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ICAR-National Bureau of Agricultural Insect Resources

Molecular biology: the future of entomology

Molecular biology and genome studies over the last 30 years have unravelled the mystery of insect genetics with computational biology and bioinformatics skills that could be potentially tapped for insect pest management. The constant improvements in sequencing technologies and increased availability of insect genomic resources, which have opened an avenue for advanced research on comparative genomic approaches, molecular mechanisms of insecticide resistance, DNA barcodes, molecular phylogeny, gene silencing through RNAi, gene and genome editing, gene switching using nuclear receptors, gene drives and BioBricks, have eventually unravelled options for the development of novel alternative and beneficial pest control measures.

Various computer algorithms are used for the identification of structural elements (e.g., genes) within the genome, but to assign a function to each gene, researchers must search databases for genes with similar sequences (which are likely to be homologous, i.e. to have a common ancestor) for which a function has been established or proposed. With this information in hand, it becomes possible to compare insect genomes with one another and with those of other organisms. The results of these analyses can help in understanding the evolution of insects, including the phylogenetic relationships among different orders, and of the molecular underpinnings of insect-specific processes. This comparative genomics approach will not only help to elucidate basic processes involved in insect biology but can also lead to the identification of many insect- or pest-specific proteins that can be targeted for the development of insecticides. NBAIR has done whole genome sequencing of brinjal shoot and fruit borer, *Leucinodes orbonalis*, for the first time in the world, and has also analysed the whole transcriptomes of seven agriculturally important insects and the mitochondrial genome of EPN which enabled the annotation of the molecular mechanisms and pathways of important biological traits, including that of insecticide resistance.

A DNA barcode is a short sequence from standardised portions of the genome (648 bp of mtCOI). DNA barcoding is technically a simple and rapid approach in which a small DNA fragment is amplified by PCR from the total genomic DNA and then sequenced. DNA barcoding is a useful tool for species identification, as long as taxonomists are supported to discover and describe species that are subsequently identified by the use of DNA barcodes. At NBAIR, DNA barcoding for many insects and other arthropods such as spiders and mites, and entomopathogenic nematodes, has been carried out for the last five years. More than 3,000 species have been DNA barcoded so far.

Integrated taxonomy involving molecular tools (DNA barcodes) has enabled us in the identification of insects from body parts such as legs or antennae, and also from different metamorphic stages. NBAIR has used this technique to resolve many insect identity-related issues. Recently, an Asian subterranean termite species, *Coptotermes gestroi*, was identified as a contaminant in an imported packaging material through molecular tools.

Microgenomic identification systems, which permit species discrimination through the analysis of a small segment of genome, represent an extremely promising approach for the genetic identification of biological diversity at the species level. DNA barcoding is exceptionally useful for unambiguous identification of biological specimens and more efficient management of species diversity in GenBank. Insect genome analysis paves way to identify the underlying molecular mechanisms of insecticide resistance through gene expression studies (quantification of mRNA related to insecticide resistance) so as to identify novel targets with unique mode of action, which aid in insecticide resistance management and ultimately contribute towards effective management of insect pests.

Recent advancements in sequencing technologies have led to the generation of voluminous genomic information, which can provide a template to manipulate them for the

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development of novel pest management technologies. NBAIR has developed a Molecular Database of Indian Insects (MoDII) wherein all the information pertaining to insect molecular/genomic resources along with their biological parameters are available for researchers, students and farmers.

Gene silencing is one among the technologies which put into use RNAi-based approaches. RNAi-mediated silencing of different insect genes involved in various physiological processes was found to be detrimental to insects' growth, development and survival. At NBAIR, gene silencing research is being carried out for agriculturally important insects through externally funded projects.

Lately, genome editing (CRISPER/Cas9-based) approaches have been effectively utilised for pest management via direct as well as indirect methods by improving the efficiency of biocontrol agents. This revolutionary technology has ramified applications in pest management, and attempts have been made on different species of insects, including *Aedes aegypti*, where conditional sterility or pathogen-free insects can be developed.

Investment in terms of dedicated manpower and infrastructure for carrying out such high-tech research will improve the applications of molecular biology in insect pest management in future.

