



## International Conference on Biotechnology

*on application of Biotechnology  
in addressing the development needs of Bangladesh*

## Souvenir

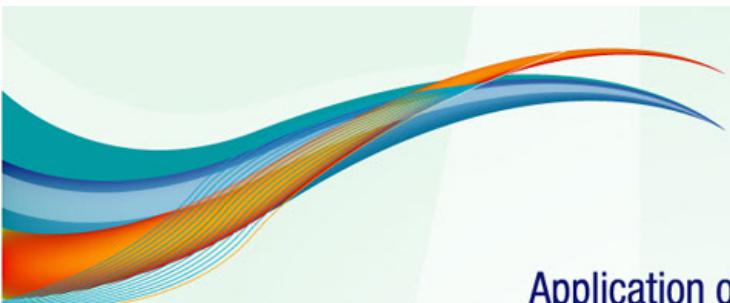
- Programme
- Speakers
- Abstracts

25-26 May, 2013  
The Westin Dhaka, Dhaka



**CARES**

Committee of Action for Research, Extension and Services



# Application of Biotechnology in addressing the development needs of Bangladesh



in collaboration with



&

*American Society of Bangladesh-affiliated Microbiologists*

---

Platinum partner:



Print media partner:



Electronic media partner:



## A brief introduction of CARES and its first International Conference

CARES has been born from an idea that there is a need to establish in Bangladesh a common platform where in the best among our farmers, the scientists and the industrial entrepreneurs will evolve a common course of action. In pursuit of the idea a group of likeminded people, spearheaded by Dr. A. Majeed Khan, an academic entrepreneur with a background in field action research, representing constituent interests resolve to establish an "apex" body in the scheme of a "Virtual University" and to call it a **Committee of Action for Research, Extension and Services (CARES)**. The overall scheme is to promote research and active liaison amongst institutions and programs in the role of a catalyst to achieve a higher and faster growth of economy based research, and extension of evidence based knowledge. It is a non government, non political, not for profit charitable organization dedicated to the development of national wealth and high skilled manpower.

### **Its vision:**

CARES vision is to promote and support basic, applied and prototype research of immediate economic value in the fields of agriculture, life sciences, small and medium agro-enterprises, green technology and rural enterprises. We consider applications of Biotechnology, if made sustainable and affordable, the key for changing livelihood of our farmers and for shaping an inclusive society in Bangladesh.

### **Objectives:**

The principal objectives of **CARES** are:

1. Encourage and advice the stake-holders to outline a dynamic scientific policy for development including prioritization in the fields of agriculture, medical bio-technology and related technology to cope with environmental bio-diversity;
2. Promote internationally competitive research capacity in the country through pooling of resources and expertise, and through inter-disciplinary institutional collaboration;
3. Help establish a number of "centers of excellence" at different academic and research institutions that could serve as regional foci for research and training for young scientists in advanced bio-diversity and related social sciences;
4. Raise funds and donation and liaison grants to support research and institutional development; and work closely with academia, government, R & D institutions, industries and the private sector to achieve the aims of the organization;
5. Work closely with the University Grants Commission (UGC) and relevant government ministries, professional organizations and foundations to promote and ensure excellence in the higher education and enhance opportunities for young scientists;
6. Form a network of Centers of Excellence with complementary expertise and laboratory facilities to help meet the national scientific and related economic development priorities; and

- 
7. Actively promote university-farm-industry tripartite relationship so that the fruits of research optimize public well being and bring positive impact of the growth of national per capital income.

**Programs and Functions:**

**Programs:** **CARES** is a tertiary organization accented on "Action" through a network of organizations, institutions and entrepreneurs. It promotes and assists the following types of programs:

- i. Research,
- ii. Action Research/ pilot Projects,
- iii. Training, Workshops, Dialogues and Seminars,
- iv. Institutional development and
- v. Net-working and dissemination of information.

**Functions:** Based on the above programs, **CARES** is engaged in the following function organized as divisions, each under a Director responsibility to the Director General, the committee and net-work. These divisions are:

- i. Communication and Information technology (net-working)
- ii. Finance and Fiduciary
- iii. Grants and Special programs
- iv. Nuclei projects/ institutions
- v. Public Relation and Human Resources.

**CARES** hopes to play a pivotal role in establishing a scientific culture in the country. It has a vision to see a scientific Bangladesh by creating an enabling knowledge and information driven economy whereby our national growth is facilitated through the application of innovative research in different spheres of human engagement. **CARES** is a platform for our scientists, researchers, academics, professionals and business persons to work collectively and produce results of national and international importance.

**First Executive Committee (2011-2013):**

**Dr. A. Majeed Khan**

*Chairman*

**Professor Dr. M. Shamsher Ali**

President

Bangladesh Academy of Sciences (2008 - 2011): Vice Chairman

**Professor Dr. K.M. Sultanul Aziz**

Fellow, Bangladesh Academy of Sciences - Secretary General



**Prof. Dr. Md Abdur Razzak**  
Former Executive President  
Bangladesh Agriculture Research Council - Assistant Secretary General

**Commodore (Rtd) M. Ataur Rahman**  
Chairman  
Business & Management com. Ltd.(BAMCO)

**Members:**

**Dr. Salehuddin Ahmed**  
Former Governor, Bangladesh Bank  
Professor, North South University

**Dr. Mahabub Hossain**  
Executive Director  
BRAC

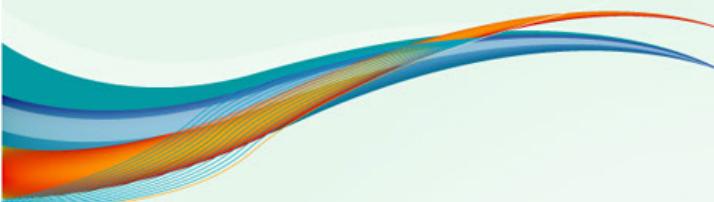
**Lt. Gen. (Rtd) Dr. M. Aminul Karim**  
Senior Research Fellow  
University of Malaya

**Professor Dr. Haseena Khan**  
Department of Biochemistry and Molecular Biology  
Dhaka University

**Mr. Wali-ul- Maroof Matin**  
Chairman and Managing Director  
Alliance Capital Asset Management Ltd.

**Dr. M. Muhit**  
Pro - Vice Chancellor  
University of South Asia, Dhaka

**Maj. Gen. (Rtd) Prof. Dr. ASM Matiur Rahman**  
Physician, Former Adviser  
Caretaker Government  
Fellow, Bangladesh Academy of Sciences.



### **International Conference on Biotechnology (ICB):**

The idea of hosting an international conference on biotechnology has been on the agenda of CARES for a fairly long time. Chairman and some distinguished members of CARES had discussed in various meetings and caucuses during the past one year as to how such a conference would meet the CARES objectives. They had agreed the following to be the conference objectives: (1) taking stock of Biotechnology research in the country, (2) seek encouragement from progress made in the SAARC and ASEAN regions, (3) identification of priority areas, (4) current research capability and international collaborative research, and (5) policy recommendations. The conference will be the formal launching of CARES as well.

### **Expectations from the ICB:**

The conference should conclude with recommendations from participants from home and abroad which CARES could then submit to the Government of Bangladesh and international organizations like the United Nations, The UNESCO, UNICEF, WHO,UNIDO, WTO, WIPO, specialized scientific bodies-the Third World Academy of Sciences (TWAS), the Bangladesh Academy of Sciences (BAS), and, also to the stake holders in the field of sciences in the country.

Our science and scientists need resources and congenial supportive facilities in order to shape the country's future. The support needs to come both from the government as much as from the private sector. CARES seeks to address our business-industry sectors, which are engines of economic growth along with our farmers. Through the Conference, CARES wants to call upon the entrepreneurs to make investment in Biotech industries and enterprises. We believe that Bangladesh truly has the potential to catch up with the achievements of some of our neighboring countries in the field of biotechnology. Together we must take this journey!

## **INTERNATIONAL CONFERENCE ON BIOTECHNOLOGY**

### **Committees**

#### **Organizing Committee**

Chairman : Prof. K.M. Sultanul Aziz  
Secretary : Dr. S.K. Roy  
Members : Prof. M. Shamsher Ali  
Prof. Md. Abdur Razzak  
Commodore (Rtd) M. Ataur Rahman  
Dr. Salehuddin Ahmed  
Dr. Mahabub Hossain  
Lt. Gen. (Rtd) Dr. M. Aminul Karim  
Prof. Haseena Khan  
Mr. Wali-ul-Marоof Matin  
Dr. M. Muhit  
Maj. Gen. (Rtd) Prof. A.S.M. Matiur Rahman  
Prof. Md. Rafiqul Hoque  
Prof. Naiyyum Chowdhury  
Prof. Mesbahuddin Ahmad  
Prof. Mesbahuddin Ahmed  
Dr. M. Zainul Abedin  
Prof. Anwar Huq  
Dr. Firdausi Qadri  
Prof. M.A. Rahim

#### **Core Committee**

Chairman : Dr. A. Majeed Khan  
Members : Prof. K.M. Sultanul Aziz  
Commodore (Rtd) M. Ataur Rahman  
Mr. Wali-ul-Marоof Matin  
Lt. Gen. Harun-Ar-Rashid (Rtd)  
Prof. M.A. Rahim  
Dr. S.K. Roy  
Dr. Saidul Islam

#### **Reception Committee**

Chairman : Dr. A. Majeed Khan  
Chairman, Executive Committee, CARES

**Members :**

Prof. M. Shamsher Ali  
President, Bangladesh Academy of Sciences (2008- 2011)

Prof. K.M. Sultanul Aziz  
Fellow, Bangladesh Academy of Sciences

Prof. Md. Abdur Razzak  
Former Executive President  
Bangladesh Agriculture Research Council

Commodore (Rtd) M. Ataur Rahman  
Chairman, Business and Management Co. Ltd.  
BAMCO

Dr. Salehuddin Ahmed  
Former Governor, Bangladesh Bank  
Professor, North South University

Dr. Mahabub Hossain  
Executive Director, BRAC

Lt. Gen. (Rtd) Dr. M. Aminul Karim  
Senior Research Fellow, University of Malaya

Prof. Haseena Khan  
Department of Biochemistry and Molecular Biology  
Dhaka University

Mr. Wali-ul-Maroof Matin  
Chairman and Managing Director  
Alliance Capital Asset Management Ltd.

Dr. M. Muhit  
Pro-Vice Chancellor  
University of South Asia, Dhaka

Maj. Gen. (Rtd) Prof. A.S.M. Matiur Rahman  
Physician, Former Adviser, Caretaker Government  
Fellow, Bangladesh Academy of Sciences

Mr. Mahbubul Alam  
Editor, The Independent

Mr. Moazzam Hossain  
Editor, The Financial Express

### **Scientific Committee**

Chairman : Prof. Zeba I. Seraj  
Secretary : Dr. S.K. Roy  
Members : Maj. Gen. (Rtd) Prof. A.S.M. Matiur Rahman  
Prof. M.A. Rahim  
Prof. A.K.M. Faziul Haque  
Prof. Ahmed Abdullah Azad  
Dr. Mahabub Hossain  
Dr. Saidul Islam  
Dr. M. Zainul Abedin  
Dr. Firdausi Qadri  
Dr. Munirul Alam  
Mr. M. Shamsul Islam Khan  
Dr. Md. Riazul Islam

### **Abstract Review Committee**

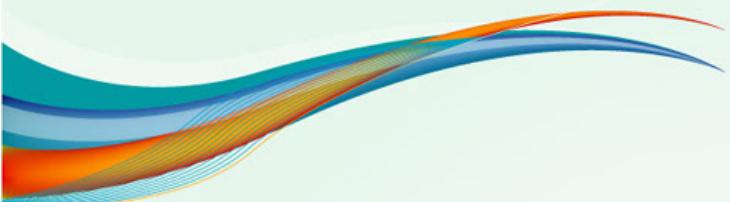
Chairman : Prof. Zeba I. Seraj  
Members : Dr. S.K. Roy  
Prof. K.M. Sultanul Aziz  
Dr. Firdausi Qadri  
Mr. M. Shamsul Islam Khan  
Dr. Munirul Alam  
Dr. Md. Riazul Islam

### **Poster Committee**

Chairman : Dr. Munirul Alam  
Members : Dr. Sucharit Basu Neogi  
Mr. Imran Ali  
Mr. M. Shamsul Islam Khan  
Ms Sadia Faruque  
Mr. Abu Ayub Khalid

### **Accommodation Committee**

Chairman : Mr. Quaiyum Khan  
Members : Mr. Shibli Kaiser Aziz  
Ms. Sadia Faruque  
Mr. Abu Ayub Khalid



### **Media, Communication, and Publication Committee**

Chairman : Mr. M. Shamsul Islam Khan  
Secretary : Dr. Md. Nazim Uddin  
Members : Prof. K.M. Sultanul Aziz  
Dr. S.K. Roy  
Mr. A.B. Manjoor Rahim  
Mr. Shibli Kaiser Aziz  
Ms Sadia Faruque  
Mr. M. Hossam Haider Chowdhury  
Col. Lutful Haque (Retd.)  
Ms Dilara Begum  
Mr. Md. Moniruzzaman  
Mr. Syed Atiar Rahman Sabuz  
Mr. Abu Ayub Khalid

# **International Conference on Biotechnology**

**25-26 May 2013**

On

Application of Biotechnology in addressing the development needs of Bangladesh

At

The Westin, Gulshan 2, Dhaka.

---

## **Programme**

**25 May 2013**

### **SESSION 1**

**Topic / Theme                          Medical Biotechnology**

Chair: H.E Prof. Tissa Vitharana, Sr. Minister of Scientific Affair, Sri Lanka

Co-chair: Prof Mesbahuddin Ahmad, President, Bangladesh Academy of Sciences.

<b>Time</b>	<b>11.15 am to 12.45 pm</b>	<b>Keynote Address:</b>
	11.15 am to 11.40 am	Distinguished Professor Rita Colwell University of Maryland, USA
	11.40 am to 11.55 am	Dr. N.K Ganguly, Distinguished Biotechnology Research Professor, NII, India
	11.55 am to 12.10 pm	Maj. Gen. Prof. Dr. ASM Matiur Rahman, Fellow, Bangladesh Academy of Sciences.
	12.10 pm to 12.20 pm	Dr. Zakir Hossain, Senior Scientist, Transgenic & Gene Targeting, NUS, Singapore
	12.20 pm to 12.30 pm	Dr. Firdausi Qadri, Director, Center for Vaccine Science, senior scientist and head Immunology unit, ICDDR,B
	12.30 pm to 12.40 pm	Q. A.
	12.40 pm to 12.45 pm	Summing up:
	<b>12.45 pm to 14.15 pm</b>	<b>Buffet Lunch</b>

## **SESSION 2**

### **Topic / Theme              Agricultural *Biotechnology***

**Chair:** Prof. Naiyyum Choudhury, Professor and Coordinator, Biotechnology, BRAC University

**Co-chair:** Prof. Dr. Ahmed Ismail Mustafa, Chairman, BCSIR

**Time        14.15 pm    to    15.45 pm**

14.15 pm    to    14.30 pm	Prof. Asis Datta, Professor of Eminence, National Institute of Plant Genome Research, Delhi
14.30 pm    to    14.45 pm	Prof. Dr. S.K. Sopory, Vice Chancellor, Jawaharlal Nehru University
14.45 pm    to    15.00 pm	Prof. M.R.S.Rao, President, Jawaharlal Nehru Centre for Advanced Scientific Research, Bangalore, India
15.00 pm    to    15.10 pm	Dr. Sirimali Fernando, Ministry of Scientific Affairs, Government of Sri Lanka
15.10 pm    to    15.25 pm	Dr. Dhirayos Wititsuwannakul, Assistant President for Research, Siam University, Bangkok, Thailand.
15.25 pm    to    15.35pm	Dr. M.A. Rahim, Professor of Horticulture, Bangladesh Agricultural University
15.35 pm    to    15.40 pm	Q. A
15.40 pm    to    15.45 pm	Summing up
<b>15.45 pm    to    16.15 pm</b>	<b>Tea Break</b>

## **SESSION 3**

### **Topic / Theme                      Economy, Industry and Environment**

**Chair:** Prof. Dr. Md. Rafiqul Hoque, Vice-Chancellor, Bangladesh Agricultural University, Mymensingh

**Co-chair:** Prof. Dr. Dhirayos Wititsuwannakul, Assistant President for Research, Siam University, Bangkok, Thailand

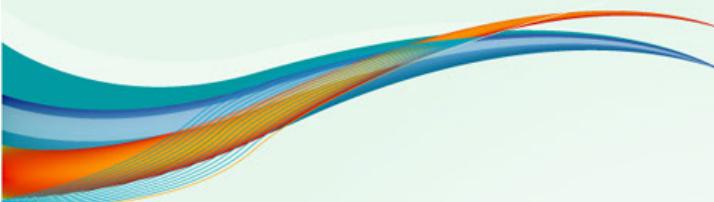
<b>Time</b>	<b>16.30 pm to 18.00 pm</b>	
	16.30 pm to 16.45 pm	Dr. Mahabub Hossain, Executive Director, BRAC Bangladesh
	16.45 pm to 17.00 pm	Mrs. Rokia Afzal Rahman, President, Metropolitan Chamber of Commerce, Dhaka
	17.00 pm to 17.15 pm	Mr. Anis Ud Dowla, Chairman, ACI
	17.15 pm to 17.30 pm	Mr. Amir Khasru. Mahmud Chowdhury, former Minister of Commerce and President, Chittagong Stock Exchange
	17.30 pm to 17.45pm	Dr. M Saif Islam, Director, Northern California Nanotechnology Center University of California - Davis
	17.45 pm to 18.00 pm	Dr. Anwar Huq, Professor, Maryland Dept of Cell Biology and Molecular Genetics, University of Maryland, USA

## **Dinner**

<b>Time</b>	<b>19.30 pm to 21.30 pm</b>	<b>Grand Ball Room, The Westin Dhaka</b>
-------------	-----------------------------	--

## **Programme**

- Launching of CARES
- Award of Fellowship
- Concert
- Dinner



**26 May 2013**

At

The Westin, Gulshan 2, Dhaka.

---

**SESSION 4**

**Topic / Theme**

**Biotechnology: Research and Development**

**Chair:** Prof. Asis Datta, Professor of Eminence, National Institute of Plant Genome Research, Delhi

**Co-chair:** Dr. Hossain Zillur Rahman, Executive Chairman, Power and Participation Research Center.

**Time**      **8.30 am to 10.00 am**

8.30 am	to	8.40 am	Dr. Golam Morshed, Clinical Professor, University of British Columbia (NRB)/ Dr. K.M.S Aziz
8.40 am	to	8.55 am	Dr. John Clemens, Executive Director, ICDDR,B
8.55 am	to	9.05 am	Dr. Firdausi Qadri, Director, Center for Vaccine Science, ICDDR,B
9.05 am	to	9.15 am	Ms Jaishree Sijapati Pandey, Sr. Scientist, Nepal Academy of Sciences and Technology
9.15 am	to	9.25 am	Prof. Kasturi Datta, Distinguished Biotechnology Professor
9.25 am	to	09.40 am	Dr. Mominul Huq, Sr. Vice President, Bangladesh Association of Pharmaceutical Industry.
09.40 am	to	10.00 am	Poster Browsing and Tea.

## **SESSION 5**

### **5.1 Parallel**

**Topic / Theme                          Medical Biotechnology**

**Chair:** Distinguished Prof. Kasturi Datta

**Co-chair:** Dr. Zakir Hossain (NRB)

**Time            10.00 am    to    11.00 am**

**Paper Presenters:**

1. Dr. Sharif Akhteruzzaman, University of Dhaka
2. Mr. Subhagata Choudhury, BIRDEM, Dhaka
3. Mr. Iqbal Hassan Khan, Incepta Pharmaceutical Ltd., Dhaka
4. Dr. Mohammad Alimul Islam , BAU, Mymensingh
5. Ms. Tanvira Afroze Sultana, BIRDEM, Dhaka
6. Ms. Shamsun Nahar Khan, East-West University, Dhaka
7. Muhammad Tufazzal Hossain Howlader, BAU, Mymensingh
8. Dr. Abul B.M.M.K.Islam, University of Dhaka

### **5.2 Parallel**

**Topic / Theme                          Agriculture and Environment**

**Chair:** Prof. S. K. Sopory, Vice Chancellor, JNU

**Co-chair:** Prof. M.R.S. Rao, President, JNCASR

**Time            10.00 am    to    11.00 am**

**Paper Presenters:**

1. Ms. R.H. Sarker, University of Dhaka
2. Prof. Dr. M.A.Rahim, BAU, Mymensingh
3. Prof. Zeba I. Seraj, University of Dhaka
4. Dr. Mirza Mofazzal Islam, BINA, Mymensingh
5. Prof. Dr. M.S.A. Fakir, BAU, Mymensingh
6. Prof. Lutful Hassan, BAU, Mymensingh
7. Prof. K.M.S. Aziz, BAS, Dhaka
8. Prof. Muhammad Manjurul Karim, University of Dhaka

## 5.3 Parallel Topic / Theme

**Microbial and Environmental Biotechnology**

Chair: Prof. Partha Majumder, Director, National Institute of Biomedical Genomics

Co-chair: Mr. Jaishree Sijapati, Senior Scientist, Nepal Academy of Science and Technology

**Time**      **10.00 am**    to    **11.00 am**

## **Paper Presenters:**

1. Prof. Md. Mozammel Huq, University of Dhaka
  2. Mr. Anwar Huq, University of Maryland, USA
  3. Mr. Nur A. Hasan, University of Maryland, USA
  4. Mr. Munirul Alam, ICDDR,B, Dhaka
  5. Mr. Abul Kalam Azad, Shahjalal University of Science & Technology, Sylhet
  6. Mr. M. R. Islam, BAU, Mymensingh
  7. Mr. Md. Bazlur Rahman Mollah, BAU, Mymensingh
  8. Mr. A. K. M Golam Sarwar, BAU, Mymensingh

## **CONCLUDING SESSION**

**Time**      **11.00 am**    to    **12.30 pm**

1. Chief Rapporteurs' summary - Dr. Md Saidul Islam and Dr. Mirza Mofazzal Islam
    - a. Biotechnology, an interdisciplinary science, holds enormous prospect of individual career development as well as institutional collaboration within the region and beyond;
    - b. The Government as well as the private sector have enormous scope and responsibilities to help develop biotechnology research and industrial entrepreneurial collaboration;
    - c. Biotechnology is a multidisciplinary subject of tremendous potential for the development of food, medicine and horticultural products: fruits, orchids, flora etc.
  2. The interface role of CARES in accelerating the industrial, agricultural and entrepreneurial development of national economy and quality human resources – Dr. A. Majeed Khan
  3. Address by the Guest of Honor H.E. Architect Yeafesh Osman, State Minister for Sciences and Technology, Government of Bangladesh



## International Conference on Biotechnology

*on application of Biotechnology  
in addressing the development needs of Bangladesh*

25-26 May, 2013

**ABSTRACTS OF ORAL PRESENTATIONS**

## Probiotic Technology: A new solution for Sustainable Aquaculture in Bangladesh

Abu Md. Ramim<sup>1</sup>, Sabikunnahar<sup>1</sup>, Shafiqur Rahman<sup>1</sup>,  
M. Niamul Naser<sup>2</sup>, and Muhammad Manjurul Karim<sup>1</sup> (manjur@univdhaka.edu)

<sup>1</sup>Department of Microbiology and <sup>2</sup>Department of Zoology, Faculty of Biological Sciences,  
University of Dhaka, Dhaka 1000, Bangladesh

**Background:** *Macrobrachium rosenbergii*, the giant freshwater prawn, is one of the most important commercially-produced crustaceans in Bangladesh. A significant limitation to the industry is loss of productivity due to the emergence of various pathogenic bacteria and viruses and their resistance to chemotherapeutic drugs, resulting in mass mortality and consequent crop failure. Finding a public-health and environment-friendly alternative is, therefore, a burning question to ensure the sustainability of this industry in Bangladesh. **Objective:** Probiotics-the friendly bacteria with a host of benefits that work by competitive exclusion of pathogenic bacteria-can be a suitable alternative. This study was undertaken to test probiotic bacteria able to inhibit the growth of pathogenic bacteria that cause mortality of prawn. **Methods and materials:** Following an outbreak of disease in prawn hatcheries of the Sathkhira and Khulna regions during May 2012, samples were collected immediately from dead prawn larvae, rearing water, and fish feed to isolate and characterize the pathogenic bacteria. The antibiogram of the isolated pathogens was conducted before they were challenged to compete with probiotic bacteria, *Lactobacillus* spp., isolated from curd. **Results:** The bacterial count was significantly higher ( $p<0.05$ ) in the animal samples than that in the water and fish feed samples, indicating that mortality was due to bacterial infections. The dead animals harboured heavy loads of pathogenic bacteria and were identified as *Vibrio* spp. and *Shigella* spp. based on the morphological, biochemical and physiological characteristics of the pathogens. Twenty-six antibiotics were tested to study the drug-resistance patterns of the isolates. All the tested isolates ( $n=36$ ) showed remarkable resistance to virtually all the drugs tested ranging from moderate to complete resistance, and the highest sensitivity of the isolates (75%), however, was recorded for doxycycline. In an *in vitro* attempt to control the growth of pathogens in solid media, the cell-free supernatant (CFS) of the two-day old *Lactobacillus* spp., soaked in blank discs, clearly produced at least 15 mm zone of inhibition. In broth cultures, CFS, collected at intervals of different days, produced a dose- and age-dependent reduction of bacterial count. **Conclusion:** The inhibition of bacterial growth could be attributed to some extracellular substances released from *Lactobacillus* spp., demonstrating its potential to be an excellent probiotic candidate for possible applications in prawn aquaculture. If successfully transferred to rearing environment, this will be an eco-friendly approach to counter bacterial infections without compromising the quality of prawn, thereby ensuring food safety in the prawn industries of Bangladesh.

