FOUNDER DIRECTOR

ANNUAL REPORT

OF

THE BOSE INSTITUTE

FOR

1998-99

Bose Institute

93/1, Acharya Profulla Chandra Road
Calcutta-700 009

(3) Studies on structure, function and dynamics of biomolecule.
(4) Studies on ecology, environmental pollution and related problems.
(5) Studies on microbes and parasites for industrial and medical applications.
(6) Problems in condensed matter, intermediate energy physics and related interdisciplinary areas.

Besides these, Bose Institute is also conducting Course Work Programme for pre and postdoctoral research students and in different areas of Biotechnology.

Through years of its existence this prestigious Institute has proved itself to be a pioneer in the frontiers of modern Physical and Life Sciences. Its work and achievements have attracted attention of Scientists from our country and abroad. Many of our Faculties are Bhatnagar awardees, Fellows of National Academics and recipients of National and International awards, and have intra, inter-institutional, International research collaborations and their own funded projects related to above-mentioned areas. Facilities and infrastructure developed in the Institute are also used by scientists of many universities and Institutes of the country. Our scientists also serve the nation through their participation in many national and international scientific committees/organisations, to selection committees, and acting as examiners of many university and national tests.

From the Director's Desk

I am pleased to present the Annual Report of Bose Institute for the year 1998-99 before you. During this period the Institute is on the way of its continued progress and a lot of developmental activities so far as creation of infrastructural facilities, instrumental capabilities and renovation and repair of various old Buildings and Laboratories are concerned. It is now well-known to everybody that Bose Institute is a multi-disciplinary research institute having ten Departments/Sections doing scientific research in the area of Physics, Chemistry, Botany, Biochemistry, Microbiology, Biophysics, Animal Physiology, Plant Molecular and Cellular Genetics, Immunotechnology and Environmental Sciences. It is quite advantageous on the part of our scientists that under one umbrella so many disciplines of science exist so that they can explore the possibilities of using them, collaborating with like-minded scientists from other disciplines in other Departments and Sections as well.

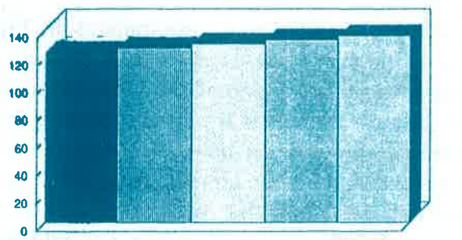
With the incorporation of quite a large number of sophisticated equipments in the Institute and induction of various equipments to upgrade the departmental and sectional instrumental capabilities, the Institute has emerged as a potentially well-equipped laboratories in the entire North-Eastern region of our country. It is very heartening for me to announce that all of these newly acquired equipments are being well maintained by a group of scientists on whom the responsibilities of each of the equipment for maintenance has been given. Such gadgets not only facilitates the research of our own scientists, but helps scientists from other Institutions, Universities also. We are extending this instrumental support to others also, keeping in mind that this would help promote scientific research in general to other organizations as well.

We have taken up the matter with the Department of Science & Technology of the Government of West Bengal to even organize a course on usage of various equipments for advanced research work for the post M.Sc. students. If everything goes alright then we might be able to initiate such course from January 2000 on a three-monthly basis. Several faculty members from different departments are expected to take part in this programme. The institute also had taken a stride to initiate a 5-year M.Sc.–Ph.D. Programme as a part of the proposed Technological University, which have been initiated by the Government of West Bengal. This 5-year comprehensive programme towards giving an M.Sc.–Ph.D. Degree by the Technology University in the area of Biotechnology will be fully organized by Bose Institute on their behalf after a necessary agreement between the institute and the Technology University is signed. Necessary measures in this regard and discussions pertaining to this proposal are presently going on. The Institute is also entering into a collaborative programme with the Department of

Agriculture, Calcutta University (sponsored by ICFRE) for multilocation trial of Poplar tree in our Madhyamgram Experimental Farm after signing an MOU that the cost of such plantation will be borne by the project funded by the Calcutta University, but the trees, in course of time (about 3 to 5 years), will be the Bose Institute property. These trees have high value and obviously will fetch certain amount of funds to the Institute. Commercial cultivation of medicinal plants have been initiated in the Madhyamgram Experimental Farm by Prof. P. Bhattacharyya. It is anticipated that large scale cultivation of the medicinal plants will also fetch certain amount of funds. I am happy to inform you that the Institute has been able to hand over quite a large number of plantlets developed by tissue culture technique, such as Kaal Megh and Teak plant as also bamboo developed by tissue culture, to the hands of the Department of Science & Technology, Government of West Bengal for large scale monoclonal propagation. This programme was undertaken as a part of our "Lab to Land Programme". The Institute has been looking forward to all avenues for generation of outside cash flow in the event of the serious financial problems, which it faced during the financial year

Number of Publications

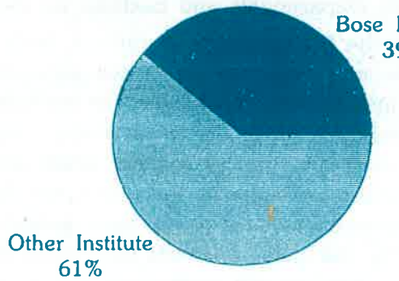
(Year wise)



Figure—1

Publication in Highimpact Journals

1982-1998 (Plant Science)



IICB, JU, CU, SINP, CRI

Calcutta

Figure—2

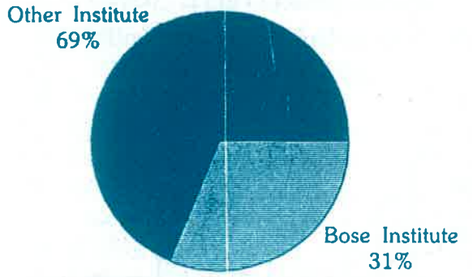
1998-99. The minimum requirements of the Institute (Rs. 991.43 lakh) could not be procured from the Department of Science & Technology, Government of India, while the funds provided by them (Rs.702.66 lakh) were grossly inadequate. But the Institute had to incur expenditures on account of salary at the increased rate subsequent to the 5th Pay Commission recommendations, arrear payment, payment of increased rate of pension to the pensioners etc.

While looking at the various scientific progress and achievements of the Institute it may be noted that there is a progressive increase in the number of scientific research publications from Bose Institute ***(Fig.1)***. Out of the major Institutions and Universities

in the Calcutta region, Bose Institute alone showed largest amount of its contributions in high impact factor journals comprising 39% of the total of all other Institutions in the Calcutta region ***(Fig.2)***. As per the MEDLINE search, out of a total of about 10 high impact factor journals, Bose Institute's contributions stand 1/3rd of the total number from

Publication in Highimpact Journals

1993-1998 (MEDLINE)



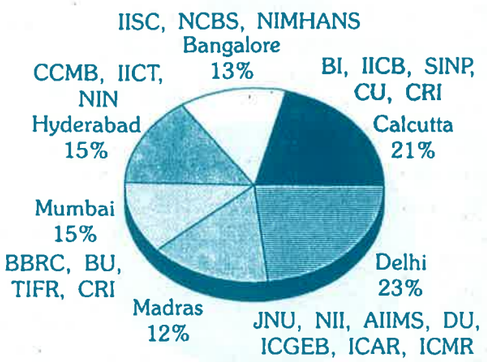
Other Inst. = IICB, JU, CU, CRI, SINP

JBC, JMB, J. Bact, PNAS, Gene, Biochem
Prot. Sc., Port.Eng., Plant Physiol
Phytochem, BBRC, BBACTA, Bioche-Mol-Bio-Int

Figure—3

Publication in Highimpact Journals

1992-1998



Plant Science (Agricola)

Figure—4

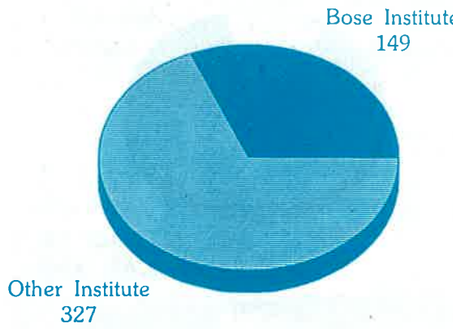
Calcutta region. Two Universities and three major research institutions were included in this comparative search ***(Fig 3)***. So far as Plant Science is concerned, as per AGRICOLA search by comparing and contrasting major Institutions from Calcutta, Delhi, Bombay, Madras, Hyderabad and Bangalore, Calcutta alone contributed 21%, while Delhi only were higher than Calcutta with 23% of its publications ***(Fig. 4)***. Publications from all other major cities were lower than Calcutta. It is heartening to note that the Bose Institute among other institutions in Calcutta contributed about 1/3rd of the total publications in the top ranking high impact factor biomedical journals ***(Fig. 5)***.

The Bioinformatics Centre (Distributed Information Centre), which had been functioning at Bose Institute with the support from the Department of Biotechnology, Government of India are catering to the requirements of not only West Bengal but also for those from Bihar, Orissa, North-Eastern States, Madhya Pradesh, Maharastra, Uttar Pradesh, Andhra Pradesh, Karnataka, Assam etc. ***(Fig. 6)***. People come from these cities to do computer mediated search, sequence analysis, and also to carry out biomolecular modelling studies involving Silicon Graphics, etc. A National Facility for biomolecular modelling has been set up by DBT, Government of India. DIC is also extending its area of activities, and are also adding to our revenue earnings touching about Rs. 2.5 lakhs per annum during 1998-99 ***(Fig. 7)***.

I very strongly feel that the Institute has to give a fresh look as to how the expertise and capabilities of our scientists, and also the tremendous instrumental facilities created in the Institute, could be extended to the other organizations, and such consultancy projects would not only help them earning some extra money but also help the Institute to tide over the financial constraints. Such ventures should be taken up seriously otherwise the Institute might come up to a situation which would be difficult to tide over. On the Institutional Administration and Accounts side also, all round efforts have already been initiated to stop excess expenditures on projects, to recover advances to various individuals timely and efficiently, and also to ensure that avoidable expenditures should be reduced as much as practicable. We need co-operations from all concerned so that the Institute does not have to face financial contraints and difficulties. As you are well aware that the problems arising out of our severe financial constraints have been brought to the attention of all authorities, but up until this date nothing really happened, although certain amount of verbal promises have been received.

Publication in Highimpact Journals

1982-1998 (Plant Science)



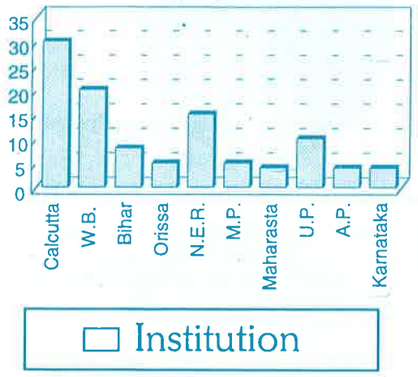
Other Institute = IICB, SINP, CU, CRI, JU

Figure—5

BIOINFORMATICS CENTRE

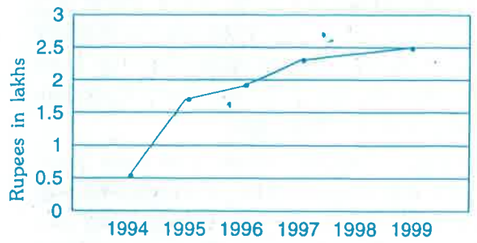
BOSE INSTITUTE (DIC)

(Institutions/Universities/Organization using DIC facilities)



Figure—6

Revenue earned by D.I.C.



Figure—7

I am happy to let you know that the Institute now figures in the international publication "Profilies of Institutions for Scientific Exchange and Training in the South"

published by the 3rd World Network of Scientific organizations in collaboration with South Centre and 3rd World Academy of Sciences.

I am extremely happy to announce that Dr. Siddhartha Roy of the Department of Biophysics has recently been awarded the coveted **Shanti Swarup Bhatnagar Prize** for his contributions in the area of Biophysical Studies of Molecular Structures. I am quite sure that the inputs, which have gone into the Institute for the past several years, are beginning to blossom. I am quite certain that in future years many more laurels will be brought by our scientists. Dr. Pratima Sinha of the Department of Biochemistry has been awarded for Fellowship of the National Academy of Sciences, Allahabad. Dr. Pinak Pani Chakraborty has been awarded the Fellowship of National Academy of Sciences, Allahabad. Prof. Parimal Sen has been awarded the Fellowship of the National Academy of Sciences, Allahabad. Dr. Siddhartha Roy has been awarded the Fellowship of the Academy of Sciences, Bangalore.

Recently, we have noted in a publication from NISSAT, New Delhi, wherein they have compared and contrasted the scientific contributions in terms of publication of papers in reputed journals after selecting out 50 major research institutions in the country. Although there could be questions and comments regarding the procedure and method they followed for this national survey but taking their omission and commission granted, the Bose Institute's contributions in the area of Biomedical Research appear to be as one of the five top ranking institutions out of the 50 such organizations in the country. In the field of Biological Research, Bose Institute figures among the first 13 research institutions. After going through their methods it appears that if we would have been accurately represented, our position could have gone further up the ladder.

The research areas of the Department of Physics includes high energy particle and nuclear physics, astrophysics and cosmology, radiation physics, condensed matter and atomic physics, quantum mechanics, fluid dynamics, statistical physics, ultrasonics, acoustics, chemical physics and materials science. The achievements of the Department are tables of stage-of-the art precise elastic scattering cross sections of photons ($13<Z<104$, $50\ KeV<Er<1500\ Kev$), discovery of new interfacial instabilities in stratified fluid flows, study of long range quantum interaction between deformable one-dimensional structures, connection between geometric phase and the exclusion principle of Quantum Mechanics, use of biochemical of statistical methods to understand anomalies in bacterial evolution (punctuated equilibrium), development of an exactly solvable spin model, study of randomness and disorder in the formation of turning patterns in reaction-diffusion systems, estimation of baryonic cold dark matter in the Universe from the QCD phase transition, electromagnetic diagnostics of Quark Gluon Plasma, development of the superheated drop technology for further novel uses, systematic study of hadronic properties at extreme conditions of high temperature/density.

The Department of Chemistry has concentrated its research efforts in 3 major directions, such as Chemistry, Biophysical Chemistry, Biochemistry and Molecular Biology. The Department has submitted 3 Patent applications and has been successful to obtain significant amount of funds from various national and international funding agencies

worth crores of Rupees. Both Synthetic and Natural Product Chemistry, Lipid Chemistry, Drug resistance mechanism involving Tubuculous and shigella organisms, mechanism of transport and its regulation involving Na-K ATPases and others are the present focus of study. The significant new work of the Department includes molecular chaperones in folding of globular proteins and to understand the structural and conformational basis of chaperone function, identification of cholesteric, chiral smectic and smectic B states in F-actin using optical techniques (experimental and theoretical) and development of molecular theory of the smectic C state of matter and use of continuum theories to account for the phase transition involved in F-actin (theory).

The Department of Botany has been carrying out its research in a number of directions of modern plant sciences. Some of the areas are – marker assisted breeding in rice to develop PCR based DNA markers linked with submergence tolerance, conservation of economically useful plants using micropropagation techniques, isolation and characterisation of bio-active substances from marine and terrestrial sources, molecular mechanism of polymine action under abiotic stress, to delay fruit ripening by over expressing SAM decarboxylase and anti sense ACC synthetase RNA in banana, development of molecular markers for seedling vigor in rice and selection of appropriate germplasm for rainfed lowland condition, molecular documentation of tea germplasm, free radical damage and scavenging enzymes in plants, mangrove, coastal quaternary and sea level change in Bengal basin, seed physiology and seed screening, improvement of Brassica and micro-propagation of forest trees, DNA polymerase from plants; characterisation and cloning, salinity stress and gene expression in rice, air born spores and pollen and their allergenicity. A new technique for micropropagation of plant using coir in place of agar has been developed in the Development, for which a Patent application has also been made.

In the Department of Biochemistry, the major emphasis of research are primarily in 2 major directions, such as Molecular Biology and Genetics and Structural Biology. The Department is carrying out significant work, for which various members of its faculty have been recognised at the national and international level. An account of the work includes–P gene lethality in regulation of gene expression in mycobacteria and its phase, chromosome replication and segregation in the budding yeast, control of cell division, differentiation and molecular epidemiology of parasitic protozoa, protein self assembly, folding and protein-drug interactions, small molecule and macromolecular crystallography; database analysis; energy calculations on model systems and molecular design, crystallographic studies of modified DNA oligomers, lipase, repressor, model building studies of repressor molecules, protein-DNA interactions; transcriptional regulation in *E.coli* etc.

The Department of Microbiology has been carrying out research in different areas, which include understanding of the molecular basis of cholera and related diarrhoea with a view to develop appropriate diagnostic and prophylactic measures, molecular and cellular mechanisms of host parasite interaction in visceral leishmaniasis, biotransformation of organic compounds by microbes : Production and synthesis of

important drug intermediates, mechanism of protection of impairment of P 450 during host parasite interactions in *Visceral leishmaniasis* : Role of nitric oxide synthase, Development of improved strains of rhizobium for efficient nodulation and nitrogen fixation: Development of biofertilizer, microbial beneficiation of ores and minerals : Understanding the molecular mechanisms of oxidation of sulfur compounds by *Thiobacillus*, signal transduction and apoptosis : Understanding the role of ceramide in altering the signalling mechanism in host defence system and molecular biology of *Mycobacterium:* Understanding the replication mechanism of plasmid pAL 5000. The Department filed Patent for "Development of a microbial based arsenic assay kit."

In the Department of Biophysics, the Scientists are publishing in high impact factor journals and are also able to obtain research grants from various funding agencies. The emphasis of the Department is on structural Biology studies and on structure-function analysis of both large and small molecules.

The Animal Physiology Section is carrying out its research activities in the area of understanding the basic physiological mechanisms involved in various life processes of animals, development of technologies for improved production of economically important animals and to evaluate different states related to human health and disease.

The Plant Molecular & Cellular Genetics Section has been contributing in the field of Molecular and Cellular Genetics including Biotechnology of Plants. The highlights of the achievements of the Section include yellow stem border resistant IR64 rice cultivars generated with modified BT gene, successful introduction of alien genes in different cultivars of Mung Bean, Mustard and Chickpea, introduction of agronomic traits from wild and allied germplasms into Mustard cultivar through somatic hybridization and establishment of antiquity of the inositol synthase protein/gene.

The newly created Immunotechnology Section has been concentrating their research work on selective induction of apoptosis in cancer cells with simultaneous potentiation of immune cells of the host by Protein A, amelioration of AZT induced toxicity by Protein A using combinatorial therapy, mapping of peptide sequence of *S. aureus* Protein A having immunomodulatory motifs, Protein A induced activation of antitumor responses–involvement of nitric oxide, studies on Heat Shock Protein–bound superantigen chimeric protein as immunogen to develop antitumor immunity, cloning and expression of Protein A into mammalian system, studies on the functions of different truncated polypeptide fragments of Protein A, development of a more sensitive methods to quantita mRNA of cytokines and their receptors based on competitive PCR and development of a more sensitive method for diagnosis of malaria parasites. The Section has submitted 2 Patent applications.

The Environmental Sciences Section studies the environmental impact and toxicology profile of various environmental chemicals. It has applied for a Patent for the development of a method for the elimination of organochlorinated pesticide (HCH) from soil water and food through a microbial process. It has also published several papers in different journals and has been consistently helping a large number of

industries and other establishments including West Bengal Pollution Control Board, Calcutta Port Trust, Johnson & Nicholson, Indian Rare Earths Ltd., Shalimar Paints Ltd., MECON, Ranchi, India Government Mint, Fort William Industries Ltd., Pharma Intel, Bharat Breaks and Valves Ltd., SMG High, Indian Foundry Association, Barger Paints Ltd., Exide Industries, Santaldi Thermal Power Station, Bandel Thermal Power Station etc., and many others. The Section could bring a total of about Rs. 80.00 lakhs worth of small and big projects through extension of its consultancy, analytical testing, research etc.

During the year Prof. N.K. Ganguly, Director-General, ICMR, delivered the Acharya J. C. Bose Memorial Lecture on November 30, 1998 on a very promising topic "Present and Future of AIDS Scenario in India". The Department of Botany organised a Training Workshop on "Coastal Quaternaries, Bengal Basin" and the Department of Chemistry organized a National Symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules." Both of them were very successful. The Institute organized a Farmer's meet at our Madhyamgram Experimental Farm and the awareness programme was very successful.

Like previous years, the Institute celebrated the National Science Day on February 28, 1999, with quiz contest among the School Students, exhibition etc. A Scientists' retreat was organized at the Director's Bungalow at Falta to discuss about ways and means as to how to attract outside cash flow to the Institute. Several ideas have come up out of those discussions and are being actively pursued. I am quite sure that with the active interest, hard work and participation of all sections of our employees and the infrastructural support being received from our Library, Workshop, Experimental Farms, RSIC, DIC, Administration, Accounts, J.C. Bose Meseum and from other corners the Institute looks forward with great hopes and aspirations to the next millennium. This being the last Annual Report of the present millennium will remain as a historical document for the Institute and the nation for future researchers, who would look for great scientific institutions of the nation and their work, activities, successes and failures.

I wish you all a great and successful year ahead of us.

(P.K. RAY)
DIRECTOR

CONTENTS

MANAGEMENT COMMITTEE

Members of the Governing Body

1. Shri Sanjoy Sen
2. Prof. Sunando Bose
3. Shri Debabrata Bose - Secretary
4. Prof. R.N. Chakraborty
5. Dr. (Mrs.) Debi Chakraborty
6. Prof. D. Banerjea
7. Prof. B.B. Baliga
8. Prof. S.N. Chatterjee
9. Shri Somnath Sanyal
10. Dr. A. Chatterjee
11. Director, Bose Institute

Members of the Council

1. Shri Sanjoy Sen
2. Prof. S. Bose, Upto 31.10.98
   Dr. A Chatterjaee—w.e.f. 01.11.98
3. Shri Debabrata Bose
4. Prof. R.N. Chakraborty
5. Prof. (Mrs) Debi Chakraborty
6. Mr. Somnath Sanyal
7. Prof. S. N. Chatterjee

} Nominees of the Governing Body

8. Secretary, Department of Science & Technology, Government of India, or his nominee
9. Director General, C.S.I.R. or his nominee
10. Joint Secretary and Financial Adviser, Department of Science and Technology, Govt. of India or his nominee
11. Representative of the Lok Sabha
12. Principal Accountant General (A & E), West Bengal
13. Secretary, Higher Education (Nominee of the Govt. of West Bengal)
14. Municipal Commissioner, Representative of the Calcutta Municipal Corporation
15. Director, Bose Institute - Ex-officio
16. Deputy Director, Bose Institute - Ex-officio
17. Prof. Sandip K. Basu
18. Prof. G. Padmanaban

} Two scientists from outside West Bengal nominated by DST, Govt. of India

Registar, Bose Institute - Non Member Secretary

Invitees :
1. Representative of the Academic Council
2. Representive of Bose Institute Employees Association.

Members of the Finance Committee

1. Shri Sanjoy Sen - (Representative of the Council) - Chairman
2. Shri Debabrata Bose - (Representative of the Governing Body)
3. Joint Secretary and Financial Adviser to the Department of Science and Technology, Govt. of India, or his nominee
4. Principal Accountant General (A & E), West Bengal
5. Director, Bose Institute - Ex-officio
6. Registar - Non-member Secretary

Acknowledgement

The Council takes the opportunity to express its grateful thanks to the Department of Science and Technology, Government of India, for providing generous funds in the form of Grants-in-aid, and for the support of research projects, and also to other external agencies such as the Government of West Bengal, Department of Atomic Energy, Council of Scientific and Industrial Research, Indian Council of Agricultural Research, Indian Council of Medical Research, etc., for their support in several forms. The Council also records its thanks to the British Council, Federal Republic of Germany, United States of America, the Rockefeller Foundation, the ECU and others for their support in several forms, such as research grants, gift of equipments, books, travel grants, collaborative project, etc. The Council further records its thanks to the Governing Body, Bose Institute, and to the members of the various Selection Committee for their help and interest.

SCIENTIFIC AND TECHNICAL REPORTS

Physics

Faculty : Prof. S. C. Roy, Prof. M. H. Engineer (Chairman, til 31.12.98), Prof. S. Raha (Chairman, 1.1.99 till date), Dr. A. K. Chatterjee (till 31.8.98), Dr. S. N. Changdar, Prof. I. Bose, Prof. D. Home, Dr. (Mrs.) B. Roy, Dr. B. K. Chatterjee, Dr. S. K. Saha, Dr. T. P. Sinha

HIGHLIGHTS

An ultrasensitive neutron dosimeter has been developed and patent application filed. Significant progress has been made in various areas, like the cosmological cold dark matter problem, CP violation and Bohmian quantum mechanics, viscosity divergence in polymeric fluids, statistical model of biological evolution and others. Several academic staff (faculty as well as research associates/ scholars) were invited to attend international conferences/workshop abroad with full financial support from the organisers. The department played a major role in organising a meeting of the National Task Force for Science at High Altitude during May 1998.

INSTITUTIONAL PROJECT VI

A. Acoustics

A.1. *Ultrasonic studies on conducting polymers :* **B. Roy, T. P. Sinha ,** Sonali Saha and S. K. Siddhanta (Dept. of Polymer Science and Technology, Calcutta University)

Ultrasonic velocity measurements using interferometric method have been made in stable aqueous solutions of conducting polymers like polyaniline (PANI) and polyorthotoluidine (POT), prepared using water soluble support polymers like polyacrylamide (PAAm) and polyvinyl pyrrolidone (PVP). Using the measured ultrasonic velocity, density and viscosity of pure polymers (PANI and PVP) and composite polymers (e.g. PANI-PAAm, PANI-PVP, POT-PAAm and POT-PVP), the different thermodynamic parameters like adiabatic compressibility, intermolecular free length and ultrasonic absorption have been determined. From analysis of the results it has been concluded that hydrogen bonding plays an important role in the molecular interactions between the conducting polymers and the support polymers (PAAm, PVP).

A.2.1 *Auditory Illusions :* **M. H. Engineer,** with the technical assistance from Subrata Das (Junior Laboratory Assistant) and help in circuit designing from Dr. Anutosh Chatterjee (RSIC).

Visual illusions affecting humans are well studied objects. for e.g., the well known Necker's Cube. The corresponding auditory case has not been effectively explored. I have conceived a new experiment which extends the well known Deutsch Illusion. Two distinct, pure tones are fed separately to the two ears for a period of, say, one quarter of a second. During the next quarter second the locations of the same pure tones are switched - what was in the right ear is now in the left and vice versa. This alternating sequence of clashing information is continued indefinitely. What is actually perceived? Only one pure tone is heard at any time and, when heard, is heard in only one ear. Both the tone heard, and the ear in which it is heard, change together abruptly i.e., when the frequency heard changes, the ear in which it is heard also changes! By providing suitable markers , electronically, we are now trying to resolve whether the signals heard are amongst those presented at the time i.e., whether there are time delays. There are good reasons to believe, based on the work of Benjamin Libet, that time delays are a feature of human consciousness. The experiments underway will throw light on this important question.

A.2.2 *Fluid Mechanics :* **M. H. Engineer**

The theory behind the viscous fingering instability reported by Engineer and Roy was explored. It was established that, when the sound source fills the tube cross-section, as in the Kundt's Tube case, convective cells spaced every one half wave length apart are formed. But, when the sound source acts only over a part of the cross section the flow pattern is more complicated. In the first case, analytic theory has been worked out, but the second remains

intractable. Some work has also been started on the stability of the steady flows.

B. Applied Radioactivity and Liquid Dynamics.

S. N. Changdar

Studies were continued to observe and explain the effect of coupled flow on diffusive motion of ions and molecules in complex liquid systems, pareticularly in solutions using P-32 as the tracer atom. The experimental data were obtained by using the sliding cell method, sheared boundary technique and capillary technique developed in our laboratory.

C. Radiation Physics and Condensed Matter

C.1. *Viscosity divergence in polymeric liquids :* **B.K. Chatterjee , S. C. Roy** and N. K. Halder

Recent experiments on the viscosity divergence with concentrations of suspended particles lead to a new concept of gelation. Further study using suspended particles of more regular shape and size has been made. Poppy seeds of average size of 0.104 cm and three varieties of mustard seeds with average size ranging from 0.13 cm to 0.24 cm have been used to measure the threshold concentrations at which the viscosity diverges. Three different grades of polydimethyl siloxane have been used. A distinct correlation between the two seemingly uncorrelated parameters, threshold concentration and the viscosity exponent, has been observed. Further investigations to understand this unexpected observation are needed.

C.2. *Neutron spectrometry using superheated drops :* **B. Roy, B. K. Chatterjee, S. C. Roy** and Mala Das

The effect of temperature variation on neutron induced nucleation in superheated drops made of R-12 exposed to Am-Be neutron source has been studied over a wide range of temperature (-17oC to 60oC) using the horizontal tube volumetric method developed in our lab. The apparatus consists of several long glass tubes with narrow bores placed horizontally on a graduated platform with one millimetre spacing. The tubes contain a coloured water column as an indicator of the volume of vapour formed on nucleation. The temperature of the sample was controlled by a sensitive temperature controller involving cold finger system developed in our lab. The temperature can be made stable within ± 0.1 oC. From this study, the results on the neutron detection efficiency of superheated drops of R-12 have been utilised to construct the neutron energy spectrum of Am-Be source. The energy resolution achievable by this method is better than the other methods tested for neutron spectrometry using superheated liquid..

C.3. *Development of ultrasensitive neutron dosimeter :* **B. Roy, B. K. Chatterjee, S. C. Roy** and Mala Das

An ultrasensitive neutron dosimeter with SDD has been developed in our lab. by using horizontal volumetric method mentioned above. This device can detect neutron dose as low as 0.1 mSv. This dosimeter does not require any power source and therefore suitable for long term measurement like area monitoring etc. A patent application for this device has been filed.

C.4. *Development of gamma ray sensitive superheated drop detector :* **S. C. Roy, B. K. Chatterjee, B. Roy** and Mala Das

A gamma sensitive superheated drop detector has been developed in our lab. The sensitivity of the said gamma detector has been tested with photons of energies in the range 60 keV to 1332 keV at room temperature (24oC). This detector has the potentiality to be used as a gamma dosimeter in area monitoring or other field applications. Its application as a gamma ray spectrometer will be explored.

C.5. *A new method of determining limit of superheat of a liquid :* **S. C. Roy, B. K. Chatterjee, B. Roy** and Mala Das

A new method of determining the limit of superheat of liquids has been developed here. A compact electronics to detect each drop vaporisation acoustically has been developed and a remote data aquisition system has been set up to automatically acquire data in a computer through MCS. The whole data acquisition system has been successfully standardized. Using this acoustic detection system, the limit of superheat of three refrigerant liquids have been found out. The agreement between the theory and the present experimental results is better than the other available methods.

C.6. *Superparamagnetic behaviour of nano-materials :* **T. P. Sinha** (in collaboration with Prof. D. Banerjee of Calcutta University, Calcutta.)

Mössbauer spectroscopic method has been used to determine the magnetic behaviour of Fe-Ni fine particles (≈ 200 Å) in the concentration range 10 to 35 at% Ni. The Mössbauer spectra of the alloys having 20, 22.5 and 25 at% Ni show the existence of a broad central line along with a sextet whereas the spectrum of Fe72.5Ni27.5 alloy shows only a broad central line. The central line in each of the above cases disappears for large particles, suggesting that this line originates from the superparamagnetic behaviour of ferromagnetic fine particles. The spectra reveal that ferromagnetic order appears in fine particles (≈ 200 Å) of Fe70Ni30 and Fe65Ni35 alloys. The experimental results suggest that the anisotropy energy changes with the change of Ni concentration in these alloys.

C.7. *Structural and ferroelectric studies of modified* lead-*lanthanum-zirconate-titanate :* **T. P. Sinha,** in collaboration with S. R. Shannigrahi, Prof. R. N. P.Choudhary and Prof. H. N. Acharya of Indian Institute of Technology, Kharagpur.

Polycrystalline samples of Pb0.92(La1-zLiz)0.08(Zr0.60Ti0.40)0.98+0.04zO3 (z = 0.0, 0.1, 0.3, 0.5, 0.7) have been prepared using the metal-alkoxide sol-gel technique. Differential thermal analysis and x-ray diffraction studies show that the above materials can be formed in a single-phase perovskite structure at 550 oC. The formation of grain structure, particle size and their distribution have been studied by scanning electron microscopy and transmission electron microscopy. Systematic studies of compositional dependence of ferroelectric properties of the compounds have been done. High value of dielectric constant ~ 104, remanent polarization (14-24 mC cm-2), pyroelectric coefficient (46-57 nC cm-2 K-1) and small piezoelectric parameters have been observed for all the compounds. The density of the sample increases with increasing Li+ concentration. A combination of donor and acceptor doping in lead-zirconate-titanate in the studied materials lead to stable ferroelectric states over a wide usable range.

C.8. *Dielectric study of barium-strontium titanate single crystal :* **T. P. Sinha** and Sonali Saha

Dielectric measurements of Ba1-xSrxTiO3 crystals grown by Remeika technique using KF flux have been performed. The tetragonal (ferroelectric) to cubic (paraelectric) transitions occur at 72 and 48 oC for x = 0.15 and 0.20 respectively. The activation energies have been determined in the paraelectric region. Since the size of Sr is smaller than that of Ba, it has been argued that the impurity atoms may take up off-centre positions, giving rise to local dipoles and random local fields.

D. High Energy Physics, Astrophysics and Cosmology

D.1.1. *High Energy Particle and Nuclear Physics :*

(a) Finite Temperature Field Theory : **Sibaji Raha** and Sanjay Kumar Ghosh, in collaboration with Dr. Abhijit Bhattacharyya of Variable Energy Cyclotron Centre, Calcutta.