N. Bakthavatsalam
Director (Acting)

Research Highlights

New species of *Foenatopus*

A new species, *Foenatopus achterbergi* (Hymenoptera: Stephanidae) (Fig. 1), was described and illustrated from Karnataka. It was also compared with all the closely related species from the Oriental region. The genus *Foenatopus* is known from the Afrotropical, Neotropical, Oriental and Palaearctic regions.

Fig. 1: *Foenatopus achterbergi*

Redescription of *Gynaikothrips microchaetus*

Gynaikothrips microchaetus (Fig. 2) was described from Dharwad in Karnataka, and until now was known only from southern India. The species was illustrated and redescribed with additional features, based on the type material together with specimens freshly collected on leaves of *Ficus curtipes* in Manipur and Mizoram.

Fig. 2: *Gynaikothrips microchaetus*

Redescription of *Aduncothrips asiaticus*

The female of *Aduncothrips asiaticus* (Fig. 3) was redescribed along with the description of previously unknown male based on specimens collected on an agriculturally important new host plant, *Moringa oleifera*. Redescription included a detailed explanation of setal arrangement on head, thoracic area, number and arrangement of tibial spines, abdominal tergites and sternites, with illustrations which were lacking in the original description.

Fig. 3: *Aduncothrips asiaticus*

First record of *Rhene flavigera*

Rhene flavigera (Figs 4 & 5), a member of Salticidae, was recorded for the first time in Bengaluru and Bhubaneswar. This species can be distinguished from other members of the genus in having large copulatory openings in the epigyne, a pocket in the vicinity of epigastric furrow and spirally coiled distal part of internal canals. Variations in the epigyne of Indian species were also documented.

Fig. 4: Female *Rhene flavigera*

Fig. 5: Male *Rhene flavigera*

Predators for red spider mite management

A predatory mite, *Neoseiulus indicus*, and an anthocorid bug, *Blaptostethus pallescens*, were evaluated against the red spider mite in rose (Fig. 6). The bioagents reduced the mite population by 82% and 65%, respectively.

Fig. 6: Release of predators for red spider mite management

Biocontrol-based management of the fall armyworm

A field trial (Fig. 7) was laid out in farmers' fields at Manchanahalli village in Chikkaballapura district of Karnataka with installation of controlled-release fall armyworm pheromone traps, four releases of *Trichogramma pretiosum*, two sprays of neem oil and one spray each of *Bacillus thuringiensis* (NBAIR BT25) and *Metarhizium anisopliae* (NBAIR Ma-35) for managing the fall armyworm (Figs 8 & 9). At 60 DAT, there was 76% and 71.6% reduction in egg masses during kharif and rabi seasons, respectively.

Fig. 7: Layout of field trial

Fig. 8: Scientists and farmers displaying components of biocontrol-based field trial

Fig. 9: Scientist evaluating field trial

NBAIR pollinator garden as a refugium for the migratory blue tiger

Blue tiger, *Tirumala* sp., is one of the most abundant urban-dwelling butterflies recorded from India. This is also a known migratory species, migrating twice a year during the pre-monsoon (April–May) and post-monsoon (September–October) seasons. NBAIR maintains two patches of a pollinator garden with over 100 plant species belonging to diverse families. Recently, the migratory population of this butterfly was seen congregating in large numbers in the pollinator garden. Presence of large numbers of wild *Crotolaria* sp. and other flora, which are attractive to these migrating butterflies, should have contributed to the congregation in large numbers (Fig. 10). These butterflies are similar to the monarch butterfly (*Danaus plexippus*), which is known for transcontinental migration. The concept of establishing pollinator gardens amidst urban areas helps in conserving these butterflies as pollinators and also in enhancing the aesthetics.

Fig. 10: Congregation of *Tirumala* sp. in wild *Crotolaria* sp.