**Acknowledgement:** The study was supported by the Department of Fisheries, Ministry of Fisheries and Livestock, and the Ministry of Science and Technology, Government of Bangladesh.

## **Development of a Novel Universal Multiplex RT-PCR Kit for Rapid Serotyping of Dengue, Chikungunya, Japanese Encephalitis, and West Nile Viruses from Clinical and Field Samples by Single-step-single-tube Reaction**

**Mohammed Alimul Islam<sup>1,2</sup> (alim\_bau@yahoo.co.in), Md. Mortuza Ali<sup>3</sup>,  
Shingo Inoue<sup>1</sup>, and Kouichi Morita<sup>1</sup>**

<sup>1</sup>Department of Virology, Institute of Tropical Medicine, Nagasaki University, 1-12-4 Sakamoto, Nagasaki 852-8523, Japan, <sup>2</sup>Department of Microbiology and Hygiene, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh, and <sup>3</sup>Department of Community Medicine, Gonoshasthaya Samaj Vittik Medical College, Gono Bishwabidyalay, Mirzanagar, Savar, Dhaka 1344, Bangladesh

The study was designed to develop a rapid and simultaneous detection of most important arboviruses of human being of the tropical and sub-tropical countries of the world. ELISA and other serological tests, RT-LAMP, NASBA, and conventional RT-PCR have been used for the detection of antigens and antibodies of dengue, Chikungunya, Japanese encephalitis, and West Nile viruses all over the world. All these tests are time-consuming, laborious, and costly, and the result of diagnosis is sometimes confusing. Any of the above-mentioned tests, which have been designed for the detection of one viral genome from other viral genomes, remains undetected. The sensitivity and specificity of a multiplex RT-PCR depends on the design of primers, uses of enzymes, and condition of nucleic acid amplification. In this study, three DNA-polymerase (LA-Taq, r-Taq, and Tth) and two reverse-transcriptase (AMV-RT and RT-ACE) were used. Highly gene-specific primer was designed against each of the four serotypes of dengue and other serotypes of Chikungunya, Japanese encephalitis, and West Nile viruses to increase the sensitivity and specificity of MRT-PCR. Of the six combinations, the AMV-RT (reverse transcriptase) and La-Taq (DNA polymerase) combination was found to be the best in terms of sensitivity and specificity of the MRT-PCR, which could detect a minimum number of any of the four species of arboviruses obtained either from the clinical or laboratory or field samples. The test was found to be cost-effective (Tk 50.0/US\$ 0.60), rapid (1.5 hours), and sensitive (0.1 FFU can be detected). Anyone, either from the developed or developing countries, having minimum knowledge on PCR can easily perform the test either in the hospital or in a diagnostic centre dealing with bulk samples where the diseases are endemic.

**Acknowledgement:** The study was supported under the Core Research for Evolutional Science and Technology (CREAST) programme, Japan

## Biodegradation of Reactive Textile Dyes by Bacterial Isolates

Abul Kalam Azad (dakazad-btc@sust.edu), **Md. Zobaidul Hossen,**  
Kamrul Islam, and Mohammad Majharul Islam

Department of Genetic Engineering and Biotechnology, Shahjalal University of  
Science and Technology, Sylhet 3114, Bangladesh

Most textile industries in Bangladesh dispose of reactive dyes in the environment without any treatment and pollute the environment severely. To obtain bacteria having a remarkable ability to decolorize and degrade reactive textile dyes, 29 bacterial strains were isolated from the effluents collected from two textile mills and two leather industries. Screening of these isolates for dye decolorization and degradation capability was performed in the nutrient broth medium using eight structurally different reactive textile dyes. Of these bacterial isolates, 12 showing one or more dye decolorizing ability within 48 hours of incubation were identified. Morphological, cultural and biochemical characterization indicated two isolates as *Aeromonas*, three as *Pseudomonas*, three as *Bacillus*, two as *Serratia*, one as *Citrobacter*, and one as *Morganella*. The decolorization and degradation capability of *Aeromonas*, *Pseudomonas*, and *Bacillus* was optimized using Novacron Super Black G, one of the eight reactive dyes used. Physicochemical conditions for decolorization of Novacron Super Black G by the *Aeromonas*, *Bacillus*, and *Pseudomonas* isolates were optimized. These bacteria decolorized the reactive dyes and grew well in a high concentration of the dye up to 500 mg/L. *Aeromonas* sp., *Bacillus* sp., and *Pseudomonas* sp. showed significant dye decolorization by 93%, 92%, and 91% respectively at 200 mg/L dye concentration after 96 hours of incubation under optimum conditions. Biodegradation and decolorization of reactive dye were confirmed using UV-VIS spectrophotometry and Fourier transform infrared spectroscopy. Peaks of the parent dye compound completely disappeared after 96 hours of incubation. This result clearly indicates that the dye had been catabolized and used by the bacterial isolates. High decolorization efficiency and facile conditions of these bacterial isolates indicate their potential in the biological treatment of dyeing mill effluents.

## Functional Structures of Cry4Aa Toxin Responsible for Mosquitocidal Activity against *Culex pipiens*

Mohammad Tofazzal Hossain Howlader<sup>1,2</sup> (tofazzalh@bau.edu.bd),  
Saori Nakao<sup>2</sup>, Hiroshi Sakai<sup>2</sup>, and Tohru Hayakawa<sup>2</sup>

<sup>1,2</sup>Department of Entomology, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh  
and <sup>2</sup>Laboratory of Gene Engineering, Department of Bioscience and Biotechnology, Graduate  
School of Natural Science and Technology, Okayama University, Japan

**Background:** Cry4Aa is a mosquitocidal protein produced by *Bacillus thuringiensis* subsp. *israelensis*. Cry4Aa exhibits potent and specific toxicity to *Anopheles*, *Aedes*, and *Culex* mosquito larvae. These mosquitoes are vectors of malaria and viral haemorrhagic fevers. Hence, Cry4Aa attracts great interest for developing bioinsecticide. The three-dimensional structure of Cry4Aa is similar to the structures of previously-characterized Cry toxins that are composed of three domains (domain I, II, and III). **Objective:** The study was undertaken to determine the functional structures of Cry4Aa involved in mosquitocidal activity. **Methods and materials:** Loop structures exposed on the surface of Cry4Aa molecule were analyzed by alanine scanning. At the same time, the polypeptides derived from domain II and III of Cry4Aa were expressed as alkali-soluble inclusions using the expression vector p4AaCter. The polypeptides were purified using Ni-charged affinity column. Interactions between these polypeptides and the brush border membrane (BBM) proteins prepared from *C. pipiens* larvae were analyzed using the quartz crystal microbalance (QCM) device. Involvement of sugars in Cry4Aa toxicity was also analyzed by bioassay using *C. pipiens* larvae. **Results:** Alanine scanning analyses revealed that the loops exposed on the surface of Cry4Aa (loops between beta1-alpha8, beta2-3, beta4-5, beta6-7, beta8-9, and beta10-11) could be modified without the loss of its mosquitocidal activity. The analyses using the QCM device revealed relatively high affinities of the polypeptide of beta1-alpha8, beta2-3, and domain III to BBM proteins, and their KDs were estimated as 59, 54, and 63 nM respectively. Bioassay using the Cry4Aa pretreated with various monosaccharides revealed the enhancement of toxicity by GalNAc pretreatment. On the other hand, the pretreatment with fucose inhibited the toxicity of Cry4Aa. **Conclusions:** Multiple subsites that work cooperatively for receptor binding may be spread out in domain II and III of Cry4Aa. Especially, the domain III of Cry4Aa may interact with BBM proteins via GalNAc and/or fucose residues of its sugar-side-chain. Thus, the mechanism of Cry4Aa is a unique and may be quite different from that of well-characterized other Cry toxins.

**Acknowledgement:** The work is supported by the Japan Society for the Promotion of Science (KAKENHI Grant No. 2301392 and 24380034).

## Creation of Knock-out Mouse Models by Transgenesis to Understand Human Physiology and Diseases

Md. Zakir Hossain (csmzh@nus.edu.sg)

Transgenic and Gene Targeting Facility, Cancer Science Institute of Singapore,  
National University of Singapore, Centre for Translational Medicine,  
14 Medical Drive 12-01, Singapore 117599

**Background:** The anatomy, genetics, and physiology of mouse model is very close to higher mammals, including human; thus, mouse is a rational and relevant model for biomedical research. The rapid advancement in genome bio-informatics and transgenic technology enables the fine mapping and precise engineering of mouse genome. **Objective:** Loss or gain of gene function is the fundamental mechanism in biological process and in various human anomalies and diseases. Therefore, we exploited the transgenic approaches to create knock-out or knock-in mouse models to understand the function of homology gene in human (patho-) physiology. **Methods and materials:** Candidate genes were selected based on experimental and bio-informatics evidences. Gene-targeting vectors were constructed; genes of interest were targeted in mouse embryonic stem (ES) cell line. Chimera mice were generated from targeted ES cell clones and bred to establish knock-out or knock-in mice for gene of interest. Phenotypic (survival/behavioural, histological and/or molecular) analysis was conducted in knock-out mice and wild-type/heterozygous controls. Animal breeding, tissue harvesting and analysis were performed according to the National Advisory Committee for Laboratory Animal Research and the Institutional Animal Care and Use Committee. **Results:** Subtle or severe phenotypes correlated with the gene-expression pattern and molecular function of a specific gene of interest. Homeo-domain genes Emx/Otxare involved in early patterning of embryonic brain, knock-out or knock-in of these genes caused regionalization defects in the brain. Zo2/Tjp2, a scaffolding protein associated with tight junctions required in early embryogenesis and targeted inactivation of Zo2, caused embryonic lethality at embryonic day 6.5 (E6.5). Testis-specific cold-shock-domain gene (Msy4) is important for spermatogenesis, and inactivation of these gene caused male infertility in mice. A transcriptional co-activator Taz/Wwtr1, highly expressed in kidney and lung, is a candidate gene for polycystic kidney diseases and pulmonary emphysema. Aberrant expression of Taz is associated with non-small cell lung cancer. **Conclusion:** Knock-out and knock-in mice are powerful tools to understand the genetic link in the biological process and diseases, which can, thus, serve as a rational pre-clinical model for the therapeutic target discovery and validation of many diseases.

**Acknowledgement:** Institute of Molecular Embryology and Genetics (IMEG), Japan; Agency for Science Technology and Research (A\*STAR), Singapore; Ministry of Education (MOE), Singapore; National Research Foundation, Singapore (NRF).

## Rubber Latex Coagulation Mechanism Elucidated and the Biotechnology Implication to Field Applications

Dhirayos Wititsuwannakul (dhirayos@siam.edu)

Siam University, Petkasem Road, Bangkok 10163, Thailand

An *in vitro* aggregation of washed lutoid membrane with rubber particles, prepared from the bottom (lutoid) fraction and rubber layer of centrifuged fresh latex, leading to the rubber coagulum formation and the latex coagulation, was demonstrated. A Triton X-100 extract of lutoid membrane protein was isolated and prepared examining for its role in the latex coagulation process. Aggregation of the rubber particles (RP) was shown to be induced by a specific Hevea latex lectin-like protein (HLL) present on the lutoid membrane. The purified HLL was 17 kDa monomer whereas the natural active form of HLL in the latex was found to be 267 kDa lectin, with a pI value of 7.2. A binding protein (BP) ligand bound on the RP surface as a counterpart for HLL was identified and characterized as RP-HLLBP, for its specific interaction with the HLL. The RP-HLLBP activity was assayed by its ability to compete with erythrocytes in the standard haemagglutination inhibition (HI) assay to its monitor-specific interaction. It was with a strong HI titre (high lectin recognition), optimum activity at pH 5-8 and was thermostable up to 60 °C. It was a glycoprotein with Mr of 24 kDa. The released free RP-HLLBP was shown to inhibit latex coagulation via competition with the RP bound glycoprotein BP. Chitinase abolished its HI activity and inhibited HLL-induced RP aggregation in a competitive dose-dependent manner, indicating the presence and the role of N-acetylglucosamine (NAG) as a key factor in the HLL binding recognition. These results were, thus, final elucidation of the long puzzle RP coagulation mechanism.

**Acknowledgement:** The study received supporting grants from USAID (USA) and the Thailand Research Fund (TRF)



## **Prospects and Issues for Developing Biotechnology Industries in Bangladesh: Laboratory Perspectives**

Muhammad G. Morshed (Muhammad.Morshed@bccdc.ca)

Department of Pathology and Laboratory Medicine,  
University of British Columbia, Vancouver, BC, Canada

Bangladesh is one of the most densely-populated countries in the world and became a global partner in pharmaceutical sectors, however, far behind in other biomedical industries, such as biotechnology, medical devices, and laboratory diagnostics. Like pharmaceuticals, all these segments are science-driven and should be highly regulated. Among the SAARC countries, India is well ahead of laboratory diagnostic segments of biomedical industries. Evidence-based patient management become routine, and, thus, small and medium-scale laboratory diagnostic companies can play a crucial role nationally and globally. Small and medium-scale laboratory diagnostic companies in Bangladesh can grow in three stages: test development, commercialization, and marketing. Test development is the crucial part, and it should be based on trial and error methods, and funding should mostly come through the government sector. Commercialization and marketing can be channeled through venture capital or diversification fund (share market). Biomedical graduates are underused in Bangladesh, and they are highly capable in basic, applied, developmental and product-oriented research with proper research facilities. Quality product and work environment would be a challenge. A few examples of the prospects and challenges of small- and medium-scale laboratory diagnostic companies will be discussed.

## High-Tech Science for a Low-Tech Solution: Sari Filtration for Cholera Prevention

Anwar Huq and Rita Colwell

University of Maryland, College Park, Maryland, USA

**Introduction:** *Vibrio cholerae* is a natural inhabitant of the aquatic environment and when present in drinking-water can cause cholera in humans. Person-to-person transmission is accelerated by contact with the bacterium under poor hygienic conditions. Environmental parameters have been determined that are associated with increased numbers of the bacteria comprising an infectious dose. Using contaminated water for bathing, washing fruits and vegetables, and drinking, can lead to serious epidemics of cholera. Those at the lowest economic stratum are at the greatest risk as they tend to be malnourished, lack proper sanitation, and have no access to potable water. **Methods and materials:** A clear understanding of the ecology of *V. cholerae* was essential to develop strategies for prevention and/or intervention of the disease. One major impediment, however, was the inability to detect *V. cholerae*, the causative agent of cholera because it can enter into a viable but non-culturable state, when bacteriological media will fail to support growth. Using a direct detection method, optimized in our laboratory that has since become a major tool in ecological studies, we demonstrated that cholera bacteria in the natural environment are associated with free-swimming, small microscopic plankton, and copepods. Each individual copepod can carry up to 104 cholera bacteria. Ingesting 1-10 such copepods in untreated water or by eating fresh fruit or salad contaminated with the cholera bacteria can cause the disease. Cholera bacteria are attached to their host, the copepod, in the aquatic environment, which can be removed from the water by simple filtration, a method developed in our laboratory at the University of Maryland and was field-tested to determine its efficacy. **Results:** Using the sari filtration method, 99% of bacteria attached to copepods and particles were removed from water. A field trial in a population of 45,000 in Matlab, Bangladesh, using four layers of sari cloth resulted in 48% reduction of cases of cholera. A follow-up study was carried out in the same villages five years after the original trial and showed that the filtration method is sustainable, continuing to protect villagers, with 25% fewer cases of cholera where filtration was continuing to be used. Additional analysis showed an indirect herd protection, i.e. protection for households from cholera when surrounded by those families who did filter their water. **Conclusion:** Simple filtration proved to be effective and was based on ecological studies carried over the past 30 years. It can save lives without financial burden to the community, if implemented properly.

**Acknowledgement:** The work was supported by the U.S. National Institutes of Health and the Thrasher Research Fund.

## Current Trends of Biosimilar Growth Open Opportunities for Bangladesh

Md. Shawkat Hossain, Raquibur Rahman, Ataur Rahman, Mohammed Zakiur Rahman, and  
**Md. Iqbal Hassan Khan** (iqbal@inceptapharma.com)

Research and Development Department, Biotechnology Derived Product Facility,  
Biotech Division, Incepta Pharmaceuticals Ltd., Bara Ranagmatia, Zirabo, Savar,  
Dhaka 1341, Bangladesh

Biosimilars or follow-on biologics (FOBs) of many blockbuster biopharmaceutical products will lose patent protection of 21 products within 2019 having a total market value of over US\$ 50 billion. Since the top 25 biologics are driving 83% of global sales, patent expiry of many of these products is opening up new possibilities for FOB players in the next five years. Shifting in disease patterns, product demand, and better tertiary care boost enormous commercial opportunity for the companies interested in FOBs. However, due to the high clinical development and manufacturing costs (US\$ 40-80 million), the price difference between biosimilars and corresponding originator products is still a challenge. It needs at least 40-50% price reduction from branded products to meet the customer's expectation. Being a new field based on a new regulatory pathway, FOBs are in direct competition with some very large, well-established innovator companies with enormous budgets. Moreover, the development of second-generation biopharmaceutical products in the market perhaps with improved safety and efficacy than original first-generation products is again a challenge for FOB marketing. But in the case of Bangladesh which meets her 97% of pharmaceutical local market demand, FOB production will significantly reduce the dependency on imported, expensive biotherapeutics. For instance, FOBs in India, attracting large investments in areas of research, clinical trials, and manufacturing, are expected to grab at least 20-25% of global market share in biosimilars because of its established infrastructure, talent pool, and consistent quality compliance within the next five years. With the advent of potential niche, low-cost power in unregulated and semi-regulated markets, Indian players are sensitized to adopt in line with the US and EU stringent biosimilar guidelines. Similarly, Bangladesh can enjoy facility and development costs than peers in developed countries and add another pillar of success achieved in small molecule generics by partnering with large multinational corporations for clinical trials and regulatory approval process in EU/US. Incepta Pharmaceuticals Ltd. has developed the country's first GMP compliant research-cum-commercial integrated facility to synthesize FOBs using r-DNA technology. The pilot facility currently simulates the potential process for production of insulin, filgrastim, interferon, erythropoietin, etc, in pilot scale, which will be followed by dosage form design (formulation) and commercialization of FOBs.

## DNA Marker-assisted Breeding and Genetic Transformation for Producing Salt-tolerant Rice for Bangladesh

Zeba I. Seraj (zebai@univdhaka.edu)

Plant Biotechnology Laboratory, Department of Biochemistry and Molecular Biology,  
University of Dhaka, Ramna, Dhaka 1000, Bangladesh

The major focus of our laboratory is the development of salt-tolerant high-yielding rice varieties using molecular breeding and genetic transformation techniques. These varieties are essential for cultivation in the saline coastal areas of Bangladesh, which occupy a tenth of our total cultivable land. The saline-prone areas are gradually increasing both in severity and area due to climate change?therefore, the need for highly-tolerant rice cannot be over-emphasized. Marker-assisted backcrossing was then used to introgress a large region or rice chromosome 1, called Saltol, into the mega-rice varieties, BR11 (T. Aman) and BRRIdhan28 (Boro), and several near isogenic lines (NILs) were produced in a collaborative effort with BRRI. Some of these salt- tolerant BR11 lines and BBRI dhan28 lines have given yield advantage over their parents in both normal soil and salinity-affected areas. Novel salt tolerance determinants from Rice Landraces adapted to the coastal areas of Bangladesh, such as Boilam and Horkuch, are being identified. These QTLs can then be introgressed into existing varieties to produce a higher level of salt tolerance. Different genes shown to confer salt tolerance, including Na/H antiporters, detoxification genes, RNA/DNA unwinding genes. and transcription factors, have been cloned and transformed into rice and are in various stages of development. The best-performing plant is selected at T2. The transgene is then backcrossed into farmer-popular high-yielding varieties. The Agrobacterium tumefaciens mediated in Planta method is being tried to avoid tedious tissue culture and plant regeneration steps. Using this method, we have introduced several genes into BRRI dhan 27, 28, 36, 47, and 55.