The effective masses and decay widths of the pseudoscalar mesons (pion and kaon) in a hot quark medium have been studied in the framework of the Nambu-Jona-Lasinio (NJL) model. It has been observed that decay widths of both pions and kaons become very large above a common temperature of about 200 MeV, which may be interpreted as the deconfinement temperature. A paper with these results has been published.

Modification of the NJL model to include explicit gluonic degrees of freedom is currently being looked into.

(b) Microscopic Approaches to Hadronization :

Sibaji Raha and Sanjay Kumar Ghosh, in collaboration with Dr. Jan-e Alam and Dr. Abhijit Bhattacharyya of Variable Energy Cyclotron Centre, Calcutta and Prof. Bikash Sinha of Variable Energy Cyclotron Centre and Saha Institute of Nuclear Physics, Calcutta.

Stochastic pionization in a non-equilibrium, highly excited system of quarks and gluons produced in ultrarelativistic heavy ion collisions is being investigated in the framework of the Smoluchowski theory of coagulation. Work on this project is continuing. A manuscript has been submitted for publication.

(c) Hadronic Structure Functions and Quantum Chromodynamics : **Sibaji Raha** and Sanjay Kumar Ghosh.

Studies on incorporating the dynamical effects of colour confinement on hadronic structure functions have been continued. A manuscript has been submitted for publication.

Coupled equations for valence and sea quark and gluon distributions are being solved numerically.

(d) Meson mass renormalization in nuclei : **Sibaji Raha** and Sanjay Kumar Ghosh, in collaboration with Dr. Abhijit Bhattacharyya of Variable Energy Cyclotron Centre, Calcutta.

The modification of the effective mass of r meson in the ground state of light nuclei, especially *He3* , has been investigated. In the light of very recent experimental data, we have shown that the incorporation of the tensor interaction can explain the observed shift of the effective mass of the r meson, without the assumption of restoration of chiral symmetry. A manuscript with this result has been accepted for publication.

D.1.2. *Astrophysics and Cosmology - theoretical and observational :*

(a) Many-body Theory of Strongly Interacting Matter and Equation of State : **Sibaji Raha**, Sanjay Kumar Ghosh, (in collaboration with Prof. S. C. Phatak of Institute of Physics, Bhubaneswar. and Dr. Abhijit Bhattacharyya of Variable Energy Cyclotron Centre, Calcutta.)

The Walecka model for superdense hadronic matter, although very popular due to its simple structure, has many drawbacks, the most serious ones being the high incompressibility at nuclear matter density and the dropping of the baryon masses to zero at moderately high densities and temperatures. Efforts have been made to improve on these by including the mesonic corrections self-consistently. It has been shown that the inclusion of in-medium meson mass renormalization causes a substantial softening of the equation of state at higher densities. Also the variation of baryon masses with density is found to be much slower than in the original Walecka model. A manuscript with these results has been submitted for publication.

(b) Study of Strangelets in Cosmic ray fluxes : **Sibaji Raha,** Shibaji Banerjee and Sanjay Kumar Ghosh, in collaboration with Dr. Debapriyo Syam (Presidency College, Calcutta)

Propagation of strangelets in the upper atmosphere, taking account of the earth's magnetic field at the latitude of Darjeeling and Sandakphu, has been studied. Ionisation loss as well as charge and mass capture reactions have been included. A manuscript has been accepted for publication and another one submitted for publication. Relativistic treatment of the process is being carried out.

(c) QCD Phase transition in the early universe and baryonic dark matter : **Sibaji Raha,** in collaboration with Dr. Jan-e Alam, Dr. Abhijit Bhattacharyya, Dr. Pradip Roy and Mr. Saurav Sarkar of VECC, Calcutta, Prof. Bikash Sinha of VECC and SINP, Calcutta and Dr.Pijushpani Bhattacharjee of Indian Institute of Astrophysics, Bangalore.

The size distribution of primordial strange quark nuggets, formed in the cosmological QCD phase transition, has been calculated and shown to peak at a baryon number which is comfortably above the threshold for stability on cosmological time scales. It has been demonstrated that the surviving nuggets can indeed account for all of the cold dark matter. For the evaporating nuggets, it has been shown that neutrino inflation washes out the density inhomogeneity sufficiently fast so as to leave the prediction of Big Bang nucleosynthesis essentially unchanged. A manuscript has been submited for publication.

D.2.1. *Bohmian Approach to Quantum Mechanics and CP-Violation Experiments :* **Dipankar Home** in collaboration with Dr. A.S. Majumdar , S.N. Bose National Centre for Basic Sciences, Calcutta.

We argue that the inference of CP violation in experiments involving the k0-k-0 system in weak interactions of particle physics is dependent on the assumption of "particle trajectories" for the decaying particles and the decay products. A consistent explanation in terms of such trajectories is naturally incorporated within the Bohmian model of quantum mechanics.

D.2.2. *Testing a Dynamical Model of Wavefunction Collapse in the Cosmological Scenario :* **Dipankar Home,** in collaboration with Dr. A.S. Majumdar, S.N.Bose National Centre for Basic Sciences, Calcutta.

We consider the continuous spontaneous localization (CSL) of baryonic wavefunctions using a dynamical model of wave function collapse. The total value of the associated energy liberation which contributes to the energy density of the universe in the form of non-baryonic dark matter is calculated from the time of baryogenesis up to the present era. Taking into account the standard cosmological expansion, it is found that for a suitable choice of parameters, the CSL contribution matches the critical density of the universe.

D.2.3. *Quantum Zeno Effect and Quantum Nonlocality :* **Dipankar Home,** in collaboration with Dr. M.A.B. Whittaker of Dept. of Pure and Applied Physics, The Queen's University of Belfast, UK.

We study the hitherto unexplored possibility of using Quantum Zeno Effect to uncover a new type of quantum effect embodying a certain nonclassical "action at a distance".

D.2.4. *Broadcasting Quantum Entanglement :* **Dipankar Home,** Somshubhro Bandyopadhyay, (in collaboration with Dr. Guruprasad Kar, Indian Statistical Institute, Calcutta.)

Broadcasting of entanglement is studied where we use universal quantum cloners (in general less optimal) to perform local cloning operations. It is shown that there is a lower bound on the fidelity of the universal quantum cloners that can be used for broadcasting. We prove that an entanglement is optimally broadcast only when optimal quantum cloners are used for local copying. We also show that broadcasting of entanglement into more than two entangled pairs is forbidden using only local operations.

D.2.5. *Disentanglement by Local Cloning :* Somshubhro Bandyopadhyay, in collaboration with Dr. Guruprasad Kar and Mr. Anirban Roy, Indian Statistical Institute, Calcutta

Disentanglement of pure bipartite quantum states is shown to be possible within the framework of the schemes developed for entanglement splitting and broadcasting of entanglement.

D.2.6. *The Geometric Phase :* **M. H. Engineer**

The work of Maslov and Fediruk was studied. It relates to the general understanding of *semiclassical* quantum physics. I hope to be able to use the available techniques to study in detail the behavior of the Geometric Phase in this limit and also to further understand quantum transitions that appear when the classically approximated dynamical variables of a part of an interacting system , approach points which make the energy levels of other, quantum, parts of the system degenerate.

D.3.1. *Strongly correlated systems :* **Indrani Bose** and Emily Chattopadhyay

Recently, some mixed-spin antiferromagnets have been discovered which exhibit an unusual phenomenon. Below an ordering temperature, long range antiferromagnetic order coexists with Haldane-gap (HG) excitations. We have constructed a spin model for which this can be shown in a rigorous manner. The model describes a system of spins of magnitude 1/2 and 1. Using the method of matrix products, we have been able to determine the ground state exactly. The coexistence of long-range order and HG excitations is also shown explicitly. Studies have been initiated on the SO(5) theory of strongly correlated systems. This theory predicts the coexistence of superconductivity and antiferromagnetism and may be relevant for some heavy fermion systems. Work on spin ladders doped with holes is continuing.

D.3.2. *Cross-disciplinary Physics :* **Indrani Bose** and Indranath Chaudhuri

Recently, an experiment on biological evolution has been performed which shows clear signatures of punctuated equilibrium (PE). The death and replication rates of bacteria are high, so in four years, bacteria evolves through 10000 generations. In the experiment, part of the bacterial population was removed every fifteen days and kept frozen. Two quantities that were monitored are the average cell size and the replication rate. The experimental data show that these two quantities remain more or less the same over two hundred generations and then exhibit a rapid increase. Periods of stasis (~ 200generations) punctuate short periods of rapid

growth. We have modified the well-known Bak-Sneppen model of biological evolution and have shown that this modified model can reproduce the qualitative features of the experimental curves.

Work on reaction-diffusion systems is in progress.

PUBLICATIONS

1. Chatterjee BK and Roy SC 1998 Tables of elastic scattering cross sections of photons in the energy range 50 keV to 1500 keV for all elements in the range 13<Z<104 ; *Journal of Physical and Chemical Reference Data* **27** 1011.

2. Carney JPJ, Kissel Lynn, Pratt RH, Roy SC and Sengupta SK 1998 Elastic photon scattering from excited states of atoms and ions ; *Radiation Physics and Chemistry* **51** 371.

3. Roy B, Chatterjee BK, Das Mala and Roy SC 1998 Study of nucleating efficiency of superheated droplets by neutrons ; *Radiation Physics and Chemistry* **51** 473.

4. Roy B, Chatterjee BK and Roy SC 1998 An accurate method of measuring lifetime of superheated drops using differential manometer ; *Radiation Measurements* **29** 173.

5. Chatterjee BK, Das Mala and Roy SC 1998 Tabulations of elastic scattering cross sections of all elements 13<Z<100 in the energy range 50 keV to 1000 keV ; *Radiation Physics and Chemistry* **51** 369.

6. Das Mala, Roy B, Chatterjee BK and Roy SC 1998 Superheated drops of Freon-502 and Freon 21 as neutron detector ; *Proc. of DAE Symposium on Nuclear Physics (India)* **41B** 402.

7. Das Mala, Roy B, Chatterjee BK and Roy SC 1999 Efficiency of neutron detection of superheated drops of Freon-22 ; *Radiation Measurements* **30** 35.

8. Roy B and Engineer MH 1998 A new viscous fingering instability : the case of forced motions perpendicular to the horizontal interface of an immiscible liquid pair ; *Indian Journal of Physics* **72A** 1.

9. Saha Sonali, Siddhanta SK, Roy B and Sinha TP 1998 Ultrasonic studies on stable aqueous polyaniline prepared using water soluble support polymer ; *Journal of the Acoustic Society of India* **XXVI** 97.

10. Chakrabarty D, Santra S C and Roy B 1998 Survey of community annoyance due to traffic noise exposure in Calcutta metropolis ; *Journal of the Acoustic Society of India* **XXVI** 39.

11. Santra S C, Chakrabarty D and Roy B 1998 Urban traffic noise abatement with vegetational barriers ; *Journal of the Acoustic Society of India* **XXVI** 1.

12. Kundu TK, Mukherjee M, Chakravorty D and Sinha TP 1998 Growth of nano-a-Fe2O3 in a titania matrix by the sol-gel route ; *Journal of Material Science* **33** 1759.

13. Kumar S, Roy K, Maity K, Sinha TP, Banerjee D, Das KC and Bhattacharya R. 1998 Superparamagnetic behaviour of Fe-Ni alloys at low Ni concentration ; *Physica Status Solidi (A)* **167** 175.

14. Shannigrahi SR., Choudhary RNP, Acharya HN and Sinha TP 1999 Characterizations of sol-gel grown (PbLaLi) (Zr0.60Ti0.40)O3 ; *Journal of Applied Physics* **85** 1713.

15. Shannigrahi SR., Choudhary RNP and Sinha TP 1998 Phase transition in sol-gel derived (Pb-La-K) (Zr Ti) 03 ceramics ; *Solid State Physics (India)* **41** 149.

16. Kumar Viresh, Pattabiraman NS, Das D, Ghugre SS, Sinha TP and Chintalapudi SN 1998 Effect of 40 Mev alpha particles on hexagonal boron-nitride ; *Solid State Physics (India)* **41** 197.

17. Saha Sonali, Sinha TP and Majumdar CK 1998 Mössbauer study of Fe (2+) ions in hypersthene ; *Solid State Physics (India)* **41** 448.

18. Ghosh SK 1998 Strangeness, neutrinos and neutron stars, in : *Science at High Altitudes* (Eds. S Raha, PK Ray and B Sinha) ; *Allied Publishers New Delhi,* 101.

19. Bhattacharyya A and Ghosh SK 1998 Model study of hot and dense baryonic matter ; *International Journal of Modern Physics* **E7** 495.

20. Ghosh SK and Phatak SC 1998 H-Dibaryon in chiral color dielectric model ; *Physical Review,* **C58** 1714.

21. Ghosh SK 1998 Kaon condensation superdense baryonic matter, in *Physics and Astrophysics of Quark - Gluon Plasma (ICPA-QGP'97)* ; pp 515 (eds. B Sinha, DK Srivastava and Y. P. Viyogi), *Narosa Publishing, New Delhi.*

22. Raha S 1998 The QCD phase transition in the early universe ; in *Physics and Astrophysics of Quark - Gluon Plasma (ICPA-QGP'97)*, pp 223 (Eds. B Sinha, DK Srivastava and YP Viyogi), *Narosa Publishing, New Delhi.*

23. Alam J, Raha S and Sinha B 1998 Closing the universe with primordial quark blobs ; *Nuclear Physics* **A638** 523.

24. Alam J, Raha S and Sinha B 1999 Quark nuggets as baryonic dark matter ; *Astrophysical Journal* **513**, 572.

25. Bhattacharyya A, Ghosh SK and Raha S 1999 Pion and Kaon dissociation in hot quark medium ; *Modern Physics Letters* **A14,** 621.

26. Home Dipankar and Majumdar A.S. 1998 Testing a Dynamical Model of Wave Function Collapse in the Cosmological Scenario ; *Quantum Coherence and Decoherence*, pp 291 (Eds. K Fujikawa and YA Ono) *Elsevier, Amsterdam,*

27. Home Dipankar and Whittaker MAB 1998 Quantum Zeno Effect : Relevance for Local Realism ; *Physics Letters* **A 239** 6.

28. Home Dipankar and Majumdar AS 1999 On the Importance of the Bohmian Approach for Interpreting CP-Violation Experiments ; *Foundations of Physics (USA)* **29** 7.

29. Bose I 1998 Quantum magnetism: novel materials and phenomena ; *Indian Journal of Physics* **72A** 1.

30. Bose I 1998 Models with spin-gap : some exact results ; *Communications in Theoretical and Mathematical Physics* **1** 219.

31. Przewoski BV, Daehnick WW, Doskow J, Dzemidzic M, Flammang R, Lorentz B, Meyer HO, Pancella PV, Pollock RE, Quin P, Rinckel T, Saha SK, Schwartz B, Sperisen F, Thörngren-Engblom P, Ugorowski P and Wise T 1998 Polarization lifetime near an induced depolarizing resonance ; *Review of Scientific Instruments* **69** 3246.

32. Meyer HO, Balewski JT, Dzemidzic M, Doskow J, Pllock RE, Przewoski BV, Thörngren-Engblom P, Wolanski M, Haeberli W, Lorentz B, Rathmann F, Schwartz B, Wise T, Daehnick WW, Flammang RW, Saha SK, Tedeschi DJ and Pancella PV 1998 Dependence of pp ® pp p^0 near threshold on the spin of the colliding nucleons ; *Physical Review Letters* **81** 3096.

Books :

Raha S, Ray PK and Sinha B (Editors), 1998 **Science at High Altitudes,** Allied Publishers, New Delhi.

Participation in Conferences/Symposia/ Training Programmes/Awards and Honours received :

Prof. S.C. Roy presented an invited talk in the European Conference on Dispersive X-ray Spectrometry (EDXRS98) held at Bologna, Italy during June 7-12, 1998, chaired one technical session in the EDXRS 98 held at Bologna, Italy, presented an invited talk in the National Conference on Recent Trends in Modern Physics (NCRTMP99) held at Kollam.

Prof. M. H. Engineer Participated in the Sixtieth Birthday Conference in honor of V. Singh held at TIFR , December 1998, participated in the Centenary Celebrations of the American Physical Society, March 1999 at Atlanta Georgia, Spent two weeks on invitation at the Depertment of Physics, University of South Carolina in collaborative research with Prof. Jeeva Anandan during March 1999, delivered an invited seminar on *"The geometric phase in electron-phonon systems"* at the Physics Dept., University of South Carolina during March 1999, delivered an invited seminar on "Auditory Illusions" at the Physics Depertment, Viswabharati University, Shantiniketan in August 1998.

Prof. Sibaji Raha acted as the Member Secretary for the National Task Force Science at High Altitude. Organised and participated at a meeting of the Task Force in Darjeeling during May 1998, attended and chaired a session at the national workshop on Relativistic Heavy Ion Collision at Institute of Physics, Bhubaneswar during May 1998, served in the TPSC Calcutta Centre sub-committee as a member.

Prof. Indrani Bose was invited to give a talk at the International Conference on Quantum Magnetism and Strongly Correlated Systems in I.I.Sc., Bangalore (September 1998), invited to give four lectures at the School on strongly

correlated systems at I.I.T., Kharagpur (October 1998), invited to give a talk at the International Symposium on Recent Trends in Theoretical Physics held at T.I.F.R., Mumbai (January 1999), invited to give a talk at the International Conference, Statphys III, held at the S.N.Bose National Centre, Calcutta (January 1999), invited to give a talk at the Indo-Israeli Workshop on Condensed Matter Physics and Materials Science, organised by INSA and held at New Delhi (February 1999), invited to give six lectures in the Refresher Course for College Teachers, organised by Calcutta University (March 1999), have taught in the M.Sc. Solid State Physics Course of Calcutta University (March-April 1999), invited to give a talk at the one-day Workshop on Protein Folding held at S.I.N.P., Calcutta (1999), invited to give a talk at the Workshop on Bioinformatics, organised by D.I.C., Bose Institute (1999)

Prof. Dipankar Home visited School of Mathematics, Macquarie University, Sydney, Australia from 24 Sept. - 30 Oct. 1998 for research collaboration and delivered a series of Lectures on the Foundational Issues of Quantum Mechanics at Macquarie University.

Dr. S. N. Changdar attended and presented a paper at the National Symposium on Acoustics (NSA-98), held in Calcutta, attended the Seventeenth National Symposium on Cryogenics at Nagpur University, Nagpur, Feb. 19-21, 1999 and was invited to chair a scientific session.

Dr. B. Roy presented a paper in the symposium, **"Condensed Matter Days - 98"** held at T. M. Bhagalpur University, Bhagalpur during August 27 - 29, 1998, presented papers in the "National Symposium on Acoustics - 98" held at IACS, Calcutta during Dec. 18-20, 1998.

Dr. T. P. Sinha participated in the International School on Powder Diffraction held at Indian Association for the Cultivation of Science, Calcutta from 7th to 10th October 1998, presented four papers in the meeting "Condensed Matter Days - 1998" held at Department of Physics, Bhagalpur University, Bhagalpur from 27th to 29th August 1998, presented three papers in the Annual DAE Solid State Physics Symposium held at Department of Physics, Kurukshetra University, Kurukshetra from 27th to 31st December 1998.

Dr. Sanjay Kumar Ghosh participated in the Task Force meeting of Science at High Altitude at Darjeeling during May 1998, participated in the national workshop on Relativistic Heavy Ion Collision at Institute of Physics, Bhubaneswar during May 1998 and delivered a talk, participated in the International workshop on quantun fields in & out of equilibrium, at Brookhaven National Laboratory, USA during October 1998, visited University of Maryland, USA during November 1998 and delivered a seminar, visited University of Virginia, USA during November 1998 and delivered a seminar, visited Indiana University, USA during November 1998 and delivered a seminar.

Mala Das participated in III SERC school on Nuclear Physics held at VECC, Calcutta during Nov. 2 -21, 1998.

Sonali Saha participated in the International School on Powder Diffraction held at Indian Association for the Cultivation of Science, Calcutta from 7th to 10th October 1998, presented two papers in the meeting "Condensed Matter Days - 1998" held at Department of Physics, Bhagalpur University, Bhagalpur from 27th to 29th August 1998.

Ph.D. Awarded:

Name	University	Year	Title	Supervisor
Abhijit Bhattacharjee	Jadavpur University	1998	Finite Temperature field theory and its phenomenological applications	Prof. Sibaji Raha
Aniruddha Deb	Jadavpur University	1998	Experimental and theoretical determination of Compton profile of compounds and alloys	Dr. Arun Kumar Chatterjee
Debashis Chakraborty	University of Kalyani	1998	"Studies on road traffic noise, its impact responses and planning for future noise reduction in Calcutta metropolis"	Dr. B. Roy, jointly with Prof. S. C. Santra of Dept. of Ecological Studies, School of Environmental Science, University of Kalyani.

RESEARCH SCIENTISTS/RESEARCH ASSOCIATES/POST-DOCTORAL FELLOWS/ SENIOR RESEARCH FELLOWS/JUNIOR RESEARCH FELLOWS

Dr. Sanjay Ghosh, Aniruddha Deb (till 13.12.1998), Mala Das, Indranath Chaudhuri, Shibaji Banerjee, Som Shubhro Bandyopadhyay, Sonali Saha, Emily Chattopadhyay, Susim Maity (from 15.6.98 till 30.10.98), Mousumi Ghosal (from 10.06.98 till 28.2.99), Ujjwal Sen.

SUPPORTING STAFF MEMBERS

N. C. Dey, A. K. Ganguly, K. C. Das, S. P. Singha, Shyam Sundar Mallick, Subhasis Banerjee, Manas Datta, Subrata Das, Tarun Kumar Maji, B. P. Ram, B. Dutta, D. K. Thakur, Kissan Das, S. K. Basu, R. K. Mourya, Prabir Halder.

Chemistry

Faculty : Prof. P. Chakrabarti, Prof. A. Ghosh, Prof. P. Bhattacharyya, Prof. Parimal C. Sen (Chairman), Dr. Manas Chakrabarty, Dr. Suniti Misra, Dr. K. P. Das, Dr. Joyoti Basu, Dr. Manikuntala Kundu, Dr. Pradip Das.

HIGHLIGHTS

Clinical trial of Eicosapentaenoic Acid (EPA) on volunteer patients suffering from cardiovascular disorders has been started. A new protein kinase (PKx) has been purified and characterised from goat testis is seemed to be involoved in fertility regulation. Anti bacterial and anti diabetic compounds have been isolated from Indian Medicinal plants. Mechanisms of drug resistance in *Shigella dysenteriae* and mycobacteria have been proposed. The mechanism of molecular chaperone activity of α-crystallin at different temperatures has been studied.

INSTITUTIONAL PROJECT II

A. *Stuides on the liver lipids and fatty acids of the Sting Ray, Dasyatis bleekeri (Blyth) :* **Amitabha Ghosh**, Debasish Pal, Dipankar Banerjee, Tarun K. Patra and Amarendra Patra

The Sting ray, *Dasyatis bleekeri* (Blyth) has been studied for lipids and fatty acids of its liver. The neutral lipids identified were hydrocarbons, wax esters, sterol esters, 1-*O*-alkyl-2,3-diacylglycerols, triacylglycerols, and sterols. Neutral lipids were predominant (91.8%), major components being triacylglycerols (92.7%). Polyenoic fatty acids of n-3 series, viz. EPA and DHA, were high in the phospholipid and neutral lipid fractions. Cholesterol was the major component (67.9%) in the steryl ester fraction. Glyceryl ethers, with chainlengths upto 30 carbons, were recorded with unsaturated, anteiso, iso, and normal chains. In wax ester alcohols, upto 32 carbon chains were recorded. Hydrocarbons were upto 36 carbon chains with anteiso, iso, and normal chains. Among branched chain hydrocarbons, pristane was the major component (6.7%) and squalene was present at the level of 3.5%. Chimyl and batyl alcohol backbones were the major components found in 1-*O*-alkyl-diacylglycerols.

B. *Analysis of fatty acid composition and oil content in different tissues of Mango to elucidate the high stearate deposition in its fat :* **Prantosh Bhattacharyya** and Aniruddha Aich

Mango is a non-traditional oil seed plant with unusually high stearate content (~ 40%) in its matured seed. To elucidate the nature of high stearate deposition in Mango seed kernel, analysis were carried out in different developmental stages of mango seed lipid. The analysis showed that from immediate stage, the stearic acid content is very high (~ 30-35%). The main fatty acid constituents are C18:0 (stearic acid) and 18:1 (oleic acid). It is found that average oil content is 1-3%. In tissues like bud, flower leaves, the stearic acid accumulation is very much lower (4-6%) but main constituents are palmitic acid (40%) and linoleic acid (~20%). This shows that major organ for synthesis and storage of stearate rich fat is seed tissue.

INSTITUTIONAL PROJECT III

A. *Lipid peroxidation in human uterine tumors :* **Parul Chakrabarti** and Subhansu Ray

Our work in this area shows that altered lipid peroxidation in human uterine tumors is due to loss of a delicate balance between the available substrates (polyunsaturated fatty acids) and inhibitors (a-tocopherol, protein thiol, glutathione etc.). Low levels of substrates and high levels of inhibitors may account for low susceptibilities of the uterine tumors to lipid peroxidation, thus corroborating the general hypothesis that a decreased rate of lipid peroxidation may be related to an increased rate of cell division in the undifferentiated rapidly proliferating system.

B.1.A *75 kilodalton competitive inhibitor protein of Na^+, K^+-ATPase from rat brain cytosol binds to a site different from ouabain-binding site :* **Parimal C. Sen**, Koushik Roy and Atin K. Mandal

A sodium, potassium-ATPase inhibitor protein has been purified to homogeneity from rat brain cytosol by ammonium sulphate precipitation, DEAE anion-exchange chromatography and hydroxyapatite adsorption column chromatography. The purified protein migrates as a single polypeptide band of 75 kDa on 7.5% SDS-PAGE. Amino acid composition data shows the presence of high amount of acidic amino acids in its molecule relevant to its pI value of 4.6. The inhibitor binds Na^+, K^+-ATPase reversibly and blocks ATP binding sites at micromolar range with an I_{50} of about 700 nM. As a result, phosphorylated intermediate formation of Na^+, K^+-ATPase is hindered in presence of the inhibitor. It does not affect p-nitrophenylphosphatase activity. Tryptophan fluorescence study and CD analysis suggest conformational changes of Na^+, K^+-ATPase on binding to the inhibitor.

B.2. *Role of protein kinase C in regulation of membrane bound guanylate cyclase activity stimulated by E. coli heat stable enterotoxin (STa) in rat intestinal epithelial cells :* **Parimal C. Sen**, Koushik Roy, Atin K. Mandal and Arindam Basu (in collaboration with Alok Ghoshchoudhury, Chandan Jana and Uma Ganguly of NICED, Calcutta)

STa inhibits the inward ion transport and stimulates the secretion of water and chloride into the small intestinal lumen by activating the membrane-bound guanylate cyclase type C (GC-C). Previous studies from our laboratory have demonstrated that apart from activating the membrane-bound GC-C, STa action involves a chain reaction associated with the rise of cytosolic Ca^{2+} and stimulation of Ca^{2+} and diacyl glycerol (DAG)-dependent protein kinase C (PKC) activity.

In the present study, we have demonstrated further the translocation of PKC from cytosol to the membrane after incubation of the epithelial cells with STa. Thereafter, a gradual loss of both membrane-bound and total PKC activities were noticed upto 5 min, indicating that the PKC was down-regulated in parallel with the sustained rise of $[Ca^{2+}]i$. On the other hand, staurosporine and H7 besides inhibiting the PKC activity in STa-treated cells, significantly reduced the STa-stimulated cyclic GMP formation, while phorbol ester (b-PMA) potentiated both STa-stimulated PKC response and GC activity. All these studies suggested the involvement of PKC in STa-induced cyclic GMP response in rat enterocyte.

B.3. *Potential synthetic peptides as substrates of protein kinase C :* **Parimal C. Sen**, Koushik Roy and Atin K. Mandal (in collaboration with F. Hudecz of Eotvos Lorand University, Budapest, Hungary)

Protein kinase C is believed to phosphorylate 'Ser' and 'Th' residues of proteins and peptides. Two synthetic peptides of the sequences (I) VRKGTLRRL and (ii) VRKGSLRRL (both are modified PKC specific substrate, VRKRTLRRL) are found to be phosphorylated by PKC isolated from rat brain. VRKGTLRRL is phosphorylated more than VRKGSLRRL and is comparable with VRKRTLRRL. However none of them competes with the phosphorylation of histone by PKC. The result suggests that though PKC phjosphorylates 'Ser' and 'Th', the amino acid sequence of proteins/peptides are important in controlling the extent of phosphorylation by PKC.

C. *Synthetic Heterocyclic Chemistry*

C.1. *Newer Syntheses of Pyrrolocarbazoles :* **Manas Chakrabarty**, Shampa Khasnobis and Nandita Ghosh

In continuation of our ongoing work on the development of new synthetic routes to pyrrolocarbazoles, 9-methyl-3-formylcarbazole, reported last year, was condensed with ethyl azidoacetate to produce the corresponding arylidene azidoester. Thermal cyclisation of this substrate in different solvents is being attempted.

C.2. *Hetero Diels-Alder Reactions :* **Manas Chakrabarty** and Shampa Khasnobis

In continution of our ongoing work on the hetero Diels-Alder route to the synthesis of condensed heterocycles, the reaction of the two indolic azadienes, stated in previous Annual Reports, was extended to two other dienophiles, viz. diethyl azodicarboxylate and methyl propiolate. Some of the products have been identified and the rest are being characterised. None of the identified products is the desired Diels-Alder adduct.

C.3. *Studies on Reactions of Carbazoles :* **Manas Chakrabarty** and Shampa Khasnobis

With a view to finding out a plausible mechanism of the formation of the products from the base-catalised reaction of carbazoles with formalin, stated in the Annual Report of 1996-97, the reactions were repeated with paraformaldehyde instead of formalin. Altogether different results were obtained. Attempts are being made to evolve a mechanism which could be compatible with the two sets of results.

D.1. *Studies on molecular chaperones :* **K.P. Das** and J. Bhattacharyya

All the molecular chaperones known till date are well organised, folded protein molecules whose subunit structure and three dimensional organisation are believed to play key role in the mechanism of substrate recognition and assistance in protein folding. We have reported for the first time that α_s-casein, abundant in mammalian milk, having no well defined secondary and/or tertiary structure, chaperones *in vitro* a variety of unrelated proteins/enzymes against heat, chemical or light induced stress. We have shown that α_s-casein unlike other chaperones is highly surface active and also possesses some features of cold shock proteins.

We have also made an attempt to study the reversible folding and unfolding reactions of a 800 kDa multimeric protein α-crystallin. The fast folding/unfolding pathway of α-crystallin follows a two-state process without populated intermediates. To further investigate the kinetics of secondary, tertiary and quarternary structure formation during refolding, stopped-flow kineties studies are in progress.

D.2. *Secondary structure of aggregated proteins :* **K.P. Das** and S. Bhattacharyya

Attempts are being made to study the secondary structure of the aggregates of substrates formed under various stress condition such as heat, oxidative stress, exposure to uv light etc. by FT-IR spectroscopy.

E. *Structure and function of the human erythrocyte membrane :* **Joyoti Basu**, Raja Bhattacharyya, Biswajit Pal and Prasun Moitra

Evidence accumulated over the years suggests that human erythrocyte membrane protein 4.2 is one of the proteins involved in strengthening the cytoskeleton-membrane interactions in the red blood cell. Deficiency of protein 4.2 is linked with a variety of hereditary hemolytic anemia. However, the interactions of protein 4.2 with other proteins of the erythrocyte membrane remain poorly understood. The major membrane binding site for protein 4.2 resides on the cytoplasmic domain of band 3 (CDB3). In order to characterize the domain involved in interaction with the CDB3, protein 4.2 was subjected to proteolytic cleavage and gel renaturation assay, and the 23 kDa N-terminal domain was found to interact with band 3. This domain contained two putative palmitoylable cysteine residues, of which cysteine 203 was identified as the palmitoylable cysteine. Recombinant glutathione S-transferase (GST)-fusion peptides derived from this domain were characterized with respect to their ability to interact with the CDB3. While these studies do not rule out the involvement of other subsites on protein 4.2 in interaction with the CDB3, the evidence suggests that the region encompassing amino acid residues 187 to 211 is one of the domains critical for the protein 4.2-CDB3 interaction. This is also the first demonstration that palmitoylation serves as a positive modulator of this interaction.

F.1. *Lyotropic Polymorphism in F-actin :* **Pradip Das**, J. Roy and N. Mitra

F-actin exhibits an extended lyotropic polymorphism which includes an athermal nematic phase, a thermal cholesteric phase, a couple of thermal chiral smectic phases and a smectic B phase. Optical rotatory dispersion studies have been completed on the cholesteric phase. The birefringence has been quantitated, and order parameter calculations are now under way.

F.2. *Molecular Model Calculation of the nematic to smectic C transition in F-actin :* **Pradip Das** and N. Chakrabarti

McMillan molecular model of the smectic-A phase has been extended to the smectic-C case. The phase sequence being studied is as follows : Isotropic → Nematic → Smectic A→ Smectic C. Order parameter vs temperature variations are being numerically calculated for different compositions. Entropy and heat capacity variations with temperature will be calculated next.

