Biotechnologist, Apex,

Laisa Lisa, Assoc. Prof., Jagannath U and

Rokeya Begum, PhD fellow CARS, DU

#### **Research associates**

##### **MS students**

Taslima Haque

Arif Ashraf

Samsad Razzaque

Umme Habiba

Sudip Biswas

Sarah Sarker

Shabnam Zaman

Afroza Ferdous

#### **Research assistants**

Md. Shamim Hossain

Rabin Sarker

Nazrul Islam

Al Amin

Raju Ahmed

#### **Focus**

Our focus is on producing rice tolerant to saline stress, suitable for growth in the coastal areas of Bangladesh. Any improved rice variety for the coastal region will make major impacts in the livelihoods of the resource-poor farmers of the region, in addition to increasing total rice production, which is important to maintain self-sufficiency in our staple food.

Our laboratory has been successful in establishing DNA marker-based breeding for development of salt tolerant strains of rice. While Bangladesh Rice Research Institute has done the breeding, we have identified suitable progenies having the salt tolerance loci, and thus helped speed up the breeding process using molecular technologies. We are also transforming rice with regulatory genes like transcription factors to produce both salt and drought tolerant rice.

#### Specific areas of work

- Marker-assisted backcrossed rice undergoing field trials in the Southern Coastal regions
- Search for salinity tolerance QTLs from Bangladesh rice landraces, using next generation sequencing technologies in collaboration with University of Texas at Austin
- Cloning and subcloning of genes reported to confer salt tolerance for *Agrobacterium*-mediated rice transformation, both via tissue culture and *in planta* transformation
- Rice transformation with genes reported to confer salt tolerance; assessment of tolerance and characterization of agronomic properties of modern rice after incorporation of transgene
- Use of the rice seed to produce stable Cholera and TB Antigens for vaccination purposes
- Use of Bioinformatics tools to characterize promoter sequences

#### Achievements

- DNA-marker assisted breeding lines produced and currently undergoing field trials
- Salt tolerant traditional rice produced with the full length vacuolar antiporter gene
- Salt tolerant transgenic rice with the Helicase gene produced in the genetic background of high yielding rice, BR28, 29, 36 and 47
- Salt tolerant BR27 rice produced with the HARDY Transcription factor
- Salt tolerant BR55 produced with the SNAC1 transcription factor gene

### Department of Genetic Engineering and Biotechnology

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Jessore 7408, Bangladesh

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Department of Genetic Engineering and Biotechnology was established in 2010. Genetic Engineering and Biotechnology Department launched four years undergraduate program aiming at offering B.Sc. in Genetic Engineering and Biotechnology degree. The department started one year M.S. in Genetic Engineering and Biotechnology degree program from 2014. Within a short period the department has developed a well organized Biotechnology Laboratory which was equipped with sophisticated instruments including PCR, gel electrophoresis, centrifuge machine, shaking water bath, table top micro centrifuge, digital micropipettes, vortex machine, incubator, UV trans-illuminator for doing molecular biology research. To conduct plant tissue culture research with modern facilities including growth chamber, laminar airflow cabinet, autoclave machine, distilled water plant, pH meter, analytical balance, magnetic stirrer, microwave oven, refrigerator and different types of culture vessels were also available in our Laboratory.

#### On-going research

1. Protocol development for Gerbera (*Gerbera jamesonii*) Tissue Culture: A perspective for cost efficient plantlets production.
2. Biotechnological approaches for micropropagation of strawberry (*Fragaria ananassa* Duch.) in Bangladesh.
3. Productions of virus free potato plantlets and micro-tuber through biotechnological approaches.
4. Identification and characterization of elite traits of native date palm (*Phoenix sylvestris*) for better juice and fruit yield through integrated biotechnological and molecular biological approaches.

#### Future plan

1. Improvement of important vegetables and ornamentals plants through *in vitro* and

*Agrobacterium* mediated gene transfer techniques for disease, flood, salinity and drought resistance.

2. Improvement of cereals and rice varieties for the tolerance of arsenic, cadmium and chromium.

#### Faculty Member

1. Professor Shaikh Mizanur Rahman, Ph.D
2. Dr. Md. Ziaul Amin, Associate Professor
3. Dr. A.M. Swaraz, Assistant Professor
4. Md. Nazmul Hasan, Assistant Professor
5. Aneesa Ansari, Assistant Professor
6. Dr. Md. Mashiar Rahman, Assistant Professor
7. Md. Abdur Rauf Sarkar, Lecturer
8. Md. Shahedur Rahman, Lecturer

### Biotechnology Division Bangladesh Rice Research Institute (BRRI) Gazipur-1701, Bangladesh

#### Introduction

Biotechnology Division is one of the major components of rice varietal development program area in BRRI. Since its inception, the division has been working for generating rice breeding lines through different biotechnological tools. Its major thrust includes the varietal development activities for high yield, quality, stress tolerance and biofortification of rice. Currently, it is mainly involved in rice tissue culture, genetic transformation, marker assisted selection (MAS), gene pyramiding, QTL identification and DNA fingerprinting of the modern rice varieties, advanced breeding lines and local land races.

#### Existing capacities

Sl. No.	Categories	Existing capacities
1	Laboratory facilities	Well equipped laboratory for Tissue culture, Marker assisted selection (MAS), QTLs identification, genetic transformation study, gene expression study etc.
2	Technical skill	Tissue culture, MAS, QTLs identification, genetic transformation, gene expression etc.
3	Human resources	Currently six scientists are working in this division. Among them four scientists having PhD degree in advanced biotechnological research including molecular biology and genetic engineering.
4	Local/ foreign collaboration	IRRI, USAID

#### Background of Biotechnological Research in BRRI

Biotechnological research was first initiated in BRRI with a small facility of the Plant Pathology Division in late September, 1982. A team of researchers of Plant Breeding, Plant Physiology and Plant Pathology was assigned to tissue culture for rice improvement. Later in 1987 with the development of a separate facility, tissue culture research was continued as a component of the Plant Breeding Division. A full-fledged division known as Biogenetic Engineering was established from April 1991. In April 1996, this division was renamed as Biotechnology Division to cover all aspects of biotechnological research for rice improvement.

#### Major objectives of the division

- Development of modern rice varieties suitable for both favorable and unfavorable environments through different biotechnological techniques.
- Molecular characterization of local germplasms to identify desired traits for incorporating them in future variety development programme.
- Identification of the desired tissue culture derived lines from seed, embryo and anther culture through pedigree selection, observational trails and primary yield trails.
- Establishment of efficient genetic transformation system for Bangladeshi rice genotypes.
- DNA fingerprinting of BRRI released varieties and advance breeding lines to protect biopiracy.
- Identification of useful QTLs /genes for high yield, disease resistance, salinity tolerance etc. for varietal improvement.
- Gene pyramiding for resistance to diseases and insect.
- Construction and screening of cDNA/ genomic library to characterize and use important genes for rice improvement.



## Progress/achievements

The major achievements are stated below:

- Methods and protocols have been established on culturing explants, such as seed, embryo, young panicle and anther of *indica* rice
- Higher regeneration rates from callus of rice tissue culture have been achieved in both *indica* and *japonica* rice by using various salts of sodium
- DNA fingerprinting was done on 50 BRRI released varieties to protect biopiracy
- Efficient genetic transformation system was established for Bangladeshi rice genotypes
- Two Bacterial Blight resistance genes (*xa13* and *Xa21*) have been pyramided in BRRI dhan29
- Molecular characterization of 127 local Aus germplasms has been completed
- *Sub1* gene has been introgressed into BRRI dhan44 for submergence tolerance
- Confined green house facilities were developed for transgenic research

## Future Plan

1. To develop manpower skill in rice modern biotechnological research to undertake frontier research programs appropriate to the future need.
2. Introduction and validation of transgenic rice events: Bio fortified rice, biotic and abiotic stress tolerant rice.
3. Identification, introgression and validation of agronomically important QTLs for high yield, biotic and abiotic stress tolerant rice. Positional cloning and sequencing of the target QTL region leading to the development of gene based markers.
4. Molecular characterization of existing germplasm, land races and related varieties for identification and usage in breeding program. Explore and utilization of available QTLs through Marker Assisted Selection in popular rice varieties.
5. Development and introduction of transgenic rice having useful genes for nutritional important, and biotic and abiotic stress tolerant.
6. Construction of cDNA/ genomic library to characterize important genes.
7. Development of short duration, stress tolerant, fine grain, high nutritional qualities rice varieties through anther culture and seed culture.

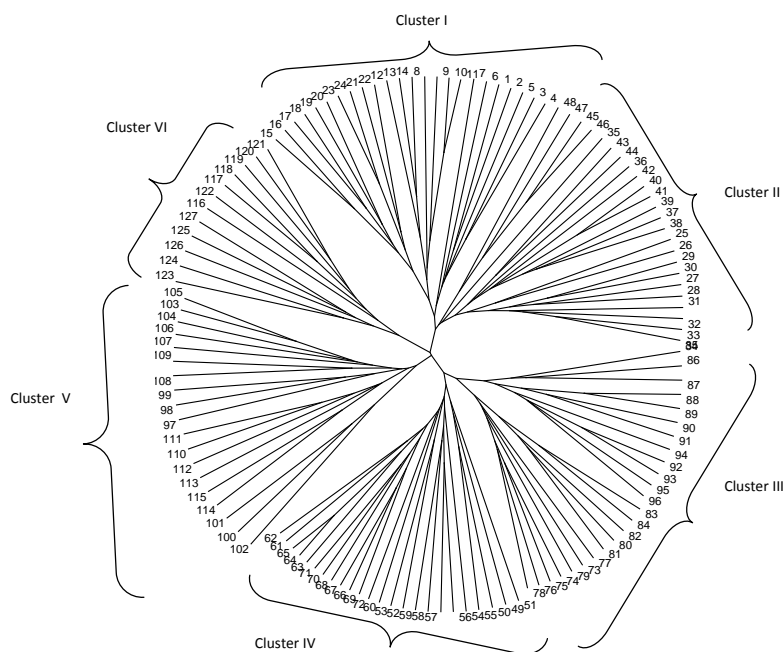


Fig. 1. An unrooted neighbour-joining tree showing the genetic relationships among 127 Aus genotypes (12 BRRI released and 115 Aus landraces) based on the alleles detected by 41 microsatellite markers

**Legend :** 1=Ajab Bett, 2=Agun Ban, 3=Atithi dhan, 4= Aalo Sate, 5= Begun Bahar, 6= Boilum, 7= Bailum-3, 8= Barmulka-2, 9=*Benaful*, 10=*Benaful*, 11= Bathuri, 12=Ausaloi, 13= Baismuguria, 14= Bador jota, 15= Bawoi, 16= Beri, 17= Beni muri, 18= BR319-1-HR-12, 19= Bora dhan, 20= Baisha Muri, 21= Bar Pa, 22= Balion, 23= Bil Kalae, 24= Balam, 25= Bhatkarari, 26= Boaila, 27= Barga Dhan, 28= Bali Bokri, 29= Chenri, 30= Chamka, 31= Chiknal, 32= Chitri, 33= Chapila, 34= Chakulya, 35= Dhula Biz, 36= Darial, 37= Goreswar, 38= Gutle, 39= Hidi 2, 40= Holat, 41= Holae, 42= Haita saita, 43= Honuman jota, 44= Hijoli Aus, 45= Haji Sail, 46= Hati Bajor, 47= IR19746-28-2-2, 48= Jhora, 49= Jamri saity 50= Jamurus, 51= Jagli, 52= *Japanese #7*, 53= *Japanese #3*, 54= Joba, 55= Korcha Muri, 56= Katar, 57= *Kali Bori*, 58= *Kali Boro*, 59= Kamani sail, 60= Koi juri, 61= Koblerash, 62= Khusni, 63= Korcha, 64= Kala, 65= Kalo Hizli, 66= Kheri Jamri, 67= Khamar Mundu, 68= Kaika, 69= Kadar Chap, 70= Laksmi lofa, 71= Lada Moni, 72= Lagi jota, 73= Lakhi Lata, 74= Manik Modu, 75= Malshira, 76= Mary satia 77= Manik Mondal, 78= Manik Mondol, 79= Mazra, 80= Modhu mala, 81= Manik Jor, 82= Magi Sarsa, 83= Moush Doll, 84= Morich Boti, 85= Mi-Mandi, 86= Mi-mandisarang, 87= Matia, 88= Nayan Tara, 89= Noroi, 90= Nusha Ratoi, 91= Nordi, 92= Porangi 7, 93= Parangi, and 94= Paik Juta, 95= Pankliiras, 96= Pipe Sail, 97= Panburi, 98= Padma Moni, 99= Padha Moidu, 100= Panchash, 101= Parija, 102= Ranga Moni, 103= Ranga Moni, 104= Rathail, 105= Sribalium, 106= Saribail, 107= Soloi, 108= Sodai Soru, 109= Soda, 110= Sail bogi, 111= Tarabali, 112= Tapa sail, 113= Tusha, 114= Udobali, 115= Zamir Saita, 116= BR1(chandina), 117= BR2 (Mala), 118= BR3(Biplob), 119= BR6, 120= BR7 (Bri Balam), 121= BR8 (Aasa), 122= BR9 (Sufala), 123= BR12 (Mayana), 124= BR15 (Mohinye), 125= BR16 (Sahya Balam), 126= BR20 (Nizamy), and 127= BR21(Niamat)