**Acknowledgement:** Funds from the U.S. Department of Agriculture and Bangladesh Academy of Sciences (BAS) to my laboratory for the above work are gratefully acknowledged.

## Development of Fungal Disease Resistance in Lentil (*Lens culinaris* Medik.) Following Agrobacterium-mediated Genetic Transformation.

Subroto Kumar Das, M.I. Hoque, and R.H. Sarker (rhsarker2000@yahoo.co.uk)

Plant Breeding and Biotechnology Laboratory, Department of Botany,  
University of Dhaka, Ramna, Dhaka 1000, Bangladesh

The present investigation was aimed to integrate fungal diseases-resistant gene in lentil (*Lens culinaris* Medik) plants through *Agrobacterium*-mediated genetic transformation. As an integral part of *Agrobacterium*-mediated genetic transformation, three different explants, namely cotyledonary node, decapitated embryo, and cotyledon attached decapitated embryo, were used for developing a transformation compatible regeneration system. Regeneration of multiple shoots was achieved via direct organogenesis from the above-mentioned explants on MS medium supplemented with 0.5 mg/L BAP + 0.5 mg/L Kn + 0.1 mg/L GA3 + 5.5 mg/L tyrosine. Among the three explants, cotyledonary node explants showed the best result towards *in vitro* regeneration. However, the regenerated shoots failed to achieve an effective root system. Encouraged by the reports of previous workers, it was tried to induce *in vitro* flowering on regenerated shoots to overcome the problem of rooting in obtaining complete plantlets. The best response regarding the development of *in vitro* flowering was obtained by culturing regenerated shoots on half strength of MS medium containing 20 mg/L IBA and 0.5 mg/L NAA. Transformation experiments were performed using two strains of *Agrobacterium*; one was with marker genes (denoted as strain I) and other with antifungal gene (denoted as strain II). The marker strain LBA4404 containing binary plasmid pBI121 conferring  $\beta$ -glucuronidase (GUS) and *nptII* gene was resistant to kanamycin. Antifungal strain EHA105 harbouring bar gene was resistant to phosphinothricin and chitinase gene. Considering transformation and regeneration efficiency, cotyledon attached decapitated embryo was found to be best among all the explants studied. For strain I, transformed shoots were selected using 200 mg/L kanamycin. On the other hand, 2.0 mg/L phosphinothricin was found to be optimum to select transformed shoots containing fungal resistance genes. Transformation frequency for strain I and strain II was 1.06% and 0.49% respectively. The selected shoots developed *in vitro* flowers following their subculture on half strength of MS medium containing 20 mg/L IBA and 0.5 mg/L NAA with 50 mg/L ticarcillin. After 12-15 days, most of these flowers produced fertile seeds on the same medium. Seedling germinated from these *in vitro* raised seeds was successfully transplanted to soil for the development of further progenies. Genomic DNA was isolated from these transformed lentil shoots for molecular analysis through polymerase chain reaction analysis. In the case of strain I, stable integration of GUS gene was confirmed by PCR analysis. For strain II, it confirmed the integration of fungal diseases-resistant gene within the genomic DNA of transformed shoots of lentil. This technique of *in vitro* flowering and seed formation can be exploited to develop transformed seeds in lentil since *in vitro* root formation appears to be a major constraint in obtaining complete plantlet under *in vitro* condition.

**Acknowledgement:** This investigation was supported by the U.S. Department of Agriculture through a project.

## **Promise of Bioinformatics in Pharmaceutics and Diagnostics**

Abul B.M.M.K. Islam (khademul@du.ac.bd)

Department of Genetic Engineering and Biotechnology,  
University of Dhaka, Dhaka 1000, Bangladesh

The combination of high-throughput technologies with information technologies has produced an enormous amount of information relating to biomedicine. However, extensive growth in biomedical data generation has not yet been translated proportionately for clinical returns. Bioinformatics holds immense promise in this area by developing new tools to efficiently capture, curate, and analyze these huge data, thereby helping diagnose diseases, identify drug targets, and develop new medicines. Integration of multi-dimensional genomic data and genome-wide association studies (GWAS) may contribute profoundly to explore the mechanism of complex diseases. However, it requires correct record of phenotypic information. Bioinformaticians can play a role to develop software for collecting, integrating, and extracting clinical information; database development, and data management. They further can develop a database of novel mutations/SNPs and their associations with drug responses. Such high-throughput studies with Bangladeshi patients, however, are still very limited and should be explored. While conventional diagnosis may sometimes be erroneous, bioinformatic methods/tools may help us find out disease-specific gene signature or biomarkers for accurate and specific molecular diagnosis. This method may also classify diseases, find new disease genes, delineate the pathogenic pathways, predict patients' survival time, predict functional consequences of mutations, identify the mode of action of candidate drugs, and improve therapy by detecting and clustering important disease subtypes. Recently, the Open Source Drug Discovery initiative for system-level understanding has drawn much attention. Computer-aided drug design can dramatically reduce time and cost of effective biopharmaceuticals. Fortunately, we have enormous varieties of plants having medicinal value, whose active compounds can be tested computationally for rational drug design. Recently, epigenetic enzymes have drawn much attention as drug target due to their reversible nature. Wealth of data generated world-wide can be analyzed to find epigenetic drug target and predict clinical outcome and possible side-effects. Correlation- and network-based analysis is becoming more promising for designing combinational therapy. As the new era of personalized medicine is approaching, development of new bioinformatic systems and databases is needed for individualized therapies. Therefore, translational bioinformatic applications in genome medicine are expected to generate a great hope for future medicines.

## Genetic Polymorphism of 30 InDel Markers for Forensic Use in Bangladeshi Population

Sudipta Arka Das<sup>1</sup>, Ismail Hosen<sup>1</sup>, Ahmad Ferdous<sup>2</sup>, and Sharif Akhteruzzaman<sup>1</sup>

<sup>1</sup>Department of Biochemistry and Molecular Biology, University of Dhaka, Ramna, Dhaka 1000, Bangladesh and <sup>2</sup>National Forensic DNA Profiling Laboratory, Dhaka Medical College, Dhaka 1000, Bangladesh

**Background:** Insertion-deletion polymorphisms (InDels or DIPs) represent a large portion of all polymorphisms in human genome. These are basically length polymorphisms created by insertion or deletion of one or more nucleotides, combining the common features of both SNPs and STRs. Their low mutation rate, smaller amplicon size, and multiplexing capability made the polymorphisms suitable for use in forensic and parentage testing. This study investigated 30 diallelic autosomal insertion-selection polymorphisms in a Bangladeshi population sample. **Objective:** The purpose of the study was to assess the suitability of 30 InDel polymorphisms in forensic applications and population studies. **Materials and methods:** In this study, a set of Bangladeshi population sample ( $n=132$ ) was genotyped using 30 InDel markers included in Investigator DIPplex PCR amplification kit (QIAGEN, Germany). The analytical methods involved the extraction of genomic DNA from peripheral blood samples. The extracted DNA was PCR-amplified in a multiplex fashion. The resultant PCR products were separated by capillary electrophoresis for genotype assignment. **Results:** Allele frequency, observed and expected heterozygosity, polymorphism information content, probability of match, power of discrimination, typical paternity index (TPI), and power of exclusion were calculated for these loci. The Hardy-Weinberg equilibrium tests demonstrated no significant deviation from expected values ( $p>0.00167$ , after Bonferroni correction for multiple testing). The random probability of match was  $2.87 \times 10^{-12}$ , and the cumulative power of exclusion was 0.99470 for the 30 loci. **Conclusion:** The high level of power of discrimination (0.999999875) makes it well-suited for the identification of individuals. However, a relatively-low range of TPI (0.791-1.179) and PE (0.0685-0.263) limits its usefulness in paternity and kinship investigations. The studied 30 InDel loci, however, offers a good supplementary tool for resolving challenging kinship studies and an efficient alternative to SNP typing in the studied population.

## Molecular Diagnostic Tests in Bangladesh: Challenges and Opportunities

**Subhagata Choudhury, Tanvira Afroze Sultana, M. Sohrab Alam, and Rubyyat Hassan**

BIRDEM General Hospital, 122 Kazi Nazrul Islam Avenue,  
Shahbagh, Dhaka 1000, Bangladesh

**Introduction:** Molecular diagnosis is rapidly becoming an inseparable part of diagnosis of diseases. This cutting-edge technology can be used for diagnosing both malignant and infectious diseases and help in determining the drug dosage, tissue types for organ transplant, and risk of inherent disorders. An added advantage is that it provides an indication of therapeutic choices and prognosis of diseases. **Methods and materials:** A survey was conducted in some diagnostic laboratories and research institutes of Bangladesh to observe the existing range of molecular diagnostic tests in Bangladesh. The challenges faced in establishing and sustaining these tests were noted, and opinions of stakeholders regarding further opportunities were recorded to improve this area. Results: Hospitals, namely BSMMU, BIRDEM, CMH, Square, Apollo, and icddr,b and diagnostic laboratories, such as DNA Solutions, Labaid, Medinova, and Popular Diagnostics Ltd., have introduced molecular diagnostics in Bangladesh. PCR-based tests are available for MTB, HBV, HCV, HIV, and HPV and for a few types of leukemia. Immunophenotyping has been introduced in one institute and one private hospital but the service is interrupted by various limiting factors. FISH is not available in the country. **Discussion:** Challenges in the development of molecular diagnostics are variable. There is limited knowledge on molecular genetics among physicians. The meagre human resource in the country is inappropriately used. Strong advocacy and marketing strategies of neighbouring countries favour medical services abroad. Customs regulations are not favorable in terms of high-tax rates and complicated procurement mechanisms. Strong policies are lacking to ensure after-support from the suppliers. Service available is centred in the capital. Most of all, there is no reference laboratory in the country. **Conclusion:** Bangladesh has an immense potential for the development of molecular diagnostics in the country. Review of policies relating to import and support of cutting-edge technology in the diagnostic sector with the involvement of experts in this field is essential to make the sector viable. The establishment of a specialized institute on molecular diagnostics can yield a score of professionals. Strong commitment from physicians, molecular biologists, financial companies, and government policy-makers together are required to bring molecular diagnostics to the doorstep of every citizen of the country.

**Acknowledgement:** The authors acknowledge the support of BSMMU, BIRDEM, CMH, Square, Apollo, icddr,b, DNA Solutions, Labaid, Medinova, and Popular Diagnostics Ltd.

## Molecular Epidemiological Analysis of Highly-pathogenic Avian Influenza Viruses (H5N1) of Bangladesh

M.E. Haque<sup>1</sup>, M. Giasuddin<sup>2</sup>, E.H. Chowdhury<sup>1</sup>, and M.R. Islam<sup>1</sup>  
(mrislam\_bau@yahoo.com)

<sup>1</sup>Department of Pathology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh 2National Reference Laboratory for Avian Influenza, Bangladesh Livestock Research Institute, Savar, Dhaka, Bangladesh.

**Background:** Highly-pathogenic avian influenza (HPAI) viruses of the H5N1 subtype have been devastating the poultry industries across the world since 2003. The virus also may transmit to humans with high case fatality. In Bangladesh, H5N1 HPAI virus was first detected in February 2007. Since then, the virus has become entrenched in poultry of Bangladesh and caused six waves of outbreaks. So far, there have been seven human cases of H5N1 HPAI infection in Bangladesh with one case fatality. **Objective:** The study was conducted to investigate the molecular epidemiology of H5N1 HPAI viruses circulating in Bangladesh since 2007. **Materials and methods:** A fragment of HA gene of 21 selected isolates obtained at different time points was amplified by RT-PCR and subjected to gene sequencing and phylogenetic analysis along with reference strains of different clades and sub-clades. Full-length genes of different genome segments of the selected isolates were also amplified by RT-PCR and sequenced either directly or after cloning in plasmid vectors. In addition, available gene sequences of other Bangladeshi isolates, established at international reference laboratories, were also downloaded from the GenBank. Phylogenetic tree was constructed for each genome segment. Molecular analysis was also performed on multiple alignments of deduced amino acid sequences of each protein. **Results:** The results revealed that clade 2.2 virus was first introduced in Bangladesh in 2007, followed by the introduction of clade 2.3.2.1 and 2.3.4 viruses in 2011. Interestingly, in 2012, only clade 2.3.2.1 viruses were isolated. Phylogenetic analysis of individual full-length genome segments revealed at least two events of segment re-assortment. In three different H5N1 isolates, either M or PB1 gene was substituted by the corresponding segment of low pathogenic H9N2 virus. Point mutations were also acquired at the potential active sites of different proteins. **Conclusion:** Implications of these HA clade shift, segment re-assortment, and point mutations will be discussed.

## **Food Security in Bangladesh: Cassava (*Manihot esculenta* Crantz) could be a Potential Supplementary Crop to Rice and Wheat**

**<sup>1</sup>M.S.A. Fakir<sup>1</sup>** (fakirmsa@yahoo.com), M.G. Mostafa<sup>1</sup>, H. Seal<sup>2</sup>, and A.K.M.A. Prodhan<sup>1</sup>

**<sup>1</sup>Department of Crop Botany and <sup>2</sup>Department of Agricultural Chemistry,  
Bangladesh Agricultural University, Mymensingh, Bangladesh**

Cassava tubers (roots) are rich in starch (25-30% FW), and leaves are rich in proteins (25-30% DW). Leaves and tubers are used as staple food, feed, and raw materials in industries. In cassava, all plant parts are rich in cyanogens, which produce hydrogen cyanide (HCN) (100-300 mg kg<sup>-1</sup> fresh tissue) during post-harvest processing. Lethal dose for human-being is only 50 mg HCN 50 kg<sup>-1</sup> of body weight. Processing techniques, such as chopping, boiling, and drying eliminate 99% of HCN. Hence, cassava leaves and tubers are used after processing. Cassava matures in 6-8 months after planting stem cutting. Cassava is a drought- and heat-tolerant crop. It produces much greater fresh tuber yield (50-70 t ha<sup>-1</sup>, 40% moisture vs 6-10 t ha<sup>-1</sup> in rice and wheat 12-14% moisture) in poor soils where most other crops fail. Further, cassava can be grown in hill slope, roadside, homestead, borders (ails) of rice fields. All these qualities make cassava a climate-resilient crop. Fresh tubers are boiled and eaten like sweet potato and cooked like potato. Processed fresh leaves are an excellent source of protein-rich vegetable and compost. Flour and starch extracted from cassava tubers are used as an alternative to wheat flour. Flours and pellets are widely used as animal food. Fresh tubers were sliced into particular size and boiled at 100 °C and were served with salt. Fresh tubers were sliced and sun-dried to desirable moisture content. Dried tubers were milled into flour. For starch extraction, chopped fresh tubers were blended with adequate water, and the filtrate was decanted to starch. The starch obtained was then dried in the sun and was ready for use. *Chapati*, cake, pie, etc. were prepared using 10-40% of cassava flour mixed with wheat flour. These cassava-based baking products were equally liked by most people. It appears that cassava could be a potential supplementary food for rice and wheat in Bangladesh.

## Indiscriminate Use of Chemicals in Fruits and Their Effects on Human Health

M.A. Rahim (marahim1956@yahoo.com)

Department of Horticulture, Bangladesh Agricultural University,  
Mymensingh 2202, Bangladesh

Fruits are highly nutritious and form a key food commodity in the human consumption. Fruits are highly perishable due to their low shelf-life. These food commodities are contaminated with toxic and health hazardous chemicals. Chemicals such as calcium carbide and higher doses of ethylene and oxytocin are used for artificial ripening of fruits and for increasing their size. The use of edible coating, like chitosan, Aloe vera gel, non-chemical ripening processes, non-chemical process of extending shelf-life of different fruits, has also been suggested as per research findings in BAU-GPC. Residual limit of different chemicals for safe fruits has been standardized. Awareness of consumers for these toxic chemicals has also been suggested. Moreover, formalin, also used for extending the shelf-life of fruits, also causes several health problems. Calcium carbide, more commonly known as masala, is a carcinogenic agent and banned under the PFA Rules. In Bangladesh, people have been consuming toxic fruits ripened using calcium carbide, a hazardous chemical. This poses great health risks to consumers. Calcium carbide has cancer-causing properties and causes neurological disorders. It can result in tingling sensation and peripheral neuropathy. A significant number of pregnant women consume fruits ripened with carbide, resulting in delivery of babies with abnormalities. Moreover, the widespread use of formalin in Bangladesh and elsewhere in the world in the preservation of fruits is posing a threat to public health. The chemical used as a solution in water makes fruits such as mangoes attractive. The use of non-edible colouring and toxic-coating materials in fruits for extending their shelf-life and attractiveness also causes serious health hazardous in Bangladesh. This paper mainly focuses on the indiscriminate use of chemicals and their effects on health. Moreover, results of research conducted by the postgraduate students at the BAU-GPC were also discussed, e.g. issues like producing attractive, chemical-free safe fruits for the consumers. The results suggest the use of edible coating like chitosan, Aloe vera gel, non-chemical ripening processes, non-chemical process of extending shelf-life of different fruits. Residual limit of different chemicals for safe fruits is standardized. Awareness of consumers for these toxic chemicals is also suggested.

**Acknowledgement:** The author acknowledges the logistic support provided by the BAU-GPC, Mymensingh.

## Providing Safe Drinking-water Sources in Rural Bangladesh

Sonia N. Aziz<sup>1</sup> (aziz@moravian.edu ) and K.M.S. Aziz<sup>2</sup>

<sup>1</sup>Moravian College, 210 Comenius Hall, Bethlehem, PA 18018, USA and

<sup>2</sup>Bangladesh Academy of Sciences, Agargaon, Sher-e-Bangla Nagar,  
Dhaka, Bangladesh

Widespread arsenic contamination of groundwater in Bangladesh places the health of millions of Bangladeshis in jeopardy while pathogen-contaminated surface water serves as an alternative. Water sources without high levels of arsenic, or pathogen contamination are scarce, affecting peoples' time available for work and other activities when they have to seek safe water to drink. Limited information and heavy constraints on resources may preclude people in developing countries from taking protective actions. The focus of this paper is to report on factors affecting protective actions taken by rural Bangladeshi villagers. Results show that people respond to information campaigns by taking protective actions, furthermore parents take more protective actions than individuals without children, and water procurers walk an average of 44 minutes one way to procure safe drinking-water, although arsenic-contaminated water is available from nearby tubewells. The factors affecting protective actions are taken in conjunction with a stated preferences survey asking villagers their willingness to pay for a more convenient water source. The data can be used for supporting any public-health mitigation policy, including biotechnology efforts for tubewell-head mitigation, to make convenient but arsenic-contaminated tubewells viable again as a water source.

## Salinity-tolerant Rice Variety for Combating Climatic Disaster and Assuring Food Security

Mirza Mofazzal Islam (mirza\_islam@yahoo.com), Shamsun Nahar Begum, and Md. Nazmul Hoque

Plant Breeding Division, Bangladesh Institute of Nuclear Agriculture,  
Mymensingh 2202, Bangladesh

**Background:** More than one million hectares of rice land in Bangladesh are affected by salinity, including 53% of the coastal areas, due to global warming and recent climate change. Further degradation will cause detrimental effect on food security. The use of salt-tolerant varieties is the most economical and effective way of increasing crop production on saline soils. Salt tolerance trait is complex genetically and physiologically. Biotechnology with conventional breeding can facilitate the development of salt-tolerant varieties. **Objective:** The main objective of the research was to develop salt-tolerant rice varieties to enhance rice production for food security and improve livelihood of the farm community. **Methods and materials:** Sixty rice germplasm were evaluated for salinity tolerance in hydroponic system at the seedling and reproductive stages using the IRRI standard protocol. SSR markers also showed salinity tolerance in tested line PBRC-37. Evaluation of salt tolerant line 'PBRC-37' was done in the Boro season of 2011-2012 in the coastal saline areas of Satkhira, Bagerhat, and Khulna by the Seed Certification Agency (SCA). **Results:** Of the 60 rice germplasm, 24 were selected based on salt tolerance. At the seedling stage screening, nine lines were found to be tolerant at the EC level of 12 dS/m while 15 lines were moderately tolerant. Of the elected 24 germplasm, three strains, viz. Pokkali, PBRC-37 (salt-tolerant variety, Binadhan-10), and FL-478 were found to be salt-tolerant, four lines as moderately tolerant among 24 tested entries at the EC level of 6-12 dS/m. Binadhan-10 gave a higher yield (5.6 t/ha) under salt stress (10-12 dS/m) than the released salt-tolerant variety Binadhan-8. **Discussion:** Binadhan-10, a new salt-tolerant rice variety, can tolerate up to 12 dS/m of salinity which can be cultivated in 40-50% of fallow lands in both Boro (dry season) and Aman (wet season) seasons. The farmers can get yield of 5.5-6.50 t/ha in saline land and 7.5-8.5 t/h in non-saline land. Farmers can increase their income per hectare by Tk 45,000 by cultivating Binadhan-10 other than the salt-susceptible variety. **Conclusion:** Binadhan-10 can be cultivated in a large part of saline-prone 13 coastal districts, and more 4-5 million tonnes of additional rice can be produced a year. Dissemination of this variety in a large-scale would enable farmers to get higher productivity, ensure food security, and improve livelihood.

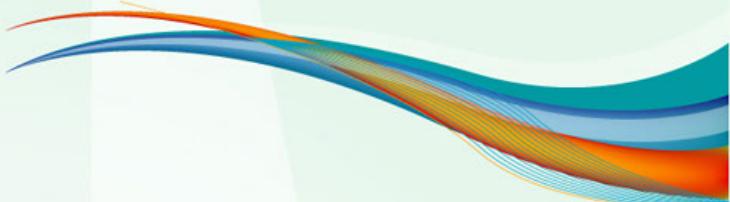
**Acknowledgement:** The support of PIU-BARC: NATP Phase-1 and International Rice Research Institute is acknowledged.

