

# **Insecticides Resistance in Insect Pests or Vectors and Development of Novel Strategies to Combat Its Evolution**

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## **Abstract**

The purpose of this presentation is to explore about insecticides resistance, in what way it occurs and how to manage this so that pesticides can continue to be used as crop management tools in the future. Resistance is an important concept to understand when attempting to manage a vector or pest that is the situation in which the insects are no longer killed by the standard dose of insecticide (they are no longer susceptible to the insecticide) or manage to avoid coming into contact with the insecticide. Insecticide resistance is an increasing problem faced by those persons who need insecticides to efficiently control medical, veterinary and agricultural insects. The development of resistance in the fields is influenced by various factors, and these are biological, genetic and operational issues. Biological factors are generation time, number of offspring per generation and migration. Genetic factors are frequency and dominance of the resistance gene, fitness of resistance genotype and number of different resistance alleles. These factors cannot be influenced by man; however, operational factors such as treatment, timing and dosage of insecticide application, persistence and insecticide chemistry, all are equitable. With cases of resistance on the rise and insecticide resources declining, it has become apparent that chemical pest control, as practiced today, may no longer be sustainable without the availability of specific strategies and tactics for the prevention or management of resistance. The emergence of insecticide resistance in insect populations is an evolutionary phenomenon and without taking actions to delay or minimize it now, pesticide management tactics currently used may someday no longer work. Generally recognized approaches to resistance management are grouped under three principal categories, first, low selection pressure supplemented by a strong component of non-chemical measures (management by moderation), second, elimination of the selective advantage of resistant individuals by increasing insecticide uptake through the use of attractants or by suppressing of detoxication enzymes through the use of synergists (management by saturation), and third, application of multi-directional selection by means of mixtures or rotations of unrelated insecticides or by use of chemicals with multi-site action (management by multiple attack). In such cases, the strategy chosen to resistance management must be based on thorough knowledge of the resistance implications of the candidate insecticides and the biology of resistant insect pests or vectors.

## **Keywords**

Agriculture, Insecticide, Pesticide Resistance, Public Health, Managing Resistance

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## **1. Introduction**

Worldwide, the major economic and environment losses due to the application of pesticides, impacts on public health,

livestock and livestock product losses have increased control expenses resulting from pesticide-related destruction of natural enemies and the development of pesticide resistance in pests. Moreover, crop and crop product losses; honeybee

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losses; crop pollination problems; bird, fish and other wildlife losses; and governmental expenditures to reduce the environmental and social costs of the recommended application of pesticides are further severe issues. Many of the major pest species are already resistant to members of each of the principal classes of insecticides. Furthermore, efforts to control resistant *Heliothus* spp., (corn ear worm) exert a cost on other crops when large, uncontrolled populations of *Heliothus* and other pests disperse onto other crops (Sarwar, 2013 a; 2013 b; Sarwar et al., 2011 a; 2009). In addition, the cotton aphid and the whitefly have been exploded as secondary cotton pests because of their resistance and their natural enemies exposure to high concentrations of insecticides (Sarwar, 2013 c; 2013 d; 2013 e; Khalid, et al., 2015).

A population of insect pests in a crop production field can be made up of different biotypes that are organisms of the same species, but has genetic variances. This natural genetic difference within a pest can allow some members of the species to survive of pesticide applications and may protect the organism from damage by the pesticide. The surviving members of that population become resistant to the pesticide. Thus, pesticide resistance is the natural ability of a biotype of an organism to survive exposure to a pesticide that would normally kill an individual of that species, which occurs with insects and other pests after repeated exposure to a single type of pesticide. This is because only the resistant organisms are left to reproduce with other resistant organisms, the new resistant biotype then becomes the dominant biotype of the pest population and the pesticide is no longer useful for managing that specific pest. Using one type of pesticide over and over again can produce resistance in a population of insects since the surviving biotype is nowadays resistant to the pesticide (Sukhoruchenko and Dolzhenko, 2008).

## 2. Propensity of Resistance

Today, insect pests are major threats to human health and agriculture, which are brought under control by pesticides; however, these are on the rebound. Resistance does not evolve at the same rate in every species or population, but may develop rapidly in some populations and slowly in others. The extensive use of pesticides has often resulted in the development and evolution of pesticide resistance in insect pests, plant pathogens and weeds. An early report by the United Nations Environmental Program (UNEP, 1979) suggests that pesticide resistance is ranked as one of the top 4 environmental problems of the world. About 520 insect and mite species, a total of nearly 150 plant pathogen species, and about 273 weeds species are now resistant to pesticides (Stuart, 2003).

