



Orchids

Newsletter



राष्ट्रीय आर्किड अनुसंधान केन्द्र
(भारतीय कृषि अनुसंधान परिषद्)
पाक्योग - 737 106, सिक्किम

National Research Centre for Orchids
(Indian Council of Agricultural Research)
Pakyong - 737 106, Sikkim, India

Vol. 13(1)

January - June, 2012

Contents

Research Highlights	01	Training Imparted	10
Institutional Activities	07	Education and Training	10
Republic Day Celebration	07	Distinguished Visitors	10
Participation of Scientists in Symposia/ Seminar etc.	09	Publications	10
		Appointments	11

RESEARCH HIGHLIGHTS

Molecular analysis for genotyping of *Cymbidium* orchids

19 *Cymbidium* species were used for the study. PCR amplifications using three nuclear (ITS) and ten plastid loci (microsatellite markers) showed significant variability. The amplified samples were stored for sequencing for development of molecular data base of *Cymbidium* orchids (Fig.1).

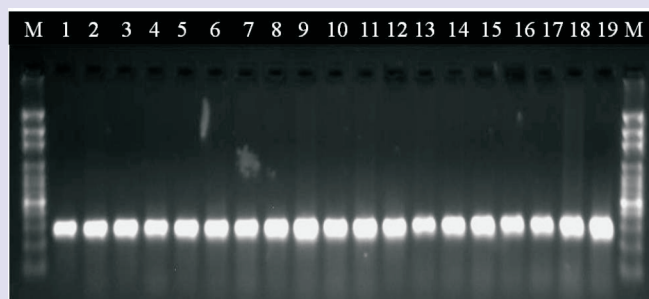


Fig 1. Banding pattern of *Cymbidium* species using forward and reverse primer accD1f5A and accD4rR5

Molecular analysis of *Dendrobium* orchids

610 accessions of 59 species of *Dendrobium* orchids were collected from different locations of India. A

set of 28 STM primers which were already optimized for two important parameters of PCR i.e. annealing temperature of primers and MgCl₂ conc. are used for PCR amplifications. The result of PCR amplifications of all the primers indicated a high potential to study the genetic variability of the *Dendrobium* species.

Germplasm collection

Five hybrids of *Paphiopedilum* namely *Paph.* Jolly Green Jem Mikkabi x Okayama Fresh 'Fast River', Pacific Ocean Ann x Small World, *Paph.* Helas Jim x Vallarow 'Red Challenge' *Paph.* Silhouette 'Chanson' and Startler x Hamana Emy x Teafor Twa were also procured during the reported period.

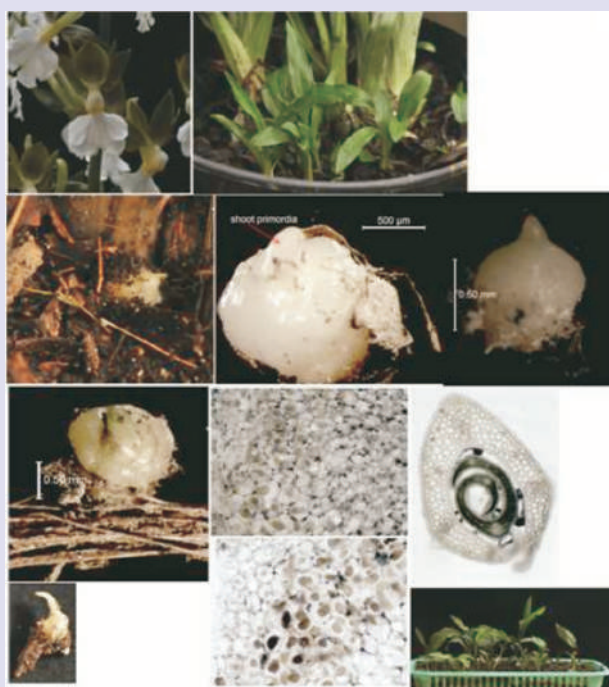
Four new records for orchids to West Bengal

During the field exploration in hitherto under or unexplored areas in Darjeeling district of West Bengal under orchid germplasm conservation programme several orchid species have been collected and conserved at National Research Centre

for Orchids, Darjeeling Campus, Darjeeling. Out of them, upon critical studies based on literature King and Pantling (1898), Pradhan (1979), Lucksum (2007) and Chowdhary (2010) and consultation of regional herbaria LBG and CAL, 4 species have been identified as *Calanthe mannii* Hook f. *Calanthe yucksomnensis* Lucksum, *Calanthe pachystylis* and *Cymbidium cyperifolium* Lindl., which were not recorded earlier from the state.