Hirsutella thompsonii to manage the broad mite in mulberry

Of late, mulberry has been under attack by the broad mite, *Polyphagotarsonemus latus*, in Karnataka, Tamil Nadu and Andhra Pradesh (Fig. 11). The infestation is widespread in Ramanagara, Mandya and Chamarajanagar districts of Karnataka, with losses in the range of 20–70%. In a series of field trials in Ramanagara district, the acaropathogenic fungus *Hirsutella thompsonii* [MF(Ag)66], sprayed at 1% concentration of a mycelial–conidial formulation, not only outperformed the chemicals used alongside in the experiments but also spread to a considerable distance, thus causing widespread epizootics within a week of treatment.

Fig. 11: Mulberry crop affected by the broad mite

Host range expansion of *Eocanthecona furcellata*

The native pentatomid predatory bug *Eocanthecona furcellata* was observed to have expanded its host range by predating on larvae of the fall armyworm. The female bug was found to be capable of feeding on 69–126 second to sixth instar larvae of the pest. However, the field performance of *E. furcellata* was restricted due to the presence of secondary parasitoids like *Gryon* sp., *Telenomus* sp. and *Trissolcus* sp.

Leaf whorls of *Dracaena* sp. as nesting sites for the leafcutter bee, *Megachile laticeps*

The leafcutter bee, *Megachile laticeps*, was observed to have constructed its nests inside the leaf whorls of *Dracaena* sp. (Fig. 12). The mother bee carrying a leaf bit was seen entering the leaf whorl where a partially lined nest was observed. The whorl occupancy rate of *M. laticeps* was 73.2% on an average. The bee constructed a single cell per leaf whorl and several leaf foraging trips were made to line as well as to cap the cell. The bee was found to be carrying copious amount of pollen in its abdominal scopa during the foraging trips. It made an inspection flight in a circular fashion twice lasting for about two minutes after making the final cap in its nest (Fig. 13). The bee made oblong and circular cuts on mature leaves of *Adenanthera pavonina* (Fig. 14), which were used in nest construction.

Fig. 12: *Megachile laticeps* constructing cell in leaf whorl of *Dracaena* sp.

Fig. 13: Capped nest cell

Fig. 14: Leaves of *Adenanthera pavonina* used for cell lining

Engineered β-cyclodextrinylated MEMS devices as sensors for pheromone of *Bactrocera oleae*

A novel sensor that allows the monitoring of *Bactrocera oleae*, an olive fruit pest, was developed. It facilitates the timely application of suitable remedies, before substantial onset of pest infestation. Covalently functionalised β-cyclodextrinylated MEMS devices were made for the selective and sensitive detection of the female sex pheromone of *B. oleae*. Two of the MEMS devices, with silicon dioxide surface-micromachined cantilever arrays and zinc oxide surface-microfabricated interdigitated circuits, were used to selectively capture the major pheromone component, 1,7-dioxaspiro[5,5]undecane (Fig. 15). The non-covalent capture of olive fruit fly pheromone inside the β-cyclodextrin cavity leads to the reduction of the resonance frequency of the cantilevers, whereas an increase in resistance was found in the case of zinc oxide-derived MEMS devices. Sensitivity of the MEMS devices to the olive fruit fly pheromone was found to be directly correlated with the increasing availability of β-cyclodextrin moieties over the surface of the devices and thus the detection limit of the devices was achieved to a value as low as 0.297 ppq of the olive fruit fly pheromone. The reversible nature and the capability of suitably functionalised MEMS devices to detect the presence of the female sex pheromone of *B. oleae*, before the substantial onset of pest infestation in an orchard, makes the technology quite attractive for viable commercial application.