INSTITUTIONAL PROJECT V

A. *Drug targets and drug resistance mechanisms in mycobacteria :* **Parul Chakrabarti, Joyoti Basu, Manikuntala Kundu**, Jasimuddin Ahamed, Sanjib Bhakta, Susmita Sen, Baisakhee Saha Chaudhuri and Anuradha Bhaduri

(i) Isoniazid accumulation in Mycobacterium smegmatis is modulated by proton motive force-driven and ATP-dependent extrusion systems

Resistance to isoniazid (INH), a frontline, antituberculosis drug, presents a major problem in the chemotherapy of tuberculosis. Although several targets of INH have been identified, the mechanism of INH resistance remains incompletely understood. We demonstrate that INH accumulation in *Mycobacterium smegmatis* is enhanced both upon addition of a proton motive force (pmf) uncoupler, carbonylcyanide *m*-chlorophenylhydrazone (CCCP) and upon addition of *ortho*-vanadate, an inhibitor of ATP-dependent efflux pumps. Both the $\Delta\Psi$ and ΔpH components of the pmf are likely to be involved as judged by the effects of valinomycin and nigericin respectively. Reserpine, an inhibitor of the human MDR1 P-glycoprotein, enhances INH accumulation in a manner similar to *ortho*-vanadate. Verapamil, a calcium channel blocker, also enhances INH uptake. Taken together, the results provide evidence of the involvement of both pmf- and ATP-dependent extrusion systems in INH efflux in *M. smegmatis*, making it important to evaluate the role of such systems in INH resistance in pathogenic mycobacteria.

(ii) Cloning and overexpression of the components of a putative doxorubicin efflux pump of Mycobacterium tuberculosis

The *drrAB* genes encode DrrA and DrrB proteins which together function as a doxorubicin efflux pump, and share characteristics similar to the P-glycoprotein multidrug transporter. The DrrA protein harboring the nucleotide-binding domain of this ABC-type transporter has been cloned and overexpressed in *E. coli*. Its characterization is in progress.

(iii) Cloning, overexpression and purification of the high molecular mass penicillin-binding protein 1 of Mycobacterium tuberculosis

One of the two high molecular mass PBPs of class A of *M. tuberculosis* has been amplified by PCR from a cosmid carrying this gene. The gene has been sequenced and introduced into the vector pET28a. It has been expressed as a His-tag PBP, purified on a Ni^{2+}-affinity column, and characterized with respect to its penicillin-binding properties. It represents one of the β-lactam-sensitive targets of this organism. Its detailed characterization is in progress.

B.1. *Antidiabetic activity of an Indian Medicinal Plant :* **Prantosh Bhattacharyya**, Debasish Ghosal and S. Sinha

The active hypoglycemic principle isolated from the leaves of a plant of Genus *Lagastromia* is identified to be a protein. Further studies are going on to characterize the protein.

B.2. *Hepatoprotective studies of an Indian medicinal plant :* **Prantosh Bhattacharyya**, Debasish Ghosal, S. Datta and S. Sinha

The herbal protein CI-1, purified from a leguminous plant *Cajanus indicus* showed dose dependent (1.5 - 6.0 mg/Kg x 7 days) protective activity on isolated hepatocytes (*ex vivo*) against β-galactosamine HCl induced hepatic damage in rats. It enhanced the percent viability of the hepatocytes following β-galactosamine treatment.

B.3. *Immunomodulatory effect of the rodent bone marrow cytokine BIM in human carcinoma :* **Prantosh Bhattacharyya** and Debasish Ghosal

The immunosuppressive role of advanced carcinoma is now well documented. The low WBC count paves the pathway for opportunistic infections. Moreover, chemotherapy could not be initiated also in advanced malignancy due to immunosuppression. To upregulate the suppressed immune system Granulocyte Macrophage-Colony Stimulating Factor is now in use with certain limitations. Administration of the non-species specific cytokine BIM has shown definitely in all the 12 patients under trial that leukopoiesis and neutropoiesis occur within 96 hours preventing infections of lungs and g.i. tract. The high WBC count makes the patient competent for chemo and/or radiotherapy. Results indicate that BIM, as adjuvant to chemo and/or radiotherapy could down regulate tumor growth spuially in brain, spleen, uterus, liver and lung.

B.4. *Immunomodulatory effect of two herbal preparation in patients suffering from tuberculosis :* **Prantosh Bhattacharyya**, D. Ghoshal, J. Chattopadhyay and P.K. Debnath

The clinical improvement observed within 28 days in patients suffering from tuberculosis and treated with herbal preparations and ATD has shown a definite immunological basis over those treated only with ATD. The experimental group do not suffer from liver dysfunction as is observed in those only on ATD. Significant weight gain was observed and these experimental patients also showed absence of T.B. bacteria in sputum within 28 days and when compared to control only on ATD.

C.1. *Microbial Transformation of Condensed Heterocycles :* **Manas Chakrabarty** and Subho Ghosh (in collaboration with Prof. P.K. Chakrabartty, Department of Microbiology, Bose Institute)

As a pre-requisite to the microbial transformation of pyrrolocarbazoles - our ongoing D.B.T.-P.D.F. programme - a few carbazoles were subjected to several species of fungi. None of the substrates was found to furnish any transformation product.

C.2. *Effect of the anti-depressant drug, lithium chloride, on the chromaffin cells of domestic pigeon : an ultrastructural finding :* **Manas Chakrabarty** and Subho Ghosh (in collaboration with Prof. Asok Ghosh and Dr. (Mrs.) Santasri Sengupta of the Department of Zoology, University of Calcutta)

In continuation of our earlier work on the histophysiology of adrenal medulla during lithium chloride treatment in domestic pigeons (*Columba livia*), we further studied the ultrastructure of chromaffin cells by TEM. A drastic depletion of catecholamine granules and a degeneration of their membranes were observed, which further demonstrated the deleterious effect of lithium chloride on the adrenal medulla.

D. *Drug resistance mechanisms of Shigella dysenteriae :* **Manikuntala Kundu**, Jasimuddin Ahamed and Jayanti Gangopadhyay

(i) *An extended spectrum β-lactamase from a clinical isolate of S. dysenteriae*

β-lactamases are primary responsible for β-lactam resistance in Gram-negative bacteria. We have cloned, sequenced and characterized an extended spectrum beta-lactamase capable of hydrolysing several cephalosporins, from a clinical isolate of *Shigella dysenteriae.* Nucleotide sequencing of the plasmid harboring this β-lactamase of *S. dysenteriae* revealed an open reading frame encoding an SHV-type β-lactamase termed SHV-11.

This β-lactamase has one single mutation at position 35 (leu→gln), compared with the SHV-11 enzyme (which has a narrow substrate spectrum). This mutation leads to the SHV-11 enzyme hydrolysing oxacillin and oxyiminocephalosporins. There is little similarity of the nucleotide sequence upstream of the -10 region of the promoter with that of the prototype SHV-1 *bla* gene. Changes in the -35 and -10 regions have strong influence on the promoter strength. In *E. coli*, the promoter is stronger the closer the sequence is to the consensus (-35 consensus sequence, TTGACA; -10 consensus sequence TATAAT). The promoter comprising 5'TTGCAA'3' (-35 box) and 5'TATTCT3' (-10 box) identified in the *S. dysenteriae* SHV-11 *bla* gene has likely influenced β-lactam susceptibilities in the present instance also.

(ii) Mechanisms of quinolone resistance in S. dysenteriae

Quinolones are widely employed to treat shigellosis. However, quinolone resistance has already been reported necessitating an understanding of the mechanisms of development of resistance. In China, upto 50% of *S. dysenteriae*, *S. sonnei* and *S. boydii* are resistant to these drugs. DNA gyrase and topoisomerase IV, the bacterial type II DNA topoisomerases, are known to be the targets of the fluoroquinolones. The increase in clinical use of quinolones, has led to the gradual emergence of resistant strains with alterations in the genes mentioned above. In Gram-negative bacteria such as *Escherichia coli*, quinolone resistance mutations occur in discrete regions termed the quinolone resistance determining region (QRDR) of GyrA and to a such lesser extent, GyrB. Similar mutations have been recently identified in analogous regions of ParC. We have studied mechanisms of quinolone resistance in several clinical isolates obtained from the International Center for Diarrheal Diseases, Dhaka, and the National Institute of Cholera and Enteric Diseases

(NICED), Calcutta. The Ser83→Leu substitution in GyrA appeared sufficient to confer high-level nalidixic acid resistance (MIC > 250 μg/ml). Four strains DS-1, DS-2, CI-1 and CI-2 (NICED) for which the norfloxacin MICs were 0.5 μg/ml, harbored the mutation Asp87-Gly in *gyrA*. None of the isolates examined had any mutations in the *parC* gene.

Accumulation of norfloxacin was studied using carbonyl cyanide *m*-chlorophenylhydrazone (CCCP) (100 μM) as proton motive force (pmf) uncoupler. Addition of CCCP increased the level of accumulation in these resistant strains to a level comparable to that of AK19520 (a sensitive strain), suggesting a role of a pmf-dependent efflux pump in the development of resistance. Since these strains lacked *gyrA* or *parC* mutations, increased efflux pump activity appears likely to be sufficient to confer 20-80-fold resistance to the fluoroquinolones compared to AK19520.

PUBLICATIONS

1. Saha Choudhuri B, Sen S & Chakrabarti P 1999 Isoniazid accumulation *in Mycobacterium smegmatis* is modulated by protonmotive force-driven and ATP-dependent extrusion system; Biochem. Biophys. Res. Commun. **256** 682-684.

2. Ghosh AS, Kar AK & Kundu M 1999 Impaired imipenem uptake associated with alterations in outer membrane proteins and lipopolysaccharides in imipenem-resistant *Shigella dysenteriae*; J. Antimicrob. Chemother. **43** 195-201.

3. Ghosh AS, Kar AK & Kundu M 1998 Alterations in high molecular mass penicillin-binding protein 1 associated with beta-lactam resistance in *Shigella dysenteriae*; Biochem. Biophys. Res. Commun. **248** 669-672.

4. Bhattacharya R, Das AK, Moitra PK, Pal B, Mandal I & Basu J 1999 Mapping of a palmitoylable band 3-binding domain of human erythrocyte protein 4.2; Biochem. J. **340** 505-512.

5. Pal D, Banerjee, D. Patra TK, Patra A & Ghosh A 1998 Liver lipids and fatty acids of the Sting Ray, *Dasyatis bleekeri* (Blyth); JAOCS **75** 1373-1377.

6. Ghoshal D, Sinha S, Sinha A & Bhattacharyya P 1998 Immunosuppressive effect of vestibulo-cerebellar lesion in rats; Neuroscience Letters **257** 89-92.

7. Datta S & Bhattacharyya P 1998 Role of bone marrow and thymus secretory protein in maintaining immune homeostasis and haemopoiesis in control and malnourished mice; Ind. J. Exp. Biol. **36** 1233-1239.

8. Ghosh R, Sen PC & Biswas S 1998 *Mimosa pudica* Apyrase requires polysaccharide and Ca for the activity; Mol. Cell Biochem. **187** 47-85.

9. Sen PC 1998 Nitric Oxide : A wonder molecule; Science & Culture **64** 265-267.

10. Roy K, Mandal AK & Sen PC 1999 A 75 kilodalton competitive inhibitor of Na^+, K^+-ATPase from rat brain cytosol binds to a site different from ouabain binding site; Eur. J. Biochem. **261** 84-89.

11. Chakrabarty M, Batabyal A & Patra A 1998 Application of phosphorous trichloride in attempted bis-indolisation of cyclohexanedione and dimedone; Indian J. Chem. **37B** 1104-1108.

12. Chattopadhyay G, Saha TK & Chakrabarty M 1998 New reactions of oximinocyanoacetamides with benzylamine : A one-pot entry into isoxazole nucleus; Indian J. Chem. **37B** 861-864.

13. Chakrabarty M, Ghosh S, Sengupta S & Ghosh A 1998 Interplay of adrenal catecholamines and phospholipids during water-deprivation stress in mice; Biogenic Amines **14** 607-614.

14. Pandit M, Khasnobis S & Sengupta D 1998 Effect of GABA Analogues on membrane parameter in experimental animal; Biomed. Res. **9** 47-54.

15. Bhattacharyya J & Das KP 1998 α-Crystallin does not require temperature activation for its chaperone - like activity; Biochem. Mol. Biol. Intl. **46** 249-258.

16. Guha S, Manna TK, Das KP & Bhattacharyya B 1998 Chaperone-like activity of tubulin; J. Biol. Chem. **273** 30077-30080.

Visit abroad

Prof. P.C. Sen visited Hungary under an Indo-Hungarian collaborative projective during April 24 - May 20, 1998.

Participation in Conferences/Symposia, Training Programmes/Awards and Honours received

Prof. Parul Chakrabarti

Chaired a session and delivered an invited talk on "Application of fluorescent technology in mycobacterial research" in the XIV National Symposium of Indian Photobiology on "Photo-induced molecular phenomena", Calcutta, October, 1998 ; Chaired a session and delivered an invited talk on "Mycobacterial Cell Envelope : A Treasure House of Drug Targets" in the National Symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules", Calcutta, February 1999 ; Delivered an invited lecture on "Molecular mechanisms of drug resistance in bacteria in the Fresher course for university and college teachers on Molecular Biology and Biotechnology" organised by the School of Life Sciences, Sambalpur University, Orissa, November, 1998 ; Delivered an invited talk on "Drug targets and drug-resistance mechanisms in mycobacteria" in the Biochemistry Department, Delhi University in October, 1998 ; Member of the Tenth Advisory Committee for 3-Semester (integrated) M. Tech. course in Biotechnology and engineering at the Indian Institute of Technology, Kharagpur.

Prof. Parimal C. Sen

Delivered invited lecture in the Hungarian Academy of Sciences at Pecs, Szeget Medical School and Eotvos Lorand Universit, Budapest, Hungary ; Co-chaired a Scientific Session in the Symposium "Computer in Medical Technology" at BITM ; Co-chaired a Scientific Session in the National Conference on "Recent Trends in Spices and Medical Plant Research" at Bose Institute ; Represented Bose Institute in DBT Group Monitoring meeting of PD Training Programme in July, 1998 at IIT, Mumbai ; Acted as Convenor to organise the National Symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules" organised by the Department of Chemistry, Bose Institute in February, 1999 ; Delivered an invited lecture at Indian Science Congress Association at Chennai in January, 1999 ; Delivered a lecture at the National symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules" at Bose Institute in February, 1999 ; Koushik Roy and Atin Mandal attended a Symposium on "Computer in Medical Technology" at BITM, Calcutta and presented a paper at the National Symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules" at Bose Institute ; Koushik Roy received the Best Poster Award in Life Science in the National Symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules".

Dr. Manas Chakrabarty

Delivered an Invited Lecture entitled, "A Peep into the Kingdom of Alkaloidal Glycosidase Inhibitors" on 14th January, 1999 in the Zoological Society at the Deopartment of Zoology, University of Calcutta ; Participated, along with Miss Shampa Khasnobis and Dr. Subho Ghosh, both of whom presented Posters, in the National Symposium on 'Recent Advances in Structure, Synthesis and Function of Biomolecules' on February 4-6, 1999 at the Department of Chemistry, Bose Institute. Miss Shampa Khasnobis obtained the Best Poster Presentation Award in Chemistry in this Symposium ; Participated in the 35th Annual Convention of Chemists held at the Karnatak University, Dharwad during November 4-7, 1999; (along with S. Khasnobis) Symposium on 'Current Awareness in Organic Chemistry' held at the Department of Chemistry, University of Calcutta on October 28, 1998; (along with S. Khasnobis and S. Ghosh) the 6th Dipta Memmorial symposium held on September 1, 1998 at Bose Institute ; As a Guest Lecturer, taught in the B. Tech. (1st Year) Course in Polymer Science & Technology, University of Calcutta and in the M.Sc. (2nd year) Course in Chemistry (Organic Special) in Presidency College, Calcutta during 1998-99 ; Acted as an Evaluator of Research Papers submitted for the Khosla Research Awards of the University of Roorkee, Roorkee ; Dr. Subho Ghosh participated in the 'Workshop on Structure and Sequence : Data Base Analysis' held at Bioinformatics Centre, Bose Institute during March 11-12, 1999.

Dr. Manikuntala Kundu

Delivered a lecture in the National Symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules" organised by the Department of Chemistry, Bose Institute in February, 1999 ; Served as Joint Convenor of the National Symposium on "Recent Advances in Structure, Synthesis and Functions of Biomolecules" organised by the Department of Chemistry, Bose Institute in February, 1999 ; Presented a paper in the 6th Western Pacific Congress of Chemotherapy and Infectious Diseases, Kualalumpur, Malayasia in November, 1998.

Dr. Joyoti Basu

Presented a paper in the FEBS Meeting in Copenhagen, Denmark in July, 1998 ; Delivered a lecture in the 15th International Leprosy Congress, Beijing, China in September, 1998 ; Delivered a lecture in the National Symposium on "Recent Advances in Structure, Synthesis and Functions of Biomolecules" in February, 1999.

Dr. Anupa Bhattacharyya, Ms Chaitali Bhattacharjee and Ms. Sandipta Bhattacharjee participated at the National Symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules" organised by the Department of Chemistry, Bose Institute, Calcutta during February 4-6, 1999.

Dr. Jaya Bhattacharyya presented a paper at the National Symposium on "Recent Advances in Structure, Synthesis and Function of Biomolecules".

GRANTS-IN-AID SCHEMES

Investigator/ Co-investigator	Title	Financed by
Prof. Parul Chakrabarti Dr. Manikuntala Kundu	Molecular Basis of Non-specific Resistance in Mycobacteria : Role of Efflux Pumps	DBT
Prof. Parul Chakrabarti Dr. Joyoti Basu	Drug Permeation through Mycobacterial Cell Wall	DST
Dr. Joyoti Basu Dr. Manikuntala Kundu	Protein palmitoyl transferase : a potential regulator of dynamic protein palmitoylation	DBT
Dr. Manikuntala Kundu	Role of outer membrane proteins in drug resistance in *Shigella dysenteriae*	CSIR
Dr. Joyoti Basu Dr. Manikuntala Kundu	Molecular analysis of the functional role of palmitoylation of human erythrocyte membrane protein 4.2	Indo-French
Dr. Joyoti Basu	Structural and funcfional analysis of a high molecular mass class A penicillin-binding protein in Mycobacterium leprae	CSIR
Dr. Manikuntala Kundu	Mechanisms of beta-lactam resistance in *Shigella dysenteriae*	DST
Prof. Prantosh Bhattacharyya	Antihepatotoxic agents from some Indian medicinal plants	CSIR
Prof. Parimal C. Sen	Amphiphilic drug induced alteration of ion transporting ATPase activities modulated through protein kinase and endogenous regulators	CSIR
Dr. Manas Chakrabarty	Synthesis of pyrrolocarbazoles, an emerging class of bioactive heterocycles	CSIR
Dr. Pradip Das	Quantitation of Orientational Ordering in the F-actin Smectic B phase	CSIR
Dr. K. P. Das	Conformational specificity of α-crystallin for its function as molecular chaperone	CSIR

Ph. D. Awarded

Name	University	Year	Title	Supervisor
Anindyasundar Ghosh	Calcutta	1998	Studies on beta-lactam antibiotic resistance in *Shigella*	Dr. M. Kundu
Subhansu Roy	Calcutta	1998	Studies on human uterine tissues	Prof. P. Chakrabarti

RESEARCH SCIENTISTS/RESEARCH ASSOCIATES/POST-DOCTORAL FELLOWS/ SENIOR RESEARCH FELLOWS/JUNIOR RESEARCH FELLOWS

Shampa Khasnobis, Sarmistha Dutta, Atin K. Mandal, Jasimuddin Ahamed, Soumita Das, Sandipta Bhattacharya, Nilanjana Chakrabarti, Sudipan Karmakar, Biswajit Pal, Koushik Roy, Alok Kanti Kar, Jayanti Gangopadhyay, Sandip Pal, Sanjib Bhakta, Jitamanyu Chakrabarty, Pinakiranjan Jana, Susmita Sen, Baisakhee Sahachowdhury, Nandita Ghosh, Somesh Baranwal, Debasish Maiti, Dr. Debasish Ghosal, Dr. Swati Sinha, Dr. Anupa Bhattacharyya, Dr. Jaya Bhattacharyya, Dr. Nita Mitra, Dr. Subho Ghosh, Dr. Prasun Kanta Moitra Dr. Anuradha Bhaduri.

SUPPORTING STAFF MEMBERS

Nirmal Kumar Das, Prasanta Pal, Aniruddha Aich, Debaprasad Roy (till 31.01.99), Sukumar Maitra, Gopinath Mukherjee, Mrityunjoy Gayen (till 30.11.98), Paresh Bhattacharya, Alok K. Kundu (till 26.06.98), Pranab K. Dey, Bhabani Prasad Banerjee, Chittaranjan Ghosh, Debdas Nandy, Rama Mukherjee, Gopal K. Biswas, Tapas Ghosh, Arindam Basu, Dipak Chandra Konar, Subhas Chandra Pal, Amarnath Bhatta, Kodan Das, Asoke Kumar Maity, Sachchidanand Ram, Kalyan Das.

Botany

Faculty : Prof. S. Gupta (Chairman), Prof. S. N. Ganguly, Prof. B. Ghosh, Prof. K. K. Mukherjee, Prof. S. Sen Mandi, Dr. B. B. Mukherjee, Dr. P. K. Saha, Dr. D. N. Sengupta, Dr. S. Bhattacharya, Dr. S. Gupta Bhattacharya, Dr. T. K. Ghose

HIGHLIGHTS

Molecular analysis of plant response to salinity stress specially on polyamine biosynthesis and ABRE. Analysis of the Rice genome in relation to submergence tolerance and aroma. Study on genetic variabilities of *Beta palonga* and *Brassica* by RAPD. Clonal propagation of some medicinal and aromatic plants. Airborne spores and pollen and their allergenecity. DNA finger printing and developing molecular markers in elite tea clones

INSTITUTIONAL PROJECT I

A.1. *Studies on DNA Polymerase from Rice:* **Dibyendu N. Sengupta**, Sankar Bakshi, Sailendranath Sarker

Purification of ddNTP sensitive, single polypeptide of 69kDa size, DNA polymerase beta type enzyme from shoot tips of rice was repeated several times, using column chromatographic procedure, as standardized before. The purified enzyme will be pooled and production of antibody will be attempted. The YDP IV gene from yeast for similar enzyme has been amplified, subcloned in pBSKS and verified by DNA sequencing. The cDNA will be used as probe to select homologous transcript from rice. Oligonucleotides have been synthesized based on amino acid sequence of a ddNTP sensitive DNA polymerase from wheat germ (Luque et al. 1998) which will also be used as probel to detect homologous transcript and DNA fragment in rice. PolyA+ RNA has been prepared from wheat and rice seedlings, attempts will be taken to amplify cDNA for Pol beta by RT-PCR.

A.2. *Salinity stress and gene expression in rice:* **Dibyendu N. Sengupta**, Kakali Banerjee, Abdur Rouf

As reported earlier that by RT-PCR we have amplified the entire coding region of the cDNA for a factor called OSBZ8 (bZIP class). The cDNA was then subcloned in bluescript and nucleotide sequence was determined from the 5' end and homology with the published sequence (Hattori's group from Japan) was detected. The 3' end contains GC rich region as a result effort to sequence with T7sequanase has failed. To produce antibody against the N-terminal domain of OSBZ8, the 5' end 550bp area was amplified using new primers. The new partial OSBZ8N will be subcloned in pGEX3X to produce GST:0SBZ8n fusion protein in E.coli, which will be used for development of antibody, a reagent essential to detect the interaction of OSBZ8 with ABRE promoter. Synthetic ABRE based promoter has been synthesized using oligonucleotide containing 4 tandem repeats of ABRE (Emla sequence from wheat Em gene) and 35S basal promoter, which has been verified by subcloning in TA vector followed by sequencing. The Rab 16A gene has been amplified from the genomic DNA of Pokkali, salt tolerant rice by PCR using sequence specific oligonucleotide primers.Both synthetic ABRE-based and natural Rab 16A promoter will be used to overexpress stress inducible genes.

A.3. *Delay of Fruit Ripening by Antisense RNA approach :* **Bharati Ghosh, D.N. Sengupta**, Mausumi Sengupta, Pallab Kundu, Tapan Bera

To delay fruit ripening in tomato and banana our goal is to inhibit endogenous ethylene biosynthesis in fruit by antisense RNA approach. For that, a partial antisense construct and a full length cDNA construct of ACS2 (ACC Synthase2), the key enzyme of ethylene biosynthesis pathway have been generated. The partial antisense construct has been cloned in the plant transformation binary vector pBI121 and mobilized into *Agrobacterium tumefaciens* for transformation of tomato. Regeneration

method of tomato from different tissues has been standardised and the transformation experiments with the construct are going on. The tomato plants will also be transformed with the antisense full length construct of the ACS2 gene. Along with this we have developed the protocols for multiple propagation from shoot tips and generation of profused calluses from the leaf bases of tissue culture grown young banana plants and suspension cell culture of banana to produce somatic embryos.

A.4. *Mechanism of Polyamine Action and Molecular cloning of cDNAs for the Enzymes of PA Biosynthesis :* **Bharati Ghosh, Dibyendu N. Sengupta**, Papia Roy, Saswati Laha, Kamala Niyogi

In DST, Govt. of India, Project our goal is to find out the mechanism of polyamines (PAs) under abiotic stress. As polycationic PAs stabilises membrane, we are trying to find out the role of PAs on H^+ ATPase, H^+ATPase was inhibited by NaCl and reversed by Spd 3^+ and Smp 4^+. Phosphorylation of a 55kD polypeptide in S80 fraction of rice root extract was detected and found to be enhanced by $Ca2^+$ or Spd 3^+ or Spm 4^+, but inhibited by Na^+ or Li^+ suggesting the involvement of $Ca2^+$ dependent protein Kinase.

In DBT funded CPMB programme our goal is to overexpress SAM decarboxylase, and Spd synthase in salt sensitive rice cultivars to make it tolerant. The cDNA for SAMDC has been amplified from Poly A^+ RNA of salt treated rice roots by RT-PCR. The cDNA was cloned in pBSKS and verified by sequencing from two end. For making antibody, the cDNA has been subcloned in pEZZ (Pharmacia).

B.1. *Cytogenetical investigations in Beta spp. and studies on wild gene pool of Chenopodium and Ipomoea spp :* **K. K. Mukherjee,** S. K. Mitra and S. Das

Direct organogenesis through bud formation was recorded from petiole and leaf explants of *Beta vulgaris* (both sugar beet and table beet). The buds were induced from blade petiole transition (BPT) zone. The frequency of organogenesis was high in both the varieties of *Beta vulgaris*.

Studies on the isozymes (Esterase, Peroxidase, Acid phosphatase and GOT) of leaf tissue of two diploids (2n = 18), one hexaploid (2n = 54) of *Chenopodium album* and *C. murale* (2n = 18) manifested both commonarities and diversities between the species studied. The isozyme profile of this species are suggestive of their close relationship and also indicate contribution of the three diploids in the evolution of the hexaploid *C.* album.

Twelve species of *Ipomoea* comprising nine wild, two horticultural and one semicultivated were subjected to RAPD analysis to estimate genetic distance between these species. The level of polymorphism between the species was very high. From twelve decamer random primers used, total 458 fragments were amplified (comprising the monomorphic bands in different species). All the primers revealed appreciable monomorphism in banding spectra in different groups of species. Percentage based monomorphism between different species pair was determined followed by cluster analysis and principal component analysis (PCO) which suggest a common genetic base for each group of species.

B.2. *Phenotypic and genotypic studies with some species of the family Fabaceae and Brassica juncea :* **P.K. Saha**, B. Das and A. Roy

Pure line populations of *Tephrosia purpurea* were raised at Madhyamgram Experimental Farm. In the seeds of F3 population, about 90% homozygous seed population (based on testa colour) was obtained. Changes in the seed status with respect to their storage period was also investigated. In case of *Albiziza procera* seed, effect of dormancy breaking treatments were studied and it was found that changes in ultrastructures occur during the physical as well as chemical treatments which ultimately shorten the water uptake phases. This shortening of phases leads to successful germination of *A. procera* dormant seeds. Another leguminous seed, *Sesbania cannabina* was also investigated and ultrastcuctural differences among a single seed lot was observed. In all the three species, it may be possible to recommend few presowing measures to have maximum utilisation of a given seed lot.

The hybrid (recombinant) seeds from two cultivars of *Brassica juncea* were sown in Madhyamgram Experimental field for isolation of lines having higher yield potential. Plants could

not grow well due to rainfall and fluctuation of temperature.

B.3. *Germination and Demonstration of tissue culture raised planting materials from Andrographis paniculata :* **S. Gupta**, D. Sinha Roy and R. Poddar

Aseptic culture of *A. paniculata* (Kalmegh) was established throughout the season taking explants from different populations collected from different parts of 24-Parganas. Protocol for establishment, multiplication and rooting as described in the report of previous year was reproducible and was found to be efficient enough to develop large number of micropropagated plants from different populations. A transplantation protocol of high survival rate (90%) was also established in a pilot scale. Micropropagated clones of different populations of *Andrographis* are now ready for hardening in a large scale to the green house of the funding agency within a short duration.

B.4. *Genetic approach for the improvement of submergence tolerance in Rice :* **S. Gupta** and Sarat Kumar Raj

The androgenic double-haploid (DH) submergence tolerant lines developed from our laboratory were maintained at the Madhyamgram Farm. Replicated yield trial was conducted in the field. Straw yield and Harvest Index (HI) was determined. Significant variation among the lines have been observed in relation to grain yield. Recovery percentage after 10 days complete submergence under 50 cm water depth of each line had been scored during rainy season in Green house tank condition. All the lines show high recovery percentage (more than 80%). Physico-chemical characterization of the grains of each line are in progress.

To characterize all the lines at the DNA level, experiments have been conducted using polymerase chain Reaction (PCR). Lines possessing two disease resistant genes viz. Bacterial Blight of Rice (Xa-21) and Blast of Rice (Pi-25) have been screened using STS primers linked to the particular diseases. To analyse the transfer of major locus for submergence tolerance (QTL) mapped on rice chromosome 9 from the source genotype (FR 13A & *O. rufipogon*) into the androgenic lines, RAPD markers linked to the QTL have been successfully amplified in the respective tolerant & susceptible parents. Selection of lines using these molecular markers are in progress.

B.5. *Salt stress in rice cultivars :* **B. B. Mukherjee** and Sangita Basu

Mechanism of salinity tolerance by tolerant a rice variety was investigated in comparison to sensitive variety. Exchange pattern of Sodium and Potassium and its relationship with the accumulation and leaching of proline were studied and statistically evaluated and a distinct relationship was established indicating some clue of the mechanism of salinity tolerance.

B.6. *Studies on the Bengal Basin :* **B.B. Mukherjee**

Some geomorphological and biostratigraphical data from the southern part of the Bengal Basin were considered in relation to some temporal photograph (remotely sensed) to evaluate the Holocene history of sea level change in the Bengal Coast.

A training workshop on geomorphology and biostratigraphy & Geographical Information system of the Bengal basin was organised in botany Department to train man power capable to collect data from the Bengal basin and develop GIS for the region.

B.7. *Clonal propagation of medicinal plants :* Rajasri Bhattacharya and **Sabita Bhattacharya**

In view of cost effectiveness of protocols of micropropagation, low salt media were used thoroughly for growth and rooting of individual shoots obtained from multishoot bunch for both the species. 80% of such shoots of *W. somnifera* survived in small plastic pots containing soilrite and soil mixture at 1:1 ratio under room temperature.

In a detailed study on hormonal effect on rooting of *W. somnifera* shoots, IAA was proved best. Root initiation can be influenced even from the cut petiolar end of the entire leaf and provided a second source of mass production of roots, the source of withanolide, the important secondary metabolite.

B.8. *Identification & study of Biochemical components of seed vigour for direct seeding cultivation of rice :* **Swati Sen-Mandi**

Studies were made on large number of rice varieties obtained from Chinsurah Rice Research

Station & Directorate Rice Research, Hyderabad, ICAR, for variation in isozyme pattern of Esterase (respiration associated enzyme), & Super Oxide Dismutase (SOD), peroxidase(free radical scavenging enzymes) at different developmental stages of germination.These have shown a correlation with germination performance viz. seed vigour. Attempts are now being made to develop precise molecular marker for vigour trait in rice seed.

B.9. *Germplasm Screening and Development of Molecular Markers for seed vigour in rice using RAPD/AFLP Techniques :* **Swati Sen-Mandi &** Subhra Talai

From RAPD analysis of genome in rice varieties recognised as high and low vigour through use of physiological and biomolecular parameters , some PCR products could be identified that correlate with vigour quality. One band representing the product amplified by primer AB-17 was found to be absent in low vigour seed. Sequencing of this DNA fragment for designing primers for screening of germplasm is now underway in our laboratory in order to design specific primers that. would help in screening rice germplasm for seed vigour quality.

B 10. *DNA Finger Printing and Cattaloguing of Tea Germplasm Developing Molecular Markers in Elite Tea Clones :* **Swati Sen-Mandi** & Rajan Kumar Mishra and Partha Talukdar

A component of the cellulase enzyme system has been located to be useful as a molecular marker for screening tea germplasm. Purification and characterization of this molecules is now under progress.

DNA fingerprinting have been documented in several tea clones using random primers. Studies on correlation of DNA polymorphism with desirable characters in tea is now underway.

INSTITUTIONAL PROJECT II

A. *Chemistry of Natural Products :* **S.N. Ganguly**

A.1. From the methanolic extract of the defatted roots of *Camellia sinensis*, a white solid crystallised from chloroform-methanol, m.p. 239-40o, $C_{29}H_{44}O_2$ (M^+424) was isolated. On the basis of physical evidences coupled with chemical evidences, the compound was identified as a new pentacyclic triterpene and its structure was given.

A new diterpene, $C_{21}H_{32}O_3$, m.p. 89-90° was isolated from the acetone extract of the pods of *Cassia fistula.* On the basis of physical and chemical evidences its structure was established.