Natural seed germination in *Calanthe Yucksomnensis* S. Z. Lucksum under captivity

Seeds of orchids are small measuring less than a millimetre in length and less than a tenth to a quarter in widths. They also have little or no food reserves to carry out normal metabolism during germination. Hence, they require assistance of fungus to mobilize



certain nutrients and growth factors. Though the seed germination in *Calanthe* under captivity were achieved by spreading seeds around potting mixture when the role of the fungal associates were not known. The efforts resulted in development of a hybrid named *Calanthe Dominii* in 1852. The study

of fungal associates is not only essential for multiplication of a species but also for an accurate understanding of the ecology of orchids, improved management, and translocation opportunities for terrestrial orchids particularly with aim of rebuilding wild population. *Calanthe yucksomnensis* is a terrestrial orchid species first reported from Yucksom in Sikkim Himalaya. The species flowers in the month of March and April and seeds are matured and dehisce during August- October. A luxurious sapling growth of plantlets was observed in 2011 and it was comprehended that seeds may be germinating in the potting mixture. The plants were monitored for germinating seeds. The protocorms of various stages were found near the clump base and roots during the month of April – May. Histological observation from the roots of mature plants and protocorms suggested presence of mycorrhiza. No fungus association was observed in leaf primordial region. The isolation and identification of fungal associate may be useful in seed germination and hardening of tissue cultured plants.

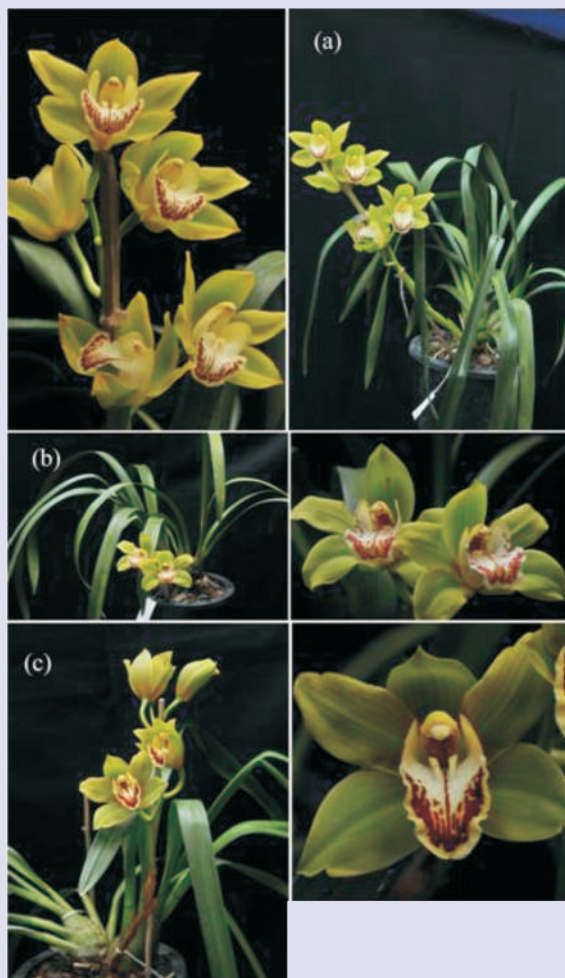
Collection, characterization, evaluation and conservation of orchids

Characterization of 28 species was done as per common descriptors .The characterized species were *Phalenopsis lobbii*, *Phiaus flavus* , *Tainia hookerianum*, *Phalenopsis mannii* , *Papilionanthe vandarum* , *Lycaste cruenta* , *Phiaus tankervilleae*, *Coelogyne nitida* , *Ascocentrum ampullaceum*, *Calanthe triplicata* , *Eria flava* , *Bulbophyllum odoratissimum* , *Eria pannea*, *Thunia marshalliana*, *Hygrochilus parishii*, *Ascocentrum ampullaceum*, var *auranticum*, *Kingidium taenialis* , *Thunia bracteata* , *Phreatia elegans*, *Liparis bistrata* , *Thelasis longifolia* , *Cleisostoma subulatum*, *Platanthera insectifera*, *Podochilus khasianus*,

Microvipera obtusa , *Bulbophyllum maculosum*,
Bulbophyllum ornatissimum, *Coelogyne flaccida*.