Fig. 2. Opening ceremony of transgenic green house in BRRI



Fig. 3. outside view of transgenic green house



Fig. 4. Scientists working at Molecular Laboratory

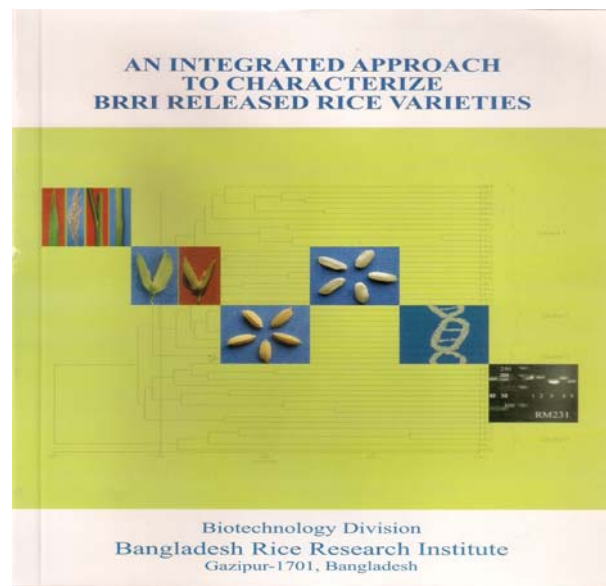


Fig. 5. Publication on molecular & physiological characters of 50 BRRI released varieties



Fig 6. Putative transformants of BRRI dhan29-TPSP plants grown in earthen pot

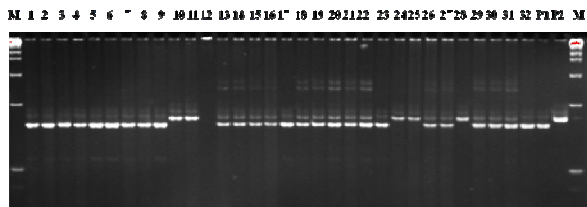


Fig. 7. DNA profile of homozygous plants selection with *SUB1C* 173 in BC<sub>5</sub>F<sub>2</sub> generation of BRRi dhan44 × BRRi dhan52, Legend: Lane 1-32 = individuals of BC<sub>5</sub>F<sub>2</sub> generation, lane P1= BRRi dhan44, lane P2= BRRi dhan52



Fig.8. Promising advanced anther culture derived rice lines.

#### Scientific manpower of the Biotechnology Division

1. Dr. Md. Enamul Hoque, PSO & Head
2. Dr. Shahanaz Sultana, SSO
3. Dr. Jannatul Ferdous, SSO
4. Dr. Nilufar Yasmin Shaikh, SSO
5. S.M. Hisam Al Rabbi, SO
6. Ripon Kumar Roy, SO

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## DEPARTMENT OF GENETIC ENGINEERING AND BIOTECHNOLOGY UNIVERSITY OF CHITTAGONG

The Department of Genetic Engineering and Biotechnology was established on Session of 2004-2005, under the leadership of Professor Dr. Mohammad Al-Forkan. The Department started academic function with the admission of fifteen Students to first year B.Sc (Hons). course in 2004-05. The Postgraduate course was first offered in Session of 2008-09 with the admission of only eleven Students, who passed B.Sc (Hons). examination from this department.

Fourteen teachers having academic background are now engaged in teaching and guiding research students. Of them one is Professor, two Associate Professor, Six Assistant Professors and five Lecturers.

At Present 101 students are studying in different years of B.Sc (Hons) and MS. courses. B.Sc (Hons.) course having a four year duration. The Department has been updated with the introduction of Research facilities in different aspects of Plants Sciences and courses included Genetic Engineering and Biotechnology, Cell Biology and Cytology, Microbiology, Biology, Bio-physical Chemistry, Fundamentals of Computer and IT, Plant Breeding, Enzymology, Organic Chemistry, Biostatistics, Pharmaceutical Biotechnology, Molecular Biology, Proteomics and Protein Engineering, Immunology and Industrial Biotechnology, Immunodiagnostics, Virology and Oncology, Food and Nutrition Biotechnology, Clinical Biotechnology etc.

Over the last 4 years, about 53 students have been awarded M.S. degree from this department. At present 10 research students are engaged in higher research leading to M.Phil and Ph.D. degree in this department. Many research projects are going on which are funded by different funding agencies including USDA, UGC and P&D-CU, etc. Mrs Sabrina Shamin Alam has been awarded “Prime Minister Gold Model” for her academic excellence in B.Sc (Hons.) examination. and also many students have received NST fellowship.

**Plant Biotechnology and Genetic  
Engineering Division, Institute of Food and  
Radiation Biology, AERE,**

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Fax: 88-02-7789620

Plant Biotechnology and genetic Engineering Division is one of the pioneer laboratories in the country to initiate research on plant tissue culture in the early eighties with micropropagation of forest trees. Later on other crop plants like cereals, fruit, medicinal, ornamental, vegetables, etc. have been included in the program. In the mid-nineties research on *Agrobacterium*-mediated genetic transformation has been initiated with the fiber crop jute and this technique is expanded gradually to other plants of economic importance viz. rice, sugarcane and Papaya.

#### **Objectives**

- To improve different economic important plants through *in vitro* mutagenesis viz. crop, ornamental, timber etc.
- To apply *in vitro* culture techniques for cloning of economic and endangered plants.
- To establish and exploit genetic transformation techniques for improvement of specific traits in plants of economic importance.

#### **On-going research and development programs**

1. Development of micropropagation techniques for multiplication and conservation of different crop plants (e.g. cereals, medicinal, ornamental, forest, fruit, vegetables, etc.).
2. Induced mutation for higher protein, amylase and yield in salt tolerant rice landraces by gamma irradiation.
3. Improvement of crop quality and stress tolerance for sustainable crop production using mutation techniques and biotechnology (RAS/5/045).
4. . Supporting Mutation Breeding Approaches to Develop New Crop Varieties Adaptable to Climate Change (RCA) RAS5056.
5. Application of genetic transformation techniques for the improvement of specific traits in plants of

interest (e. g. rice, jute, papaya, sugarcane, mustard, etc.).

6. Improvement of *Jatropha* using mutation techniques and biotechnology.

#### **Research Activities**

Research activities of this division are mainly focused on three aspects: (i) Plant Tissue Culture (Micropropagation) (ii) Improvement of Plant species through gamma irradiation and (iii) genetic transformation. Regeneration of plant from different explants (leaf, shoot tip, nodal segment, cotyledon etc) under appropriate conditions on defined nutrient media is called the plant tissue culture while alteration of a phenotype of an individual by the insertion of foreign DNA is called genetic transformation. Several research groups have been conducting research on Plant Biotechnology. Their aim is to develop regeneration protocol for multiplication of plants as well as application of gamma ray in *in vitro* culture system for the improvement of economic plants. Micropropagation techniques have been applied to plants of economic importance that include cereals, forest, fruit, medicinal, ornamental plants etc. In genetic transformation, *Agrobacterium tumefaciens* harboring plasmid is used as vector and is mostly used for the development transformation system as well as improvement of specific trait by the insertion of specific gene. Scientists of this division have also been engaged to improve of banana through *in vitro* mutagenesis and doubled haploids techniques and also to improve rice for salinity tolerance, composition or quality of grain, etc using mutation techniques and biotechnology in collaboration with IAEA and FNCA (Forum for Nuclear Cooperation in Asia).

Scientists have also been continuing their efforts to develop improved varieties of economically important plants by exploiting *Agrobacterium* –mediated genetic transformation techniques.

#### **Laboratory facilities**

Plant Biotechnology and Genetic Engineering Division equipped with different modern Plant Tissue culture and Molecular biology related instruments. These are as follows: Top Loading Balance, Ultra-cold Freezer,



Vortex Rota mixer, Water Still (FStream), Analytical Balance, Autoclave, Compound Microscope, Digital pH Meter, Fluorescent Microscope, Laminar Flow Bench, Magnetic Stirrer with Hot Plate, Microwave Oven, Refrigerator, Shaking Water Bath, Temp. & Light controlled incubator, Top Loading Balance, Ultra-cold Freezer, Centrifuge (Portable), Gel Electrophoresis (Horizontal), Low Temp. Incubator etc.

The PBGE division often uses 50 kCi Co<sup>60</sup> batch type gamma irradiator situated at IFRB for Radiation breeding research activities in different plants.

### Recent publication

1. P.K. Roy, A.N.K. Mamun, M. H. Kabir., M.R. Islam, M.T. Jahan and M.Z. Rahman. Development of an efficient *in vitro* regeneration protocol on an Orchid, *Phalaenopsis amabilis*. Nuclear Science and Applications. Vol.20.No.1&2.2011 (Published in December 2013).
2. M.T. Jahan, M.R. Islam, A.N.K. Mamun, P.K. Roy and M. H. Kabir. Organogenesis in gladiolus (*Gladiolus imbricatus* L. cv. Violet) using corm and cormel explant . Jahangirnagar University J. Biol. Sci. 2(1):105-111, 2013(June).
3. A.N.K. Mamun, A.K. Azad, M.H.Kabir, P.K.Roy, M.R.Islam, M.T.Jahan, M.A. Azam, M.L.Hakim and G. Ahmed. High yielding mutants with shorter life cycle selected in rice irradiated with carbomn ion beam. FNCA/MEXT technical Report. Achievement sub-project on composition or quality in rice (2007-2012). Mutation breeding project, Forum for Nuclear Cooperation in Asia (FNCA). March, 2013.
4. M. R. Islam, M. T. Jahan, A.N.K. Mamun, P.K.Roy, M.H. Kabir, and M. Z. Rahman. *In vitro* clonal propagation of *Musa sp.* Cv. Agnishwar- a rare Banana plant variety of Bangladesh. Jahangirnagar University J. Biol. Sci. 1(1): 63-71, 2012.
5. P.K.Roy, A.N.K. Mamun, M.H. Kabir, M. R. Islam, M. T. Jahan and M. Z. Rahman. *In vitro* indirect regeneration of Sugarcane ( *Saccharum officinarum*) var. Isd 16 through apical leaf culture. Bangladesh J. Life. Sci.23 (1): 123-128, 2011 (June).

### Achievements

- For the first time among Asian countries, this laboratory succeeded to produce doubled haploid plants in banana through anther culture.
- Irradiation dose has been optimized for irradiation of different plant species viz. rice, banana, jatropha etc.
- *In vitro* cloning systems of a number of forest, fruit, medicinal and ornamental plants have been developed. These are as follows:-

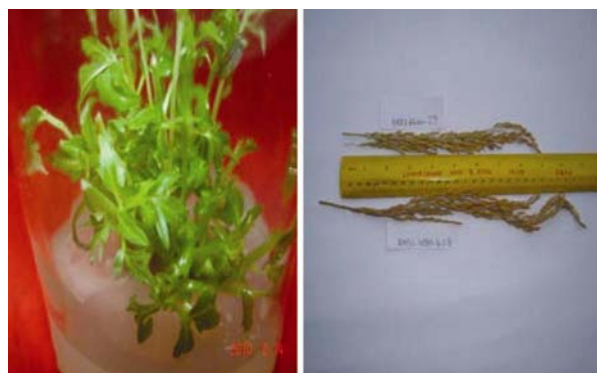


Fig. 1. Multiple shoot development in Stevia. Fig. 2. Ion beam mutant of BARRI Dhan 29

**Forest trees:** Banyan (*Ficus benghalensis*), Sisoo (*Dalbergia sissoo*), Teak (*Tectona grandis*), Peepul (*Ficus religiosa*), Ipil-Ipil (*Leucaena leucocephala*).

**Fruit Plants:** Carambola ( *Averrhoa carambola*), Apple ( *Malus domestica*), Kul ( *Zizyphus jujuba*), John's breed ( *Ceratonia siliqua*), Wood apple ( *Aegle marmelos* ) , Banana ( *Musa spp*), Orange ( *Citrus limon*),



Fig. 3. Development of Callus in Rice. Fig. 4. Gamma ray treatment in Sugarcane



**Medicinal Plants:** Neem ( *Azadirachta indica*), Nishinda ( *Vitex negundo*), Kurchi ( *Holarhena antidysentrica*), Periwinkle ( *Catharanthus roseus* ), Kalokeshi ( *Eclipta alba*), Tulshi ( *Ocimum sanctum*), Lajjabati ( *Mimosa pudica*), Thankuni ( *Centella asiatica*), Sheuli ( *Nyctanthes arbortrisris*), Dandokalosh ( *Leucas aspera*), Khayer ( *Acacia catechu*), Thuja ( *Thuja occidentalis*), Stevia ( *Stevia rebaudiana* ), Aloevera ( *Aloe barbadensis* ), Paulownia ( *Paulownia tomentosa* ) etc.



