भाकृ अनुप
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CENTRAL INSTITUTE OF BRACKISHWATER AQUACULTURE, CHENNAI

CONTENTS

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Chennai - 600 028.

PCR - BASED DIAGNOSIS OF INDIAN WHITE SPOT VIRUS

White Spot Syndrome Virus (WSSV), known to cause white spot disease, is the most virulent virus affecting cultured shrimps *viz.*, *Penaeus monodon* and *Fenneropenaeus indicus*. Till date, no treatment is known to control this viral disease. Early diagnosis / detection and adoption of health management practices are the only alternatives for tackling it. Conventional diagnostic tools using histopathological techniques are time consuming, less sensitive and fail to detect early stages of infection. Hence, a rapid, specific and sensitive diagnostic tool is necessary for diagnosis of WSSV disease in shrimps.

Penaeus monodon affected with white spot disease virus

DNA - based diagnostics

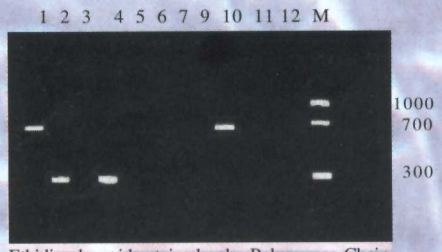
Advances in molecular biology enabled researchers to design a new generation of DNA based diagnostic tools, such as the Polymerase Chain Reaction (PCR) technique and Gene Probes.These tools are known for their speed, sensitivity and simplicity and are capable of identifying a number of viral, bacterial and parasitic pathogens even at a very early stage (asymptomatic / carrier stage) of infection. Recently, the PCR technique has been proved to be the most sensitive and specific diagnostic tool for the screening of WSSV disease in shrimp seed and mother shrimps (spawners / broodstock).

PCR

PCR is a relatively simple technique by which a DNA or cDNA template of target pathogen is amplified a thousand or million fold, quickly and reliably, in a short period of 3-4 hours. So far, no other technique has equaled PCR in sensitivity.

The PCR process uses a specialized DNA-polymerase enzyme which can synthesize complementary strands, using a DNA fragment of about 20 bases long as primers, incorporating 4 nucleotides flanking the target sequence. The mixture is heated in an automated instrument called thermocycler, to separate the double stranded DNA containing the target sequence by thermal denaturation and then cooled to allow (1) the primers to bind to their complementary sequences on the separated strands i.e. annealing of oligonucleotide primers to the complementary sequence and (2) primer-directed DNA syntheses with the help of thermostable taq DNA polymerase, resulting in formation of new complementary strands. By repeated heating and cooling cycles, the target DNA is multiplied exponentially. Each new double strand separates to become two templates for further syntheses. Thus after one cycle, there will be 2 copies, after two cycles, there will be 4 copies and after three cycles 8 copies and so on. In this way, the target is amplified several folds, producing enough DNA to be visualized, to enable diagnosis. The product obtained, can be analyzed by a number of methods such as agarose gel electrophoresis. The products also can be readily visible under UV trans-illumination of an ethidium

bromide stained gel.(Fig.1).



Ethidine bromide stained gel - Polymerase Chain Reaction (PCR) diagnosis of WSSV infected *P. mondon* broodstock.
Lane 1.Sample No.1 WSSV 1st step PCR positive, Heavy infection
Lane 2 Sample No.2 WSSV Nested PCR positive, mild infection
Lane 3 Sample No.3, PCR 1st step negative
Lane 4 Sample No 3, Nested PCR positive, mild infection
Lane 5,6 and 7 nested PCR negative, WSSV free shrimps
Lane 8 Negative tissue control Nested PCR product
Lane 9 Positive control, 1step WSSV positive
Lane 10 Positive control, 2nd step WSSV positive
Lane 11 Negative reagent control
Lane 12 Negative tissue control
M Molecular weight marker

Nested PCR

Nested PCR or two step PCR is useful in reducing or eliminating unwanted products and simultaneously increasing the sensitivity significantly. An aliquot of the first PCR product is then subjected to an additional round of amplification using primers complementary to the sequences internal to the first set of primers. Only the legitimate product is amplified in the second round. This approach of two step amplification is often useful even if the designed product is initially below the level of detection by ethidium bromide staining.