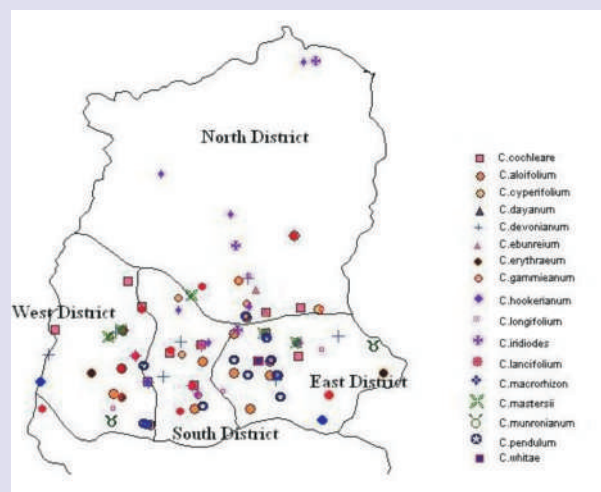
Development of new hybrids

Out of 500 progenies of a cross (*Cym* Sleeping Nymph 'Glacier' x *Cym lowianum*), the five flowered during the year. The colour of flowers was green with shades and faint lining of brown on sepals and petals. The flowers were semi round in shape and their number on floral spike varied from 2-5. Marked differences in lip banding pattern was observed. The three different lip banding pattern were present in F₁ population (Fig.). . Most of the progenies are expected to flower next year when it would be possible to make a selection of superior genotype from the progenies on the basis of their morphological desirable traits.



Mapping of area of *Cymbidium* species

The species diversity and richness was studied from point to point grid analysis option of DIVA GIS. Rainfall and altitude map of the state was prepared using BIOCLIM model of DIVA GIS. The results indicated that altitude and rainfall are the most important factors influencing the distribution of *Cymbidium* species. High richness was found in the areas located between 1500-2000 msl having annual precipitation between 1500-3000mm.



Development of complete protocol for mass multiplication of *Coelogyne cristata* through seed culture

In order to develop a protocol for mass multiplication for *Coelogyne cristata*, the seed pods were sterilized and culture was initiated on Gamborg B5, Murashige and Skoog, Knudson C & Nitsch media. Different parameters like swelling, globule formation, greening, germination were observed. Gamborg B5 media was found best for seed germination which took the least number of days for swelling (6days), globule formation (12days), greening (21days) and germination (41days). Shoot initiation was high in case of Gamborg media containing BAP 0.5mg/l whereas the lowest was in

Development of Protocol for Mass Multiplication of Rare Endangered and Threatened Orchid Species

To collect the planting material of *Renanthera imschootiana* & *Vanda coerulea* a trip was made to Mizoram and 16 plants of *Renanthera imschootiana* and 7 plants of *Vanda coerulea* were collected from Champhai District. The collected plants were planted in pots and grown under fibre house. The flowers *Renanthera imschootiana* were selfed for seed formation. Experiments on initiation of cultures using leaf and root tips are under progress.

Influence of frequency and mode of application of different growth regulators on *Dendrobium* hybrid 'Thongchai Gold'

Growth regulators like GA₃ (50,100,200 ppm) and BA (50, 100 ppm) were applied in two frequencies (morning and evening spray) and in two different modes like foliar spraying and drenching. Maximum flower spikes (2.08) and (2.0) were found in drenching and morning spraying of BA 50 ppm. Drenching of BA 100 ppm increased spike length (42.87cm), rachis length (30.31cm), spike girth (0.61 cm), and number of flowers per spike (15.25 cm). Overall flowering was found more in morning spray as compared to evening spray and drenching.