Fig. 5. Acclimatization of tissue culture derived Paulownia plants

**Ornamental plants:** Rose (*Rosa spp.*), Orchid, Gerbera (*Gerbera jamesonii*), Anthurium (*Anthurium andraeanum L.*), Chrysanthemum ( *Chrysanthemum morifolium*), Dhalia ( *Dhalia hybrida*), Gladeolus ( *Gladeolus grandifolius*).

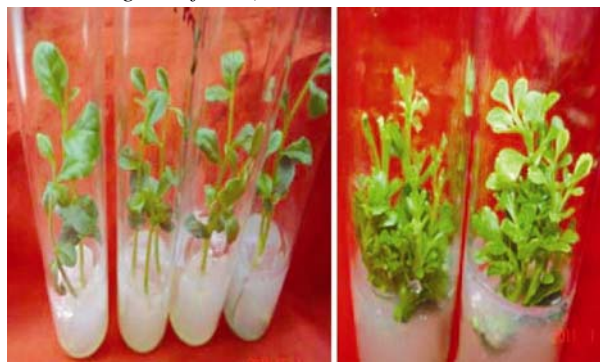


Fig. 6. *In vitro* rooting of Coddell. Fig. 7. Multiple shoot regeneration in Chrysanthemum

- *Agrobacterium* – mediated genetic transformation systems in tobacco, potato and papaya have been established.



Fig. 8. Gamma ray treatment in rice. Fig. 9. Control and mutant of sugarcane var. china

### Academic training and manpower development

Each year students from different public and private universities of Bangladesh receive research facilities and guidance from this division to complete their M. Sc., M. Phil. and Ph. D. thesis.

### Collaboration

PBGE division has collaboration with International Atomic Energy Agency (IAEA), Austria and Forum for Nuclear Cooperation in Asia (FNCA), Japan.

### Future plan

- Establishment of green houses / glasshouses for acclimatization of plants derived through tissue culture as well as confined trial of genetically transformed plants
- Establishment of a bio-diesel plant in AERE compound for semi-pilot scale production of bio-diesel from high yielding mutants of jatropha ( *Jatropha curcas L.*)
- Exploitation of genetic transformation techniques with the goal of producing transgenic plants having characteristics of disease resistance, stress tolerance, and high yield ability in sugarcane, rice, papaya, banana etc.
- Set up a molecular biology laboratory

### Present manpower ( Scientists)

Dr. A. N. K. Mamun , PSO and Head  
Dr. Protul Kumar Roy, PSO  
Mr. Mohammed Rafiqul Islam, SSO  
Dr. Md. Humayun Kabir, SO  
Mrs. Mustari Taslim Jahan, SO  
Mr. Md. Ziaur Rahman, SO

**Department of Biotechnology**  
**Sher-e-Bangla Agricultural University**  
**Sher-e-Bangla Nagar**  
**Dhaka-1207, Bangladesh**

Department of Biotechnology is one of the promising Department of Sher-e-Bangla Agricultural University. It has skilled and experienced faculty member headed by Dr. Md. Ekramul Hoque. We are doing researches

on the area of plant tissue culture and plant molecular biology. The Department had established regeneration protocol of various agricultural crops like potato, banana ginger, zerbera etc. Advanced cell culture technology researches like anther and microspore culture for haploid plant regeneration, meristem culture have been conducted in the Department. We have now been able to developed anther culture protocol in Rice and *Brassica*.

**Some research activities of Biotechnology Department**

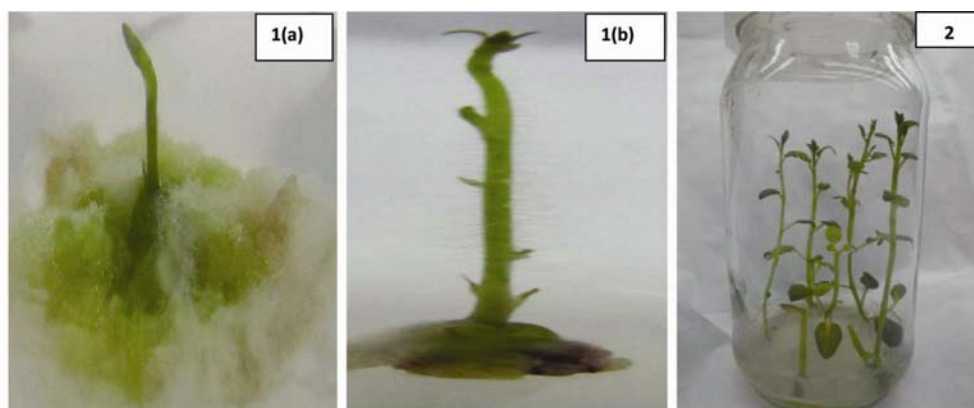


Fig 1 (a+b). Shoot and Plantlet regeneration from callus in potato

Fig 2. Micro propagation in potato.

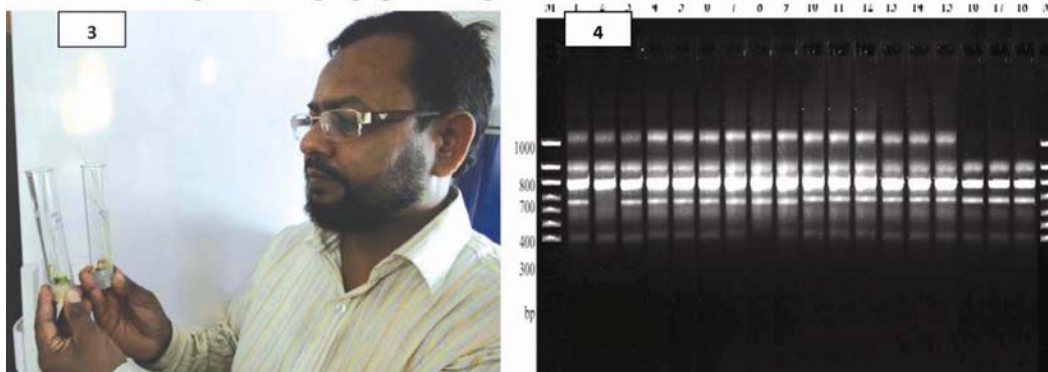


Fig 3. Anther culture and haploid plant regeneration in Rice.

Fig 4. Molecular diversity analysis in lentil

In addition, Biotechnology Department has done DNA fingerprinting, molecular characterization of germplasm and molecular diversity analysis of potato and lentil. Five National and International Projects on tissue culture and molecular diversity have been successfully run by Dr. Md. Ekramul Hoque which ultimately enriched the Department with modern sophisticated equipments like PCR machine, Electrophoresis system,

Gel documentation System, -20°C and -86°C deep Freezer. The Department is working smoothly on cell and molecular biology. Our mission is to develop resistant varieties for biotic and abiotic stresses in economically important agricultural crops. Genome sequencing and Bioinformatic works for crop improvement is our next vision.

**Plant Breeding & Biotechnology  
Laboratory  
Department of Botany  
University of Dhaka**

Research in plant tissue culture and biotechnology was initiated at the Department of Botany by Professor A. S. Islam back in 1977. Professor A. S. Islam, Professor M. Mozammel Haque, Professor Syed Hadiuzzaman, Professor Rakha Hari Sarker, Professor M. Imdadul Hoque and Dr. Mohammad Nurul Islam have received formal training on various areas of plant tissue culture and plant biotechnology from various International Institutes and Universities abroad.

Along with the development of expertise, the tissue culture and biotechnology laboratory of this department obtained substantial grants from UNESCO, USAID, JICA in the form of equipment and glassware. UNESCO helped in setting up the tissue culture growth room. This department has also received funds from the Ministry of Science & Technology, Govt. of Bangladesh to create facilities for genetic engineering and biotechnology. US Department of Agriculture (USDA) is supporting to conduct research through a project for the genetic transformation of lentil and other grain legumes. Alexander von Humboldt Foundation is also supporting a project for the development of fungus resistant chickpea and lentil.

A course on plant tissue culture and biotechnology was introduced in 1986 in M. Sc. classes as a special paper. Since then in this stream some 8-12 students pass out every year with special training on tissue culture and plant biotechnology. Most of the M. Sc./M.S. students carry out research in various aspects of plant tissue culture and genetic transformation. This department also offers M. S. and M. Phil programs in plant tissue culture and biotechnology. Recently M.S. program has been offered in three special branches in this department and plant biotechnology is one of them. Under this group students are given chances to do research on various aspects of biotechnology. The following students were awarded Ph.D. and one M. Phil. degrees during the last few years. The title of their thesis is mentioned along with the name of supervisors:

**Ph.D.**

1. Ratan Lal Banik of Forest Research Institute (FRI), Chittagong. "Biology and Propagation of Bamboos of Bangladesh" (Supervisor: Professor Syed Hadiuzzaman)
2. Md. Tozammel Hossain of BCSIR, Dhaka. "Somaclonal variation in triticale and wheat" (Supervisor: Professor A. S. Islam)
3. Shafiul Alam Bhuiyan, Biotechnology Division, BARI, Joydebpur, Gazipur. "Evaluation and Production of Somaclonal Variants in Groundnut (*Arachis hypogaea* L.)" (Supervisor: Professor A. S. Islam).
4. Usha Rani Das. BARI, Joydebpur, Gazipur. "Combining ability of seven inbred lines of maize and regeneration potentiality of F<sub>1</sub> hybrids through anther and embryo culture" (Supervisors: Professor Syed Hadiuzzaman and Professor Rakha Hari Sarker)
5. Shelima Begum, Dhaka Cllege, "Regeneration potentiality of four species of Amaryllidaceae and Liliaceae and the effect of gamma radiation on their regeneration." (Supervisor: Professor Syed Hadiuzzaman).
6. Salim Khan, BCSIR, Dhaka, "Development of efficient *in vitro* microtuber formation in potato (*Solanum tuberosum* L.) for its commercial exploitation". (Supervisors: Professor M. I. Hoque, Professor R.H. Sarker and Professor H- P. Muehlbach)
7. Bethee Das, Sherpur College, "Development of *in vitro* regeneration system in cultivated *Corchorus* spp." (Supervisors: Professor M.M. Haque and Professor M. I. Hoque)
8. Rahima Khatun, Jute Research Institute, "Improvement of fibre yield and associated traits in white jute (*Corchorus capsularis* L.)". (Supervisors: Professor R .H. Sarker and Dr. M.A. Sobhan)

**M. Phil.**

1. Ms. Bithi Das, Eden University College. "Somaclonal variation in two jute species: *Corchorus capsularis* and *C. olitorius* (supervisor: Professor M. M. Haque).

At present a good number of candidates have been awarded M. Phil. and Ph.D. from this lab.

### Current Research Programs

- (a) Development of suitable regeneration system for *Brassica* spp., Gerbera, Tea, Rice, Chrysanthemum, Potato, Tomato, Peanut, Lentil, Chickpea, Mungbean and Brinjal etc.
- (b) *Agrobacterium* mediated transformation of local varieties of Lentil, Chickpea, Mungbean and Brinjal, Tomato, Brassica and Jute for the development of fungal disease resistant varieties.
- (c) Induction of variation through gamma radiation, molecular characterization for identifying the variation by isozymes and RAPD analysis in Gerbera.
- (d) Induction of variation through gamma radiation and molecular characterization for identifying the variation by RAPD analysis in Chrysanthemum.
- (e) Detection and analysis of variation among field grown and in-vitro grown plants of peanut varieties.
- (e) Molecular characterization by RAPD analysis and direct gene transfer via Electroporation in aromatic rice varieties growing in Bangladesh.
- (f) Identification of superior clone using molecular marker and establishment of suitable organogenesis protocol in Tea.
- (g) Study of horizontal gene transfer from transgenic to wild relatives of brinjal through pollen pistil interaction.

### Micropropagation

Micropropagation of various ornamental, medicinal, timber and fruit plants have been carrying out in this Department. Protocols have been developed for large scale production of banana, orchids, chrysanthemum and potato micro tuber for commercial exploitation.

Earlier the technology of micropropagation of a number of species of bamboo, particularly, *Bambusa baluca* has been developed by Dr. Ratan Lal Banik. The results of field trial conducted by Dr. Banik both at FRI and at selected farmers' field show the superiority of this innovative *in vitro* protocol over conventional age-old methods so far used for propagation. Methods developed for initiation of rhizomes at the base of

branches of some bamboo species have been successful. Farmers have adopted this technique for mass multiplication of the varieties of their choice. This method has also proved to be a successful method for bamboo afforestation projects.

### Improvement of grain legumes

*Agrobacterium*-mediated transformation has been carried out for the insertion of desired genes into the three important grain legumes namely, lentil, chickpea and mung bean as well as peanut. Genetic transformation compatible *in vitro* regeneration protocols has been established for peanut, lentil and chickpea using various explants. Mung bean has also been included in this program. Transformation experiments have been conducted using *Agrobacterium tumefaciens* strain LBA4404 harboring binary plasmid pBI121, containing *GUS* and *nptII* (neomycin phosphotransferase II) genes. Stable integration of *GUS* - *glucuronidase* (and *nptII* (neomycin phosphotransferase II) genes was confirmed by polymerase chain reaction and histochemical assay.