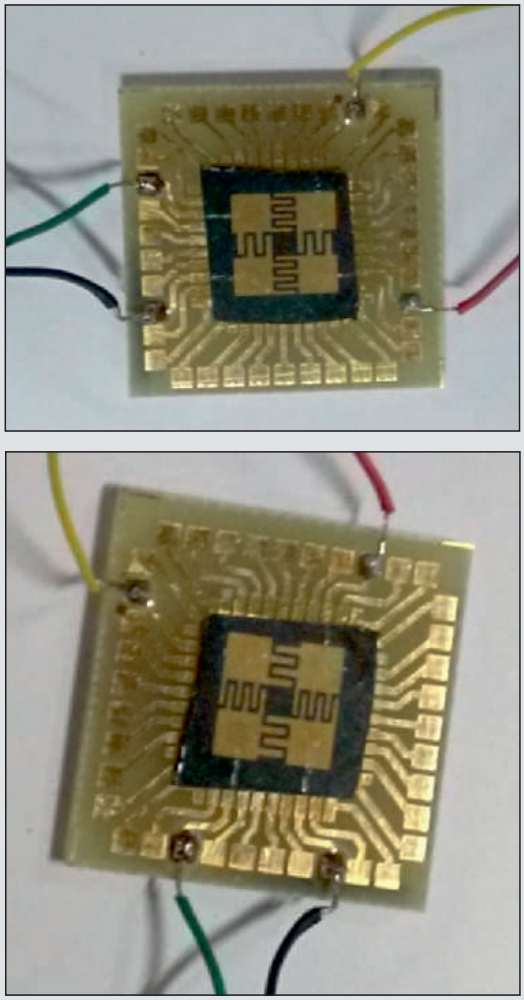


Fig. 15: Device fabrication: inter-digitated circuits

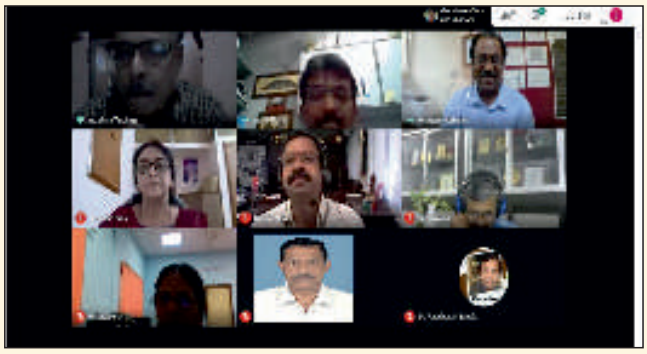
NBAIR celebrates National Moth Week 2020

NBAIR celebrated the "National Moth Week 2020". The National Insect Museum (NIM) of the bureau showcased the moth collections from across the country housed since decades. Hundreds of these moths were mounted delicately and preserved with utmost care and affection to depict the rich biodiversity. The collections were arranged family-wise and also according to the economic importance for the species which were associated with agriculturally important crops. The present moth collection is in a growing stage and the taxonomists aim to collect and accommodate many uncovered taxa from various parts of the country. NIM houses many of the enchanting species, the most beautiful and vivid atlas moth, *Attacus atlas*, one of the largest saturniid moths known; the enchanting and mesmerizing *Actias selene*, the Indian moon moth or Indian luna moth; the weird notodontid moth *Neostauropus alternus* and the confusing sphingid moth *Cephonodes hylas*. Dr Ankita Gupta and the museum staff coordinated the celebration of the event by preparing an informative and delightful video on glimpses of the moth repository in the museum added over the years with classic audiovisual inputs from Dr M. Sampath Kumar.

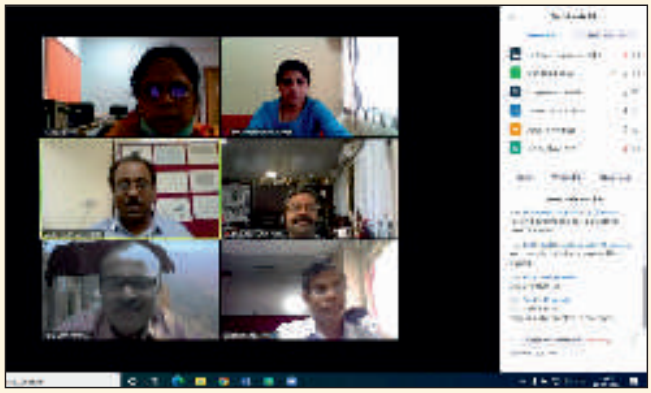
NBAIR organises Town Talk Series

NBAIR has created a platform titled "Town Talk Series" so as to host talks on wide-ranging topics from scientists, research managers, policymakers, research scholars and farmers.