When a resistant population occurs, the pesticide is no longer useful for managing that specific pest and other management options must be sought out. However, if pest resistance is managed effectively, a pesticide can remain useful for growers to continue using into the future. When a pest becomes resistant, the insecticide is used more frequently and the insecticide must ultimately be replaced as insect control diminishes. The development of insecticide resistance can be directly associated with genetic (frequency of R alleles, number of R alleles, dominance of R alleles, penetrance, expressivity, interactions of R alleles, past selection by other chemicals, extent of integration of R genome with fitness factors), biological and ecological factors (biotic- generation turn-over, offspring per generation, monogamy or polygamy, parthenogenesis; behavioural, isolation, mobility, migration, monophagy or polyphagy, fortuitous survival, refugia) and to operational practices (the chemical- chemical nature of pesticide, relationship to earlier used chemicals, persistence of residues, formulations; the application- application threshold, selection threshold, life stages selected, mode of application, space-limited selection, alternating selection) used to control them. The genetic factors that influence the development of resistance are probably beyond the control of most pest management programs. However, certain biological or ecological factors, especially those dealing with movement and untreated refugia, may be exploited to reduce the likelihood of resistance. Of all the factors influencing the development of resistance, the pest management specialist has the most control over the operational aspects (Rust, 1996; Sarwar, 2015 a; 2015 b; 2015 c; 2015 d; 2015 e; 2015 f; 2015 g; 2015 h).

## 3. Types of Resistance

There are three ways of looking at insecticide resistance, each of which is useful in a different context (World Health Organization, 2012):-

### 3.1. Molecular Genotyping of Resistance

This situation is the identification of the underlying genes that confer the inherited trait of resistance. Identification of a resistance gene provides evidence of the underlying evolutionary process. Depending on the type of resistance mechanism, this provides understanding of both the degree of resistance expressed in individual insects with the resistance gene and the frequency of such insects in the population.

### 3.2. Phenotypic Resistance

This is the basic expression of the genetic cause of resistance, shown by a insect's ability to resist and survive the effects of the insecticide. Phenotypic resistance is measured in a

susceptibility test of vector mortality when subjected to a standard dose of the insecticide. It is the development of an ability in a strain of insects, to tolerate doses of toxicants, which would prove lethal to the majority of individuals in a normal population of the same species. Phenotypic resistance is the phenomenon most commonly referred to in public health.

### 3.3. Resistance Leading to Control Failure

Resistance leading to control failure is the selection of heritable characteristics in insect population that results in repeated failure of an insecticide product to provide intended level of control when used as per recommendation. Resistance leading to control failure is the phenomenon most commonly referred to in agriculture. However, national dengue or malaria control programs should not wait for control failure to occur before implementing strategies to manage insecticide resistance. There is no acceptable level of control failure in public health and its waiting could result in delaying action until this is too late.

## 4. Types of Resistance Mechanisms

Normally, an insecticide penetrates rapidly through the insect's integument, reaching the site of action, and the site may be a vital enzyme, nerve tissue, or receptor protein. Insecticide molecules bind to the site and when they have attained threshold concentrations, they cause the insect's death. Resistance may be selected at each step of this pathway, the integument may be selected for lower permeability, thus reducing the rate of entry of the insecticide; new or more abundant metabolic enzymes may be selected, which break down the insecticide more efficiently; and altered target sites may be selected on which the insecticide no longer binds. Of these three types of mechanisms, metabolism and insensitivity at the site of action are the most important. A reduction in the rate of cuticular penetration aids both types of mechanisms in a synergistic way (Georghiou, 1994).

When an arthropod develops resistance to one insecticide, it has gene that may allow it to be resistant to another related insecticide. This is cross resistance, for example, insects that become resistant to one organophosphate tend to be resistant to all other organophosphates or have partial resistance to carbamates or pyrethroids. When an arthropod has more than one mechanism of resistance it is said to have multiple resistances, for example, the resistant insect's cuticle may be thicker to reduce insecticide's penetration than normal and same insect may also have specialized enzymes inside its body to break down an insecticide once it gets through the cuticle (Goodell et al., 2001). Resistance mechanisms can be

grouped into six categories, but target-site resistance and metabolic resistance being the primary focus of this document.

### 4.1. Target-Site Resistance

It occurs when the site of action of an insecticide (typically within the nervous system) is modified in resistant strains, such that the insecticide no longer binds effectively and the insect is therefore unaffected, or less affected by the insecticide. Resistance mutations known as knock-down resistance mutations, can affect acetylcholinesterase, which is the molecular target of organophosphates and carbamates, or may be voltage-gated sodium channels (Williamson et al., 1993).