## Bioprocess Development for Eco-friendly Microbial Products and Its Impacts on Bio-industry Establishment in Bangladesh

Md. Mozammel Hoq (mhoq@univdhaka.edu)

Department of Microbiology, University of Dhaka,  
Ramna, Dhaka 1000, Bangladesh

**Introduction:** The biotechnology industry can spark industrial development in Bangladesh not only due to her major dependency on agricultural productivities but prior and judicial commercial exploitation of microbial processes both genetically and culturally on local cheap raw materials could deliver agro-industrial products, such as biofertilizer, biopesticides, bio-energy, including industrial enzymes (on use in textiles and garments, leather processing, and poultry feed formulations), bio-pharmaceuticals, vaccines, etc. profitably, with impact on saving foreign currency, cleaner environments, food security, reduction of GHG, and development of critical manpower. **Methods and materials:** Indigenous microorganisms were obtained through rationale screening for specific product synthesis capability with high titre; strain was improved by genetic manipulation and optimized for cultivation conditions on cheap agro-industrial raw materials, at both lab scale and pilot-scale level bioreactors. The recovery and purification process developed are simple based on the technical uses in the commercial sectors. **Results:** We have worked on the development of bioprocesses for production and technical applications of eco-friendly industrial enzymes and biopesticides based on indigenous microorganisms. The enzymes include proteases customized for tanneries (soaking enzyme, unhairing enzymes, bating enzymes), cellulase-free xylanases from thermophilic fungi for softening of low-quality jute fibres (fibres with high content of xylanase, pectin, and lignin) processing and in poultry feeds, cellulases from *Trichoderma viridiae* for biopolishing and keratinase from *Bacillus* spp. for conversion of poultry waste feathers to feed protein. Our microbial products, along with the imported enzymes, were tested for their respective technical applications at laboratory and pro-type pilot scale level, which demonstrated comparable performance. Furthermore, *Bacillus thuringiensis* biopesticide has been developed, and a large-scale production of Bt-endotoxin on cheap local substrates was done and applied in the field to control caterpillar-like pests with brinjal, cauliflower, and cabbage. The factors affecting the economic feasibility and sustainability of the processes worked in establishing bio-industries in Bangladesh will be discussed. **Conclusion:** The results will be a useful basis for commercial applications and development of bio-industries in Bangladesh.



## Engineering Ultra-sharp Nanostructures for Highly Selective Biological, Chemical and Agricultural Sensing, Pollution Monitoring and Control

M. Saiful Islam (sislam@ucdavis.edu)

Northern California Nanotechnology Center and Department of Electrical and Computer Engineering, University of California-Davis, California 95616 USA

Recent progress in nanotechnology has enabled the realization of engineered material structures with unrivaled dimensions in which quantum confinement, high surface to volume ratio and entwined nanoscale metal/semiconductor building blocks are woven together in a highly-integrated fashion to explore innovative applications. Ultra-sharp and thin nanostructures with distinct material-dependent properties exhibit important functionalities in devices, including gas ionization sensors, field emission devices, ion-mobility spectrometry, electrostatic precipitators, and biological, chemical and agricultural sensors. We will present examples of applications of engineered semiconductor nanowires in ultra-selective and high-performance sensors and discuss their importance in chemical and biological detection; diagnosing medical symptoms of certain diseases; monitoring and controlling agricultural and industrial green house gas emission, indoor air quality of homes, public places, manufacturing plants, automotive emission, waste disposal, and treatment plants. We will also show how such devices can dramatically reduce the design complexities of pollution monitoring and controlling systems. In the first example, a sensor based on charged particle beams will be presented, for which the geometrical and surface properties of the constituent semiconductor and oxide nanotips are engineered with controlled introduction of metallic impurities to realize more than three orders of magnitude reduction in the electric-field strength for gas ionization. We demonstrate that nanoscale pristine tips can be controllably decorated with atomic metal impurities to enhance the electron tunneling properties of a field ionization gas sensor under extremely low bias voltage. An advantageous combination of field enhancement on nanoscale tips, surface states introduced by defects along with controlled impurities and bandgap widening through quantum confinement contributes to such lowering of ionization voltages. These structures belong to a new class of devices that capitalizes on the notion that nanostructures offer great potential to be rationally tailored in a myriad of useful ways for accurate fingerprinting a broad range of biological and chemical analytes with ultra-high selectivity. In the second example, we will demonstrate biochemical sensors enabled by ion-sensitive field effect transistors with integrated nanowires. Increased surface area of these simple devices exhibits an order of magnitude larger gate capacitance, helps achieve pH sensitivity close to the theoretical value, and offers a strong potential to overcome the miniaturization and integration challenges of chem-bio sensors.

**Acknowledgments** The work was partially supported by the National Science Foundation and The Center for Information Technology Research in the Interest of Society (CITRIS).

## Diarrhoeal Disease Epidemiology and Ecology: Changing Climate and Concerns

Munirul Alam<sup>1</sup>, Anwarul Iqbal<sup>1</sup>, Abdus Sadique<sup>1</sup>, Marzia Sultana<sup>1</sup>, Fatema-tuj Johura<sup>1</sup>, Shah M. Rashed<sup>1</sup>, Tarequl Islam<sup>1</sup>, Kabir U. Ahmed<sup>1</sup>, Mahamudur Rashid<sup>1</sup>, M. Saiful Islam<sup>1</sup>, Shirajum Monira<sup>1</sup>, Sucharit B. Neogi<sup>1</sup>, Nur A. Hasan<sup>3</sup>, Anwar Huq<sup>3</sup>, David A. Sack<sup>4</sup>, R. Bradley Sack<sup>4</sup>, and R.R. Colwell<sup>3,4</sup>

<sup>1</sup>icDDR,b, Mohakhali, Dhaka 1212, Bangladesh, <sup>2</sup>National Institute of Infectious Diseases, Tokyo, Japan, <sup>3</sup>University of Maryland, USA, and <sup>4</sup>Bloomberg School of Public Health, Johns Hopkins University, USA

Diarrhoeal diseases, namely cholera, cause millions of morbidity and mortality worldwide, mostly in developing countries where the sources of pure drinking-water are scarce. Although diarrhoea has been well-established as a seasonal disease with its annual recurrence in defined time periods in many endemic areas, the patterns of infections in Bangladesh differ regionally in terms of both seasonality and number of annual peaks, as opposed to single annual peaks occurring in most endemic areas of the world. For example, in Dhaka, the capital city, and Matlab, a rural area 50 km apart, recurrent diarrhoea occurs twice in the form of two distinct peak patterns: the first peak of the year occurs during spring months, March-May, and the second peak occurs in the fall during September-November. As a result, diarrhoeal disease research and intervention in Bangladesh have primarily progressed focusing on the unique bimodal mode of transmission in Dhaka and Matlab, although of special interest is climate and related physico-chemical and biological factors that drive the seasonal outbreak peaks occurring world-wide, including Bangladesh. Data obtained from routine environmental and clinical surveillance carried out biweekly/monthly for more than a decade on the epidemiology and ecology of *Vibrio cholerae* involving regional hydro-climatology, plankton abundance, bacterial community dynamics in water, microbiology and molecular biology of the bacterium generated an overwhelming amount of information, including strong evidence of regional climate link to outbreaks of cholera and that *V. cholerae* thrives and modulates its life-cycle in the estuarine ecosystem of Bay of Bengal, which is well-known as the home of Asiatic cholera. Our molecular microbiological data (including DNA sequencing of targeted genes and comparative genomics) also reveal that *V. cholerae*, an estuarine bacterium, which is evolving in endemic areas by laterally acquiring virulence and related genes, can differ spatio-temporally and cause more severe disease by transmitting worldwide, adapt and evolve independently in diverse aquatic ecosystems. Finally, our meta-genomics data reveal the mysterious world of microbes showing how *V. cholerae* interacting with the community in the ecosystem can initiate seasonal epidemics of cholera and how the changing climate and the consequent burden of seasonal and off-season diarrhoea can affect human-beings by predisposing them to multidrug-resistant bacteria.

## **Role of 7SK snRNA in HIV1 Replication: Future Drug Designing**

**Shamsun Nahar Khan<sup>1,2</sup>** (nahar305@yahoo.com)

<sup>1</sup>Molecular and Cellular Biology Department, Harvard University, USA and

<sup>2</sup>Department of Pharmacy, East West University, Plot A/2, Jahurul Islam City,  
Aftabnagar, Dhaka 1212, Bangladesh

RNA plays crucial roles in the pathogenesis of many diseases and has gained a lot of interest as a tool for functional genomics. It is equally important as a promising therapeutic approach for the treatment of various diseases, e.g. HIV-1 infection, diabetes, etc. In the present study, we focused on the 7SK small nuclear RNA (snRNA), which is abundant with 331-nucleotide. It has one of the important functions as a transcriptional regulator during the elongation phase of HIV-1 virus. Immunodeficiency virus (HIV) exploits host's cellular proteins during its replicative cycle and latent infection. The positive transcription elongation factor b (P-TEFb) is a key cellular transcription factor critical for these viral processes. 7SK RNA binds to HEXIM1 regulatory domain and promotes the binding of the HEXIM C-terminal domain to cyclin T1/T2 of P-TEFb. P-TEFb shows little CTD kinase activity during its sequestering with 7SK and HEXIM1; it indicates that 7SK snRNA, in collaboration with HEXIM1, functions as an inhibitory factor of P-TEFb. Successive viral replication requires recruitment of P-TEFb by HIV-1 TAT protein for the completion of the viral RNA transcription process. Thus, one of the burning hypotheses is 7SK snRNA and HIV-1 protein Tat interaction. The study reports the evidence of strong binding interaction of 7SK snRNA and HIV-1 protein Tat with promising data of 2D NMR spectroscopic studies and ITC (isothermal calorimetric analysis) and future direction of drug designing.

## Progress in Generating Bio-engineered Crop Plants for Sustainable Agriculture

**Lutful Hassan** (lutfulhassan@yahoo.co.uk), G.S. Jahan, R. Gain, and S.N. Islam

Biotechnology and Genetic Engineering Laboratory, Department of Genetics and Plant Breeding, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh

We attempted genetic transformation of the local varieties of *Brassica campestris* and *B. napus* using *Agrobacterium*-mediated transformation. *Agrobacterium rhizogenes* strain LBA 9402 was used for the production of hairy roots. For co-transformation experiments, the strain LBA 9402 with the binary vector pBIN19 containing the p35S GUS INT gene was used. For plant regeneration, 0.5 mm sections of root material were excised and treated with a liquid callus-inducing medium (C23Y) for three days. These were then placed on N5 medium with antibiotics. The GUS staining was carried out according to Jefferson *et al.* (1987). A. *tumefaciens* strains: (i) GV3101 with the vir plasmid pMP90 and (ii) the strain C58C1 ATHV with the vir plasmid pTiBo542, a strain similar to EHA101, were used. The selectable marker gene, *nptII* (*neomycin phosphotransferase*) was used. The reporter gene  $\beta$  Glucuronidase (GUS) under control of the Ubi and the 35S-Promotor and with an Intron was used. Stem segments proved to be the best explant. Shoot regeneration in *A. rhizogenes* transformation experiments was not successful. Regeneration from *A. tumefaciens*-mediated transformation proved to be successful. Insertion of salt-tolerant genes (*AtNHX1*) from *Arabidopsis thaliana* in the popular varieties of *Brassica* genotypes is in progress. For sustainable agricultural practices, the transformed rapeseed varieties will be available for the farmers of the coastal wetland of Bangladesh.

## Harnessing Heterosis for Growth through Intergeneric Hybrid: Molecular Cytogenetic Studies on Avian Hybrid Sterility

Fhamida Binte Islam<sup>1</sup>, Md. Bazlur Rahman Mollah<sup>2</sup> (mbrmollah.ps@bau.edu.bd ), and Yoichi Matsuda<sup>1</sup>

<sup>1</sup>Department of Applied Molecular Bioscience, Nagoya University, Japan, <sup>2</sup>Poultry Biotechnology and Genomics Laboratory, Department of Poultry Science, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh

Harnessing heterosis for growth and reproductive traits is a major target area for improving productivity in livestock and poultry. Intergeneric and/or interspecific hybrids are often used for improving productivity in livestock and poultry. However, hybrids between different species are often inviable or, if they live, they are sterile. The use of sterile hybrids (mule) in human civilization has a long history because mule animals often perform better than their parents in respect of growth, endurance, and draught power. To date, the molecular mechanism of hybrid sterility in avian species is poorly understood. To understand the molecular mechanism of hybrid sterility in mule duck, an intergeneric sterile hybrid between domestic duck (*Anas platyrhynchos*) and muscovy duck (*Cairina moschata*), we performed histological and cytogenetic characterization of the sterility by chromosomal morphology, meiotic chromosome configuration, histology of testicular cross sections, and apoptosis of germ cells. We also performed cross species chromosome painting, fluorescence in situ hybridization (FISH) mapping of functional genes, telomeric repeats, and molecular cloning of repetitive DNA. Karyotypic analysis revealed that there is a morphological difference in chromosome 1, 2, and Z and size difference in chromosome 5 between two species. Meiotic chromosome configuration revealed high frequency of primary spermatocytes with highly-condensed chromosomes at late pachytene to first meiotic metaphase (MI). The spermatocytes with meiotic abnormality were consequently eliminated by apoptosis, resulting in the absence of post-meiotic cells after MI stage. These results suggest that the failure of meiotic chromosome pairing in primary spermatocytes by mutually incompatible difference in the chromosome morphology, which is followed by meiotic arrest at MI, might be a main cause of male sterility in mule ducks. FISH analysis of novel centromeric repeats indicates that the difference in the centromeric repeats in chromosome 1 and 2 between t *A. platyrhynchos* and *C. moschata* might be the cause of the failure of chromosome pairing in primary spermatocytes. The findings of these studies in mule duck will help improve our knowledge on the mechanism of sterility in avian species and assist in the production of mule duck for better meat production.

## Genomics of Immune-Response to Vaccines against Enteric Infections

Partha P. Majumder

National Institute of Biomedical Genomics, Kalyani, India

Differences in immunological response among recipients of vaccines against infectious diseases are determined by both their genetic differences and environmental factors. Enteric infections, such as typhoid and cholera, are a major public-health burden in many regions of the world. With the goal of understanding the role of genomic factors in the determination of protection to vaccinees, as measured by immunological response, conferred by typhoid and cholera vaccines, we have conducted two large studies in India. **Typhoid:** Significant associations of response with SNPs in seven genes (*DEFB1*, *TLR1*, *IL1RL1*, *CTLA4*, *MAPK8*, *CD86*, and *IL17D*) were discovered and cross-validated. Overall, (a) immune response to polysaccharide antigens is qualitatively different from that to protein antigens and (b) polymorphisms in genes involved in polysaccharide (LPS) recognition, signal transduction, inhibition of T-cell proliferation, pro-inflammatory signaling, and eventual production of antimicrobial peptides are associated with antibody response to the LPS vaccine for typhoid. **Cholera:** Significant associations of four SNPs and haplotypes in three genes (*MARCO*, *TNFAIP3*, and *CXCL12*) with response were discovered and validated. *CXCL12* is a neutrophil and lymphocyte chemoattractant that is upregulated in response to *V. cholerae* infection. LPS in the vaccine possibly provides signals that mimic those of the live bacterium. *TNFAIP3* promotes intestinal epithelial barrier integrity and provides tight junction protein regulation; possible requirements for adequate response to the vaccine. LPS is a potent activator of innate immune responses and a ligand of MARCO. Variants in *MARCO* have been found to be associated with LPS response but not with high vibriocidal titre level.

## Change, Exchange, and Community Interactions in Emergence and Evolution of *Vibrio cholerae*

**Nur A. Hasan<sup>1</sup>, Seon Young Choi<sup>1</sup>, Munirul Alam<sup>2</sup>, M. Mozammel Hoq<sup>3</sup>, Anwar Huq<sup>1</sup>, and Rita R. Colwell<sup>1,4,5</sup>**

<sup>1</sup>Maryland Pathogen Research Institute, University of Maryland, College Park, MD, USA;

<sup>2</sup>icddr,b, Mohakhali, Dhaka 1212, Bangladesh, <sup>3</sup>Department of Microbiology, University of Dhaka, Ramna, Dhaka 1000, Bangladesh, <sup>4</sup>Johns Hopkins Bloomberg School of Public Health, Baltimore, MD, USA, and <sup>5</sup>Center of Bioinformatics and Computational Biology, University of Maryland, College Park, MD, USA

For centuries, cholera has been one of the most feared diseases, and *Vibrio cholerae*, a water-borne pathogen, is the causative agent of this severe watery diarrhoeal disease. It is the only enteric pathogen that has the potential to produce pandemic disease. Many impressive advances have been made in the past few decades on understanding the biology, mechanism of pathogenesis, ecology, and epidemiology of *V. cholerae* and of the disease cholera. Despite all these advances, the global incidence of cholera is increasing steadily. Since *V. cholerae* is autochthonous to the aquatic environment, it is not possible to eradicate the disease but prevention is an achievable goal. To this end, we employed a holistic approach for acquisition of knowledge useful for lowering the burden of disease. A combination of microbiological and molecular methods, along with high-throughput genomics and metagenomic analyses, dissecting the micro niche of the bacterium to elucidate interaction within microbial populations in the environment and human, and investigating the genomic evolution of the bacterium to catalogue virulence markers and track their dissemination within and between toxigenic, non-toxigenic, and archived reference isolates. Investigating the distribution of a pool of virulence genetic markers, among 794 clinical and environmental *V. cholerae* samples, we demonstrated that the distribution of certain virulence genes is similar among most *V. cholerae* O1, O139 and non-O1/O139 whereas the distribution of the major virulence markers markedly differed among toxigenic and non-toxigenic strains. Metagenomic analysis demonstrated that aquatic environments harbour genetically diverse and complex microbial communities, including primary members of both freshwater and coastal ecosystems. The abundance and diversity of the microbial community, including *Vibrio* population, exhibited a complex seasonal distribution, and specific consortia among the natural community, perhaps stimulated as a result of positive selection, might be crucial to trigger *V. cholerae* to overcome the natural competition and emerge in epidemic proportion. Comparative genome analysis of over 150 *V. cholerae* and its phylogenetic near neighbours demonstrated various genomic events, including inter- and intra-chromosomal re-arrangements, gene absence and acquisitions, chromosomal integration of phages, and other mobile genetic elements (MGEs), all of which are the important driving forces in the evolution of species *Vibrio*. The study also demonstrated that *V. cholerae* harbours an open pan genome, and the current pandemic is caused by strains belonging to a single phyletic line, diversified mainly via horizontal gene transfer from a dynamic *Vibrio* gene reservoir in the natural environment.

## Genera Represented by Single Species in the Angiospermic Flora of Bangladesh and Their Conservation

A.K.M. Golam Sarwar (drsarwar@bau.edu.bd)

Department of Crop Botany, Bangladesh Agricultural University,  
Mymensingh 2202, Bangladesh

**Introduction:** Genera represented by a single species are one of the important groups of plants that are interesting not only in floristics study but also in phytogeography and phylogenetic studies. They need critical taxonomic evaluation and documentation and also deserve special attention from the conservation point of view. **Methods and materials:** In this paper, the list of genera represented by a single species in Bangladesh has been reviewed and updated. **Results and discussion:** In total, 777 genera of 160 families of flowering plants are represented by a single species in the flora of the country. Of these, 13 genera are monotypic in the strict sense, i.e. represented worldwide only by the 'Type species'. The number of genera represented by a single species should be increased as the inventory of plants of Bangladesh is far beyond the completion. Four of the genera represented by a single species are endemic in Bangladesh. Fifty-one families are represented only by a single species. Hitherto, 21 taxa have not been collected again after their original collection, and 82 taxa are threatened as per the IUCN Red list category. **Conclusion:** The genera represented by a single species occupy ca. 22% of the angiospermic taxa of Bangladesh, a challenging and stimulating group that merits considerable further attention of both taxonomists and conservation biologists.



## International Conference on Biotechnology

*on application of Biotechnology  
in addressing the development needs of Bangladesh*

25-26 May, 2013

**ABSTRACTS OF POSTERS**

## Isolation of Lead and Chromium-resistant Bacteria from the Buriganga and Shitalkhya Rivers in Bangladesh

Tahmina Shammi<sup>1</sup> and Sangita Ahmed<sup>2</sup> (sangita@du.ac.bd)

Department of Microbiology, University of Dhaka  
Ramna, Dhaka 1000, Bangladesh

**Background:** Water pollution in Dhaka city has been a serious problem for years, and increasing urbanization is worsening the situation. In recent years, the rivers Buriganga, Turag, Shitalakhy, and Balu, which provide the main water supply in Dhaka, have been loaded with heavy metals, such as lead, chromium, and mercury, to such an extent that it is imposing a great threat to public health. However, removal of heavy metals from wastewater mainly involves expensive chemical technology, and as a cheaper alternative, bioremediation of heavy metals using metal-resistant microorganisms is growing attention globally. **Objective:** The present study was aimed to isolate and identify lead and chromium-resistant bacteria from the Buriganga and the Shitalkhya and investigate their potential for removing these two harmful heavy metals from water. **Methods and materials:** In total, 61 isolates resistant to 50 ppm Pb and Cr were obtained from the two sites (Buriganga and Shitalkhya). **Results:** Morphological and biochemical analyses revealed that, of the total isolates, 34 (56%) were *Bacillus* sp., 18 (30%) were *Staphylococcus* sp., and 9 (15%) were Gram-negative rods. Initially, the *Bacillus* sp. isolates were characterized for their resistance to increasing concentrations of Pb and Cr. It was observed that 75% of these isolates were resistant to 1,000 ppm Pb and Cr. A further study identified one *Bacillus* sp. isolate with minimum inhibitory concentration value of 1,500 ppm for Pb and 3,000 ppm for Cr. This heavy metal-resistant bacterium could be useful for the bioremediation of Pb and Cr from heavily-contaminated waters of the two major rivers of Bangladesh. **Conclusion:** Further characterization of this isolate is necessary to reveal the detail mechanism of heavy metal resistance, which would enable the development of a bacterial bioremediation-based method of the water-treatment system to remove heavy metals from the polluted aquatic bodies.