Uses of PCR

The PCR can be widely used in detecting the entry of lethal pathogens such as viruses in shrimp hatchery and grow out systems. It can also be used in epizootiological studies of pathogens in an effort to draw-up disease control measures.

RESEARCH HIGHLIGHTS

Studies on the treatment of shrimp farm waste water, using Bagasse (Sugarcane by-product), indicated that it is effective in the removal of NH_3-N, NO_2-N and other metabolites .

VISITORS

- Dr.R.A.Selvakumar, Retd. Assistant Director General (M.Fy.), ICAR, New Delhi, 10 July 2001.
- Shri V.Venkatesan, Retd., Director, MPEDA, Kochi, 10 July 2001.
- Dr. Subba Rao, Retd. Principal Scientist, CIFE, Mumbai, 10 July 2001.
- Shri R.Mathivanan, Principal, Tamil Nadu Fisheries Staff Training Institute, Chennai, 10 July 2001.
- Shri Y.C.Thampi Sam Raj, Project Director, Rajiv Gandhi Centre for Aquaculture (RGCA), MPEDA, Mayiladuthurai, 10 July 2001.

ENGAGEMENTS

Dr.G.R.M.Rao, Director, CIBA, attended the following Meetings / Conferences / Workshops:

- 22nd Meeting of the Aquaculture Authority, at Kolkata, 11 July 2001.
- 5th Meeting of National Committee on Introduction of Exotic Aquatic Species in Indian Waters, at New Delhi, 17 July 2001.
- Meeting of Directors' of ICAR Institutes, at NBPGR, Pusa, New Delhi, 23-24 July 2001.
- National Workshop on Sustainable Aquaculture for Future India, at University of Madras, Chennai, 26-27 July, 2001.
- Sensitisation Workshop under NATP (Coastal Agro-ecosystem), at University of Agricultural Sciences (Hebbal), Bangalore, 31 July 2001.
- 9th Scientific Advisory Panel Meeting of NATP, at University of Agricultural Sciences (Hebbal), Bangalore, 1 August 2001.
- 23rd Meeting of the Aquaculture Authority, at MPEDA, Kochi, 8 September 2001.
- National Conference on "Environmental Biodiversity and Bioethics - Current Trends and Future Direction", at Loyola Institute of Frontier Energy, Loyola College, Chennai, 20 September 2001.
- Meeting of Directors' of ICAR Fisheries Research Institutes, at CIFE, Mumbai, 21-22 September 2001.
- 15th Annual General Body Meeting of OSSPARC, 19th Executive Committee and 8th Annual General Body Meeting of RGCA, at MPEDA Regional Office, Chennai, 26 September 2001.
- Fourth World Conference on the Shrimp Industry and Trade and Buyer and Seller Meet, at Chennai, 27-29 September 2001.

The scientists attended the following Seminars / Workshops:

- Dr.K.O.Joseph, Senior Scientist, delivered a guest lecture on "Changing patterns in the soil and water conditions of the coastal region due to intensification of shrimp farming", at the International Workshop on Aquaculture and Environment, at Kochi, 13-14 July 2001.
- Dr.P.Ravichandran, Dr.T.C.Santiago, Principal Scientists and Shri K.Ponnusamy, Scientist, attended the Sensitization Workshop under NATP (Coastal Agro-ecosystem), at University of Agricultural Sciences (Hebbal), Bangalore, 30-31 July 2001.

SPECIAL FISH FARMERS' DAY CELEBRATIONS AND FARMERS' MEET

A Farmers' Meet was organized by CIBA in connection with the Special Fish Farmers' Day Celebrations, at Muttukadu Experimental Station, on 10 July 2001. Dr.G.R.M.Rao, Director, presided over the function. More than sixty fish farmers from Chennai, Kancheepuram and Tiruvallur districts of Tamil Nadu participated in the Meet. Other participants included officials from the State Fisheries Department of Tamil Nadu