With an attempt to develop fungal disease resistance lentil varieties two constructs with antifungal protein genes (vitis stilbene synthase and chitinase ) are being used following the transformation protocol developed through marker genes. Moreover another plasmid harbouring the selectable marker gene *bar*, which encodes the enzyme phosphinothricin acetyltransferase (PAT) and a *pgip* gene from raspberry (*Rubus idaeus* L.), coding for polygalacturonase inhibitory protein was used for lentil transformation. The expression of this recombinant gene can confer resistance against fungal





pathogens (*Colletotrichum*, *Botrytis* etc). A number of transformed shoots have been recovered from the decapitated embryo explant of Barimasur-4 following their transformation and selection using PPT (phosphinothricin).

### Improvement of peanut:

Transformation system has been developed for a local cultivars namely, DM-1 and Dhaka-1 of peanut using *Agrobacterium tumefaciens* LBA4404 harbouring binary plasmid pBI121( $\beta$ -glucuronidase) gene and nptII containing GUS (neomycin phospho transferase II) gene conferring resistance to kanamycin. Regeneration of multiplication shoots was achieved from infected young leaflet explants on MS medium-supplemented with 5.0 mg/l BAP + 0.5 mg/l Kn via organogenesis. The recovery of transformed shoots was achieved through a selection pressure of 300 mg/l kanamycin. The selected kanamycin resistant shoots were rooted on half the strength of MS medium containing 0.2 mg/l NAA and 50 mg/l kanamycin. PCR analysis revealed the integration of trans genes. Transformed rooted plantlets (To) were successfully transferred to soil where these plants produced viable seeds. The stable expression of GUS gene was observed among the various tissues of leaf, stem and pollen grains of the T1 plants. The seed developed from these T1 plants were normal in size and were found to be viable. The major constraints of peanut production in



Bangladesh include the incidence of several types of fungal diseases. Therefore two fungal disease resistance gene viz. vst (stilbene synthase) and vst-chitinase are now being used for the introduction of fungal resistance by applying the above mentioned transformation protocol.

### Jute transformation :

*Agrobacterium*-mediated genetic transformation method has been developed for different varieties of white jute (*Corchorus capsularis* L.). The regeneration system based on direct organogenesis from cotyledon with petiole, cotyledonary node, and mature embryo explants has been developed for the varieties of CVL-1, CVE-3, BJC-7370 and BJC-83. Different explants such as petiole attached cotyledon, cotyledonary node, mature embryo and mature embryo with cotyledon were tested with *Agrobacterium tumefaciens* strain (LBA4404/pBI121) with GUS reporter gene and nptII





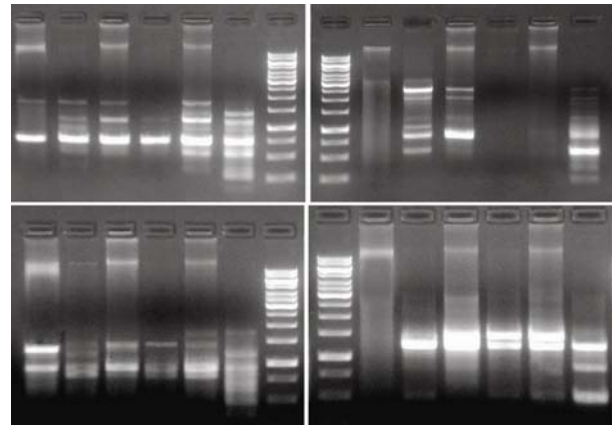
gene conferring resistance to kanamycin. Maximum transformation ability was obtained from petiole attached cotyledon and mature embryo explants. An *in vitro* regeneration protocol suitable for genetic transformation was also established. The shoots were subjected to rooting on MS with 0.3 mg/l IBA. The stable expression of the GUS gene was observed among the various tissues of shoot, stem and root of transformed plantlets. Genomic DNA isolated from these transformed shoots exhibited the stable integration of GUS and *nptII* genes through PCR analysis.

### Workshop on the improvement of tropical fruit trees

In collaboration with Winrock International and BAPTC&B this Department organized a practical workshop on improvement of tropical fruit trees through tissue culture and genetic engineering from



March 27 - April 6, 2003. Winrock International also supported the visits of two US scientists, namely, Professor Richard Litz of University of Florida and Professor Alan McHughen of University of California, Riverside.



### Collaboration with foreign universities :

- Plant Breeding & Biotechnology Laboratory of this Department has very active collaboration with the Research group of Prof. Hans-Jörg Jacobsen of University of Hannover, Germany (Grain legume project) & Prof. Hans-Peter Mühlbach's group of University of Hamburg (Sissoo die back project). Many of our students and faculty members obtained Ph.D., MS as well as short term training on molecular biology, genetic transformation from the above labs.

### Affiliated centre of ICGEB

This Department has been functioning as an affiliated centre of International Centre for Genetic Engineering and Biotechnology (ICGEB). With the support from ICGEB and in collaboration with the Department of Biochemistry and Molecular Biology of Dhaka University this affiliated centre has organized an International Practical Course on Novel Genetic Markers for Crop Improvement (4-13, November, 2001). Scientists from home and abroad participate in that workshop.

This affiliated center is involved in selecting potential candidates regularly for pre- and post-doctoral fellowships awarded by ICgeb. By now 11 Bangladeshi scientists received pre- and post-doctoral fellowships from ICgeb.

**Staff members engaged in Plant Tissue Culture and Biotechnology Research:**

Professor Rakha Hari Sarker

Professor M. Imdadul Hoque

Dr. Mohammad Nurul Islam

Rita Sara Borna

**Molecular Biology Laboratory  
Department of Biochemistry and  
Molecular Biology  
University of Dhaka, Dhaka-1000,  
Bangladesh**

Molecular Biology Lab is led by Professor Haseena Khan with three Research Associates, one PhD student and seven MS students from the Department of Biochemistry and Molecular Biology.

Jute, because of its immense influence on the economy and culture of Bangladesh is considered to be the country's identity. Despite its important role in the economy and environment, jute research and development has been neglected in Bangladesh. However, in recent years jute research has been progressing at a potential rate and many aspects of jute at the molecular level have been discovered.

Our lab's goal is mainly to develop improved jute variety which can produce quality fibre and withstand environmental stresses. Use of jute as the source of other commercially important products is included in the primary objectives. Recent studies have provided fascinating information about a putative *vps 51* (vacuolar protein sorting associated protein 51) gene showing strong correlation to low temperature tolerance in jute. Further studies have also identified a novel miRNA that is entrenched in the coding sequence of VPS 51 gene in jute.

Studies of stress responsive pathways like ABA biosynthetic pathway, cold acclimation pathways etc. are also of vital importance. These involve comparative gene expression analyses. However before consistent expression data is produced, selection and validation of reference genes to be used as internal control for qPCR for jute is essential, which is also a topic of intense observation. In conjunction to stress responsive pathways, studies have been conducted to show different temporal expression patterns for miRNAs in response to abiotic stresses. Deliberate changes in the expression levels of many miRNAs have been linked to tolerance to different stress conditions in many plants.

Studies are also underway on a comprehensive approach to characterize jute endophytes and establish their implicit growth assistance to the plant. This will aid us to better understand the mutual relationships between host and endophytes. It will help in designing a successful bio-control mechanism paving the way for the development of stress-tolerant jute in the future. Research experiments are also being conducted to identify potential role of jute in phytoremediation of heavy metals.

Meanwhile, high lignin content of jute hinders processing for industrial purposes (viz., paper pulp, textile, forage digestibility and biofuels). So, a reduction of the content of lignin in jute will help to boost its commercial usability. Study is aimed at reducing lignin in jute, by introducing artificial miRNA based vector in jute plants for down-regulation of lignin biosynthetic gene(s) in jute.

**Reduction of the lignin content of jute by gene silencing:**

Following genes of the lignin biosynthetic pathway have been selected for reduction in expression

1. *COMT*-Caffeic acid O-methyltransferase
2. *C4H*- Cinnamate 4 Hydroxylase
3. *C3H*- p- coumarate 3 Hydroxylase
4. *F5H* – Ferulate 5 Hydroxylase

An effective RNAi (SiRNA and amirRNA) technique has been chosen for reducing the lignin content of jute by introducing antisense RNA based vector in jute

plants for silencing the genes (individually) of lignin biosynthetic pathway.

After the introduction of the gene silencing vectors a reduction in the expression of the COMT gene was observed in the transgenic generations.

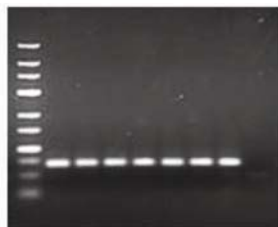


Fig: PCR showing Normalization with reference gene



Fig. Semi quantitative Expression of COMT gene in transgenic plants



Fig. RNA for Northern

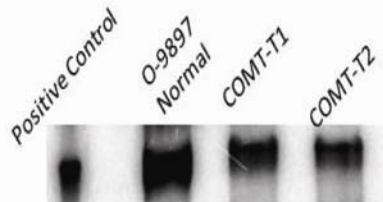


Fig. Northern blot analysis

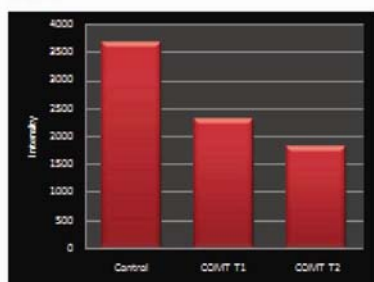
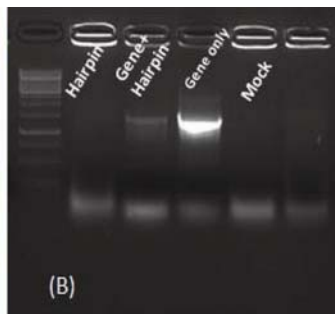


Fig. Graphical representation of Lignin depletion in Transgenic generations

A reduction of COMT expression (by northern blot) was observed in transgenic plants. The lignin content of the transgenic plants were also found to decrease.



(A)



(B)

Fig. Showing Agro-infiltration(A) and RT-PCR out comes of different infiltrated zones (B)

amiRNA (artificial miRNA) based hairpin constructs were designed for C3H and F5H genes. For the proof of functionality of the amiRNA construct a transient assay has been carried out. The C3H-amiRNA construct has been mobilized in tobacco through *Agrobacterium tumefaciens* mediated infiltration method. The functionality of the construct has been monitored by means of RT-PCR followed by small RNA northern.

The expression of the target genes has been monitored along a time-gradient in the transgenic generations and a reduction in the expression level has been observed.

A chemical lignin estimation method has been carried out both for transgenic and normal lines. The acid insoluble lignin content has been found to decrease by as much as 16-27 % in the lignin content of transgenic jute stems when compared to the normal lines.

**Biotechnology Laboratory, Molecular Biology Laboratory and Plant Genetic Engineering Laboratory, Institute of Biological Sciences, University of Rajshahi, Rajshahi-6205, Bangladesh**

Telephone: +880-721-750928 (Off.), Fax: 880-721-750064, [www.ru.ac.bd/ibsc](http://www.ru.ac.bd/ibsc)

The Institute of Biological Sciences (IBSc), University of Rajshahi, Bangladesh is the only national biological research institute established in 1989 to promote and provide facilities of higher studies and co-ordinate researches on advanced biological fields leading to M. Phil. and Ph D. degrees. The Institute of Biological Sciences acts as the core institute liked and supported by the departments of Botany, Zoology, Pharmacy, Biochemistry and Molecular Biology, Genetic Engineering and Biotechnology, Medical Sciences departments of the university. Main area of biotechnological research work presently institute of biological sciences has developed three (03) research laboratories namely i) Biotechnology Lab., ii) Molecular Biology Lab. and iii) Plant Genetic Engineering Laboratory. This Biotechnology laboratory is one of the best laboratories in the University of Rajshahi and by this time 31 Ph. D and 6 M. Phil degree have been awarded from these labs, Professor M. A. Bari Miah is the founder and head of the

Biotechnology Lab of the institute with group members Dr. Parvez Hassan, Professor and Dr. S. M. Shahinul Islam, Associate Professor of the Institute. Presently around eighteen Ph.D fellows have been pursuing research works with this group in three labs.

### Objectives

1. Callus induction, cell culture and studies on molecular phylogeny of banana, potato, orchids by molecular markers.
2. Development of arsenic resistance in crop plants through genetic engineering to combat food chain contamination in Bangladesh.
3. To establish a suitable *in vitro* techniques for biodiesel production and their conservation
4. Anther and microspore culture of different cereal crops and their genetic transformation with different agronomics traits.
5. Indigenous knowledge of herbal medicine and *in vitro* propagation and conservation of medicinal plants with isolation of secondary metabolites from cell culture.
6. Isolation and characterization of abiotic stress tolerance gene especially for drought and salinity tolerance in cereal and other crops.
7. To establish an efficient genetic transformation system for cereal and other crops by *Agrobacterium*-mediated genetic transformation using the targeted gene.