**Acknowledgment:** The study was funded by the Ministry of Science and Technology, Government of Bangladesh.

## Structural Analysis of Interacting Domains of RNA-dependent RNA Polymerase Complex in Nipah Virus

Md. Jahangir Alam, **Saleha Sultana Rima** (rima.sust@yahoo.com), Bijenro Khadka, Abdullah Zubaer, Simrika Thapa, and Mohammed Lashkar

<sup>1</sup>Department of Genetic Engineering and Biotechnology, Shahjalal University of Science and Technology, Sylhet, Bangladesh and <sup>2</sup>Swapnojaatra Bioresearch Laboratory, DataSoft Systems BD Ltd., Dhaka, Bangladesh

**Background:** Nipah virus is a non-segmented negative stranded RNA virus. The genome is packaged by the nucleoprotein (N) within nucleocapsid that recruits the polymerase protein by connecting the phosphoprotein-large (polymerase) (P-L) interaction through nucleoprotein-phosphoprotein (N-P) interaction forming the RNA-dependent RNA polymerase (RdRp) complex having importance in RNA replication and transcription. **Objective:** The study was conducted to understand the structural aspects of the complex. **Methods and materials:** Interaction between interacting domains of Nipah virus RdRp complex was identified through disorder analysis and homology modelling based on secondary structure and template identification. **Results:** The evidence that proteins of RdRp complex are highly disordered under native conditions is expected to be invaluable for future structural studies of whole proteins. Homology modelling and protein-protein docking studies of four domains forming two interactions (alpha MoRE-PXD and PMD-domain I) were performed with certain bioinformatic tools which facilitate the execution of weak interaction. **Conclusion:** The results suggest the alpha MoRE-PXD interaction as a valuable drug target as it is made/broken repeatedly to give access to polymerase.

**Acknowledgement:** The study was supported by the Swapnojaatra Bioresearch Laboratory, DataSoft System BD Ltd. for three months under authorization of the Department of Genetic Engineering and Biotechnology, Shahjalal University of Science and Technology.

## Pyramiding of Genes for Resistance to Bacterial Blight in Rice

M. Monirul Islam, Shahanaz Sultana, **M. Enamul Hoque** (hoque2003@yahoo.com),  
and M. Shamsher Ali

Biotechnology Division, Bangladesh Rice Research Institute,  
Gazipur, Bangladesh

**Background:** Rice is the staple food crop in Bangladesh. Bangladesh has made tremendous progress in food production, especially in rice production, due to the introduction and adoption of high-yielding varieties and associated technologies. However, BRRI dhan28 and BRRI dhan29, the most popular two rice varieties of the Boro season, have shown susceptibility to bacterial blight (BB). The existence and emergence of new pathotypes has resulted in the breakdown of resistance in varieties possessing a single resistance gene. Durable resistance to BB can be achieved by pyramiding of several resistance genes into a single genetic background. **Objective:** The study was conducted for pyramiding of genes (*xa13* and *Xa21*), which confer BB resistance in the background of BRRI dhan28 and BRRI dhan29 for developing high-yielding cultivars with resistance/higher levels of tolerance to BB that limit the rice production in Bangladesh. **Methods and materials:** Two backcrosses were made between the recurrent parents (BRRI dhan28 and BRRI dhan29) with the donor (IRBB60) containing two BB-resistant genes (*xa13* and *Xa21*). Plants having both the BB-resistant genes (*xa13* and *Xa21*) were confirmed by molecular screening. Two tightly-linked molecular markers (marker Xa13 for *xa13* and marker Xa-21 for *Xa21*) were used for molecular screening. Plants having both the BB-resistant genes were used for further backcrossing up to backcross generation five (BC5). Molecular screening was done on BC5F1 progenies of BRRI dhan28/IRBB60 and BRRI dhan29/IRBB60 cross. **Results:** Three plants possessing both the resistant genes (*xa13* and *Xa21*) were identified using molecular marker from each of the cross. The identified pyramided lines having the *xa13* and *Xa21* genes showed resistance to BB after their exposure to the BB pathogen (*Xanthomonas oryzae* pv *oryzae*). **Conclusion:** These BB-resistant pyramided lines can further enhance the rice production in Bangladesh.

**Acknowledgement:** The study was supported by a research grant of BARC.

## Development of Abiotic Stress-tolerant Rice (*Oryza sativa L.*) to Attain Food Security with Environmental Safety

Shamsul H. Prodhan<sup>1,2</sup> (shamsulhp@yahoo.com), Tsuyoshi Motohashi<sup>2</sup>, and Atsushi Komamine<sup>3</sup>

<sup>1</sup>Department of Genetic Engineering and Biotechnology, Shahjalal University of Science and Technology, Sylhet 3114, Bangladesh, <sup>2</sup>Tokyo University of Agriculture, 1737 Funako, Atsugi 243-0034, Japan, and <sup>3</sup>Research Institute of Evolutionary Biology, 2-4-28 Kamiyoga, Setagaya, Tokyo, 158-0098, Japan

Crops are affected by many types of abiotic stresses, such as salt, drought, and freezing. Among these stresses, salinity is one of the major factors limiting crop production in the world. Development of salt-tolerant rice is, therefore, a significant research objective in agricultural biotechnology. An important requirement for crop and environmental improvements is the introduction of new genetic material into the cultivated lines through the introduction of single/multiple genes and through genetic engineering. In this research, we were able to establish an efficient *Agrobacterium*-mediated transformation method in both Japonica (cv. Nipponbare) and Indica rice (*Oryza sativa L.* cvs. BR-5, Kasalath, and Kataribhog). The *Agrobacterium tumefaciens* strain-E-HA101—harbouring the binary vector pIG121Hm/Km/katE was used for the transformation of Nipponbare, Kasalath, and BR-5 cultivars, and pIG121Hm/Km/GUS were used for the transformation of Kataribhog rice. The vector contains the  $\beta$ -glucuronidase (GUS) gene as a reporter gene, hygromycin resistance (HPT) and kanamycin resistance gene (NPTII) as a selection agent, and katE the desired gene in the T-DNA region. Total genomic DNA was isolated from the transgenic plants; the presence of the transgenes was confirmed by PCR and Southern blot analysis; and expression of the katE gene was detected by RT-PCR. The catalase activity of the transgenic T1 plants was about 1.5 to 2.5-fold higher than that of the wild rice plants. Landrace Indica rice Kasalath was able to grow for more than 20 days in the presence of 250 mM sodium chloride and produced seeds for more than three months in the presence of 100 mM sodium chloride. On the contrary, the nontransgenic rice plants could not survive even for 10 days in the presence of 50 mM sodium chloride. The introduction of a single gene significantly improved the salt tolerance of Indica and Japonica rice. In this research work, we report the application of genetic engineering in staple food rice for the improvement of stress areas to bring food security with safe environment.

## Construction of an Antiviral Vector Targeting Mungbean Yellow Mosaic Virus

Sujay Kumar Bhajan, Sonia Khan Sony, Rita Sarah Borna, Mohammad Nurul Islam,  
M.I. Hoque<sup>4</sup>, and R.H. Sarker (rhsarker2000@yahoo.co.uk)

Plant Breeding and Biotechnology Laboratory, Department of Botany,  
University of Dhaka, Dhaka 1000, Bangladesh

Mungbean yellow mosaic virus (MYMV) is responsible for the yellow mosaic disease and causes maximum yield loss of mungbean in Bangladesh. The yield potential of mungbean can be achieved through the development of yellow mosaic virus-resistant mungbean. In this context, MYMV coat protein (CP) and silencing suppressor gene (AC2)-specific primer sets were designed from the conserved regions after alignment of the available respective gene sequences in the NCBI database. Part of the MYMV CP (~750 bp) and AC2 (~450 bp) gene was PCR amplified using CP and AC2-specific primers from the template DNA of the MYMV-infected leaf samples. Sequence analysis of PCR-amplified DNA confirmed the presence of the MYMV CP and AC2 gene. The amplified MYMV CP gene (~750 bp) was cloned into pBI121 vector replacing the GUS gene by using BamHI and SacI restriction recognition sites in antisense orientation resulting asCP/pBI121 construct. The AC2 gene was then sub-cloned using BamHI restriction recognition sites in antisense orientation into the asCP/pBI121 vector resulting asAC2-CP/pBI121 construct driven by CaMV35S promoter. Cloned construct asAC2-CP/pBI121 was confirmed through restriction digestion, PCR amplification and sequencing. The sequence analysis of the CP and AC2 gene showed 98% sequence similarities with MYMV-BD strain during NCBI BLAST search. Finally, the cloned construct asAC2-CP/pBI121 was transferred to Agrobacterium tumefaciens strain LBA4404 with an aim to exploit this in transforming local mungbean varieties.

**Acknowledgement:** This investigation is supported by the U.S. Department of Agriculture through a project.

## RID Domain of *Vibrio cholerae* MARTX Toxin Alludes Bi-functional Activity with Three Sub-domains in Structure

Abdullah Zubaer<sup>1,2,3</sup>, Simrika Thapa<sup>1,2,3</sup>, **Bijendra Khadka**<sup>1,2,3</sup>  
 (bijendra.khadka@swapnojaatra.org), and Mohammad Lashkar<sup>1</sup>

<sup>1</sup>Swapnojaatra Bioresearch Laboratory, DataSoft Systems, Dhaka 1215, Bangladesh,  
<sup>2</sup>Department of Genetic Engineering and Biotechnology, Shahjalal University of Science and Technology, Sylhet, Bangladesh, and <sup>3</sup>CANSi Research Institute, Sylhet, Bangladesh

**Background:** A Rho-GTPase inactivation domain (RID) is regarded as a domain of the multi-functional, autoprocessing Rtx toxin RtxA from *Vibrio cholerae*. The RID domain causes actin depolymerization and rounding of host cells through inactivation of the Rho family of small GTPases.

**Objective:** The study was carried out to elucidate the functional and structural organization of the RID domain to understand its possible role in Rho GTPase inactivation and subsequently develop an antimicrobial drug for the treatment of cholera. **Materials and methods:** The amino acid sequence of the *V. cholerae* Rtx RID domain (residues 2552-3099) was used for performing a homology search using PSI-BLAST and HHpred. To identify the domain architecture, the domain database servers, such as Pfam, CDD, and SMART, were used. The three-dimensional structure of the entire RID domain has been modelled using the I-TASSER server and MODELLER. **Results:** Based on sequence analysis of RID to identify the functional sites, it was found that the domain could be divided into three sub-domains (R1, R2, and R3) regarding homology inference, structure, and functional prediction. The study disclosed that the first sub-domain (R1) of RID is responsible for membrane localization being homologous to the C1 domain of *Pasteurella multocida* toxin, and the remaining two sub-domains have distinct catalytic activity. Based on homology inference, it was proposed that the second domain (R2) of RID functions similar to the adenylyl cyclase domain of *Bordetella pertussis* toxin and oedema factor of *Bacillus anthracis* for targeting the Rho-GTPase regulation. The third domain (R3) is distantly related to the peptidase C58 family, thus opposing the previously-referred activity of RID only as protease. Furthermore, using ab initio and template-based methods, a reliable three-dimensional structure of the RID domain was constructed. **Conclusion:** The study reports the presence of three different sub-domains within the Rtx RID domain, each contributing to the overall function of RID. These results provide organizational, functional, mechanistic and structural insights into important toxin proteins involved in bacterial pathogenesis.

**Acknowledgement:** The authors are thankful and deeply indebted to DataSoft System Bangladesh Limited for their infrastructural and financial support to conduct the study.

## Appropriate Bating Enzyme for Use in Bangladesh Tanneries

**Md. Arafat Al Mamun<sup>1</sup>, Md. Ruhul Amin<sup>2</sup>, Syed Rubel Hossain<sup>2</sup>, Md. Nahinur Rahman Akand<sup>3</sup>, Sabur Ahmed<sup>2</sup>, Fatema -Tuj-Zohra<sup>2</sup>, Shakila Nargis Khan<sup>3</sup>, and Md. Mozammel Hoq<sup>3</sup> (mmzhoq@gmail.com)**

<sup>1</sup>Pilot Plant Research Laboratory, Centre for Advanced Research in Sciences,

<sup>2</sup>Institute of Leather Technology, and <sup>3</sup>Department of Microbiology,

University of Dhaka, Ramna, Dhaka 1000, Bangladesh

**Background:** Bating is one of the main operations in tanneries for the removal of non-structural proteins with hides and skins, which render the leather produce soft, pliable, and permeable. Enzymatic bathing offers an eco-friendly process replacing many hazardous chemicals and quality products. In view of this, a *Bacillus* bacterium (*Bacillus* MAS-6) was developed for the production of high-level alkaline serine protease, capable of removing nonstructural proteins in bathing operation of cow hides successfully. **Methods and materials:** *Bacillus* MAS-6, developed by mutation, was cultivated in laboratory scale and pilot plant level for the production of alkaline protease in both defined and complex soy meal media at 37°C and pH 7.5. To make a cheaper medium for protease production, glucose was replaced by molasses in the soy meal medium. Thus, the soy meal medium with molasses was used successfully for the large-scale production in bioreactor (7.5 l and 120 l) with *Bacillus* MAS-6 under 30% dissolved oxygen concentration controlled by cascading mode maintained by both agitation and aeration automatically. Both *Bacillus* MAS-6 and commercial enzymes (OROPON K, bate enzyme) were tested for bathing of cow hides under comparable conditions at theproto-type application facility in the laboratory of the Institute of Leather Engineering and Technology (ILET), Hazaribagh, Dhaka. The bated products were evaluated using standard qualitative tests at the ISO laboratory, ILET. **Results and discussion:** The modified soy meal medium, which was developed with molasses, supported both growth and enzyme production satisfactorily in both shake flask and bioreactor cultivations. The organism showed a maximum growth of  $8.2 \times 10^9$  CFU/mL after 28 hours, with a productivity of 19,000 U/L/hour of the protease in bioreactor while it was 6,773 U/L/hour in the shake culture. This protease enzyme hydrolyzed albumin, globulin, and elastin efficiently but did not degrade collagen, the composition of skin itself. This result is important in leather processing because the non-structural proteins were removed selectively without affecting the collagen of hides. The qualitative tests (thumb test, bubble test, pH and phenolphthalein test) of bated pelts demonstrated acceptable results within the standard quality limits. In addition, tensile strength, percentage of elongation, stitch tear strength, tongue tear strength, grain crack strength, flexing endurance, softness, and water vapor permeability tests of crushed leather finally produced by the two enzymes also showed satisfactory results. **Conclusion:** The results of both production and application of the enzyme from *Bacillus* MAS-6 may successfully be used for the commercial production of bathing enzyme in the tanneries of Bangladesh. It will not only reduce the dependency on foreign currency but will reduce the use of harsh chemicals and will promote the development of biotechnology industry in Bangladesh.

**Acknowledgement:** The research project was partially supported with financial grant by the Ministry of Education, Government of Bangladesh.

## **Anthropogenic Stress in Rodents: Emergence of Hantavirus and Altered Host-parasite Interaction**

**Abdullah Mahmud-Al-Rafat** ([rafat\\_0907@yahoo.com](mailto:rafat_0907@yahoo.com)) and **Mahbub-E-Sobhani**

Biotechnology and Genetic Engineering Discipline,  
Khulna University, Khulna 9208, Bangladesh

The emergence and re-emergence of infectious diseases got considerable attention in the last few decades during which three-fourths of emerging human infectious diseases were caused by zoonotic pathogens. Hantavirus is one of the deadly zoonotic viruses harboured by rodents. Hantavirus produces severe fatal infection in human-beings while normal host rodents show no signs of diseases. The emergence of Hantavirus could be attributed to anthropogenic stress. Several anthropogenic stressors, such as deforestation, urbanization, noise, introduced species, electromagnetic fields, light pollution, etc., contribute to the alteration of the endocrine balance in rodents. These are also responsible for immune, nervous and physiological alteration. Chronic anthropogenic stressed rodents have elevated levels of glucocorticoid, a major immunosuppressive hormone. Interactions among the endocrine, nervous and immune systems determine the outcome of host-parasite interactions. Glucocorticoid is responsible for reducing the resistance of wild animals against viruses and increases the tolerance of animals to harbour the virus in a minimal range without causing any disease. By activating the organ-specific regulatory mechanism, it also plays a major role in host-parasite interaction. Understanding the viral persistence in rodents would provide insights into future therapy of such infectious diseases.

## Fermentation of Municipal Solid Wastes Using Potential Bacterial Isolates and Production of Commercially-important Protease

Abul Kalam Azad (dakazad-btc@sust.edu), Kamrul Islam, Akhikun Nahar,

Md. Mahbub Hasan, Md. Faisal Azim, Md. Saddam Hossain, and Mohammad Rejaur Rahman

Department of Genetic Engineering and Biotechnology, Shahjalal University of Science and Technology, Sylhet 3114, Bangladesh

Over 10,000 tonnes of municipal solid wastes (MSWs) generated from six major cities of Bangladesh are generally dumped either to fill low land or are spread here and there without proper management causing severe environmental pollution. The organic material of MSWs can be used as substrate for fermentation to produce commercially-valuable enzymes, biofertilizer, biofuel, biopesticides, etc. To produce extracellular bacterial proteases using MSWs, 12 proteolytic bacterial isolates from the MSWs were screened, and three isolates-*Bacillus* sp., *Pseudomonas* sp., and *Serratia marcescens*-were selected based on their morphological, cultural and biochemical characteristics. With a basal medium, a maximum level of protease was produced from the *Bacillus*, *Pseudomonas*, and *S. marcescens* isolates after 48, 42, and 24 hours of fermentation respectively. The conditions for protease production by the three isolates were optimized using the basal medium. With the basal medium in which carbon and nitrogen sources were replaced by cellulosic and proteinous materials of MSWs, a maximum level of protease was obtained from the *Bacillus* and *Pseudomonas* isolates after 60 hours of fermentation. However, under the same conditions in this medium, *S. marcescens* produced a maximum level of protease after 48 hours. However, in bioreactor with the same MSW medium, *S. marcessens* produced almost 2-fold protease after 72 hours of fermentation compared to that at shake flask fermentation. Crude proteases produced from the three bacterial isolates using cellulosic and proteinous MSWs were stable at pH 6.0-8.0 and at temperatures between 25 °C and 40 °C. The protease from the *Bacillus* isolate was of metallo and cysteine type whereas the protease from the *Pseudomonas* isolate was of cysteine type, and the protease from the *S. marcescens* was of serine and cysteine type. The physical changes of chicken flesh treated with crude proteases were recorded and proteases from *Bacillus*, *Pseudomonas*, and *Serratia marcescens* degraded 20%, 45%, and 50% weight of the chicken flesh respectively. The results support the notion that the MSWs might be a suitable raw material for the production of bacterial protease.

## **Adaptation of Peste des Petits Ruminants Virus of Bangladesh in Tissue Culture: A Step Towards Development of a New Vaccine Candidate**

**E.H. Chowdhury** (emdad001@yahoo.com), M.M. Rahman, J.A. Begum,  
A.R. Bhuiyan, M. Giasuddin, and M.R. Islam

Department of Pathology, Faculty of Veterinary Science,  
Bangladesh Agricultural University, Mymensingh 2202, Bangladesh

Peste des petits ruminants (PPR), an acute and highly-contagious viral disease of small ruminants, is endemic in Bangladesh and causes huge economic losses due to its high morbidity and mortality in Black Bengal goats. Post-mortem tissue samples were collected from a Black Bengal goat having a clinical history of PPR. Initially, samples were checked by reverse transcription-polymerase chain reaction (RT-PCR) for PPR virus. The samples found to be positive by RT-PCR were subjected for isolation in Vero cell culture and reconfirmed in Vero cell culture by RT-PCR. Twelve isolates of PPRV from different parts of Bangladesh were sequenced, and subsequently, phylogenetic and homology analyses of the sequence were carried out. It was found that PPRV circulating in Bangladesh for the last three years belong to the same lineage IV along with other Asian isolates, which indicate their common ancestry. The similarity of N and F genes among all the Bangladeshi isolates was 94.5-99.6% and 98.7-100% respectively. It was also found that percent identity and divergence of Bangladeshi isolates with other isolates from distant geographical regions varied to a greater extent in their N gene sequences than F gene sequences. The N protein of recent lineage IV isolates of Bangladesh and other countries have two unique amino acid substitutions, which are not evident in earlier lineage IV isolates. Three of the genetically best matched PPRV isolates were selected for attenuation in Vero cell. PPR virus grown in Vero cell produced characteristic CPE. Infected cover slip cultures stained with H&E showed vaculation, rounding, and aggregation of cells. Numerous acidophilic intra-cytoplasmic and intra-nuclear inclusion bodies were also observed, indicating that virus is replicating well in Vero cell. So far, 35 serial passages were completed. The study is in progress. A 65-70 passage will completely attenuate the virus.

**Acknowledgement:** The authors acknowledge SPGR, PIU, BARC, and International Atomic Energy Agency, Vienna, for fund and instrument support.

## Prevalence of Multidrug-resistant Zoonotic *Salmonella* and *Enterobacter* spp. in Poultry of Bangladesh

Shuvro Prokash Nandi, Munawar Sultana, and M. Anwar Hossain (hossaina@du.ac.bd)

Department of Microbiology, University of Dhaka, Dhaka 1000, Bangladesh

**Background:** Zoonotic bacterial pathogens are directly transmissible from animals to humans. Food-producing animals can serve as reservoirs of antibiotic-resistant organisms (and their genes). This is particularly an important issue in Bangladesh due to the concurrent use of antibiotics in the poultry feed, irrational use of antimicrobials in the treatment of diseases, and unhygienic practices in farming and marketing. **Objective:** The study was undertaken to investigate the prevalence of multidrug-resistant zoonotic *Salmonella* and *Enterobacter* spp. in poultry of Bangladesh. **Methods and materials:** The study investigated zoonotic pathogenic bacteria involved in poultry based on their serotypes, resistotypes, and genotype. The study analyzed the drug-resistant profile against 15 major groups of common antibiotics and molecular characterization of pathogenic bacteria from poultry samples in Bangladesh. **Results:** The prevalence of *Salmonella* spp. and *Enterobacter* spp. was 21.1% and 3.6% respectively. A large number of the isolates were plasmid-free and resistant to 10-15 groups of antibiotics. Some of these multidrug-resistant (MDR) isolates also depicted typical phenotype for the presence of extended spectrum beta-lactamase (ESBL). Antibiotic resistance-encoding genes, such as vanA for vancomycin resistance and blatem, blactx-M, and blashv for ESBL production, were also detected from the isolates. Genetic fingerprinting methods and corresponding sequencing of 16S rRNA genes of the representative isolates detected a close similarity to *Salmonella Typhimurium*, *S. Enteritidis*, *S. Paratyphi*, *Enterobacter cloacae*, *E. hormaechei*, and *E. cancerogenous* in the poultry samples, indicating a significant zoonotic hazard. **Conclusion:** The prevalence of MDR-ESBL isolates in poultry, along with their zoonotic relevance, implicates the necessity of hygienic practices and continued monitoring of poultry and poultry products in Bangladesh.

