International Code of Conduct on the Distribution and Use of Pesticides

Guidelines on Prevention and Management of Pesticide Resistance

September 2012

The Inter-Organisation Programme for the Sound Management of Chemicals (IOMC) was established in 1995 following recommendations made by the 1992 UN Conference on Environment and Development to strengthen cooperation and increase international coordination in the field of chemical safety. The participating organizations are the Food and Agriculture Organization of the United Nations (FAO), the International Labour Organization (ILO), the Organisation for Economic Co-operation and Development (OECD), the United Nations Environment Programme (UNEP), the United Nations Industrial Development Organization (UNIDO), the United Nations Institute for Training and Research (UNITAR) and the World Health Organization (WHO). The World Bank and the United Nations Development Programme (UNDP) are observers. The purpose of the IOMC is to promote coordination of the policies and activities pursued by the participating organizations, jointly or separately, to achieve the sound management of chemicals in relation to human health and the environment.

This publication was developed in the IOMC context. The contents do not necessarily reflect the views or stated policies of individual IOMC participating organizations.

E-ISBN 978-92-5-107348-3 (PDF)

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Abbreviations

Bt	*Bacillus thuringiensis*
FAO	Food and Agriculture Organization of the United Nations
FRAC	Fungicide Resistance Action Committee
FRAG	Fungicide Resistance Action Group
GMO	Genetically Modified Organism
HRAC	Herbicide Resistance Action Committee
IRAC	Insecticide Resistance Action Committee
IRAG	Insecticide Resistance Action Group
IPM	Integrated Pest Management
kdr	knock-down resistance
MoA	Mode of Action
OCP	Onchocerciasis Control Programme
OP	Organophosphate
R individuals	Resistant individuals
RMP	Resistance Management Plan
RR	Homozygous resistant
RRAC	Rodenticide Resistance Action Committee
RS	Heterozygous for resistance
S individuals	Susceptible individuals
SS	Homozygous susceptible
WHO	World Health Organization

Definitions

Behavioural resistance – any modification in pest behaviour that helps the pest to avoid the lethal effects of pesticides. The pest organism is still sensitive to the pesticide and will be killed if exposed to a lethal dose. Consequently, those individuals evading exposure survive and reproduce. This may lead to the development of a behaviourally resistant strain.

Cross-resistance – when resistance to one pesticide confers resistance to another pesticide, even where the pest has not been exposed to the latter product. Cross-resistance occurs because two or more compounds are acting on the same target site and/or are affected by the same resistance mechanism. Cross-resistance develops most commonly with compounds having the same mode of action and that are usually, but not always, chemically related from the same chemical group. It may be complete or partial (if more than one mechanism is responsible for the resistance).

Diagnostic dose – a dose that is used to determine if the pests collected and tested were resistant to the point that field failures of control are possible.

Diploid – having two homologous chromosomes in pairs in the nucleus so that twice the haploid number is present, usually written as 2n.

Discriminating dose – a dose that is used to discriminate between resistant and susceptible individuals and is not directly related to field efficacy.

Haploid – having a single set of unpaired chromosomes in a somatic cell. Haploidy is a characteristic of sex and germ cells.

Metabolic resistance – resistance inferred by a metabolic process, e.g. in insects that are able to detoxify or break down the toxin faster than susceptible insects, or that quickly rid their bodies of the toxic molecules. Insects use their enzyme systems to break down insecticides and resistant strains may possess higher levels of these enzymes or of enzymes that are more efficient at detoxification. In addition to being more efficient, these enzyme systems also may have a broad spectrum of activity, i.e. they can degrade many different pesticides.

Mode of Action (MOA) – the biochemical process by which a pesticide disrupts normal pest biology usually resulting in the death of the pest. Normally this is a target binding site or a key biological process.

Multiple resistance – the simultaneous presence of several different resistance mechanisms in the same organism. The different resistance mechanisms may combine to provide resistance to multiple classes of pesticides. In the field, multiple resistance and cross-resistance may appear, but the former developed from separate selection events, while the latter is the result of shared resistance mechanisms.

Multisite compound – a compound that affects more than one target site. To become resistant, the organism would then need to develop resistance at more than one target site, which is more difficult than developing resistance to a **single site compound** that affects only one target site.

Penetration resistance – a resistance mechanism essentially limited to insects, in which the cuticle slows the penetration of the pesticide into the body of the pest. Penetration resistance is usually present along with other forms of resistance and the reduced penetration intensifies the effects of those other mechanisms.

Pesticide – any substance, or mixture of substances, or micro-organisms including viruses, intended for repelling, destroying or controlling any pest, including vectors of human or animal disease, nuisance pests, unwanted species of plants, or animals causing harm during or otherwise interfering with the production, processing, storage, transport, or marketing of food, agricultural commodities, wood and wood products or animal feeding stuffs, or which may be administered to animals for the control of insects, arachnids or other pests in or on their bodies. The term includes substances intended for use as insect or plant growth regulators; defoliants; desiccants; agents for setting, thinning or preventing the premature fall of fruit; and substances applied to crops either before or after harvest to protect the commodity from deterioration during storage and transport. The term also includes pesticide synergists and safeners, where they are integral to the satisfactory performance of the pesticide.

Resistance (technical) – a genetic change in an organism in response to selection by pesticides, which may impair control in the field.

Resistance (practical) – a heritable change in the sensitivity of a pest population that is reflected in the repeated failure (more than one instance) of a product to achieve the expected level of control when used according to the label recommendation for that pest species and where problems of product storage, application and unusual climatic or environmental conditions can be eliminated as causes of the failure.

Resistance mechanism – biological processes used by the pest to avoid the lethal action of the pesticide. Resistant organisms may have more than one resistance mechanism.

Resistance selection – the survival of resistant individuals in a population while susceptible individuals are killed by the pesticide treatment. The resistant individuals are "selected" to survive and produce resistant offspring. The net result is that continued use of the pesticide "selects" a pest population that becomes less and less susceptible to the pesticide. The selection process can be rapid, one or two seasons, or develop slowly over a number of years, depending on the pest, its exposure to the pesticide, and the genetics of resistance to a particular pesticide.

Guidelines on Prevention and Management of Pesticide Resistance

1 Introduction

1.1 Scope of the guidelines

These guidelines address the problem of pesticide resistance in agriculture and how to limit its development while continuing to protect crops from pests. The guidelines are intended for scientific, technical, and policy experts who prepare or evaluate pesticide resistance management plans, and for pesticide regulators who assess the risk of resistance development during registration of new pesticides or renewal of already approved products.

The guidelines are organized as follows:

- this chapter briefly outlines the problem and its causes, and identifies the objectives and challenges in managing pesticide resistance;
- chapter 2 describes the factors that affect the development of resistance and tells how to assess the risk, or likelihood, that resistance to a pesticide will develop;
- chapter 3 describes practices and strategies for preventing and managing resistance;
- chapter 4 explains how to detect and verify resistance in the field;
- chapter 5 describes how resistance has been prevented in Bt crops;
- chapter 6 briefly touches upon resistance in vectors of human diseases;
- annex 1 provides a list of further reading and online resources;
- annex 2 provides Internet links to real-life examples of resistance management plans.

These guidelines focus on the management of resistance to chemical pesticides in agriculture (including in transgenic crops), in insects, weeds, fungal diseases, and rodents. Although many of the principles described are also valid for other pesticide uses, such as in public health or forestry, these other uses are not addressed in detail, as guidance on pesticide resistance management for public health pests and disease vectors is available elsewhere.

1.2 The problem and its causes

Resistance is a genetically-based characteristic that allows an organism to survive exposure to a pesticide dose that would normally have killed it. Resistance genes occur naturally in individual pests because of genetic mutation and inheritance. They spread throughout pest populations due to a process of selection brought about by repeated pesticide use. Resistant populations develop because the resistant individuals survive and subsequently reproduce, and the trait for resistance is “selected” in the next generation, while the susceptible individuals are eliminated by the pesticide treatment. If the treatment continues, the percentage of selected survivors will increase and the susceptibility of the population will decline to a point that the pesticide no longer provides an acceptable level of control.

Certain pest control practices have consistently been shown to exacerbate the loss of susceptible pest populations and the development of resistance.

These include:

- continued and frequent use of a single pesticide or closely related pesticides on a pest population;
- the use of application rates that are below or above those recommended on the label;
- poor coverage of the area being treated;
- frequent treatment of organisms with large populations and short generation times;
- failure to incorporate non-pesticidal control practices when possible; and
- simultaneous treatment of larval and adult stages with single or related compounds.

In addition, failure to adhere to good farming practice such as crop rotation and cleaning of farm equipment, which helps prevent the spread of pest seeds and spores, can exacerbate the spread of resistance.

1.3 Objectives and challenges in resistance management

The objective of resistance management is to prevent or at least slow the accumulation of resistant individuals in pest populations, so as to preserve the effectiveness of available pesticides. Resistance management can also be thought of as susceptibility management, as the aim is to maintain a high percentage of susceptible genes within the pest population while keeping genes for resistance at a minimum. The challenge is to reduce the selection pressure for resistance while providing the necessary level of crop protection.

If the principle of resistance management is relatively simple, putting it into practice for a given crop or pest is often not. There is unfortunately no single resistance management prescription that can be applied globally to all pesticides, pests, and crops. Nor is resistance solely a technical problem that can be readily overcome with the right new pesticide with a new mode of action, or an adjustment in the way conventional pesticides are used.

Managing resistance requires: first, the use of rational pest control strategies based on the principles of integrated pest or vector management, which reduce pesticide use and hence the selection pressure for resistance; and second, the implementation of a comprehensive and tailor-made Resistance Management Plan (RMP) that is adapted to the pest, the crop and the region, and that forms an integral part of the Integrated Pest Management (IPM) strategy for the cropping system. A key principle of IPM is to use pesticides only when absolutely necessary, and to use alternative pest management techniques whenever possible. IPM therefore constitutes a fundamental approach to resistance management by minimizing the selection pressure that leads to resistance.

1.4 Farmer education

Socioeconomic and infrastructure factors affect the success of any resistance management plan. There is always a concern about the cost. For all pesticides, the preference is very nearly always for the least costly product. While it seems obvious that preventing resistance development is the best option, this is not always apparent to growers, particularly if the pesticide being used is relatively inexpensive and resistance takes a long time to develop. If it is clear that adherence to IPM and to a well-designed RMP will increase profitability, then farmers will be much more likely to follow these practices. But if no advantage to the programmes is observed or the recommendations cannot be afforded, the likelihood of implementation is low and the probability that resistance will develop is high. Grower education and information access are therefore critical to implementing any RMP. Growers must know what needs to be done in order to practice IPM and prevent resistance development, and why it is important. The information must be widely accessible and understandable to them.

2 Evaluating the risk of resistance

To effectively prevent and manage resistance, both the principles underlying the origin of resistance and the factors that can influence its development and spread need to be understood.

2.1 Resistance fundamentals

What is resistance?

Resistance is defined as a genetic change in an organism in response to selection by toxicants. The development of resistance does not automatically lead to impairment of pest control. For instance, low levels of resistance may be observed in the laboratory without immediate problems arising in the field. However, if prevention is to be encouraged, resistance should be detected and addressed in an early stage, before pest control failure occurs in the field.

When pest control fails in the field due to pesticide resistance, we speak of “practical resistance.” This is a heritable change in the sensitivity of a pest population that is reflected in the repeated failure (more than one instance) of a pesticide to achieve the expected level of control when used according to the label recommendation for that pest species and where problems of product storage, application, and unusual climatic or environmental conditions can be eliminated as causes of the failure. This second definition is therefore narrower than the first, and though it is only at this stage that economic problems due to resistance may arise, it may be late for setting up resistance management measures.

Genetic basis of resistance

Resistance occurs when naturally occurring genetic mutations allow a small proportion of the population to resist and survive the effects of the pesticide. If this advantage is maintained by continually using the same pesticide, the resistant organisms will reproduce and the genetic changes that cause resistance will be transferred from parents to offspring. Through this “selection process,” resistant organisms eventually become numerous and control by the pesticide may fail (Figure 1). Resistance should not be confused with the tolerance that can occur after sub-lethal exposure to insecticides but is not genetically passed on to offspring.

Resistance development is a genetic process. The characteristic or “trait” which confers resistance is contained in one or more genes. A gene is a portion of a chromosome in the organism’s cell. When individuals reproduce, they pass along unique combinations of genes to their offspring. An allele is one of two or more varieties of a gene. For instance, one allele may be the resistant trait (R); the other the susceptible one (S).

Most multicellular organisms have two sets of chromosomes, that is, they are diploid. Diploid organisms have one copy of each gene (and therefore one allele) on each chromosome. If both alleles are the same, they are homozygotes. If the alleles are different, they are heterozygotes.

Some organisms (e.g. many fungi, during the vegetative part of their life cycle) are haploid; they have a single set of unpaired chromosomes.

Resistance alleles can range from dominant through semi-dominant to recessive. If dominant or semi-dominant, only one parent need possess the characteristic for it to be fully or partially expressed in the offspring. If recessive, both parents must possess the trait. Fortunately, most resistance mechanisms are controlled by recessive or semi-dominant alleles, which slows their spread within the population.

The genetic trait that allows the organism to survive exposure to the pesticide will be found in one or both of the gene's alleles. When the trait is in both alleles (written RR), the pest is homozygous resistant; the pest will likely be highly resistant to the pesticide and will pass on one resistant allele (R) to its offspring. If the offspring also receive an R from their other parent, they too will be RR. If the trait for resistance is found in just one of the gene's alleles (RS), the pest is heterozygous resistant; the pest will be less resistant to the pesticide, and may or may not pass on the gene for resistance to its offspring. Individuals that are homozygous susceptible, SS, are susceptible to the pesticide.