### Existing research facilities for biotechnological work at IBSc, RU

#### Biotechnology Laboratory

The Biotechnology Lab dimension is 30 × 40 ft, inoculation chamber (10 × 10 ft) and growth chamber (10 × 12ft). There is a modern and well equipped **Cell Culture Room**. Dimension of the room is 20 × 20ft. There are six (06) shakers machine and dark room facilities. For plant tissue culture facilities some necessary equipments namely PCR (Bio-Rad), -86°C Ultra low freezer, -27°C medical freezer, cooling incubator, fluorescence microscope, binocular microscope, leaf area meter, electric analytical balance, hot plate with magnetic stirrer, refrigerator, medical

deep freezer (-30°C), microtome machine, muffle furnace, laminar air flow, water distillation plant, autoclave, refrigerated high speed centrifuge, ultrasonic cell disintegrator, pH meter rotary vacuum evaporator, micro-oven, orbital shaker, air cooler, etc.

#### Molecular Biology Laboratory

Dimension of the molecular biology lab is 20 × 22 ft. Basic equipments and other research facilities of this laboratory: horizontal and vertical gel electrophoresis system, cold chamber for protein isolation. Olympus Photographic phases contact microscope, UV-VIS spectrophotometer, OSK Refrigerated Hi-speed centrifuge, Eppendorf top centrifuge, laminar air flow, freeze dryer, ultrasonic cell disintegrator, TLC facilities, freeze medical deep freezer (-30°C), Temperature controlled water bath, both with Shaker, High precision electronic digital pan balanced etc.

#### Plant Genetic Engineering Laboratory

Dimension of the molecular biology lab is 20 × 22 ft. Basic equipments for tissue culture, genetic transformation and related other facilities of this laboratory: PCR (Bio-Rad), laminar air flow cabinet, two layers incubator with shakers, refrigerator (24 Cu. Ft), mini or micro centrifuge (Eppendorf), analytical balance, vertical autoclave with quick-open door, quick spin centrifuge, benchtop pH meters, -26°C refrigerator, distilled water machine, digital magnetic stirrers hot plate, cooling incubator, vortex mixer, electrophoresis power supply unit (mini and midi), ordinary centrifuge (6 well), oven, nano lum mono/photo UV transilluminators, peristaltic pump, handled homogenizer. There is a separate inoculation chamber and a culture room also.

**Other facilities for biotechnological research at IBSc, RU:** The institute has eight (08) acres of land for its field and experimental work. Under HEQEP project, recently we established a culture and inoculation room and modern research facilities for applied research in the field laboratory. We also established a glass house for working with different crops round the year. Besides the above mentioned laboratories the institute has a modern seminar room,

store room, computer and data processing room, equipment and storage rooms.

#### **Collaboration:**

1. Japanese University Collaboration: Tokyo University of Technology, Japan
2. Swedish University Collaboration: We have research collaboration programme with Skövde University, Sweden
3. Research collaboration with ICGB, New Delhi, India with the research group of Dr. Narendra Tuteja under the CRP-ICGB Research grant.
4. ACI-IBSc collaboration for the commercial utilization of biotechnological research products.

#### **On-going international projects**

Third World Academy of Science (TWAS, Italy) declared the institute one of their TWAS Research Unit (TRU) and provided research grant on “Genetic transformation of antifungal genes into *Dalbergia sissoo*” 2. The institute has developed a collaborative research with Skövde university, Sweden and the Swedish Research Council for Environment Agricultural Science and Spatial Planning (FORMAS, 2008) provided research grant on “Developed of a genetically modified variety of rice (*Oryza sativa L*) for effective prevention of arsenic contamination of the major human food chain in Bangladesh”.

#### **On-going research activities in the area of plant biotechnology and genetic engineering:**

1. Callus induction, cell culture and studies on molecular phylogeny of banana (*Musa spp.*) cultivars by DNA fingerprinting.
2. Indigenous knowledge of herbal medicine and *in vitro* propagation and conservation of medicinal plants with isolation of secondary metabolites from cell culture.
3. Development of arsenic resistance in crop plants through genetic engineering to combat food chain contamination in Bangladesh.
4. Biodiesel Producing plants and Biotechnological approaches for their conservation.

5. Biodiversity and conservation of medicinal plants cultivated in Oushodi Gram Natore, Bangladesh.
6. Anther and microspore culture of major cereal crops and their genetic transformation with different agronomics traits.
7. Production of doubled haploid lines through androgenesis and identification of plants by molecular markers in maize (*Zea mays L.*).
8. Production of *in vitro* microtuberlet through bioreactor and development of transgenic plant using abiotic stress tolerant gene in potato (*Solanum tuberosum L.*).

#### **Marketing status of tissue culture products including export (if any):**

We already marketed virus-free potato plants to the farmers that produced by tissue culture in different areas of Bangladesh.

#### **Main constraints**

- a. Still we have some shortage of lab equipments.
- b. Lack of greenhouse for maintaining crops round the year.
- c. Lack of transport vehicle.

#### **Future planning**

- a. Development of suitable protocols for applied research specially for potato, orchid, stevia and some other commercial crops to fulfil our national goals for food security.
- b. Development of abiotic stress tolerant major cereal and other crops in Bangladesh.





## Biotechnology Program at BRAC University (BRACU)

Biotechnology programme at BRAC University (BRACU) was launched in Spring Semester 2007 with an aim to develop highly competent biotechnologists to serve in different academic and R&D institutions in public and private sectors at home and abroad. Three years later in Summer 2010 BSc in Biotechnology was launched at BRACU.

So far 41 students have completed their post graduate degree while first batch of biotechnology graduates will

be passing out this year. All BRACU graduates are employed in various industries and universities. Several of our students after completion of their degree have started their research abroad leading to PhD.

The BRACU has also developed good laboratory facilities for plant tissue culture and transformation work. In 2010 Plant Biotechnology laboratory received a grant from Bangladesh Academy of Sciences. Under the BAS-USDA-PALS Endowment fund lab facilities has been establish to do research in the field of molecular biology.



Both at undergraduate and post-graduate levels students have to conduct a research project on problems relevant to Bangladesh. In the field of plant biotechnology, research is going on to establish micropropagation and transformation protocols to produce various transgenic agricultural crops. Also research is going on to isolate and characterize several salt tolerant genes in the plant biotechnology lab of BRACU. So far, micropropagation of both commercially valuable potato varieties and environmental stress tolerant potato varieties have been done. For improvement of peanut and mungbean research is going on micropropagation of these legume crops. A strong concentration has been given in developing salinity stress tolerant transgenic tomato variety under the BAS-USDA Endowment fund. Some of these works are going on in collaboration with other institutes, like, Dept. of Biochemistry and Molecular Biology, DU, Bangladesh Agricultural Research

Institute (BARI), Sher-e-Bangla Agricultural University (SAU).

After completing post graduate degree many students have began their PhD abroad like in Canada, various European countries etc. BRACU also encourage students to get training from abroad. As-a-whole graduates of BRACU are well equipped with theoretical and practical experience to carryout any assignment at home and abroad.

### Agricultural Biotechnology Support Project II (ABSPII)

House 24, Road 7, Sector 4, Uttara Model Town, Dhaka-1230, Bangladesh

Agricultural Biotechnology Support Project II (ABSPII) is funded by the United States Agency for International Development (USAID) and led by Cornell University, Ithaca, New York, USA. It is a collabo-

native research project involving public-private partnerships for agricultural biotechnology to address constraints to food crops production. ABSPII focuses on the safe and effective development and commercialization of bioengineered crops as a complement to traditional and organic agricultural strategies in developing countries. It is a global program operating in East & West Africa, Indonesia, India, Bangladesh and the Philippines.

In South Asia (India and Bangladesh), managed by Sathguru Management Consultants Pvt. Ltd., India, ABSPII helps in capacity building towards research, policy development, licensing and outreach. The coordinating office of ABSPII for Bangladesh is at Uttara, Dhaka.

ABSPII has trained over 150 scientists/policy makers from Bangladesh abroad for short and long term on different aspects of biotechnology, seeds, technology transfer, etc. Initially, ABSPII started working on four projects: Brinjal resistant to shoot and fruit borer, potato resistant to late blight, chickpea resistant to pod borer and drought and salinity tolerant rice. Currently, concentration mainly focuses on the first two projects.

Based on the govt. approval, the partner institution, BARI has already completed two seasons multilocation field trials for Bt brinjal and transgenic potato.

(Source: Souvenir of 6th Intl. PTC&B Conference)

**Alpha Biotech Ltd.**  
**Kacharipara, Rajendrapur, Gazipur**  
**Jatiya Scout Bhaban (9<sup>th</sup> Floor)**  
**70/1, Purana Paltan Line, Kakrail, Dhaka-1000.**  
**Tel: 9336499, Fax: 8315335, Email: aal@gtlbd.com**

**Alpha and Associate Group** is a multidisciplinary private company committed to agricultural development in Bangladesh.

Alpha Biotech Ltd. (ABL) is a sister concern of Alpha group established in 2001 and working on various crops for its development.

Bangladesh is an agrarian country with a huge scope for development of different crops through conventional and biotechnological approaches.

During last three decades, potato production has increased significantly. But, the accumulation of viruses and other pathogens make the crop vulnerable to degeneration and yield loss over time. Moreover, cost of seed tuber is the main obstacle for the majority marginal factors discouraging the cultivation of potato. ABL has developed poor-farmers friendly cultivation with reduced cost of quality healthy seed.

Scientists are producing healthy and quality seeds of leading potato cultivars - Diamant, Cardinal, Granula and Multa through meristem culture. ABL has Memorandum of Understanding (MOU) with Bangladesh Agricultural University (BAU) for setting necessary technical assistance. Two Ph.D. and four MS students of BAU have done their partial research in ABL Lab.

Many private sectors and organizations have already come forward with production of seed and table potato. last year, ABL has cultivated potato in 250 bighas of land at Domar, Nilphamari and produced minituber, breeder, foundation, certified, TLS seeds of potato. ABL's tissue culture laboratory at Rajendrapur, ABL's produced 12,00,000 plantlets to produce disease free and healthy potato seeds. The microplants were tested through ELISA to ascertain that those microplants were disease free. This year the number of plants have been increased these are to be supplied to cultivate potato in 400 bighas of land and for this plan has been initiated to produce 25,00,000 plantlets. At present, the workers of laboratory are subculturing the microplants to regenerate more new plantlets and microtubers of Granula, Diamant, Cardinal, Multa varieties of potato.

It is worth mentioning that the first generation tissue culture materials produced last year in the netted land by ABL was far ahead of the total quantity of that planted by all other private and public sectors in Bangladesh.

It is expected that present innovation will increase the quality seed supply to the growers for upliftment of farm income and alleviation of poverty in Bangladesh.

It is that others hoped cooperation and contribution will make the workers more confident to develop and increase food security in the country.

(Source: Souvenir of 6th Intl. PTC&B Conference)

## SAARC Agro-Biotech

4 Shah Ali Bag, Mirpur 1, Dhaka-1216, Bangladesh

Starting with only 8 (eight) acres of land during 2007, this year we are producing seed potatoes in about 100 (one hundred) acres at Panchagarh. Our three main varieties are Diamante, Cardinal and Granula and some new varieties like Courage, Asterix, Lady Rosetta etc. are in pipe line. We have been supplying seed potatoes to Munshigonj, Comilla, Rangpur, Thakurgaon, Panchagarh, Rajshahi, etc. districts for last several years with very good reputation. Besides, we are also working with Banana, Strawberry, Carnation, Gerbera, etc. plants and fruits.



Net house

This company aims to develop as a seed industry. We have two units: (a) a plant tissue culture laboratory and (b) a seed production unit. Through tissue culture laboratory we can produce about 3-4 lakh of plantlets. Our Laminar Air Flow Cabinets, Autoclaves, Water De-ionizing Plant, Hot Bead Sterilizers, Magnetic Stirrer, Growth Racks, Timers and some other important machineries were manufactured by our another sister concern SAARC Engineering, which cut our laboratory establishment cost for about some folds. Tissue culture laboratory produces and supplies quality plant materials to seed production unit and seed production unit follows every rules and regulations strongly for producing quality seeds. We have a very organized technical person power team with very well and long professional track records with different govt., NGO and private organizations.



Inside test tube

We are planning to go for organic cultivation system soon and to reach this target this year we are in trial production of seed potatoes in about 10 (ten) acres of land.



Harvested

We are committed to produce and supply quality seeds and plant materials. If we get financial support and training assistance from any local or foreign organizations we are hopeful that we can play a vital role to develop country's agricultural economy.