## Antibiotic Pollution and Occurrence of Multidrug-resistant Bacteria in the Environment of Bangladesh

Munawar Sultana (munawar@du.ac.bd), Nazratan Naeem, Fahmida Sultana, Shuvro Prokash Nandi, and M. Anwar Hossain

Department of Microbiology, University of Dhaka, Ramna,  
Dhaka 1000, Bangladesh

**Background:** Clinical waste water (CWW) possesses the risks of spreading antibiotic-resistant bacteria in the environment causing R-gene pool pollution. In Bangladesh, liquid discharges from hospitals are directly released into the municipal sewage system and pollute the surroundings water bodies/rivers. **Materials and methods:** The CWW effluents (hospital outflow) from the Dhaka Medical College Hospital (DMCH) and Sir Salimullah Medical College Hospital (SSMCH) and urine samples from UTI (urinary tract infection) patients of the Enam Medical College Hospital, Savar, were collected for the isolation and genotyping of multidrug-resistant (MDR) bacteria. The antimicrobial susceptibility patterns of the isolates were determined, and resistance (R) genes of  $\beta$ -lactamases (bla), extended spectrum  $\beta$ -lactamases (ESBL), quinolones (qnr), and vancomycin (vanA) were detected by PCR and sequencing. The antibiotic concentrations of the SSMCH effluents directly released into the river Buriganga were measured by liquid chromatography-mass spectroscopy (LC-MS). **Results:** Sixty of 271 CWW and UTI isolates were characterized as MDR-ESBL producers. About 32, 30, and 26 times higher ciprofloxacin, cefixime, and both ciprofloxacin and cefixime-resistant bacterial counts were observed in the CWW over the control samples. The isolates showed varying ranges of plasmids and even absence of plasmid under experimental condition; however, the isolates were resistant to 10-15 groups of antibiotics. Genotypically, the isolates were categorized into three classes of ESBLs (TEM, SHV, and CTX-M), with an abundance of CTX-M ESBL genotypes and three classes of quinolone-resistant (qnrA, qnrB, and qnrS) genes. The isolates belonged to the genus *Escherichia*, *Klebsiella*, *Enterobacter*, *Proteus*, and *Pseudomonas*, with a high abundance of *Escherichia* spp. A high concentration of ciprofloxacin was detected from the direct outflow of the SSMCH entering the river Buriganga. **Conclusion:** CWW exposed to the surrounding freshwater bodies is polluting the environment with MDR-ESBL bacteria that are potent to harbour and disseminate resistance properties. Therefore, the indiscriminate and unrestricted use of antibiotics and the release of CWW through clinical outflow can pose a great threat to the environment.

## Isolation and Molecular Detection of Foot-and-Mouth Disease Viruses of the 2012-2013 Outbreak, Circulating in Bangladesh

Mohammed Alimul Islam<sup>1</sup>, **Mohammed Ehsanul Haque<sup>1</sup>** (faysalvet@yahoo.com),  
Mohammed Golam Moktadir<sup>1</sup>, Mohammad Muradul Islam Chowdhury<sup>1</sup>,  
Mohammad Mostofa Kamal<sup>1</sup>, Mohammad Anwar Hossain<sup>2</sup>,  
Yohei Kurosaki<sup>3</sup>, and Jiro Yasuda<sup>3</sup>

<sup>1</sup>Department of Microbiology and Hygiene, Bangladesh Agricultural University,  
Mymensingh 2202, Bangladesh, <sup>2</sup>Department of Microbiology, University of Dhaka,  
Dhaka 1000, Bangladesh, and <sup>3</sup>Department of Emerging Infectious Diseases,  
Institute of Tropical Medicine, Nagasaki University, Japan

A study was designed to know the status of circulating foot-and-mouth disease viruses (FMDVs) in the cattle population of Bangladesh. In total, 180 samples (tongue epithelium-140 and tissue from interdigital spaces-40) were collected from 10 different districts of Bangladesh during 2012-2013. An attempt was made to isolate the FMDVs from the field samples in BHK-21 cell line and confirmation of the viruses by RT-PCR using type-specific primers against four types (O, A, Asia1, and C) and one subtype A22. Of the 180 samples, 50 were positive for virus isolation and 120 by RT-PCR. Of the 50 isolates of FMDV, only three types (type A, O, and Asia-1) were confirmed by RT-PCR. Of the three confirmed types of FMDV detected by RT-PCR, the predominant types were type O (70, 58.33%), followed by type A (35, 29.17%), and Asia-1 (15, 12.5%) respectively. The cytopathic effects were in the BHK-21 cells rounding, cluster formation, granulation, individualization, and elongation of the cells manifested by any of the three types of FMDV investigated in this study. Of the three types of FMDV, the Asia-1 and A type viruses revealed faster CPE (with 26-36 hours of post-infection) compared to that of O type virus (36-60 hours). The higher rate of virus detection by RT-PCR indicates that the live virus is not always required for type and sub-type determination of FMDV from the field samples. The results clearly indicate that there are three types of FMDV circulating either as a single or multiple types of outbreak of diseases in the cattle population of Bangladesh. Further investigation relating to the origin of the viruses needs to be made.

## **Future Prospect of Bio-energy Use in the Tea Industry of Bangladesh Through Jatropha Plantation**

**Md. Asraful Alam (sharif7bd@yahoo.com)**

School of Life Science and Biotechnology, Dalian University of Technology,  
Dalian 116024, China

Bangladesh is a fast-developing country and rated to be one of the few countries that have the fastest rate of economic growth, which will lead to it attaining the status of a 'middle-income' nation within the next decade. Hence, the energy needs are not only to be sustained but also to be greatly enhanced. The global growth of fossil-based oil is causing a drastic environmental damage, which will have a serious impact on countries such as Bangladesh. With the advancement in the agricultural sector to use farm equipment, heavy vehicles, and machineries, the demand for diesel fuel has increased drastically. All these elements lead our policy-planners to look for other viable alternatives. Compared to other bio-fuels, 'Jatropha curcas' is favourably identified as a potential source of bio-diesel. Both primary and secondary data show that there is approximately 8,600 hectares (ha) of land available for Jatropha cultivation in the tea estates all over Bangladesh. The Bangladesh tea industry currently produces 56-60 million kg of tea per year and consumes 3,200 tonnes of diesel at an import cost of Tk 120 million, which reflects Tk 2.14 per kg of the production costs whereas these costs could be potentially reduced to Tk 1.00 per kg, thereby making a potential saving of Tk 1.14 per kg overall, making a saving of Tk 63.84 million on account of Jatropha-based bio-diesel with the consequent impact on selling prices favourable to buyers. The geographical position of the Tea Estates in Bangladesh, the availability of unused land, manpower, infrastructure, and the willingness of the Tea Estate owners to cultivate Jatropha demonstrate that the prospect of not only energy self-sufficiency in the tea industry through Jatropha plantation is a very viable proposition but also an avenue to reduce the Government's import bills on account of diesel and make savings in our foreign currency spending.

**Acknowledgment:** The author is grateful to the Green Gold Platinum Ltd. to provide necessary fund and support for the study.

## Cold or Heat-Shock Positively Modulates Oxidative Protection of Salt- and Drought-stressed Mustard (*Brassica rapa* L.) Seedlings

Mohammad Anwar Hossain<sup>1,2</sup> (hossainma@gmail.com), Mohammad Golam Mostofa<sup>2,3</sup>, and Masayuki Fujita<sup>2</sup>

<sup>1</sup>Department of Genetics and Plant Breeding, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh, <sup>2</sup>Laboratory of Plant Stress Responses, Department of Applied Biological Science, Faculty of Agriculture, Kagawa University, Miki-cho, Kita-gun, Kagawa 761-0795, Japan, and <sup>3</sup>Department of Biochemistry, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur 1706, Bangladesh

A large number of studies have shown the existence of cross-stress tolerance in plants but the exact physiological and biochemical mechanism(s) is/are poorly understood. Importantly, a large number of recent studies in plants demonstrated that the simultaneous induction of both reactive-oxygen species (ROS) and methylglyoxal (MG) detoxification system are essential in inducing abiotic oxidative-stress tolerance. Therefore, to explore the possible involvement of antioxidative and glyoxalase systems in cross-stress tolerance, we conducted a laboratory experiment with mustard (*Brassica rapa* L.) seedling by applying a short-term cold-shock (6 °C, 5.5 h) or heat-shock (42 °C, 5 h) and test their cross-adaptation to salt and drought stresses. Seven-day-old seedlings were subjected to salt (150 mM NaCl, 48 h) and drought stress (induced by 20% PEG, 48 h) with or without cold- or heat-shock. The results showed that both salt and drought stresses abruptly increased the hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and lipid peroxidation (malondialdehyde, MDA) levels. Drought stress treatment resulted in a significant increase in ascorbate (AsA) content. The reduced glutathione (GSH) and oxidized glutathione (GSSG) contents increased in response to both salt and drought stresses; however, the GSH/GSSG ratio decreased significantly in response to drought stress. Salt stress treatment resulted in a significant increase in ascorbate peroxidase (APX), monodehydroascorbate reductase (MDHAR), glutathione reductase (GR), glutathione S-trasnsferase (GST), and glyoxalase (Gly I) activities whereas catalase (CAT) and glyoxalase (Gly II) activities decreased. In contrast, drought stress treatment resulted in a significant increase in MDHAR, dehydroascorbate reductase (DHAR), glutathione peroxidase (GPX), and Gly I activities whereas APX, CAT, and Gly II activities decreased. Surprisingly, cold- or heat-pretreated salt and drought-stressed seedlings maintained higher AsA, GSH contents and GSH/GSSG ratio, higher activities of APX, DHAR, GR, GST, GPX, CAT, Gly I and II, and lower the levels of GSSG, H<sub>2</sub>O<sub>2</sub>, and MDA compared to the control and in most cases seedlings subjected to salt and drought stress without cold- or heat-shock. The findings showed that retention of the imprint of previous stress exposure (short-term cold- or heat-shock) rendered the plant more tolerant to salt- and drought-induced oxidative stress by modulating antioxidative and glyoxalase systems.

## Development and Application of a Multiplex PCR Assay for the Detection of Pathogenic *Vibrio cholerae*, *Vibrio parahaemolyticus*, and *Vibrio vulnificus* Species

Sucharit Basu Neogi<sup>1,4</sup> (sbneogi@icddrb.org), Farhana Akther<sup>1</sup>, Rubén Lara<sup>2,3</sup>, Shinji Yamasaki<sup>4</sup>, and Munirul Alam<sup>1</sup>

<sup>1</sup>Centre for Food and Waterborne Diseases, icddr,b, Dhaka 1212, Bangladesh,

<sup>2</sup>Leibniz Zentrum für Marine Tropenökologie, 28359 Bremen, Germany,

<sup>3</sup>Argentine Institute of Oceanography, Bahia Blanca 8000, Argentina, and

<sup>4</sup>Osaka Prefecture University, Osaka 598-8531, Japan

**Background:** *Vibrio cholerae*, *V. parahaemolyticus*, and *V. vulnificus*, the three most important bacterial species linked to diseases in humans, shrimps, and fish, can co-exist in the aquatic environment, diseased animals, and seafood. The rapid, precise, and simultaneous detection of these pathogens is very important in preventing outbreaks of diseases. **Objective:** The study was undertaken to develop an effective multiplex PCR assay for simultaneous detection of the three most important *Vibrio* species, particularly from environmental samples. **Methods and materials:** Species-specific PCR primers were designed, and the primer concentration and PCR conditions were optimized. The assay was validated with 260 *Vibrio* strains, including 190 *V. cholerae*, 5 *V. vulnificus*, and 15 *V. parahaemolyticus*, 20 other *Vibrio* species, and 17 other bacterial species. This rapid method was applied for the detection of the pathogenic species from estuarine waters of Vietnam and Bangladesh and marine waters of the Atlantic in parallel to biochemical screening and 16S rRNA gene sequencing. **Results:** The assay showed high fidelity in detecting the target bacteria and also showed good efficiency in detecting co-existing target species in the water samples spiked with the target species. The detection limit of all the target species was 10 cells per PCR tube. Identification by this simple assay was more precise than the laborious biochemical screening and expensive 16S rRNA gene sequencing techniques. Of the *Vibrio* isolates (n=462) from the Bangladesh coast, a high abundance of *V. cholerae* (15-50%) and *V. parahaemolyticus* (12-38%) was observed while, in Vietnam mangrove, the dominating isolates (n=82) were *V. parahaemolyticus* (66%) and *V. vulnificus* (13%). The Atlantic isolates (n=215) were devoid of the target species, indicating that the pathogenic *Vibrio* species might have adapted themselves in the estuarine environment nearby human activities rather than the oceanic environment. **Conclusion:** The specificity and sensitivity of the multiplex PCR is 100% each and sufficient for the simultaneous detection of these potentially pathogenic *Vibrio* species in environmental samples. This simple and cost-effective method can be used for the rapid detection of the major pathogenic *Vibrio* species, which may aid in predicting and preventing outbreaks of diseases.

**Acknowledgement:** The study was supported by the Monbukagakusho scholarship scheme of the Ministry of Education, Culture, Sports, Science and Technology of Japan, the German Science Foundation (DFG/BMZ) (Grant No. LA 868/5-1), and the National Institutes of Health, USA (Grant No. 1 R01 A139129-01).

## Introduction of Efficient Indigenous Bt Biopesticide in Integrated Pest Management to Control Vegetable Pests in Bangladesh

**Md. Asaduzzaman Shishir, Md. Boduzzaman, M. Nahinur Rahman, Arafat Al Mamun, Alamgir Rahman, Mohammad Ilias, Shakila N. Khan, and Md. Mozammel Hoq**  
(mhoq@univdhaka.edu)

Department of Microbiology, University of Dhaka, Ramna, Dhaka 1000, Bangladesh

**Background:** *Bacillus thuringiensis* (Bt) biopesticides are very efficient to play important roles in crop protection by controlling specific pests and are, therefore, being used in the form of biopesticide and transgenic plants worldwide. Resistance of pests and food security are the burning issues regarding pesticides for which integrated pest management (IPM) is a solution, although locally-produced Bt biopesticide is not yet included as an important component in Bangladesh. **Objective:** The present work is aimed at the production and application of locally-developed potential *Bt* biopesticide in Bangladesh agriculture. **Methods and materials:** *Bt* was isolated from different eco-systems of Bangladesh by acetate selection-heat treatment- lecithinase production and characterized by haemolytic activity, plasmid profiling, endotoxin profiling, and toxin gene profiling. *Bt* isolates were classified into different biotypes and checked for the presence of *cry1*, *cry1A*, and *cry2A* genes by PCR identification. Bioassay was performed with the PCR-positive isolates against *Bactrocera cucurbitae*, *B. dorsalis*, and *B. tao spp.* A large-scale production of Bt biopesticide with potential strain was carried out in the Defatted Mustard Seed mill medium in the bioreactor. It was readily sprayed on cauliflower and cabbage farming under comparable conditions besides control and chemical insecticide currently in use. **Results and discussion:** In total, 303 *Bt* isolates obtained were, thus, classified into 16 biotypes, e.g. *Bt thuringiensis* (10%), *kurstaki* (14%), *indiana* (20%), etc. 12-15 kb plasmids were present in the isolates, and 28, 21, and 16 isolates contained *cry1*, *cry1A*, and *cry2A* genes respectively. Performing bioassay, indigenous *Bt* JSc1 was found to be highly toxic ( $LC_{50}=6.81$ ) against *B. cucurbitae*, and the biopesticide preparation protected 95% and 83% of cauliflower ( $n=50$ ) and cabbage ( $n=50$ ) respectively from the infestation and damage by the pests in the field whereas chemical pesticide protected 86% and 82% respectively. Thus, in controlling the pests with cauliflower and cabbage, *Bt* JSc1 biopesticide preparation was proved to be more efficient based on the results analyzed statistically. **Conclusion:** The promising results, thus, obtained with the indigenous *Bt* clearly suggest the prospect of production and use of biopesticide for controlling the vegetable pests successfully through IPM, thereby increasing the food security in Bangladesh.

**Acknowledgement:** The work is financially supported by the United State Department of Agriculture under the Biotech Agricultural Research Projects and partially by the Government of Bangladesh.

## Enhancing Salt Tolerance of Rice Cultivars by Transformation with Regulatory Protein-like Transcription Factors

**U.S. Mahzabin Amin**, Sudip Biswas, Rumana Sultana Tammi,  
Shanaz Parvin, Wasifa Kabir, and Zeba I. Seraj (zebai@univdhaka.edu)

Department of Biochemistry and Molecular Biology, University of Dhaka,  
Ramna, Dhaka 1000, Bangladesh

**Background:** Stress-related transcription factors (TFs) activate cascades of genes that act together in enhancing tolerance towards stresses. This property makes TFs an attractive target for the manipulation of abiotic stress tolerance. Soil salinity is one of the major abiotic stresses that influence the productivity of many important crops of Bangladesh. Two transcription factors-SNAC and HARDY-are of particular significance for abiotic stress tolerance. The plant-specific SNAC1 or stress-responsive NAC (NAM, ATAF1/2, and CUC2) transcription factors play diverse roles in plant development and stress responses. The *HARDY* gene belongs to the stress-related AP2/ERF (APETALA2/ethylene responsive element binding factors) super family of transcription factors and has been reported to improve the ratio of biomass production to the water used-known as water-use efficiency (WUE)-by reducing transpiration under drought stress and improving photosynthetic assimilation under salt stress. **Objective:** Attempts were, therefore, made to enhance the tolerance of rice by genetic transformation using these two transcription factors. **Methods and results:** *SNAC1* cDNA was isolated from the salt-tolerant rice landrace Pokkali and transformed into the local landrace Binnatoa. The *SNAC1*-positive transgenic plants were advanced to the T3 generation and their tolerance to 12 dS/m salt stress confirmed at the seedling stage. Two lines with significant tolerance were selected for further analysis. The *HARDY* coding region was isolated from *Arabidopsis thaliana* cDNA and again transformed into BA. Three selected transgenic plants are being analyzed. As BA is not a farmer-popular variety, the transgenes have to be backcrossed into farmer-popular cultivars. To bypass the tissue culture regeneration and the backcross steps, some farmer-popular varieties were also transformed with these genes using the In Planta transformation method by bypassing the tissue culture and regeneration steps in the laboratory. The status of the transgenes was confirmed by PCR analysis, resistance to hygromycin B and GUS detection, and positive plants were advanced to T1 generation. Bioinformatics analyses were performed to develop a gene network model to understand the downstream effect of these transcription factors. **Conclusion:** Salt-tolerant rice was successfully produced using transcription factors but needs to be functionally validated further.

**Acknowledgement:** The study was supported by the U.S. Department of Agriculture and the Bangladesh Academy of Sciences.

## Promoter Characterization of Two Salt Stress-related Genes in Tobacco Leaves and Rice Callus

**Sudip Biswas**, Md. Muntasir Ali, U.S. Mahzabin Amin, Taslima Haque, and Zeba I. Seraj (zebai@univdhaka.edu)

Department of Biochemistry and Molecular Biology, University of Dhaka,  
Ramna, Dhaka 1000, Bangladesh

**Introduction:** Crop production is greatly affected by abiotic stresses, such as drought, submergence, salinity, etc. Therefore, there is a dire need to develop abiotic stress-tolerant crops to cope with the situation, particularly rice, in the context of Bangladesh. One approach to produce stress-tolerant rice is by its transformation with genes known to confer tolerance. These genes, however, need to be driven by efficient promoters. Most commercial vectors used for transformation, however, contain the suboptimal constitutive promoter CaMV35S (Cauliflower mosaic virus35S) to drive the transgenes. Therefore, identification and characterization of both inducible and constitutive novel promoters are necessary for use in transformation to produce stress-tolerant rice. **Objective:** The present study involves characterization of the promoters of two stress-responsive genes: ascorbate peroxidase (APX) from rice and RD29A from arabidopsis. **Methods and results:** Two different fragments of the APX promoter (APX-large, 2000 bp) and APX-small, 1000 bp) were amplified from salt-tolerant rice variety (Pokkali) and cloned into the promoter-less binary vector (pHGWFS7.0) containing the GUS reporter gene. *A. tumefaciens* containing the cloned promoters from Pokkali: APX-large GUS, APX-small GUS, and CaMV GUS (positive control) were infected on tobacco leaves and rice callus to monitor their transient expression. Both qualitative and quantitative analyses denoted that the APX-large promoter showed higher expression compared to both APX-small and CaMV promoters in rice callus. However, in tobacco leaves, the CaMV promoter showed higher expression than the other two promoters. For the RD29A promoter (containing a number of abiotic stress-related cis-regulatory elements), a 592 bp region upstream of the rd29A gene has been cloned from *Arabidopsis thaliana* genomic DNA into a Gateway® compatible binary vector. The activity of the  $\beta$ -glucuronidase (GUS) gene will be assessed when driven by rd29A under salt-stress conditions, and subsequent comparison of expression patterns will be carried out using the CaMV35S promoter. **Discussion:** The goal is to determine the susceptibility of these two promoters for effective expression of transgenes in salinity-susceptible rice varieties.

**Acknowledgement:** The study was supported by the U.S. Department of Agriculture and the Bangladesh Academy of Sciences.

## **Secondary Metabolites from Endophytic Fungi**

Nilufar Nahar (nilufarnahar@yahoo.com)

Department of Chemistry, University of Dhaka,  
Ramna, Dhaka 1000, Bangladesh

The word endophyte means 'in the plant' and refers to all microorganisms that live in the intercellular spaces of stems, petioles, roots, and leaves of plants causing no apparent symptoms of disease. The host and the fungi are in harmony and can spend the whole time. Endophytic fungi spend the whole or parts of their life-cycle colonizing inter- and/or intra-cellularly inside the healthy tissues of the host plant. But, in stress condition, fungi can come out. Plant endophytic fungi are good sources of bioactive secondary metabolites than terrestrial plants. Isolation of small amount of bioactive compounds can give the lead to large-scale production by fermentation for industrial and pharmaceutical application and will be helpful for conservation of biodiversity. The flora of Bangladesh is rich in aromatic and medicinal plants, and their endophytic fungi were not studied before. To discover novel pharmaceutical candidates, several endophytic fungi were isolated from medicinal plants of *Terminila chebula* Retz, *Ocimum basilicum* L, *Aquilaria malaccensis* Lamk, and *Magnifera indica* L. Healthy single fungal strains isolated from these plants were sub-cultured in large scales and were extracted with ethyl acetate and fractionated by chromatographic techniques. From the ethyl acetate extract of selected fungal strains, several novel and known compounds, including terminatol, terminanone, secalonic acid A, secalonic acid D, ergosterol, derivative of 4, 7 dimethyl-1, 3-dioxa cyclohepta-2-one, coriloxin, derivative of coriloxin, cis-4-hydroxy-hexadec-6-enoic acid methyl ester, 1,7-dihydroxy-3-methoxyanthraquinone, and propyl p-methoxy phenyl ether 6-methoxy-7-O-(p-methoxyphenyl)-coumarin, were isolated. The structure of the isolated compounds was elucidated by spectroscopic techniques, i.e. UV, IR, 1D and 2D NMR, and mass studies.