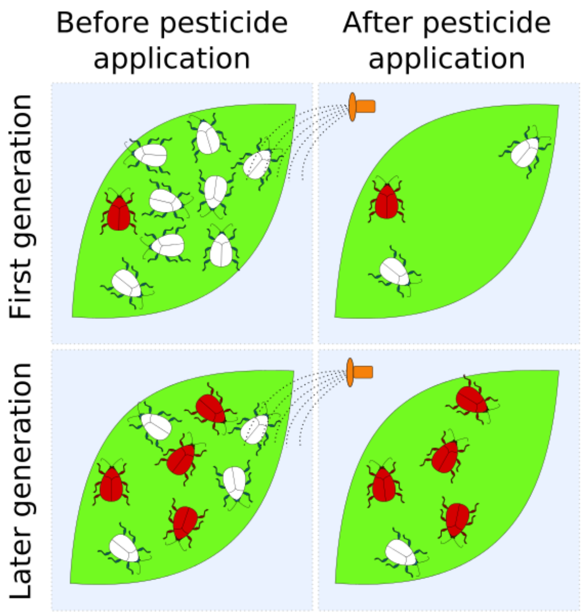

Figure 1 Pesticide applications can select for resistant pests. In this figure, the first generation happens to have an insect with a heightened resistance to a pesticide (red). After pesticide application, its descendants represent a larger proportion of the population because susceptible pests (white) have been selectively killed. After repeated applications, resistant pests may comprise the majority of the population (source: Wikipedia; 11 Jan 2012)

Populations of organisms that have never been exposed to pesticides are usually fully susceptible, and resistance genes within those populations are very rare. This is generally due to a "fitness cost," which means that organisms which are resistant lack some other attribute or quality which has been "traded off' against the resistance trait. A reduction in fecundity or overall robustness may for instance be found in resistant organisms. Because of the fitness cost, resistant organisms are at a disadvantage compared to susceptible organisms once the pesticide is removed. Susceptible organisms will then have the selective advantage and the pest population will, in principle, revert to its susceptible state.

This reversion to susceptibility is the underlying assumption behind resistance management. However, reversion rates are variable and may be very slow, particularly when a pesticide has been used for many years. This is one of the reasons why prevention of resistance development is better than attempting to “cure” resistance after it has developed.

2.2 Resistance mechanisms

Agricultural pests use a variety of mechanisms to survive exposure to toxicants. Resistance can develop more easily when two or more of these mechanisms are used at the same time. The resistance mechanisms fall into the following general categories:

Metabolic detoxification (enzymatic)

Resistance through metabolic detoxification is most often found in insects and is less common in weeds and pathogens. It is based on enzyme systems that insects have developed to detoxify naturally occurring toxins found in their host plants and in the blood ingested by blood feeding insects. These systems include esterases, cytochrome P450 mono-oxygenases, and glutathione S-transferases. Resistant insects may have elevated levels of a particular enzyme or altered forms of the enzyme that metabolize the pesticide at a much faster rate than the non-altered form. In either case the resistant insect can detoxify the pesticide before the pesticide kills it.

Metabolic resistance can range from compound specific resistance to very general resistance to a broad range of compounds. Similarly, the level of resistance provided to the insect can range from very low to very high, and can vary from compound to compound. This mechanism often cleaves the pesticide molecule or adds molecules to the pesticide, e.g. glutathione transferase, which detoxifies the compound.

Enhanced metabolism is also a common resistance mechanism in weeds. For example, enhanced rates of metabolism of acetyl-CoA carboxylase (ACCase), acetolactate synthase (ALS), and photosystem 2 (PS2) herbicides have been reported.

Reduced sensitivity at the target site

With this mechanism the binding site of the pesticide is changed so that it cannot effectively bind to the target site, thus eliminating or significantly reducing the pesticide’s effectiveness. This is the most common mechanism in fungi and weeds, and is also very common in insects. There are four general categories of target site resistance in insects:

- *kdr* (knock-down resistance) interferes with the sodium channel in nerve cells. This is a common mechanism used for resistance to DDT and pyrethroids, e.g. in *Anopheles gambiae*, *Blattella germanica.* There are several mutations that produce *kdr* and super *kdr*.
- *MACE* (modified acetylcholinesterase) modifies the structure of acetylcholinesterase so that it is no longer affected by the insecticide. This is, for example, the mechanism for pirimicarb resistance in *Phorodon humuli* and is responsible for resistance in *Tetranychus urticae.*
- *Rdl* (resistance to dieldrin) is a point mutation that reduces dieldrin binding at the GABA receptor. It is responsible for dieldrin resistance in *Anopheles quadrimaculatus* mosquitoes and in *Lucilia cuprina*, the sheep blowfly.
- *Bt* resistance occurs through loss of cadherin, which has important roles in cell adhesion, ensuring that cells within tissues are bound together. This mechanism is found, for instance, in *Bt*-resistant diamondback moth (*Plutella xylostella)*

There are many examples of target site resistance in weeds. The most important of these include:

- *ALS* (Acetolactate synthase) inhibitors, which cause a change in target site enzyme ALS
- *ACCase* (Acetyl-CoA carboxylase) inhibitors
- *PS2* (Photosystem 2) inhibitors

Reduced penetration

This mechanism slows the penetration of the pesticide through the cuticle of resistant insects. Alone, this mechanism produces only low levels of resistance. However, by slowing the penetration of the toxicant through the cuticle it can greatly enhance the impact of other resistance mechanisms. For example, an insect without any penetration resistance might be 25-fold resistant, whereas if penetration of the pesticide were reduced two-fold then the overall resistance could be nearly 50-fold.

Sequestration

In plants, the pesticide is removed from sensitive parts of the organism to a tolerant site, such as a vacuole, where it is effectively harmless to the target organism. This type of resistance has been demonstrated for the herbicides glyphosate, paraquat and 2,4-D. In insects (aphids, *Culex* mosquitoes, etc.) metabolic enzymes are significantly amplified (up to 15% of the total body protein) and bind to the insecticide but the insecticide is not metabolised, i.e. the insecticide is sequestered.

Behavioural resistance

Behavioural resistance is limited to insects, mites and rodents. It refers to any modification in the organism's behaviour that helps to avoid the lethal effects of pesticides. 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 into the crop canopy, or fly away from the target area). Behavioural resistance has also been reported in mice.

Behavioural resistance does not have the same importance as the physiological resistance mechanisms mentioned above but can be considered to be a contributing factor, leading to the avoidance of lethal doses of a pesticide.

2.3 Key factors in resistance development

The risk of resistance development is quite variable among and within pesticide groups and pest species but is particularly high for many of today's selective pesticides with specific modes of action. In general, pesticides with a single target site that are applied numerous times to a large population of pests with a high population turnover will be more at risk of resistance development than pesticides that attack several target sites and are used less frequently on a pest that has a low population turnover. In the first situation the selection pressure would be very high; in the latter it would be much lower. That being said, resistance does not always develop as predicted.

For current pesticides a considerable amount of information related to resistance on a variety of crops and pests is often available. This information can be used to estimate the risk of resistance development for new uses and uses in new geographic locations. For new pesticides, however, especially if they represent new chemical groups, assessing the risk of resistance development is more difficult. Experience with similar chemistry and target pests, as well as the mode of action of the compound, will provide some insight. However, there is still a lot to be learned. At present it is really only possible to estimate whether the risk of resistance development is low, medium, or high.

The factors that affect resistance development can be grouped into three categories: the pest's genetic make-up, the pest's biology, and "operational factors" including cropping practices and the pesticide characteristics and application (see Table 1). While it is not possible to precisely predict the development of resistance to a particular compound, it is possible to assess the risk generally by evaluating these factors for each pesticide-pest-crop situation. That is why it is critical to gather as much information as possible on the biology of the pest, the characteristics of the compound, the use of the compound, and the specific situation in which the compound will be used. Similarities will exist between compounds, pests, and uses but each situation will be different. Taking all these factors into consideration when designing a resistance management programme will go a long way toward ensuring its success.

Table 1 Biological, genetic, and operational factors in resistance development.

Factor	Potential for resistance development	
	Lower	Higher
Biological factors		
Population size	Small	Large
Reproductive potential	Low	High
Generation turnover	One or less generations per year	Many generations per year
Type of reproduction	Sexual	Asexual
Dispersal	Little	Much
Seed bank	Large	Small or none
Pesticide metabolism	Difficult	Easy
Number of target sites of the pesticide	Multiple sites	Single, specific
Pest host range	Narrow	Wide
Genetic factors		
Occurrence of resistance genes	Absent	Present
Number of resistance mechanisms	One	Several
Gene frequency	Low	High
Dominance of resistance genes	Recessive	Dominant
Fitness of "R" individuals	Poor	Good
Protection provided by the "R" gene	Poor	Good
Cross resistance	Negative or none	Positive
Past selection	None	Significant
Modifying genes	Absent	Present
Operational factors		
Activity spectrum of the pesticide	Narrow spectrum	Broad spectrum

Factor	Potential for resistance development	
	Lower	**Higher**
Pesticide application rate	Label rate; heterozygotes killed (If R gene is incompletely dominant)	Less than label rate: heterozygotes survive More than label rate: Only some homozygous resistant individuals survive and reproduce (especially if there is little immigration)
Application coverage	Good	Poor
Systemicity	Effect of factor is variable; may increase or decrease risk of resistance	
Treatment frequency	Low	High
Presence of secondary pests	Absent (only the target pest is treated)	Present (not targeted (potential) pests are also treated)
Life stages treated with related pesticides	Single	Multiple
Proportion of population treated	Effect of factor is variable; may increase or decrease risk of resistance	
Persistence	Short	Long
Number of crops treated	One	Many
Crop sequence	Crops separated by time or geography	Crops inter-planted; no break between planting; continuous
Pest control tactics	Multiple control tactics (chemical, biological, cultural}	Continuous use of single method or compound
Non target effects	Selective activity, no effect on natural enemies	Non selective, natural enemies also killed

2.3.1 Biological factors

Population size

Population size is a major factor in the development of resistance. In insects, the larger the population is, the greater the chance that resistance will develop. If the population is large, even if the percentage of resistant individuals is low, the number of survivors following a pesticide application could be a rather large. If repeated pesticide treatments eliminate most of the susceptible individuals, the odds of the resistant survivors finding mates and passing on the resistance genes could be very good. Conversely, if the pest population is small, the odds of successful mating of the very few resistant survivors could be very low and resistance development would be slow.

The situation is similar for fungal populations. Most 'natural' populations of fungal pathogens will contain a very small proportion of resistant individuals. Application of a fungicide will select for these individuals while not completely eliminating all the susceptible individuals (because of the inadequacies of spray applications). Thus the population left after fungicide application will contain a higher proportion of resistant individuals but will not be totally resistant. In the absence of a resistance management plan, this process will be repeated with each subsequent application until the pest population contains enough resistant individuals to create a problem. The process can be slowed by the ingress of susceptible individuals from outside the application area.

In the end, resistance management is a question of numbers. The extent of the pest control problem is directly related to the number of resistant individuals. If a pest infestation is low (i.e. a relatively small number of individuals), there is unlikely to be a control problem even if there is a high level of resistance (i.e. the resistant pests are strongly resistant). Conversely, if there is a high pest infestation, there will be an obvious field control problem even if the level of resistance is moderate.

Reproductive potential

Reproductive potential, or the number of offspring, seeds or spores per “parent”, has a significant effect on the development of resistance in pest populations. For all sexually reproductive pests that are targeted by pesticides, and with other factors being equal, the greater the number of offspring per organism, the larger the number of resistant individuals there will be.

The reason for this in insects is that producing a large number of offspring increases the chances of there being more individuals carrying the resistance gene and hence, if pesticide use continues, the odds of selecting individuals that carry one or two resistant alleles. The larger the number of survivors carrying resistance genes, the greater the potential is for heterozygote or homozygote individuals to mate. This can result in an increase in the frequency of the resistance genes in the population.

In weeds, annual species with high seed production and genetic diversity have a better chance of resistance development than species with lower seed production and genetic diversity.

In fungi, most propagation occurs through asexual spores transmitted by water splash or wind during the season with each disease lesion capable of releasing vast numbers of spores. Such spores can travel distances of a few metres to many hundreds of miles, depending on the pathogen. Sexual spores, where produced, are usually formed in late season and released early in the subsequent season after over-wintering. They can be important sources of new variation in the fungus but their role in resistance development is not known.

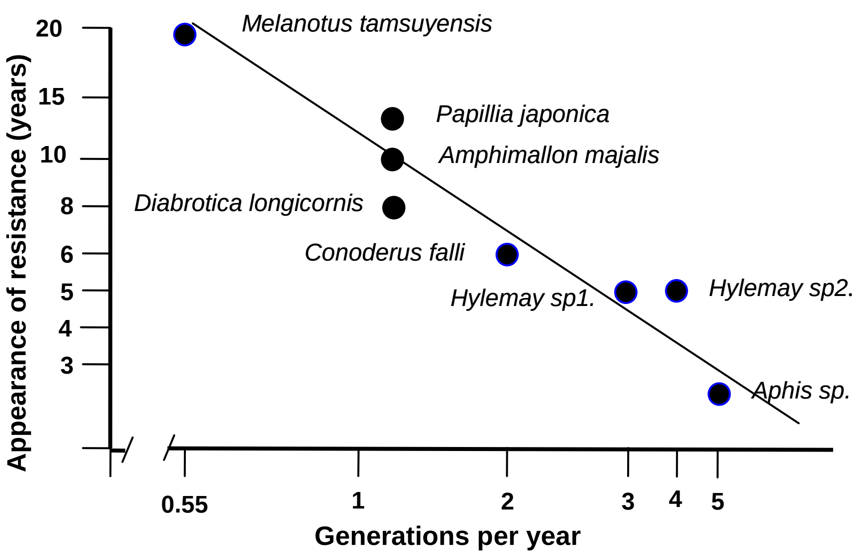

Figure 2 Relationship between the number generations per year of a pest insect and the appearance of resistance selected by soil applications of aldrin/dieldrin [Source: NRC (1986)]

Generation turnover

Generation turnover has an important role in the speed of resistance development. For insects, weeds and plant pathogens, resistance development will be relatively slower if there is only one generation per year rather than several, because the pest population is selected only once a year. Figure 2 compares the rates at which different insect populations developed resistance to the insecticide aldrin/dieldrin, ranging from 2 years for aphids with five generations per year to 20 years for *Melanotus tamsuyensis*, an insect with a 2-year life cycle.

Type of reproduction

Both sexual and asexual reproduction can contribute to the development of resistance. Sexual reproduction provides the impetus for genome rearrangements. However, once resistance has been selected it is more likely to be spread rapidly via asexual reproduction. For example, in aphids most reproduction is asexual throughout the year, and most fungal pathogens spread through asexual spores (conidia). Essentially the offspring are clones of their parent. If a portion of an aphid or fungal population has a resistance gene, this portion will survive while the susceptible portion of the population will be eliminated by intensive pesticide application, and the surviving resistant segment may quickly increase, leading to a rapid development of resistance unless a resistance management programme is implemented.