(Source: Souvenir of 6th Intl. PTC&B Conference)

**Plant Biotechnology Laboratory**  
**Biotechnology and Genetic**  
**Engineering Discipline**  
**Khulna University, Khulna-9208,**  
**Bangladesh**  
www.ku.ac.bd

Biotechnology and Genetic Engineering Discipline (Formerly “Biotechnology Discipline”) launched four years undergraduate program in June, 1995 aiming at offering B. Sc. in Biotechnology degree, which was first of its kind in Bangladesh. From August 2003, the Discipline has started offering 4-year B.Sc. in Biotechnology and Genetic Engineering degree. The Discipline started one year Master degree program from 2000 and 18 months duration (consists of three terms) M.S. degree in Biotechnology and Genetic Engineering from 2007-2008 session. The nature of this Discipline is multidisciplinary i.e., it covers various branches of Biotechnology viz. Agricultural /Plant Biotechnology, Animal Biotechnology, Medical and Pharmaceutical Biotechnology, Environmental Biotechnology, Fermentation/Bioprocess Technology, Enzyme Technology, Food Biotechnology etc. Plant Biotechnology Laboratory was established during 1989-1999 with the financial support of Khulna University.

In undergraduate course-curricula, it has educational and research program on Plant Biotechnology in addition to other basic and applied courses. Plant Biotechnology related courses are being taught in the undergraduate program include: Evolutionary and Functional Botany, Plant Physiology, Plant Breeding, Plant Pathology, Plant Cell and Tissue Culture, Molecular Genetics, Genomics and Proteomics, Nitrogen Fixation and Biofertilizer, Plant Biotechnology and Genetic Engineering etc. Moreover, basic courses like Principles of Genetics, Cytology, Microbiology, Biochemistry, Molecular Biology etc. are also being taught. At M.S. level, several courses like (i) Advanced Plant Biotechnology, (ii) Biosafety and Bioethics, Biotechnology in Hybrid Seed Production and (iv) Molecular and Resistance Plant Breeding courses are related to Plant Biotechnology are also being taught.

Plant Biotechnology Laboratory is equipped with modern instruments required to conduct undergraduate practical classes and researches of undergraduate and graduate students. The list of major equipments available in the laboratory is given below:

(1) Precision electronic digital balance (Readability: 0.0001g)(2) Digital top pan balance (Readability: 0.1g) (3)Autoclave (4) Split type microprocessor system air cooler (5) Microflow laminar hood (6) Fristem water still (7) De-ionized water plant (8) pH meter (9) Microwave oven ((10) Microprocessor system orbital shaking incubator (11)Shaking water bath (12)Digital water bath (13) Table top microcentrifuge (Capacity: 18000rpm) (14)Magnetic stirrer (15) Camera fitted binocular microscope (16)Digital camera (Sony Cybershot) (17) Refrigerator (18) Freezer (-20°C) (19) Digital timer (20) Lux meter (21) Gel Doc

Beside the above equipments, “Plant Biotechnology Laboratory” was modernized with the financial support of Ministry of Science & ICT, Government of Bangladesh during 2003-2005. The following equipments were procured for the purpose of conducting advanced research in the field of molecular markers (RAPD, AFLP) assisted breeding as well as variety fingerprinting. (1) PCR machine (2) Digital micropipettes of different capacities (3) Electrophoresis apparatus (5) Ice Flake maker /machine (6) Liquid nitrogen container (7) Vortex machine etc. Necessary molecular biology grade chemicals, enzymes and primers were also procured from the above fund to conduct research on molecular markers and *Agrobacterium* mediated transformation.

**List of M.S. thesis/research conducted at Plant Biotechnology laboratory**

1. *In Vitro* Propagation of Banana (*Musa sp.*) cv. Baro Kulpak- An Indigenous Variety of Southern Bangladesh (Student No. MS- 070701, Session: 2006-2007; Supervisor: Prof. Md. Raihan Ali; Co-supervisor: Prof. Dr. S.M.M. Rahman)

**List of M.S. thesis/research conducted at Plant Biotechnology laboratory:**

1. Response to *In Vitro* Callus Induction and Shoot Tip Culture of Pumpkin (*Cucurbita maxima*)



- Duch.) (Student ID: 060736; session: 2008-2009; Supervisor: Prof. Md. Raihan Ali; )
2. Morphological and Molecular Characterization of Selected Pumpkin (*Cucurbita maxima*) Varieties in Khulna Region Using RAPD Markers.(Student ID:060733; Session: 2008-2009; Supervisor: Dr. Sayda Reahana; Co-supervisor: Prof. Dr. S.M.M. Rahman)
  3. Morphological and molecular Characterization of Selected Lemon (*Citrus aurantifolia*) Varieties with RAPD Markers (Student ID:060703; Session: 2008-2009; Supervisor: Prof. Dr. S.M.M. Rahman)
  4. Analysis of Morphological and RAPD Marker Based Genetic Diversity of Selected Promising Brinjal Varieties. (Student ID:060714; Session: 2008-2009; Supervisor: Prof. Dr. S.M.M. Rahman)
  5. Effect of 2,4-D and NAA on Callus Induction in Mature Seed Derived Embryo of Aromatic and Jhum Rice (*Oryza sativa* L.) (Student ID: 060715; session: 2008-2009; Supervisor: Prof. Md. Raihan Ali; Co-supervisor: Dr. Sayda Reahana)
  6. Lack of facilities for repair of instruments or private service centre at Khulna district
  7. Lack of fund for short term and long term training at ICGEB or any other advanced Lab/University for personnel working at this lab.
  8. Shortage of supporting staff like lab. attendant, nursery man or field attendant to carryout field experiment /green house/net house management

#### Future plan

1. *Agrobacterium* mediated transformation of various economically important crops.
2. Development of protocol for *in vitro* regeneration of local and improved varieties of chickpea, mungbean and lentil.
3. Development of somaclonal variants of pulse crops.
4. Development of anther culture protocol of local and improved varieties of rice.
5. Development of strawberry variety through somaclonal variation.
6. Development of protocol for micropropagation of export quality potato var. lady rosetta
7. Cryopreservation of economically important and endangered plant species.
8. Insect resistant variety development through the use of systemic RNAi technology.

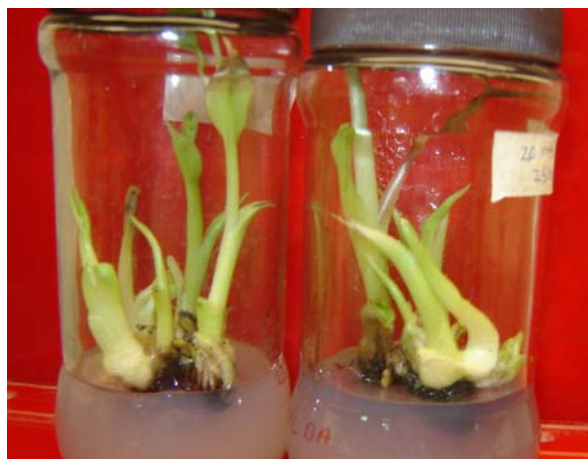
#### List of academic staff involved in research at Plant Biotechnology Laboratory

1. Professor Md. Raihan Ali (Lab in-Charge)
2. Professor Dr. S.M. Mahbubur Rahman
3. S.M. Abdul Awal, Assistant Professor
4. Dr. Sayda Rehana, Assistant Professor
5. Ahsan Habib, Assistant Professor

#### Main constraints:

We are facing following problems to carryout research at our laboratory. These are as follows:

1. Insufficient fund for recurrent cost of the lab.
2. Shortage of consumable items like molecular biology grade high quality chemicals.
3. Lack of -86°C freezer to keep enzymes, primers and other samples for long time.
4. Lack of UV-VIS spectrophotometer, ELISA reader and its chemicals, Inverted microscope .
5. Lack of green house, net house and hardening room for in vitro regenerated plant materials.



*In vitro* regeneration of local “Baro kulpak” variety of banana

(Source: Souvenir of 6th Intl. PTC&B Conference)



**Plant Breeding and Gene  
Engineering Laboratory  
Department of Botany, Rajshahi  
University, Rajshahi, Bangladesh**

Plant biotechnological research activities of the Department have now been expanded to a considerable extent. Trained manpower, equipments, chemicals and laboratory setup are now available for conducting advanced research in the field of *in vitro* crop improvement, micropropagation and pathogen-free seed and seedlings production. Initial work was conducted to standardize plant regeneration protocols for different fruits and timber plants that resulted in the establishment of micropropagation protocols for many of them. The programme also included mass propagation of some endangered and eco-friendly plant species of medicinal and ethnobotanical importance.

The present work emphasizes on *in vitro* conservation of plant genetic resources under local biodiversity and manipulation and regeneration of cereal crops through *in vitro* techniques. Somatic embryogenesis and plant regeneration have been successfully established in some vegetable crops and native fruit plants. Protocol for meristem culture to produce virus-free potato seed has been standardized and the technology are being transferring to the private commercial tissue culture laboratories established in Rajshahi metropolis and elsewhere in the country. The plant breeding and gene engineering laboratory significantly contributing agri-industry development in Bangladesh and producing a handsome amount of pathogen free seeds and saplings.

At present a reasonable number of M.Sc., M. Phil. and Ph.D. students have been involved in plant tissue culture and genetic engineering research. Thirty Ph.D. and twenty five M. Phil. students have already been awarded with the degrees and a pretty numbers are preparing their theses in this field.

**Running Research Projects:**

**Strawberry**

- Development of strawberry varieties suitable for Bangladesh through somaclonal variation

- Micropropagation and field evaluation of newly developed strawberry varieties at commercial scale

**Potato**

- Production of virus free seed potato and commercial evaluation
- Nutritional improvement of potato by genetic transformation
- Production of somatic hybrids between tetraploid potato (CV Desiree) and wild diploid potato (*S. chacoense*)
- DNA finger printing for the identification of potato varieties grown in Bangladesh.
- Potato microtuberization and profiling key factors effective for nuclear seed production
- ELISA test for virus detection and disease indexing

**Rice**

- Parental evaluation of rice varieties for the production of hybrid seeds
- Genetic transformation of rice with PRP gene(s)
- Seed priming for induction of epigenetic changes in rice for early maturity and high yield

***Arabidopsis thaliana***

- Genetic transformation and chromosomal mapping of transgene in *Arabidopsis thaliana* L.

**Lettuce**

- Production of Salmon Calcitonin protein in Lettuce
- Production of Human Calcitonin related peptides blood pressure depressant.
- Transgenic expression of *PAT* (Pesticidal gene) in lettuce.

**Water chestnut**

- Mass production of seedlings of water chestnut with hormonal treatment
- Genetic diversity study in water chestnut with molecular markers (isozyme, RAPD, Matk and RbcL Sequencing)

**Teasel gourd**

- Micropropagation and induction of triploid bisexual flower and crossing among different ploidy levels and production of female seed and continuous fruiting

**Tomato**

- Diallel and single cross analysis in tomato
- Production of hybrid tomatoes
- QLT analysis with RFLP & PAPD in tomato

**Maize**

- Parental evaluation and production of hybrid maize

- Genetic transformation in maize with PRP gene(s)

#### **Melon**

- Micropropagation of Japanese melon

#### **Wheat**

- Evaluation of wheat genotype for drought and salt tolerance

#### **Sugarcane**

- Transgenic sugarcane with stem borers and red rot resistance genes

#### **Flowers**

- Micropropagation of orchids and inca marigold

#### **Banana**

- Mass production of banana plantlets through micropropagation

#### **Pineapple**

- Mass production of pineapple plantlets through micropropagation

#### **Medicinal plants**

- Micropropagation and conservation of threatened medicinal plants

#### **Fruits**

- Establishment of *in vitro* protocols for large-scale propagation of horticultural plants



Harvested strawberry fruits (RABI-3)



Potato field (Foundation seeds)

Future research programme of the laboratory includes the improvement of crop plants through *in vitro* mutation and *Agrobacterium* mediated gene transfer techniques. About 250 research papers have been resulted based on the findings of plant breeding, tissue culture and genetic engineering oriented research problems and published in highly reputed national and international journals, edited books and international conferences proceedings.

#### **Members Presently Engaged in Research**

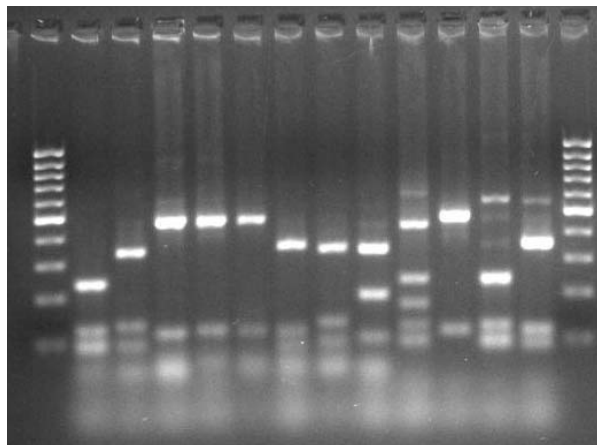
Professor A.K. M. Rafiul Islam  
 Professor M. Monzur Hossin  
 Mr. Rezaul Karim, Lecturer  
 Mr. F.M.A. Haydar  
 M. Nasiruddin

(Source: Souvenir of 6th Intl. PTC&B Conference)

### **Tissue Culture Laboratory Department of Botany, University of Chittagong Chittagong-4331 Bangladesh**

Under the leadership of Professor S. K. Bhadra an extensive research programme on Biotechnological research was first initiated in the Botany Department in mid 1980's. Although the programme was mainly centered around genetic improvement of mung bean with the use of *in vitro* techniques such as embryo culture, cell and tissue culture, later on in 1990's the programme was extended to a wide range including detailed study of different biotechnological aspects such as micropropagation, *Agrobacterium* mediated genetic transformation, development of microtuber and virus free seedling production techniques in potato, tissue

culture based conservation of indigenous, rare and endangered orchids and medicinal plants. So far efficient and reproducible protocols for rapid propagation and conservation of 50 different indigenous orchid species, 20 medicinal plants and some economically important crop plants have been developed.