## Determining the *In vitro* Responses from Hypocotyls Explants in Tomato under Red and Blue Light-emitting Diode Irradiation Source

Most Tahera Naznin ([tnaznin2009@yahoo.com](mailto:tnaznin2009@yahoo.com)) and Mark Lefsrud

Bioresource Engineering Department, McGill University, Canada

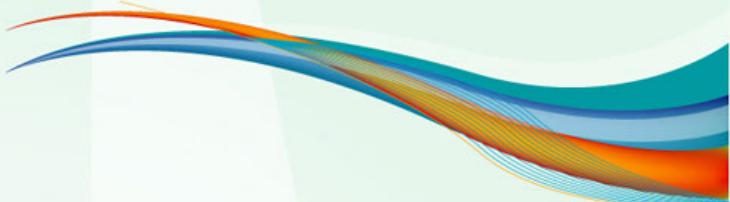
In micropropagation, fluorescent lamps are mostly used as a light source. Fluorescent lamps have peaks throughout the spectrum but lack emissions in the far red. However, specific wavelengths of light energy can be wasted when it is not used by the plant. Light-emitting diode (LED) lamps are anticipated to replace fluorescent lamps in most applications due to their reduced electricity consumption, improved quality of light, small size, durability, long-operating lifetime, relatively cool emitting surfaces, and the possibility for customization of the light spectrum. This study was conducted to explore the regeneration potential of tomato hypocotyls segments under LED and fluorescent lamps to examine whether the light source has any effects on plant tissue culture. Four light treatments: fluorescent lamp (control), red LED, blue LED, and 10:1 red and blue LED were used in the study. The LED array was controlled (current controlled) using a single channel controller to produce uniform intensity of specific wavelengths of light over the PAR spectrum. Specifications of the controller is a 24VDC, 2.0A maximum, 48 watt unit with current selected and displayed (0-1.92ADC), automatic voltage control. Surface sterilization of tomato seeds was done by soaking into the solution of 8% clorox (sodium hypochlorite) for 10 minutes, followed by three times rinsing with sterilized distilled water. Seeds were inoculated in test tubes containing MS medium and were transferred in a dark room for germination. Hypocotyl segments (1-2 cm) of 18-21-day old *in vitro* plants were excised under aseptic conditions. The excised explants were cultured in MS medium supplemented with 30 g of sucrose and 2 mg/L of BAP. The cultures were incubated under different light treatments in growth chamber (16/8 light/dark regime) having the same temperature for six weeks. Data were collected and evaluated in terms of shoots number and length. Among the four treatments of light, 10:1 red and blue LED was found to be superior in growth traits (number of shoots/explants and shoot length). The lowest significant growth was observed in control. This research will facilitate the improved selection of LEDs for plant tissue culture.

## Genetic Divergence, Induced Mutation, and Inter-relationships Among Yield Components of Mango (*Mangifera indica L.*)

D.A.N. Majumder, M.L. Hasan, and M.A. Rahim (marahim1956@yahoo.com)

Bangladesh Agricultural University,  
Mymensingh 2202, Bangladesh

**Introduction:** Mango is the king of fruits, and it grows in both tropics and subtropics. Bangladesh produces a wide range of mango varieties. Mutation breeding may help produce regular and good-quality varieties. **Objective:** The investigation was carried out to study the genetic diversity and induced mutation of mango. **Methods and materials:** The study was conducted at the Bangladesh Agricultural University, Mymensingh, during 2007-2009. Evaluation and morphological characterization of 60 mango genotypes were performed using different qualitative and quantitative characters. Variations among the genotypes in respect of plant, leaf, inflorescence, fruit, and stone characters were observed. **Results:** In all the traits, genotypic coefficient of variation (GCV) was always smaller than phenotypic coefficient of variation (PCV). A narrow difference between GCV and PCV and high heritability coupled with high genetic advance confirmed a least environmental effect on % flowering shoot, number of fruits per plants, and yield per plant, which offered a better scope for selection. Genetic diversity was assessed using 15 morphological characters. Based on multivariate (D2) analysis, the 60 mango genotypes were grouped into eight clusters. Principal component analysis revealed that the first nine axes accounted for 88.30% of the total variation, and the remaining six characters accounted for 11.70% of variation. Principal coordinate analysis showed that the highest inter-cluster distance (151.82) was between cluster II and cluster VIII, and the lowest (55.47) was between cluster VII and cluster VIII. Considering the univariate and multivariate study, 10 promising genotypes, viz. MI01, MI04, MI09, MI23, MI24, MI25, MI26, MI28, MI94, and MI95, were selected for a further breeding programme. Three enzymes, such as gulutamate oxaloacetate transminase (GOT), malate dehydrogenase (MDH), and peroxidase (PER), were used for investigating the genetic diversity of the 60 mango genotypes at the protein level. Eight zymotypes with GOT, 10 with MDH, and seven with PER were formed by 22, 39, and 12 bands respectively at different Rf values. Again, based on three isozymes banding patterns, the 60 genotypes were grouped into eight clusters. Compared to the three studied DNA extraction protocols of mango, such as SDS, CTAB, and water saturated ether (WSE) method with NaCl, it was found that the WSE method with NaCl had the highest value of average percentage (85.44%) in DNA content of the mango genotypes. RAPD analysis of the 60 mango genotypes was done to detect the genetic diversity at the DNA level. Of 40 primers, 11 decamer primers amplified 104 bands, of which 101 (97.12%) were polymorphic. Nei's (1973) highest genetic distance (0.883) was observed between MI01 and MI18. The mean genetic diversity among all the accessions was 0.323, and Shannon's information index was 0.489 across all loci. The UPGMA dendrogram, based on Nei's (1972) genetic distances, revealed that the 60 mango genotypes were grouped into seven clusters. Clustering of accessions, based on morphological characters, did not match with the clustering obtained from isozyme and RAPD analyses. It was evident from morphological, isozyme and RAPD analyses that there was no relationship between genetic divergence and geographical distribution of genotypes. From the correlation study between yield and yield contributing characters, plant height, % flowering



shoot, % perfect flower, and number of fruits per plant were found to be positive and significantly correlated with yield. Genotypic correlation coefficient was higher than phenotypic ones for most cases, indicating a strong inherent relationship among the characters. Path coefficient analysis showed that plant height, panicle per shoot, and % perfect flower had a maximum direct effect on yield per plant, followed by fruit weight at both genotypic and phenotypic levels. Induction of mutation on eight selected genotypes, derived from a morph-molecular diversity study, was performed applying three doses of radiation, such as 20 Gy, 30 Gy, and 40 Gy. Sixteen mutants of mango, derived from 20 Gy and 30 Gy radiation, were characterized by RAPD analysis. With primer OPA-10, the amplified bands OPA-10-675 bp, OPA-10-850 bp, OPA-10-875 bp, and OPA-10-925 bp; with primer OPB-18, the amplified bands OPB-18-458 bp, OPB-18-500 bp, OPB-18-825 bp, and OPB-18-950 bp; with primer OPC-12, the amplified bands OPC-12-1100 bp; and with primer OPC-13, the amplified bands OPC-13-350 bp, OPC-13-600 bp, and OPC-13-1500 bp were found in mutants but were not formed that of their parents. None of the five primers alone was able to detect all the 24 mutants. Conclusion: The findings suggest that the bands may reflect the existence of inter-allelic interactions in RAPD markers. For distinguishing the mango genotypes at the intra-species level, more RAPD primers may be included in future studies.

**Acknowledgement:** The authors acknowledge logistic support provided by the BAU-GPC, Bangladesh Agricultural University, Mymensingh.

## Emerging Pathogenic *Vibrio cholerae* non-O1/O139 with Reservoir Potential of Drug-resistance Genes

Mohammad Tarequl Islam<sup>1</sup> (tarequl@icddrb.org), Feroz Ahmed<sup>1</sup>, Afroza Akhter<sup>1</sup>, Abdus Sadique<sup>1</sup>, Shah M. Rashed<sup>1</sup>, Shahnewaj Bin Mnnan<sup>1</sup>, Zillur Rahman<sup>1</sup>, Sabita R. Rahman<sup>2</sup>, and Munirul Alam<sup>1</sup>

<sup>1</sup>Centre for Food and Waterborne Diseases, icddr,b, Mohakhali, Dhaka 1212, Bangladesh and <sup>2</sup>Department of Microbiology, University of Dhaka, Ramna, Dhaka 1000, Bangladesh

**Background:** Pathogenic strains of *Vibrio cholerae* non-O1/O139 are known to cause outbreaks of diarrhoea worldwide and serve as an important environmental reservoir for drug-resistance genes. SXT-related conjugative, self-transmissible elements, and different classes of Integrons are known to encode resistance in *V. cholerae* towards multiple commonly-used drugs. Thus, assessment of distribution and type of these elements in *V. cholerae* population is important to understand the dynamics of continuously evolving drug-resistance problem. **Objective:** The study was conducted to assess distribution and to characterize SXT element and integrons in potentially-virulent *V. cholerae* non-O1/O139 strains isolated from the rural coastal areas of Bangladesh using standard tools.

**Materials and methods:** Antimicrobial resistance profile of *V. cholerae* non-O1/O139 strains (n=200), isolated form environmental samples, carrying putative virulence genes (toxR, rtxA, rbcC, ompU, hlyA, hap, nanH) pre-confirmed by PCR, were assessed using the disc-diffusion method with seven commonly-used drugs. PCR was carried out to screen and characterize SXT element and for integrons. **Results:** Antimicrobial resistance profile revealed that 34% (n=67) of the tested strains were resistant to more than three drugs, including trimethoprim/sulphamethoxazole (SXT), furazolidone, tetracycline, and erythromycin (E). Of the multidrug-resistant strains, 87% (n=59) possessed the SXT element and 11% (n=7) contained class I integron (intI). Three strains of *V. cholerae* were positive for both SXT element and class I integron; 97% of the SXT element containing strains possessed integrase gene (SXTint), gene for sulphamethoxazole (sullI), trimethoprim (dfrA1), and streptomycin (strAB) resistance. All the class I integron-positive strains also possessed the 3'-CS end of the element and had different sizes of inserts ranging from 600 bp to 1.4 kb within the variable region, which can serve as a docking site for resistance genes. **Conclusion:** SXT and integron-mediated resistance and distribution data of the elements emphasize the continuous monitoring of drug-resistance profile and genetic markers to handle the threat posed by the continuous emergence of multidrug-resistant *V. cholerae* non-O1/O139 strains, which can serve as reservoirs of resistance properties to be disseminated to pandemic *V. cholerae* and other microbial populations involved in infectious diseases.

## **Computational Approach to Search for Plant Homologues of Human Heat-shock Protein**

Animesh Sarker and **AI Amin** (niloygenetics@gmail.com)

Department of Biotechnology and Genetic Engineering, Mawlana Bhasani Science and Technology University, Tangail, Bangladesh

A bioinformatics finding shows that various human heat-shock protein (HSP) homologues are frequent in plants. HSPs, which are exposed to high temperature or other stresses, have chaperone activity and belong to four conserved classes: HSP60, HSP70, HSP90, and HSP100. A bioinformatics blast search revealed that each of the classes contains a number of HSP plant homologues. The closest identified homologue of human HSP\_7C is a protein of unknown function (NCBI Accession XP\_002332067) from *Populus trichocarpa*. Comparative analysis of secondary and three-dimensional structure of this predicted protein confirms its functional relationship with the class of HSP70.

## **Voltage Generated from Mangrove Forest Sediment Microbial Fuel Cell Through Modification of Design and Components**

S.M. Monsur Musa, Sabiha Rahman, and D.I. Sharif

Biotechnology and Genetic Engineering Discipline,  
Khulna University, Khulna, Bangladesh

Electricity can be generated from naturally-occurring electro potential differences through the burial of an anode in sediment and the immersion of a cathode in the overlying body of water. Such a set-up is called sediment microbial fuel cell (SMFC) or benthic MFC. The mangrove forest sediment of the Sunderbans was used for constructing SMFC, and an open circuit voltage was measured over 40 days. Voltage was found to increase over 14 days after which it remained steady for about seven days and then declined. The highest voltage of 610 mV was obtained from a single SMFC on the 24th day. Comparative analysis using an earthen pot as a proton-exchange membrane showed the voltage to increase gradually over an initial period, with a highest increase to 100 mV. An analysis of different cathode materials showed that the highest voltage was obtained from carbon rod over carbon brush and stainless steel as a cathode material. Several SMFCs were added to make a series connection, which gave enough voltage (3.8 volts) to turn-on a LED bulb and a scientific calculator. The findings suggest that the soil sediment of the Sundarbans can be a potential source for further studies in the development of SMFC.

## Study on Underutilized Fruit Species of Coastal Areas in Bangladesh for Year-round and Off-season Fruit Production

A.K.M. Ashraful Alam<sup>1</sup> (kazalashraf@gmail.com), M.A. Rahim<sup>2</sup>  
(marahim1956@yahoo.com), and Md. Habibur Rahman<sup>2</sup>

<sup>1</sup>School of Agriculture and Rural Development, Bangladesh Open University, Gazipur 1705, Bangladesh and <sup>2</sup>Department of Horticulture, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh

The coastal areas of Bangladesh are divided into three regions, namely eastern, central, and western. According to the coastal zone policy of the Government of Bangladesh, 19 of the 64 districts are in the coastal zone, covering 147 sub-districts. The total number of people living in the coastal zone is 35.1 million that represent 28% of the total population of the country. The population density in the exposed coast is 482 persons per square kilometre compared to 1,012 for the interior coast. Fishing, agriculture, shrimp farming, salt farming, and tourism are the main economic activities in the coastal area. The fruit-production scenario of Bangladesh in 2009-2010 was 45.25 lakh tonnes in the land of 1.46 lakh hectares. The deficit of fruit production was 9.60 lakh tonnes. For the physiological balance of human body, the typical Bangladeshis consume 76 g/day/capita, which are behind the requirement of 85 g. Most people of the country cannot afford to buy even the average requirement of fruits due to their unavailability and high price. The consequence of this event is, therefore, widespread malnutrition throughout the country. The use of an appropriate amount of fruits can play a vital role in minimizing the malnutrition situation in the country. In the coastal areas of Bangladesh, mainly Khulna, Bhola, Bagerhat, Borguna, Satkhira, Patuakhali, and Barishal regions, many underused fruits, such as river ebony (*Diospyros peregrina*), velvet apple (*Diospyros discolor*), cowa (*Garcinia cowa*), sapota (*Manilkara zapota*), golden apple (*Spondias dulcis*), wax jambu (*Syzygium samarangense*), monkey jack (*Artocarpus lakoocha*), bullock's heart (*Annona reticulata*), elephant apple (*Dillenia indica*), wood apple (*Feronia limonia*), star gooseberry (*Phyllanthus acidus*), and aonla (*Phyllanthus emblica*), are growing round the year without much care largely in the homestead, fallow and forest areas and at the roadside and railway lines. These fruits are well-adapted to the local climate, available in off-season, and highly nutritious, contribute to the reduction of poverty, protection against natural disasters, balancing the coastal ecosystem, and household food security of the rural people, and play a significant role in herbal medicine.

## **Application of PCR Technology in Agriculture in Bangladesh**

Mahmudur Rahman (Mahmud\_reaz@yahoo.com)

Pharmacy Discipline, Khulna University,  
Khulna, Bangladesh

The polymerase chain reaction (PCR) technique is an in vitro and rapid method for amplifying nucleic acids. Since its inception, this technique and its modified methods, such as loop-mediated isothermal amplification (LAMP), nucleic acid sequencing-based amplification (NASBA), self-sustained sequence replication (3SR), rolling circle amplification (RCA), phylogenetic analyses, random amplified polymorphic DNA (RAPD-PCR), nested multiplex PCR, phylotype-specific multiplex PCR, restriction fragment length polymorphism (PCR-RFLP), real-time quantitative PCR (RT qPCR), reverse transcriptase (RT) PCR, and repetitive sequence-based PCR (REP-PCR), had been applied in different branches of agriculture. This article describes the application of this technology that has brought revolution in different sectors of agriculture, including farming corps, such as paddy, wheat, jute, cotton, tea, vegetables, sugarcane, maize, pulses, potato, oilseeds, spices, fruits, tobacco, etc. and animal farming, including dairy, beef, meat, poultry, fishery, shrimp, apiculture, sericulture, etc.

## A Conserved Cell Surface Protein, SMU.518, Is Required for Virulence Expression in *Streptococcus mutans* UA159

Mohammad Shahnoor Hossain

Department of Genetic Engineering and Biotechnology,  
University of Dhaka, Ramna, Dhaka 100, Bangladesh

**Introduction:** Gram-positive genus-*Streptococcus*-includes various formidable pathogenic species. *Streptococcus* causes various diseases, such as pneumonia, meningitis, septicaemia, endocarditis, and oral caries. The prevention of streptococcal diseases is hindered by the limitation of well-conserved target for the development of universal vaccine. Using bioinformatics tool, a cell surface protein-SMU.518-was predicted, which is conserved among all streptococci. **Objective:** The present study was conducted to investigate this well-conserved streptococcal surface-associated protein for its role in virulence expression in *S. mutans* UA159, a causative agent of dental caries. **Methods and materials:** Using an advanced Cre-loP-based gene knock-out approach, SMU.518 mutant strain was constructed, and the expression of various virulence genes, bacteriocin production, transformation efficiency, and biofilm formation by *S. mutans* UA159 was studied. **Results:** The results suggest that the SMU.518 mutant is unable to produce bacteriocins and is defective in competence development compared to the wild-type strain. Moreover, its biofilm-formation ability was greatly impaired, and the mutant was more susceptible to antibiotic and acid stresses compared to the wild-type bacterium. Our complementation studies with the plasmid-borne introduction of SMU.518 to mutant background could fully restore the mutant phenotypes. **Conclusion:** The results indicate that SMU.518 is essential for the production of bacteriocins, competence development, and biofilm formation in *S. mutans* UA159. Further study is necessary to evaluate its role in other streptococci.

## **Cronobacter: A Unique Bacterial Species Identified in Bangladesh that Threatens to Increase the Burden of Disease**

Ashfaqul Hoque, Tahmeed Ahmed, Mohammad Shahidullah, Anowar Hossain,  
Mohammad Ilias, and **Dilruba Ahmed** (dahmed@icddrb.org)

icddr,b, Mohakhali, Dhaka 1212, Bangladesh

**Background:** *Cronobacter* species, formerly known as *Enterobacter sakazakii*, is implicated in outbreaks of meningitis and enteritis, especially in infants, preterm, and low-birthweight babies. Its presence in powdered infant formula (PIF) has been linked to outbreaks of disease. There is a huge market for imported PIF in Bangladesh; however, there is no information from Bangladesh regarding the contamination of PIF with *Cronobacter* spp. **Objective:** The study was carried out to investigate the presence of *Cronobacter* spp. in PIF and clinical samples (blood, stool, and CSF) collected from infants aged 0-24 months admitted to three different hospitals (BSMMU Hospital, Mirpur Shishu Hospital, and Dhaka Medical College Hospital) in Dhaka with diarrhoea, septicaemia, and/or meningitis and in PIF samples collected from retail markets. **Methods and materials:** All the PIF and blood, stool, and CSF samples from the study infants were tested for *Cronobacter* spp. In brief, the PIF samples were pre-enriched in buffered peptone water, followed by enrichment in Enterobacteriaceae enrichment broth, and *Cronobacter* screening broth. Isolates of *Cronobacter* spp. were identified in chromogenic DFI agar (Oxoid Ltd., UK). All the PIF samples were tested anonymously. **Results:** Of 32 PIF samples (8 brands) collected from hospitalized neonates, 16 were culture-positive but all were negative for *Cronobacter* spp. Of the 32 PIF samples (8 brands) from retail markets, 17 were culture-positive, and one was positive for *Cronobacter* spp. The isolate was confirmed with API 20E bioMérieux (France) biochemical profile and a standardized conventional PCR targeting the alpha-lucosidase and 16S rRNA gene sequence of *Cronobacter sakazakii*. *Cronobacter* spp. was not detected in any of the blood, stool, or CSF samples. **Conclusion:** *Cronobacter* spp. was detected for the first time in PIF in Bangladesh. This should caution healthcare providers about the potential outbreaks of serious disease caused by these unique bacteria in neonates and young infants. An active surveillance for this emerging foodborne pathogen is warranted to prevent the increased burden of septicaemic and gastrointestinal illnesses in Bangladesh.

**Acknowledgement:** The study was funded by the icddr,b and the Government of Bangladesh through the IHP-HNPRP.

***In Vitro Shoot Proliferation and Plant Regeneration of Plumbago indica L. (Ractochita),  
a Rare Medicinal Shrub of Bangladesh***

**Fowzia Haque, A.K.M. Sayeed Hassan, Miskat Ara Akhter Jahan, and Shyamal K. Roy**

Biological Research Division, BCSIR Laboratories,  
Dhaka 1205, Bangladesh

A protocol was established for *in vitro* shoot proliferation and plant regeneration of a rare medicinal shrub of Bangladesh, *Plumbago indica* L. (Plumbaginaceae), using shoot tip and nodal explants. Best shoot induction was observed on MS basal medium supplemented with 0.5 mg/L of BAP, in which 92% of the nodal explants responded to produce a maximum number ( $42.8 \pm 1.18$ ) of shoots per culture. *In vitro* raised shoots rooted on half strength MS medium with 0.5 mg/L of IAA. For acclimatization and transplantation, the plantlets in the rooting culture tubes were kept in normal room temperature for seven days before transplanting in pots where the plantlets were reared for three weeks. The survival rate of regenerated plantlets was 82%.

**Callus Induction and Plant Regeneration of *Paederia foetida* L., a Widely-used Medicinal Vine in Bangladesh**

A.K.M. Sayeed Hassan, **Chapol Kumar Roy,**  
Rebeka Sultana, and Rahima Khatun

Biological Research Division, BCSIR Laboratories,  
Dhaka 1205, Bangladesh

An efficient protocol was established for *in vitro* plant regeneration of *Paederia foetida* L. (Family: Rubiaceae), a widely-used medicinal vine of Bangladesh through callus induction in nodal segment. Yellowish green nodular callus was observed from nodal segments on MS basal medium supplemented with 1.5 mg/L of BAP + 0.5 mg/L of NAA within three weeks. The callus produced a large number of shoots ( $14.4 \pm 1.29$ ) when sub-cultured on MS medium with 0.5 mg/L of BAP. *In vitro* raised shoots rooted on half strength MS medium with 0.5 mg/L of IBA. For acclimatization and transplantation, the plantlets in the rooting culture tubes were kept in normal room temperature for seven days before transplanting in pots where the plantlets were reared for three weeks. The survival rate of plantlets was 85%. Regenerated plants were morphologically uniform having normal leaf shape and growth.