The presence of a sexual cycle in plant pathogens is often considered to increase the possibility of resistance developing by new gene combinations being formed. Conversely, it is equally possible that such sexual recombination could break gene sequences and lead to a loss of resistance factors. In practice it appears that resistance to most fungicides is present in untreated pathogen populations but at a very low level. Such resistance is then selected out by exposure to the fungicide. In weeds, the potential for the spread of resistance is lower in species that are self-pollinators or can reproduce vegetatively than in species that are cross pollinators.

Dispersal

Both long range and relatively short range movement of pests can affect the susceptibility of a particular population in a field or an area. Insects, spores, and seeds can be dispersed by the wind, can be imported with seed, soil, equipment, plant roots, containers, plant products, etc., or in the case of insects, can fly to new areas. For weeds, the potential for resistance to persist or spread will be much greater for species whose seed can be easily disseminated by the wind. For insects and diseases, the arrival of heterozygous or susceptible individuals will generally dilute resistance in the population of concern, as incoming insects can mate with treatment survivors and incoming plant pathogen spores can develop lesions and produce new colonies of susceptible types. This is the basis of the use of refugia to maintain susceptible pest populations. On the other hand, it is also possible for resistance genes to be introduced into a population by individuals migrating from an area where resistance is a problem. For example, a resistant strain of insects selected in a greenhouse may move (migrate) to surrounding fields and introduce the resistant gene to the field population.

In situations where resistance is operationally recessive only a few homozygous resistant (RR) individuals will survive after treatment with an insecticide. As homozygous susceptible (SS) individuals move into the area and mate with the survivors many offspring will be heterozygotes (RS) or (SS) susceptible individuals. If a treatment is made and the proper dose is used and good application coverage is achieved, the SS and most if not all of the RS individuals will be killed. However, if a reduced rate is used and/or coverage is poor, subsequent applications can result in the survival of many RS individuals and result in faster selection of a resistant population.

Seed bank

Resistance will be slower to appear in weed species that have higher levels of seed dormancy, i.e. a greater number of seeds in the soil which may emerge over time. While the seed produced after each application of an herbicide may contain a higher proportion of resistant individuals, susceptible seed from the seed bank will dilute resistance levels.

Pesticide metabolism

Increased metabolic degradation of a pesticide is one of the resistance mechanisms found in certain organisms, in particular insects and mites (see 2.2). Pesticides which are relatively easily metabolized by common biotransformation processes run a higher risk of becoming less effective through resistance development than pesticides which are more difficult to detoxify in the organism.

Number of target sites of the pesticide

Resistance develops more quickly when a pesticide has a single target site. If a pesticide has multiple target sites, the pest has to develop resistance at all of these sites. If a pesticide has only one target site, a single mutation at a single gene can lead to resistance.

Pest host range

Pests with a wide host range, infesting more crops, may have a higher risk of developing resistance than pests which are very crop-specific. This is particularly relevant for insecticides, as one insect species can infest several different crops. In many instances, strategies have been designed for a specific crop without regard for nearby or rotational host crops or insect movement, and thus have underestimated the number of treatments the insects receive. For example, a particular insect pest species may receive three to four treatments on cotton and four to five applications on a nearby, or subsequent, vegetable crop. The cotton specialist sees three to four selections; the vegetable specialist four to five selections; but the insect is actually receiving seven to nine selections. That is why it is important, particularly for insecticides, to design management strategies that are area-focused rather than crop-focused.

Such considerations are less relevant for fungicides as the plant pathogen is almost always crop specific and problems will only arise if the neighbouring crops are the same as the treated crop. However, defining spray programmes by geographical area rather than being simply farm based is worth consideration. Ideally, the best approach would be to coordinate spray application programmes so as to minimise the selection pressure on the pathogen caused by continuous exposure to the fungicide.

2.3.2 Genetic factors

Occurrence of resistance genes

For resistance to be selected in a pest population, at least some of the individual pests must have a gene for resistance. The degree of resistance and the speed at which it develops in the population depend on the effectiveness of the gene(s) in protecting the pest. In general, the greater the protection provided by the gene(s), the lower the fitness cost, and the higher the frequency of the resistance gene, the faster resistance can be selected.

Number of resistance mechanisms

As described above (see 2.2), there are several mechanisms that allow agricultural pests to survive exposure to toxicants, and resistance can develop more easily when an organism has more than one of these mechanisms. In insects in particular, there are many instances where pests may use more than one mechanism to develop resistance, even though one mechanism may be more pronounced than the others.

The combined effect of two mechanisms may also greatly increase the degree of resistance. For example, if an insect is 10-fold resistant to a pesticide through enzymatic detoxification and two-fold resistant because of reduced penetration, the overall resistance level could be 20-fold rather than 12-fold. (For fungicides, such additional complications are not relevant.) Also, if several different resistance mechanisms are simultaneously present in the same organism, this may result in resistance to more than one class of pesticide. This is referred to as multiple resistance.

Gene frequency

Gene frequency (often also referred to as allele frequency) is the proportion of all copies of a gene that is made up of a particular gene variant (or allele). The frequency of the resistance allele has a significant effect on the development of resistance. In most instances the frequency of homozygote individuals that are resistant to a new pesticide is very low, e.g., 10^{-4} or lower, while the frequency of heterozygous individuals may be higher. While other factors also affect the selection of resistance in a population, in general, the higher the resistance gene frequency the faster resistance will develop.

For fungi the situation is a little different as all except the oomycetes (characteristically the downy mildew fungi) exist in the haploid state. The selection of resistant mutants from the natural population for these is thus a simple consideration of the selection pressure.

Dominance of resistance gene(s)

Resistance genes can range from dominant through semi-dominant to recessive. If a trait is dominant or semi-dominant, only one parent has to possess the trait for it to be fully or partially expressed in the offspring. If it is recessive, both parents must possess the trait. If the resistance is genetically dominant, it can rapidly become established within the population and be difficult to manage. Fortunately, most resistance mechanisms (for example *kdr*) are controlled by recessive or semi-dominant genes, which increases the chance of managing resistant populations. For example, in the case of the carboxylic acid amide fungicides, resistance is recessive in *Plasmopara viticola* such that only the homozygous recessive individuals are resistant. This is thought to explain why resistance does not build up in *Plasmopara viticola* populations.

In insects, incompletely recessive or dominant genes can be made functionally dominant when the individuals carrying those genes are exposed to reduced rates of the pesticide. This lower dose can result from the deliberate use of a low rate, inadequate coverage of the plant or area being treated, or exposure to pesticide residues that are degrading on the treated surface. When this occurs, heterozygote individuals survive and pass on the resistant gene when they mate with other heterozygote or susceptible individuals.

Fitness of “R” individuals

Individuals carrying a gene for resistance may suffer a fitness cost, such as reduced vigour and/or a timing difference in the life-cycle which makes it difficult for mating to occur with individuals not having the R gene. If the fitness cost of the resistance gene is low, resistance genes may accumulate quite rapidly in a population. However, if the fitness cost is high, then only in the presence of the pesticide will resistant individuals have a significant advantage over the susceptible ones. In the absence of the pesticide, the resistant forms may be non-competitive and be lost very quickly. It is this factor that permits rotation to be a successful resistance management tool.

However, it is not always the case that resistant individuals suffer a fitness cost. For fungicides, it is wrong to assume that resistant fungi will be less fit than susceptible fungi, as this is not always the case. It is also possible for “less fit” individuals to mutate and be selected further for increased fitness.

Protection provided by the “R” gene

If the resistance gene provides a high degree of protection from the pesticide, then individuals carrying that gene have a very high probability of surviving a pesticide application and passing the resistance gene on to the next generation. However, if the resistance gene provides only a moderate level of protection, then the individuals carrying the resistant gene will be protected from lower doses of the pesticide but not high doses. This is another reason to ensure that full label rates of a pesticide are used and that the best coverage possible is achieved. Lower doses and poor coverage permit the accumulation of the resistance genes in the population.

Cross-resistance

Cross resistance means that resistance to one pesticide confers resistance to another pesticide, even where the pest has not been exposed to the latter product. Its presence therefore increases the risk of resistance. Cross-resistance occurs because two or more compounds are acting on the same target site and/or are affected by the same resistance mechanism. Cross-resistance develops most commonly with compounds having the same mode of action and that are usually, but not always, chemically related from the same chemical group. It may be complete or partial (if more than one mechanism is responsible for the resistance).

Some resistance mechanisms can affect compounds in different chemical classes but this phenomenon is largely restricted to insecticides. For example, both DDT and the pyrethroids are affected by the *kdr* gene which interferes with the sodium channel in nerve cells. Intensive use of pyrethroids on a population which had an earlier resistance problem with DDT could result in the development of pyrethroid resistance in that population.

In some cases negative cross-resistance occurs, when a resistance mechanism makes the organism resistant to one pesticide but increases its susceptibility to another pesticide.

Past selection

Past selection of resistance genes may facilitate the development of resistance to new compounds because the previous use has most likely increased the resistance gene frequency. That does not necessarily mean the new compound will be ineffective or that resistance to it will develop quickly. It simply means that the potential for resistance development is higher than it would have been had no related compounds been used previously. However, if there is a high level of cross-resistance and there was a serious resistance problem in the past, then the potential for resistance developing rapidly to the new compound is high.

Modifying genes

Resistance genes may be deleterious to the pests that have them to a greater or lesser degree. However, with time and continued selection the lowered fitness of the resistant individuals may be overcome as ancillary, or modifying, genes associated with improving fitness are acquired. In some instances the fitness cost of the resistance gene is nearly entirely overcome; the resistant gene continues to appear in the pest population and reversion to the original susceptible gene occurs very slowly or not at all. In other cases, the fitness cost cannot be overcome and reversion, in the absence of selection, occurs rather quickly. The role of modifying genes is best understood in insects and weeds. Relatively little work has been done on modifying genes in fungi, although it is known that resistance can develop to the de-methylation inhibitor fungicides as a result of the accumulation of many genes of minor effect as well as being due to major genes or major genes together with modifiers.

2.3.3 Operational factors

Activity spectrum of the pesticide

Broad spectrum pesticides that are effective against a wide range of pests or species are more likely to cause resistance problems than narrow spectrum pesticides for the simple reason that they are likely to be used much more often in a given area because they control more pest species. In most cropping situations where there are other target pests to be controlled, the narrow spectrum product will be used less frequently, and the selection pressure will be lower.

Broad spectrum pesticides should also be used with care because they may be selecting resistance in non-target pest species that happen to be in the treated area along with the target pest, at sub-treatment threshold levels. For example, use of a carboxylic acid amide, a very narrow spectrum fungicide, to control downy mildew on vines will not affect other vine diseases and will thus not exert any selection pressure other than on the downy mildew. In contrast, a DMI or QoI fungicide applied to a cereal crop to control one particular disease may well exert a selection pressure on other diseases present because of the broad spectrum of activity of these fungicide groups. This situation is exacerbated if the secondary pest requires a higher rate for control than the primary pest targeted by the treatment. If the secondary pest becomes a primary problem at a later date, resistance may develop rather quickly.

Application rate

Although pesticide application rates are not set with regard to resistance, it is important to apply the recommended rate and not underdose. Ideally this rate should eliminate all susceptible and essentially all heterozygous resistant individuals from the pest population while reducing pest numbers below the economic threshold. If the dose is too low, the susceptible individuals will be eliminated but the partially resistant heterozygotes will survive. A dose that is too low will also have the effect of making the resistance gene functionally dominant and resistance may develop rather quickly. However, attempting to eliminate heterozygote individuals is most effective if the population is not extremely large, consists mostly of susceptible individuals, and is subject to immigration by susceptible individuals; then highly resistant homozygous resistant individuals should be rare and will likely suffer from reduced fitness because of the resistance genes.

The use of higher than recommended application rates is not recommended either. This is because if there are any survivors from a high rate, these are likely to be mainly homozygous resistant. In particular when there is no immigration of susceptible individuals, high dose rates are then very likely to increase the development of resistance. Higher dose rates will also kill more natural enemies, which may in turn result in increased pest populations.

Coverage

Coverage of the substrate (e.g. crop, commodity) being treated is very important. If coverage is good, with the correct amount of pesticide applied to the entire area, the pests will more likely encounter the desired, lethal rate. If coverage is poor, with some areas receiving more pesticide and others less or none at all, the result will be similar to what happens when below-label rates are used. Homozygous individuals will be selected and the development of resistance will be promoted.

Systemicity

The use of systemic rather than contact pesticides can both hasten and slow the development of resistance. Systemic insecticides generally have a much lower impact on beneficial insects associated with the pest. Thus, after an insecticide treatment, predators are still present and may eliminate many of the surviving pests and prevent further transmission of their resistance genes to the pest population. However, systemic compounds also have drawbacks.

The impact of systemicity depends on the compound in question. Systemic insecticides may provide the infesting pest with a more uniform dose and can reach pests that would have been protected from foliar application of a contact pesticide, because they were under a leaf. While this is good for pest control it can increase the selection of resistance, because it eliminates the possibility that some susceptible pests escape treatment and continue to contribute their genes to the pest population. For fungicides, systemicity allows the fungicide to permeate the plant as leaves expand, thereby protecting plant tissue that wasn't directly treated. Generally, systemic compounds persist within the plant longer than contact pesticides, and thus create more selection pressure for resistance, particularly if there is a continuous influx of the pest.

There is a great temptation to use systemic fungicides as curative applications, to control fungal infections after they have established in the plant tissue. In general this is not considered to be good practice and most resistance management guidelines specifically advise against the use of curative applications because of the increased selection pressure they present.

Treatment frequency

Pesticide treatment frequency should be limited to the number of treatments necessary to protect the crop or control the pest, as unnecessary treatments increase the selection pressure on the pest population. Particularly unadvisable is the use of frequent treatments at suboptimal rates (for example to reduce treatment costs), which can lead quickly to the development of resistance. Only the susceptible individuals will be eliminated from the population, while the heterozygotes will be functionally resistant and will consequently be selected along with the homozygote resistant individuals. There are cases where spectacular pest control has been achieved (temporarily) when frequent applications were made, only to be followed by the development of a very serious resistance problem. In situations where the continual movement of untreated individuals into an area makes frequent applications necessary, it is wise to rotate pesticide treatments with unrelated compounds in order to reduce the selection pressure on the pest population.