Town Talk Series 001 was organised as a webinar on "Paradigm Shift in Plant Disease Management for the Millennium" with three invited talks on 24 July 2020. The programme started with the introductory remarks by Dr N. Bakthavatsalam, Director, NBAIR. He elaborated on the need to exchange ideas across the disciplines to generate novel ideas that could lead to innovations, and initiatives like Town Talk Series would address it. The first talk was by Dr B. Ramanujam, Principal Scientist and Head-in-Charge of Division of Germplasm Conservation and Utilisation, NBAIR, on 'My memoirs on crop protection research'. In his talk, he discussed the isolation, multiplication and bioefficacy of entomopathogenic fungi, and the need for registering them to benefit farmers. This was followed by a talk on 'Plant disease diagnostics and certification' by Dr R. Selvarajan, Principal Scientist, ICAR–National Research Centre for Banana, Tiruchirappalli. The diagnostic techniques developed in his laboratory to benefit banana farmers were briefed by him. Implementation of the diagnostics and certification programme in increasing banana production in the country was discussed. The third speaker was Dr Aundy Kumar, Principal Scientist, ICAR–Indian Agricultural Research Institute, New Delhi, who delivered a talk on 'New insights on rice phyllobiome'. He presented the role of microbes on the phylloplane, their diversity and the economic benefits that could be exploited for disease management in rice. A total of 63 participants from ICAR institutes, AICRP-BC centres and state agricultural universities participated in the webinar. Former Directors of the bureau Drs Abraham Verghese and Chandish R. Ballal participated in the discussion. Drs K. Subaharan, U. Amala, M. Mohan and M. Pratheepa organised the event.



Town Talk Series 002 as a webinar on "Making Smallholder Farming Climate Resilient" was organised on 14 August 2020. The webinar was chaired by Dr N. Bakthavatsalam, Director, NBAIR. In his introductory remarks, he stressed on the importance and impact of watershed development projects considering the current farming scenario. The guest speaker, Mr Crispino Lobo, Founder, Watershed Organisation Trust (WOTR), Pune, delivered a talk on 'Making Smallholder Farming Climate Resilient'. He elaborated upon the concept of water budgeting, water stewardship and the role of WOTR in transforming the lives of people living in villages across the country through participatory watershed development, ecosystem restoration and climate-resilient, sustainable agriculture with an action plan towards the concept of 'more crop per drop'. The webinar was organised by Drs K. Subaharan, U. Amala, M. Mohan and M. Pratheepa.

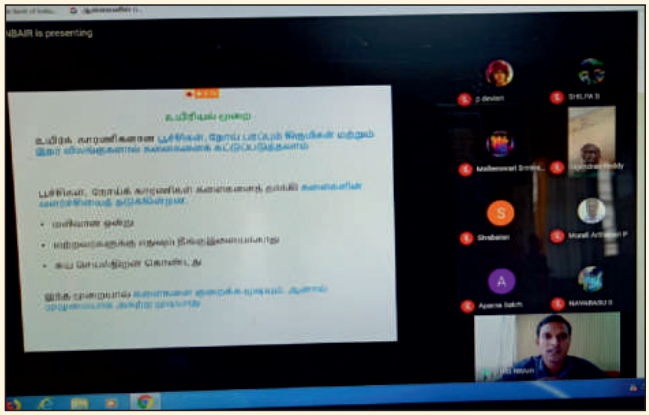


NBAIR in collaboration with The Alliance of Bioversity International and the International Center for Tropical Agriculture (CIAT), India, organised a webinar on "The Agrobiodiversity Index in India: Leading the Path for Other Countries" as part of **Town Talk Series 003** on 17 August 2020. Dr Natalia Estrada Carmona, Associate Scientist with The Alliance of Bioversity International and CIAT, Italy, delivered the lead talk. She elaborated on the skeleton of agrobioversity index (ABDI) with the three measurement categories, viz. market and consumption, production and genetic resources. She explained that the sub-indicators such as *ex situ* and *in situ* conservation of diversity, seed banks, species diversity, seed diversity and varietal diversity are the major parameters of genetic resources for the estimation of ABDI.