### 4.2. Metabolic Resistance

Resistant insects may detoxify or destroy the toxin faster than susceptible insects, or quickly rid their bodies of the toxic molecules. Metabolic resistance is the most common mechanism and often presents the greatest challenge. Insects use their internal enzyme systems to break down insecticides. Resistant strains may possess higher levels or more efficient forms of these enzymes. In addition to being more efficient, these enzyme systems also may have a broad spectrum of activity (i.e., they can degrade many different insecticides). This resistance is related to the enzyme systems that all insects possess to detoxify foreign materials. It occurs when increased or modified activities of an enzyme system prevent the insecticide from reaching its intended site of action. The three main enzyme systems are esterases, mono-oxygenases and glutathione S-transferases, and while metabolic resistance is important for all four insecticide classes, different enzymes affect different classes. Metabolic and target site resistance can both occur in the same vector population and sometimes within the same individual mosquito. The two types of resistance appear to have different capacities to reduce the effectiveness of insecticide based on vector control interventions, with metabolic resistance being the stronger and more worrying mechanism (Insecticide Resistance Action Committee, 2011).

### 4.3. Behavioral Resistance

This is any modification in insect's behaviour that helps it to avoid the lethal effects of insecticides. Several publications have suggested the existence of behavioural resistance and described changes in vectors' feeding or resting behaviour to minimize contact with insecticides. Studies showed that *Anopheles farauti* vector stopped biting later in the night (23:00-03:00) and instead bit only in the earlier part of the evening, before humans are protected by sleeping in a sprayed room. In most cases, however, the resistant insects may detect or recognize a danger and avoid the toxin. This

mechanism of resistance has been reported for several classes of insecticides, including organochlorines, organophosphates, carbamates and pyrethroids. Insects may simply stop feeding if they come across certain insecticides, or leave the area where spraying occurred, for instance, they may move to the underside of a sprayed leaf, move deeper in the crop canopy or fly away from the target area, or may follow all these activities (Mouchet et al., 2008).

#### 4.4. Cuticular Resistance

Resistant insects may absorb the toxin more slowly than susceptible insects. Penetration resistance occurs when the insect's outer cuticle develops barriers which can slow absorption of the chemicals into their bodies. This can protect insects from a wide range of insecticides. Penetration resistance is frequently present along with other forms of resistance, and reduced penetration intensifies the effects of those other mechanisms. Generally, it is a reduced uptake of insecticide due to modifications in the insect's cuticle that prevent or slow the absorption or penetration of insecticides. Examples of reduced penetration mechanisms have suggested correlation between cuticle thickness and pyrethroid resistance in *A. funestus* due to two genes that encode cuticular proteins that are upregulated in pyrethroid-resistant mosquitoes. Behavioural and cuticular resistance mechanisms are rarer than the other mechanisms and are perceived by most experts to be a lesser threat than chemical resistance (Wood et al., 2010).

#### 4.5. Altered Target-Site Resistance

The site where the toxin usually binds in the insect becomes modified to reduce the insecticide's effects. This is the second most common mechanism of resistance. The site of action has been altered to decrease sensitivity to toxic attack. Alterations of amino acids responsible for insecticide binding at its site of action cause the insecticide to be less effective or even ineffective. The target of organophosphorus (malathion, fenitrothion) and carbamate (propoxur, sevin) insecticides is acetylcholinesterase in nerve synapses, and the targets of organochlorines and synthetic pyrethroids are the sodium channels of the nerve sheath.

#### 4.6. Physical Resistance Mechanism

The pickup or intake of toxic agent is slowed or reduced by modification to the insect skeleton, or the rate of excretion of the toxic compound is increased (Karaagac, 2012).

## 5. Strategies for Managing of Resistance

The objectives of resistance managing strategies are intended

to maintain the effectiveness of vector's control, despite the threat of resistance. Resistance management is not a novel concept, but, it has been used in agriculture and to address some public health situations over the past century. Several strategies have been used or proposed for managing resistance to insecticides for vector or pest control, including rotations of insecticides, combination of interventions, miscellany of spraying and use of mixtures. In certain settings, non-insecticidal tools, such as non-insecticide-based larviciding and environmental management, can also be used to reduce the overall mosquito population and limit the number and size of breeding sites without selecting for resistance. Integrated vector management (IVM), by reducing reliance on chemical control, can also be considered a mean of integrated resistance management (IRM) (Sarwar, 2013 e; Sarwar et al., 2003; 2011 b)