RFLP Profile of Fungal DNA



Somatic embryogenesis in *Dendrobium* orchid

The research team is now composed of three teachers; Professor S. K. Bhadra, Md. Mahbubur Rahman and Dr. M. Musharof Hossain with their research students and fellows. The research programme has recently been further extended with inclusion of a project on conventional and molecular characterization of mycorrhizal fungi and their use in biological hardening of the tissue culture raised plants. The biotechnology

laboratory is now well equipped with automated temperature controlled culture room, high speed liquid filter sterilization system, different kinds of high resolution microscopes, molecular analytical facilities, hardening laboratory established with the financial assistance of different native and international agencies.



*In vitro* germination of orchid seeds  
(Source: Souvenir of 6th Intl. PTC&B Conference)

### **BRAC Plant Biotechnology Laboratory Joydebpur, Gazipur, Bangladesh**

Today, BRAC (Building Resources Across Communities) is the largest non governmental development organization in the developing world. BRAC has given top priority on agricultural development because of its stronger role in poverty alleviation and its potential for generating income employment for the poor and marginal farmers. It has significant contribution in the national production and also in improving nutritional status. BRAC believes that the demand for high quality seed production and distribution will help meeting the present demand of Bangladesh. With this objective in mind we have been using Biotechnology as a frontier technology which has a great potential to provide new solution to the problems.

The shortage of high quality seeds/seedlings and tubers is one of the major constraints in increasing the productivity of agriculture in Bangladesh. Only 8 - 10% of the seed available to the farmers is produced under controlled conditions to ensure high quality. The

majority of seeds are produced by indigenous methods without recourse to technology to ensure high yield and disease free varieties. A large part of the demand for the high yielding varieties met from imports and thus a vast market exists for cheap, high quality, locally adaptable seeds/seedlings and tubers. These challenges of increased food production can be achieved through improvement of presently available technologies and intensification of crop production in marginal lands.

BRAC's objective is to claim a substantial portion of this market helping farmers to increase their productivity and income. Therefore, a project has been taken up in view of the need for healthy stocks and disease free planting materials for the farmers, plant propagators and seed producers of Bangladesh. BRAC started its activity with a set up of a small scale tissue culture laboratory in June 1997 to undertake the micropropagation of banana, potato plantlets/micro tubers and ornamental plants. Two years of the experimentation and observation have shown that potato, ornamental plants and banana tissue culture are quite promising. Based on the success of the TC lab, and the continued demand for high quality disease free tubers and plantlets, BRAC has set up a new Biotechnology laboratory at Gazipur, 35 kilometers from Dhaka, where quite a good number of govt. agricultural research institutes are there. These government institutes offer help to BRAC lab in many ways.

BRAC's biotechnology laboratory is well equipped with modern tissue culture facilities. Equipment, machineries and chemicals are available for the establishment of meristem culture and micropropagation. In addition to media preparation (600 sq. ft.), three growth rooms (each 300 sq. ft.), washing room (500 sq. ft.), transfer room (400 sq. ft.) media storage room (300 sq. ft.) , R & D (300 sq. ft.) sorting, counting and checking room (300 sq. ft.) and totally are 7000 sq. ft. The tissue culture production facilities comprise 3 US class growth rooms each with a capacity for accommodating 25,000 culture bottles at a time, 10 Laminar air flows, a medium kitchen facility with a capacity of preparing 4000 bottles ( 60-70 liters) of medium/day.

The laboratory uses most modern instruments including virus testing kit (ELISA). It is headed by highly qualified and experienced staff for generating over two million plantlets of international standard per year. In addition four green houses each with an area 3000 (30 × 100 ft.) sq. ft have been built utilizing most advanced equipment such as automatic shade system, fogging, blackout and irrigation system and movable benches and in addition 12000 sq. ft. of 50 and 75% shading net houses for hardening of tissue culture raised plantlets.

Among professional staff, there are one Ph. D. (plant tissue culture), four Masters (plant tissue culture), 30 laboratory assistants to maintain laboratory and 25 agriculture graduates to maintain the field.

BRAC biotechnology is the largest unit in Bangladesh and has successfully adopted TC technique for mass multiplication covering a wide range of plant species such as potato, banana, starfruit, jackfruit, olive, sweet karamcha, wood apple, lemon, strawberry, papaya, orchids, carnation, gerbera, anthurium, agave, ornamental taro, neem, stevia etc. selected according to the priority crops in the country.

BRAC biotechnology laboratory started the production of potato plantlets since its inception (1998) to ensure quality seed potatoes for growers assisting increased potato production as well as saving foreign currency for importing seed potato from overseas. The laboratory produces plantlets ( $G_0$ ) which are grown in two seasons in the net houses for mini tuber ( $G_1$  yield 3.5 - 4MT/acre) and pre-foundation seed ( $G_2$  yield 6-7 MT/acre) production. Pre-foundation seeds are multiplied in another two seasons in the open field for foundation ( $G_3$  yield 9-10 MT/acre) and certified seed ( $G_4$  yield certified seed 8-9MT, total production 13-14 MT/acre) .The certified seeds are distributed to the growers for table potato production. It is to be noted that plantlets (10000 - 12000) yielded 400-450 MT certified seeds after four generations. During the last winter, BRAC's biotech lab. produced 6000 tons of disease free potato seeds in the field using BRAC certified seed potato. It is reported that average potato yield is 35 to 45 tons/ha whereas the yield is only 1/3 (11 to 12 tons/ha) using seed from local sources. BRAC's biotech lab. has currently fixed up the target of seed production of five varieties: Diamant, Asterix,



Cardinal, Felsina and Granola in the winter season (2007-2008) to 10,000 tons. BRAC's seed potato multiplication in the field is being assisted and monitored regularly by internal and external experts to maintain the seed quality.



Greenhouse



Potato



Strawberry



Natural Light



Transfer Room



Natural Light Room



Tissue Culture Lab



Through tissue culture ornamentals such as imported anthurium, double and single gerbera, carnation and orchids, the four popular flowers are being multiplied. It is to be noted that it has already captured domestic markets stealing the hearts of the flower lovers. Also included in the program multiplying *en masse* fancy cacti, grafting various colored scions onto the green mother stocks. Through tissue culture superior varieties of different fruit plants are also being multiplied superior. During the current year BRAC's bio-tech lab. has a plan to produce 3 lacks of fruit plantlets, including banana, jackfruit, strawberry, sweet karamcha, and lemon.

BRAC tissue culture laboratory has developed a protocol to micropropagate an extremely useful medicinal plant - stevia in recent years. Stevia contains compounds called steviosides that are 250-300 times sweeter than sugar. Because the body does not metabolise steviosides they do not contribute any caloric value to food. Diabetic and others, unable to tolerate sugar, can take stevia with immunity. Dieters love stevia because they can continue to enjoy sweets without counting calories. Stevia can be used as sweetening agent, it has also been found to have hypertensive, hypoglycemic and bactericidal properties. High demand of stevia plant is an indication of success of the BRAC tissue culture lab. It represents a new opportunity for researchers and farmers too. Because farmer can earn Taka 3 - 4 lacks/acre as net profit per year.

#### **Low cost technologies for the commercial micro-propagation of vegetatively propagated plants.**

Dr. F. J. Zapata Arias, Advisor to BRAC's plant Biotechnology group, began to work on April 2009 on the development of low cost technologies for the commercial micropropagation of vegetatively propagated plants.

*In vitro* cultures are usually maintained under artificial lighting at controlled light and temperature regimes. It is costly to equip growth rooms with fluorescent tubes and temperature control units that they entail high running costs and maintenance. Moreover, the operation may be frequently disrupted by power cuts or lack of spare parts and timely service.

We are proposing the total replacement of these practices by using natural light to reduce the cost of production of micropropagules.

We aim to achieve this without compromising quality or quantity and are using potatoes, bananas and orchids in our experiments.

If successful this technology could also be use by farmers in tropical and sub-tropical agro-ecological regions similar to Bangladesh and will contribute to the efforts in achieving food security by producing cheaper clones of potato.

Keeping the national priorities in mind, BRAC biotech lab. continues research and developmental program in the area of plant biotechnology. With the basic infrastructure we have for biotechnology work, we may initiate genetic engineering work in near future to develop disease and pest resistant varieties of potato, chickpea, papaya, egg plant, rice and maize etc.

#### **Staff members**

Dr. M.A. Razzaque Shah, Tissue Culture Spacialist

Maksuda Khatun, Program Organizer

Shahana Akther, Program Organizer

S.M. Asadul Haque, Program Organizer

S.M. Shahin, Program Organizer

Suvra Majumder, Program Organizer

(Source: Souvenir of 6th Intl. PTC&B Conference)

### **Square Agro Development and Processing Limited AgriBiotech Division**

Tissue culture is a newly emerging, highly rewarding technology with large potential application in crop improvements which is highly appropriate for developing countries like Bangladesh. Conventional plant breeding techniques have made considerable progress in the development of improved varieties but they have not been able to keep pace with the increasing food demand of the country. Annual population increased in the country outpaced annual increases in food production. The present population of the country is about 160 million and at the present growth rate it is expected to touch 180 million by the year 2011. The situation necessitated to produce more

food for the increased population of the country which is not possible by the existing traditional crop improvement program. Tissue culture, when integrated with conventional crop improvement techniques will be more efficient environmentally compatible and ultimately cost effective utilization of resources for improved agricultural productivity. So, this is the high time to introduce biotechnology to speed up the crop improvement processes.

**Plant issue culture** means a collection of techniques by which isolated cells or tissue from either root stem leaf or any parts of the plant body, if provided with suitable condition would develop into plantlets in test tube. Then in the control environment of a laboratory growth room, the growth of this culture is directed towards the production of a large number of true to type shoots which can either be rooted *in vitro* in sterile medium mixture according to the specified requirements of the customers.

**SQUARE GROUP**, a leading business organization of Bangladesh, has set up a plant tissue culture laboratory under Square Agro Development and Processing Ltd. in 2003 to develop agriculture & agro processing sector in the country and is committed to bring improved quality planting materials and year round production through AgriBiotech. By its fifth year AgriBiotech division turned into a profit making organization.

#### **Aim**

- To produce disease free, high yielding seeds and seedlings such as potato banana, strawberry, stevia, ginger, turmeric, and flowers including varieties of orchids.
- To create a supply chain and distribute the above product to the root level farmers and nurserymen.
- To solve the insufficiency of quality seeds and seedling in the market.
- To reduce the dependence on imported seeds.
- To increase productivity and production that will help poverty alleviating activities by improvement in income and employment.

#### **Milestones AgriBiotech Division**

- 2002 : Feasibility test and budget approval
- 2003 : Start laboratory renovation
- 2004 : Laboratory operation start  
Potato plantlet sowing for breeder seed production
- 2005 : Start banana sapling sale  
Continue seed potato production (breeder and foundation class)
- 2006 : Continue banana sapling sale  
Continue seed potato production (breeder, foundation and certified class)
- 2007 : Continue banana sapling sale  
Continue seed potato production (breeder, foundation and certified class)  
Start seed potato sale  
Start orchid sapling sale
- 2008 : Continue banana sapling sale  
Continue seed potato production (breeder, foundation and certified class)  
Continue seed potato sale  
Start orchid sapling sale
- 2009 : Continue banana sapling sale  
Continue seed potato production (breeder, foundation and certified class)  
Continue seed potato sale  
Continue orchid sapling sale  
Start strawberry sapling sale

#### **Present Activities**

Production of breeder seed from potato plantlet in net house has been started since 2004. Certified seed potato has been being produced and being supplied to the root level farmers since 2007. Besides this we are engaged in production and marketing of banana saplings, orchid pot plant, chrysanthemum, carnation, eustoma, ginger, turmeric etc from 2007.

#### **Laboratory**

Well equipped around 4000 sq-ft tissue culture laboratory is situated in Dhaka at Uttara, a convenient location having careful designing and planting facilities

to produce 1 million seedlings per year which includes growth room, media preparation room, aseptic transfer area, sterilization room, laboratory office room, glass ware washing room, store etc.

#### **Shade / poly shade house/nursery area**

Orchid - 1000 sq ft

Banana - 3000 sq ft

Others - 500 sq ft.

#### **Multiplication / seed production Field**

At about 125 acres fields are cultivated at present which will be increased to 200 acre within 2012 for potato seed production. Breeder seed from *in vitro* plantlet has been started since 2004.