NBAIR organises webinars as part of Parthenium Awareness Week

The Society for Biocontrol Advancement and NBAIR jointly organised a webinar on "Biocontrol of Parthenium" as part of NBAIR's "Parthenium Awareness Week" on 21 August 2020. The webinar was chaired by Dr N. Bakthavatsalam, Director, NBAIR. He opined that adoption of a community-level approach will aid in containing the spread of the weed. Dr K. Dhileepan, Principal Entomologist, Biosecurity Queensland, Australia, delivered a talk on 'Biocontrol of parthenium'. The potential of biological agents in containing the parthenium menace was discussed by him. He called upon the Indian researchers to look for biocontrol agents attacking different parts of the parthenium plant as additional agents that would help in the control of the weed along with *Zygogramma bicolorata* in the long run. Former Directors of the bureau, Drs N.K. Krishna Kumar, Abraham Verghese and Chandish R. Ballal, also gave their remarks. Around 107 participants from ICAR institutes, AICRP-BC centres and universities attended the webinar. Drs K. Subaharan, G. Sivakumar, M. Sampath Kumar, M. Mohan, U. Amala and M. Pratheepa organised the event.

ICAR–Krishi Vigyan Kendra (KVK), Needamangalam in Tamilnadu, and NBAIR jointly organised a webinar on "Dimensions of Invasive Parthenium Weed Eradication" as part of the KVK's "Parthenium Awareness Week" on 21 August 2020. NBAIR scientists Drs Prakya Sreerama Kumar and M. Sampath Kumar participated in the programme. Dr Sreerama Kumar elaborated on the negative impact of parthenium weed on crops, human beings and animals. Dr Sampath Kumar enumerated the benefits and ecological advantages of using biological control, particularly *Z. bicolorata*, for parthenium weed management. A total of 54 participants, comprising mostly farmers and students, attended the webinar. The webinar was moderated by Drs M. Ramasubramanian and V. Radhakrishnan of ICAR–KVK, Needamangalam.



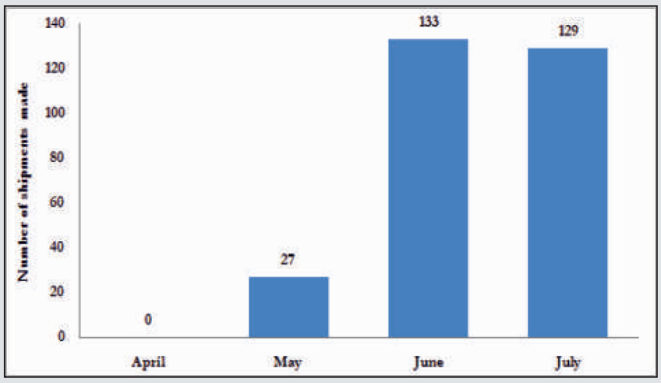
Independence Day at NBAIR

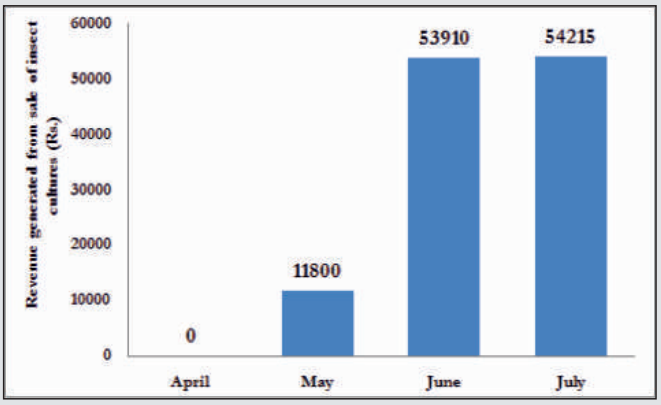
NBAIR celebrated the 74th "Independence Day" on 15 August 2020 with the hoisting of the national flag and singing of the national anthem by the staff at Hebbal. Addressing the staff members, Dr N. Bakthavatsalam, Director, NBAIR, urged the scientists of the bureau to work hard for the betterment of Indian agriculture. Dr T.M. Shivalingaswamy, farm in-charge, unfurled the national tricolour on the other campus of NBAIR at Yelahanka.