Influence of plant growth regulators and inorganic nutrients on flower regulation of *Dendrobium* hybrids 'Emma White'

Different treatments consisting of inorganic nutrients (NPK 20:20:20 and 30:30:30 along with Ca, Mg and Mn) and growth regulators (BA 10, 25, 50 ppm and GA₃ 50, 100, 200 ppm) and distilled water as control were experimented to regulate

flowering in winter season. Plants treated with NPK 20:20:20 with Ca, Mg and Mn along with BA 50 ppm and GA₃ 100 ppm increased number of spike (2.3), spike length (35.80cm), rachis length (22.7cm), number of flowers (11.1) and intermodal length (3.0). Whereas stalk length (4.25cm) and flower size (40.75cm) were found maximum in the plants treated with NPK 30:30:30 with Ca, Mg, Mn along with BA 25 ppm and GA₃ 50 ppm.

Evaluation and biochemical analysis of backbulbs and mature bulbs of *Cymbidium* hybrids

Out of sixteen hybrids of *Cymbidium* orchids, fourteen hybrids came into flowering. Maximum width (9.0cm) and length (14.5cm) of flowering pseudobulb and spike length (81cm) was recorded in 'Fire Storm Blaze'. Longevity of flowers on plants was recorded highest in 'Caripepper Peachy Keen'(70 days). In backbulbs, carbohydrate content ranged from 70mg/g in 'Pink' to 152 mg/g in 'Fire Storm Blaze' whereas in flowering pseudobulbs it varied from 88mg/g in 'Pink' to 180mg/g in 'Fire Storm Blaze'. Fire storm Blaze had maximum chlorophyll content (71.20 mg/g) followed by 'Hazel Fay Tangerine' (58.80mg/100g) and minimum in 'Red'(19.20mg/100g).

Role of weeds in virus transmission

Like any other crops, weeds are very common in orchids. Some of the weeds like *Drymaria cordata*, *Crassocephalum crepidiodes*, *Oxalis corniculata*, *Oxalis stricta* and two more unidentified weeds are observed very common on orchid plantation. These weeds were tested for the presence of CymMV and ORSV. Samples were processed and total viral RNA was isolated using Qiagen RNeasy mini isolation kit and RT-PCR was performed using virus specific

primers designed from CP gene sequences of CymMV and ORSV available in GenBank. RT-PCR results showed that these weeds are positive with CymMV and ORSV. Therefore, it is concluded that weeds are the hosts of CymMV and ORSV and can play important role in virus transmission.



Fig. 3a. Weeds growing with the orchids in pots

Molecular characterization of orchid anthracnose

Anthrachnose disease of orchids caused by *Colletotrichum gloeosporioides* is most destructive disease of orchids and is serious production constraint in orchid production in Sikkim. To confirm the pathogen 25 isolates from different hosts were collected from NRCO germplasm house and characterized on the basis of morphological and molecular basis. The culture of *C. gloeosporioides*

varies from white, grey, orange and pink colour. The polymerase chain reaction (PCR) assay with the *C. gloeosporioides* species specific primer (CgINT) with ITS4 yielded a single band of 450 bp and ITS1 and ITS4 primer combination amplified 560 bp products. Nucleotide sequencing of the ITS region of ribosomal DNA of all isolates showed 100% homology with *C. gloeosporioides* isolates available in GenBank. This showed that the pathogen involved with anthracnose disease of orchids is *Colletotrichum gloeosporioides*.