Presence of secondary pests

Another consideration, as mentioned in the discussion of pest host spectrum, is the presence of non-target but susceptible pests that happen to be in a crop at sub-economic levels while another pest species has reached a treatment threshold. Although the second pest species is targeted, the first will also be selected by the pesticide treatment. This is why detailed pesticide application records should be kept and consulted. Unfortunately, crop specialists often fail to consider the treatments made when the pest was at sub-economic levels when designing a pest control programme.

Life stage(s) treated

Resistance is less likely to develop if insects can be treated at life stages when they are especially vulnerable to pesticides (e.g. in Lepidoptera, the neonate or first instar larvae or adult males are much less able to metabolize insecticides) or if different life stages can be treated with unrelated compounds. In the latter case, the reason is that if some individuals prove to be resistant to a pesticide at one life stage, they are likely to be eliminated if the next stage is treated with an unrelated compound. Such an approach is generally difficult to achieve, however, unless the generations are very synchronous or the larvae and adults live in different environments. Most often there is a mixture of life stages in a field situation.

Proportion of population treated

Generally, resistance does not develop at the same time over the entire geographical range of a pest species. It is much more likely to develop locally. In the case of fungi, for example cereal powdery mildew, resistance often appears first in the areas of highest fungicide use and highest disease intensity, e.g. northern Europe, and then spreads southwards. For insecticides and herbicides, if only particular fields or localized areas are treated, susceptible individuals or seeds may move into the treated area and any resistant genes present will be diluted when the susceptible individuals mate or pollen from susceptible plants crosses with the resistant survivors. Conversely, for all pesticides, a local area can be overly treated and a localized resistant population created. If the species is highly mobile, e.g. spores, seeds and arthropods carried by the wind, it can carry resistant genes to other areas where the gene may not be present or present at only a very low frequency.

Persistence

All things being equal, resistance is less likely to develop with less persistent pesticides, because the selection pressure is lower. However, in many instances, long residual products are desired because fewer applications are required. Nevertheless, if the pesticide dissipates slowly, a rate which selects resistant individuals will be reached at some point and resistance selection will occur.

In insects, the amount of selection is partially dependent on the movement of the insect pest. For example, if the original application eliminated most of the pests and there are few untreated individuals encountering the treated areas, then there will be little selection. However, if there is a continuous influx of untreated individuals from adjacent areas, or emerging from the soil, then the number of resistant individuals selected could be high. If treatments are not frequent and persistence is short, the resistance genes in the population may be diluted by the influx of susceptible individuals. The best situation is a highly active pesticide that produces the desired pest control and then degrades very quickly. Persistence and frequency of application are related. Short residual pesticides tend to exert less selection pressure and resistance development is slower. However, frequent applications can negate the benefits of a short residual pesticide.

Number of crops treated

If many different crops are treated with the same pesticide, the risk of resistance development is higher, in particular for pests with a wide host range. Insects may be selected by the pesticide on various subsequently grown crops, and refuges with susceptible individual will be smaller.

Crop sequence

If crops grown in the same area are separated in time (e.g. with fallow periods between subsequent cropping cycles) or if they are grown in distinct geographical areas, the risk of resistance development will be lower. On the other hand, if continuous cropping is applied, the number of selection events by a pesticide may be high and resistance will develop faster. Similarly, if crops are planted over a large continuous area, refuges where susceptible individuals can survive will be smaller.

Pest control tactics

The continued use of a single pesticide or a unique reliance on chemical control will likely increase the risk of resistance development to pesticides. This is why resistance prevention and management strategies tend to prescribe the use of multiple control tactics, based on chemical, biological and cultural practices.

Non-target effects

Specifically for insecticides, control methods which have little effect on natural enemies of crop pests, such as the use of selective insecticides and/or alternative pest management techniques, tend to slow resistance development. This is because natural enemies will kill both resistant and susceptible pests, thereby reducing the frequency of resistant genes in the population if resistance is not yet predominant.

2.4 Fungicide resistance risk

The risk of resistance development depends largely on the chemical class of the fungicide, the pathogen involved, and how and where the treatment is made. Each chemical class is characterized by a typical resistance pattern. Table 2 categorises the major chemical classes and compounds according to whether they are at high, moderate, or low risk of resistance development

Table 2 Inherent risk of resistance associated with different fungicides and chemical classes of fungicides.

Resistance risk	Chemical class or compound
High	Benzimidazoles, dicarboximides, phenylamides, strobilurin analogues (e.g. methoxyacrylates, oximino acetates)
Moderate	2-Amino-pyrimidines, aminies (including morpholines), anilinopyrimidines, aromatic hydrocarbons, azoles, carboxanilides, carboxylic acid amides, carpropamid, cymoxanil, fenhexamid, kasugamycin, phenylpyrroles, phosphorothiolates, quinoxyyfen
Low	Chlorothalonil, coppers, dithiocarbamates, fosetyl-Al, pyroquilon, phthalimides, suplhurs, tricyclazole
[Sources: Brent & Hollomon (2007a,b), FRAC (2011)]	

In addition to the risk of resistance associated with various fungicides there are also a number of pathogens which have demonstrated a propensity for developing resistance over the years. These are shown in Table 3.

Table 3 Examples of important plant pathogens considered to present a high risk of resistance development.

Pathogen	Crop
Phytophthora infestans	Potatoes (phenylamides only)
Plasopara viticola	Grapes
Erysiphe gaminis	Wheat and barley
Uncimula necator	Grapes
Sphaerotheca spp.	Various
Mycosphaerella fijiesis	Bananas
Pyricularia oryzae	Rice
Gibberella fujikuroi	Rice
Botryotinia fuckeliana	Various, especially grapes
Venturia spp.	Apples and pears
[Source: OEPP/EPPPO (2002)]	

With new fungicides that do not fit into the categories above, it is necessary to consider the individual factors that can lead to resistance development. The framework provided in Table 4 can be used to do this.

Table 4 Framework for predicting the development of resistance to a new fungicide

Factor	Positive indication of resistance risk
Inherent characteristics of the fungicide	
Fungicide class	When the fungicide is a member of a class which has a record of resistance problems.
Target site	If there is a single target site; or if the site is known to be capable of change to a form that is unaffected or less affected by other fungicides.
Cross-resistance	If there are target pathogen strains resistant to existing fungicides which also resist the new fungicide.
Response to mutagenic agents	If treatment with mutagenic agents causes the target fungus to produce resistant, fit mutants.
Response in sexual cross experiments	If sexual crossing causes the target fungus to produce resistant, fit recombinants.
Response to repeated fungicide application	If repeated exposure of the target fungus to the fungicide, in the laboratory or in field plots, causes the appearance of resistant, fit strains at detectable levels.
Inherent characteristics of the plant pathogen	
Generation time	If the target pathogen multiplies rapidly, and hence fungicide applications are frequent
Amount of sporulation	If sporulation of the pathogen is abundant
Spore dispersal	If spores spread readily between plants, crops and regions
Genetic adaptability	If the pathogen is haploid, has a gene structure that allows expression of mutations to resistance, has an obligatory sequence of sexual and asexual reproduction in the disease cycle, or shows other signs of genetic adaptability
History of resistance	If the pathogen has a record of developing resistance to fungicides (of any kind)
Conditions of use (locally determined)	
Application of the fungicide	If fungicide applications will be repetitive, if the fungicide (or fungicides related to it by cross-resistance) will be used continually and/or widely on crops in the region
Complementary measures	If other types of fungicide (as mixtures or in rotation) or if non-chemical disease-suppressant measures, (e.g. crop-rotation, resistant varieties, hygienic precautions) are not to be used
Pathogen incidence ("disease pressure")	If the pathogen is present in large amounts and/or over large areas, and/or is multiplying quickly over long time periods (short generation time)
Pathogen isolation	If populations of the target pathogen are isolated and/or non-migratory, e.g. in glasshouse crops
[Source: Brent & Holloman (2007a)]	

Figure 3 illustrates how the inherent risk of resistance associated with specific fungicides and pathogens can be categorised. The risk categorization is approximate and the scores are arbitrary, but they are probably the best estimates that can be made in the light of current knowledge. The Fungicide Resistance Action Committee (FRAC), a specialist technical group of CropLife International that provides fungicide resistance management guidance, regularly reviews the resistance risk of the chemical groups and maintains a List of Plant Pathogenic Organisms Resistant to Disease Control Agents, which indicates whether an organism has shown any resistance in the field or laboratory, including in mutation studies.

Figure 3 Risk of resistance development for specific fungicide-pathogen combinations. The inherent risk of resistance associated with specific fungicides and pathogens can first be categorised separately as high, medium, low, or non-existent (correspondingly scored as 3, 2, 1, or 0.5), and then combined for a fungicide-pathogen score (from 9 to 0.5). [Source: Brent & Holloman (2007a)]

Combined risk: 1 = low, 2-6 = medium, 9 = high				
High Benzimidazoles QoIs Phenylamides Dicarboximides	3	**3**	**6**	**9**
Medium Carboxanilides DMIs Phenylpyrroles Phosphorothiolates Anilinopyrimidines MBI-Ds	2	**2**	**4**	**6**
Low Coppers, Sulfur Chlorothalonil Dithiocarbamates Phthalimides MBI-Rs Probenazole	0.5*	**0.5**	**1**	**1.5**
Fungicide Risk **Pathogen Risk**		1	2	3
		Low *Rizoctonia* Rusts Soil borne pathogens Smuts & Bunts	**Medium** Eyespot *Mycosphaerella graminicola* *Rhyncosporium*	**High** *Bptrytis* *Blumeria* *Magnaporthe* *Venturia* *Plasmopara* *Penicillum* *M. fijiensis* *Phytophthora infestans***

* The low score reflects the long standing record of "no resistance" in this low risk group.

** *Phytophthora infestans* is considered by some to present a medium risk as the high risk classification is based largely on the reaction to phenylamides.

The latest information is available from the FRAC website at: www.frac.info.

Finally, the development of resistance in a country or region also depends on the conditions of fungicide use. These conditions are sometimes referred to as risk modifiers, but in fact they are important determinants of resistance development and must always be included as an integral part of the assessment. The most important conditions of use that affect resistance development are considered to be:

- the number of applications – the more frequently a particular compound is applied to a pathogen population, the more rapid the selection of resistance;
- exclusive use of a single mode of action – the more exclusive the use of a single mode of action, the more sustained the selection pressure for resistance;
- the 'dose' of fungicide used – application of less than the dose recommended on the product label may increase resistance selection pressure;
- the extent of pathogen populations exposed to the fungicide – if disease incidence within a particular region is relatively low, sporadic, or irregular from season to season, the selection of resistance is reduced;
- size of the treated plots and proportion of the area or region treated – the larger the plots treated and the greater the proportion of the overall area where the fungicide is used, the more widespread the selection and build-up of resistant variants;
- reliance on fungicides only and failure to use integrated disease management – will result in increased fungicide resistance selection pressure; and
- isolation of pathogen populations (e.g. in greenhouses or polythene tunnels, or in isolated agronomic regions), preventing re-entry of sensitive forms – can favour development of resistant populations.

2.5 Herbicide resistance risk

Resistance to herbicides has evolved less rapidly than resistance to insecticides and fungicides but it has been reported worldwide. This has been attributed to the:

- relatively slow reproduction of plants, often only one generation per year;
- incomplete herbicide selection pressure from herbicides;
- soil seed reserves (seed bank);
- plasticity of weedy plants;
- multiple modes of action of early herbicides; and
- use of non-chemical weed control methods in conjunction with herbicide use.

In many instances weeds appear to suffer no fitness cost for resistance genes. Consequently the frequency of these genes may be high even before they are selected by herbicide use. Most cases of herbicide resistance involve a single mutation or modification in some function so that the weed is resistant or cross-resistant. Instances of multiple resistance have been reported, but it seems to be quite rare for a single plant to possess multiple resistance mechanisms.

Resistance to herbicides is currently increasing at an exponential rate. This may be due to the fact that many of the newer, very active herbicides affect only a single target site.

The principal herbicide resistance mechanisms are:

- *altered target site:* due to a change in the structure of the target site, the herbicide no longer binds to its normal site of action, allowing the plant to survive the herbicide treatment;
- *enhanced metabolism:* the resistant plant can degrade the herbicide to non-phototoxic substances faster than a normal sensitive plant, thereby surviving an herbicide treatment

in much the same manner as many crop plants; and

- *compartmentalism/sequestration:* the herbicide is removed from the sensitive parts of the plant cell to a tolerant site, such as a vacuole, where it is effectively harmless to plant growth.

The most important factor in the development of herbicide resistance is the frequent use of herbicides with similar modes of action. Other factors include the:

- intensity of selection pressure;
- use of crop rotations that rely primarily on herbicides for weed control – crop rotation is important because it will determine the frequency of treatment and type of herbicide used. It is also the major factor in the selection of non-chemical weed control options, and it has a strong impact on the weed flora present;
- lack of non-chemical weed control practices – cultural or non-chemical weed control techniques, incorporated into an integrated approach, are essential to a sustainable crop management system;
- frequency of resistant genes in the weeds being treated; and
- size and viability of the seed bank, i.e. weed seeds lying dormant in the soil, which can act as a buffer delaying the development of resistance.

Table 5 shows how different operational factors affect the development of herbicide resistance, and specifically whether the practices present a low, moderate, or high risk of selecting resistance in the treated weeds.

Table 5 The impact of operational factors on the development of herbicide resistance.

Operational factors	**Risk of resistance development**		
	Low	**Moderate**	**High**
Cropping system	Full rotation	Limited rotation	No rotation
Herbicide mix or rotation in cropping system	>2 modes of action	2 modes of action	1 mode of action
Weed control in cropping system	Cultural, mechanical and chemical	Cultural and chemical	Chemical
Use of same mode of action per season	Once	More than once	Many times
Weed resistance to the mode of action	Unknown	Limited	Common
Weed infestation	Low	Moderate	High
Control in last 3 years	Good	Declining	Poor

[Source: HRAC (2011)]

Some classes of herbicides are more likely to have resistance problems than others. Table 6 shows the speed and likelihood of resistance development for various herbicide groups, classified according to site of action by the Herbicide Resistance Action Committee (HRAC), a specialist technical group of CropLife International that provides herbicide resistance management guidance.

Table 6 Years required for resistance development for the HRAC herbicide groups and the risk of resistance.