### 5.1. Thresholds for Susceptibility and Resistance

The test procedures for monitoring susceptibility and resistance of vectors or pests exposed to insecticides on the basis of testing for the mortality rates defines susceptibility that is an observation of more than or equal to 98% mortality rate among insects tested for resistance. Whereas, an initial observation of less than 98% insects mortality in bioassays conducted methods indicates possible resistance. Once this observation has been made, further testing is required to confirm resistance. Additional tests should be conducted to determine whether the vector's mortality rate is consistently lower than 98% and to understand the extent of resistance. Countries that are not in a position to conduct biochemical and molecular tests in the short term will have to make decisions on the basis of bio-assays only. The standard method of detection is to take sample of insects from the field and rear them to the next generations. Larvae or adults are tested for resistance by assessing their mortality after exposure to a range of doses of an insecticide. For susceptible and field populations, LD<sub>50</sub> or LC<sub>50</sub> values are calculated by using Probit analysis. The results are compared with those from standard susceptible populations (World Health Organization. 2012). In this presentation it is learned that pesticide resistance in a population of pest organisms results from using the same pesticide repeatedly. This enables individuals within a species with the natural or genetic ability to survive a pesticide exposure to reproduce and become the dominant biotype within a pest population. There are several ways that resistance can be managed. It is important to use pesticides only when needed, follow label directions, rotate pesticides and use pesticide alternatives so that these chemicals can remain a useful way for growers to manage pest organisms in crop production fields (Georghiou, 1994; Keith, 1996). The current available strategies for resistance control are:-

## 5.2. Rotations of Insecticides

Two, or preferably more, insecticides with different modes of action are rotated from one year to the next.

## 5.3. Combination of Interventions

Two or more insecticide based vector control interventions are used in a house (pyrethroids on nets and an insecticide of a different class on the walls), so that the same insect is likely, but not guaranteed, to come into contact with the second insecticide if it survives exposure to the first.

## 5.4. Mosaic Spraying

One compound is used in one geographic area and a different compound in neighbouring areas, the two being in different insecticide classes, but further research is required on use of mosaics.

## 5.5. Mixtures

Two or more compounds of different insecticide classes are mixed to make a single product or formulation, so that the insect is guaranteed to come into contact with the two classes at the same time. Mixtures are not currently available for vector control, but will become the future of IRM once they are available. In addition, synergists, which can considerably enhance the potency of an insecticide and could be used in combinations or mixtures, should continue to be investigated and rigorously tested for their usefulness.

## 5.6. General Approaches to Resistance Management

Three strategies for managing insecticide resistance have been proposed and implemented with varying levels of success.

### 5.6.1. Management by Moderation

Proponents of management by moderation recommend tactics that result in low selection pressure, conservation of susceptible insects, and supplemented by a strong component of non-chemical measures such as insect-resistant varieties, improved timing of planting and harvesting, and encouragement of biological controls. Management by moderation recognizes that susceptibility genes are a valuable resource and it attempts to preserve them by limiting the chemical selection pressure that is applied. Measures in this category include lower insecticide rates, infrequent applications, localized treatments, non-persistent chemicals, leaving some generations or sites untreated and preservation of refugia. This is why planting a refuge of susceptible corn along with Bt corn is important, so that some biotypes of the susceptible pest survive and mate with the resistant organisms, delaying the ability of the pest to

develop a resistant population.

### 5.6.2. Management by Saturation

This term does not imply saturation of the environment with pesticides, but it is intended to indicate saturation of the insect's defences by means of on-target dosages that are high enough to overcome resistance. It is elimination of the selective advantage of resistant individuals by saturation of defence mechanisms, increasing insecticide uptake through the use of attractants, rendering R genes functionally recessive through higher doses or by suppressing of detoxication enzymes through the use of synergists that can cancel out specific detoxication enzymes. Management by saturation proposes an aggressive approach by eliminating the presumably selective advantage of resistance, wherein pheromones or other semiochemicals and improved formulations are important elements in such a strategy.

### 5.6.3. Management by Multiple Attacks

Multi-directional and multi-site selection reduces degree of pressure leading to resistance. It is an application of multi-directional selection by means of mixtures or rotations of unrelated insecticides or by use of chemicals with multi-site action. The third strategy, management by multiple attacks, uses mixtures or rotations of different categories of insecticides or the use of insecticides that have several modes of action. Selection of the proper insecticides is of utmost importance. Factors such as previous exposure histories and the mechanism of resistance may affect the selection process.