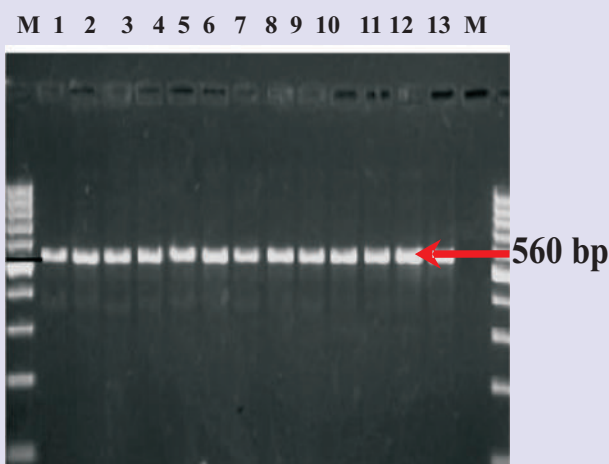


Fig. 4a. PCR using ITS 1 and ITS 4

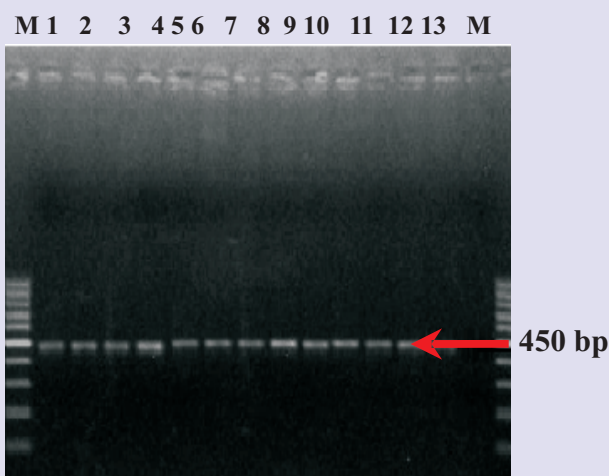


Fig. 4b. Species specific primer CgINT and ITS4

Studies on bacterial rot of orchids

The causal organism of bacterial rot was studied in detail. Three bacterial species were found associated

with the disease including saprophytic bacteria. All the three organisms were sequenced and pathogenicity test is being conducted for the confirmation of the actual pathogen.

Incidence of black spot of Aranda, Cattleya, Oncidium and Mokara hybrids

Hybrids of Aranda, *Cattleya*, *Oncidium*, *Mokara* and *Renanthera* imported from Thailand showed severe incidence of black spot disease on leaves and stem. The disease initially appears as small yellow or brown or black spots on the leaves. Under optimum environmental conditions the spots may enlarge and coalesce to form leaf blight. The disease also produces dieback symptoms in several hybrids. The incidence of the disease is ranging from 33 to 100%. The causal organism is tentatively identified as *Pestalotia* sp. The sample and culture of the fungus have been sent to Indian Type Culture Collection at Division of Plant Pathology, Indian Agricultural Research Institute, New Delhi for identification.



Fig. 5a. Black spot symptoms on Aranda hybrid

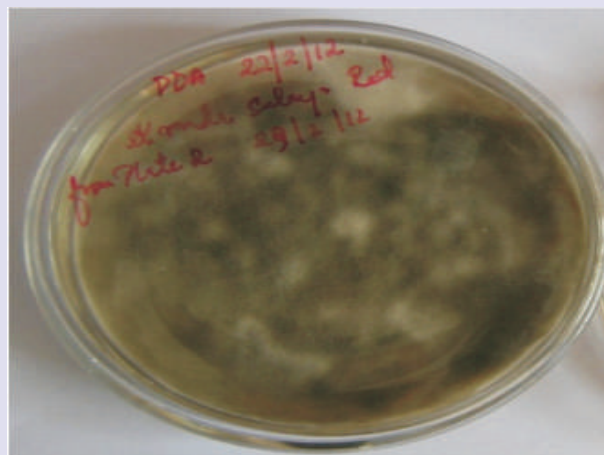


Fig. 5b. Fungal colony on PDA

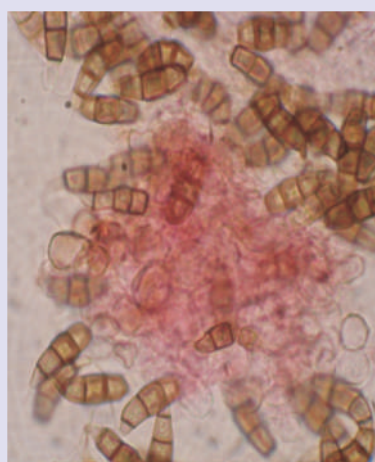


Fig. 5c. Conidia of *Pestalotia* sp

Field evaluation of efficacy of bio-pesticides against aphid on flowers of *Cymbidium*

Efficacy of botanicals and bio-pesticides were evaluated against aphids on the flowers of *Cymbidium* “H C Aurora”. Nine treatments (including control) viz., neem oil 0.03% EC (5 ml/lit.), garlic extract 5%, chilaune leaves extract (*Schima wallichii*) 10%, dhatura leaf extract 10%, econeem 3000 ppm 2ml/lit., titapat extract (*Artemisia*) 10%, tobacco extract 5%, and cow urine 50% were applied. The maximum percent reduction (78.12%) in aphid population was recorded in plants sprayed with econeem followed by neem oil (77.54%).