Herbicide groups (HRAC classification)	**Years of application before resistance develops**	**Risk of resistance**
A	6 - 8	High
B	4	High
C	10 – 15	Medium
D	10 – 15	Medium
F	10	Medium
I	Unknown	Low
L	>15	Low
M	15	Low

[Source: HRAC herbicide classification, FAO (2008)]

As a general rule, a herbicide with low selection pressure, which is used sporadically and alternated with non-chemical control practices, will have a low risk of resistance. There are also a number of weed species which have shown a propensity to develop herbicide resistance. These are listed in Table 7.

Table 7 Ten major weeds affected by herbicide resistance worldwide

Species	**Common Name**
Lolium rigidum	Rigid ryegrass
Avena fatua	Wild oat
Amaranthus retroflexus	Redroot pigweed
Chenopodium album	Lambsquarters
Setaria virdis	Cola de rata
Echinochloa cus-galli	Barnyard grass
Kochia scoparia	Common kochia
Conyza canadensis	Canadian horseweed
Amaranthus hibridus	Smooth pigweed

[Source: HRAC (1999)]

2.6 Insecticide resistance risk

There is a long history of resistance to insecticides. In fact, resistance develops to every major insecticide sooner or later. Much of the research on resistance to date has been conducted to develop tactics to overcome or delay insecticide resistance.

Most insect resistance problems have been linked to the factors shown in Table 8.

Table 8 Factors affecting resistance development in insects.

Factor	Effect on resistance development
Insect related factors	
Short life cycle	Insect population receives several or many treatments per crop per season which may shorten the time to resistance development.
High infestation/population levels	Even with high levels of control there can be relatively high numbers of selected survivors, leading to faster resistance development.
Large number of offspring per female	Permits a relatively low number of insects to quickly re-establish large populations from selected survivors which carry the resistant gene(s).
Wide host range	Insect may be selected on several crops per year.
Operational factors	
Use of lower than label use rate	Selects heterozygote resistant individuals increasing frequency of resistance genes in the pest population.
Inadequate coverage	Equivalent to low use rate which increases the survival of resistant heterozygotes and thus the frequency of the resistant gene(s).
Improper application timing	Less sensitive stages targeted or population can grow to overwhelming size. This may lead to the selection of heterozygotes among the less sensitive stages and treatment of large populations will result in the selection of large numbers of resistant individuals.
Use of a singe class of chemistry	High level of selection, i.e. increases resistance selection pressure.
Nearly total reliance on chemical control	High selection pressure on insecticides; kills predators and parasites thereby permitting the frequency of resistance genes to increase in the pest population.
Focus on single target pest and crop	Ignores insects present at lower than threshold levels and treatment of other crops, and increases resistance selection in non-target species.
Use of long residual compounds	Compounds degrade permitting the survival of heterozygote individuals thus increasing the frequency of resistance genes.
Use of broad spectrum products	Eliminates predators and parasites which may contribute to target pest control and may select for resistance in non-target pests present in the same area

[Sources: IRAC (2011), Whalon et al, (2008), NRC (1986)]

A number of insect species have developed resistance more often than others. Shown in Table 9, these include many of the most difficult and economically damaging arthropod species in the world. These species tend to have high population numbers with short generation turnover. Consequently, infestations tend to receive a large number of insecticide applications annually. Over the years many of these species have been treated with almost every new insecticide or acaricide developed. Although they are not necessarily resistant to all insecticides or across their entire range, these species require extra care. If any of the species becomes a target pest, it is critical to develop a RMP which includes as many good IPM practices as possible, before treatment with either an existing or a new insecticide. In fact, caution should also be used when treating species not included in this list; it should not be assumed that they are unlikely to develop resistance to a new insecticide that belongs to a new class of chemistry. Eventually, they may well do so.

Table 9 The top 20 arthropods for which resistance has been reported in agriculture and public health. The ranking is based on the number of insecticides the insects are resistant to, from 1 (resistant to the largest number of compounds) to 20.

Order	Family	Species	Rank	Host
Acari	Acaridae	*Rhizoglyphus robini*	19	Ornamental plants, stored onions
Acari	Ixdidae	*Boophilus microplus*	6	Cattle
Acari	Tetranychidae	*Panonychus ulmi*	9	Fruit trees
Acari	Tetranychidae	*Tetranychus urticae*	1	Cotton, flowers, fruits, vegetables
Coleoptera	Chrysomelidae	*Leptinotarsa decemlineata*	4	Potato, eggplant, tomato
Coleoptera	Tenebrionidae	*Tribolium castaneum*	17	Stored grain, groundnuts, sorghum
Dermaptera	Blatteliidae	*Blatella germanica*	7	Urban
Diptera	Calliphoridae	*Lucilia cuprina*	18	Cattle, sheep
Diptera	Culicidae	*Anopheles albimanus*	20	Human
Diptera	Culicidae	*Culex pipiens pipiens*	11	Human
Diptera	Culicidae	*Culex quinquefasciatus*	15	Human
Diptera	Muscidae	*Musca domestica*	5	Urban
Hemiptera	Aleyrodidae	*Bemisia tabaci*	8	Greenhouse, cotton, cucurbits, crucifers and vegetables
Hemiptera	Aphididae	*Aphis gossypii*	10	Cotton, vegetables
Hemiptera	Aphididae	*Myzus persicae*	3	Fruit, vegetables, trees, grains
Hemiptera	Aphididae	*Phorodon humuli*	12	Hops, plum
Lepidoptera	Noctuidae	*Helicoverpa armigera*	13	Cotton, maize, tomato
Lepidoptera	Noctuidae	*Heliothis virescens*	14	Chickpea, cotton, maize, tomato
Lepidoptera	Noctuidae	*Spodopotera littoralis*	16	Lucerne, cotton, potato, vegetables
Lepidoptera	Plutellidae	*Plutella xylostella*	2	Crucifers

[Source: Michigan State University (undated)]

2.7 Rodenticide resistance risk

The process of resistance development in rodents is similar to that found in the other pest species, i.e. it is the result of overuse, under-dosing, and the use of a single class of chemistry. However, two factors are unique to rodenticide performance. These are:

- the ability of some rodents to learn to avoid treated baits and traps, called bait-shyness or learned food aversion. This occurs most frequently with acute poisons. Modern rodenticides are therefore limited to the delayed-action anticoagulants;
- the size of rodent populations, which are typically much smaller than those of fungi, insects or weeds. The main means of control is poison baits, which each individual rodent must decide to eat. There is no spraying of large areas as with herbicides, insecticides etc.

Anti-coagulant resistance in rodents has proven to be rather complex. The major cause of resistance in Norway rats (*Rattus norvegicus*) appears to be mutations on the VKOR gene which affects vitamin K metabolism. Increased detoxification by cytochrome P450 has also been linked to resistance. As in arthropod pests, resistance in rodents is affected by reproductive characteristics, the characteristics of the pesticide, and the past history of the population.

While resistance to some anticoagulant compounds exists in certain localities, particularly in the countries of North America and northern Europe, and in the species *R. norvegicus, Mus musculus, M. domesticus* and *R. rattus*, it is nevertheless possible to control rodents satisfactorily with currently available rodenticides, even in places where resistance exists. This will likely remain the case for the foreseeable future.

3 Pesticide resistance prevention and management

3.1 Developing a resistance management plan

A RMP describes the tactics or measures that should be taken to prevent and/or manage pesticide resistance for a specific pest. The objective is to reduce the selection of resistance genes in a pest population. The tactics should be designed to maintain a high frequency of susceptible genes and a low frequency of resistance genes in the pest population by reducing selection pressure, while providing the required level of pest control. These tactics will be different for each pest group, but a number of general principles apply to all RMPs.

3.2 General principles

Pesticide resistance management as part of IPM

It is highly recommended for a resistance management plan to be developed within the framework of an overall integrated pest management approach for a given pest and cropping system. This should ensure that rational pest control strategies based on IPM principles - including the use of pesticides only when necessary and the use of alternative pest management techniques whenever possible - are designed to manage resistance.

Implement resistance prevention and management programmes when new pesticides are introduced

RMPs should be implemented before resistance becomes a problem and should be applied uniformly over large areas in order to obtain their full biological benefit. When the first noticeable symptoms of resistance appear, the frequency of the resistance gene(s) will have already increased substantially. This will make it more difficult to maintain the overall susceptibility of the pest population. Unless there is a very heavy fitness cost, the resistance gene(s) may gradually accumulate in the pest population.

Focus on the pest

In designing a RMP, it is important to learn as much as possible about the biology of the pest and its hosts. This information is essential for understanding the loss of susceptibility and development of resistance in the target pest. The RMP should address the entire area where the pest is found, not just the crop of concern. Ideally, it should be implemented across an entire cropping region, focusing on the pest rather than on any particular crop, with widespread adoption by all growers in the area. Even a small amount of non-compliance can negate the efforts of a large RMP. In the case of fungicides, the RMP should be implemented over wide geographical areas, usually whole regions or countries. In the case of herbicides, the RMP should focus on weed management in the entire crop rotation.

Consider adjacent host crops

To manage resistance in insects, in particular, RMPs should consider pesticide treatment of alternate host crops located in the vicinity of the main host crop. Many of the same insect pests are likely to be present on other crops that are growing in close proximity or in sequence, or wild hosts. If the same, or related, pesticides are used in all the crops, the population is under much heavier selection pressure than might be calculated.

For example, *Bemisia sp.* occurs on both cotton and vegetables and easily moves from one crop to another. If there were five applications on cotton and five on a vegetable crop, the *Bemisia sp*. population would receive ten applications or selections annually. If each crop were evaluated separately, it would appear that the population was only receiving only five selections per year. It is important to take this into account in designing a RMP. If each crop is considered separately it is quite likely that the selection pressure exerted on the pest population will be underestimated, particularly if different growers and crop specialists are involved.

Consider alternative (non-chemical) pest management measures

In keeping with IPM principles and strategies, a RMP should comprise as many alternative, non-chemical pest control tools and methods as possible, as long as they contribute effectively to managing the pest. These can include biopesticides, biological control agents such as predators and parasitoids, resistant crop varieties, the timing of planting so as to reduce the risk of infestation, use of crop rotation and other cultural practices that interfere with pest reproductive cycles, attention to hygienic practices such as equipment cleaning to stop the spread of seeds and spores, etc.

Use more than one class of pesticide

A RMP should incorporate as many different classes of pesticide as possible to avoid the development of cross-resistance, when resistance to one pesticide confers resistance to another, even where the pest has not been exposed to the latter product. The more non-cross resistant compounds are used the lower resistance selection pressure will be on any one compound or class of compounds. Such different classes can be applied in sequence (alternating applications) or as co-formulated mixtures or tank mixes containing compounds with different modes of action and different modes of resistance. The mode of action classification of the various fungicides, herbicides and insecticides can be found through the links provided in Annex 1.

Consider all treatments made during the year

RMPs should consider all pesticide treatments made to a crop during the year, including treatments with different compounds and of different pest life stages. Some selection for resistance occurs each time a pesticide is applied. Generally, the more treatments made, and the more insect life stages and plant pathogen generations treated, the faster susceptibility will be lost and resistance will increase, unless measures are taken to mitigate the selection of resistance genes.

For example, if a soil insect is treated with a soil insecticide, the larvae will have selection pressure for resistance. Some heterozygote larvae may survive, because it is difficult to obtain a uniform concentration of the pesticide in the soil. If the adults that develop from the treated larvae are treated again with the same or a related insecticide, then a second selection of that generation will occur. Thus, in this situation two stages of the pest will have been selected. Some of the heterozygote individuals surviving the soil treatment may be killed when the adults are treated, but over time there will be a build-up of resistant individuals in the population. To avoid this, unrelated compounds should if possible be used to treat the larvae and the adults.

Similarly, if a pest infests several crops over the course of a year and the same, or related, compounds are used in all the crops, the population is under much heavier selection pressure than might be calculated, if all crops and treatment times are not considered.

Apply only recommended pesticide application rates

The correct application rate should always be used. Reducing pesticide application rates to reduce costs may appear to provide the pest control desired, but this is only temporary. Continuous use of below label rates will result in the increased selection of heterozygote and homozygote resistant individuals thus increasing the development of a resistant population. The properly applied label rate should eliminate the heterozygote resistant individuals from the pest population and significantly slow the development of a resistant population.

Involve stakeholders

To have a chance of success any resistance management strategy should be agreed on by all stakeholders, including growers, the pesticide registrar, pesticide companies/distributors, the ministry of agriculture and extension services. In particular the strategy must be understandable and acceptable to farmers. For RMPs that cover large areas, such as those designed for fungicides, local and regional cooperation are essential elements for the successful development and implementation of a RMP.

Evaluate and refine the RMP

The development of resistance is a dynamic process and is continually evolving; consequently, RMPs should be flexible. To remain effective, they should continually be re-evaluated and adapted to the changing situation, which can include changes in the level of pest resistance, the availability of new pesticides with new modes of action, or the availability of new pest resistant crop varieties.

3.3 All types of pesticides – resistance management tactics

Mixtures of pesticides with different modes of action or mechanisms of resistance

Mixtures of pesticides with different modes of action can be effective in managing resistance development. Various types of pesticide mixtures are used in agriculture and pest control - for example, two pesticides with different pest spectra, the combination of a pesticide and a synergist, the combination of an insecticide and a fungicide, the addition of micronutrients to an insecticide, etc. Only mixtures for pest resistance management are considered here.

Pre-formulated mixture products and some tank mixes have proven to be relatively successful in controlling insect pests and in delaying resistance development. However, as with the use of single compounds, mixtures should always be part of a RMP. Successful mixtures or pre-formulated mixture products have been designed for specific situations and only after careful consideration of the cropping system, the effects on beneficial arthropods, and the pest complex. If the target pest population has substantial resistance to any of the components in a pesticide mixture, application of the mixture could exacerbate the situation by selecting for multiple resistance in the pest population.

Pre-formulated mixtures have the advantage that resistance management is built in by the manufacturer. Tank mixes give the user more flexibility but are effective only if the user is able to design them correctly. A fungicide mixture traditionally contains a 'high risk' (of resistance development) fungicide in mixture with a 'low risk' fungicide, the low risk component providing resistance management for the high risk component. However, carefully designed mixtures containing two high-risk components can be very effective if used correctly. The use of *ad hoc* mixtures of insecticides is not encouraged; improperly designed mixtures may not provide any delay in resistance development and may even exacerbate it.