From the Pet. Ether extract of the air dried leaves of *Sonneratia apetala,* a new triterpene, $C_{32}H_{50}O_3$ (M^+428), m.p. 268^0 was isolated as a crystalline solid. On the basis of physico-chemical evidences, its structure was determined.

INSTITUTIONAL PROJECT IV

A.1. *Environmental biopollution in relation to human health :* **Swati Gupta Bhattacharya**, Pampa Chakraborty, Indrani Chowdhury, Atin Adhikari, Moonmoon Sen and Debajyoti Ghosh (In collaboration with Prof. Sunirmal Chanda).

In addition to the continuous airborne pollen and spore survey at Madhyamgram and Domjur, another one had started in the Bose Institute from January 1999 to make a comparative analyses. Besides direct aeroallergen recording is being continued to form an aeroallergen calendar for the help of asthmatics.

A survey on respiratory allergic patients is being carried out with allergen extracts of different airborne fungal spores by skin test, lung function test and IgE-ELISA. The major allergenic spores recorded were *Aspergillus flavus, A. niger, Alternaria*, etc., which are prevalent in the air of different indoor environments.

Protein extract of *Catharanthus roseus* pollen had found to be sufficiently allergenic by skin tests and ELISA. Characterisation programme of its allergen by ammonium sulphate fractionation, gel filtration, PAGE and IgE-immunoblotting is in progress.

PUBLICATIONS

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2. Mondal N and Gupta S 1997 Anther culture of an interspecific rice hybrid and selection of fine grain type with submergence tolerance *Plt. Cell Tissue and Organ Culture* **51** 79-82.

3. Basu Sangita, Gangopadhyay G, Mukherjee BB and Gupta S 1997 Plant regeneration of salt adapted callus of indica rice (var. Basmati 370) in saline conditions. *Plant Cell Tissue & Organ Culture* **50** 153-159.

4. Gangopadhyay G. Basu S and Gupta S 1997 In vitro selection and physiological characterization of NaCl- and mannitol- adapted callus lines in *Brassica juncea*. *Plant Cell Tissue and Organ Culture* **50** 161-160.

5. Basu Sangita, Gangopadhyay G, Gupta S, and Mukherjee BB 1997 Physiological changes of *Oryza sativa* L. callus due to salt stress. *Plant Tissue & Cell Culture* 381-386.

6. Mandal N, Roy K and Gupta S 1998 Nature of Genetic Control of Submergence Tolerance in Rice. *Indian J. Genet.*, **58**(3) 285-290.

7. Gangopadhyay G, Poddar S and Gupta S 1998 Micropropagation of Sesame (*Sesamum indicum* L.) By in vitro Multiple Shoot Production from Nodal Explants. *Phytomorphology*. **48** 83-90.

8. Chakraborty P, Gupta-Bhattacharya S, Chakraborty C, Lacey J & Chanda S 1998. Aerobiology of allergenic pollen grains on a farm in West Bengal, India; *Grana* **37**(1) 53-57 (Scandinavian University Press, Sweden).

9. Chowdhury I, Chakraborty P, Gupta-Bhattacharya S & Chanda S 1998. Allergenic relationship among four common and dominant airborne palm pollen grains from Eastern India; *Clinical and Experimental Allergey* **28** 977-983 (Blackwell Science Ltd., UK).

10. Chakraborty P, Gupta S, Gupta Bhattacharya S & Chanda S 1999. Airborne pollen of *Borassus flabellifer:* Quantification of allergen and antigen; *Aerobiologia* **15** 49-55 (Kluwer Academic Publ., The Netherlands).

11. Chowdhury I, Chakraborty P, Gupta Bhattacharya S & Chanda S 1999. Antigenic relationship between four airborne palm pollen grains from Calcutta, India; *Annals of Agricultural and Environmental Medicine* **6** 53-56 (Institute of Agricultural Medicine, Poland).

12. Mitra, SK, Das S, Roy PK and Mukherjee KK 1998 Characterisation and inheritance studies on the seed coat colour mutants of two species of *Corchorus Phytomorphology* **98** 237-253.

13. Das S, Ganguly SN and Mukherjee KK 1999. Fatty acids and phytochemical components of *Ipomoea* spp. seeds *Natl. Product Sci.* **5** 1-3.

14. Roy A and Saha PK 1997 Factors involved during in vitro culture of *Calamus rotang*. *Jour. of Tropical Forest Science*; **10(2)** 225-232.

15. Saha, PK, Bhattacharya A, Chetti MB, Hiremath SM and Nawalagatti CM 1997 Effect of physical and chemical treatments on testa of *Cassia tora.* In First International Conference on *Parthenium* **(Vol.II)**; Ed. M Mahdevappa and VC Patil; Vishwakala Printers, Bangalore, pp. 65-69.

16. Das B, Bhattacharya A, Saha PK, Chetti MB and Hiremath SM 1997 Water uptake patterns in *Cassia tora* L. as influenced by different seed types. *Ibid.*, pp. 70-73.

17. Bose A, Tiwari BS, Chattopadhyay MK, Gupta SR and Ghosh B 1999 Thermal stress induces differential degradation of Rubisco in heat sensitive and heat tolerant rice. *Physiol Plant* **105** 89-94.

18. Ghosh B 1998. Prospects of salinity mrkers from mangroves and there biotechnological application. *Sci. and Cult.* **64** 165-166.

Participation in Conferences / Symposia / Training Programme / Awards and Honours received.

Dr. Swati Gupta Bhattacharya : Attended workshop on biosafety issues emanating from use of genetically modified organisms organised by Biotechnology Consortium India Ltd., New Delhi and Department of Biotechnology, Ministry of Science & Technology held on October 23, 1998 at Bose Institute, Calcutta; attended the "Symposium on Biotechnology for Health, Agriculture & Environment" organised by Center for Applied Sciences and Technology in collaboration with Bose Institute and Indian Science Congress Association held on Nov. 10-12, 1998, Calcutta.

Ms. Pampa Chakraborty participated in the 5th International Congress on Aerobiology, 1998, held in Perugia, Italy. She obtained 1000 USD Travel Grant Abstract Award for young scientists to attend the Congress by International Association of Aerobiologists.

Dr. P.K. Saha attended the 3rd International Plant Tissue Culture Conference during March 8-10. 1999 in Dhaka, Bangladesh and presented a paper. He also became a member of the New York Academy of Sciences in 1998.

Dr. D.N. Sengupta has presented an invited talk in National Symposium on "Cellular Response to Stresses and Defence System" organised by the Institute of Life Sciences, Bhubaneswar, Orissa on Nov. 11-13, 1998. Dr. Sengupta has presented an invited talk in University & College Teachers Refresher Course on Molecular Biology & Biotechnology, organised by School of Life Sciences, Sambalpur University at Sambalpur on 16th Nov., 1998. Dr. Sengupta was selected by DST, Govt. of India to visit Cairo, Egypt, as a member of Indian Delegates in Indo-Egypt Inter-Governmental Exchange programme. Dr. Sengupta has visited different research Institute in Cairo and participated in the group discussion between the scientists of Egypt and Indian Delegates.

Prof. Bharati Ghosh presented invited talks in the following Seminars/symposia : On Biotechnology for Health, agriculture and environment and chaired a session on Nov 10-12 organised by CAST, BI and ISCA. On recent trends in application of biotechnology in food quality, safety, preservaion at 86th Indian Science Congress organised by Biochemistry, Biophysics and Molecular Biology session during 3 to 7 January, 1999 at Chennai. In 10th Reunion of Botany Department, Visva Bharati University, Santiniketan on 17th January 1999. On Recent advances in structure and function of Biomolecules organised by Dept. of Chemistry, Bose Institute during 4 to 6 February, 1999. On Plant Physiology and Biochemistry in relation to Agriculture and Environment and chaired a scientific session in the symposia organised by Life Science, Devi Ahilya Viswavidhyalaya, Indore, 13 to 18 February 1999. In S.M. Sircar memorial conference by Plant Physiology forum, Calcutta. Prof. Bharati Ghosh visited the USDA, Beltsville, Maryland, USA, in 1998 (Aug-Sept) as part of the programme of Indo US collaborative project. Prof. Bharati Ghosh is honoured for her significant contributions by Society for Plant Physiology and Biochemistry at Indore.

Ms. Mausumi Sengupta and Saswati Laha attended symposia at Indore and presented a poster in the poster session.

Prof. Swati Sen Mandi : (i) Participated as Chairperson in a session of invited papers and acted as Judge for young Scientists' best paper presentation at the National Symposium of Cellular Response to Stresses And Defence Mechanism (CRSDM) held during 11-13 Nov. 1998 at Bhubaneswar ; **(ii)** Participated in discussion at the National Dialogue : Issues In Management Of Plant Genetic Resources organised by the Indian Society of Plant Genetic Resources and National Bureau of Plant Genetic Resources, New Delhi 110 012, India : Dec. 1-2, 1998. ; **(iii)** Participated as a M. Sc. Course, Guest Lecturer in Agricultural Department ,Calcutta University. : **(iv)** Nominated Indian representative in the Steering Committee of the International Society for Seed Science (ISSS).

Dr. B. B. Mukherjee : Organised a Training Workshop on Bengal Basin in January 1999 in Bose Institute during 4-9 January 1999.

Prof. S. Gupta : Convened National Workshop on Enterpriunorship Development based on Chemical and Agro Technologies at Vivekananda Institute of Biotechnology, Nimpith 24 Pgs(N), W. B.

Sabita Bhattachaya : Attended workshop on biosafety issues emanating from use of genetically modified organisms organised by Biotechnology, Ministry of Science and Technolgy held on October 23 1998 at Bose Institute, Calcutta ; Symposium on Biotechnology for Health, Agriculture and Environment organised by Centre for Applied Sciences and Technology in collaboration with Bose Institute and Indian Science Congress held on 10-12, 1998 in Calcutta ; attended 2nd All India People's Technology Congress organised by FOSET and Department of Science and Technology, Govt. of W. Bengal, held on 12-14 Feb. 1999 at Science City, Calcutta.

GRANTS-IN-AID SCHEMES

Investigator/ Co-investigator	Title	Financed by
Prof. B. Ghosh Dr. D.N. Sengupta	Use of Antisense RNA to delay fruit ripening	DBT
Prof. B. Ghosh Dr. D.N. Sengupta	Molecular mechanism of polyamine action in protection against abiotic stress in Indica rice	DBT
Prof. B. Ghosh Dr. D.N. Sengupta	Cloning of Molecular Markers involved in salt tolerance and their overexpression to enhance salt tolerance in rice plants	DBT (CPMB) Phase II
Prof. B. Ghosh	Metabolisn in relation to environmental stress in rice	Indo-US
Prof. S. Gupta	Generation and demonstration of tissue culture raised planting materials of *Andrographis paniculata* and *Tectona grandis*	W.B. SC & ST
Dr. D.N. Sengupta	Molecular analysis of regulation of salinity stress and abscisic acid inducible gene in rice. (Upto Sept. 1998)	DST
Dr. D.N. Sengupta	Characterisation and cloning of plant DNA polymerase	CSIR
Prof. Swati Sen-Mandi	DNA Finger Printing And Cataloguing Of Tea Germplasm Developing Molecular Markers in Elite Tea Clones	DBT TEA BOARD
Dr. Sabita Bhattacharya	Generation and demonstration of tissue culture raised planting materials of *Abroma augusta*	W.B. SC & ST
Dr. Swati Gupta Bhattacharya	Environmental biopollution and its impact on human health	DST, W.B.
Dr. B.B. Mukherjee	Holocene and Biostratigraphy of South Bengal with reference to Geoidal changes	D.O.D.

PH.D. AWARDED

Name	University	Year	Title	Supervisor
Soubhik Das	Calcutta	1998	Cytomorphological, Chemical and Biochemical investigations on some *Ipomoea* species	Prof. K.K. Mukherjee
Pampa Chakraborty	-do-	1999	Aerobiology of suburban fringe of Calcutta with reference to chemical characterisation of some common allergenic pollen	Dr. Swati Gupta Bhattacharya
Amitabha Roy	-do-	1998	Studies on genetic variabilities of the recombinants of mustard *(Brassica juncea* Czern and Coss.) raised through *in vivo* and *in vitro* techniques	Dr. P.K. Saha

RESEARCH SCIENTISTS/RESEARCH ASSOCIATES/POST-DOCTORAL FELLOWS / SENIOR RESEARCH FELLOWS / JUNIOR RESEARCH FELLOWS

Sreejit Kr. Mitra, Sarat Kr. Raj, Debjani Sinha Roy, Sangita Basu, Pampa Chakraborty, Atin Adhikari, Moon Moon Sen, Debajyoti Ghosh, Sanghamitra Bhattacharya, Biswajit Das, Tapan Bera, Sankar Bakshi, Rajasri Bhattacharya, Subhra Talai, Abdur Rouf, Saswati Laha, Papiya Roy, Mousumi Sengupta, Pallab Kundu, Kakali Banerjee, Kamala Niyogi, Rajan Kr. Mishra, Partha Talukdar, Indrani Bhattacharya.

SUPPORTING STAFF MEMBERS

N. N. Chowdhury, Sankar Basu, Sukumar Majumdar, Dr. Ramit Poddar, Chanchal Chakraborty, Jadab Kr. Ghosh, Binoy Krishna Modak, Chaitali Roy, Kaberi Ghosh, Shyamal Kr. Ghosh, Dilip Kr. Chakraborty, Shyamapada Halder, Kanailal Chatterjee, Sukadeb Das, Sudhir Jana, Sasanka Sekhar Bera, Sambhunath Debnath, Dhiren Kumar Das, Rabin N. Roychoudhuri, Bipul Kr. Nag, Kanan Singh Jagannath Das, Subhas Mandal, Munna Turi.

Microbiology

Faculty: Prof. Timir B. Samanta (Chairman from January 1, 1999), Prof. P.K. Chakrabartty (Chairman upto December 31, 1998), Prof. A.C. Ghose, Dr. Roma Chakraverty (upto July 31, 1998), Dr. Pradosh Roy, Dr. S. Majumdar, Dr. S. K. Dasgupta

HIGHLIGHTS

A β-lactamase free penicillin amidase produced by *Alcaligenes* sp., which can act as both deacylating and acylating agent, has been characterised.

INSTITUTIONAL PROJECT I

A. *Interaction of aluminium with iron transport system in rhizobia* : **P. K. Chakrabartty**, Anup Halder (in collaboration with Dr. Nupur Roy)

Under iron depleted condition iron transport system of *Rhizobium* sp. (*Cicer arietinum*) BICC 651 is induced. Under this condition aluminium at 100μM causes more than two-fold increased siderophore production but no change in outer membrane protein profile. It is hypothesised that aluminium also binds to siderophore and this causes reduced binding of iron to siderophore in competition. The binding of aluminium to siderophore was confirmed in modified CAS assay where aluminium is used in place of iron. Addition of siderophore to modified CAS changes the spectrum indicating binding of aluminium to siderophore. The result was further confirmed by binding of aluminium to siderophore and recording the change in the spectrum of siderophore.

INSTITUTIONAL PROJECT V

Molecular and cellular basis of bacterial and parasitic diseases

A.1. *Cholera and Vibrio cholerae* : **A. C. Ghose**, R.K.Nandy, S.Mukhopadhyay and B. Nandi

The distribution and function of a 21-kD outer membrane protein (OMPw) of *Vibrio cholerae* was studied. PCR analysis using different sets of primers showed that the *ompW* gene was present in *V. cholerae* strains irrespective of their biotypes/serogroups but not in other vibrios. The *ompW* gene sequence of the non-01/non-0139 *V. cholerae* strain 10325 was determined and compared with those of an 01 classical strain 569B. The two sequences differed in only 5 nucleotides and 3 amino acid residues in their predicted protein sequences. A recombinant suicide vector pBN490 was constructed which contained a 220-bp fragment of the *ompW* gene. This vector was used to generate insertion mutants of *V. cholerae* 01 strains lacking functional *ompW* gene. Competitive colonization experiments carried out with the wild type strains and their OMPw-negative mutants did not show any significant difference in their intestinal colonization potential.

A.2.1 *Leishmaniasis* : **A. C. Ghose** and A. Mookerjee (in collaboration with Prof. F. Hudecz of E. L. University, Budapest, Hungary)

The antileishmanial effect of the drug methotrexate (MTX) coupled to branched chain polypeptide carriers was evaluated *in vivo* and *in vitro*. The drug carrier conjugates used were poly{Lys-(MTX-DL-Ala)} (MTX-AK), poly{ Lys-(MTX-Ser -DL-Ala)} (MTX-LAK) and poly{ Lys-(DL-Ala -Leu)} (MTX-ALK). Preliminary experiments showed that the antileishmanial properties of MTX-ALK conjugate was superior to the other two conjugates. Therefore detailed studies were carried out with the MTX-ALK conjugate. Treatment of *L. donovani* infected Balb/c mice with this conjugate showed significant reduction of parasite load in the liver of infected animals as compared to those observed with MTX (alone), ALK (alone) or even MTX+ALK (mixture) treated animals. Comparable results were also obtained when leishmania infected mouse macrophages were treated with MTX-ALK conjugates *in vitro*.

A.2.2. *Leishmaniasis* : **Timir B. Samanta**, Nilanjana Das, Himan Mukherjee and Manika Das

The therapeutic effect of the traditional drug for leishmaniasis, stibanate alongwith the immunomodulator protein A (PA) was investigated together with their role in control of impairment of Cyt P450. Stibanate treatment (10 mg/kg body wt x 7 days) alone decreased parasitemia and protected impairment of P450 quite considerably. The liver marker enzymes except ACP i.e., ALP, GOT and GPT remained unaffected. Treatment with PA alone (50 mg/kg body wt x 5 days) was better in terms of protection of Cyt P450 impairment and ACP/ALP normalisation. But the parasitemia did not decrease appreciably in comparison to stibanate treatment. Combination of stibanate and PA treatment produced best results in terms of all these parameters. ACP/ALP/GOT/GPT returned to normal, Cyt P450 isozymes (aniline hydroxylase and aminopyrine demethylase) were 70-80% of normal while parasitemia decreased to 25% of infected control. There were similarities between hepatic histologies with the biochemical data in all these cases.

A.3. *Down stream effector molecules of TNF-α in human neutrophil* : **Subrata Majumdar** and Sonali Das

Recent evidence suggests that branching pathways of sphingolipid metabolism may mediate either apoptotic or mitogenic responses depending on the cell types and the nature of the stimulus. Mammalian systems respond to environmental stress by either adapting or undergoing programmed cells death.

Ceramide, a novel intracellular lipid second messenger produced upon TNF-α induction plays a major role in its signalling cascade and is reported to be a mediator of TNF-α induced apoptotic death signal. When human neutrophil was treated with 500 and 1000 U/ml of TNF-α for different time periods, enhanced intracellular ceramide concentration was observed. In some cellular system, ceramide initiates differentiation or cell proliferation, while in other systems ceramide signals apoptosis. The ceramide signalling pathway is activated by the sphingomyelinase (SMase) mediated hydrolysis of cell membrane sphingomyelin to ceramide. Recent investigations link the activation of neutral SMase to the extracellular signal regulated kinase (ERK) cascade and pro-inflammatory responses and acidic SMase to the stress activated protein kinase cascade and apoptotic response. When we assayed the SMase activity in TNF-α treated neutrophil at 1000 U/ml concentration we observed that ASMase activity increased at 4 hrs of incubation whereas at 16 hrs of incubation the NSMase activity increased to some extent, but ASMase activity decreased.

A.4. *Molecular genetics of Mycobacteria* : **Sujoy K. Das Gupta,** Shreyasi Chatterjee, Abhijit basu and Mahasweta Mitra

In order to understand the molecular genetics of mycobacteria in more details and to develop efficient tools for genetic engineering of mycobacteria we have been investigating the replication of *E. coli*- mycobacteria shuttle vectors derived from pAL5000, a mycobacterial plasmid. Primer extension analyses have been performed to map the promoter region of ORF1 which encodes the replication protein RepA. Several transcription initiation sites were found to be present in this region indicating that there could be multiple promoters involved in the expression of ORF1. Further, to identify the DNA protein interactions that take place in this region, a gel retardation assay was performed. The specific binding of a factor to a 200bp fragment spanning the promoter region has been demonstrated and is being characterized further.

In another area we have been continuing our attempt to construct a convenient phage based transformation system for mycobacteria. Accordingly earlier we constructed a cosmid shuttle vector for mycobacteria utilizing the cohesive end (cos) site of a mycobacteriophage L1. The initial cosmid vector which featured only the L1 cos site has been further modified to include the λ cos site. Using this modified vector the relative efficiency of packaging of the vector using λ and L1 lysogens are presently being assessed. We have also characterized the L1 cos site further. Using primer extension analysis it was possible to demonstrate that the cos site cleavage pattern of L1 was identical to that of the related mycobacteriophage L5. A deletion mutant of L1 has been obtained in which a large region (about 8kb) spanning the early regulatory region was found to be deleted. The properties of this phage are being investigated

to obtain deeper insight into the molecular biology of mycobacteriophages.

B. Industrially important microbial enzymes

B.1. *Biotransformation of organic compounds by microbes : Studies on penicillin amidase* : **Timir B. Samanta** and Anjan Pal

Penicillin G amidase produced by a strain of *Alcaligenes*.sp (A-13) was purified by salt precipitation, ion exchange and gel filtration chromatography. Sodium dodecyl sulphate (SDS) gel electrophoresis of the purified enzyme showed two major bands of 66 and 36 kDa respectively.

In another experiment cells of A-13 entrapped in calcium alginate and agar-agar separately were tested for penicillin G amidase activity. The calcium alginate was found to be the better matrix. Calcium alginate immobilized cells of A-13 were also tried for acylation of 6-APA with phenylacetic acid and it furnished penicillin G.

B.2. *Molecular Recognition of chiral compounds by microbes* : **Timir B. Samanta** and S. Das

During our work on molecular recognition of chiral molecules by microbes attempts were also made for transformation of prochiral molecules both cyclic and acyclic in nature by microbos. A bacterial strain F-4 was found to transform some specific prochirals to chirals. The chirals so produced were resolved on a C18-bondapack column (reverse phase) by high pressure liquid chromatography and subsequently partially characterised by chemical methods. Both the epimers were formed.

B.3. *Degradation of poly-hydroxyalkanoate by bacteria* : **P. K. Chakrabartty** and A. K. Halder

A large number of strains of rhizobia were tested for their ability to metabolize poly-hydroxyalkanoate (PHA) using the compound as the sole source of carbon or in combination with succinate. In succinate supplemented medium growth was significantly higher than in the presence of PHA alone. Poor PHA metabolizing capability of the strains resulted in low growth and on solid medium escaped visual detection of any PHA degradation.

For isolation of efficient PHA degrading bacteria enrichment cultures were established with soil inoculum. Three bacterial strains using PHA as the sole source of carbon were isolated. In presence or absence of succinate, all the three strains produced clear zones on solid agar plates containing PHA. Degradation of PHA by the strains seems to be constitutive and is caused by extracellularly produced enzymes. The bacteria are pale yellow to yellow-brownish in colour, leathery in texture, aerobic, gram -ve, rod shaped, sheathed, mostly single or in chains of two or more.

B.4. *Microbial starch degrading enzymes* : **P. K. Chakrabartty** and Mithu De

Starch enriched cultures of soil suspension afforded isolation of a large number of bacterial isolates producing amylase activity. Activity studies of enzymes of the isolates showed that in general growth in medium with 2% starch as carbon source yielded higher enzyme activity than with 0.15% starch. Several isolates produced two peaks of activity, one at 40°C and the other at 60 - 70°C. Product profile analyses after growth at 50°C showed that most of the isolates produced α-amylase.

B.5. *Microbial Proteases* : **Roma Chakraverty** and Papia Mitra

A *Streptomyces nogalater* strain (Ac 80) produced extracellular protease when grown on wheat bran substrate by the solid state fermentation method. The protease could dehair goatskin within 18 h of incubation. Two mutants, Mt C and Mt L were isolated by UV irradiation of Ac 80. The former (Mt C) had the highest proteolytic activity.

The enzyme from the mutant strain, Mt C was purified by ammonium sulphate precipitation, gel filtration and ion exchange chromatography. The purified enzyme showed a single band on SDS-PAGE. The molecular weight was estimated to be about 66,000 dalton. The purified protease could dehair goatskin after 14 h of incubation.

C.1. *Identification of lithotrophic sulfur oxidation gene cluster in a sulfur lithotrophic bacterium* : **Pradosh Roy** and Chandrajit Lahiri.

Seven transposon insertion sulfur-lithotrophic mutants were mapped and found to be dispersed

within a 6.3 kb *Eco*R1 fragment of the genome of a sulfur - lithotrophic new bacterium. The transposon flanked genome of one of these mutants was sequenced. The sequence (1125 bp) had a potential open reading frame of 265 amino acids and found to be homologous to a putative protein of *Aquifex aeolicus*. Sequence analysis of the putative protein showed two potential cytochrome c-heme binding sites which could be one of the 4 subunits of thiosulfate dehydrogenase identified from an another sulfur lithotrophic bacterium.

PUBLICATIONS

1. Datta J, Ghosh S, Maiti AK, Das AK, Modak DP, Bhattacharyya P, Ghosh A, Mandal NC, Chakrabartty PK and Ray PK 1998 Biodegradation of organochlorine pesticides by soil bacteria; J Sci Indus Res **57** 838-843.

2. Halder AK, Mohanty BK, Ghosh S and Mishra AK 1998 Biobeneficiation of silica containing ores. In Microbes : For Health, Wealth & Sustainable Environment. pp 719-724. Ed. Ajit Verma (New Delhi : Malhorta Publishing House).

3. Ghosh Sanjukta, Paul Soumitra, Das Sonali and Majumdar Subrata 1998 Lipoarabinomannan induced cytotoxic effects in human mononuclear cells; FEMS Med Microbiol and Immunol **21(3)** 181-187.

4. Ghose AC, Mookerjee A, Sengupta K, Ghosh AK, Dasgupta S and Ray PK 1999 Therapeutic and prophylactic uses of Protein A in the control of *Leishmania donovani* infection in experimental animals; Immunology Letters **65** 175-181.

5. Dasgupta S, Mookerjee A, Chowdhury SK and Ghose AC 1999 Immunosuppression in hamsters with progressive visceral leishmaniasis; an evaluation of the the role of nitric oxide toward impairment of the lymphoproliferative response: Parasitology Research **85** 594-596.

6. Mukhopadhyay Srirupa, Sen Pradip, Bhattacharya Sandip, Majumdar Subrata and Roy Syamal 1999 Immunoprophylaxis and Immunotherapy against experimental *Visceral leishmaniasis*; Vaccine **17** 291-300.

7. Chawla, M and Das Gupta, SK 1999 Transposition-induced structural instability of *Escherichia coli* - mycobacteria shuttle vectors; Plasmid **41** 135-140.

8. Pal Anjan and SamantaTimir B. 1999. B-lactamase free penicillin amidase from *Alcaligenes sp.* : Isolation strategy, strain characterization and enzyme immobilization ; Curr. Microbiol **29**, 244-248.

9. Dutta T K and Samanta T B, 1999 Bioconversion of progesterone by the activated immobilized conidia of A. Ochraceus TS Curr. Microbiol. **39** 309-312.

Participation in Conferences/Symposia/ Training programme/Awards and Honours received

Prof. A. C. Ghose visited Eotvos Lorand University (Research Group of Peptide Chemistry), Budapest, Hungary during Aug. 1-15, 1998, under the Indo-Hungarian Collaborative Research Programme. He also presented an invited lecture in the International Symposium on Biotechnological Approaches to Bacterial and Parasitic Diseases held during the IICB, Calcutta, on November 4-6,1998; delivered a talk and chaired a session in the Symposium on Biotechnology of Health, Agriculture and Environment held at Calcutta, during November 10-12, 1998; attended and delivered a lecture in the National Symposium on Recent Advances in Structure, Synthesis and Function of Biomolecules held at Bose Institute during February 4-6, 1999 ; delivered a series of lectures to the M.Sc.(Microbiology special) students of the Biochemistry department of Calcutta University.

Prof Timir B. Samanta acted as a Resource person in Academic Staff College and delivered a lecture in the department of Zoology, Calcutta University on December 15, 1998; acted as a General Secretary in the symposium on Biotechnology for Health, Agriculture & Environment organised by the Centre for Applied Science & Technology (CAST) in collaboration with Bose Institute and Indian Science Congress Association during Nov 10-12, 1998.

Prof. P.K. Chakrabartty acted as a Resource person in Refresher courses for College and University Teachers and delivered two invited lectures at Sambalpur University on 13th and 14th November, 1998, one invited lecture at Kalyani University on 21st January, 1999 and one invited lecture at Calcutta University on 12th March, 1999.

Shri Ananda Mookerjee presented a poster in the National Symposium on Recent Advances in Structure, Synthesis and Function of Biomolecules held at Bose Institute in February,1999.

Dr. Ranjan Nandy attended the International symposium on "Biotechnological Approaches to Bacterial and Parasitic Diseases" held at IICB, Calcutta during Nov 4-6, 1998; attended a symposium on "Recent Developments in Molecular and Cell Biology" held at the Biophysics Department, Calcutta University during Dec 15-16, 1998; participated in a Workshop on "Structure and Sequence Database Analysis at the Bose Institute, Calcutta during March 11-12, 1998.

GRANT-IN-AID-SCHEME

Investigator	Title	Financed by
Prof.A.C.Ghose	Characterization of a new type of pilus expressed by pathogenic strains of non-01 *Vibrio cholerae* and evaluation of its role in intestinal colonization as well as protection.	CSIR
Prof.A.C.Ghose	Use of an immunomodulator protein A in the treatment of visceral leishmaniasis.	ICMR
Prof.A.C.Ghose	Development of bioconjugates for diagnosis and treatment of microbial infections. (Indo-Hungarian Collaborative Project).	DST
Prof. Timir B. Samanta	Studies on protection of impairment of cytochrome P450 during experimental leishmaniasis in golden hamster : Role of nitric oxide synthase.	CSIR
Dr. P. Roy	Development and evaluation of a micorbial based arsenic assay kit.	DBT
Dr. S. Majumdar	Signal transduction mechanism in Visceral leishmaniasis in mice : Role of TNF in leishmaniasis .	DAE
Dr. S. Dasgupta	Understanding the mechanism of replication of the mycobacterial plasmid pAL5000 using an *in vitro* approach.	CSIR
Dr. S. Dasgupta	A cosmid vector for cloning genes linked to viability of virulent mycobacteria in macrophages.	DBT

Ph.D. AWARDED

Name	University	Year	Title	Supervisor
Ranjan K. Nandy	Calcutta University	1998	Immunobiological characterization of the epidemic isolates of *Vibrio cholerae* 0139 : A comparative study with *Vibrio cholerae* 01 and other non-01 organisms.	Prof.A.C.Ghose
Subhajit Dasgupta	Calcutta University	1998	Stuides on the immunological mechanisms in golden hamsters experimentally infected with *Leishmania donovani*	Prof. A.C. Ghose

RESEARCH SCIENTISTS/RESEARCH ASSOCIATES/POST DOCTORAL FELLOWS/ SENIOR RESEARCH FELLOWS/ JUNIOR RESEARCH FELLOWS

Dr. Anup K. Halder, Dr. Eliza Chakrabarty (upto 08.12.98), Dr. Indrani Chakrabarty (upto 31.12.98), Nilanjana Das, Papia Mitra, Sonali Das, Ananda Mukherjee, Ranjan Nandy, Anupama Saha, Anjan Pal, Mamta Chawla (upto 30.10.98), Shreyoshi Chatterjee, Sarmistha Mukherjee (upto 4.9.98), Sandip Bhattacharya, Sanjukta Ghosh, Biswaswar Nandi, Shuvendu Das, Chandrajit Lahiri, Manika Das, Abhijit Basu, Mahashweta Mitra.

SUPPORTING STAFF MEMBERS

S.K. Das (I), S.K. Das (II), Gita Dey (upto 31.12.98), Dr. Bandana Banik, N.K. Malakar, B.N. De Sarkar, B.N. Mandal, P.K. Chattopadhyay, P. Gupta, S. Gazi, D. Sarkar, Babli Marik, Prabir Halder, Debashis Majumdar, B. Bahali, N. Patra, J. Prasad, B. Bhattacharyay, R. Prasad, B. Ram, B. Das, S. Das. Rabin Pal, Sankar Bari, Purnendu Manna, Narayan Patali.

Biochemistry

Faculty : Prof. B. Bhattacharyya (Chairman), Prof. N.C. Mandal, Prof. S. Biswas (upto 31.5.98), Dr. P. Sinha, Dr. P. Chakrabarti, Dr. A. Lohia, Dr. R. Chattopadhyaya, Dr. P. Parrack.

HIGHLIGHTS

Goat brain tubulin has been shown to prevent aggregation of several proteins unrelated in structure or sequence. This chaperone-like activity for tubulin has been demonstrated for the first time.
It has been shown that there are two distinct cyclic AMP dependent functional forms of CRP, a prokaryotic transcription factor.

INSTITUTIONAL PROJECT I

A.1. *Seed Protein Genes and Transgenic Plants :* **R.K. Mandal** (Retired) and Surekha Mandal.

The work on the transgenic plants expressing a trypsin inhibitor gene from mustard and thereby showing insect resistance has been completed and phased out. The T_3 generation transgenic tobacco and tomato seeds have been kept for field trial.

A.2. *An unique Apyrase from Mimosa pudica :* **Susweta Biswas**, Mita Sen, & Niladri S. Kar

Tubulin protein, when seperated from MAPs, is polymerized in presence of taxol or DMSO, to form microtubule. Purified apyrase isoform I and II, from sensitive plant *Mimosa pudica*, seperately can induce tubulin polymerization. This has been studied spectrophotometrically. The microtubule, formed either in presence of apyrase isoform I and II, is depolymerized at 0°C indicating the retention of its specific characteristic. These microtubule structures were examined under electron microscope. Both isoform I & II are found to be associated with microtubules showing that these proteins are involved in the tubulin polymerization. Pattern of the MT formation is different in case of isoform I & II.

Enzymatic parameters (Km & Vmax) and thus substrate specificity for various nucleotide di- and tri-phosphates were obtained in presence of various divalent cations with the help of partially purified *Mimosa* apyrase. For example, in presence of Mn^{++}, the substrate-specificities in descending order are as follows: ADP>GTP> ATP >GDP (9.4: 4.3: 3.7: 1). In case of ATP, the substrate specificity showed dependence on the divalent cation present in the assay condition: Mn^{++}>Mg^{++}> Ca^{++} (3:2.2:1). Since most of the apyrases reported so far have higher substrate specificity for ATP rather than ADP, the *Mimosa* apyrase may represent a novel type.

INSTITUTIONAL PROJECT III

A.1. *Studies on Developmental Regulation in Mycobacteriophage L1 :* **N. C. Mandal**, Prajna Mandal, Chandrani Chattopadhyay, and Rajat Bhattacharyya

One important improvement in the purification of L1-coded exonuclease was made. Earlier, during purification the exonuclease was losing activity. By trial experiments, it was observed that partially purified enzyme showed maximum activity (about 5 to 6-fold higher) in the presence of both Mn^{++}(5mM) and Mg^{++}(10mM) compared to that in presence of 3 mM Mg^{++} as was used earlier. Also, the activity of the partially purified enzyme was completely inhibited in presence of 10 mM EDTA. This property may be utilized in further purifying the enzyme by affinity chromatography.

Earlier studies showed that the G27 gene positively regulates the transcription of delayed early genes and two genes, G23 and G25 regulate the transcription of late genes of L1. To get further support in favour of these conclusions, study of L1-coded protein synthesis by L1G27ts901, L1G23ts23, and L1G25ts889 mutants at 32 and 42oC was attempted. The results showed that the former mutant could synthesize only the immediate early proteins but none of the delayed early and late proteins at 42oC. While each of the two latter mutants could make early and delayed early proteins like wild-type L1, G23ts was defective in the

synthesis of some of the late proteins and G25ts in all of the late proteins. This suggests a complicated control of late gene expression.

The nucleotide sequence of the early regulatory gene G27 of L1 was determined and confirmed by sequencing both the strands. An analysis of the sequence reveals that there is a putative promoter with its -10 and -35 regions and its structure looks like that of *E. coli* promoter. Several ORFs with their putative SD sequences upstream have been indicated. Computer search shows that several blocks of this DNA bear about 98% homology with different segments of mycobacteriophage L5 DNA. One interesting point to note that all those homology blocks have GATC at both the ends. A 200 bp from the N-terminal part of the coding sequence is repeated directly after the C-terminal ends of all the possible ORFs. Identification of the actual ORF, the upstream promoter and downstream terminator (if there is any) and upstream operator (if any) are in progress.

Cloning of five L1 promoters was reported earlier. One of these five promoters, P5 was shown to be a strong early promoter of L1. Preliminary in vitro transcription using the 700 bp DNA from subclone pCM8 containing the above promoter and mycobacterial RNA polymerase produced a 380 b long transcript suggesting the possible position of the promoter in the DNA. Location of the promoter in the L1 genome has been determined. Sequencing of a portion of this 700 bp DNA has been done. Properties of L1G23ts23 mutant suggested the existence of a third late regulatory gene of L1 which is possibly regulated by G23. In this venture, one ts mutant of L1 was isolated which did not complement L1G23ts23 phage but could make functional exonuclease at 42oC. The wild-type counterpart of this mutant gene has been cloned. This gene in 800 bp DNA segment is being sequenced. (Supported by CSIR, Govt. of India)

A.2. *Tubulin substructure and Identification of its Structural. Functional and Regulatory Domains* : **B. Bhattcharyya**, Suranjana Guha, Tapas Kumar Manna, Sankar Bhattacharyya and Taradas Sarkar

Tubulin, a ubiquitous protein of eukaryotic cytoskeleton, is a building block unit of microtubule. Although several cellular processes are known to be mediated through the tubulin-microtubule system, the participation of tubulin or microtubule in protein folding pathway has not yet been reported. Here we show that goat brain tubulin has some functions and features similar to many known molecular chaperones. Substoichiometric amounts of tubulin can suppress the non-thermal and thermal aggregation of a number of unrelated proteins such as insulin, equine liver alcohol dehydrogenase and soluble eye lens proteins containing β- and gamma-crystallins. This chaperone-like activity of tubulin becomes more pronounced as temperature increases. Aging of tubulin solution at 37°C also enhances its chaperone-like activity. Tubulin loses its chaperone-like activity upon removal of its flexible hydrophilic C-terminal tail. These results suggest that both electrostatic and hydrophobic interactions are important in substrate binding by tubulin and that the negatively charged C-terminal tails play a crucial role for chaperone-like activity.

A.3. *Structure Function Relationship of Tubulin-Drug Interaction* : **B. Bhattacharyya**, Pradip Mahapatra & Shalmali Chakraborty

Non covalent hydrophobic probes such as bisANS has become increasingly popular to gain information about protein structure and conformation. However, there are limitations as bisANS binds nonspecifically at multiple sites of many proteins. Successful use of this probe depends upon the development of binding conditions where only specific dye-protein interaction will occur. In this study, we have shown that the binding of bisANS to tubulin occurs instanteneously, specifically at one high affinity site when 1 mM GTP is included in the reaction medium. Substantial portions of protein secondary structure and colchicine binding activity of tubulin are lost upon bisANS binding in absence of GTP. BisANS binding increases with time and occurs at multiple sites in absence of GTP. Like GTP, other analogs, GDP, GMP, ATP also displace bisANS from the lower affinity sites of tubulin. We believe that these multiple binding sites are generated due to the bisANS - induced structural changes on tubulin and the presence of GTP and other nucleotides protect those structural changes.

A.4 *Chromosome Transmission in Yeast* : **P. Sinha**, Niranjan Das, Kaustuv Sanyal, Atasi Poddar, Santanu Ghosh and Sujata Hajra.

Work on *mcm* mutants of the yeast *S. cerevisiae* defective in chromosome segregation was continued last year in my laboratory. We published our results on the cloning and characterization of *MCM16, MCM21* and *MCM22* genes. Given below is a brief summary of this work .

(i) *MCM18/CTF19 and MCM19*

These genes were cloned by complementation using the same selection scheme as described for *MCM16*. Subcloning and sequence analysis of the complementing clone showed that *MCM18* codes for the same ORF as *CTF19*, a gene required for chromosome segregation (Hyland K *et al.* (1999) J Cell Biol. 145 15-28) *MCM19* codes for a novel protein - ORF YBR107C. Our studies, similar to those performed for *mcm16*, *mcm17* and *mcm21* and *mcm22* have also confirmed the roles of these genes in chromosome segregation.

(ii) *Genetic interactions of MCM16 and MCM21 with microtubule-organising genes*

In a further attempt to understand the molecular mechanisms by which these genes act, we have begun screens for the identification of other genes with which *MCM16*, *MCM21* and *MCM22* interact. It has been found that *MCM16* and *MCM21*, both interact genetically with *TUB1*, coding for alpha-tubulin in this yeast. That is, while each of the single mutants *tub1*, *mcm16* and *mcm21* can grow at 18°C, without any loss in cell viability, the double mutants *tub1 mcm16* and *tub1 mcm21* fail to grow at this temperature. Similar interactions were found with *CIN1*, a gene putatively required for beta-tubulin folding in this yeast. Thus, the effects of *mcm16* and *mcm21* mutations get enhanced in the presence of microtubules which may be compromised in their structure but are, nevertheles, functional. We have shown that both *MCM16* and *MCM21* are required for kinetochore function (Sanyal *et al.*, 1998; Poddar *et al.*, 1999) but cells carrying deletions in these genes are viable. Therefore, small perturbations in the microtubule structure or in the kinetochore function can be tolerated by a cell, but only when these defects are present singly. A simultaneous presence of these defects can completely breakdown kinetochore-microtubule interactions. An understanding of the process by which *mcm16* and *mcm21* affect kinetochore structure will help in understanding the molecular basis of kinetochore microtubule interactions.

Other screens, such as identification of interacting proteins by two-hybrid analysis, synthetic lethality and dosage suppression are also ongoing in my laboratory.

(iii) *Subcellular localization of Mcm proteins*

We are concentrating on the subcellular localization of all the Mcm proteins. For this we have constructed fusions of GFP (green fluorescent protein) with the Mcm protein in high copy expression vectors. Preliminary experiments have localized Mcm21p to the nucleus and others (Mcm16 - Mcm19 and Mcm22 proteins) to the cytoplasm. These observations are being confirmed by using expression vectors which have lower copy numbers or by using conditions under which protein expression can be regulated.

A.5. *Structural analysis to explain helical propensities of amino acid residues* : **P. Chakrabarti** and Debnath Pal

All known structures from the Brookhaven Protein Data Bank have been analysed to find out the correlation of the main and side-chain conformations of different residues. There is no general explanation for the difference in the helical propensity values for different residues. Our work has shown that a parameter, CS_{AX}, which indicates the similarity in the conformational features (Phi, Psi and chi1 angles) of different residues (X) with Ala (A) (which has the highest propensity value), has a very strong correlation with the propensity of the residue to occur in helices.

A.6. *Protein structure visualization* : **P. Chakrabarti** and Debnath Pal.

Visualization of protein structure is an important aspect of any study pertaining to structure-activity correlation. We have devised a plot in which the three-dimensional information of a structure can be represented in two dimensions by depicting the regions in the

conformation space (phi, psi and chi1 torsion angles) to which the residues belong. Secondary structural features, presence of any *cis* peptide bond or disulfide linkage are also shown, and the plot can also be used to compare two structures and to highlight the effect of mutation on structure.

A.7. *Analysis of the environment of tryptophan residue :* **P. Chakrabarti**, Uttam Kumar Samanta and Debnath Pal

The environment of Trp residues, which are very important for studying protein structures and dynamics using fluorescence spectroscopy, has been found out from an analysis of known protein structures. Trp residues are also found at the binding sites of many enzymes, and important lessons on molecular recognition and binding affinity can be derived from our analysis.

A.8. *Aromatic-aromatic interactions in and around helices and their use in the design of helical peptides :* **P. Chakrabarti** and Ajoy Koomer

Stacked and edge-to-face interactions between two aromatic residues are known to be stabilizing. We are analysing if such motifs are found when a pair of aromatic residues belonging to a helix or a helix and its immediate neighbourhood are spatially close, and if so, if there is any pattern in the sequence in the intervening region. The results will be used to design sequences with high helicity brought about by aromatic-aromatic contacts.

A.9. *Crystallographic studies of lipase from Aspergillus niger :* **Rajagopal Chattopadhyaya** and V.M.H. Namboodiri.

Protease digested fragments of the purified lipase after denaturation were purified for protein sequencing. Properties of the enzyme upon removal of covalently attached carbohydrate were studied. The lipase was further characterized by pI determination and compared with another fungal lipase obtained from Amano, Japan in their biochemical properties. Crystallographic trials with the lipase have not yielded X-ray diffracting crystals so far. Trials are also being performed with the carbohydrate depleted lipase.

A.10. *Sequence determination of lipase :* **Rajagopal Chattopadhyaya** and Swati Ray

Sequence determination efforts for the lipase have just commenced starting from total cellular RNA and subsequent RT-PCR using the N-terminal specific oligo primer. N-terminal sequencing of the lipase was previously determined.

A.11. *Crystallographic studies of lambda repressor and mutants :* **Rajagopal Chattopadhyaya** and Kausik Ghosh

Last year the crystallization of the wild type and a mutant carboxyterminal domain were reported. The former diffracted very weakly and the latter weakly, and a data set was collected at the facility in SINP. The x-ray data are being processed to determine the unit cell parameters. Interpretation is difficult due to weak diffraction. Improved diffraction may be obtained by low temperature data collection or synchrotron radiation.

A.12. *Model building studies of intact lambda, P22, 434 and LexA repressors :* **Rajagopal Chattopadhyaya** and Kausik Ghosh

The complete models of all four repressors were deposited at RCSB (http://www.rcsb.org), the new headquarters of PDB, and accession codes obtained. Several manuscripts have been prepared and two already submitted.

A.13. *Structural Basis of Control of Transcription Initiation in E coli :* **P. K. Parrack**, Jayanta Mukhopadhyay & Sibes Bera

In *E. coli*, the transcription activator protein CRP (Cyclic AMP Receptor Protein) adopts an active conformation only when it is complexed with cyclic AMP (cAMP). Recent x-ray crystallographic studies indicate that there are two types of binding sites for cAMP in CRP, with different affinities. Thus, different cAMP-CRP complexes are formed at low and high concentrations of cAMP. Using *in vitro* transcription experiments, we have shown that the low cAMP complex can activate transcription. However, the high cAMP concentration complex, whose conformation resembles that of unliganded CRP, loses the ability to activate. Further, we have correlated the above properties of CRP at low and high cAMP concentrations with the DNA binding property of the protein, and shown that at high cAMP, the cAMP-CRP complex cannot bind efficiently to specific sites.

A.14. *Role of upstream sequences in promoter recognition by RNA Polymerase :* **P. K. Parrack** & Runa Sur

Binding of *E. coli* RNA polymerase to certain nonpromoter sequences in the *E. coli* gal promoter region has been studied in detail. One such sequence has been located in the promoter-upstream region of the gal operon, by DNase I footprinting. It covers 44 base pairs, centered around -100, contains a classical Pribnow box TATAAT, and can give rise to run-off transcripts in vitro. By primer extension experiments, we have seen that transcripts corresponding to this promoter-like element (called P3), occurs *in vivo*, in wild type as well as in cya- cells. *In vivo* experiments wherein the promoter activity of the wild type promoter (including P3) has been compared with a "P3-less" or "P3-only" situation using the promoterless vector pTAC have also been carried out. It is observed that while P3 alone has very weak promoter activity, its presence does contribute to the promoter strength of galP1/P2.

A.15. *Structural Studies on the cII protein of bacteriophage lambda :* **P. Parrack** & Ajit Bikram Datta

The cII protein of lambda phage is a transcription activator which activates the pRE and pint promoters in lambda phage and promotes lysogeny. CII is a small, tetrameric protein with M.W. 4 x 11 KDa. Not much is known about its structure. To obtain the protein in milligram quantities required for structural studies, the cII gene has been cloned into a high expression vector (pET3a) under T7 promoter control. The protein expressed from this clone (pAB305) has been purified. At higher concentrations required for structural studies like crystallography or NMR, there was a tendency of the cII protein to form aggregates. This problem has been surmounted, and sufficient quantities of the protein are now available in required concentration. Crystallization of cII is in progress, in collaboration with Dr. P. Chakrabarti of our department.

INSTITUTIONAL PROJECT IV

A.1. *Fish Genetic Stock Identification and Conservation :* **R.K. Mandal** (Retd.)

The work on this project has been phased out after successful completion.

INSTITUTIONAL PROJECT V

A.1. *Phosphoinositide signalling and Ca2++ Homeostasis in Entamoeba histolytica :* **Susweta Biswas** & Jayasree Basak

The formation of both Ins(1,4,5)P_3 and Ins(1,3,4,5)P_4 in *E. histolytica* was demonstrated. Phosphatidyl inositol, Phosphatidyl 4-monophosphate and Phosphatidyl inositol 4,5-bisphosphate could be formed by the phosphoinositides kinase(s), presence of which was detected in the membrane fraction of *E. histolytica*. The phosphatidyl inositol 4,5-bisphosphate was rapidly hydrolysed by phospholipase C to produce Ins(1,4,5)P_3. This was further phosphorylated to form Ins(1,3,4,5)P_4 by inositol(1,4,5)P_3-3 kinase which was detected in the cytosol of this organism. Calcium uptake in *E. histolytica* was mediated by calcium dependent ATPase which was found to be located in the membrane. Protein kinase C activated by diacylglycerol (DAG) and MAP kinase involved in the downstream network of signal transduction were found to be present in *E. histolytica*.

A.2. *Molecular genetics of Entamoeba histolytica :* **Anuradha Lohia**, Anasuya Ganguly, Suchismita Das, Sulagna Ghatak

We have been studying the regulation of cell division in amoeba for the past few years. Our earlier studies have identified several homologues of eukaryotic cell cycle controlling genes in amoeba such as cdc2, ras, rap, rho, rac, gamma tubulin, MCM3, inositol 1-phosphate synthase, mos and p21 activated kinase. We have also used multiparameter flow cytometry to study the cell cycle phases in amoeba and the expression of the above named proteins during the cell cycle. We have demonstrated that unlike other eukaryotes where DNA duplication occurs only once per cell cycle, amoeba cells replicate their DNA more

than once per cycle leading to a heterogenous polpulation of euploid and polyploid cells in culture. Thus the paradigm of checkpoint controls which regulate the progression of the eukaryotic cell cycle appears to be different in amoeba. Furthermore, eukaryotic chromosomes segregate on a single microtubule spindle assembly after the centrosome/ Microtubule organising centre has duplicated. In amoeba cells we have multiple extranuclear MTOCs. To better understand the regulation of cell division in amoeba we have therefore asked the following questions : 1) What are the molecular events regulating cytokinesis in dividing amoeba? 2) Does the amoeba genome need a 'license' to replicate? 3) Are ali the multiple microtubule organising centres required for spindle formation in amoeba? In our studies with the regulation of cytokinesis we have identified two genes - *Eh dia* and *Eh septin* - which are sequence homologues of a) the Drosophila gene coding for the diaphanous protein and b) the yeast *cdc3* family of genes. Both these eukaryotic genes regulate cytokinesis. We have cloned their homologues in amoeba, expressed them in *E.coli* and are at present immunising rabbits to raise polyclonal antisera against these proteins. We plan to localise these proteins by immunofluorescence and to study their biochemical functions once the antisera is ready. We have observed that the levels of free intracellular Calcium change during the different cell cycle phases and during differentiation of trophozoites into cysts in *Entamoeba invadens* by flow cytometry. Our earlier results show that p21 rho accumulates to a high concentration in polyploid cells or cells which have replicated their genome several times without undergoing cytokinesis. We plan to follow the p21 rho mediated signal transduction pathway which controls both cytokinesis and encystation in amoeba.

We have completed the full length sequences of *Eh MCM2* and *Eh MCM5* - homologues of *MCM2* and *MCM5* which along with *MCM3* act as licensing factors for the initiation of eukaryotic DNA replication. In the light of our findings that amoeba DNA replication can occur more than once per cell cycle, we are studying the role of these proteins in regulating DNA duplication in amoeba. We have found that these proteins are only present when DNA synthesis is arrested by serum starvation in the nucleus. They rapidly disappear when serum is added to starved cells. We plan to further characterise their function with the use of suitable inhibitors at different phases of the cell cycle.

We have cloned the beta tubulin gene from amoeba (previously cloned by another group) and expressed it in *E.coli*. Rabbits have been immunised with the recombinant beta tubulin protein for raising polyclonal antisera. Using a combination of anti-gamma tubulin antibody and anti-beta tubulin antibody we would like to visualise spindle formation from MTOCs to determine if each of them forms a functional spindle.

We have isolated chromosome sized DNA moelcules and restricted it *in situ* in agarose blocks for the purpose of constructing Yeast artificial chromosomes of amoeba.

PUBLICATIONS

1. Mandal S and Mandal RK 1999 Application of Plant Tissue Culture and Biotechnology for Improving Agricultural Productivity. *Sci. Cult.* **65** No. 3-4.

2. Raychaudhuri S, Basu M and Mandal NC 1998 Glutamate and cyclic AMP regulate the expression of galactokinase in Mycobacterium smegmatis. *Microbiology* **144** 2131-2140.

3. Jana NK, Roy S, Bhattacharyya B, and Mondal NC 1999 Amino acid changes in the repressor of bacteriophage lambda due to temperature-sensitive mutations in its cI gene and the structure of a highly temperature-sensitive repressor. *Protein Engineering* **12** 225-233

4. Chakraborty S, Sarkar N and Bhattacharyya B 1999 Nucleotide-dependent bisNAS lrinding to tubulin Biochem.Biophys Acta, **1432** 350-355.

5. Ghosh R, Biswas S and Ray S 1998 An apyrase from *Mimosa pudica* contains N5, N10-methenyl tetrahydrofolate and is stimulated by light, *Eur. J. Biochem.* **258** 1009-1013.

6. Sanyal K, Ghosh SK and Sinha P 1998 The *MCM16* gene of the yeast *Saccharomyces cerevisiae* is required for chromosome segregation. *Mol. Gen. Genet* **260** 242-250.

7. Poddar A, Roy N and Sinha P 1999 *MCM21* and *MCM22*, two novel genes of the yeast *Saccharomyces cerevisiae* are required for chromosome transmission. *Mol. Microbiol.* **31** 349-360.

8. Pal D and Chakrabarti P, Different types of interactions involving cysteine sulfhydryl group in proteins. *J. Biomol. Struct. Dyn.* **15** 1059-1072.

9. Chakrabarti P and Pal D, Main-chain conformational features at different conformations of the side-chains in protein. *Protein Engng.* **8** 631-647.

10. Samanta U, Chakrabarti P and Chandrasekhar J 1998 Ab initio study of energetics of X-H...pi (X=N,O, and C) interactions involving a heteroaromatic ring. *J. Phys. Chem.* **A102** 8964-8969

11. Chakrabarti P and Chakrabarti S, 1998 C-H...O hydrogen bond involving proline residues in alpha-helices. *J. Mol. Biol.* **284** 867-873.

12. Lohia A, Hait NC and Majumder AL 1999 L-myo-Inositol 1-phosphate synthase from *Entamoeba histolytica*: characterisation of the gene and the enzyme. *Mol. Biochem. Parasitol.* **98** 67-79.

13. Chattopadhyaya R and Ghosh K Knowledge based model of a LexA repressor dimer (theoretical model) bound to RecA operator. RCSB000408, PDB code 1QAA, processed 2.2.99

14. Chattopadhyaya R and Ghosh K Knowledge based model of a lambda repressor tetramer (two dimers) bound to two adjacent operator sites. RCSB000598, PDB code 1LMD, processed 9.3.99.

15. Chattopadhyaya R and Ghosh K, Phage 434 repressor tetramer (two dimers) model bound to two adjacent operator sites. RCSB000681, PDB code 1RPD, processed 19.3.99.

16. Chattopadhyaya R and Ghosh K P22 c2 dimer model bound to operator DNA. RCSB000693, PDB code 1QAR, processed 22.3.99.

17. Ghosh K 1998 Apoptosis or programmed cell death : the current scenario, *Science and Culture*, **64** 244-248.

Participation in Conferences/Symposia/ Training programme/ Awards and Honours received

Prof. S. Biswas Delivered invited talk at National Symposium on recent advances in Structure, Synthesis & Function of Biomolecules from 4-6, Feb., 1999 organized by Dept. of Chemistry, Bose Institute, Calutta.

Prof. N. C. Mandal Gave a course of lecture on Molecular Biology to the M.Sc. final year students of Biochemistry Department and M.Sc. Bio-technology students (first year) of the Calcutta University, served as a member of the Sectional Committee of Indian National Science Academy, Acted on several occasions as a member of the Assessment Committee of Indian Institute of Chemical Biology, Jadavpur, Calcutta, acted as a member of the Ph.D. Committee in Biochemistry of the Calcutta University, acted as examinar of Ph.D. thesis of different Indian Universaities, attended Guha Research Conference -1998 held at Port Blair, Andaman & Nicobar Islands, served as a member of Programme Advisory Committee (PAC) of the Department of Science & Technology, Govt. of India, invited to chair a scientific session in the Symposium organized by Dr. B. C. Guha Centre for Genetic Engineering and Biotechnology in December, 1998, served as paper setter and examiner for M. Sc. examinations in Calcutta and Visva Bharati Universities.

Prof. B. Bhattacharyya Delivered invited talk at the Annual Meeting of Society of Biological Chemists, India at New Delhi, delivered invited talk at National Symposium on recent advances in structure, synthesis & function of Biomolecules from 4-6 Feb., 1999 organized by Dept. of Chemistry, Bose Institute, Calcutta, elected member of Sectional Committee IX (Biochemistry and Biophysics), INSA ; Elected member of the Sectional Committee of General Biology, Indian Academy of Science, Bangalore; Elected member of the National Committee for IUBMB, INSA.

Dr. Pratima Sinha Elected Fellow of the National Academy of Sciences (Allahabad) in 1998 for contributions in the field of Molecular biology and Genetics.

Dr. P. Chakrabarti Delivered talks at: Guha Research Conference, Port Blair, December 1-7, 1998, 64th Annual Meeting of the Indian Academy of Sciences, Kottayam, October 28-30, 1998, workshop on sequence analysis and molecular modelling, New Delhi, December 17-18, 1998, 67th Annual Meeting of Society of Biological Chemists, New Delhi, December 19-21, 1998, XXIX National Seminar on Crystallography, Chennai, December 21-23, 1998, national Symposium in Chemistry, Bangalore, January 27-30, 1999, discussion meeting on protein folding, SINP, Calcutta February 12, 1999, reviewed thesis for Indian Institute of Science, University of Hyderabad and manuscripts for Protein Science, Protein engineering and Acta Crystallographica D.

Dr. Anuradha Lohia Elected as member of Guha Research Conference, delivered taeks at "Biotechnological approaches to Bacterial and Parasitic diseases" at IICB, Calcutta, "Colloquium on Biotechnology for Women: A vision for the 21st Century", Organised by DBT, India, Guha Research Conference, Port Blair, Andaman Islands, Cancer Research Institute, Mumbai: "Signal Transduction in Biology", 21st Century at Rotary Club, Calcutta; Bhartiya Sanskriti Sansad, Calcutta.

Dr. P. Parrack taught X-ray crystallography to M.Sc. part II students of the Dept. of Biophysics, Kalyani University, Kalyani, W.B. Dr. P. Parrack, Dr. Sibes Bera, Mr. Jayanta Mukhopadhyay, Ms. Runa Sur and Mr. Ajit Bikram Datta attended the "Discussion Meeting on Protein Folding" held at the Saha Institute of Nuclear Physics, 12th February 1999.

GRANTS-IN-AID SCHEMES

Investigator/ Co-investigator	Title	Financed
Prof. S. Biswas	Study of Signal Transduction in plants: Inositol trisphosphate receptor from *Vigna radiata*, its gene cloning and characterization of different domains. (Terminated on Oct., 1998)	CSIR
Prof. S. Biswas	Signal Transduction pathways operative in *Entamoeba histolytica* mediated by Inositol phosphates	DAE
Prof. S. Biswas	Biochemical and Biophysical studies on the novel NTP-NDPase (apyrase) linked with the sensitivity of *Mimosa pudica* leaflets.	DST
Prof. N.C. Mandal	Studies on the mechanism of action of gene G23 in post-transcriptional regulation of bacteriophage L1 gene expression.	CSIR
Prof. B. Bhattacharyya	Studies on carboxy termini of tubulin: Its structural and functional implications (Terminated on 28.2.99)	CSIR
Dr. Pratima Sinha	The role of Mcm family of proteins in initiating DNA replication of eukaryotic replication origins (terminated on Aug., 1998).	DAE
Dr. Pratima Sinha	Characterization of *MCM* genes required for DNA replication and segregation in the yeast *S. cerevisiae*.	CSIR
Dr. P. Chakrabarti	Protein structure and function through data base analysis and crystallography.	DST
Dr. P. Chakrabarti	Analysis of interactions in helices in proteins and their use in the design of helical peptides.	CSIR

Investigator/ Co-investigator	Title	Financed by
Dr. A. Lohia	Studies on the expression of ras superfamily proteins in *Entamoeba histolytica*.	DBT
Dr. A. Lohia	Identification of pathogenic strains of *Entamoeba* by a PCR based diagnostic assay.	CSIR
Dr. A. Lohia	Construction of Yeast Artificial Chromosomes of *Entamoeba histolytica*.	CSIR
Dr. A. Lohia	Transformation of *Entamoeba histolytica* to study the role of Fund proteins involved in the growth, virulence and drug sensitivity of the parasite.	US-India
Dr. A. Lohia	Role of tocotrienols as anti-prolferative agents against *Entamoeba histolytica*.	UNESCO Special Grant
Dr. Rajagopal Chattopadhyaya	Biophysical characterization of DNA oligomers containing UV induced thymine dimers (Terminated on 31.7.98)	DAE
Dr. Rajagopal Chattopadhyaya	Sequence and three dimensional structure determination of lipase from Aspergillus niger (Started on 12.3.99)	CSIR
Dr. P.K. Parrack	Structural Basis of control of transcription initiation in Bacteria by the cyclic AMP receptor protein	CSIR

Ph.D. AWARDED

Name of the candidate	University	Year	Title	Supervisor
Biswajit Roy	Calcutta University	1998	Studies on structure and evolution of seed proteins with special reference to trypsin inhibitor	R.K. Mandal
Nrisinha Dey	Calcutta University	1998	Studies on seed storage proteins of some economically minor plants	R.K. Mandal
Somnath Bhattacharya	Calcutta University	1999	Genetical studies of certain novel yield characters of *Brassica campestris*	R.K. Mandal
Dr. Lopa Adhikary	Calcutta University	1998	Studies on lethal interaction of the bacteriophage lambda replication gene P with its host *Escherichia coli*	N.C. Mandal
Pampi Sarkar (nee Basu)	Jadavpur University	1998	Tubulin Sulfhydryl Groups as Probes to understand its Structure-Function Relationship	B. Bhatta-charyya
Sib Sankar Ray	Jadavpur University	1998	Studies on the growth and pro-liferation of *Entamoeba histolytica*	Dr. A. Lohia
Samudra Saurabh Gangopadhyay	Calcutta University	1998	Identification and characterization of genes regulating growth and proliferation of *Entamoeba histolytica*	Dr. A. Lohia

RESEARCH SCIENTISTS/RESEARCH ASSOCIATES/POST-DOCTORAL ELLOWS/ SENIOR RESEARCH FELLOWS/ JUNIOR RESEARCH FELLOWS

Dr. Ardhendu K. Maji, Dr. Dr. K.K. Singh Chauhan, Dr. Amitabha Chakraborty, Dr. Jayashri Basak, Dr. Bhaswati Goswami, Dr. Mita Sen, Dr. Pradip Kr. Mahapatra, Dr. Niranjan Das , Dr. Sibes Bera, Dr. Suranjana Guha, Mr. Niladri Sekhar Kar, Ms. Surekha Mandal, Mr. V.M.H. Namboodri, Ms. Runa Sur, Ms. Anasuya Ganguly, Mr. Ajoy Koomer, Mr. Tapas K. Manna, Ms. Sujata Hajra, Ms. Sulagna Ghatak, Mr. Samarpan Majumder, Mr. Banabehari Giri, Ms. Suroopa Banerjee, Ms. Shalmali Chakraborty, Ms. Prajna Mandal, Mr. Kausik Ghosh, Mr. Kaustav Sanyal, Mr. Jayanta Mukherjee, Ms. Chandrani Chattopadhyay, Ms. Indrani Datta, Ms. Atasi Poddar, Mr. Santanu Kr. Ghosh, Mr. Shankar Bhattacharyya, Mr. Taradas Sarkar, Mr. Uttamkumar Samanta, Mr. Debnath Pal, Mr. Ajit Bikram Datta, Mr. Rajat Bhattacharyya, Ms. Suchismita Das

SUPPORTING STAFF MEMBERS

Mr. Sukumar Saha, Mr. Subhash Debroy, Mr. Durga Charan Das, Mr. Swapan Kr. Das, Mr. Asim Kr. Banerjee, Mr. Narayan Ch. Datta, Mr. Susanta Kr. Das, Mr. Subhash Chakraborty, Mr. Asim Poddar, Ms. Debarati Kanjilal, Sri Durjyodhan Roy, Sri Biswanath Bez, Md. Asraf Ali Molla, Sri Sudhangsu S. Maiti, Sri Basudeb Ghorui , Sri Dulal Ch. Mandal, Ms. Ranubala Das, Sri Ranjit Das

Biophyics

Faculty: Prof. A. K. Banerjee (Chairman), Dr. S. Roy, Dr. G. Basu

HIGHLIGHTS

The crystallographic structure of Calotropin DII has been solved and Protein sequencing and electron density fitting continuing. Co-crystallization and structure elucidation of a series of small model complexes of amino acids and nucleotides. A few new complex structures have been solved and reported. New insights into molecular basis of multi-partite operator recogntion. Elucidation of thermodynamic basis of cognate tRNA recognition by the synthetases. NMR solution structure determination of several peptides. Identification of novel structural motifs in proteins.

INSTITUTIONAL PROJECT I

Structure, function, dynamics & characterisation of macromolecules & some bio-active molecule

A.1 *Structural elucidation of the catalytic mechanism and interaction at active site of two enzymes: a) Structural studies on Nature's another swiss army knife.Calotropin DII, a thiol protease b) studies on beta lactam-beta lactamase and acetanilide-cyclooxygenase interaction :* Suparna Bhattacharya, Sibani Chakravarty, Asok K. Pal, Umesh Haldar, Sreya Ghosh, Bishnu Mukhopadhyay, **Asok Banerjee**

Proteolytic enzymes are involved in various biological processes with wide importance in medicine and industry. Recently they are implicated in anti cancer and DNA binding activity. We are working on a novel cysteine protease,Calotropin DII.The crystallographic structure of Calotropin DII has been solved by molecular replacement method using Papain as phasing model. The Sequencing and refinement of the Protein is progressing with prime objective of visualizing its substrate specificity to compare it with other cysteine proteases to rationalise a conclusion. Successful sequencing from N-terminal and electron density map confirmed that N-terminal is not blocked as suggested by other worker. A series of inhibitor complexes have been done for crystallization.

One beta lactam and one acetanilide structures have been solved by crystallography and modelling study of the complexes of the beta lactamase and cyclooxygenase respectively with them are being studied . The results are encouraging and a receptor based drug design protocol is taking shape.

A.2 *Visualization of the role of water in protein-nucleic acid interaction at atomic resolution :* Asim K. Bera, Suparna Bhattacharya, Sreya Ghosh, B. P. Mukhopadhyay, U. Haldar, **Asok Banerjee**

Molecular recognition is a subject of fundamental importance in biology. Protein - Nucleic acid complexes embrace many aspects of cellular DNA function and genetic expression.The nature of interaction of these complexes are emerging recently but none at atomic resolution. Our group has succeeded to solve a series of model complexes of amino acids and nucleic acids at atomic resolution by x-ray crystallography. These structures show interesting findings many of which are common in macromolecular complexes and some interaction modes are still to be characterized in macromolecular complexes. Water mediated specific and non-specific interactions of the partners are found out which play a key role in recognition. It is unlikely that these mode of interactions are not exploited in vivo by Nature.

A.3 *Modelling & Structural studies of I) peptidoglycan - a chemoreceptive constituent of Mycobacterium cell wall and II) small molecules of biological importance :* Suparna Bhattacharya, Sibani Chakravarty, Asim Bera, Sreya Ghosh, Bishnu P. Mukhopadhyay, **Asok Banerjee**

i) The cell wall constituents of Mycobacterium are important and are prime targets for chemotherapeutic drugs. Peptidoglycan, a constituent, has been studied and a pseudo two fold symmetry along glycan chain has been found out. This along with X-ray diffraction data and our study excludes many proposed

hypothetical models and endorses the stereochemically consistent and acceptable model "Knox and Murty". The kinetic and binding affinity studies of the specific chemical agents/ligands with model peptidoglycan system has clarified some mode of interaction at molecular level. ii) Structural studies of a series of small molecules of biological interest are continuing with a hope to correlate these structures to their functions. Several of these structures include : one acetanilide analog, and beta lactum analog etc.

A.4. *Molecular Basis of Binding Cooperativity in l-repressor :* Sunanda Deb, Sumita Bandyopadhyay and **Siddhartha Roy**

A non-cooperative mutant of l-repressor, Y210C, has been purified and characterized. The mutant protein does not show any evidence of cooperative interaction as judged by difference near UV circular dichroism spectra of DNA. The mutant protein also shows much weaker self-assembly as revealed by fluorescence anisotropy measurement. Far UV circular dichroism spectrum of the protein shows modest but significant reduction in the 220 nm range suggesting a structural change. Lehrer plot of acrylamide quenching of Y210C repressor at a pre-dominantly dimeric concentration (0.5 mM), is almost identical with that of the wild-type protein at the same concentration. Transmission of operator induced conformational change is also preserved in the mutant protein. Like that of the wild-type protein, cysteines of the mutant protein are unreactive to sulfhydryl reagents under native conditions. Most importantly, C210 is unreactive to sulfhydryl reagents under native conditions. This fact coupled with the structural change observed in far UV CD suggests that C210 is located at the interior of the protein and exerts its effect indirectly on cooperative contact probably through destabilization of a reverse turn, of which it is an important part.

A site-directed mutation, F235C, was created at the penultimate residue of the l-repressor. Under non-denaturing conditions, Cysteine 235 shows half-of-the-sites reactivity, i.e. only one out of two cysteine 235 residues in the dimer shows reactivity towards sulfhydryl reagents. The measured second order rate constant from stopped-flow experiments is only one-fourth than that of the free cysteine, suggesting exposed location of the reactive sulfhydryl. Measurement of dimer-monomer dissociation constant suggested that dimer-monomer dissociation of the mutant repressor is similar to that of the wild-type. Affinity towards a single operator O_R1 is also similar to that of the wild-type repressor. The mutant repressor gene in a multi-copy plasmid confers immunity towards infection by a cI lambda phage, suggesting preservation of functional integrity.

Far UV circular dichroism spectra show no major change in the secondary structure. Fluorescence quenching experiments suggest increased exposure of some tryptophan residues. Urea denaturation profile indicates decreased stability of the C-terminal domain, suggesting that the C-terminal tail region may interact with the protein core.

The relevance of the implicated structural asymmetry of the F235C for the wild-type repressor was tested with a dimer-monomer defective repressor S228N. Dimer-monomer dissociation constant of the hybrid wild-type/ S228N repressor is identical to that of the wild-type, suggesting an asymmetric monomer-monomer interface. We propose that wild-type l-repressor may have a head-to-tail arrangement of identical subunits.

Binding of regulatory proteins to multipartite DNA binding sites often occurs with protein-protein interaction resulting in cooperative binding. The operators of bacteriophage l have several pairs of repressor binding sites (O_R1-O_R2, O_R2-O_R3, O_L1-O_L2 and O_L2-O_L3) separated by a variable number of basepairs and thus, are a model system for studying multipartite operator recognition by DNA binding proteins. Near-UV Circular Dichroism spectra show that the DNA is distorted in O_R1-O_R2 and O_L2-O_L3 but much less so in O_R2-O_R3. Upon titration of l-repressor with single operator sites O_R1, O_R2 and O_R3, it was observed that the tryptophan fluorescence quenches to different degrees, suggesting different conformations of the protein in the three DNA-protein complexes. Acrylamide quenching of tryptophan fluorescence of l-repressor bound to these single operators also show different Stern-Volmer constants, supporting above conclusions.

Titration of l-repressor with oligonucleotides containing pairs of operator sites also causes different degrees of fluorescence quenching. In particular, fluorescence quenching induced by O_R1-O_R2 binding is less than the quenching induced by either of the single operators alone, suggesting additional conformational changes upon establishment of protein-protein contact. Stern-Volmer constants obtained from acrylamide quenching of tryptophan fluorescence of l-repressor bound cooperatively to pairs of operator sites are different from that of the single operator site bound repressors. For example, O_R2-O_R3 bound repressor has significantly higher acrylamide quenchable components than either of the O_R2 or O_R3 bound proteins, again suggesting additional conformational changes upon establishment of protein-protein contact. We conclude that the strategy of recognition of multipartite operator by l-repressor is complex and varied, involving conformational changes in both DNA and protein that is determined by the separation of the binding sites as well as the nucleic acid sequence.

A.5. *Structural Basis of recognition of cognate tRNAs by Aminoacyl-tRNA synthetases :* Amit Mandal, Rajat Banerjee and **Siddhartha Roy**

Discrimination of cognate and non-cognate tRNA by aminoacyl-tRNA synthetases occurs at several steps during the catalytic cycle. The initial binding step certainly discriminates between cognate and non-cognate tRNAs, although the structural and molecular basis of such discrimination is not understood. We have studied the effect of ionic strength on association constants of cognate and non-cognate tRNAs by two aminoacyl-tRNA synthetases, glutamyl and glutaminyl, from *E.coli*. The slopes of the log K_a versus log [MX] plots are all similar and less that 1. There is also no significant difference between association constants in potassium chloride and potassium glutamate. These results are in sharp contrast to specific and non-specific protein-DNA interactions, where both types of interactions are stronger in glutamate than in chloride and, in general, ionic strength effects are much stronger on non-specific complexes than on the specific complexes. Water release stoichiometries were calculated from the effect of glutamate on the association constants using the method of Record and co-workers. With both the synthetases, cognate tRNA binding leads to release of about 10-20 water molecules. Unexpectedly, non-cognate tRNA binding to both synthetases, lead to larger water release stoichiometries, suggesting additional interactions between non-cognate tRNAs and the synthetases. It appears that although the non-cognate tRNAs bind to the synthetases with only a modest decrease in affinity, when compared to the cognate ones, the nature of the interactions are significantly different.

A.8. *Studies of Peptide and Protein Structure and Dynamics Stability and folding properties of 2S mustard seed protein :* **Gautam Basu**, Manasi Ghose, Lipika Pal, Jayati Sengupta and Raja Bannerjee (In collaboration with Prof. R. K. Mondal of the Biochemistry department) We have initiated studies on the stability and folding behaviour of the 2S mustard seed protein. The protein has been succesfully purified and preliminary equilibrium folding / unfolding behaviour has already been completed. Now we are investigating the kinetic behaviour of the folding/unfolding transition.

A.8.1 *Sudies of peptides and proteins exhibiting the 3_{10}-helical secondary structure :*

Analysis of the PDB for the occurrence and stability of 3_{10}-helices has been completed. Several interesting features characteristic of the 3_{10}-helices have emerged from our study including identification of novel supersecondary structural motifs containing a 3_{10}-helix. Synthesis and spectroscopic studies of a series of peptides containing the amino acid Aib is continuing for establishing a thermodynamic scale for the relative preferences of amino acids between the 3_{10}- and the a- helical states. Simultaneously, peptide fragments from proteins that exhibit a 3_{10}-helical conformation in the protein have been synthesized. Preliminary results from spectroscopic studies, CD and NMR, have revealed the conformational variabilty of these peptides. More peptides are being synthesized for a comprehensive understanding of the conformational preferences of peptides which show a 3_{10}-helical conformation in the protein.

A.8.2. *NMR solution structure of a 20 residue peptide from the B domain of Protein A that exhibit unique biological properties :*

Protein-A (42 kDa, 395 residues) is a cell wall binding protein from *S. aureus* which shows diverse array of immunomodulatory properties. It has got a unique property of binding to the Fc region of different IgG molecules. Different proteolytic fragments of this protein which retain Fc binding activity are thought to be responsible for different biological properties shown by protein-A. The solution conformation of one of these fragments (20 amino acid), is being studied by NMR. Along with CD experiments we have tentatively determined the solution conformation of this peptide to be non-helical and extended, with a turn in the middle. More work is going on and in this project the solution conformation of several other related peptides will also be studied.

PUBLICATIONS

1. Bera AK, Mukhopadhyay BP, Pal AK, Haldar U, Bhattacharya S and Banerjee A 1998 Hydrogen bonding co-operativity of water to nucleotide recognition. Structure of Octadecahydrated inosine 5'-monophosphate, 2(C10H12N4O8P).18H2O at atomic resolution ; J. Chem. Crystallogr. USA **27** 509-516.

2. Roychowdhury C, Banerjee A, Bag P and Roy Shamal 1998 Topological shape and size of peptides : Identification of potential Allele Specific Helper T Cell Antigenic sites ; J. Chem. Inf. Comput. Science. **69** 402-408.

3. Deb S, Bandyopadhyay S and Roy S 1998 A spectroscopic study of Y210C l-repressor: Implications for cooperative interaction; Prot. Engg. **11** 481-487.

4. Roy S, Garges S and Adhya S 1998 Activation and Inhibition of transcription by differential contact: Two sides of a coin; J.Biol.Chem. **273** 14059-14062.

5. Ghosh R, Biswas S and Roy S 1998 A complex apyrase implicated in the response of mimosa pudica to stimuli contains methenyl tetrahydrofolate and acts as a blue light receptor; Eur J Biochem. **258** 1009-13.

Participation in conferences/symposia/ training/programme/Award and honours received.

Prof. A Banerjee participated as an organizing Committee member of International school on powder diffraction held at IACS, Calcutta in October 1998.

Dr. S. Roy visited Bruker-Spectrospin in Switzerland for two weeks in February 1999 for training programme in multidimensional NMR spectroscopy.

Dr. G. Basu visited Bruker-Spectrospin in Switzerland for two weeks in January 1999 for training programme in multidimensional NMR spectroscopy, spent two and a half months begining October 15 1998 in the laboratory of Prof. Dinshaw Patel of Memorial Sloan Kettering Cancer Center, New York, as a visiting investigator.

GRANTS-IN-AID SCHEMES

Investigator/ Co-investigator	Title	Financed by
Prof. A. Banerjee	Structural basis of differences in catalytic mechanism of different Cysteine Proteases, Calotropin DII by X-ray and modelling	DBT
Dr. S. Roy	A study of structural and molecular basis of co-operative binding of operator sites of repressor.	CSIR
Dr. S. Roy Dr. G. Basu	Molecular basis of recognition of tRNAs by Aminoacyl-tRNA synthetases	DST
Dr. G. Basu	Theoretical studies of peptide and protein dynamics from a collective motion viewpoint	CSIR
Dr. G. Basu	A Thermodynamic Scale for the Relative Preferences of Amino Acids between the 3_{10}- and the a- Helical States.	DST
Dr. G. Basu (with Dr. P. Chakrabarti)	Analysis of interactions in helices in proteins and their use in the design of helical peptides	CSIR

RESEARCH SCIENTIST/RESEARCH ASSOCIATES/POST DOCTORAL FELLOWS/ POOL OFFICERS/SENIOR RESEARCH FELLOWS/JUNIOR RESEARCH FELLOWS

Dr. Jayati Sengupta, Mr. Amit Kr. Mondal, Mr. Rajat Banerjee, Ms. Sunanda Deb, Ms. Manashi Ghosh, Ms. Lipika Pal, Ms. Ruchira Chattopadhyay, Ms. Suparna Bhattacharya, Ms. Sibani Chakravarty, Ms. Sanchari Kar

OTHER SUPPORTING STAFF MEMBERS

A. Bhattacharyya, B. Majumdar, B. Marick, B. Munshi, S. Jogsharma.

Animal Physiology Section

Faculty : Prof. A. K. Ray (In-Charge), Dr. Rama Ghosh (upto April, 1998), Dr. G. Sa.

HIGHLIGHTS

It has been established that endothelin (ET) stimulates differentiation as well as maturation of glial cells to microglial cells, the resident macrophage of brain through the activation of differentiation specific gene expression. The factors involved in these signaling pathways are 91 kDa transcription factor called signal transducer and activator of transcription or STAT. After tyrosine and serine phosphorylation p91 STAT homodimerizes and translocates to nucleus where it binds to specific nucleotide sequence (SIE) and activates specific gene transcription. Thyroid hormone affects adult mammalian brain function through rise in intrasynaptosomal Ca 2^+ level and activation of NOS activity.

INSTITUTIONAL PROJECT I

A. Environmental Physiology and Hormonal Regulation :

A.1. *Carbofuran induces cytochrome P450 1A1 gene in primary culture of fish hepatocytes : Effect of trace elements :* **Rama Ghosh, Arun K. Ray** and Manik Ch. Ghosh

In course of our investigation on the activities of the biomarker enzyme ethoxyresorufin-o-deethylase (EROD) we have demonstrated the stimulatory role of carbofuran on the enzyme activity. In our subsequent experiment we have demonstrated the inhibitory effect of copper added as trace element in the hepatocyte primary culture from Singi fish (*Heteropneustes fossilis* Bloch). The data suggest that copper could down-regulate the EROD activity by decreasing the stability of glutathione which helps in maintaining the strucutural integrity of EROD.

A.2. *Production of triploid and unisex fish: Precocious maturation of ovary in unisex Indian Catfish (H. fossilis) :* **Arun K. Ray** and Basant K. Tiwary

Treatment of testosterone to the one day old hatchlings of Singi fish at an appropriate dose for a month caused paradoxical feminization (100%) and precocious maturation of ovary, within 6-7 months in laboratory condition. The average diameter of oocytes in ovary of precociously matured fish (PMF) was significantly higher than the control females. The ovary in PMF showed large ova at ripe or mature stage.

Highly immunoreactive GnRH cells were found in preoptic area of hypothalamus with higher levels of GtH-II and sex steroids in plasma of PMF in preparatory phase of ovarian cycle, in comparison to the control female fish. The unisex production of catfish with precocious ovarian maturation by hormonal manipulation may be useful for "out-of-season" fish production.

A.3. *Role of estrogen in ovarian development in freshwater prawn (Macrobrachium rosenbergii) : Estrogenic Induction of vitellogenin synthesis in hepatopancreatic cells in primary culture :* **Arun K. Ray,** Anuradha Chaudhuri and Sharmila Chatterjee

Primary cultures of the hepatopancreatic cells of freshwater prawn has already been established in our laboratory. After further fine tuning of the culture conditions for optimum viability and attachment, the cells were used to study *in vitro* effects of different growth factors and hormones on their growth , viability and biosynthetic activities. Among the growth factors studied, hepatocyte growth factor was found to be more effective in inducing growth and proliferation and maintaining viability of the cells as confirmed by flow cytometric analysis and the usual dye exclusion test and LDH assay. The cultured cells when subjected to estradiol- 17β insult , released a 76 kD protein into the culture medium similar to vitellogenin (Vg), the egg-yolk precursor present in hemolymph as shown by SDS-PAGE. Further analyses using antibodies raised against Vg will reveal the identity of the protein secreted by prawn hepatocytes in response to exogeneous esrtrogen and the type of the cells that synthesise it.

A.4. *Thyroid hormone regulation of mammalian brain function :* **Arun K. Ray** and Nilkanta Chakraborty

To evaluate the rapid non-genomic action of thyroid hormone we have already demonstrated the stimulation of Ca^{2+}- ATPase, AChE and rise in $(Ca^{2+})i$ in adult rat cerebrocortical synaptosomes. Further study demonstrated the T_3- induced rise in $(Ca^{2+})i$ to be responsible for activation of nitric oxide synthase (NOS) enzyme. Such activation of the enzyme is considered to be a delayed (10- 30 sec) effect preceded by the rise in $(Ca^{2+})i$ (within 5 sec),in depolarised condition. In non-depolarised condition such T3-induced changes did not occur in synaptosomes. Such observations are much convincing evidences for T_3 to be responsible for non-genomic neuronal Ca^2 -induced signal generation for neuronal -NOS activation and NO production.

A.5. *Phenylhydrazine hydrochloride (PHH) -thyroid hormone interaction in rat: A dose-specific dual role of PHH on thyroid gland :* **Arun K. Ray** and Mitali Pramanik

In course of our investigation on PHH effects in juvenile male rat we have earlier observed a generalised goitrogenic role of this compound. Further, on critical analysis of he data , a thyrotropic nature of PHH was visualised at lower dose (10µg/g). PHH behaved like a thyroid stimulator leading to a significant rise in serum thyroid hormone levels (T_4, T_3) with typical hyperactive structure of thyroid gland. At higher doses (20 or 30 µg/g) PHH caused abrupt fall in T_4 and T_3 levels with typical goitrous appearance of thyroid gland in juvenile male rats. The fact indicated a dose -specific dual role of PHH on thyroid gland, the mechanisms of which are being investigated.

B. Control of growth and development of silkworm and silk production.

B.1. *Nature of variations of estradiol-17beta level in haemolymph of 5th instar silkworm larvae :* **Arun K. Ray** and Bela Keshan

During our investigation on the metabolic role of estrogen (E_2) in silkworm we have found anabolic effect of the vertebrate female sex steroid in silkgland of *Bombyx mori* L. Further studies were undertaken to know the age-dependent changes in the E_2 titre to correlate its necessity with the normal events of metamorphic changes in the 5th instar. Although, a regular pattern of oscillation in the haemolymph level of E_2 was observed in different days in control larvae at 5th instar, a sharp high level of E_2 was identified on the 10th day preceded by a small increase on the 9th day. In E_2-treated larvae an exaggerated but control-like oscillations of the hormone level was found. The fact may indicate presence of a fine regulatory mechanism for E_2 titre in the haemolymph of 5th instar larvae.

C. Cell Signalling

C.1. *Molecular mechanisms of endothelin-induced differentiation and maturation of Microglial cells, the resident macrophages of brain :* **Gaurisankar Sa**, Tanya Das (Immuno-technology section), Suman Pal and Tathagatha Chaudhury

Endothelins (ETs) are a class of homologus 22 amino acid regulatory polypeptides that elicit various responses in many tissues. For the first time, we have demonstrated that ET-1 elicits a distinct signalling mechanism by which it mediates glial cell differentiation. The initial event is ligand-induced ET receptor aggregation which may involve single or multiple receptor subtypes. Receptor aggregation concomitantly activates JAK-1 (Janus Kinase 1, a non-receptor tyrosine kinase) through tyrosine phosphorylation. Phosphorylated and activated JAK-1 then tyrosine phosphorylates p91 STAT (Signal tranducer and activator of transcription) protein. The tyrosine phosphorylated p^{91} STAT is then homodimerized forming a sis-inducible factor (SIF) and translocates to the nucleus. The SIF is then additionally phosphorylated at the serine residue by p^{42} MAPkinase, activated by ET-1 through a protein kinase C-dependent signalling pathway. The phosphorylated and activated SIF then binds to sis-inducible element (SIE), which alone is sufficient, under certain circumstances to mediate endothelin-induced transcriptional activation of differentiation specific genes.

C.2. *Mimicry of microglial cells as the macrophages of brain :* **Gaurisankar Sa**, Tanya Das (Immuno-technology section), Suman Pal and Tathagatha Chaudhury

Microglia are a stable population of highly ramified or dendriform cells of bone marrow origin that are generally accepted to represent the

resident macrophages of brain, owing to their many functional and phenotypic similarities. The transformation of microglia into intrinsic brain macrophages appear to be under strict control and takes place under different stresses including infection. The differentiation of microglia is accompanied by the release of several secretory products e.g. proteinase, cytokines, reactive oxygen intermediates and reactive nitrogen intermediates. Activation of microglia is a multistep pathway in which responsive microglia sequentially become primed and triggered by a variety of stimuli. We have found that one of the microglia activator is endothelin (ET). Our preliminary data showed that ET stimulates the secretion of various cytokines and nitric oxides from microglia.

C.3. *Molecular mechanisms of curcumin-induced tumour cell killing :* **Gaurisankar Sa**, Tanya Das (Immunotechnology section), Tathagatha Chaudhury and Suman Pal

Turmaric has been used as an anti-inflammatory agent in Ayurvedic Medicine from ancient time. Recently it has been published that the active component of turmaric has an anti-tumour property. Our laboratory is actively engaged to find out the mechanism(s) of anti-tumour activity of curcumin using Ehrlich's ascites tumour cells (EAC) in the peritoneal cavity of mice as a model. Our preliminary observation revealed that curcumin indeed reduces the tumour burden in the host. Further works are in progress.

PUBLICATIONS

1. Keshan B & Ray AK 1998 Action of estradiol-17beta on the synthetic activity of the silkgland in *Bombyx mori* L.,; J. Insect Physiol. **44** 491-498.

2. Keshan B & Ray AK 1998 Subcellular localisation of trehalase in different tissues of the silkworm *Bombyx mori* L.: Demonstration of the highest activity in mitochondria; Sericologia **38** 411-418.

3. Chaudhuri AB, Krishnan N, Sarkar PK, Sinha AK, Sinha SS & Ray Arun K 1998 Octopamine titer in the circulating fluid of tropical tasar silkworm, *Antheraea mylita* Drury (Lepidoptera; Saturnidae) and its response to injected estrogen during critical phase of diapause termination; Current Sience **74** 695-699.

4. Tiwary BK, Kirubagaran R & Ray AK 1998 Testosterone-mediated complete Feminization with precocious ovarian maturation in catfish, *Heteropneustes fossilis* (Bloch); Aquaculture Research **29** 333-336.

5. Sarkar PK & Ray Arun K 1998 Specific binding of L-triiodothyronine modulates Na^+-K^+-ATPase activity in adult rat cerebrocortical synaptosome; Neuroendocrinology **9** 1149-1152.

Participation in conferences/Symposia/ Training Programmes/Awards and Honours Received.

Prof. Arun K. Ray delivered lecture in the National Workshop on "Recent Advances in Hormonal Physiology of Fish and Shellfish Reproduction" during 16-18 April, 1988 at Bhubaneswar, attended Alexander von Humboldt Conference at Hyderabad during 20-22 November, 1988, acted as an external examiner for M.Sc. Zoology/Life Sciences in Calcutta University and Tripura University. Prof. Ray delivered lectures in Refresher's Course in Zoology and Physiology of Calcutta University.

Dr. Gaurisankar Sa made several deliberations in Society of Biological Chemist's Symposia, 1988; 10th International Congress of Immunology, 1988; Satellite Symposium .on Tumor Immunology of 10th International Congress of Immunology, 1988; The Symposium on Biotechnology for health, Agriculture & Environment, 1988; National Symposium on Recent Advances in Structure, Synthesis and Function of Biomolecules, 1999. Dr. Sa also acted as instructor in the Refreshers course in Zoology, Academic Staff College, University of Calcutta in 1998-99. He is also instructor for the pre- & post-doctoral course work in Bose Institute in 1998-99.

Dr. Anuradha Chaudhuri presented paper at National Symposium on Trends in Research on Hormones, Reproduction and Animal Productivity at University of Delhi, in October 10-12, 1988.

Ph. D. AWARDED

Name of student	University	Year	Title	Supervisor
Ms. Mitali Pramanik	Calcutta	1998	Role of thyroid hormone on the phenylhydrazine-induced haematological changes in sexually immature rats.	Prof. Arun K. Ray
Mr. Basant K. Tiwary	Calcutta	1999	An approach for genetic and endocrine manipulation for better production of catfish, *Heteropneustes fossilis*(Bloch)	Prof. Arun K. Ray

GRANT-IN-AID SCHEME

Investigator/ Co-Investigator	Title	Financed by
Dr. G. Sa	Intracellular signalling mechanisms that mediate Endothelin-induced transcription of immediate early Genes.	DST
Dr. G. Sa	Intracellular signalling mechanisms of basic fibroblast Growth factor-stimulated early gene transcription.	CSIR

RESEARCH SCIENTISTS/ RESEARCH ASSOCIATES/ POSTDOCTORAL FELLOWS/ SENIOR RESEARCH FELLOWS/ JUNIOR RESEARCH FELLOWS

Dr. Anuradha Chaudhury (upto 30.6.98), Dr. Sharmila Chatterjee, Mr. Manick Ch. Ghosh, Mr. Suman Pal and Mr. Tathagatha Chaudhury.

SUPPORTING STAFF MEMBERS

Dr. A.K. Dasmahapatra, Mr. I. Ray, Mr. S.K. Ghosh, Mr. A.K. Santra and Mr. N. C. Jana

Plant Molecular & Cellular Genetics

Faculty : Prof. A.N. Lahiri Majumder (In-Charge), Dr. Amita Pal, Dr. Sampa Das, Dr. S.R. Sikdar.

HIGHLIGHTS

Antiquity of the inositol synthase protein gene has been established. Identification and characterisation of insecticidal genes having binding affinity to specific insect mid-gut, for their expression in susceptible crop sps were made. Micropropagation of *Pelargonium X Citrosum* Van Leeni and *Clerodendron colebrookainum* walp Cl var. *denticulata* was successfully established.

INSTITUTIONAL PROJECT I

A.1. *Inositol metabolism in relation to salinity stress: Implications in the chloroplast functions :* **A.N. Lahiri Majumder,** Shilpi Ghosh, Nitai C. Hait, Sanjoy Guha Roy and Monoj Majee

Previous work from this laboratory demonstrated that salt-mediated increased synthesis of inositol during NaCl stress in the salt-tolerant varieties of rice *(Oryza sativa)* may turn out to be a mechanism through which chloroplastic and cytosolic Fructose-1, 6-bisphosphatase (Frup2ase) function may be protected during NaCl stress. For an in-depth study of this, purified chloroplastic Frup2ase from rice and *Porteresia* were subjected to NaCl treatment *in vitro* and the intrinsic tryptophan fluorescence monitored. Frup2ase from the salt-sensitive rice cultivars exhibited increased tryptophan fluorescence with increasing NaCl (50-400 mM) which is prevented by preincubation of the enzyme-protein in presence of various concentrations of inositol ranging from 20-100 mM. In contrast, Frup2ase from the halophytic *Porteresia* does not exhibit any change in the tryptophan fluoresecnce in presence of increasing NaCl (50-300 mM) when compared to that without any salt. Such results are comparable to those obtained from the *in vitro* enzyme-inhibition studies, adducing evidence that the Frup2ase(s) from cultivated sensitive and wild tolerant species of rice may be structurally different.

For further study in this direction, full length cDNA clones of *Oryza* and *Porteresia* cytosolic Fructose-1 ,6,bisphosphatase have been obtained by RT-PCR, based on sequence information of the rice gene. The PCR-products have been cloned inPGEM-Easy vector end sequenced for confirmation of the identity of the gene. Attempts are presently made to express the full length cDNAs of inositol synthase and for fructose-1,6-bisphosphatase from *Oryza* and *Porteresia* in suitable prokayotic system for elucidation of a probable structure-function relationship with respect to halotolerance.

A.2. *Development of insect resistant Brassica spp with potential plant lectin genes :* **Sampa Das,** Jayasree Banerjee, Anita Roy and Santanu Bandyopadhyay

Every year a wide range of economically important crops suffer from yield loss due to sucking type of insect attack of which aphid is the most damaging one. Few lectins have been isolated from different plant members of Alliaceae and Amaryllidaceae and found to be effective against aphids. After extracting and purifying through affinity chromatography insect bioassay have been carried out with different doses of lectin supplemented in artificial diet. Garlic leaf lectin have been proved to be more potent than bulb lectin.

Garlic bulb lectin coding genes have been isolated from garlic genome through PCR using gene specific primer designed from published sequences and cloned. Sequencing of the cloned gene is under process. Attempts are made to clone garlc leaf lectin gene.

So far there is no report on the mode of action of lectin to control the insect multiplication. Attempts have been made to isolate the Brush Border Membrane Vesicle of the lectin fed red cotton bug a model sucking

type of insect and through western analysis specific receptor protein have been identified capable of binding with garlic bulb lectin. In a parallel approach *in situ* localization of lectin bound insect gut receptor are also being documented, which suggest the possible mechanism of toxicity of lectin to control insect growth and development.

A.3. *Standardization of protocol for plant regeneration from leaf tissue end mesophyll protoplasts of Rorippa indica (L.) hiren :* **S. R. Sikdar** and Palas Mandal

High frequency plant regeneration from leaf tissue (40-50/leaf explant) and mesophyll protoplast derived calli (100%) of *Rorippa indica (L.)* hiren, a wild crucifer, has been achieved. Plant regeneration protocol from protoplast will help us in transferring Aphid tolerance and summer growing property of this wild crucifer in oil yielding *Brassica* cultivar through somatic hybridization.

Other Projects :

A.1. *Biochemical analysis of two cotyledons of Vigna radiata :* **Amita Pal** and Shamik Das (in collaboration with **Dibeyendu N. Sengupta**)

In continuation of our earlier work following investigations were made: Qualitative and quantitative changes in cotyledonary protein during cytokinin mediated adventitious meristem differentiation from the 1 st day after inoculation till the 12th day were studied. Experimental results showed rhythmic rise and fall of protein content at $\geq$ 24 hr interval and the amount of protein in cot was always higher than cot (e) from 5th to 8th day. During this period, induction of adventitious shoot differentiation was observed only from the proximal end of the cot (e) explant, this perhaps suggests participation of proteins in the shoot regeneration.

Further *in vitro* studies were conducted to understand the biochemical basis of differential behaviour of two cotyledon types. ^{35}S Methionine was used to study the pattern of *de novo* protein synthesis in the two explant types, both under the influence of one another and individually. It was observed that protein synthesis initiated within 6 hrs. in cot (e), under the influence of cytokinin, where as, no *de novo* protein synthesis was recorded even after 12 hrs in the cot explants, when grown without the influence of cot (e). The influence of cot (e) on cot in *de novo* protein synthesis needs further investigation. It is now confirmed that the cultural response of one of these explants mimics that of the other depending on the direction of the nutrient pool, which is regulated towards more number of explants. Hence, the cultural response of less number of explants always influence that of the more number of explants and suggests migration of some 'metabolite' or 'factor' governed by the nutrient pool.

Another attempt was made to study the development of embryo under *in vivo* condition. Different stages of zygote formation and cotyledon development are being studied and the days after fertilization are scored. These information are essential to study the endoreduplication pattern of cotyledon at the different stages of embryo development and for the analysis of DNA polymerase.

A.2. *Nuclei isolation from the cotyledonary explants of Vigna radiata :* **Amita Pal** and Shamik Das

A method was standarized to isolate intact nuclei mechanically from the cotyledonary explants of *Vigna radiata*. This will enable us to study the endoreduplication pattern of cot and cot (e) explants in future.

B.1. *Micropropagation of Bamboo :* **Amita Pal, Samir Ranjan Sikdar** and Pinaki Chowdhury

Method for *in vitro* root induction from the basal end of the culture regenerated *Dendrocalamus strictus was* perfected. The plantlets also developed basal microrhizomes under *in vitro* condition. *Bambusa balcooa*, another very important species, has multipurpose uses such as, building of huts, boat making, scaffolding. The fibers of this bamboo constitutes a rich raw material for the paper and pulp industry and in charcoal preparation. This particular bamboo species is almost in endangered condition due to overexploitation by the users and failure of maintenance of the plantation by conventional clonal propagation. Recently, success has been attained to multiply

nodal meristems of field grown *Bambusa balcooa*.

B.2. *Micropropagation of aromatic and medicinal plants :* **S. R. Sikdar** and Krittika Bhattacharya

Micropropagation of *Pelargonium X Citrosum* Van Leeni, an aromatic plant and *Clerodendron colebrookainum* Walp Cl var. *denticulata*, a medicinal plant have successfully been carried out. The micropropagated plants were rooted and approx. 500 plants of *Pelargonium* and 1,500 plants of *Clerodendron* have been successfully transferred for demonstration to the nursery plot at Madhyamgram Experimental Farm of Bose Institute. *Pelargonium X citrosum is* claimed to be a mosquito repellent plant and leaves of *C. colebrookianum* contain blood pressure lowering principle.

PUBLICATION

1. Sikdar SR, Serino G, Chaudhuri S & Maliga P 1998 Plastid transformation in *Arabidopsis thaliana;* Plant Cell Rep **18** 20-24.

Participation in conferences/Symposia/ Training Programmes/Honours and Award Received

Prof. A.N. Lahiri Majumder has been awarded the Foundation Day Award of the Bose Institute; worked as a Rockefeller Foundation Biotechnology Career Fellow in the Dept. of Biochemistry University of Arizona, Tucson between May to July, 1998; participated on invitation & presented paper in the Gordon Research conference on "Salt and water-stress in plants" in Oxford, U.K. during August 16-21, 1998 ; participated in the Plant Biology '98 meeting organized by the American Society of Plant Physiologists at Madison, Wisconsin during June 27 to July 1, 1998.

Dr. Amita Pal delivered an invited talk on "Improvement of bioactive compounds through *in vitro* culture", in the National Conference on Recent Trends in Spices and Medicinal Plant Research, held at Calcutta on 2nd to 4th April, 1998. She has served as guest faculty to teach topics of Genetic Engineering for the final year M.Sc. Biotechnology students at the Centre of Biotechnology, Nagarjuna University, A.P. She has delivered invited lectures on "Geneitc Engineering Principles and Application" and on "Genetic transformation of plants with special reference to *Vigna radiata*", in the Zoology and Botany Department, Nagarjuna University, respectively. Dr. Amtia Pal has imparted training on various tissue culture methods to one M.Sc. Biotechnology student for three months. American Biographical Institute has selected the biography of Dr. Pal in the International Directory of distinguished Leadership for the year 1998-1999.

Dr. S. R. Sikdar delivered two lectures one on "Protoplast fusion as a technique for crop imporvement" and the other on "Transgenic and Transplastomic approaches towards crop improvement" in the Refresher course organised by Sambalpur University, Sambalpur, Orissa on 6th and 7th November, 1998; served as paper seter for M.Sc. Biotechnology paper Examination of Life Science, Sambalpur University; has been awarded Rockefeler Foundation Fellowship for three months for training on rice transormation at IRRI, Manila, Philippines.

GRANT-IN-AID SCHEMES

Investigator/ Co-Investigator	Title	Financed by
Prof.A.N. Lahiri Majumder/ Prof. Bharati Ghosh	Role of inositol and polyamine metabolism in relation to osmotic stress in rice (*Oryza sativa)*	Indo-US
Prof.A.N. Lahiri Majumder/ Dr. S.R. Sikdar	Metabolic engineering of inositol pathway in rice: Implications in the chloroplast functions.	DBT (CPMB Phase II)
Prof. A.N. Lahiri Majumder	Plant Fructose-1,6-bis-phosphatase(s) : Regulation and Expression during salinity stress in rice.	CSIR
Dr.Amita Pal/ Dr. S.R.. Sikdar	Generation and Demonstration of Tissue Culture raised Planting Material of *Dendrocalamus strictus* and *Bambusa tulda.*	DBT/ WBSCST

Ph.D. AWARDED

Name	University	Year	Title	Guide
Sriparna Bagchi	Jadavpur University	1999	Study of Fructose-1,6 bis-phophatase in rice with special reference to salinity stress.	Prof.A.N. Lahiri Majumder

RESEARCH SCIENTISTS/RESEARCH ASSOCIATES/POST DOCTORAL FELLOWS/ SENIOR RESEARCH FELLOWS/JUNIOR RESEARCH FELLOWS

Dr. Krittika Bhattarcharyya, Dr. Pinaki Chowdhury, Dr. Jayasree Banerjee, Dr. Shilpi Ghosh, Dr. Amitava Roy, Santi Ranjan Dey, Anita Roy, Nitai C. Hait, Shamik Das, Tathagata Roychoudhury, Palash Mondal, Sanjoy Guha Roy, Anirban Chatterjee, Mitu De, Debabani Chakraborty, Sudarsana Biswas, Santanu Bandapadhyay and Manoj Majee.

SUPPORTING STAFF MEMBERS

Puspita Mukherjee, Ashim Kumar Nath, Bijoy Kumar Mondal, Motilal Singh, Pravash Basu Roy, Ganesh Chandra Dey, Arup Kumar Dey, Sk. Eklas Ali, Amar Nath Hela and Subal Basak.

Immunotechnology Section

Faculty: Prof. P. K. Ray, Dr. Nripendranath Mandal

HIGHLIGHTS

It has been established that Protein A induced tumor apoptosis is via the immunomodulatory circuit of the host. The molecular mechanisms underlying the antitumor effect of Protein A have also been delineated. In order to develop a more potent and risk-free therapeutic method, works are in progress to study the functional mimicry of Protein A by two proteolytically cleaved fragments.

INSTITUTIONAL PROJECT V

A.1. *Protein A-Induced Ehrlich's Ascites Tumor Cell Killing is via Immunomodulatory Circuit :* **Prasanta K. Ray**, Tanya Das and **Gaurisankar Sa**

Protein A (PA) of *Staphylococcus aureus* is a potent immunomodulatory as well as antitumor agent. This investigation was undertaken to find out the correlation between the immunomodulatory and antitumor effects of PA. Flowcytometric analysis of cell cycle pattern of Ehrlich's ascites tumor cells (EAs) revealed that the effects of this protein were associated with an increase in the number of EACs in the G0/G1cell cycle phase. The existence of an aneuploid peak suggests that PA at the dose used (1 µg/20 g mice) causes apoptosis in the tumor cell population. Further studies indicated that the levels of p53, bax, and c-myc proteins were increased and caspase-3 was activated in EAC as a result of PA treatment. However when PA was directly added to EAC culture, it failed to cause any apoptosis. Interestingly, when EAC were co-cultured with PA-treated splenic lymphocytes, EAC apoptosis was again observed indicating a role of PA-stimulated splenocytes in EAC apoptosis. Identification of cell cycle phase as affected by PA showed that cells of all phases were undergoing apoptosis (Fig. I). PA could increase the level of tumor necrosis factor-α and nitric oxide in the serum of EAC bearing mice. These results led us to conclude that PA is inducing the release of different apoptogenic factors from splenocytes which in turn initiate apoptosis in EAC. These findings may be of great help in designing immunotherapeutic measures for cancer treatment.

A.2. *Molecular Mechanisms of Immunopotentiation by Protein A superantigen. Transduction of Pro-Proliferative Signals Depending on the Dose of the Inducer :* **Prasanta K. Ray**, Tanya Das and **Gaurisankar Sa**

Superantigen Protein A (PA) of *S. aureus* is known as an immunomodulator. In search of the molecular mechanism(s) of PA-induced immunocyte potentiation, we found that PA can induce proliferative signals involving tyrosine kinase-PLC-γ1-PKC-MAPK-SRF/NFkB in mice splenic lymphocytes. The cell cycle analysis showed the shift of cells from G0/G1 to S and G2/M phases indicating the mitogenic effect of PA in these cells. The concentration of pro-proliferative protein bcl-2 was increased significantly and c-myc level also showed an increasing trend as a result of PA treatment.

Since cell proliferation and apoptosis are two related phenomena that maintain the balance between cell growth and cell death, we were interested to find out the switch, if any, that changes the action of PA. Our studies revealed a dose-dependent binding of PA (0.01 to 100 µg/ml PA) to the mice splenic lymphocytes. Interestingly, at lower dose of PA (1 µg/20 g mice), the concentrations of pro-apoptotic proteins, e.g., p53 and bax were very low and remained unaltered as compared with untreated one. But at higher dose (10 µg/20 g mice), concentrations of these apoptogenic proteins increased significantly whereas bcl-2 level decreased. C-myc concentration was also increased significantly at this dose. This is quite expected since it is known that depending on the concentration, c-myc can act both as pro-proliferative and pro-apoptotic agent. Cell cycle analysis revealed that at this dose, PA induced growth arrest and apoptosis in these cells as concluded from the decrease in the cell numbers of S and G2/M phases as well as from the appearance of an aneuploid peak (<2n DNA). All these findings indicated that depending on the dose of the inducer, the cross talk between these pro and anti-apoptotic/proliferative factors changes, which ultimately acts as a switch deciding whether a cell would follow proliferative or apoptotic pathway.

A.3. *An Approach to Increase the Immunogenicity of any Antigen to Increase the Efficiecy of Immunisation Process :* **Prasanta K. Ray**, Tanya Das and **Gaurisankar Sa**

Specific neutralizing antibodies play a majore role in protection of a host from primary and secondary infections. However, some antigens induce life-time protection against infection, with antibody titer high and persistent but others induce and maintain antibody titer very poorly. An improvement in the immunogenic response of an antigen may be achieved by properly manipulating the molecular and/or cellular events that govern the immune response. Since Protein A (PA) is known as an immunostimulator, we aimed at studying whether PA can increase the antibody producing efficiency against sheep red blood cells. When compared with untreated mice, antibody titer increased by 8 folds. The stimulation of the antibody producing ability of the host by PA was substantiated by the activation of NFkB, the transcription factor responsible for immunoglobulin gene expression. Knowledge gathered from these observations might be of immense help to use such strategy in increasing the efficiency of immunization. Further studies are in progress.

A.4. *Protein A Induces Differential Stress Responses in Ehrlich's Ascites Cells and Splenocytes of Tumor Bearing Mice :* **Prasanta K. Ray,** Saubhik Sengupta, Tanya Das and **Gaurisankar Sa**

Host bearing malignant tissue is regularly subjected to stress because of the anatomical and physiological compromises that the host has to make to accommodate for the growth of malignant cells. On the other hand the malignant cells continuously run the risk of getting killed due to the attacks of primarily the immune system. During this one to one combat between the host factors and the tumor cells, the heat shock proteins (hsps) are expressed in both tumor and host cells which play a significant role in cell survival. Therapeutic approaches towards cancer by boosting the immune system by way of immunomodulators like Protein A tune up the stress response of the immune cells of the host. In our study we demonstrated that *in vivo* when Protein A was injected in mouse, differential response in the levels of hsp70 and 84 was observed both in the immunocytes and the tumor cells. We hypothesize that a decrease in protective proteins in the tumor defense helps the lymphocyte system to cause attack on tumor cells. Further studies are in progress to demonstrate the involvement of hsps in PA-induced tumor killing.

A.5. *Development of an Enzyme-Linked Immunosorbent Assay for Organochlorine Pesticides :* **Prasanta K. Ray**, Amiya Maity, Amiya Kr. Ghosh, Tanya Das and Tapan Mondal

Pesticides, e.g., Organochlorine, Organophosphates and Carbamates, are highly effective so far as the improvement in agriculture is concerned. However, all these compounds have various toxic effects on human. It is therefore of utmost necessity to detect the presence of these toxic chemicals in different food stuff. Aim of this project was to develop an ELISA kit for detection of minute amount of γ-HCH, an organochlorine pesticide. For production of polyclonal antibody to this small molecular weight compound, we first prepared the γ-HCH-BSA conjugate. The purity was checked by capillary electrophoresis. Polyclonal antibody against this conjugate was raised in rabbits and immunosorbent assay method (indirect ELISA) was developed using this polyclonal antibody. This ELISA kit can be useful in the field for the detection of γ-HCH. Further studies are in progress to develop monoclonal antibody against γ-HCH.

A.6. *A Combinatorial Approach Towards Aids-Management :* **Prasanta K. Ray**, Amiya Kr. Ghosh, Tanya Das and **Gaurisankar Sa**

Zidovudine, the FDA approved drug, is widely used in the treatment of HIV infected

patients, but hematopoietic toxicity caused by this drug has jeopardized the clinical management of AIDS. Studies have been conducted with AZT which directly affects different progenitor cells (CFU-E), (CFU-GM) of hematopoietic system to cause anemica, leucopenia, reduction of macrophages etc. The bone marrow cells are affected because of the toxic assualts of AZT and or its metabolites, but soon gets regenerated if PA is given prior to or along with AZT. From FACS-analysis of splenocytes, it has been found that AZT causes some reduction in the number of $CD4^+$ cells whereas PA increases the % of $CD4^+$ cell (about 77%), and also increase the number of $CD8^+$ cell (about 23%). It is known that HIV infection causes a severe reduction in $CD4^+$ cells. But PA treatment appears not only to replenish the bone marrow cells which are depleted by AZT, but it also increases $CD4^+$ cells significantly. This information might help developing strategies as to how AIDS therapy involving AZT can be made more effective.

A.7. *Protein A-Induced Activation of Anti-tumor Responses : Involvement of Nitric Oxide :* **Prasanta K. Ray**, Sreya Chattopadhyay, Tanya Das and **Gaurisankar Sa**

Protein A (PA) of *Staphylococcus aureus* has been shown to reduce tumor burden and simultaneously to stimulate the intrinsic defense machinery of the host, particularly the immunological defense system. The role of Nitric Oxide (NO) in the immunological defense mechanism and in apoptotic signaling cascade has been well documented. We wanted to find out whether NO is an effective mediator in the pathway of Protein A-induced tumor killing. To this effect, our data reveal that small non-toxic doses of PA (i.p and i.v injections) increase the NO levels in the serum of Swiss albino mice (both normal and tumor bearing) from its basal level. PA treatment *in vitro* of mouse peritoneal macrophages in culture also increases NO levels significantly. PA was also shown to increase the cytotoxicity of the macrophages against tumor targets in *in vitro* conditions. This cytotoxicity was abolished when the macrophages were pre-treated with NMMA (N^{ω}-monomethyl L-arginine), which indicates a positive co-relation between NO release and PA induced target toxicity.

A8. *Immunomodulation : A Plausible Mechanism for Protein A-Induced Ehrlich's Ascites Tumor Cell Killing :* **Prasanta K. Ray** and Pratima Sinha

Cell mediated and humoral immunities are the two arms of acquired immune response. The type of immune response generated is a function of the cytokines produced during exposure to antigens. Production of type 1 cytokines (IL2, IFNγ and TNF) leads to cell mediated immunity, whereas type 2 cytokines (IL4, IL5, IL6, IL10 and IL13) are associated with antibody production. The biological response of this dichotomy of T cell responses in tumor-bearing animals have only been described recently. Antitumor response of Protein A of *Staphylococcus* aureus is well documented. Most of the workers suggested that the antitumor response was immunologically mediated, there is only one report demonstrating activation of NK cells and also spleenic lymphocyte mediated killing of tumor targets. Directed evidences of antitumor response by PA in tumor bearers, with special reference to Th1 or Th2 type of cytokines, had not been clearly demonstrated by any worker so far. The objective of the present investigation was to understand the immunological mechanism of antitumor response of Protein A in Ehrlich's Ascites (EAC) tumor bearing swiss albino mice. We observed that IFNγ, IL2, TNFα, IL1β and IL1α increased after treatment with Protein A whereas IL4 and IL6 decreased after the treatment as compared to EAC control mice. IFNγ to IL4 ratio clearly indicates TH1 type of response in EAC tumor bearing host after treatment with PA .

A.9. *A 16-MER peptide Derived from Chymotryptic Cleavage of Staphylococcus aureus Protein A :* **Prasanta K. Ray,** Pratima Sinha and Jayati Sengupta

Protein A (PA) of *Staphylococcus aureus* has various array of biological functions such as antitumor, antitoxic, anticarcinogenic, immunomodulatory, antifungal and antiparasitic. Our hypothesis is that PA, being a foreign protein, may be subjected to proteolytic cleavage in vivo. It is thus possible that such degraded products might be functionally operative *in vivo*. Our previous Molecular modeling studies

qualitatively showed that two peptides of PA, 20-mer and 16-mer, may retain Fc-binding ability from interaction energy point of view. In the present study, we have selected 16-mer peptide derived from chymotrypsin digestion (theoretical) of B-domain among all five IgG binding homologous domains of protein-A. Our aim was to understand whether this theoretically predicted 16-mer chymotryptic fragment could be formed in real experiment and also to understand biological activities of this peptide. Chymotrypsin cleavage of PA at 37°C for 24h produced four fragments on reverse phase HPLC. The aminoacid analysis of each fragment shows absence of cysteine residue from all the fragments, which justifies absence of cysteine in PA. We also observed high content of aspartic acid and glutamic acid residues in all the fragments. On gel filtration chromatography the chymotrypsin cleavage of PA showed five peaks, one of which overlaps with our theoreticaly selected 16-mer peptide on superimposition. We verified the IgG binding capacity of 16-mer peptide by capillary electrophoresis. We also observed that 16-mer peptide induces production of TNFα and IL1α in mice serum. The peptide also activates macrophages causing *in vitro* EAC tumor cell cytotoxicity. Above observations suggests that 16-mer peptide may be obtained by chymotrypsin cleavage and also that this peptide has some of the biological properties of PA, such as IgG binding, TNFα and IL1α elicitation in mice serum and *in vitro* EAC tumor cell cytotoxicity.

A.10 . *A Minimized FC binding Peptide from Protein A Induces Immunocyte Proliferation and Evokes TH1-Response in mice :* **Prasanta K. Ray**, Pratima Sinha And Jayati Sengupta

It is now well established that PA is a potent biological response modifier, showing simultaneously antitumor, antitoxic, anticarcinogenic, antifungal, antiparasitic and immunomodulatory properties. Since PA is a foreign protein, it is quite logical to assume that it may be cleaved into smaller peptide fragments in vivo which may be responsible for biological activities of whole PA molecule. The present study was undertaken to dissect out the structural entities of PA responsible for its biological properties. Protein A (PA) of Staphylococcus aureus has a unique property of binding with immunoglobulins. On the basis of molecular modeling and energy minimization studies a 20-mer tryptic fragment (theoretical) was predicted to retain IgG binding capacity which has been verified by immunoblot. This peptide sequence was selected to carry out experimental studies to show its functional mimicry of PA. We observed in the sera of 20-mer peptide treated mice that the concentrations of IFNγ, TNFα and IL1α increase to a peak level by 4h; on the other hand, there was a decrease in IL4, IL6 and IL10 concentrations at the same time (4h). The ratio of IFNγ to IL4 showed Th1 type of response wit h the peptide as well as with that of PA. The nitric oxide concentration in sera also increases and the peak increase was in 6h with both the peptide and PA. Cell cycle analysis using FACS shows that 20μ g dose of peptide was non-toxic to thymocytes and spleenocytes; on the other hand, it was immunoproliferative, shifting the thymocytes and spleenocytes from G0/G1 to S phase of the cell cycle. Further studies are in progress to evaluate other biological properties of the peptide, to evaluate if this peptide could be used as a substitute of PA to mimic at least some of its biological activities

A.11. *Antitumor Property of A 20-mer Peptide : Derived from Tryptic Cleavage of Staphylococcus Aureus Protein A by Shifting Tumor Specific T-Cell Respose form type 2 to Type 1 Cytokine Profile :* **Prasanta K. Ray** and Pratima Sinha

Protein A from *Staphylococcus aureus* (PA) has a wide variety of immunological properties, such as the stimulation of lymphocyte subpopulations, the release of interferon, TNFα, IL1α and natural killer activity. In general, exogenous antigens require processing by antigen presenting cells (APC) before their association with MHC proteins to from a compex that can be recognized by specific T cell receptors. T cells do not recognize native protein antigens, unless previously denatured or partially degraded, and subsequently displayed in association with MHC class II molecules by APC. We speculated that short synthetic peptides derived from PA might have silmilar immunological properties to those exhibited by

the native protein. We have earlier reported that 20-mer peptide from PA mimics immunomodulatory behavior of PA. In this report our aim is to verify whether 20-mer peptide fragment is formed in reality by trypsin cleavage and also as this peptide increases IFNγ, TNFα and IL1α we decided to study in vitro cytotoxic properties of this peptide as well as antitumor activity. Trypsin cleavage of PA at 37^{0}C for 24h produced nine fraggments on reverse phase HPLC. The aminoacid analysis of each fragment shows absence of cysteine residue from all the fragments, which justifies absence of cysteine in PA. On gel filtration chromatography the trypsin cleavage of PA showed four peaks, one of which overlaps with our theoretically selected 20-mer peptide on superimposition. Antitumor response of Protein A of *Staphylococcus aureus* is well documented. We wanted to understand whether the 20-mer tryptic peptide of PA, identified by us have the antitumor property and if yes, the mechanism behind the effect. We observed that the 20-mer peptide when injected to EAC tumor bearing host increases the survival mice to 40 days (experiment was terminated at 40th day) as compared to the EAC controls, where all the mice were found dead after 24 days. We also investigated the profile of the cytokines to understand the antitumor effect of the peptide and found that the peptide increases the level of IFNγ, TNFα and IL2 whereas IL4 and IL6 was downregulated. The ratio of IFNγ to IL4 clearly indicates TH1 response in mice. Thus this 20-mer peptide was found to have antitumor property by shifiting T cell response to TH1 type as that of Protein A. We also observed that 20-mer peptide activates macrophages and induces *in vitro* cytotoxicity against EAC tumor. Above observations suggests that 20-mer peptide may be obtained by trypsin cleavage and also that this peptide has some of the biological properties of PA, such as *in vivo* antitumor effect by skewing T cell response to Th1 type as well as *in vitro* cytotoxicity against EAC tumor.

A.12. *Cloning and expression of protein A gene into mammalian system :* **Nripendranath Mandal,** Srikanta Jana and **Prasanta K. Ray**

Protein A is a cell wall protein of staphylococcus aureus Cowan I. It has multifunctional properties such as antitumor, antitoxic, anticarcinogenic, immunomodulatory and mitogenic properties. Many laboratories previously used protein A to remove immune complexes from serum of cancer patients in an attempt to control the disease, it has also been used to ameliorate toxic effects of many drugs or chemicals. Nobody ever tried to use protein A gene as a therapeutic gene to express in mammalian system. Using suitable eukaryotic promoter, specially using an inducible promoter, protein A gene expression study will be conducted. In future this study will be extended to gene therapy in animal models. Apart from obtaining basic information about the gene expression of protein A into mammalian cell line, this study will focus attention to construct a suitable vector in the field of gene therapy.

At present we have cloned protein A gene into prokaryotic and eukaryotic expression vectors. More experiments are in progress to study the expression of protein A gene.

A.13. *Cloning and Characterisation of a novel protein purified from mice splenocyte and thymocyte :* **Nripendranath Mandal**, Srikanta Jana and Eliza Chakraborty

There are two arms of acquired immunity that have different sets of participants and different sets of purposes but with one common aim; to eliminate the antigen. Of these two arms of the acquired immune response. One set is mediated mainly by B-cells and circulating antibodies hence it is termed humoral immunity. The other set mediated by T cells, instead of synthesising antibodies, release various cytokines that affect other cells to mediate the others. Hence, this arm of the acquired immune response is termed cellular or cell mediated immunity.

Immunocompetent lymphocytes combine with the foreign antigen, or a portion of it, termed epitope, by virtue of their surface receptors. They are stimulated under appropriate conditions to proliferate and differentiate into clones of cells with the corresponding identical receptors to the particular portion of the antigen, termed antigenic determinant or epitope. As with B cells, each T cell bears on its surface a clonally distributed receptor that interacts with antigen. This is known as T-cell receptor (TcR).

Recently we have purified a single band protein (43kd) from mice splenocyte and thymocyte having affinity with protein A (shown in fig 2). This protein has different molecular weight than IgG as determined by Gel-electrophoresis. More experiments are in progress to study the molecular cloning and characterisation of these newly found proteins.

A.14. *Quantification of the cytokines and their receptors mRNA based on competitive PCR :* **Nripendranath Mandal**, Srikanta Jana and Eliza Chakraborty

The lymphokines or cytokines, the hormone-like growth factors and differentiation factors, provide a network of coordination for T cells, B cells and macrophages to get activated during immune responses. Cytokines mediated immune and inflammatory responses can be modulated at two stages; i.e., at the activation of Cytokine genes and at the effector phase involving Cytokine-receptor interaction. The structure and biology of cytokines, the regulation of Cytokine genes and their receptors, signal transduction and hematopoietic growth factors have been well studied during the last two decades.

Cytokine detection and quantification at the protein level is hampered by their low production and by the lack of monoclonal antibodies for each Cytokine or receptor subunit. In addition, the sensitivity of assays such as flowcytometry or various binding assays using receptor-specific antibodies or the ligand for the receptor itself is low. Consequently, Northern blot analysis, RNAse protection assays and mRNA phenotyping have been widely used for the study of short-lived, low-copy number mRNA transcripts. It has wide spread applications in genetic disease diagnosis, disease susceptibility and cancer diagnosis. However, even these techniques require at least 105-106 molecules per sample and also suffer from lack of sensitivity. Reverse transcription (RT)-PCR is an extremely powerful method for mRNA analysis. However, it can be difficult to obtain quantitative information with this technique.

Recently we are developing an assay system that will allow us to quantitate mRNA of cytokines at very low level (fg level). More experiments are in progress to construct a plasmid at the very early stage of this project.

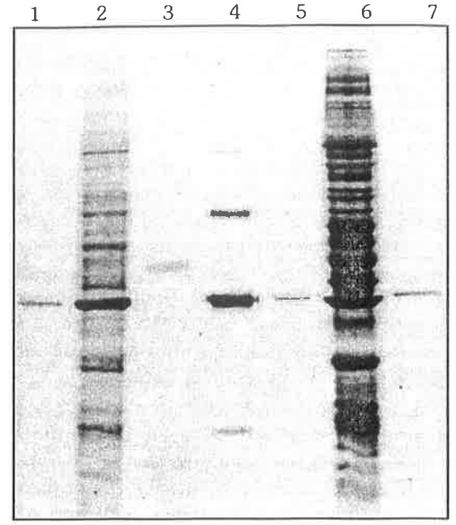

Fig.2. SDS PAGE gel (10%) electrophoresis of purified protein from thymocyte and splenocyte. The protein bands are visualised by Silver stain method. Lane1. Purified protein from splenocyte, Lane2. Crude cell lysate of splenocyte, Lane3. Mouse IgG, Lane4. Marker(medium range- Bangalore GENEI), Lane 5&7. Purified protein from Thymocyte, Lane 6. Crude cell lysate of Thymocyte.

INSTITUTIONAL PROJECT III

A1. *Solution Structure Determination of Short Synthetic Perptides having Sequences of Protein-A using CD and NMR Techniques :* **Prasanta K. Ray**, Jayati Sengupta and **Gautam Basu**

It is well established that short synthetic peptides are able to induce antibodies that recognize, with high frequency, the cognate sequence in the folded protein. This phenomenon is difficult to explain unless the peptide is able to adopt a highly preferred conformation, either free in solution or on the B-cell receptor, approaching that which it adopts in the native protein or else that protein itself approaches disorder through conformational fluctuations. Several short peptide sequences, each capable of inducing antibody that bind to the cognate sequence in the folded protein, exhibit conformational preferences for secondary structure (helix, turn) in aqueous solution. These experiments have led us to propose that highly

immunogenic peptides frequently have a high propensity to adopt folded structure.

One of the difficulties in studying peptides is the nature of the structural states. Most peptides do not exist as a single structure but as a conformational ensemble of random and structured molecules in equilibrium. Until recently the potential of NMR for conformational studies of flexible linear peptides has not been realized because of problems arising from averaging of NMR parameters over all populated conformations. Short linear peptides in water typically give 'random coil' NMR spectra, and evidence for nonrandom conformations could only be obtained by indirect methods.

In the present study, conformational preferences were investigated in synthetic peptide, 20-mer (as mentioned earlier) derived from Protein-A (B-domain), a three-helix bundle protein, using 1H NMR (1Dand 2D) and CD spectroscopy.

In summery, the structure of 20 residue peptide seems to be basically a flexible one in water with two low populated helical turn like structures (turn or nascent helix) at N- (residue 4-9) and C- (residue 16-18) terminal and the structure gets weak in the centre (near Proline).

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2. Sengupta J, Sinha P, Mukhopadhyay C, and Ray PK, 1999. Molecular modeling and experiment approaches toward designing minimalist protein having Fc-binding Activity of Staphylococcal Proten-A. Biochem. Biophy. Res. Comm. **256(1)**, 6-12.

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4. Ghosh AC, Mookerjee A, Sengupta K, Ghosh AK, Dasgupt S, and Ray PK, 1999. Therapeutic and prophylactiv uses of the protein A in the control of Leishmamia donovani infection in Experimental Animals. Immunol. Lett. **65**, 175-181.

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9. Subbulkshmi V, Ghosh AK, Das T, and Ray PK, 1999. Mechanism of Protein-A Induced amelioration of Toxicity of Anti-AIDS Drug, Zidovudine. Biochem. Biophys. Res.Comm. **250**, 15-21.

10. Ray PK, and Das T 1999. Molecular adaptation to toxic chemicals and drugs. Advances in Organ Biology (Das DK ed), Jai Press USA. **6**, 251-265.

Participation in Conferences/Symposia/ Training Programme/Awards and Honours received

Dr. Tanya Das has delivered seminars in 10th International Congress of Immunology (held in New Delhi 1998), Symposium on Biotechnology for Health Agriculture and Environment (held in Calcutta, 1998), Satelite Symposium on Tumor Immunology, 10th International Congress of Immunology, 10th International Congress of Immunology (held in Calcutta, 1998) and in National Symposium on Recent Advances in Structure, Synthesis and function of Biomolecules (held in Calcutta, 1999).

Dr Pratima Sinha presented poster in 10th International Congress of Immunology, held in New Delhi, 1998 and also in National Symposium on Recent Advances in Structure, Synthesis and Function of Biomolecules, held in Calcutta, 1999. Dr. Sinha participated in continuing education and workshop in Immunology, held in New Delhi, 1998.

Mr. Amiya Kr. Ghosh presented a paper in 10th International Congress of Immunology, held in New Delhi, 1998. Mr. Ghosh also participated in continuing education and workshop in Immunology, held in New Delhi, 1998.

GRANTS-IN-AID SCHEMES

Investigator/ Co-Investigator	Title	Financed by
Prof. P.K. Ray	Development of an Enzyme Linked Immunosorbent Assay for Organo-Chlorine Pesticides.	DBT
Prof. P.K. Ray	Development of an Immunotherapeutic Approach to Control Aflatoxin Mediated Death in Poultry	DST

RESEARCH SCIENTISTS/RESEARCH ASSOCIATES/POST-DOCTORAL FELLOWS/ SENIOR RESEARCH FELLOWS/JUNIOR RESEARCH FELLOWS

Dr. Tanya Das, Dr. Pratima Sinha, Dr. Jayati Sengupta, Dr. Elisa Chakraborty, Saubhik sengupta, Amiya Kr. Ghosh, Srikanta Jana, Sreya Chattopadhyay.

SUPPORTING STAFF MEMBERS

Uttam Kr. Ghosh, Tapan Mandal, Rina Das, Amartya Sen, Ranjit Das.

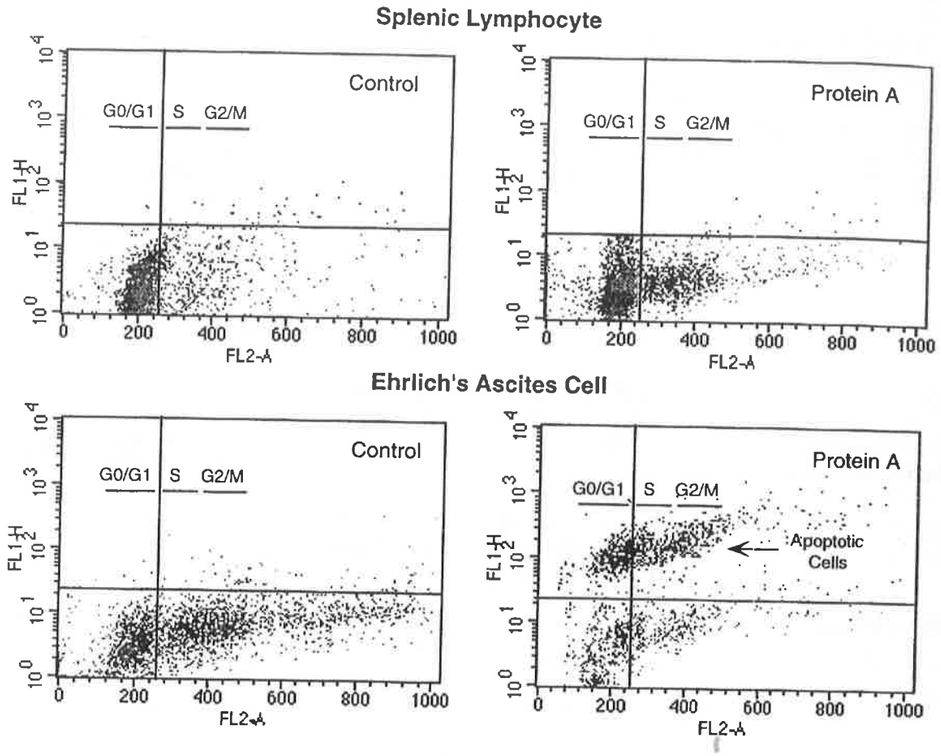

Fig. 1. Phases of cell-cycle at which Protein A induces apoptosis. Protein A-treated mice splenic cells or EAC's were tested for phase distribution of apoptotic cells using propidium iodide as well as Br-dUTP and terminal deoxynucleotyl transferase. Flowcytometric analysis showed the presence of doubly stained (apoptotic) cells at all the phases of cell-cycle in EAC of Protein A-treated mice.

Environmental Sciences Section

Faculty: Prof. P. K. Ray

HIGHLIGHTS

A microbial decontamination technology of organochlorine pesticides from oil and food stuffs has been patened. Protein A demonstrated the immunomodulatory activity in chicks. A water filter to decontaminate drinking water from arsenic has been developed.

INSTITUTIONAL PROJECT I

A.1 *Development of arsenic selective water filter for the decontamination of arsenic from drinking water :* **P. K. Ray,** D.P. Modak, Asis Dalal and Amiya K. Maiti

Arsenic has gained great notoriety historically for its toxic properties. The state of West Bengal has been suffering from arsenic contamination in drinking water for the past decade. Using oxides of various metals attempt have been made to adsorb arsenic from water. We have seen that the mixed oxides are more efficient to act as a scavenger than the individual oxides with kd values for arsenic 5.0 x 10^4. A pilot scale study showed that the filter can efficiently remove arsenic in a continuous dosing of 0.3 ppm of arsenic in water.

A.2. *A method of elimination of organochlonne pesticides from soil, water and food by using Bacterial Strains BI-100 and BI-102 :* **P.K Ray,** P.K. Chakrabartty, P. Bhattacharyya, D.P. Modak, J. Datta and Amiya K. Maiti

As we all know that γ-hexachlorocyclohexane (γ-HCH, lindane) which is an organochlorine pesticide has been widely used for both agricultural and public health purposes since long. However, there is always a tendency to use it in large excess. In spite of its long term toxicity and long persistence in soil, India and many tropical countries are still using HCH due to economic reasons resulting in continuous contamination of new sites by HCH. Since this pesticide is not biodegradable large amount of it accumulate in the environment and enter the food chain subsequently.

The object of our study was to remove HCH from soil by using the bacterial strains BI-100 and BI-102 isolated in our laboratory, thereby limiting the entry of HCH to food chain.

The bacterial strains were isolated from soils where gammexane is used periodically. These strains were listed for their ability to degrade γ-HCH that had been added to the mineral salt medium as the sole source of carbon. It has been observed that within 12h after inoculation of the bacterium BI-100 into the medium under aerobic conditions, γ-HCH disappeared completely while that occurred at 32h with BI-102. Similar results were observed when γ-HCH contaminated soil samples were treated with BI-100 and BI-102 individually. Immobilized BI-100 is a novel preparation to make the processed food stuffs, particularly liquids like water, milk, oil etc. free of γ-HCH. Interestingly, more than 90% of γ-HCH could be removed from contaminated liquid food stuff like water, milk and oil by passing them through a column packed with beads of calcium alginate immobilized bacteria.

A patent application has been filed.

A.3. *Search of immunomodulatoy compounds from exotic Indian plants :* **P.K. Ray,** M. Chakraborty, P.K. Debnath, D.P. Modak, J. Datta and Goutam K. Datta

For a long time many herbal products have been ascribed to have immunomodulatory activity in Ayurved. Some medicinal plants had been screened by various researchers, offer a significant potential in this direction.

In this study, we have isolated various fractions by solvent extraction from *Desmotrichem fimbriatum* and screened separately for their (i) Antimicrobial activities (ii) Immunopotentiating effect (iii) Antitumor effect and (iv) Antioxidant property.

We have observed antibacterial activity of the aqueous alcohol extract against *Escherichia coli, Bacillus subtilis, Staphylococcus aureus, Sarcina lutea, Mycobacterium smegmatis,* and *Vibrio cholerae,* and antifungal activity against *Aspergillus niger, Helminthus oryzae.* Besides, at a dose of 25 mg/kg b.w. of D. fimbriatum aqueous alcoholic extract indicated significant lucocytosis and neutrophilia in murine system. During the experiment of Ehrlich's ascites tumor cell both in vitro and in vivo, the drug showed remarkable decrease in tumor cells than the normal. In both cases, the observations are further supported by DNA cell cycle phase distribution studies against control.

Another study of aqueous alcohol extract of drug observed antioxidant activity during the psychophysical stress condition. The other studies are in progress.

A.4 *Development of an immunotherapeutic approach to control aflatoxin mediated death in poultry :* **P. K. Ray**, P. Bhattacharyya, D. Ghosal and S. Bid

Aflatoxin problem is considered to be crucially important in poultry industry in terms of its economic implication. The disease caused by it is aflatoxicosis. It was found that, the outbreak of disease was connected with the consumption of aflatoxin contaminated diet. Improper storage facility, use of low grade oil seeds and warm/humid climate facilitate growth of several toxin producing fungi and other microbes in poultry feed. Birds are dying in poultry farms in thousands because of this feed contaminant. In fact, once the outbreak occurs there is no specific means by virtue of which toxicity can be prevented. As aflatoxin intoxicated animals have responded positively to Protein A under different stressed conditions we are trying to extend the use of protein A to poultry immunopotentiation.

Our experimental observation reveals that Protein A has potential to protect Aflatoxin B_1 treated chicks against the general toxicity parameters. In smaller samples it seems that Protein A could protect reduction in body weight, increases RBC & WBC counts in comparison to that of AFB_1 treated chicks.

Another experiment is going on in which liver of AFB_1 treated birds were collected to test whether aflatoxin is accumulating in the liver of aflatoxin treated birds and also whether it is persisting there even after cooking so that there arises a chance of transfer of this fungal toxin indirectly to human being.

A.5. *Development of a database on pesticides :* **P.K. Ray,** D.P. Modak and Jayati Sengupta

With the advent of 'green revolution', an elevated level of different pesticides which are not easily biodegradable, are entering constantly into different environmental compartment in many ways and are persistent in nature. We have developed a database covering all pertinent informations related pesticides especially focusing attention on food contamination and toxicity.

Data were collected (for 145 organochlorine, organophoshorus and carbamate group of pesticides) mainly through existing Database (Agricola, Toxline Plus etc.) search. The above mentioned Data base is made using FOXBASE package named-"DATAPEST". It is menu-driven and user's familiar.

A.6. *Development of an enzyme-linked immnosorbent assay for organochlorine pesticides :* **P.K Ray,** A.K. Maity, A.K. Ghosh, T. Das and T. Mondal

Pesticides, e.g., Organochlorine, Organophosphates and Carbamates are highly effective so far as the improvement in agriculture is concerned. However, all these compounds have various toxic effects on human. It is therefore of utmost necessity to detect the presence of these toxic chemicals in different food stuff. Aim of this project was to develop an ELISA kit for detection of minute amount of γ-HCH, an organochlorine pesticide. For production of polyclonal antibody to this small molecular weight compound, we first prepared the γ-HCH-BSA conjugate. The purity was checked by capillary electrophoresis. Polyclonal antibody against these conjugate was raised in rabbits and immunosorbent assay method (indirect ELISA) was developed using these polyclonal antibody. This ELISA kit can be useful in the field for the detection of γ-HCH. Further studies are in progress to develop monoclonal antibody against γ-HCH.

A.7. *Societal Programme :* Besides this, Environmental Sciences Section has carried out consultancy work regarding Quality Control, Monitoring and Control of Environmental Pollution in local and national level. In this connection this year our department has audited ninety two foundries at Howrah and Calcutta, worked at Santaldih Thermal Power Station, Purulia and Bandel Thermal Power Station, Hooghly. We have also characterised and evaluated the possible hazard analysis at Dasnagar, Howrah Gas Tragedy sites as per order of the Hon'ble High Court.

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2. Datta J, Ghosh S, Maiti AK, Das SK, Modak DP, Bhattacharyya P, Ghosh A, Mondal NC, Chakroborty PK and Ray PK 1998 Biodegradation of organochlorine Pesticides by soil bacteria. J. Sci. Ind. Res. P-838.

GRANTS-IN-AID SCHEME

Investigator/ Co-Investigator	Title	Financed by
Prof. P.K. Ray	Development of an Immunotherapeutic approach to control aflatoxin mediated death in Poultry.	DST(Govt. of West Bengal)
Prof. P.K. Ray	Environmental Monitoring and control of air pollution of Santaldih Thermal Power Station.	WBSEB, (Govt. of West Bengal)
Prof. P.K. Ray	Environmental Monitoring and control of air pollution of Bandel Thermal Power Station.	WBSEB, (Govt. of West Bengal)
Consultancy projects from various industries .		Amount earned is Rs. 8,33,809.00 Lakhs

RESEARCH SCIENTISTS/RESEARCH ASSOCIATES POST-DOCTORAL FELLOWS/ SENIOR RESEARCH FELLOWS/JUNIOR RESEARCH FELLOWS

Dr. D.P. Modak, Dr. Jharna Datta, Mr. Amiya K. Maiti, Mr. S. Sengupta, Dr. G.K. Datta, Ms. S. Bid, Mrs. S. Chatterjee

SUPPORTING STAFF MEMBERS

Mr. A. Adak, Mr. A. Dalal, Mr. S.C Jana, Mr. S. Roy and Mr. S. Mondal

Madhyamgram Experimental Farm

Prof. Swati Sen-Mandi **(Scientist-in-Charge)**

Scientist-Farmers Meet : Technology Transfer Workshop cum Flower Show was held at Bose Institute Experiemental Farm, Madhyamgram on 15th and 16th Jan. 1999. Experts from Dept. of Agriculture, Calcutta University Dept. of Agricultural Chemistry and Soll Science, Bidhan Chandra Krishi Viswavidyalaya, National Bureau of Soil Survey and Land Use Planning (ICAR), Dy. Director, AgriHorticultural Society of India, Dept. of Botany and Section of Plant Molecular and Cellular Genetics, Bose Institute were resource persons who spoke and discussed on various aspects of Agriculture, Floriculture, Soil and Water Management, Hybrid rice seed production and Micropropagation of cash crops. More than 50 local farmers attended the 2-day-workshop under the supervision of Principal Agricultural Officer, and Sub-Divisional Agricultural Officer, Barasat, Dept. of Agriculture, Govt. of West Bengal. Participating farmers were awarded certificates.

SUPPORTING STAFF MEMBERS

Sri Pulak Kr. Roy, Sri Kachiram Kole, Sri Bimal Ch. Modak, Sri Basudev Jana, Sri Amal Krishna Das, Sri Rakhal Ch. Jana, Sri Dilip Pramanick, Sri Naliniranjan Malik, Sri Cittaranjan Mallick, Sri Subhas Senapati, Sri Sabita Baisya, Sri Sukumar Das, Sri Razkishor Lenka, Sri Rabin Talukdar, Sri Jagobandhu Nayak, Sri Laxmikanta Pradhan, Sri Nodiram Kayal, Sri Tapas Chakraborty, Sk. Inal Ali, Sri Sukanta Chakraborty, Sri Bhanu Kisku, Sri Mahesh Das Gupta, Sri Subrato Banerjee.

Library

Ms R. Mitra **(Librarian, Acting)**

The Library primarily serves the scholars, faculty members and staff of the institute. Researchers and scientists of other institutes can be members of the library with an annual membership fee. The Library has two wings with 14 staff members. The collection consists of both biological and physical sciences. Library subscribes 91 journal titles, both foreign and Indian. Internet facilities are available for internal use only.

Acquisition

Library contains 32,462 volumes of books, journals. Library purchase 38 books in 1998-99. 25 theses have been deposited during that period.

Photocopying services

Library provides photocopying facilities to institute as well as outside users on payment basis. 1,55,656 (RPG/RICOH) copies and 19,483 (Modi) copies in the Centenary Building Library and 59,067 copies in the Main Campus Library were produced during the period. Under Inter-Library loan system, photocopies were also supplied to INSDOC.

PUBLICATIONS

1. Chatterjee Chinmoy and Sarkar Pradip 1998. Abuse online on the net. Proc. XVIII National Seminar of IASLIC, 116.

2. Mitra Rukmini and Dutta Namita 1998 Right to information : role of libraries and information centers. Proc. XVIII National Seminar of IASLIC. 114.

Participation in Conferences/Symposia/ Training Programme/Awards and Honours received.

Pradip Kumar Sarkar participated in the XVIII National Seminar of IASLIC held at Trichur University, Trissur, from 21-24 December, 1998.

Rukmini Mitra participated in the National Annual Convention on Library and Information Networking (NACLIN98) held at Delnet, New Delhi from 16-18 November, 1998.

Rukmini Mitra delivered an invited talk on "Fulbright at Fifty : a review for 2000 A.D. and beyond" at USIS, Calcutta on September 12, 1998 ; delivered an invited lecture on "Research Library" in the Librarians Day, organised jointly by IASLIC, BLA and Jadavpur University on 9 August, 1998.

SUPPORTING STAFF MEMBERS

R. Bhattacharya, P.G. Das, K.P. Hait, S.K. Pal, P.K. Sarkar, B.S. Chaudhuri, G. Biswas, A. Kolay, G. Mukherjee, S. Bhuiyan, M. Jogsharma, A.K. Nandi, D. Das, D. Turi.

Workshop

Mr. A. Mukherjee **(Superintendent)**

The Workshop consists of the following units e.g. i) Mechanical ii) Electrical (both campuses) iii) Refrigeration (both campuses) iv) Glass blowing v) Carpentry vi) Drafting vii) Electronic & viii) Store. During the year under report Workshop received about 2000 requisitions from various departments of the Institute.

Mechanical section received about 200 job requisitions during the year 1998-99. Most of them are completed and some are on process. The Mechanical unit designed and fabricated various equipments as per requirement of the Scientists. Among the jobs undertaken by the Workshop are i) Perspex door for Laminarflow apparatus ii) Water circular gasket for P.D. Lab. iii) Gel Electrophoresis iv) Camera stand for Microscopic photography v) Stereo glass viewer for detecting structure formation. Repairing of vacuum pump, Aircompressor, Water pump for Madhyagram Farm etc. and also spare for the aforesaid job have been done. One Troly has been fabricated and handed over to the Biochem Dept.

The revenue Rs. 24,000/- has been earned by this time towards the fabrication and supply of two Crescographs to BITM during this period.

Electrical section for both campuses received about 1200 nos of jobs during 1998-99. Electrical section undertook various types of electrical instruments for repairing like voltage stabiliser, Water bath, Shakers, Autoclaves, Megnatic stirrer. Repairing of Motor for pump and other motors including rewinding and heat varnishing, repairing of fans have also been done. Moreover, Electrical section attended about 200 seminars and lectures. This section undertook the responsibility of Audio and lighting system during the symposium held in the Institute and also undertook to render services for seminar/symposium organised by outside agencies against payment basis. The Preliminary Energy Audit has been completed successfully. A stock of total no of Airconditioners, Deep freezes, Autoclaves, Ovens etc has been taken for monitoring power load of the Institute.

Besides service contract, Refrigeration unit having 1(one) Mechanic in each campus maintained some 0^0 Cold Rooms and A.C. machines of both campuses. Some welding works have also been done by the refrigeration mechanic and received about 150 requisitions and completed all the jobs.

Glass blowing section having one Glass Blower received about 200 nos of requisitions and completed all the jobs. Glass blowing section has done some special type of glass works as per requirement of the Scientists.

Carpentry section having one carpenter and one helper completed the partition work of laboratories including, making wooden box, chairs, shelves with door system, Crescograph box, Glass case, and repairing and remodeling the master clock box of Sir J.C. Bose, wooden catalog tray, door closer, key board, notice board etc. Carpentry section received 150 nos of job during the period most of them are completed.

Drafting section received more than 150 requisitions from various departments for scientific drawing and all of them have been completed. This section has done the drawing of Mechanical, Civil, Electrical works.

Internal telephone system is looked after by the Electronic unit satisfactorily and it also maintained the EPABX system of Campuses. This unit received 92 nos of job during this period. Most of them are completed. Operator Console has been introduced for better telephone facility.

Store : Purchased and issued various types of materials for maintenance of electrical, mechanical, building and refrigeration. Store also purchased tools and instruments, telephone sets etc.

SUPPORTING STAFF MEMBERS

Main Campus :

Mrinal Ch. Roy, Ashutosh Sarkar, Samar Kr. Dutta, Sisir Ghosal, Sithil Mukherjee, Tarapada Das, Dipen Sen, Bankim Dutta, Subhas Chakraborty, Sk. Md. Ehia Hossain, Basanta Shit, Bholanath Saren, Sourendra Nath Chakraborty, Abdul Rahaman Molla, Panchanan Santra, Walter D' Rozario, Rajkumar Das, Pranab Banerjee, Subrata Basak, Lochan Shee, Sairun Begum, Gopal Ch. Das, Sk. Md. Farruck, Kishori Mohan Saha, Bramdeo Prasad, Kalachand Sen, Thakur Gond, Panchugopal Karak, Kalipada Das.

Centenary Campus :

Mr. Dipak Dutta, Pitambar Patra, Anil Sengupta, Tapan Kr. Mondal, Jogendra Nath Das, Ashit Banerjee, Murari Mohan Shee, Baidyanath Murmu, Sk. Md. Raffick.

Regional Sophisticated Instrumentation Centre

Dr. A. Chatterjee (upto 31.7.98) **(Scientist-in-Charge),**
Dr. D. K. Mukherjee (from 1.8.98)

The RSIC continued to provide analytical instrumental services to the academic, R&D and Industrial users from within the Institute as well as from outside. The resume of activities are shown in the table. Efforts to replace the obsolete and worn out machines have remained futile while no new machine has also been added.

Consultancy services along with help on rectification of instrumental problems were provided to the personnel of some departments or sections of the Institute as well as outsiders.

RSIC personnel also conducted lecture courses in Calcutta and Jadavpur Universities.

RESUME OF ACTIVITIES (APRIL 1998 TO MARCH 1999)

	Internal users		External users	
Name of Instrument	**No. of Samples**	**Revenue Earned**	**No. of Samples**	**Revenue Earned**
AAS	55	5,500	315	43,100
EPR	–	–	62	3,100
FLS	217	1,560	82	480
	TIME OF USE (HRS.)		TIME OF USE (HRS.)	
GLC	11	1,350	106	7,200
SEM	60	12,728	40	11,582
TEM	311	13,040	291	36,550
HPLC	120	17,246	–	–

PUBLICATIONS

1. Mondal A, Chaudhuri S, Ghosh A, Laskar IR and Ray Chaudhuri N, 1998 Synthesis, Characterization and Thermal. Synthesis and Single-Crystal Structure of an Unusual Heptacoordinate Cadmium (II) Complex, [Cd (medien) $(No_3)_2$] (medien-bis (2-aminoethy)-methylamine). Acta Chemica Scandinavica, **52**, 1064.

2. Mondal A, Chaudhuri S, Ghosh A, Laskar IR and Ray Chaudhuri N, 1998 Synthesis, Characterization and Thermal Studies of Bis (2-aminoethyl) methylamine (medien) Complexes of Cadmimum (II). Single Crystal Structure of Cd (medien) I_2. Acta Chemica Scandinavica, **52**, 1202.

SUPPORTING STAFF MEMBERS

Dr. Dilip Kumar Mukherjee (upto 31.7.98), Dr. Siddhartha Chaudhuri, Mr. Arabinda Biswas, Mr. Tapan Datta, Mr. Jaideb Chattopadhyay, Mr. Arup Kumar Sen, Mr. Samir Kumar Mukherjee, Mr. Pradip Kumar Karmakar, Mr. Jaganmoy Guin, Mr. Subhas Chandra Maikap, Mr. Pathik Ranjan Ray, Mr. Alakananda Sarkar, Mrs. Tanima Modak Dhar, Mrs. Manisha Banerjee, Mr. Debabrata Polly, Mr. Sankar Santra, Mr. Sanjoy Santra, Ms. Saraswati Sen, Mr. Shyam Sundar Kundu, Mr. Gobinda Das, Mr. Amit Kumar Mahinder.

J. C. Bose Unit and Museum

Mr. Dibakar Sen **(In-Charge)**

During the year J.C. Bose Unit and Museum has been renovated. A wall-Clock of five feet height of Acharya J.C. Bose's period, lying in our Workshop in a near dilapidated state has been recovered and brought into working condition by the Workshop and finally installed in our Museum.

Special display in Museum were arranged on **Science Day** i.e. *28th February* and to mark the **Foundation Day** of the Institute on *November 30, 1998.*

A good number of visitors both from India and abroad including students from different Colleges and Universities visited the Museum. Besides, the J.C. Bose Unit and Museum furnished information regarding Acharya J.C. Bose and his works to eminent persons and common men on request.

Documentay films on the museum were made by Prof. S. Sen, Paschim Banga Rajya Prathamik Siksha Unnayan Sanshad and Calcutta Television Network during 1998-99.

Participation in Conferences/Symposium/ Training Programme/Award and Honour received

Dibakar Sen delivered lectures on: *"Life and Works of Acharya J.C. Bose"* in *Don Bosco School, Calcutta* on *25th April 1998* : *"J.C. Bose : Scientist, Poet and Philosopher all in one"* in Ramakrishna Mission (*Deoghar, Bihar*) on *May 1998* ; *"Works of Acharya J.C. Bose (Part I and II)"* in *Academic Staff College Refresher Course, Bardhaman University, Department of Physics* on *13th June 1998* ; Ramakrishna Mission (*Purulia*), Presidential address on *02th August 1998* in Annual Exhibition and of their Science Day; *All India Radio, Calcutta A,* a Talk on *"Patra Sahityer Aloy Acharya Jagadis Chandra on 30th November 1998* ; *"Life and Works of Acharya Jagadis Chandra Bose" in Vijnan Krishi and Gramin Silpa Mela, Garalgacha,* Dt. Hooghly on *26th December, 1998.*

Sen. D. 1999, Title "UPASAK GAACH" In "AKASH PRADEEP" (62 selected Science Articles during last hundred years) edited by Dr. Shymal Chakraborty, published by Paschim Banga Bangla Academy.

SUPPORTING STAFF MEMBER

N. Das.

Distributed Information Centre

Dr. Pinakpani Chakrabarti **(Scientist in Charge)**

A. Bioinformatics Resource Created :

CONFPLOT : For displaying the three dimensional information highlighting the salient conformation features. The software is available for downloading by anonymous ftp from our server (reference : Pal, D. & Chakrabarti, P. (1999) Prot. Engg. In press)

B. USE OF DIC :

During the last one year the userbase of our Bioinformatics Centre has increased significantly. Our Centre provides unique facilities of detailed sequence analysis, molecular modelng and bibliographic searching mainly. In addition to that we are also providing email facilitiy to the scientists of Bose Institute.

C. Development of facilities :

Networking and internet service :

We have fully operational LAN in the Centenary Building (where DIC is located) and the main campus of Bose Institute. With some additional financial input from Bose Institute DIC has installed two independent internet connection at the two campuses. A home page has been created.

D. Workshops organised :

A National Symposium-cum-Workshop on "Trends in Bioinformatics" was organised during March 24th to 27th, 1998. Elaborate hands on training was given to the participants. Number of registered participants was 42.

A workshop on "structure and sequence : Data base analysis" was organised during March 11-12, 1999. The number of registered participants was 22. The workshop was highly appreciated.

E. Project Supervision :

4 D.C.A. students from RCC (Jadavpur) has completed their projects in DIC under the supervision of DIC personnel.

F. Course Work Organized :

DIC had organized two courses, Introduction to PC and Sequence Analysis for the In-house research students and associates. The courses had been included in Bose Institute's course work curriculum as core and elective courses.

G. In House Training Programe :

A course on C++ language was organised during November-December, 1998 for the In-house students and associates.

H. Hardware/Software Augmentation :

One Leased line Internet Connectivity from ERNET

One HP Scanjet Scanner

Two Cisco Routers

Two RAD Modems

Five AcerPower 6100 PCs. 32 RAM. 21.GB HDD and Color Monitor

Three AcerAltons 330 servers, 64MB RAM, 4GB HDD and Color Monitor

Besides the following items are in the process of being procured :

i) GCG Wisconsin package for sequence analysis, version 10.0

ii) Color postscript laser printer

iii) 9 GB hard disk for Decalpha

iv) Hard disk and memory upgradation for Indigo II.

PUBLICATIONS

1. Mukhopadhyay MC, 1998 Biopolymers, Molecular Dynamics Simulation of Glycoprotein Glycans of Immunoglobin G and Immunoglobin **45**, 177-190.

2. Dey I, Biomol J 1999. Structure Dynamies. BiomalExploring the interaction of some N benzyloxycarbonyl-L-phenyl alanyl-L-alanine ketones with bovine spleen cathepsin B by molecular modelling and binding free energy calculation. **16,** 891-900.

SUPPORTING STAFF MEMBERS

Dr. T. C. Ghosh, Mr. V. S. Sundararajan, Mr. T. K. Bhattacharyya, Ms. S. Majumdar, Mr. S.K. Gupta, Mr. J. Mandal.

Publication

Mr. A. K Das Gupta, **(Superintendent)**

Since its inception in 1980 the Publication Section has been doing all sorts of publications from this institute i.e. Transactions of the Bose Research Institute : Annual Report (both English and Hindi versions) ; Memorial Lectures ; Director's Report ; Folders ; Posters and Leaflets of different Symposium, Seminars Training Programmes ; and works of Sir J. C. Bose. Beside these Publication Section provides photocopying facilities to the staff members of both the Campuses of the Institute.

M/s. Das Gupta & Go. Private Limited, 54/3. College Street, Calcutta 700 073 has been appointed as sole distributor of our publications. The following books are available for sale from their counter.

J.C. BOSE AND MICROWAVES - A COLLECTIONS :		RS. 200.00
SCIENCE AND SOCIETY - RFFLECTIONS :		RS. 1050.00
J.C. BOSE - A SCIENTIST ANE A DREAMER :		
	VOL. I.	RS. 1250.00
	VOL.II.	RS. 1250.00
	VOL.III	RS. 600.00
	VOL.IV	RS. 1500.00
	VOL.V	RS. 550.00
PATRABALI		RS. 150.00

SUPPORTING STAFF MEMBER

C.K. Sasmol

Administration

Prof. P.K. Ray (Director)

Prof. Prantosh Bhattacharyya **(Registrar, Officiating upto 31 August '98)**

Shri T. K. Ghorui **(Registrar, Officiating w. e. f. 1st September '98)**

Mr. K. Chaudhuri **(Astt. Registrar I),** Mr. R.K. Kundu **(Astt. Registrar II)**

Mr. S. Pal **(Accounts Officer),** Mr. R. N. Bhattachayya **(Audit & Finance Officer)**

SUPPORTING STAFF MEMBERS

Centenary Campus

P. K. Barman, S.P. Sit, A. Bera, P. Das, Ankur Dutta, S.R. Dechoudhury, A.K. Sengupta, T. Mukherjee, E. Pal, T. Chakraborty, N. Roy Ghatak, S. Kar, Subir Sen, A. Chakraborty, Debidas Saha, Amit Kumar Ghosh, R.R. Pandey, M.S. Mitra, Dhrubajyoti Sen, Alok Kundu, B. Goswami, Sujit Kr. Ghosh, B.K. Mandal, M.C. Das, Rita Nath, Sisir Chakraborty, Swapan Kr. Singha, Rubi Sarkar, Sudam Jana, P. Mullick, R. Chakraborty, Rina Das, Sanjay Kr. Chaki, Debashis Koley, Somnath Das, A. Bhowmik, Sujata Pandit, Sukumar Das, S. Chatterjee, R.C. Sarkar, Dipak Datta, Ranjit Ghosh, K. Sur, C. Rangwa, T.K. Banerjee, K. Turi, S. Turi, K. Basar, N. Yadav, Md. K. Ansari, S. Guchait, Kanai Hazra, P. Bhuinya, Nemai Das, Kanak B. Hazra, Duryodhan Nayak, Sanjib Das, Bablu Mondal, Kashinath Pul, Kamala Turi, Subrata Banerjee, Sukanta Chakraborty, Dulal Turi, Amar Nath Hela, Ranjit Das, Sukumar Mondal, Bulaki Das.

Main Campus

Arun Kumar Das Gupta **(Superintendent),** Samar Chakraborty **(Overseer),** Sankar Nath Sinha, Rina Roy, Sanat Kr. Dey, Mahendra Nath Shee, Santosh Patra, Ramani Kanta Jana, Tarun Chakraborty, R. Ram, S. Devi. Jubaraj Paul, Nathu Ranjan Gope, Harekrishna Thoyal, Madhusudhan Marick, Fulchand Sardar, Kalicharan Turi, Kisson Das, Munna Turi, Kudan Das, Sk. Md. Kalu, Sk. Md. Eliash, Sk. Md. Zakaria, Sm. ManjuTuri.

OTHER ACTIVITIES

Lectures

1. "TFII-I : A mediator between signal transduction and transcription" —Dr. Ananda L. Ray, Tufts University School of Medicine, Boston, USA Friday, August 21, 1998
2. "Erosion of Agricultural Biodiversity"—Dr Deval Dev, Ecologist, Centre for Interdisciplinary Studies, Barrackpore, January 29, 1999
3. "Transcendance in the post-modern world"—Dr. Pattabi Raman, Center for the promotion of Learning Abilities, Auburn, Washington, USA Feb. 26, 1999
4. "Deconstructing Bacterial Cell durision"—Dr. Debabrata Raychaudhuri, Dept. of Microbiology and Molecular Biology, Fufts University School of Medicine Boston, USA April 23, 1999

Foundation Day Celebration
and
Acharya Jagadis Chandra Bose Memorial Lecture 1998

Secretary Governing Body paying homage to the Founder Sir J. C. Bose on Foundation day

Director Bose Institute presenting Life time achievement award to Prof. Amitava Sen

The 82nd Foundation Day of Bose Institute, was celebrated on 30th November, 1998. Professor N. K. Ganguly, Director-General, ICMR, Delhi, kindly graced this occasion as guest of honour and delivered the 60th Acharya Jagadis Chandra Bose Memorial Lecture on **"Present and Future Scenerio of AIDS in India".**

Five eminent scientists Professor D. P. Burma, Prof. S. P. Sen, Prof. M. S. Sinha, Prof. P. N. Nandi and Prof. Amitabha Sen, were honoured on this day for their life-time research contributions in this Institute.

In this occasion Prof. A. N. Lahiri Mazumdar of PMCG Section and Dr. Ranjan Nandi, a Research Scholar of Microbiology Department received Foundation Day Award and Sir Nilratan Sircar Prize, respectively. Both the awardees dilivered lectures on their respctive area of reasearch. The celebration ended with a brief cultural evening.

National Science Day

Stall displying various aspects of information technology on National Science Day

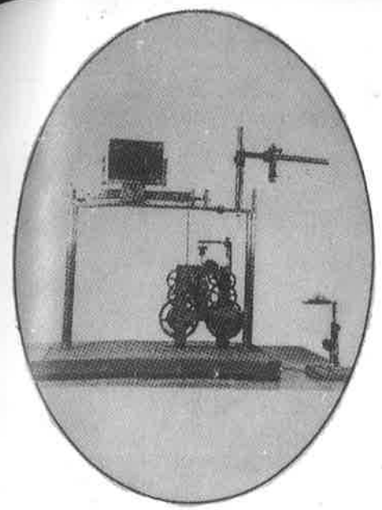
Competitor participating in debate competition on National Science Day

National Science Day was celebrated on February 28, 1999 in Bose Institute, Calcutta. On the Focal Theme "Harnessing Information Technology". Activities like debate competition, popular lectures, exhibitions, etc., related to the focal theme and 'open house' were organised

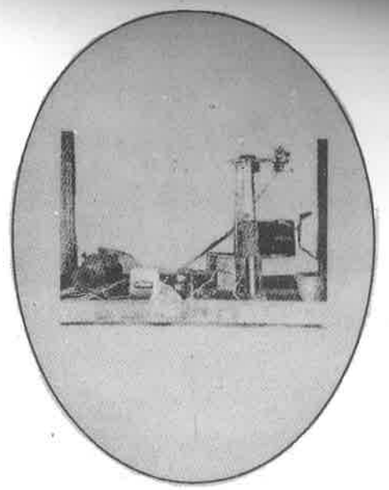
The debate competition among the school boys was held on 26th February, and several schools in and around Calcutta participated. Eminent personalities like Prof. Gautam Bhattacharyya (SINP), Mr. S. K. De (VECC) and Mr. K. B. Som (Calcutta Telephones) delivered popular lectures on various aspects of Computer Technology and its uses. Films based on applications of Computer Technology were displayed by our Library Section.

Several organizations like VSNL, VECC, British Council, Lakhotia Computers, BITM, our DIC, Publication Section, Library, etc., participated in the live demonstration of various aspects of the mirecles of Computer Technology, for public and also displayed various informative posters, books, scientific teaching devices, etc. Many visitors from different walks of life, mostly students from schools and Colleges enjoyed the day's activities and also payed their visits to different laboratories and Sir J. C. Bose Museum.

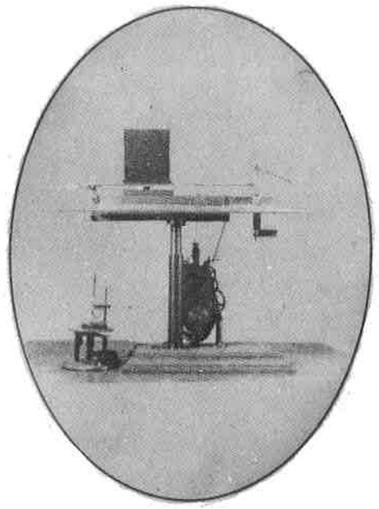
Director, Bose Institute distributed prizes to the winners of the debate competition.



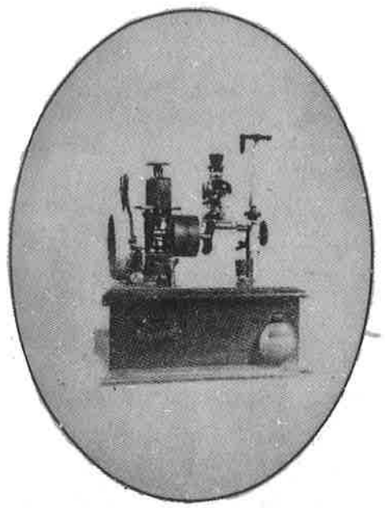
PHYTOGRAPH



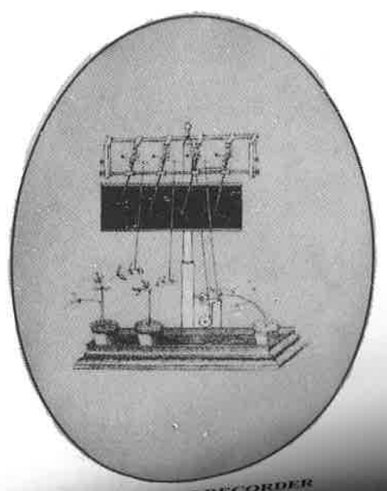
RESONANT RECORDER



PORTABLE COMPOUND
LEVER CRESCOGRAPH



PHOTOSYNTHETIC BUBBLER



FOUR PLATE RECORDER