

Insecticide resistance management: reflections and way forward*

A century ago, in 1914, Alex L. Melander was the first to document the insecticide resistance in San Jose scale, *Quadraspidiotus perniciosus* (Comstock) to lime-sulphur sprays on deciduous fruits in USA. Since then numerous cases of resistance have been confirmed for every new class of insecticides, from cyclodienes of organochlorines, formamidines, pyrethroids, *Bacillus thuringiensis* (*Bt*), spinosyns and insect growth regulators to neonicotinoids after 2–20 years of use. In order to understand the aspects of insecticide resistance and its management (IRM), a brainstorming session was organized at IIHR. An idea of having brainstorming on IRM was mooted and supported by N. K. Krishna Kumar (Indian Council of Agricultural Research (ICAR), New Delhi). A total of 65 delegates from different ICAR institutes, State Agricultural Universities (SAUs), representatives from private sector, researchers and students participated in the deliberations.

K. R. Kranthi (Central Institute of Cotton Research, Nagpur) presented an overview of insecticide resistance in arthropods with special reference to cotton bollworms and *Bt* cotton. He spoke on the rationality behind the succession of transgenic cotton hybrids like Bollgard I, II, III and expressed concern that in spite of several research efforts on IRM, the ground reality remains grim with commercial interests of pesticide firms and dealers determining the use of insecticides. B. V. Patil (University of Agricultural Sciences (UAS), Raichur) emphasized the role of young researchers in educating farmers. He mentioned that next to paddy, vegetables such as chilli, brinjal and tomato receive more pesticides. Lack of knowledge on new molecules, mode of action, misuse/overuse of insecticides are the causes for emergence

of resistance in arthropods, especially in bollworms and sucking pests. He stated that new molecules with increased efficacy are available for managing resistance. T. Manjunatha Rao (IIHR, Bengaluru) mentioned that crop production and productivity are linked to insecticide use pattern. He stated that lack of knowledge on insecticide usage over a long time has led to the development of resistance. He requested the scientific community to come out with semi-popular publications in different languages that could help policy makers and farmers to understand and implement the management strategies.

Technical sessions began with a brief presentation by Irani Mukerjee (Indian Agricultural Research Institute (IARI), New Delhi) on 'Chemical interventions for management of insecticide resistance and residues'. New classes of pesticide molecules with different modes of action and improved safety profiles such as neonicotinoids, fipronil, chlorphenapyr, sulfluramid, spinosads, buprofezin, diafenthiuron, indoxacarb, metaflumazone, pymetrozine, spirotetramat, spiromesifen, fluopyram, ipconazole and flupicolide are available for management of pests. Further, selectivity is more important in terms of safety to applicators, livestock, pets, wildlife, environment and lower residues in food. She spoke on the role of the Central Insecticide Board and Registration Committee (CIB&RC) in approving pesticides and the risks involved due to pesticides.

T. Venkatesan (National Bureau of Agriculturally Important Insects (NBAIL), Bengaluru) spoke on the 'Investigations on field-evolved insecticide resistance in diamondback moth (DBM), *Plutella xylostella* (L) in India'. DBM has gained resistance to 36 insecticides in 14 countries and it has the ability to develop resistance to a new class of insecticides in a short time of exposure. *Plutella xylostella* collected across India exhibited high level of resistance to organophosphates – chlorpyrifos and acephate, pyrethroids – cypermethrin and a new group of insecticides – indoxacarb, spinosad and novaluron. Biochemical analysis revealed resistance was due to increased

expression of esterases, GSTs and microsomal P450. Further, endosymbionts play a major role in degrading insecticides. *Bacillus cereus* strain was isolated from the gut of DBM presumed to be responsible for resistance in DBM. P. N. Krishna Moorthy (IIHR) recommended neem for managing DBM, as there are no reports of resistance against this plant product.

M. Mani (IIHR) gave a presentation on 'Insecticide resistance and its management in mealybugs'. He mentioned that due to typical waxy body cover and clumped spatial distribution pattern, to many insecticides are ineffective. Insecticide resistance has been reported in few mealybug species. The mealybug, *Pseudococcus viburni* (Signoret) was resistant to DDT, parathion-methyl and chlorpyrifos. In USA, *P. maritimus* (Ehrh.) developed resistance to parathion. Resistance has also been reported in *Planococcus citri* (Risso), *P. kraunhiae* (Kuwana) and *Maconellicoccus hirsutus* (Green). In India, there is no systematic work on monitoring and detection of resistance in mealybugs. Efforts should be made to document resistance in field populations of mealybugs, especially *M. hirsutus*, *P. citri*, *Ferrisia virgata* (Cockerell), *Paracoccus maginatus* Williams and Granara de Willink and *Phenacoccus solenopsis* Tinsley.