Relative efficacy of bio-pesticides against scale insect on *Cymbidium*

The efficacy of bio-pesticides was evaluated against scale insect infesting *Cymbidium* under polyhouse conditions. The nine treatments (including control) viz., neem oil 0.03% EC @ 5 ml/lit., econeem 3000ppm 2 ml/lit., chilaune leaf extract (*Schima wallichii*) 10%, dhatura leaf extract 10%, titapat extract (*Artemissia*) 10%, garlic extract 5%, tobacco extract 5%, and cow urine 50% were applied on four year old potted plants. The maximum reduction (92%) in scale insect population was recorded in econeem followed by neem oil.

Field evaluation of bio-pesticides for their efficacy against mite on orchid (*Dendrobium nobile*)

The efficacy of botanicals and bio pesticides was evaluated against mite under polyhouse conditions. Nine treatments (including control) viz., neem oil 0.03% EC 5ml/lit., garlic extract 5%, chilaune leaf extract (*Schima wallichii*) 10%, dhatura leaf extract 10%, B.t. (Dipel) 0.012%, titapat extract (*Artemissia*) 10%, tobacco extract 5%, and mycomite 3g/lit were applied on potted plants. The results showed that all the treatments were effective in reducing mite population over control. The maximum per cent reduction (74.30%) in mite population was recorded in neem oil treatment followed by mycomite (72.96).

Relative efficacy of bio-pesticides against shoot borer, *Peridaedala* sp. on *Dendrobium chrysotoxum*

Nine treatments viz., neem oil 0.03% EC 5ml/lit., garlic extract 5%, chilaune leaves extract (*Schima wallichii*) 10%, dhatura leaf extract 10%, B.t. (Dipel) 0.012%, tita pat extract (*Artemissia*) 10%,

tobacco extract 5%, and NPV 0.10% were applied on plants at fortnightly interval. The minimum (6.72%) shoot borer infestation was recorded on the plants treated with B.t. (Dipel) followed by neem oil (7.10%) and NPV (8.24%). at 7 days after second treatments.

INSTITUTIONAL ACTIVITIES



Celebration of Republic Day



3rd Krishi Mela (Farmer's Fair) on 7th March, 2012

The 3rd Krishi Mela (Farmers' Fair) of National Research Centre for Orchids (NRCO) was jointly organized with National Horticulture Board (NHB) on 7th March, 2012 at Pakyong of East Sikkim District for the benefit of Orchids Farmers/ Growers of Sikkim State. Shri Dawa Narbu Thakarpa, Hon'ble Minister of Agriculture, Govt of Sikkim,

inaugurated the Krishi Mela and graced the occasion as Chief guest. Shri Bhim Prasad Dhungel, Hon'ble Minister of Forest & Environment; Govt. of Sikkim was the guest of honour. Many dignitaries were present in this Mela such as Secretary, Agriculture; Principal Director (HCCDD); Dean, CA&PHT, CAU, Ranipool; and Centre-In-charge, NHB, Gangtok. In addition, numbers of State Agricultural Officers, Village Panchayet, NGOs' and farmers were also present. The progressive Orchids Growers from Kartok, Assam Lingzey, Dikling and Mirik also exhibited their produce. Approximately 300 farmers from different parts of Sikkim participated in the Krishi Mela.



First Stake Holders meeting of NRC for orchids on 26th March, 2012

The first stake holders meeting of NRC for orchids was held on 26th March, 2012 under the Chairmanship of Director, Dr. R. P. Medhi. Dean, CAU, Scientists of NRC for Orchids and ICAR research Complex for NEH Region, Sikkim Centre, Officials from other Central and State governments and R&D officials also attended the meeting.