Special fish Farmers Day Celebration at Muttukadu Experimental Station of CIBA on 10 July 2001. Left to right Shri Y.C.Thampi Samraj, Project Director, RGCA, Dr.Subba Rao, Retd. Principal Scintist, CIFE, Mumbai, Dr.R.A.Selvakumar, Retd. Assistant Director General (M.Fy.), ICAR, Dr.G.R.M.Rao, Director, CIBA, Dr.V.Venkatesan, Retd. Director, MPEDA, Kochi and Shri R.Mathivanan, Principal, Fisheries Staff Training Institute, Chennai.

and the scientists and staff of CIBA, Chennai and Muttukadu. The Best Fish Farmer Award was presented to Shri K.S.Raja Devasenapathi, a progressive fish farmer from Tamil Nadu. Dr.Subba Rao, Retd. Principal Scientist, CIFE, Mumbai,

distributed merit certificates to enterprising fish farmers. On this occasion, the CIBA hatchery produced seabass seed was distributed to private fish farmers. A special lecture was delivered in regional language (Tamil) by Dr.A.R.Thirunavukkarasu, Principal Scientist, on captive breeding of seabass. Dr.R.A.Selvakumar, Retd. Assistant Director General (M.Fy.), ICAR, New Delhi; Shri V.Venkatesan, Retd. Director, MPEDA, Kochi; Shri R.Mathivanan, Principal, Tamil Nadu Fisheries Staff Training Institute, Chennai and Shri Y.C.Thampi Sam Raj, Project Director, Rajiv Gandhi Centre for Aquaculture (RGCA), MPEDA, Mayiladuthurai, offered felicitations.

Shri K.S.Raja Deva Senapathy, a progressive fish farmer in Tamil Nadu receiving the best farmer award from Dr.G.R.M.Rao, Director , on 10 July 2001.

PARTICIPATION IN THE AGRI-INTEX 2001 EXHIBITION

Shri R.Jeevanantham, Hon'ble Minister for Agriculture, Govt. of Tamil Nadu (Second from left) at CIBA Stall in AGRI INTEX Exhibition at Coimbatore on 31 July 2001. Dr.N.Kalaimani, Principal Scientist & SIC, EEID (second from left) looks on.

The Institute participated in the AGRI-INTEX 2001 Exhibition organized jointly by Coimbatore District Small Industries Owners Association (CODISSA) and Tamil Nadu Agricultural University, at Coimbatore, during 31 July to 7 August 2001.

HUMAN RESOURCE DEVELOPMENT

Shri K.Ponnusamy, Scientist, underwent training in "Rice Production Technology" at Tamil Nadu Rice Research Institute, (Tamil Nadu Agricultural University), Aduthurai, under the NATP/ IVLP for Technology Assessment and Refinement in Coastal Agro-ecosystem of Tiruvallur district of Tamil Nadu", during 16-20 July 2001.

TRAINING

The following training programmes were organized by CIBA at Headquarters, Chennai / Muttukadu Experimental Station:

- "Brackishwater finfish breeding" (seabass *Lates calcarifer)* during 17-26 July 2001. Three officials from Marine Products Export Development Authority (MPEDA) and three private entrepreneurs participated.
- "Shrimp disease and health management" during 1-10 August 2001. 15 participants underwent the training *viz*., 8 officials from MPEDA (three from Kochi, one each from Kannur, Bhubaneswar, Kolkata, Panvel and Vijayawada), 2 private entrepreneurs (one shrimp hatchery operater from Chennai and one shrimp farmer from Andhra Pradesh), one scientist from M.S.Swaminathan Research Foundation, Chennai and four Senior Research Fellows from CIBA.

Lectures and demonstrations were arranged for the following at CIBA Headquarters, Chennai/ Muttukadu Experimental Station:

- 18 students (B.F.Sc.), from Fisheries College and Research Institute, Thoothukudi, 11 August 2001.

PARTICIPATION IN ICAR INTER-ZONE SPORTS MEET

- Shri R.Mathivanan and Shri R.Kumaresan, SS.Gr.I, represented the south zone at the ICAR Inter-zone Sports Meet at CIFT, Cochin, 4-9 December 2001 and won the 2nd prize in Carrom and 3rd Prize in Long Jump, respectively.

R.Kumerasan and R.Mathivanan reciving the third prize in long jump and second prize in carrom, respectively at ICAR Inter-Zone Sports Meet held at Kochi, 4-9 December 2001.

STAFF NEWS

Retirement

- Shri K.Devarajan, Principal Scientist, retired on superannuation, 31 July 2001.

The following Senior Scientists were promoted to Principal Scientist, w.e.f. 27 July 1998:

Dr.L.H.Rao, H.O.D., CCD,
Dr.Mathew Abraham,H.O.D, FCD,
Shri K.Devarajan,
Shri S.R.Das,
Dr.P.Ravichandran,
Shri M. Kathirvel,
Dr.S.Kulasekarapandian,
Dr.S.M.Pillai,
Dr.T.C.Santiago,
Dr.A.R.Thirunavukkarasu,
Dr.S.A.Ali,
Dr.K.Gopinathan,
Shri. R.K.Chakraborti,
Shri. S. Srinivasagam,
Dr. C.P. Rangaswamy,
Dr.B.P.Gupta,
Shri. D.Narayanaswamy,
Dr. N. Kalaimani,
Dr.M.Natarajan and
Dr.(Mrs.) Munawar Sultana.

- Dr.C.Gopal, Scientist (Senior Scale) to Senior Scientist, w.e.f. 27 July 1998.
- Shri S.V.Alavandi, Scientist (Senior Scale), to Scientist (S.G), w.e.f. 27 July 1998.
- Dr.Azad, I.S., Scientist to Scientist (Senior Scale) w.e.f. 25 March 1994 and Scientist (Senior Scale) to Senior Scientist w.e.f. 25 March 1999.
- Dr.K.K.Vijayan, Scientist (Senior Scale) to Senior Scientist, w.e.f. 5 October 1999.
- Dr.T.Ravisankar, Scientist to Scientist (Senior Scale), w.e.f. 27 July 1998.
- Mrs. D. Deboral Vimala, Scientist to Scientist (Senior Scale), w.e.f. 27 July 1998.

केन्द्रीय खारापानी जलजन्तु पालन संस्थान

पी.सी.आर. के आधार पर वाइट स्पाट सिंड्रोम वाइरस का निदान

वाइट स्पाट सिंड्रोम वाइरस, पालित झींगों (पी. मोनोडान तथा पी. इंडिक्स) को प्रभावित करनेवाला बहुत-ही विषैला विषाणु है। इस विषाणु के कारण होनेवाले वाइट स्पाट रोग के लिए आज भी कोई उपचार उपलब्ध नहीं है। पहले इस रोग की रोकथाम के लिए निदान कार्य तथा स्वास्थ्य प्रबंधन प्रयास ही एकमात्र उपाय था।

हाल ही में तैयार किया गया डी.एन.ए. पर आधारित निदान यंत्र, पालिमिरेज चैन रिएक्शन (पी.सी.आर.) तकनीक झींगा बीजों व माता झींगा (अण्डजनक/शावक संजाति) में डब्ल्यू.एस.एस.वी. के परीक्षण के लिए बहुत-ही सूक्ष्मग्राही निदान यंत्र सिद्ध हुआ है।

पी.सी.आर. बहुत-ही सरल तकनीक है जिससे जीवों के डी.एन.ए./सी.डी.एन.ए. साँचों को बहुत ही कम समय अर्थात् 3-4 घण्टों में हजार/मिलियन बार बढ़ाया गया अतः डी.एन.ए. स्पष्ट रूप से दिखाई देने लगे तथा रोग के निदान में समर्थ हुए। इसीलिए डी.एन.ए. पर आधारित निदान यंत्र जैसे पी.सी.आर., जलजन्तु स्वास्थ्य की देखभाल तथा संगरोध में बहुत बड़ी भूमिका निभा सकते हैं।

शोध विशिष्टताएँ

झींगा कृषि के गंदे पानी के उपचार के लिए बैगास (गन्ने का उपउत्पाद) के उपयोग का अध्ययन यह दर्शाता है कि यह गंदे पानी से अमोनिया नाइट्रोजन, नाइट्राइट नाइट्रोजन तथा अन्य मेटाबोलाइट्स को निकालने में प्रभावकारी है।