NBAIR organises Hindi Saptah Programme

NBAIR organised "Hindi Saptah Programme" during 14–19 September 2020. Dr N. Bakthavatsalam, Director, NBAIR, inaugurated the programme on 14 September 2020 by virtual mode and explained the importance of using Hindi language in official procedures. The message received on the celebration of *Hindi Saptah* from Mr Narendra Singh Tomar, Honourable Union Minister of Agriculture and Farmers' Welfare, was presented during the inauguration. Various competitions like elocution, essay writing, noting and singing were held and the winners were rewarded.

Bioagents Supplied During COVID Period





Awards and Recognitions

Dr T. Venkatesan

Received funding from the National Agricultural Science Fund (NASF) for the project entitled "Identification and validation of newer approaches for the management of whitefly *Bemisia tabaci* (Hemiptera: Aleyrodidae)" (Co-PIs: Drs Gandhi Gracy, K. Subaharan, R. Asokan[1], S. Subramanian[2], Vikas Jindal[3]).

[1]ICAR–Indian Institute of Horticultural Research, Bengaluru

[2]ICAR–Indian Agricultural Research Institute, New Delhi

[3]Punjab Agricultural University, Ludhiana

Dr Richa Varshney

Best Oral Presentation Award, National Conference on Agricultural Resource Management for Atmanirbhar Bharat, Central Agricultural University, Imphal, 17–19 July 2020.

Transfer of Technology

NBAIR transferred the technology "Waste to wealth: technology on black soldier fly-mediated bioconversion of farm and kitchen wastes" to Mr T.V. Anup and Abishek Reddy, entrepreneurs based in Vellore, Tamil Nadu, on 19 September 2020.

Selected Publications

Aditi, A., Venkatesan, T., Gracy, R.G., Ramya, R.S., Mohan, M. & Anil Rai. 2020. Transcriptome alterations of field evolved resistance in *Pectinophora gossypiella* against Bt Bollgard II cotton in India. *Journal of Applied Entomology,* https://doi.org/ 10.1111/jen.12805.

Gupta, A. & Gawas, S. 2020. A new species of the genus *Foenatopus* Smith (Hymenoptera: Stephanoidea: Stephanidae) from India. *Zootaxa,* 4801(2): 389–394.

Keerthi, M.C., Sravika, A., Mahesha, H.S., Gupta, A., Bhargavi, H.A. & Ahmed, S. 2020. Performance of the native predatory bug, *Eocanthecona furcellata* (Wolff) (Hemiptera: Pentatomidae), on the fall armyworm, *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae), and its limitation under field condition. *Egyptian Journal of Biological Pest Control*, 30: 69–75.

Sivakumar, G., Kannan, M., Ramesh Babu, S., Mohan, M., Sampath Kumar, M., Raveendran, P., Venkatesan, T., Rangeshwaran, R., Ballal, C.R. & Ram Kumar, P. 2020. Isolation and characterization of indigenous nucleopolyhedrovirus infecting fall armyworm *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae) in India. *Current Science*, 119(5): 860–864.

Varshney, R., Budhlakoti, N. & Ballal, C.R. 2020. Functional response of three species of predatory pirate bugs to different densities of blossom thrips, *Frankliniella schultzei* Trybom (Thysanoptera: Thripidae). *Current Science*, 118 (5): 827–833.

Veereshkumar, Kaushik, S.K., Rajarajan, K., Kumaranag, K.M., Uthappa, A., Sridhar, K.B., Badre Alam & Handa, A.K. 2020. Pollination biology of *Pongamia pinnata* (L.) Pierre: a potential biodiesel plant. *Genetic Resources and Crop Evolution*. https://doi.org/ 10.1007/s10722-020-01010-6.

Compiled and edited by: **Prakya Sreerama Kumar, U. Amala, R.S. Ramya, R.R. Rachana & N. Bakthavatsalam**
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