Figure 4 shows how use of an insecticide mixture affects an insect population in which some individuals are resistant (RR) or partially resistant (RS) to one of the two insecticides in the mixture. Individuals not killed by one component of the insecticide mixture will be killed by the other component. This assumes that the number of RRRR individuals is extremely low; they would also survive.

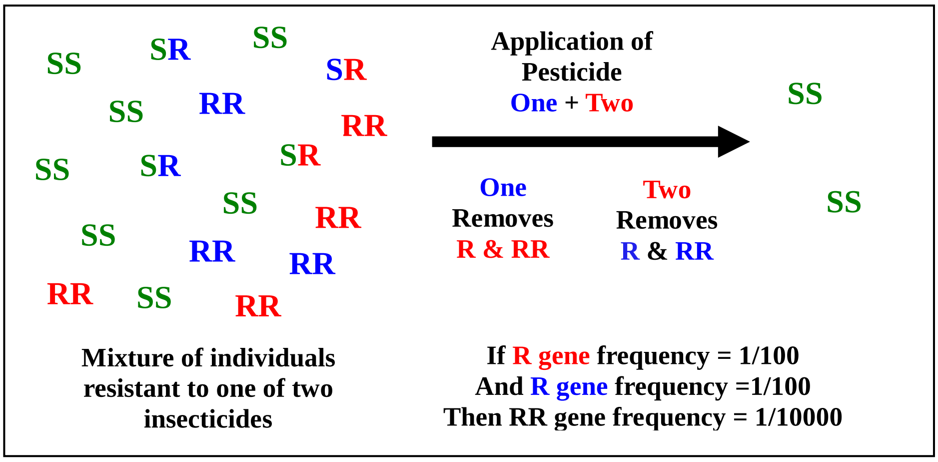

Figure 4 Illustration of the use of mixtures to reduce the accumulation of resistance genes in a pest population.

Pesticide mixtures should be used with care and are not recommended unless the mixture has been carefully researched and meets the following requirements:

- the components of the mixture are not cross-resistant, individuals with resistance to one or the other component are rare, and individuals resistant to both components are extremely rare;
- the mixture is prepared such that both pesticides are applied at their label rate. If the rates applied are only marginally effective, resistance will be much more likely to develop, because the rate used will be insufficient to kill the heterozygote individuals;
- the residual activity of both compounds is nearly the same. Otherwise, the compound with the shorter residual activity will degrade and the component with the longer residual will begin selecting for resistance to it.

Rotations or alternation of pesticides

The alternation of pesticides is another tactic used to manage resistance development.

This tactic assumes that (1) pests resistant to both pesticides are rare, hence survivors of the first pesticide application will be killed by the second, and (2) the percentage of resistant pests will decline in the absence of the pesticide because of the relative instability of the resistance mechanism. For the tactic to be effective, the following requirement must be met:

- the alternating pesticides must belong to unrelated chemical classes and they must not be cross-resistant (see Annex 1 – Modes of Action classifications);

- the two pesticides must be equally effective at their label rates;

- the interval between applications of the rotating pesticides must be long enough for the pest population to return to its original level of susceptibility, as shown in Figure 5 (where Recovery = recovery of susceptibility).

As with pesticide mixtures, alternation programmes for fungicides are often based on the use of one 'high risk' and one 'low risk' pesticide, although programmes containing only 'high risk' pesticides are also possible. This tactic depends on the alternating 'low risk' pesticide eliminating any resistant individuals or isolates that survived the previous applications of the 'high risk' pesticide.

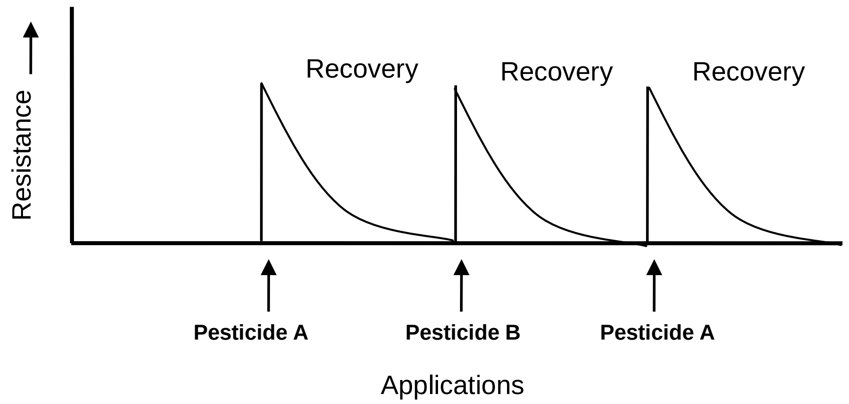

Figure 5 Illustration of the effect of compound rotation on insecticide resistance levels when used in an insecticide resistance management programme.

An example of resistance management using pesticide rotation and biological controls

Onchocerciasis Control Programme in West Africa

In West Africa, the Onchocerciasis Control Programme (OCP) managed by the World Health Organisation (WHO) was almost entirely based on weekly applications of larvicides in rivers to kill the larvae of the blackfly (*Simulium*) vector. Continuous weekly spraying was maintained for at least 15 years over eight countries, thus exerting a very high selective pressure on vector populations. Having rapidly faced very serious problems with resistance to temephos, the only larvicide used in the early stages of the programme, the OCP strengthened resistance monitoring and developed a very efficient resistance management scheme. The scheme replaced the continuous use of a single organophosphate (OP) larvicide, such as temephos, with a pre-planned rotation of unrelated products. OPs were used for limited periods along with a microbial larvicide, *Bacillus thuringiensis israelensis* (*Bti*), a pyrethroid, and a carbamate insecticide. *Bti* and the chemical larvicides were applied strategically, based on the resistance status and trends, vector population dynamics, environmental impact, cost, and logistical factors.

This strategy has been highly successful over the 17 years of its implementation. Resistance regressed to the point where it was possible to re-introduce temephos in the rotation scheme, and resistance to it never developed in areas where it had not previously been present. No resistance developed to any of the other insecticides used, even though they had the potential to select for resistance in blackflies. Extensive use of the microbial larvicide *Bti* a biological means of treating the insects with multiple toxins, has allowed successful resistance management without any measurable medium or long term detectable impact in the biological equilibrium of the treated rivers.

3.4 Fungicide resistance management tactics

A considerable number of tactics are available for managing resistance to fungicides. The tactics vary for different fungicide groups, target pathogens, crops, and geographic areas, but it is often possible and effective to integrate two or more of them together in a RMP. The tactics described below constitute the foundation for a RMP for fungicides. Specific resistance management strategies have been developed for the various fungicide groups.

Implement integrated disease management (integrated pest management)

The integrated use of cultural practices and fungicides is not only economically and environmentally beneficial but is also a major strategy for combating crop disease while avoiding or delaying fungicide resistance. Unfortunately, non-chemical methods of disease control may be weak or unavailable, so that fungicide application is the predominant, or even the sole, countermeasure for many diseases, including potato late blight, grape downy mildew, Sigatoka disease of bananas and wheat bunt.

Integrated disease management includes the following:

Cultural practices

- Use disease resistant crop varieties, biological control agents, and basic hygienic practices such as crop rotation and removal of diseased parts of perennial crop plants, to reduce the incidence of disease.
- Avoid growing large areas of the same variety, particularly if it is known to be susceptible.
- Sterilize soil and equipment to help prevent the spread of pathogenic diseases. This can be especially valid for glasshouse crops.
- Extend crop rotation intervals where possible to avoid the spread of soil-borne pathogens.
- Scout fields frequently to monitor the appearance of disease symptoms before the diseases become established.
- Become familiar with the environmental and crop conditions generally associated with disease development.

Fungicide use

- Apply fungicides only when they are really needed.
- Use fungicides at the label rate and ensure that there is good spray coverage.
- Apply fungicides to reduce the build-up of more virulent pathotoypes that may affect even (previously) disease-resistant crop varieties.
- Do not use soil applications to control foliar diseases.

Use pesticides with different modes of action where possible

The availability of different types of fungicide for each major crop disease is highly beneficial both environmentally and to overcome resistance problems. The continued use of one or a very few classes of compounds over many years presents a much greater risk of side effects and favours resistance in the target organisms.

Use fungicides with different modes of action, or in a rotation or alternation of different fungicide treatments (see Annex 1 – Modes of Action classifications). Mixtures and alternating applications or blocks of treatments of compounds that are at risk of resistance development with an unrelated companion fungicide are often used in fungicide RMPs to broaden the spectrum of the diseases controlled as well as to manage resistance.

The ‘companion’ or ‘partner’ compounds applied as either a mixture or a rotation will reduce the selection pressure exerted by the at-risk fungicide and inhibit the growth of any resistant populations. Generally, good partner fungicides are multi-site inhibitors that are highly effective against the pathogen and that have a low resistance risk. However, it is possible to use a single-site fungicide that is known to be unrelated to its partner by cross-resistance or, in the absence of known resistance, by a similar mode of action. Use of a mixture of two single-site fungicides will carry some risk of selecting dual-resistant strains, but the chance of two mutations occurring simultaneously will be very small compared to that of a single mutation.

Restrict the number of treatments per season, apply only when strictly necessary

This approach, like rotation, reduces the total number of applications of the at-risk fungicide and therefore slows down resistance selection to some extent. It can also favour the decline of resistant strains that have a fitness deficit. However, the delay in resistance may not be proportional to the reduction in spray numbers. This is because the treatments which are still applied generally coincide with the most active stages of epidemics when selection pressures are highest. On the other hand a substantial break in use at a time when the pathogen is still multiplying can allow a beneficial resurgence of more sensitive forms.

Use effective, i.e. recommended, doses

Fungicides must also be applied at the recommended dose in order to ensure their effectiveness under a wide range of conditions. Reducing the dose can enhance the development of resistance.

Avoid eradicant uses

Systemic fungicides can eradicate or cure infections, and this greatly assists their use on a ‘threshold’ basis, where application is made only when an economically unacceptable, amount of disease has already appeared. However, in certain cases, specifically where the fungicide is a mixture of a systemic and non-systemic component, a curative or eradicant treatment is not recommended as it can apply a very high selection pressure to the pathogen. In particular, eradicant use of phenylamides should be avoided, if they are applied for control of foliar diseases as a mixture with a multi-site companion fungicide. The latter does not work as an eradicant, so that the systemic component is acting alone when the mixture is applied to existing infections, which increases selection pressure.

Avoiding the use of fungicides as eradicants can delay resistance for another, more widely applicable reason. To wait until a threshold population of the pathogen appears usually means that many sporulating lesions (occupying up to 5 percent of the foliar area) are exposed to the fungicide. Opportunity for resistance selection is likely to be much greater than if the fungicide had been applied prophylactically to keep the population permanently low.

3.5 Herbicide resistance management tactics

The foundation for herbicide resistance management is use of a sustainable system that integrates physical, chemical, and biological control methods and avoids excessive reliance on any one method. In the short term, any management practice that reduces the selection pressure for resistance, for example changing herbicides, will reduce the rate of development of resistant weeds. But in the mid to long term, it is necessary to have a programme that incorporates crop management and strategic use of chemical and mechanical weed control tools. When employed in an integrated approach, these techniques will help to reduce the selection pressure and significantly reduce the chance of survival of resistant weeds.

Crop management

The following well-established crop management techniques should always be used.

- Rotate crops with different herbicide use patterns and/or growth cycles, to avoid successive crops in the same field which require herbicides with the same mode of action to control the same weed species. Different crops will allow rotation of herbicides with a different mode of action and can avoid or disrupt the growth season of the weed. In addition, crops with different sowing times and seedbed preparation can permit the use of a variety of cultural techniques to manage a particular weed problem. Crops also differ in their inherent competitiveness against weeds, and a strongly competitive crop will have a better chance to restrict weed seed production.
- Delay planting so that initial weed flushes can be controlled with a non-selective herbicide.
- Hand weed, cultivate, or plough before sowing to control emerged plants and bury non-germinated seed. These techniques exert no chemical selection pressure and assist greatly in reducing the soil seed bank.
- Use certified weed-free crop seed.
- Encourage post-harvest grazing, where practical.
- Burn stubble, where allowed, to limit weed seed fertility.
- Cut for hay or silage to prevent weed seed set in extreme cases of confirmed resistance.
- Keep equipment clean of weed seeds to avoid mechanically spreading weed seeds.

Chemical tools: herbicide rotation and mixtures

Numerous studies have reported on the advantages of and need for using multiple herbicide modes of action to prevent the onset of resistance and to address pre-existing resistance for many different crop/herbicide/weed complexes. The sequences studied include: application of herbicide mixtures; post-emergence applications used in sequence on the same crop; pre-emergence applications of soil active herbicides followed by post-emergence active products on the same crop; and the alternation of herbicides in different years/different crops within a crop rotation.

But rotation of herbicides alone is not enough to prevent the development of resistance. The chemical rotation must be employed in association with at least some non-chemical weed control measures. In cases where metabolic resistance is already present, the mode of action of the herbicide is not always the key criterion. In these cases, the mechanism of degradation can be very important and can cut across herbicide groups with different modes of action and chemistries. No classification of herbicides relating to degradation is yet available and such examples need to be handled on a case by case basis.

Products should be chosen from different mode of action groups to control the same weed species either in successive applications or in mixtures. A regularly updated classification of herbicides according to mode of action is available (see Annex 1) and can be useful for planning a weed control programme.

The following guidelines should be followed for herbicide rotation and mixtures.

- Use short residual herbicides.
- Rotate crops with different growth seasons when possible.
- Avoid continued use of the same herbicide or of herbicides having the same mode of action in the same field unless it is or they are integrated with other weed control practices.
- Limit the number of applications of a single herbicide or of herbicides having the same mode of action in a single growing season.
- Where possible, use mixtures or sequential treatments of herbicides that have a different mode of action but are active on the same target weed. For mixtures to be effective, their active ingredients should each give high levels of control of the target weed.
- Use non-selective herbicides to control early flushes of weeds prior to crop emergence.
- Always use post emergence herbicides at the recommended label rate applied at the recommended timing or growth stage of the weed.

Additional resistance management guidance

- Growers should know which weeds infest their fields or non-crop areas and where possible, tailor their weed control programme according to weed densities and/or economic thresholds.
- Follow herbicide label use instructions carefully, particularly the recommended use rates and application timing
- Routinely monitor the results of herbicide applications, being aware of any trends or changes in the weed populations present.
- Maintain detailed field records so that the cropping and herbicide history is known.