Hindi Karyashala of NRC for orchids on 21st April, 2012

One day Hindi Karyashala (workshop) was organized in National Research Centre for Orchids, Pakyong Sikkim on 21st April 2012 to enhance the efficiency of Rajbhasha of the employees of the Centre. Shri Birendra Chhetri, Director, National Informatics Centre, Gangtok, Dr. B. S. Kholia, Scientist, Botanical Survey of India, Gangtok, Shri O. P. Singh and Shri Ghanshyam, Teachers, Jawahar Navodaya Vidyalaya, Pakyong participated as resource persons.



Training Programme on 'Production technology of Orchids' – 22-24th March, 2012 and 28-30th March, 2012

2 three days training programme on production technology of orchids sponsored by NHB were conducted at NRC for Orchids, Pakyong on 22-24th March and 28-30th March, 2012. A total of 60 farmers (30 farmers in each training) from east district of Sikkim participated in the training programme.



Three days training programme on
“Commercial Cultivation of Orchids”

A three days training programme on “Commercial Cultivation of Orchids” was conducted in ICAR Research Complex for NEH Region, Basar, Arunachal Pradesh under Horticulture Mission for North East and Himalayan States (HMNEH) from 26th to 28th April 2012 in collaboration of Krishi Vigyan Kendra (KVK), West Siang, ICAR, Arunachal Pradesh. A total 25 farmers from different villages were participated in this training programme and developed their skill and gained knowledge on cultivation of orchids.

Proceedings of 1st Meeting of 5th Research Advisory Committee Meeting

The 1st Meeting of 5th RAC (Research Advisory Committee) of NRC Orchids convened on 5th and 6th June 2012 at the Conference Hall under the Chairmanship of Dr. S.N. Puri, Vice Chancellor, CAU, Imphal, Manipur and with other members present were Dr. Umesh Srivastava, ADG (Hort. II), ICAR, N. Delhi; Prof. S. P. Viji, Ex Head Deptt. of Botany, Punjab University; Dr. U.C. Pradhan, Orchid Laboratories, Abhijit Villa, Kalimpong; Dr. R.D. Rawal, Former Principal Scientist, IIHR, Bangalore; Dr. R. P. Medhi, Member, Director, NRCO, Pakyong; Shri Padam Subba, Deputy

Director, HCCD Department, Govt. of Sikkim, Gangtok; Dr. J.G. Varshney, IMC, Joint Director ICAR Res. Complex for NEH region, Sikkim Centre; Dr. N. Pathak, Executive Director, Sikkim Himalaya Flora; Mr. Nirmal Yonzon, Progressive Orchid Grower, Pakyong, East Sikkim; Shri Tapan Katham, Progressive Farmer, Kathan Bari, Malbazar and Shri Sharad Pradhan, Progressive Farmer, Yangyang, South Sikkim. Member Secretary, Dr. L.C. De invited all the esteemed members of the RAC. The Director, NRC for Orchids formally welcomed the Chairman and other members of RAC. All the Scientists of the Institute also made their presentation of research activities in the meeting.

Participation of Scientists in Conference, Meetings, Workshop, Symposia, Seminar etc in India and Abroad.

North East Agri Fair 2012 at Khanapara, Guwahati from 10-12th February, 2012 and exhibited different *Cymbidium* hybrids and other technology of NRC for Orchids, Pakyong.

R. P. Pant and N. Sailo

SFAC meeting of BPD unit at NIRJAFT, Kolkata on 13th February, 2012

S. Chakrabarti

Hortiindia Workshop cum Exhibition on Innovative Production System in Horticulture at Institute of Horticulture Technology, Greater NOIDA, U.P. from 18-19th February, 2012 and exhibited different *Cymbidium* hybrids and other technology of NRCO in the form of posters

R. P. Pant and N. Sailo

National Consultation Meet on Nano Agriculture Mission at NASC Complex, New Delhi on 12th March, 2012.

R. P. Pant and M. Chakrabarti

Review meeting of “Horticulture Mission for North East and Himalayan States” MM-I on 28th March, 2012.

R. P. Medhi

Education and Training

Scientist

Winter school on “Molecular Approaches for Allele Mining and Crop Improvement” at Division of Genetics, IARI, New Delhi from 5-25th January, 2012.