सभा

केखाजपासं. द्वारा 10 जुलाई, 2001 को "विशेष मत्स्य कृषक दिवस समारोह" के उपलक्ष्य में मुत्तुकाडु प्रायोगिक केन्द्र पर एक कृषक सभा का आयोजन किया गया।

ए.जी.आर.आई.-आई.एन.टी.ई.एक्स. 2001 प्रदर्शनी में सहभागिता

संस्थान ने 31 जुलाई - 7 अगस्त, 2001 को कोयम्बत्तूर जिला लघु उद्योग मालिक संगठन व तमिलनाडु कृषि विश्वविद्यालय (कोयम्बत्तूर) के संयुक्त तत्वावधान में आयोजित ए.जी.आर.आई.-आई.एन.टी.ई.एक्स.-2001 प्रदर्शनी में भाग लिया।

प्रशिक्षण

* 17-26 जुलाई, 2001 को सीबास एल. केलकरिफेर के विशेष संदर्भ में "खारेपानी में पखमछली प्रजनन" तथा 1-10 अगस्त, 2001 को "झींगा रोग व स्वास्थ्य प्रबंधन" पर प्रशिक्षण कार्यक्रम आयोजित किए गए।
* केखाजपासं. मुख्यालय, चेन्नई व मुत्तुकाडु प्रायोगिक केन्द्र पर व्याख्यान तथा प्रदर्शन आयोजित किए गए।

भाकृअनुप अन्तरांचल खेल-कूद मुकाबला

के.मा.प्रौ.सं. कोचिन में 4-9 दिसम्बर, 2001 को आयोजित भाकृअनुप. अन्तरांचल खेल-कूद मुकाबले में श्री आर. मदिवाणन्, एस.एस.जी.आर.-I तथा श्री आर. कुमरेसन, एस.एस.जी.आर.-I ने दक्षिणांचल का प्रतिनिधित्व किया तथा कैरम में द्वितीय एवं लांग जंप में तृतीय पुरस्कार प्राप्त किया।

स्टाफ समाचार

सेवा-निवृत्ति

* श्री के. देवराजन्, प्रधान वैज्ञानिक 31-7-2001 को सेवा-निवृत्त हुए।

पदोन्नतियाँ

निम्न वरिष्ठ वैज्ञानिकों को 27-7-1998 से प्रधान वैज्ञानिकों के रूप में पदोन्नत किया गया :

डा. एल. एच. राव
डा. मात्यू एब्रहाम
श्री के. देवराजन्
श्री एस. आर. दास
डा. पी. रविचन्द्रन्
श्री एम. कदिरवेल्
डा. एस. कुलशेखरपांडियन्
डा. एस. एम. पिल्लै
डा. टी. सी. संतियागो
डा. ए. आर. तिरुनावुक्करसु
डा. एस. ए. अली
डा. के गोपिनाथन्
श्री आर. के. चक्रवर्ती
श्री एस. श्रीनिवासगम्
डा. सी. पी. रंगास्वामी
डा. बी. पी. गुप्ता
श्री डी. नारायणस्वामी
डा. एन. कलैमणि
डा. एम. नटराजन् तथा
डा. श्रीमती मुनावर सुल्ताना ।

* 27-7-1998 से डा. सी. गोपाल को वैज्ञानिक (वरिष्ठ वेतनमान) से वरिष्ठ वैज्ञानिक
* 27-7-1998 से श्री एस.वी. अलवांडि को वैज्ञानिक (वरिष्ठ वेतनमान) से वैज्ञानिक (एस.जी.)
* 25-3-1994 से डा. आज़ाद, आई.एस. को वैज्ञानिक से वैज्ञानिक (वरिष्ठ वेतनमान) तथा 25-3-1999 से वैज्ञानिक (वरिष्ठ वेतनमान) से वरिष्ठ वैज्ञानिक
* 5-10-1999 से डा. के. के. विजयन् को वैज्ञानिक (वरिष्ठ वेतनमान) से वरिष्ठ वैज्ञानिक
* 27-7-1998 से डा. टी. रविशंकर को वैज्ञानिक से वैज्ञानिक (वरिष्ठ वेतनमान)
* 27-7-1998 से श्रीमती डेबारल विमला को वैज्ञानिक से वैज्ञानिक (वरिष्ठ वेतनमान)