Debi Sharma (IIHR) spoke about 'Insecticide residues in horticultural crops'. She mentioned that 700 pesticides are registered for use worldwide, of which 222 chemical pesticides are registered in India. About 4 million kg pesticides are consumed in India, of which 13–14% is used on fruits and vegetables. The most commonly used insecticides on fruits and vegetables in India are acephate, imidacloprid, acetamiprid, alphamethrin, chlorpyrifos and combination insecticides include acephate + fenvalerate, acephate + imidacloprid and alphamethrin + chlorpyrifos. Further, indiscriminate use of pesticides, non-observance of prescribed waiting periods, wrong advice and supply of pesticides to farmers by pesticide dealers, effluents from pesticide manufacturing units, wrong disposal of leftover pesticides,

*A report on a brainstorming session on 'Insecticide resistance management in horticultural crops – way forward' organized jointly by the Association for Advancement of Pest Management in Horticultural Ecosystems and the Indian Institute of Horticultural Research (IIHR), Hessaraghatta, Bangalore on 30 August 2014 at IIHR.

improper and incomplete cleaning of plant protection equipment are the important reasons for excess pesticide residues. She stated a project assigned by the Ministry of Agriculture, New Delhi in operation since 2005, alerts the respective state governments to monitor pesticide residues.

N. Srinivasa (UAS, Bengaluru) spoke on 'Resistance to acaricides – a challenge in horticultural system'. He stated introduction of newer varieties and newer practices of cultivation like protected cultivation have encouraged the multiplication of mites. Among mites, two-spotted spider mite, *Tetranychus urticae* Koch has developed resistance to the highest number of pesticides (92). Major resistance mechanisms involved are decreased absorption, increased excretion, target-site resistance, enhanced metabolic detoxification/degradation (by monooxygenases or esterases), conserved point mutation, etc. He suggested a pragmatic approach of critical timing of acaricide application aimed at an initial population, rotation or alternative use of acaricides with different modes of action, intelligent use of superior oils, petroleum-based horticultural mineral oils, agricultural soaps and add-on or resistant breaking compounds (synergists) in formulations can be useful for managing resistance in mites. He pointed out that petroleum-based mineral oils are widely used in Himachal Pradesh for the control of European red spider mites.

K. S. Mohan spoke on 'IRM for *Bt* crops in India – successes and lessons learnt'. He discussed on the sustainability and role of *Bt* crops in managing resistance in bollworms to *Bt* proteins. Three keys for effective IRM and to sustain any *Bt* crop are effective dose of *in planta* expressed *Bt* proteins, planting appropriate refuge and pyramiding of genes. He stressed the importance of a 'refuge' area which contains the non-*Bt* version of the host crop as it plays a pivotal role in maintaining a population of susceptible insects and maximizing the probability that rare, resistant survivors on *Bt* crop will mate with the large number of susceptible moths produced on the refuge. He explained the effectiveness of an IRM strategy by the inheritance of resistance traits and the initial frequency of resistant alleles prevalent in the target insect population. To delay resistance development, in 2002 the Genetic Engineering Appraisal Committee approved

commercial cultivation of *Bt* cotton (Bollgard®) with a requirement to plant 'refuge' crop of non-*Bt* cotton in a perimeter belt of five rows or 20% of total sown area, whichever is more. For insects like pink bollworm which feed only on cotton, it is crucially important to have non-*Bt* cotton as refuge. Further, an approach of blending *Bt* and non-*Bt* cotton seeds in a single bag is undergoing experimentation and this appears to be a robust field-viable option that needs to be encouraged and swiftly adopted.

G. M. V. Prasada Rao (Regional Agricultural Research Station, Guntur) made a presentation on 'Emerging trends in the cotton leafhopper, *Amrasca biguttula biguttula* on *Bt* cotton in Andhra Pradesh'. He mentioned that after the introduction of *Bt* cotton, leafhopper, *A. biguttula biguttula* (Ishida) has become a major production constraint on cotton. Seed treatment and indiscriminate foliar spray with neonicotinoids resulted in the development of resistance in cotton leafhopper. He explained that resistance monitoring studies conducted during the last three years indicated the development of resistance in Guntur population of *A. biguttula biguttula*. During 2013–14, the population developed 35.4-fold resistance against imidacloprid and 83.7-fold against thiamethoxam. Further, *Bt* cotton at this density is responding to higher nitrogen levels (25% more than the recommended dose of 120 kg N/ha) and producing significantly higher seed cotton yields. Higher number of plants per unit area coupled with higher nitrogen levels provide a congenial microclimate for unabated development of leafhoppers. He stressed that a systematic resistance monitoring, development and dissemination of effective management strategies against cotton leafhopper is required for achieving sustainable cotton production.