***In Vitro* Regeneration of *Ficus religiosa* L., a Multi-purpose Woody Medicinal Plant through Apical and Axillary Shoot Proliferation**

A.K.M. Sayeed Hassan, Farhana Afroz, **Miskat Ara Akhter Jahan**, and Rahima Khatun

Biological Research Division, BCSIR Laboratories,  
Dhaka 1205, Bangladesh

A protocol was established for mass propagation of the valuable medicinal plant *Ficus religiosa* L. (Moraceae) through in vitro culture. Apical and axillary buds of young sprouts from selected plants were used as explants. Best shoot induction was observed on MS basal medium supplemented with 0.5 mg/L of BAP + 0.1 mg/L of IAA, in which 78% of the explants regenerated shoots produced 16 shoots per culture. Repeated subcultures in the same medium resulted in rapid shoot multiplication with 24 shoots per culture. *In vitro* raised shoots rooted on half strength MS medium supplemented with 2.0 mg/L of IBA + 0.1 mg/L of NAA. For acclimatization and transplantation, the plantlets in the rooting culture tubes were kept in normal room temperature for seven days before transplanting in pots where plantlets were reared for three weeks. The survival rate of the regenerated plantlets was 85%.

## Transporting Fish Fingerlings: Transition between Life and Death

Md. Ibrahim<sup>1</sup>, Kazi Nazrul Islam<sup>1</sup>, A.D.A. Shahinuzzaman<sup>2</sup>,  
Kawser Ahmed<sup>3</sup>, and Gazi Nurun Nahar Sultana<sup>4</sup>

<sup>1</sup>Department of Fisheries, University of Dhaka, Ramna, Dhaka 1000, Bangladesh,

<sup>2</sup>Biological Research Division, Bangladesh Council of Scientific and industrial Research,  
BCSIR Laboratories Dhaka, Bangladesh, <sup>3</sup>Department of Oceanography, Faculty of Earth and  
Environmental Sciences, University of Dhaka, Ramna, Dhaka 1000, Bangladesh, and <sup>4</sup>Center for  
Advanced Research and Sciences, University of Dhaka, Ramna, Dhaka 1000, Bangladesh

Transportation of fish larval, fry and fingerling stages is a common procedure in aquaculture industries. The traditional method of transport uses open containers. People splash with hands or legs to put oxygen into the water during various means of transportation. Before transportation, fish would be in a state of normoxia, where fish would experience normal levels of oxygen. During transportation, lack of oxygen would trigger a physiological stress, i.e. hypoxia, where we anticipate lack of O<sub>2</sub> forces in the reduction of ATP production through the OXPHOS system with concomitant decrease in ROS production. And once released and, thus, re-oxygenation has been done, cellular ATP production would divert towards the OXPHOS system with concomitant increase in ROS production. This would lead to the death of fish fry in much higher numbers after release (normoxia again) compared to that of before release (i.e. hypoxia or transport period). To test this hypothesis, the present study investigated the formation of H<sub>2</sub>O<sub>2</sub> (via NWLSSTM-H<sub>2</sub>O<sub>2</sub> assay kit) as a representative of ROS in silver carp fingerlings during the hypoxic transportation and re-oxygenation period. To conduct the experiment, silver carp fingerlings were loaded in the transport vessels. After six hours of transport, fish were re-oxygenated and reared up to 16 days. During the hypoxic transportation and re-oxygenation period, H<sub>2</sub>O<sub>2</sub> was observed to be significantly ( $p<0.05$ ) higher compared to the control group. The results suggest that maintaining lesser ROS production in response to hypoxia, re-oxygenation is pivotal to keep fish healthy during transportation and release.

**Acknowledgement:** The study was supported by the Ministry of Science and Technology, Government of Bangladesh.

## Performance of Sweet Pepper under Protective Structure

G.M.A. Halim<sup>1</sup> and M.S. Islam<sup>2</sup> (shahidulhrt@gmail.com)

<sup>1</sup>Olericulture Division, Horticulture Research Centre, Bangladesh Agriculture Research Institute, Gazipur, Bangladesh and <sup>2</sup>Department of Horticulture, Sylhet Agricultural University, Sylhet, Bangladesh

The performance of sweet pepper was evaluated under different protective structures in two consecutive seasons of 2007-2008 and 2008-2009 at the experimental field of the Horticulture Research Centre of Bangladesh Agriculture Research Institute, Gazipur. One popular commercial capsicum variety-California Wonder-was included in the study with four protective structures (low-height poly tunnel, polytunnel with side open, poly tunnel with side closed, and poly house), including control (open field). The protective structures had a remarkable and significant influence on plant growth and yield of sweet pepper. The plants grown under the protective structures had higher plant height compared to those grown in the open field. The highest individual fruit weight (65.2 g) was recorded for the plants grown under poly house condition while it was the lowest for the open field-grown plants (3.34 g). More than five fruits were harvested when the plants were grown under the poly tunnel or poly house. The maximum fruit yield per plant (334 g) was recorded from the poly house, which was 160.4% higher than that of the plants grown under the open-field condition. The second highest yield was recorded from the plants of poly tunnel (212.5), indicating a bright scope for cultivation of sweet pepper under protective structures.

**Biotechnological Research of Insects in Bangladesh**

Masum Ahmad (masum\_bau2006@yahoo.com)

Department of Entomology, Bangladesh Agricultural University,  
Mymensingh 2202, Bangladesh

Insects are major agricultural pests and transmit many vector-borne diseases, e.g. malaria, dengue, and kala azar, to human-beings and animals. Agricultural pests are responsible for losses (30-40%), if no control technique is applied. Until now, chemicals are the only means of insect control in the country. Insecticide causes pollution, develops resistance, and poses risk of residual toxicity in commodities. Studies have been carried out to facilitate nuclear and biotechnological fields in controlling insect pest and to exploit beneficial insects. The major area covers development of eco-safe control measures for insect pests/vectors. Irradiation is an alternative quarantine treatment for horticultural crops. The use of a low-dose gamma radiation can yield higher-quality silk and lac. Radiation/microbial pathogen-induced damage of insect tissues and organs was investigated. The major achievements are: development of protocols for the increased production of quality silk and low-cost liquid diet for mass rearing of fruit fly for using in SIT, protocols for developing bacteria-based highly-efficient mosquitocidal bio-pesticide, and protocols for irradiation quarantine treatment for fresh fruits and vegetables and pest risk assessment of mango. A perspective plan is field application of bio-pesticide for the management of mosquitoes.

## Regulation of *LEE1* Promoter of Enterohaemorrhagic *Escherichia coli*

Md. Shahidul Islam<sup>1</sup>, Lewis Bingle<sup>2</sup>, Mark Pallen<sup>2</sup>, and Steve Busby<sup>2</sup>

<sup>1</sup>Department of Biotechnology, Bangladesh Agricultural University, Mymensingh 2202, Bangladesh and <sup>2</sup>School of Biosciences, University of Birmingham, Edgbaston, Birmingham, B15 2TT, UK

*Enterohaemorrhagic Escherichia coli* (EHEC) O157:H7 str. Sakai is an emerging human pathogen causing type three secretion system-based attaching and effacing lesion on gut epithelial cells. The genes, responsible for EHEC virulence, are contained in a pathogenicity island named the locus of enterocyte effacement (LEE). The LEE containing approximately 41 open reading frames (ORFs) consists of five major polycistronic operons (*LEE1-5*), which co-ordinately encode a type three secretion apparatus and effectors. *LEE1* is considered the key operon in the LEE region because it encodes Ler, which positively regulates expression of all other LEE genes. The *LEE1* promoter essential elements were determined and found that GrlA, a LEE-encoded transcription factor, positively regulates the expression from *LEE1* promoter. Deletion analysis was used for defining the essential elements at the *LEE1* promoter, and a small active promoter fragment was generated. Starting with this fragment, 'down' mutations were obtained by random mutagenesis. Most single mutations clustered in the putative-10 and 35 regions. Insertion of a single base in the spacer region that made the spacer length 19 bases reduced the promoter activity dramatically whereas deletion of a single base increased the promoter activity significantly. Potassium permanganate footprinting and *in vitro* transcription assays complemented the results of mutational analysis for the identification of the *LEE1* promoter. Results of mutational analysis further suggest that the DNA target for GrlA is located between the *LEE1* promoter-10 and 35 elements. Starting with a single base substitution that prevents induction by GrlA, a number of GrlA suppressor mutations were found. The location of the suppressors indicates a specific region on GrlA that are important for interacting with the *LEE1* promoter. Moreover, a cryptic promoter of unknown function was identified in the *LEE1* regulatory region, and a minigene was discovered in the leader region of the *LEE1*. Knocking out the minigene from EHEC genome using lambda red mutagenesis lowered the expression of downstream *ler* gene and reduced the pathogenicity confirmed by the fluorescent actin staining test. Based on the findings, it is concluded that *LEE1* has a complex regulatory region, and expression of its first cistron (*ler*) is subjected to both transcriptional and post-transcriptional regulation by GrlA and a minigene respectively.

**Acknowledgement:** The study was supported by the BBSRC project grant and Commonwealth Scholarship Commission, UK.

## A Comparison of Haematological and Cytogenetic Parameters Using Qualitative PCR Method in Monitoring CML among Bangladeshi Patients

Bikash Chandra Chanda<sup>1</sup>, Mohammad Abdul Mottalib<sup>2</sup>, Mohammad Abdul Khaleque<sup>3</sup>,

Hafizur Rahman<sup>1</sup>, Mohammad Sirazul Islam<sup>2</sup>, AND Tanvira Afroze Sultana<sup>2</sup>  
(tanvira\_sultana@yahoo.com)

<sup>1</sup>Clinical Hematology and Microscopy Unit, Clinical Laboratory Services, Laboratories, icddr,b, Mohakhali, Dhaka 1212, Bangladesh, <sup>2</sup>Hematology Section, General Laboratory, Laboratory Services Division, BIRDEM, 122, Kazi Nazrul Islam Avenue, Dhaka 1000, Bangladesh, and <sup>3</sup>Department of Life Sciences, North South University, Bashundhara Residential Area, Dhaka, Bangladesh

**Background:** The clinical significance of various isoforms of the BCR-ABL fusion gene (e1a2 and e14a2 vs e13a2 and e19a2) of chronic myeloid leukemia (CML) is still a subject of controversy.

**Objective:** A correlation between various isoforms of BCR-ABL transcripts and characteristics of CML patients, including haematological parameters, was searched. **Methods and materials:** Cytogenetic studies of bone marrow from 49 Bangladeshi CML patients, either untreated or during follow-up of treatment (Imatinib Mesylate, Gleevec) phase, were performed for messenger RNA (mRNA) expression of BCR-ABL by nested reverse transcriptase polymerase chain reaction (RT-PCR) method. **Results:** Of 30 untreated patients, four possessed the minor isoform or e1a2 transcripts, four had the micro isoform or e19a2 transcripts, and 22 had either of the two major isoforms, e14a2 or e13a2 transcripts. The distribution of e14a2 and e13a2 transcripts within the 22 patients was 16 and 6 respectively. The only difference in the clinical characteristics between the two transcripts (e14a2 and e13a2) was the platelet count, which was significantly ( $p=0.001$ ) higher in the e14a2 patients than in the e13a2 transcript. Nineteen patients were followed up for BCR-ABL expression after treatment for six months. On treatment, there was a significant decrease in white blood cell count ( $p=0.02$ ) and increase in haemoglobin level ( $p=0.03$ ) in 12 patients who showed the presence of BCR-ABL transcript compared to those ( $n=7$ ) in which the fusion gene could not be detected. **Conclusion:** The results indicate that the type of BCR-ABL transcripts correlates with the haematological parameters of CML patients.

## DNA Barcoding: A Prospective Molecular Tool for Management of Fisheries in Bangladesh

Md. Mizanur Rahman<sup>1</sup> (mizanfish@yahoo.com)  
and Mahmuda Begum<sup>2</sup> (panna\_mahmuda@yahoo.com)

<sup>1</sup>Department of Zoology, University of Dhaka Ramna, Dhaka 1000, Bangladesh and

<sup>2</sup>Zoology Section, Dhaka Laboratories, BCSIR, Dhaka 1205, Bangladesh

Bangladesh is enriched with diversified fishery resources, including more than 700 species of marine and freshwater fishes, along with a remarkable number of mollusk, crab, prawn, and shrimp species. The DNA barcoding approach can be introduced in the biological resources- management system as one of the well-timed molecular tools for the genetic characterization of vast fishery resources in the country. DNA barcode relies on short, highly, variably regions of the mitochondrial genomes and are currently used in the identification of biological matters all over the world. A short DNA sequence from a standardized position in the genome is used as a molecular diagnostic tool for species-level identification in various taxa. A region of the mitochondrial gene COI (cytochrome C oxidase subunit I) is used for barcoding animals, particularly the fishes. With thousands of copies per cell, mitochondrial sequences are readily amplified by polymerase chain reaction, even from very small or degraded specimens. The amplified sequence is submitted for sequencing in one or both directions. This tool has a great prospect to be used in different aspects of the fisheries-management system, including identification of fish for the assessment of genetic diversity, identification of threatened, endangered and protected species (conservation) and exploration of new species and possible fusions, and insights into phylogenetic relationships (fish biology, evolution). This method has an additional application, which includes the determination of biological elements from processed products, e.g. canned fish and dried fish, and even the identification of fish eggs and fish larvae for resolving the quarantine issues of fisheries management. Therefore, the DNA barcoding technique should be applied as a molecular tool for the management of fishery resources and conservation of biodiversity of Bangladesh.

## **Development of In vitro Regeneration Protocol of Important Medicinal Plant Black Pepper (*Piper nigrum L.*) and Testing of Its Antimicrobial Activity against Common Bacteria**

Nilima Das, Mousona Islam, Aleya Ferdousi, and Salim Khan

Plant Tissue Culture Section, Biological Research Division, Bangladesh Council of Scientific and industrial Research, BCSIR Laboratories, Dhaka, Bangladesh.

Black pepper (*Piper nigrum L.*), the 'King of Spices', is a universal table condiment. The black pepper is generally cultivated by seed because other vegetative propagation methods are slow and time-consuming. Therefore, the tissue culture technique is considered a more efficient and reliable method for rapid and mass propagation of this economically-important plant. The present study was initiated to develop a protocol for micro-propagation of black pepper, and the antioxidant potential of the medicinal plant *Piper nigrum L.* was investigated. Callus induction and shoot regeneration were induced from leaf explants of potted plants cultured using MS medium supplemented with different plant growth regulators. The best callogenic response was observed on explants cultured for 30 days in the MS medium supplemented with 2.5 mg/L of 6-benzylaminopurine (BAP)+2.0 mg/L of 2,4 dichlorophenoxyacetic acids. The maximum number (6.0) of shoots/explants was recorded for explants cultured in the MS medium supplemented with 2.5 mg/L of BAP. Following the transfer of shoots to an elongation medium, the longest shoots (5.0 cm) were observed in the MS medium supplemented with 2.0 mg/L of BAP +1.0 mg/L of Kn. The elongated shoots were rooted in the MS medium supplemented with different concentrations of indole butyric acid. An assay of the antioxidant potential of the in vitro grown tissues revealed the excellent inhibition of the growth of Gram-positive and Gram-negative bacteria.





### Objective:

The single goal is to significantly ensure highest level of protection, promotion & support of breastfeeding and optimal IYCF practices and optimum maternal nutrition in Bangladesh.

## Strategies:

1. Work closely with government of Bangladesh for effective national, sub-national and international impact on improved IYCF;
  2. Integration of breastfeeding promotion, protection and support with existing health care infrastructure and non government partners;
  3. Effective communication strategies and community participation to reach all segments of population;
  4. Legal framework: BMS Code implementation to protect infant and child health;
  5. Development of IYCF through research and development in Bangladesh & internationally;
  6. To abide global IYCF strategies in partnership with WABA, IBFAN, One Asia, SAIFRN, ILCI, Well Start - more.



**Bangladesh Breastfeeding Foundation (BBF) Institute of Public Health (IPH), Room # 197-200,  
(Ground Floor) Mohakhali, Dhaka, Bangladesh. Phone: +88 02 9860801, +88 02 8813734**

Fax: +880-2-9860801. Website: [www.bbf-bd.org](http://www.bbf-bd.org)

E-mail: [breastfeeding.bd@gmail.com](mailto:breastfeeding.bd@gmail.com)

Chairperson: Dr. S. K. Roy E-mail: skroy1950@gmail.com, Mobile: +8801943-220587.

**বিশ্ব মাতৃদুর্ঘট সংগঠন ২০০৯, ২০১০ এবং ২০১১  
এর উদ্বোধনী অনুষ্ঠানে মাননীয় প্রধানমন্ত্রী  
কর্তৃত প্রচলিত প্রতিক্রিয়া/সোম্বলা**

## কর্তৃক প্রদত্ত প্রাতঃক্রিয়া/ঘোষণা



● মাতৃসেবক শান্তিলাল পিতৃক স্মৃতি ●

## ମାୟେରଦୁଧ ଖାତ୍ୟାନୋର ବିଷୟେ ପାଁଚଟି ପ୍ରଧାନ ତଥ୍ୟ

- ১ জন্মের পর পরই শিশুকে মায়ের প্রথম শাল  
দুধ খাওয়ান।

২ গর্ভবতী ও প্রসূতি মাকে তার পুষ্টি এবং শিশুর  
সুস্থান্ত্রের জন্য বেশী করে খাবার খেতে হবে।

৩ ছয় মাস বয়স পর্যন্ত শুধু মায়েরদুধই যথেষ্ট,  
এমনকি পানিরও প্রয়োজন নেই।

৪ শিশুর বয়স ছয় মাস পূর্ণ হলে মায়েরদুধের  
পাশাপাশি পরিবারের অন্যান্য খাবার খাওয়ান  
এবং দুই বছর পর্যন্ত মায়েরদুধ চালিয়ে যান।

৫ যে কোন অসুবিধে শিশুকে মায়েরদুধ ও অন্যান্য  
খাবার বাবে বাবে খেতে দিন।



ମାଧ୍ୟମଦୂରେ ବିକଳ୍ପ ବାହାରକାଠ  
କରାର କାହାର ତ ଯାଏନ୍ତିରୁ  
ବୀଠିଶାଖା ଦ୍ଵୟାଶାଖା

ନିଷିଦ୍ଧ

Bangladesh Center for Communication Programs (BCCP), a successor to the Johns Hopkins Bloomberg School of Public Health/Center for Communication Programs (JHU/CCP), Baltimore, USA, is a non-profit, non-government organization operating since 1996 in Bangladesh.

Over the years, BCCP has emerged as a leading strategic communication organization with international reputation and standard having extensive technical expertise and experience in social communication. BCCP makes a difference in people's lives, bringing a unique set of skills, approaches and experience that produce results.

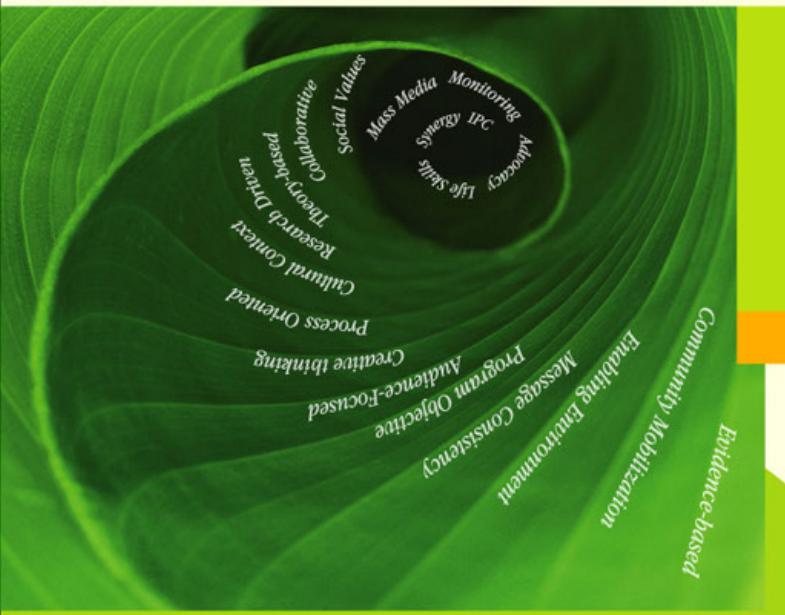
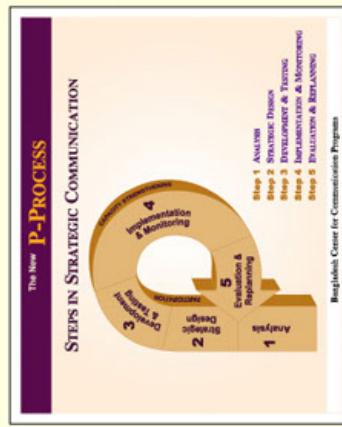
#### What We Do

BCCP designs, manages and implements large-scale communication programs and supports development initiatives of the government, NGOs, public and private sector agencies providing the services mentioned below. BCCP relies on its technical strengths, but is guided by its core values of developing result-oriented multi-sectoral collaboration and efficiency both at global and national levels.

A multi-disciplinary Research and Evaluation team deploys appropriate methodologies and approaches that best suit to the program and contributes to its success. BCCP's program evaluation methodologies also apply innovativeness that helps not only examine if change has occurred, but also how and why it has occurred.

#### Vision

To help create an empowered and enlightened Bangladesh free from poverty and exploitation based on age, sex and religion through raising awareness and increasing the knowledge of the people utilizing strategic communication.



## Strategic Communication Makes a Difference

**Our approach**  
BCCP follows "P-Process" in every stage of its operations. The P-Process is a tested and proven communication project planning tool for designing and implementing BCC programs more scientifically and effectively.

#### Areas of activity

Although BCCP is a recognized leader in the field of health communication, its work spans a broad spectrum of development issues and related other areas including: education, gender, violence against woman, anti-trafficking, agriculture, democracy and good governance, local government, anti-terrorism, public procurement reform, anti-tobacco etc. offering package of diversified services.

**Mohammad Shahjahan**  
Director & CEO  
House # 8, Road # 3, Block-A, Section-11  
Mirpur, Dhaka-1216, Bangladesh.  
Tel: +8802 9020329, 9012685  
Email: info@bangladesh-ccp.org  
Website: www.bangladesh-ccp.org

#### For details

**Bangladesh Center for Communication Programs**





in collaboration with



&

*American Society of Bangladesh-affiliated Microbiologists*

---

Platinum partner:



Print media partner:

**the independent**

Electronic media partner:

