

NBFGR





News

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From the Director's Desk....

Characterization of aquatic genetic resources is an important mandate of the NBFGR. Keeping pace with the changing needs and challenges, the Bureau is positioning itself in the field of fish genomics. Towards this objective, completing the whole mitochondrial genome sequencing of *P. chalakkudiensis* was a notable achievement. Success achieved in identification of a novel Cystatin-like gene in a hypoxia-tolerant Indian catfish, *Clarias batrachus* is also a testimony to it. The efficacy of a cell line developed from *Puntius chelynoides* as an *in vitro* model for toxicity studies was validated. A Microsatellite Database of Indian Fishes was developed which would serve as an useful resource in several areas of fisheries research. The National Consultation on Development of Surveillance programme



for aquatic animal diseases organised by the Institute in collaboration with NFDB, DAHDF & ABCS brought out some strategic recommendations, which is expected to help in formulation of a national surveillance programme. Further, two other important workshops focused on issues of national significance namely, Fish cell line: development and storage, and Strategic action plan for exploration and characterization of fish germplasm resources and indigenous knowledge in North-Eastern region of India, brought together concerned experts at a platform and generated valuable recommendations. I compliment my colleagues for their hard work in these endeavours.

J.K. Jena Director

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RESEARCH HIGHLIGHTS

Whole mitochondrial genome of *Puntius chalakkudiensis* sequenced

The Chalakkudy barb, *Puntius chalakkudiensis* is reported from a number of rivers in the southern end of the Western Ghats of India. This fish phenotypically appears to be similar to the Red-line Torpedo Fish, *Puntius denisonii*, a popular aquarium fish and was first thought to be the same species; however *P. chalakkudiensis* has a more

under slung mouth and also has a dark spot in the middle

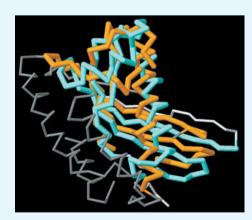
of the red dorsal fin; unlike *P. denisonii*. Initially considered endemic to the Chalakkudy River system in Kerala state, the species is now recognised to have a wider distribution in surrounding drainages including the Periyar, Manimala, and Pamba Rivers.



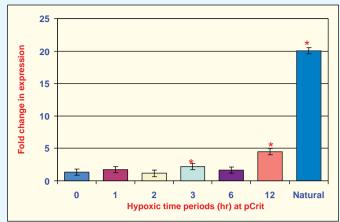
Assessment of the genetic make-up and variability of fish stocks is important for the judicious management of fishery, conservation and rejuvenation of endangered species and improvement of stocks of cultivable species. The whole mitochondrial DNA (mtDNA) sequence information is useful in analysing genetic lineage between closely important species and deepest phylogenetic analysis. Therefore, whole mitochondrial DNA sequencing of *P*. chalakkudiensis was carried out by amplifying contiguous overlapping segments of the genome using five sets of primers and also synthesizing primers from *P. chalakkudiensis* itself by primer walking. The sequence of the amplified segments was used to design new primers for amplification of interspersed regions that were not amplified. Sequencing of whole mitochondrial genome of P. chalakkudiensis was completed which generated a length of 16,997bp DNA.

A novel Cystatin-like gene in a hypoxia-tolerant Indian catfish, *Clarias batrachus* identified

As a part of programme on Bioprospecting of alleles for abiotic stress tolerance in fishes, novel genes involved in the hypoxia tolerance in *C. batrachus* were investigated. A novel transcript was identified from the head kidney subtracted cDNA library, in response to oxidation stress, the similarity search of this transcript showed no significant homology, which indicated it to be a hypoxia responsive novel sequence. Integrated genomic approaches, expression profiling and computational techniques showed it to have putative cystatin/monelin like domain, thus named *CbCystatin*. It might be a transmembrane and/or intermediate protein in signaling pathways. The



Schematic view of the CbCystatin homology model



*significant difference from normoxic control group (p<0.05)
Effects of hypoxia on the mRNA expression of *CbCystatin*in head kidney tissue of fish *Clarias batrachus*

predicted secondary structure of *CbCystatin* indicated it to be an alpha-beta type of protein, similar to that of chicken cystatin and was found to be clustered into family 2 cystatins. At transcriptional level, its expression was significantly up-regulated in response to short, as well as, long periods (more than 20 fold) of hypoxia, suggesting its positive association with oxygen concentrations lower than physiological concentrations. This work was carried out under National Agricultural Innovation Project "Bioprospecting of Genes and Allele Mining for Abiotic Stress Tolerance: Anoxia tolerance in Indian catfish".

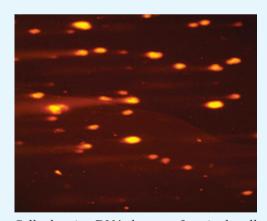
First record to demonstrate role of *in vivo* melatonin in gonadal sex differentiation in *Cyprinus carpio*

Pineal melatonin in teleostean fish is now identified to play pivotal role in biological and reproductive physiology. There are some reports demonstrating antigonadal activity of melatonin while other reports are available to show progonadal activity. However, role of pineal melatonin is not known during sex differentiation. Therefore, the possible involvement of pineal melatonin during gonadogenesis and sex differentiation was determined by analyzing the changes in the serum melatonin level of the fish treated with aromatase inhibitors to block the normal sex differentiation. It was interesting to note that serum melatonin level decreased during inhibited aromatase activity induced by the treatment of fadrozole and anastrazole (100 and 200 mg/kg). There was significant decrease in estradiol (E2) production leading to the masculinization in fish. The results of this study

suggested that fadrozole and anastrozole both effectively inhibited oogenesis and ovarian development accelerating testicular formations. Further, there was a correlation of aromatase activity and the melatonin production during gonadal development and sex differentiation in *Cyprinus carpio*.

Cell line as *in vitro* model for genotoxicity and cytotoxicity studies

A cell line developed from eye tissue of Puntius chelynoides was used for genotoxicity and cytotoxicity assessment of heavy metal salts and nanoparticles. Divalent salts used to investigate the toxic effect of two heavy metals viz., zinc and cadmium were: ZnCl₂, ZnSO₄, ZnNO₃, CdCl₂ and CdSO₄. Toxicity potential of titanium oxide, zinc oxide, silver and gold nanoparticles were also evaluated. Heavy metal salts were found to be cytotoxic, as well as, genotoxic to the studied cell line at varied concentrations. Results revealed that cadmium salts were more toxic than zinc salts. Tested nanoparticles were found genotoxic to cell line but did not exhibit any significant cytotoxicity. Results strongly validated the efficacy of this cell line as an in vitro model for toxicity studies.



Cells showing DNA damage after single cell gel electrophoresis

Microsatellite database of Indian fishes

DNA microsatellite sequences are widely dispersed in the non-coding region of eukaryotic genome and have emerged as important markers for characterization and genetic improvement of fishes. In view of the wide application of microsatellites, a DNA Microsatellite Database of Indian Fishes (MIDIF) was

developed. The database was designed using MySQL database management system on Linux platform and currently contained 1124 microsatellite sequences of 35 species belonging to 12 families, and is being updated regularly. The source data was derived from GenBank of NCBI in GenBank and FASTA file format. Several applications were implemented for allele mapping, primer designing, statistical analysis about repeats and determining the type of repeat that frequently occurs in fish genome. This database can serve as a useful resource in several areas of fisheries research such as disease diagnostics, breeding for disease resistance, genetic linkage mapping, population genetics, marker assisted selection, etc.

Exploration and assessment of fish diversity of an unexplored River Rapti in Uttar Pradesh

In Uttar Pradesh, there are several riverine areas in which status of fish diversity has not been explored. Therefore, extensive fish exploration was conducted covering the entire stretch of the River Rapti, a tributary of River Ghagra which is a major tributary of River Ganga. A total of 53 species of 41 genera belonging to 18 families were recorded for the first time from this river. The river harbors several fish species of economic and conservation value. Lengthweight relationship and condition factor of 40 species from River Rapti was studied. The growth component 'b' varied from 1.14 to 2.66 at p<0.001. The influence of environmental conditions on species occurrences and assemblage in different rivers was assessed in different microhabitats of river Rapti.



Fishing for the exploration of germplasm at river Rapti

IMPORTANT EVENTS

National Consultation on 'Development of Surveillance Programme for Aquatic Animal Diseases'

A National Consultation on 'Development of Surveillance Programme for Aquatic Animal Diseases' was organized at NBFGR, Lucknow during 17–18 April, 2012 in collaboration with DAHDF, Government of India; National Fisheries Development Board, Hyderabad and Aquatic Biodiversity Conservation Society, Lucknow. Forty experts including research scientists, development officials



and policy makers involved in aquatic animal health from all over the country, as well as, an expert from Network of Aquaculture Centers in Asia-Pacific (NACA), Thailand participated in this meeting.

Dr J. K. Jena, Director, NBFGR and Convener of the National Consultation welcomed the delegates and highlighted threats to the sustainability of the aquaculture sector due to new and emerging diseases. Dr. Jena opined that the present consultation would help in developing a roadmap for national



surveillance programme for aquatic animal diseases and manage the disease risks associated with aquatic animal trade. Prof. C.V. Mohan, Research and Development Manager, NACA, Thailand shared his experiences on the global scenario of aquatic animal disease surveillance programmes. He appreciated the efforts of NBFGR for taking this initiative and assured support of the NACA in developing a national surveillance programme on aquatic animal diseases. In her presidential address, Prof. Indrani Karunasagar, Head, Department of Microbiology, College of Fisheries, Mangalore emphasized the need for level III diagnosis particularly metagenomics in detecting the causative agents of various aquatic diseases syndromes. In technical sessions, status of diseases in finfishes and shellfishes was discussed by the participants. The development of implementation mechanisms for this surveillance programme was discussed by a panel of experts.

The Consultation recommended that a "National surveillance programme for aquatic animal diseases" be initiated in 14 selected states of fisheries and aquaculture importance in the first phase. The consultation agreed on a national list of diseases to be included under national surveillance programme in selected 14 states in first phase for the purpose of implementing national disease control strategies and meeting international obligations. Dr. P. Puina, Head, Fish Heath Division thanked all the participants for their contribution in the sucess of the Consultation.

National Workshop on 'Fish Cell Line: Development and Storage'

A National Workshop on 'Fish Cell Line: Development and Storage' was organized at NBFGR, Lucknow in collaboration with the Department of Biotechnology, Govt. of India and Aquatic Biodiversity Conservation Society (ABCS), Lucknow on 19 April, 2012. A galaxy of scientists besides scientist from the Network of Aquaculture Centre for Asia-Pacific (NACA) Thailand participated in the Workshop. The workshop was inaugurated by Dr. S.A.H. Abidi, Former Member, ASRB and Dr. Dilip Kumar, Former Director, CIFE, Mumbai presided over the function. Dr. J.K. Jena, Director, NBFGR and Convener of the Workshop highlighted the necessity of cell lines in fish disease management in the context

of higher aquaculture intensification. Guest of Honor Dr. A.S. Ninawe, Advisor, DBT, Govt. of India, New Delhi stressed upon the need for developing a perfect protocol for characterization of the cell lines in order to ensure their utility. He stressed that over 50 fish cell lines have been developed from different species in India and to store these cell lines a cell line repository is urgently required. Keeping this in view DBT has funded a project to establish of a National Repository of Fish Cell lines at NBFGR, Lucknow. participants deliberated on the status of fish cell lines, its application and storage in the repository. The workshop was aimed to prioritize the species to be used for cell line development and to make strategies for collection, characterization, storage and distribution of cell lines through a national repository of fish cell lines for R&D work. Dr. Mukunda Goswami, Sr. Scientist, NBFGR co-ordinated the Consultation.



Workshop on 'Strategic Action Plan for Exploration and Characterization of Fish Germplasm Resources and Indigenous Knowledge in North-Eastern Region of India'

A workshop on 'Strategic action plan for exploration and characterization of fish germplasm resources and indigenous knowledge in North-Eastern region of India' was organized by NBFGR, Lucknow, in collaboration with College of Fisheries (COF), Agartala during 5-6 May, 2012 at Agartala. The workshop was inaugurated by Prof S.N. Puri, Vice-Chancellor, Central Agricultural University, Imphal in presence of Dr. A.P. Sharma, Director, CIFRI, Barrakpore, Dr. J.K. Jena, Director, NBFGR, Dr. J.R. Dhanje, Dean, College of Fisheries, Agartala and Dr. K.K. Lal, Head, Fish Conservation Division, NBFGR. About 60 invited participants from different



states of the North-eastern region, including scientists of NBFGR participated in the workshop. The objectives of the workshop were to (1) prioritize aquatic areas for exploration based on identified criteria like prior knowledge on biodiversity richness or endemicity of the region, recognized protected or conservation importance, dependence of tribal or native ethnic populations utilizing such aquatic resources, knowledge gap on aquatic bioresources of the region; (2) formulate work pogrammes under network and sub network mode, and (3) discuss operational strategy and capacity building needs of the perspective collaborators. The idea was to address researchable issues through combined use of expertise available at NBFGR and various organizations in the region for exploration of aquatic germplasm resources of the North Eastern Region of India. Participants from different North-eastern states presented their research proposals under four themes viz., exploration of fish germplasm resources, biological and genetic characterization, documentation of indigenous



knowledge, and live gene banking and captive breeding.