D. Lokeshwari (IIHR) gave a presentation on 'Multiple mutations on acetylcholinesterase gene associated with dimethoate resistance in the melon aphid, *Aphis gossypii* (Hemiptera: Aphididae)'. She mentioned that chemical control using organophosphates remains the main method of melon aphid control even though resistance has been documented worldwide. She stressed the importance of understanding resistance mechanisms and their early detection for resistance management in *A. gossypii*, a potential vector of more than 75 plant

pathogenic viruses. She pointed out that *A. gossypii* is capable of developing resistance to dimethoate under selection in the laboratory. As many insect pests have developed resistance through modifications of acetylcholinesterase resulting in modified acetylcholinesterase (MACE), investigations on the dimethoate selected *A. gossypii* laboratory populations revealed the existence of two consistent non-silent point mutations (single nucleotide polymorphism), viz. A302S (equivalent to A201 in *Torpedo californica*) and S431F (equivalent to F331 in *T. californica*) in the acetylcholinesterase (AChE) gene *Ace2*. Molecular diagnostics, PCR-RFLP using *SspI* and *HaeIII* restriction enzymes was utilized to detect these point mutations. An additional novel mutation, G221A was also revealed after sequencing *Ace2* (897 bp) gene in selected resistant strains. She strongly believed these point mutations located in the active site of the enzyme are responsible to alter the ligand specificity and confer resistance.

Gandhi Gracy (NBAIL) spoke on 'Exploring endosymbionts for insecticide resistance management in *Helicoverpa armigera*'. She stated that *H. armigera* collected from Tamil Nadu, Andhra Pradesh, Karnataka, Punjab, Gujarat and Maharashtra on *Bt*-cotton, pigeon pea, chickpea and tomato was tested for resistance to emamectin benzoate, spinosad, cypermethrin, cyhalothrin and thiodicarb insecticides. Resistance ratio of third instar larvae of *H. armigera* collected from the Amreli, Rajkot, Raichur, Godavari populations was high. Selection pressure played a major role in conferring resistance in *H. armigera* irrespective of the crop and genetic make-up. Enzyme analysis revealed increased expression of GSTs and AChEs. Analysis of endosymbionts diversity in different populations of *H. armigera* by culture-dependent method identified symbionts such as *Stenotrophomonas* sp., *S. maltophilia*, *Bacillus subtilis*, *B. amyloliquifaciens*, *Acinetobacter* sp. and *Enterococcus* sp. to play role in conferring resistance in these field-collected populations.

M. Mohan (NBAIL) delivered a talk on 'Natural enemies as components of IRM strategies'. He explained the utilization of biocontrol agents in the IRM programme. Resistance in natural enemies to pesticides is an open avenue of research and opportunities to bring chemical and biological control methods together. Development of resistant strains of natural

enemies is advantageous to integrated pest management (IPM)/IRM by preservation of natural enemies during field sprays. He mentioned that surveys for resistance in natural enemy populations in plots with insecticide application and studies to characterize the tolerance/resistance in natural enemies would help in effective IRM.

S. Sriram (IIHR) in his presentation on 'Fungicide resistance and management', mentioned the role of chemical fungicides in plant disease management and resistance development by fungal pathogens. He stated that multi-site target fungicides like mancozeb and chlorothalonil (protectants) pose low risk, while single site target fungicides like benzimidazoles and strobilurins (penetrants) into plant system pose high risk. Similarly, fungi like *Rhizoctonia* and *Sclerotium* pose low risk, while powdery mildews and oomycetes (downy mildews, *Phytophthora*) pose high risk. Use of single-site target fungicides for the management of powdery mildews poses maximum risk. Both qualitative resistance (sudden lose of control) resulting from single mutation in one gene and quantitative resistance (gradual reduction in control) due to mutation in several genes that interact with each other have

been reported. Resistance to sterol biosynthesis inhibitors used for control of powdery mildews like triadimefon, tenarimol, myclobutanil, tebuconazole and triflumizole was also reported. He explained a rare genetic mutation that alters the target site(s) in the fungus to block the action of the fungicide as the origin of fungicide resistance. Use of resistant varieties, canopy management, sanitation and use of protectant fungicide are required to delay resistance development.

V. Sridhar (IIHR) spoke on 'IRM in horticultural crops – current issues and research prioritization'. He mentioned that around 600 insect species have developed resistance to pesticides. Frequent application of insecticides under protected conditions makes the problem more complicated. Use of plant oils – neem and pongamia has significant influence in increasing efficacy of pesticides like acephate and imidacloprid for management of sucking pests. Effective resistance management demands interdisciplinary approach with scientists working in different fields starting from basic ecology to molecular biology. He suggested exploiting suitable synergists, integration of bio-intensive strategies within IPM for effective management of

insect pests. There is an urgent need to characterize detoxification mechanisms, exploit biosensor tools, assess role of endosymbionts and RNAi technology for effective IRM.

M. R. Hegde (IIHR) spoke on 'Sensitization of farmers about importance of IRM and residues management'. He explained inherent problems of Indian agriculture, viz. small and marginal holdings, poor purchasing power, lower education level, lower extension support and poor infrastructure. He stressed the importance of farmers' sensitization by conducting demonstrations, interaction meetings and awareness camps. He stated innovative steps such as commodity groups, farmers' field schools, techno agents/e-extension and progressive farmers will build an effective team to sensitize farming community and manage resistance in pests.

V. Sridhar, D. Lokeshwari*, K. R. Latha and A. K. Chakravarthy, Division of Entomology and Nematology, Indian Institute of Horticultural Research, Hessaraghatta Lake Post, Bangalore 560 089, India.

*e-mail: lokibiotech@gmail.com