#### **Manpower**

About 30 numbers of permanent staffs are working at AgriBiotech division which includes researcher, management personnel, technician, skilled labor and field worker manpower.

#### **Future Plan**

- Research and development in the field of biotechnology to protect endangered medicinal plant.
- To bring some new plant such as rattan, bamboo, cane, rock melon, etc. which are almost extinct in the country.
- We are going to engage in production of processed potato variety and will be distributed to the market very soon.

(Source: Souvenir of 6th Intl. PTC&B Conference)

**AHZ Biotech Ltd.  
House No. 241, Road No.3  
Padma Housing Estate, Bhadra, Rajshahi,  
Bangladesh**

AHZ Biotech Ltd. is a private Organization. Improved, disease free and high yielding varieties of different seeds like potato are being produced. AHZ Biotech Ltd.

follows advanced tissue culture technology that is comparable to any other developed country in the world for production of seed potato. The methods are followed for production of disease free potato clones through meristem culture, their rapid multiplication through micropropagation, production of minituber and eventual production of pre-foundation, foundation and certified seed.

#### **Different Steps of Seed Production by Tissue Culture.**

##### **First step: Meristem Culture.**

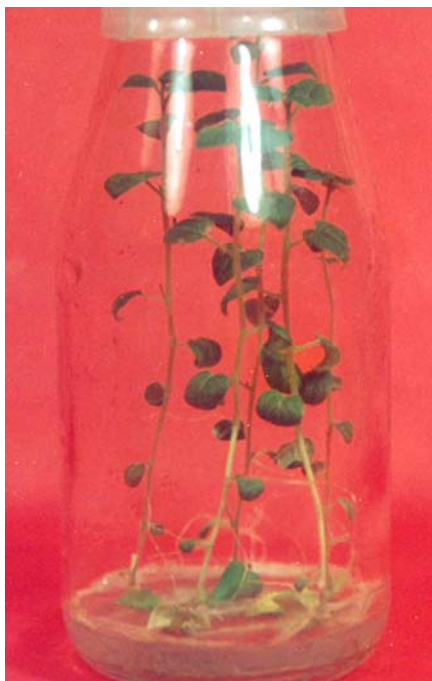
*In vitro* propagation offers excellent technique for rapid multiplication of potato plants. Meristem culture for production of disease free clones and their subsequent micro propagation are carried out. The active growing point of the plant shoot is the meristem. The dome of the shoot meristem contains truly meristematic cell and is surrounded by leaf primordia. The objective of this procedure is to produce a rooted microplant which is free from systemic virus, fungi and bacteria.



Healthy and apparently disease free buds when they developed into mini shoots, are selected, collected and brought into laboratory. Meristems are dissected aseptically under a binocular dissecting microscope. The operations are carried out at a laminar air flow sterile bench. The meristems are treated with disinfecting chemicals, washed with sterile distilled water and the meristem dome is placed in a test tube containing liquid M.S. media. In later stages (both at laboratory and field) ELISA tests and field test are done for confirmation about the presence of virus.

### Second step: Plantlet culture

The test tube containing the meristem dome is placed in laboratory called growth chamber having required temperature, humidity, and light. After 12 - 15 weeks of incubation, the plantlet is grown and attained a height of 4 - 5 cm. The plants are removed from the test tube carefully and cut into single nodal segment and plant in glass jar containing culture media. The culture can be sub-cultured for multiplication as required.



### Third step: Improved seed potato production.

When the plantlets are 5 -7 cm height and have developed a good root system, they are ready for transferring to soil beds. Before transfer, the closures of the flasks containing plantlets are removed and kept in growth chamber in natural condition for 2-3 days for acclimatization. The beds are prepared in a net house with sand and organic compost and are thoroughly disinfected by formalin to avoid bacterial infection and virus carrying vectors. The plantlets are transferred into the bed carefully followed by live irrigation. Necessary fertilizers, biocides are applied. Intercultural operations are also done as and when required. In the net house the seed potatoes are produced by the plantlets which is called breeder seed, subsequently, pre-foundation in net house, foundation and certified seeds are produced in open field with special care. In all the cases, strong

measures are done for regular spray to control disease and insects. Roguing is also done as and when required, necessary fertilizer applications and intercultural operations are also done for good growth.

During growth period, potato specialists from research institute, BADC and DAE visit the seed plots. Moreover, field officers of seed certifying agency, visit the seed plots. After getting recommendation from seed specialists and field officer of the seed certifying agency the seed plots are ready to harvest. Within 75-85 days of plantation, haulm pulling of the potato plants are done. After 10-12 days of the haulm pulling, the seeds are harvested carefully and are dried under shed for 2-3 days. After curing, the seeds are assorted, graded, bagged and stored in the special seeds chamber of Biotech's own cold storage at Mokamtala, Bogra. For good preserving of seed potatoes it requires separate chamber having 36<sup>0</sup>-38<sup>0</sup> F temperature, 90-91 % humidity and regular supply of oxygen and drain out of CO<sub>2</sub> and frequent rearrangement of bags. These facilities are available in the cold storage.



Booking for selling the seed started from 15th February, from October the preserved seed are supplied among the Govt., organisations, private agencies, and the progressive potato farmers at the fixed rate decided by the company.

**Generation and category of seed potato produced by AHZ Biotech Ltd:**

Generation	Category
Zero	Plantlet
First	Breeder seed
Second	Pre-foundation
Third	Foundation
Fourth	Certified.

Seed potatoes which produced by the AHZ Biotech Ltd are improved and cleaned. The generation -of the seeds is less even compared to imported seed potatoes. If the different agronomical practices such as timely plantation, application of fertilizers, biocides, irrigation and intercultural operations are optimized properly the tissue culture derived seed is able to produce 30 - 40 t/ha of potato in Bangladesh.

**1. Year of establishment: July 2001**

**2. Objectives**

- Disease free seed production of mainly potato and also some other crops through tissue culture.
- To increase socio-economic condition of the farmers ultimately to increase the national economy.
- To create job opportunity of different cadres.
- To select suitable cultivars among the imported potato varieties for export and industrial use.
- To conduct field experiment, demonstration, field day farmers training etc. to motivate the farmers for the use of the improved seed.

**3. Existing capacity**

One tissue culture laboratory at Rajshahi having 2 laminar airflow, 2 autoclave, 2 microscope, AC, electric balance, electric stirrer, required chemical, lab room, growth chamber etc.

**4. Scientific manpower**

8 Scientists, 8 permanent staff and 36 seasonal staff out of that one Doctorate in tissue culture, one M.Sc in Botany (specialized in tissue culture) four M.Sc (Ag.) (specialized in Biotechnology) two agriculture graduate specialization in seed production.

**5. Marketing status of tissue culture products including export,** (if any), tissue culture seed marketed among Govt. non-govt. organizations and farmers of the most of the area of Bangladesh.

**6. Main constraints**

- Non availability of permanent land for seed production.
- Lack of Agril-implements
- Shortage of lab equipments
- Lack of transport vehicle and lack of own building for laboratory.

**7. Future planning**

- Expansion of seed production to partially meet up the national requirement
- Extension of production for industrial use potato through the use of tissue culture seed and establishment of potato based food industries (like potato chips)
- To increase production of stevia and other saplings through tissue culture.

**8. Need**

- Financial support for establishment of own lab. building and expansion of laboratory and procurement of inputs.
- Agril. equipments and expansion of lab. facilities.
- Transport vehicles.
- Permanent land for seed production.
- Foreign training/visit for manpower development.

(Source: Souvenir of 6th Intl. PTC&B Conference)



## Aman Plant Tissue Culture Lab.

(A Concern of Aman Group)

Narikel Baria, Boalia, Rajshahi

### Background

AMAN Plant Tissue Culture (APTCL) has emerged as a private sector independent Research lab in the Northern belt of Bangladesh mostly covering the Rajshahi and adjacent districts with a view to meet the increasing demand for quality seeds, technical support and research support among the huge number of poor and moderate farmers of this highly potential potato growing area of the country. Potato farmers of this region are growing potato in a comparative disadvantageous situation without having any sophisticated cultivation support like quality seeds, technical advice, fertilizer supply, pest and disease control etc. Despite that the dominating share in the National production chart is the contribution of farmers of this area. Farmers were getting 12 to 15 MT productions per Hectare Land by cultivating potato in conventional way without knowing that there are rooms for even better yield by adopting some low cost technology. AMAN Plant Tissue Culture showed the farmer how to improve production and quality of potato.

Now a day potato is one of the main stable foods for us. Out of the increasing demand from farmer AMAN started realizing need for finding alternative sources of quality potato seeds since government support is severely adequate. Under the above circumstances, AMAN Group Management thought out of establishing Tissue culture lab finally with the active support from some scientists of Rajshahi University it has been possible to establish a fully equipped Tissue Culture Lab from complete AMAN's own funding.



Chief Scientific Officer Dr. Nurun Nahar in Lab Action

Since inception to first one year APTC had been involved in the advisory services to farmer and internal lab development works which has later concentrated on developments of plantlets and other research activities. A sketch on production is given below:

### Year wise Production of ATCL

Year	Plantlets	Production			
		Breeder Seeds (MT)	Pre-foundation Seeds (MT)	Foundation Seeds (MT)	Certified Seeds (MT)
2004	20000	0	0	0	0
2005	40000	2.6	0	0	0
2006	40000	5.5	42.5	0	0
2007	75000	7.5	90.00	374.00	0
2008	100000	10	110	800	1200

It should be mentioned here that AMAN from its own interest was involved in potato cultivation since long in its own land using imported seeds from Holland. Since then AMAN started selling some seeds to a limited number of potato farmers from the imported seed by which farmers known AMAN as a local source of quality potato seed. But it was badly experienced that qualities of imported seeds were not found good on several occasions. Even timely availability of imported seeds was not possible which again compelled AMAN to think otherwise. Meanwhile over a period of one decade potato business under AMAN umbrella expanded tremendously by which a good number of people were involved in growing potato. Considering the profitability and seasonal support to farmers AMAN also extended storage facilities of their potato through establishing three (03) large scale cold storages that enticed farmer to massively involve in growing potato.

APTCL is fully equipped lab where a group of technically highly skilled scientists, researchers, field

Officers, lab technicians working who are constantly busy developing and producing quality seeds. Currently Dr. Nurun Nahar who is specialized on Plant Tissue Culture is leading the technical team.

The coming year 2009 we will be able to provide 2500 M. Tons seeds to the farmers. Apart from research activities the APTCL is also involved in some other activities like Demonstration program, Training, Logistics support like fertilizer, seeds, technical support, transport and packing materials etc, and last but not the least Financial support like seasonal credit.



A growth chamber in APTC



APTCL is currently having the strengths like Well-structured small research lab 2 storied building, Moderate furnished Lab, Reasonable quantity of lab equipments, Adequate adjacent demonstration field (21 acres), Available organic and inorganic fertilizer (Being AMAN an Importer of fertilizer), Skilled Technical staff and field worker, Some Cultivation equipment, Own transport facilities, Moderate financial support, Cold storage facility (AMAN has 3 cold storages of its own), Ensured marketing outlet, Own and leased land for potato Cultivation Target beneficiaries of APTCL

are existing poor table and seed potato growers, Contact farmers, interested peoples in table and seed potato cultivation and poor, young, educated but enthusiastic men and women.

Our seeds are disease free very high yielding. Some pictures are of our seed potato given along side.

Despite having all the above, APTCL being a private sector initiative, we have still some limitations like Inadequate expatriate services, technical manpower, insufficient sophisticated lab equipments, infrastructure facility, training materials, lack of sufficient fund, lack of technical and financial collaborator and last but not the least lack of govt. support because of which we could not achieve the project target in anticipated pace.

We would welcome technical collaborator, financial support, expatriate support, training support and massive extension support.

AMAN despite having lot of limitation is trying to support poor farmer with its little endeavors which we believe in the near future with the support of outside collaboration will be minimized and a vast number of poor potato farmers will be benefited through receiving quality seed from APTCL and hope to contribute positively in the national economy.

(Source: Souvenir of 6th Intl. PTC&B Conference)

### **Department of Biotechnology and Genetic Engineering, Jahangirnagar University, Savar, Dhaka**

Cell Genetics, Genetic Engineering and Plant Biotechnology Lab

#### **Major objectives:**

- Efficient plant regeneration through somatic embryogenesis in local rice cultivars
- *In vitro* manipulations, micropropagation and stress modeling in fodder legumes
- Development of stress tolerant crops
- Micropropagation & secondary metabolites studies of endangered medicinal plants

- Pilot scale production of cells and adventitious roots through bioreactor
- Rice, Legumes, Medicinal Plants

Basic plant tissue culture facilities with controlled growth chamber, small bioreactors, cell suspension facilities.

**Human Resources:**

MPhil student: 1  
MS student: 2

**Collaborator:**

Local: 3  
Foreign: 4

- Assessment of grain qualities in local rice cultivars
- Optimization of fast and efficient somatic embryogenesis system for local aromatic rice cultivars
- Micropagation of different medicinal plants

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