International Day for Biological Diversity

NBFGR collaborated Uttar Pradesh State Biodiversity Board (UPSBB) in organization of a Conference on Marine Biodiversity on 22 May, 2012 to celebrated International Day for Biological Diversity. Speakers on the occassion were Dr. Syed Azam Khan Prof. Emeritus, Centre for Advanced Studies, Marine Biology, Annamalai University, Tamil Nadu; Shri R.K. Singh, President, UPSBB and Secretary, Ministry of Environment & Forest, Government of Uttar Pradesh; Shri Pawan Kumar, Secretary, UPSBB; Shri J.S. Asthana, Chief Conservator of Forest, Uttar Pradesh; Prof. B. Chauhan, Vice-Chancellor, Dr. Ram Manohar Lohia National Law University, Lucknow and Dr. J.K.

Jena, Director, NBFGR, Lucknow. A galaxy of eminent environmentalists, scientists, professors and teachers of various universities/institutions and students participated in the conference.

Annual Institute Research Council Meeting

The Annual IRC meeting of the Institute was conducted on 27-28 April, 2012 and 17-19 May, 2012 under the chairmanship of Dr. J.K. Jena, Director. The progress of all ongoing projects was critically reviewed by the IRC and important suggestions emerged from the discussion. Detailed presentations on all the projects were made by the respective Principal Investigators/Co-PIs. The new project proposals were also discussed and important ones were approved with suggested modifications.





PARTICIPATION IN WORKSHOPS/SEMINARS/MEETINGS/TRAININGS

Abroad

Dr. A. Gopalakrishnan, Principal Scientist and SIC, NBFGR Kochi Unit attended the Bay of Bengal Large Marine Ecosystem (BOBLME) Project Working Party Meeting on Assessing stock structure of Indian Mackerel (*Rastrelliger kanagurta*) for Fisheries Assessment in Colombo, Sri Lanka, during 28 - 29 May, 2012.

Dr. P.K. Pardhan, Senior Scientist, attended two months training under International Foundation for Science at Institute of Research in Agriculture and Food Technology (IRTA), Sant Carlos de la Rapita, Spain during 20 April to 20 June, 2012.

In India

Dr. J.K. Jena, Director participated in the following

meetings/seminars/workshops:

National Consultation Workshop on 'Coastal & Marine Biodiversity: Gaps, Challenges and Opportunities' from 12-13 April, 2012 at Gandhinagar, Gujarat.

Joint meeting of DARE/ ICAR and NBA on draft guidelines for access to genetic resources for food and agriculture and other related issues on 2 May, 2012 at NBPGR, New Delhi.

Meet on 'Higher Fisheries Education-Way Forward' on 14 May, 2012 at NASC, New Delhi.

Dr. J.K. Jena, Director and Dr. K.K. Lal, Head, Fish Conservation Division participated in the Mid-term Review of NAIP-GEF project with DG, ICAR on 16 May, 2012 at New Delhi.

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27-28 Vißy 2012 , oa17-19 eb] 2012 dks l & Fkku dsfunskd Mk-tsds tsuk dh V/; {krk eal & Fkku 'kksk ifj 'kn dh okf 'kd cBd Vk; kstr dh x; hA | Hkh ifj; kstuk Vka ds Vloskdka } kjk ifj; kstuk Vkadh folrr fjiks/Zilr f dh x; h r Fkk u; h ifj; kstuk Vka grqitrko itr f fd; sx; sr Fkk mueal segRoiw kZifj; kstuk Vkadks Vko'; d | fkki kads | kFk Vueeksnr fd; k x; kA

dk; 2 kkykvkæl xkf"B; kæcBdkæif'k{k.kkaeaifrHkkx %

fonsk % Laufku dh dkapp bdkbz ds i blkjh , oa i zkku oakkfud Mk- ,-xki kykd".ku rfkk Mk- i h-ds i zkku] ofj"B oakkfud }kjk Øe'k% Jhyadk , oa Lisu ea vk; katr cad o i f'k{k.k dk; De ea Hkkx fy; k x; kA

nsk % l & Fkku dsfunskd Mk-tsds tsuk } kjk xkdkh uxj] x¢jkr ea l enh t&fofo/krk i j vk; k&tr dk; Zkkyk eaHkkx fy; k x; kA

HKK-d-v-i- dsegkfunskd MKW , I - v; iu dslkFk , u-, -vkbZih-@th-bZ, Q- ifj; kstuk dh e/; dkfyd lek{kk cBd ea16 ebZ2012 dksubZfnYyh eal &Fkku dsfunskd MKW tsds tsuk , oaeRL; lj{k.kiłkkx dsiłkkxk/; {k MKW dsds yky ushkx fy; kA

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