3.6 Insecticide resistance management tactics

In managing insect resistance, it is important to keep in mind that the primary objective is to protect the crop or control the vector, not necessarily to kill all the insects. The overall strategy of avoiding overuse of a single insecticidal mode of action should be followed. Additional insecticide resistance management tactics are given below.

Crop-by-pest vs. regional tactics

"Crop-by-pest" resistance management tactics focus on a single crop-pest combination. They can be appropriate when the crop area is large and there is essentially one pest species (e.g. *Helicoverpa* on tomatoes) to be treated with an insecticide.

However, in horticultural and agricultural areas there is often a range of crops and a range of pests. In cases where one or more insecticides with a single MoA are used across this range of crops to control multiple pests that can readily move from crop to crop, the risk of resistance will likely increase. For example, resistance management tactics for diamondback moth on Brassica vegetables could be compromised by widespread use of similar insecticides for diamondback moth control in canola. In addition, the pest complex for a specific crop can vary within production regions and consequently, single crop-by-pest tactics may be flawed.

An alternative to the crop-by-pest tactic are “regional tactics”, where integrated resistance management plans are developed for the several crops and pests in a given geographical area and not just for single crop and pest combinations. Examples are the integrated resistance management strategies for cereals and annual horticultural crops in New South Wales and Victoria, in Australia, or vegetables in Florida, in the USA.

General practices

The following management tactics are recommended to reduce the risk of insecticide resistance developing:

Use an integrated approach

Management of insecticide resistance requires a consideration of all aspects of crop production, including agronomic practices, physical and biological control methods, and insect pest biology. Simply complying with the concepts of integrated crop management can help prevent resistance from developing. For example, monitoring and adhering to recommended pest and/or damage thresholds, respecting the usefulness of natural enemies, carrying out simple sanitation measures, removing post-harvest residues in the field, using resistant crop varieties, and simply avoiding continuous year-round cultivation of a single crop can all help to slow and even prevent resistance development.

Protect beneficial organisms

Protect natural enemies of pests insofar as possible. The contribution of beneficial organisms to pest control can be significant in many cropping systems. Beneficial organisms can also play an important part in resistance management as they help control the target pests irrespective of the pests’ degree of resistance or resistance mechanism, and thus can help slow down the resistance selection process. Natural enemies can be protected for instance by using selective insecticides, avoiding overdosing, or applying non-chemical control options.

Use recommended application rates

Use the recommended rates and treatment intervals as indicated on insecticide labels. Never apply more or less than the recommended rate, as this can result in resistance and/or unwanted effects on non-target organisms and the environment. Always make sure that spray equipment is in good condition, and that nozzles and filters are not blocked, which causes spraying of incorrect rates and can result in resistance development.

Rotate unrelated compounds

Use a variety of compounds registered for the use in question, from unrelated chemical classes that are not cross-resistant; never use a single compound or class.

Use mixtures with caution

Mixtures should be used with extreme caution and are not recommended except in very limited situations, as the incorrect use of mixtures can exacerbate resistance. In particular, mixtures should never be used if the target pest is already resistant to one of the modes of action in the mixture. If mixtures must be used, the active ingredients should be at their recommended application rates and should have similar residual activity to prevent selecting resistance to the component with the longest residual activity.

Use synergists with caution

The use of synergists, which block or delay the metabolic detoxification of insecticides, may improve their effectiveness and extend their useful lifetime if the synergists are applied at a non-toxic rate either before or at the same time as the insecticide (for example, in mixture with the insecticide). Synergists inhibit metabolic enzyme systems that can sequester or break down the insecticide and/or enhance penetration of the insecticide. Inhibition occurs because the synergist binds to metabolic enzymes and allows a larger proportion of the insecticide to reach the target site. Therefore, synergists whose only action is to inhibit metabolic enzymes are not useful if the target site is altered.

Use non-specific products

Plant protection products such as oils and soaps that have a non-specific mode of action are good resistance management tools. Where possible they should be used in rotations or mixtures with conventional insecticides, provided they effectively control both susceptible and resistant target pest populations.

Apply products with care

Apply insecticides when the opportunity for control is optimum, i.e. the infestation has reached the action threshold but is not overwhelming. Ensure that coverage is good. Do not use the same compounds with the same mode of action to control a pest that has several generations in the growing season of the crop.

Monitor problematic pests

Monitor problematic pest infestations in order to detect first shifts in sensitivity. In many instances, baseline sensitivity data for representative field populations were established before the products became widely used. Re-examining the insecticide sensitivity of these populations at regular intervals can reveal possible changes in susceptibility. Resistance monitoring carried out at regular intervals is recommended to detect possible changes in pest sensitivity before serious control problems become evident (see also chapter 4).

3.7 Rodenticide resistance management tactics

The first step is to confirm that cases of suspected resistance are indeed resistance and not just insufficient control, such as under-baiting, or migration. Remember that rodenticide resistance is characterized by the ability of the rodents to continue feeding on bait over an extended time, not by a reluctance to feed on the bait. Confirmation of resistance is best done through the use of standardized methodology. This is necessary because of the variability of rodent species and strains, differences between the response of males and females, and differences in active ingredients.

As with plant pathogens, insects and weeds, resistance management should focus on conserving the rodents' susceptibility, or reducing the phenotypic frequency of resistance to an acceptable level. This can be accomplished by placing the resistant individuals at a selective disadvantage. Unfortunately the classes of rodenticides are quite limited, so class-to-class rotation does not have the same potential for preventing rodenticide resistance as for plant pesticides.

As with other pest organisms, managing resistance in rodents involves using good RMP tactics. The basic strategy includes:

- habitat management, e.g. denial of food, harbourage and water to the rodents;
- providing barriers that will prevent rodents from reaching vulnerable crops, storage areas or buildings;
- control of rodent populations through the proper use of chemical and physical control measures.

When using chemical controls, the following actions will help to avoid the development of resistance in rodent populations.

- Use anticoagulant compounds, in good quality products, labelled for the intended use.
- Inspect all bait stations frequently and replace old bait stations as necessary.
- Follow label directions until the infestation is eliminated.
- Remove all baits once control is achieved.
- Do not use anticoagulants exclusively; permanent stations should be used only where immigration is high.
- Monitor for rodent activity routinely and keep detailed accurate records of treatment.
- Where rodent problems persist, use a variety of control measures, use alternative baits, extend the programme.
- Ensure that the infestation is completely eliminated.

4 Resistance detection and verification

4.1 Objectives of resistance detection and monitoring

When a pesticide appears not to be working as expected, the first step is to identify the problem. There are many causes of product performance problems other than resistance. These include poor application coverage, use of an incorrect rate, misidentification of the pest, adverse environmental conditions, incorrect timing of the application, and so forth. Normal field failures can too readily be attributed to resistance. These other factors should be investigated as well as possible development of resistance.

Resistance detection is the identification of a significant change in the susceptibility of a pest population to pesticides. Resistance can be detected through *ad hoc* observations made by researchers or farmers, or through systematic monitoring. Resistance monitoring attempts to measure changes in the frequency or degree of resistance in time and space. Monitoring can also be used to evaluate the effectiveness of different tactics that are employed to prevent, delay, or manage the development of resistance. Both resistance detection and monitoring are most useful when undertaken early in a resistance episode.

In principle, resistance monitoring should be carried out whenever there is a suspicion or likelihood of resistance development. For example, resistance monitoring programmes should be set up for pests and pesticides where resistance has previously been detected. For pests that have a very high risk of resistance development, a resistance monitoring programme should be established even before resistance has been detected, as an integral part of the RMP. In many countries, resistance detection and monitoring are conducted by national or regional research institutions, although pesticide manufacturers can also be involved.

Table 10 provides a basic scheme for resistance monitoring and shows how it is integrated into the RMP.

Table 10. Phases of resistance monitoring and management for a new pesticide

Timing	Resistance detection and monitoring activities	Other management activities	Actor
1-2 years before start of sales	Establish sampling and testing methods	Assess risk	Pesticide industry
	Survey for initial sensitivity data	Decide strategy of use; develop RMP	
During years of use	Monitor randomly in treated areas for resistance, if justified by risk assessment of special importance of crop/pest	Implement the RMP; watch practical performance of the pesticide closely	Research institutions, extension/advisory services, (large) pesticide users, pesticide industry
As soon as signs of resistance have been detected	Monitor to determine the extent and practical significance of resistance	If resistance problem is confirmed, review and modify RMP	Research institutions, pesticide industry
	Study cross resistance, fitness of variants of resistant organisms, assess other factors affecting the development of resistance		
Subsequently	Monitor rate of spread or decline of resistance	Watch pesticide performance; review RMPs	Research institutions, pesticide industry

Source: Adapted from NRC (1986)

Resistance development is extremely variable and is not uniform throughout an organism's range, because there are so many factors that affect the organism, the host and the pesticide application programme. Even if resistance is documented in one area, this does not necessarily warrant the removal of the pesticide overall. In addition, the detection of resistant individuals does not necessarily indicate that the pest population is resistant and uncontrollable. However, it does provide an early warning that the RMP should be adjusted in order to prevent the frequency of the resistance gene from increasing in the population and creating problems.

4.2 Resistance verification methods

Regardless of the pesticide in question, i.e. fungicide, herbicide or insecticide, there are several methods and requisites for confirming resistance in a particular organism. These include:

Discriminating dose assay

The discriminating or diagnostic-dose assay has been the most widely used method for monitoring resistance in the field, in particular for insecticides. It is easy and relatively resource efficient. The goal of the discriminating-dose assay is to determine whether the status of the population's susceptibility has changed. However, it is generally not possible to detect resistant individuals until the resistance gene frequency is greater than 1 percent.

The three important considerations for designing a single, discriminating-dose monitoring program are:

1. establishing the “diagnostic dose” to separate susceptible from resistant individuals;
2. determining the sample size to be collected at each location;
3. determining the appropriate response to a survivor of the discriminating dose.

Data can be generated from bioassays of survivors in the area that was treated, assuming it was not quickly treated with another compound.

These bioassay tests should be developed before, or soon after, a new compound is commercialized on the target pest(s), or planting of a new transgenic crop. This is often done by pesticide manufacturers, in collaboration with national or regional research institutions. The tests will be used to establish a baseline that can be used to identify the natural variability of susceptibility in the pest population and confirm resistance situations in the future. Tests should be robust, rapid and relatively easy to conduct. The procedure should be accurate and provide realistic, quantitative, reproducible and readily understandable results.

Standardized test methods that can be used to measure susceptibility can be found in the links provided in Annex 1. Ready-made resistance monitoring bioassay kits are available for important pest organisms (e.g. malaria mosquitos).

Dose-response test

The most precise method to assess the susceptibility of a population to a compound or trait is the classical dose–response bioassay. Initially, dose-response data, with a series of doses that produce mortality ranging from 5 to 95 percent in the case of insecticides and 0 to 100 percent in the case of herbicides, should be developed on a number of population samples. For herbicides, only the population in question and a population known to be susceptible need to be tested. These data can be used to determine the range of susceptibility in the population before large scale applications are made. This information can be useful later on when less than expected control is encountered.

Biochemical and immunological tests

Increasingly, biochemical tests for identifying unique detoxification enzymes associated with resistant pests are being used in the survey of both resistant individuals and populations. Immunological tests for resistance based on identification of detoxification enzymes using monoclonal antibodies have also been developed.

Baseline data

Baseline data on the pest organism’s susceptibility to the pesticide need to be collected, ideally before the introduction of the product in a given area. Irrespective of the resistance verification method used, the outcome of the tests is always compared to the baseline.

For insecticides, laboratory strains are often used to establish baseline susceptibility values. These values are of some use because they may provide information on the highest susceptibility that may be observed. However, many of these laboratory populations are actually more susceptible than any field populations because they are weakened by the rearing process. If the range of baseline values is large, this indicates there is considerable genetic diversity within the target organism population and resistance may develop more rapidly than if the range of baseline values is quite small.

For fungicides it is normal to use untreated, unexposed, field isolates to generate a sensitivity baseline. When using field populations, samples should be collected from as wide a geographic area as possible to provide a realistic view of the overall variability of the natural population. Most likely, the baseline will be a range of values rather than one absolute value against which data generated after the introduction of the pesticide can be measured. Fungicide sensitivity baselines are not usually equally distributed but are distinctly skewed to include a low proportion of individuals with EC_{50} values much higher than the mean. Such individuals are natural components of the sensitivity spectrum and are not classed as resistant; they are controlled by the normal fungicide application. Further guidance on the establishment of fungicide baselines is provided elsewhere.

For weeds, populations that are not resistant are needed to be compared to populations suspected of being resistant.

Relationship between bioassay results and field performance

As soon as possible, the correlation between the bioassay results and field performance should be established. This will allow an estimation to be made of a decline in pest susceptibility and field performance. With some compounds a small change in susceptibility, as determined by bioassays, will have a substantial impact on product field performance. With other compounds large differences in susceptibility are required before effects on field performance are observed.

4.3 Test procedures

Validated test procedures exist to assess and confirm pesticide resistance for a large variety of pests, weeds and insects. A number of these tests can be found on the various resistance action committee websites, FRAC, HRAC, and IRAC, as well as the World Health Organization (WHO) websites. These are listed in Annex 1.

5 Resistance and transgenic crops

5.1 Introduction

Transgenic crops, transformed by the insertion of one or more genes, have several advantages with respect to resistance management. One important advantage is that in transgenic plants the decline in the toxin concentration over time is minimal and rates that could cause selection pressure occur only once towards the end of the season. By contrast, with conventional pesticides the dose of pesticide available may vary between plants and over time because of coverage problems and degradation of the active ingredient. This makes repeat pesticide applications necessary and often results in many selection events. The potential for resistance selection with transgenic plants is by comparison much reduced although it is not entirely eliminated.

Although some resistance to a *Bt* toxin in the field has been reported, only a few resistance-related failures of pest control resulting from the cultivation of transgenic crops have been observed to date. Rigorous adherence to RMPs will be needed to prevent it happening in the future. Insect resistance to the *Bt* toxin could have serious effects on crop production. The weed resistance to glyphosate that is being observed in herbicide tolerant crops is essentially a normal case of herbicide resistance (i.e. it would happen for non-transgenic plants as well), but the number of resistant weed species is increasing globally. Widespread development of glyphosate resistance could be a serious threat to transgenic crops with glyphosate tolerance.