S. Chakrabarti

Distinguished Visitors

1. Shri. Dawa Norbu Thakarpa, Hon'ble Agriculture Minister, Govt. of Sikkim. - 07/03/2012
2. Shri. Bhim Dhungel, Hon'ble Forest Minister, Govt. of Sikkim – 07/03/2012
3. Prof.(Dr.) S. Rajan, ADG (Hort.- I), ICAR, New Delhi - 14/03/2012.
4. Dr. M. G. Bhat, Director, Directorate of Cashew Research, Puttur, Karnataka - 21/04/2012.

Publications

Book Chapters

1. Pant., R. P., N. K. Meena and R. P. Medhi.

2012. *Emerging Diseases and Pests of Orchids and Their Management*. In: *Sikkim Biodiversity, Significance and Sustainability*. (Eds.) P. Tamang, A. K. Srivastava and Sonam Lepcha. Sikkim State Council of Science & Technology, Gangtok, Sikkim. Pp. 110 - 127.

2. Pant., R. P and R. P. Medhi., 2012. *Advances in orchid disease management*. In: *Biotechnological and Biorational Approaches for Pest and Disease Management*. (Eds). H. Kalita, K. Kishore, H. Rahman, and L. Chatlod, Biotech Books, Ansari Road, Darya Ganj, Delhi.

Research Paper

1. Barman., D., T. U. Bharati and R. P. Medhi. 2012. *Effect of media and nutrition on growth and flowering of Cymbidium hybrid H.C. Aurora*. Indian J. Hort. 69(3): 395-398.
2. Devadas., R., D. Barman and P. Khatiwara. 2012. *First generation interspecific hybrid of Cymbidium*. ICAR News, 18 (3): 8-9.
3. Medhi., R. P., M. Chakraborti and Rampal. 2012. *Orchid biodiversity of India: conservation and utilization*. Indian J. Genet., 72(2): 148-156.
4. Pant., R. P., M. Das, M. R. Khan, K. B. Pun and R. P. Medhi., 2012. *Association of an ectoparasite nematode- Helicotylenchus microcephalus Sher, with poor growth of Cymbidium hybrids in Sikkim*. Indian Phytopath. 65 (2): 196-197.

Popular articles

1. De., L. C. and R. P. Medhi. 2012. Cymbidium – A

Diversified High Value Orchid for North Eastern India. *Agrobios Newsletter*, **10**: 30-32.

Paper presented in Seminars/Symposia:

1. De., L. C. and R. P. Medhi. 2012. Biodiversity and conservation of rare and endemic orchids of north East India. In: *Proceeding of National Seminar on 'Plant Genetic Research for Eastern and North East India'* from 11-12th May, 2012 at ICAR Research Complex for NEH Region, Umiam, Meghalaya.
2. Pant., R. P. and R. P. Medhi. 2012. Virus disease and development of diagnostics in orchids. Pp: 64-78. In: *Proceeding of National Conference on Production of Quality seeds and Planting material: Health management in Horticultural Crops from 11-14th March, 2010 at New Delhi.*

Awards/Rewards/Recognition/Bodies acquired during the reported period

1. Dr. Mridul Chakraborti received the 'IJGPB-Best Research Paper Award' from Indian Society of Genetics and Plant Breeding (Certificate and Cash Prize of Rs. 10000) during National Seminar on 'Plant Genetic Research for Eastern and North-Eastern India' jointly organized by Indian Society of Genetics and Plant Breeding and ICAR Research Complex for NEH Region, Umiam, Meghalaya on 11–12th May, 2012.

Appointments

Scientist

Dr. Ramgopal Devadas transferred from Directorate of Soybean Research, Indore and joined this Centre as Sr. Scientist (Plant Breeding) against the st of Scientist (Plant Breeding) on 3rd May, 2012.

Published by

Dr. R. P. Medhi

Director

National Research Centre for Orchids (ICAR)

Pakyong 737 106, East Sikkim

Tel. No. 03592-257954

Telefax: 03592-257289 / 257282

Editorial Committee

Rampal

M. Chakraborti

N.G. Debnath