### 5.6.4. Alternative Strategies to Manage Resistance

Alternative technologies such as non-chemical control methodologies can play an important role in management of insects having medical and veterinary importance or causing significant damage to crops. Alternative technologies and strategies such as physical or structural modifications (use of screens, caulking and sealing cracks or crevices in structures, and air vents to decrease moisture, use of polystyrene beads to reduce potential breeding sites, treatment of bed net, barriers), built-in pest control (use of organic dusts such as silica aerogel and boric acid to eliminate harbourage and breeding sites for insects), extreme cold temperature (use of cold temperatures, liquid nitrogen), heat (using carbon dioxide generated from dry ice or compressed gas cylinders), and anoxia (use of low oxygen atmospheres), may be extremely useful tools in combating the development of insecticide resistance. Toward the best of present knowledge these alternative treatments and technology might kill or mitigate both susceptible and resistant insects (Rust, 1996).

## 6. Integrated Pest Management (IPM)

Scientists and lawmakers are working towards pest control plans that are environmentally sound, effective and profitable. The best pesticide policies will reconcile environmental concerns with economic realities. Pests must be managed, farmers must survive economically and pesticides must be used as part of a planned systematic pest management program utilizing as many control techniques as applicable. This is called Integrated Pest Management (IPM), and controls such as biological, physical, cultural, genetic, environmental and mechanical techniques are just as important as pesticides. The IPM methods will continue to reduce our reliance on synthetic pesticides, and it has always implied that pesticides are one of many weapons in the pest control arsenal which includes genetics, biologic controls and plant production practices. The IPM has researched and real life success stories to keep it in the forefront of pest control, and it does not mean that IPM will not be redefined or adopt new methods as knowledge or new concepts in the area of pest management increases. The IPM methods with slightly different approaches or emphases will not eliminate the need for pesticides. The benefits of pesticides are real, and this reality will outlive the changeable winds of public opinion. Pesticides can give fast and adequate relief from pests. As the human population grows and farm acreage shrinks, food production efficiency cannot be jeopardized. We will need all of the tools at our disposal for food production, including pesticides (Keith, 1996).

Identified various strategies with the potential to delay resistance and practical successes in resistance management have relied primarily on reducing the number of insecticide treatments and diversifying the types of insecticide used. Resistance management requires more effective techniques for detecting resistance in its early stages of development. Pest resistance to a pesticide can be managed by reducing selection pressure by this pesticide on the pest population. In other words, this can be achieved by avoiding unnecessary pesticide applications, using non-chemical control techniques, and leaving untreated refuges where susceptible pests can survive. Adopting the integrated pest management (IPM) approach usually helps with resistance management. When pesticides are the sole or predominant method of pest control, resistance is commonly managed through pesticide rotation. This involves alternating among pesticide classes with different modes of action to delay the onset of or mitigate existing pest resistance. Pesticide manufacturers on product labelling may require that no more than a specified number of consecutive applications of a pesticide class be made before alternating to a different pesticide class. Tank

mixing pesticides are the combination of two or more pesticides with different modes of action in order to improve individual pesticide application results and delay the onset of or mitigate existing pest resistance. Organic farming system, avoids the use of man-made pesticides and growth hormones, and relies on crop rotation and biological pest control (Karaagac, 2012; Sarwar, 2012; 2013 f; 2013 g; 2014; Sarwar and Hamza, 2013; Sarwar and Sattar, 2016).

## 7. Conclusion

There are several thousand species of insects in the world of particular nuisance to man, either as vectors of fatal and debilitating diseases or destroyers of crops. In recent years, many of the resistance mechanisms have been detected and resistance detection methods have been developed. These mechanisms have been divided into various categories like increased metabolism to non-toxic products, decreased target site sensitivity, decreased rates of insecticide penetration, and increased rates of insecticide excretion. There are different methods to determine that the mechanisms are available in any given population and it can be seen the structure of the resistance mechanisms from these assays. Insecticide resistance management must be an integral part of all vectors and pests control programs. Using insecticides in such a way that their effectiveness is maintained, is a stewardship responsibility of the commercial companies that market them. It is also a stewardship duty of those who design and implement insect control programs. To successfully manage insecticide resistance in insects of urban or rural importance, it is necessary to consider the social and medical importance of the pest and to integrate the biology and ecology of the pest with the proposed control strategies. The ability to constantly monitor the pest population and the resistance levels as they relate to changes of susceptibility to the methodology of control is essential. Control procedures involving management by moderation should be used whenever possible. The success or failure of IPM programs and resistance management programs depends on the decision maker, the pest control personnel and the public. Management by moderation should be the basic approach and might be supplemented to the maximum possible by integrated pest management measures. Recourse to the other strategies described here will be essential in many cases, especially where high value crops or vectors of human disease are involved.

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