5.2 The history of resistance development in Bt crops

When the first transgenic plants incorporating a *Bt* toxin were being developed in the mid-1990s, there was considerable concern that resistance to the *Bt* toxin would develop. In fact, there were predictions that resistance would develop in as little as 3 to 5 years. The predictions were based on observations that:

- resistance had developed to sprayable formulations of *Bt by Plutella xylostella* in the field, thus demonstrating that resistance could develop to *Bt***;**
- resistance to *Bt*-containing insecticides and individual *Bt* Cry proteins had been selected in the laboratory;
- a consistent high dose of the toxin would be present in the plant for a considerable time resulting in very high selection pressure over multiple generations;
- the toxin would be expressed throughout the plant and the season. In addition, the transgenic crops would be widely grown thus providing very few opportunities for the dilution of any resistance genes that might be selected;
- the toxin would be in the plants from the time of germination onward thus providing a preventive rather than a curative situation. In many instances pest population levels would be below the treatment threshold and it was believed this would create unneeded selection events;
- the *Bt* toxin was essentially the only active ingredient and there were a number of resistance mechanisms possible, any one of which could be selected resulting in a *Bt* resistant pest insect population.

Actual field experience with *Bt* cotton over the last decade has demonstrated that the risk of resistance development was much lower than originally predicted. Thus far *Bt* resistance genes have only been found at low frequencies, and only limited field resistance problems have been reported so far. An important reason is the robust resistance management plans that were required for product registration. The integration of agronomic practices, bio-control methods, conventional foliar pesticides, and other integrated pest management tactics with *Bt* crops has helped to prevent resistance development. In addition, the following factors have undoubtedly contributed:

- there are relatively few areas where *Bt* crops are dominant over the entire crop area, with the exception of some intensively grown corn and cotton areas where there are large areas of *Bt* crops;
- in these areas several of the key pests have a broad range of hosts and long-range dispersal ranges, such that only a portion of the population is exposed and selected for *Bt* resistance. In addition, the RMPs have required *Bt* crops that express a single *Bt* protein to have a structured refuge of non-*Bt* crop planted next to the *Bt* crop. These non-*Bt* varieties and crops ensure the survival of large numbers of susceptible individuals in the target species;
- in insects the genes conferring resistance tend to be functionally recessive and associated with high fitness costs. So far it has been difficult to find large numbers of resistant larvae that can complete development and reproduce on *Bt* crops. In instances where *Bt* resistant populations were reared from field samples, the colonies have not survived beyond a few generations;
- the toxin is present in the *Bt* plant at a high rate, typically at least high enough to control

heterozygous insect populations in most target pests, and it persists in the plant for most of the season. This makes it more difficult for resistance to be selected than if there were repeated selections due to frequent pesticide treatments (in which product residues were declining to suboptimal levels before retreatment). In addition, a number of different *Bt* proteins with unique sites of action (i.e. insect mid-gut receptors) have now been deployed.

These factors help to explain why it has been difficult for the various pest species to develop vigorous *Bt* resistant populations. Until 2012 , field resistance (including crop failure) to Bt crops had only been documented in fall armyworm (*Spodoptera frugiperda*) in Puerto Rico, African stem borer (*Busseola fusca*) in South Africa, pink bollworm (*Pectinophora gossypiella*) in India and most recently, western corn rootworm *(Diabrotica virgifera virgifera)* in the United States. There are indications that these cases were at least partly the result of a failure to strictly follow the generally recommended RMPs for these crop-pest combinations.

It is clear that the continued viability of *Bt* crops will depend on the development and use of robust resistance management plans. It will be important to remember that resistance risk is not uniform for all products and use patterns. It cannot be assumed that experiences with *Bt* in new transgenic crops will necessarily be similar.

5.3 Tactics for preventing development of resistance to Bt toxins

Tactics for managing resistance to Bt crops are generally the same as for conventional pesticides, but with the addition of tactics to preserve genes for susceptibility in pest populations. The main tactics currently used for Bt crops include the following.

- *Crop management practices*: as for conventional pesticides, use of good crop management and integrated pest management is the foundation of resistance management. In addition to reducing the number of pesticide applications needed, good crop management helps to preserve populations of predatory and parasitic insects. Chances are good that these beneficial species will eliminate any surviving pests that remain on a transgenic crop.
- *Target pest spectrum and dose:* some insect species are more sensitive than others to the *Bt* protein and the *Bt* protein may not be expressed equally throughout the plant. As long as the *Bt* protein is expressed in the critical plant tissues and the dose is sufficient to kill all susceptible target pest populations, resistance selection would be expected to proceed very slowly. Conversely, if the level of toxin is low enough to allow some survivors including heterozygotes, resistance may evolve more rapidly.
- *Refuges for susceptible insect pests*: the provision and/or preservation of non-Bt crop refuges has been a requirement of most Bt crop resistance management plans. Placed alongside or even within the Bt crops, the refuges permit the survival of a sufficient number of susceptible pests to maintain their genes in the overall pest population. The movement of insect larvae and adults will dictate the placement of non-*Bt* crop refuges. For example, because larvae of the European corn borer (*Ostrinia nubilalis)* move readily along but not between rows of maize, an in-field refuge is the best solution (*e.g.,* eight rows of *Bt* maize, followed by two rows of non-*Bt* maize). For cotton, however, where the target pests move both along and between rows, the refuge is planted in a block and not in rows. External refuges must be close enough to the *Bt* crop to allow for random mating of the adult insects. In cotton, because the target pests (especially heliothines) are so mobile, migration from other non-*Bt* crops also provides a significant number of susceptible insects.

- *Choice of Bt crop*: attention should be paid to the type of Bt proteins present in different Bt crops, as migrating insect populations may be selected for resistance to the same or similar Bt proteins found in different crops in other growing regions. For this reason, some countries have taken steps to limit a particular Bt protein to a specific crop. Another approach is to use crops in which two or more Bt proteins with unique binding sites have been inserted into the same plant, called pyramiding. It is very unlikely that an insect could develop resistance to two different toxins. Because of the reduced resistance risk, and because the target pests are so mobile and readily migrate in from nearby non-Bt crops, a natural refuge has been allowed by the US Environmental Protection Agency for Bt cotton expressing two Cry proteins, instead of the structured refuge required for transgenic cotton with only a single Bt protein.

The strengths and limitations of various resistance management tactics used with Bt and other transgenic crops are shown in Table 11.

Table 11 Strengths and limitations of resistance management tactics for use with insect resistant transgenic crops

Tactic	Strengths and limitations
High dose to control heterozygotes	Uniform high dose is achieved against primary target pests, when possible.
Structured refuge for susceptible insects	Successfully implemented in several countries, but often are complicated and costly to deploy.
Unstructured/natural refuge (= alternate hosts)	Only significant when primary target pests are generalists.
Rotation of active ingredients	Not possible within a season, and complicated and costly to implement and verify across seasons.
Pyramided active ingredients	A successful strategy as long as the two toxins are unique in their sites of action and active against the same insect species. May also expand the insect activity spectrum.
Limit of overall area grown with transgenic crops in a given region (acreage cap)	Limited cases of successful implementation in the Philippines and Australia; may be impossible to manage in some systems.
Integrated pest management	Targeted cultural, biological and chemical IPM tools can significantly reduce the survival of resistant populations.
Monitoring of insect susceptibility	If conducted properly can measure small shifts in insect susceptibility before large-scale field failure. Collection of insect populations and insect bioassays can be difficult. Monitoring for unexpected field damage is also extremely valuable.
Stakeholder education and communication	Growers and other stakeholders should be informed of the choice of Bt crops and the importance of resistance management tactics. If a structured refuge is required, grower compliance should be monitored.
[Sources: Ferré et al. (2008)]	

6 Resistance and disease vectors

While pesticide resistance is a major problem in agriculture, it is also an important problem in the control of insect vectors that transmit diseases to humans and their livestock. Important vector-borne diseases at risk of insecticide resistance include malaria, dengue fever, leishmaniasis, and Chagas disease, among others. The severity of the diseases and the relatively few insecticides available for their control make resistance a very important issue.

The principles of resistance prevention and management in disease vectors are the same as in agriculture, but the specific practices may be different. These go beyond the objectives of the present guidelines, so readers are referred to WHO and IRAC for further information on resistance risk, detection, and management for disease vectors (see Annex 1).

An issue of particular concern, however, is the increased resistance selection pressure on vectors of human disease resulting from insecticide use in agriculture. This occurs when the insecticides applied in vector control are also used on a large scale in agriculture in the same area. Close collaboration between the agricultural and the health sectors is required to manage such risks and the elaboration of joint RMPs is recommended.

Annex 1 – Further reading and resources

Some selected further reading on the topics that have been treated in this guideline is provided. Note that certain references below may cover various topics [noted between square brackets].

Resistance risk assessment and risk factors

General

OEPP/EPPO 2002. *Resistance risk analysis*. Standards for efficacy evaluation of plant protection products, PP 1/213(2). European and Mediterranean Plant Protection Organization, Paris. (At: http://pp1.eppo.org/getnorme.php?n=213)

Fungicides

Brent, K.J. & Hollomon, D.W. 2007a. *Fungicide resistance: The assessment of risk.* FRAC Monograph 2 (revised). (At: http://www.frac.info/frac/index.htm)
[note: also “detection and verification”]

Herbicides

HRAC. Undated. *Herbicide cross resistance and multiple resistance in plants.* Monograph. Herbicide Resistance Action Committee. (At: http://hracglobal.com/Publications/HerbicideCrossResistanceandMultipleResistance.aspx)

Insecticides

Whalon, M.E., Mota-Sanchez, D & Hollingworth, R.M. (eds.) 2008. *Global pesticide resistance in arthropods.* CABI, Wallingford.)
[note: also “resistance prevention and management” and “transgenic crops”]

Rodenticides

Buckle, A.P, Prescott, C. V. & Ward, K.J. 1994. Resistance to the first and second generation anticoagulant rodenticides – A new perspective. *In:* W.S. Halverson & A.C. Crabb, eds. *Proc.16th Vertebrate Pest Conference*. pp. 137-144.Univ. of California, Davis. (At: http://digitalcommons.unl.edu/cgi/viewcontent.cgi?article=1006&context=vpc16)

Mode of action classifications

Fungicides

FRAC. 2011 FRAC Code list: (At : http://www.frac.info/frac/publication/anhang/FRAC%20Code%20List%202011-final.pdf)

Herbicides

HRAC. undated. Classification of herbicides according to site of action. (At: http://www.hracglobal.com/Publications/ClassificationofHerbicideSiteofAction.aspx)

Insecticides

IRAC. 2011. IRAC MoA Classification Scheme (At: http://www.irac-online.org/teams/mode-of-action/)

Databases of verified resistance problems and reporting

Fungicides

FRAC. 2011. FRAC list of plant pathogenic organisms resistant to disease control agents. (At: http://www.frac.info/frac/publication/anhang/List%20of%20resistant%20plant%20pathogens_Jan%202011.pdf)

Herbicides

ISHRW. Undated. International Survey of Herbicide Resistance Weeds.(At: http://www.weedscience.org/in.asp)

Insecticides

MSU. Undated. Arthropod Pesticide Resistance Database. Michigan State University. (At: http://www.pesticideresistance.org/)

Resistance prevention and management

General

NRC. 1986**.** *Pesticide Resistance: Strategies and Tactics for Management.* Board on Agriculture, National Research Council. National Academies Press, Washington, DC (At: http://www.nap.edu/openbook.php?record_id=619&page=313)
[note: also "risk assessment and risk factors" and "detection and verification"]

Fungicides

Brent, K.J. & Hollomon, D.W. 2007b. *Fungicide resistance in crop pathogens: How can it be managed?* FRAC Monograph No. 1 (revised edition). Fungicide Resistance Action Committee, Basel. (At*:* http://www.frac.info/frac/index.htm)

Damicone, J. 2007. *Fungicide resistance management.* Oklahoma Cooperative Extension Fact Sheet F-7663. Division of Agricultural Sciences and Natural Resources, Oklahoma State University. (At: http://pods.dasnr.okstate.edu/docushare/dsweb/Get/Document-2317/EPP-7663web.pdf)
[note: also "risk assessment and risk factors"]

Herbicides

HRAC. 2011. *Guideline to the management of herbicide resistance.* Herbicide Resistance Action Committee (Available at: http://www.hracglobal.com/Publications/ManagementofHerbicideResistance.aspx)

Palou, A.T., Ranzenberger, A.C., & Larios C.Z. 2008. *Management of herbicide-resistant weed populations – 100 questions on resistance*. Food and Agriculture Organization of the United Nations, Rome. (At: ftp://ftp.fao.org/docrep/fao/011/a1422e/a1422e00.pdf)

Valverde, B.E. 2003. *Herbicide resistance management in developing countries*. Food and Agriculture Organization of the United Nations, Rome. (At: http://www.fao.org/DOCREP/006/Y5031E/y5031e0h.htm)

Rodenticides

CropLife. 2003. Anticogulant resistance management strategy for pest management professionals, central and local government and other competent users of rodenticides. Technical Monograph. CropLife International, Brussels (Available at: http://www.rrac.info/downloads/technical_monograph_2003_ARM.pdf)

Insecticides

Onstad, D.W. (ed.) Insect resistance management: Biology economics and prediction. Elsevier, Amsterdam
[note: also “risk assessment and risk factors” and “detection and verification”]

Resistance detection and verification

Fungicides

FRAC. Undated. Monitoring methods to investigate possible development of resistance. Fungicide Resistance Action Committee (FRAC) (At: http://www.frac.info/frac/index.htm)

Herbicides

HRAC. 1999. *Detecting herbicide resistance*. Herbicide Resistance Action Committee. (Available at: http://www.hracglobal.com/Publications/DetectingHerbicideResistance.aspx)

Insecticides

IRAC. undated. Insecticide and acaricide resistance monitoring methods. Insecticide Resistance Action Committee (IRAC). (At: http://www.irac-online.org/teams/methods)

WHOPES. Undated. Test procedures for monitoring resistance in disease vectors. WHO Pesticide Evaluation Scheme. (At: http://www.who.int/whopes/resistance/en)

Rodenticides

CropLife. 2003. A reappraisal of blood clotting response tests for anticoagulant resistance and a proposal for a standardised BCR test methodology. Technical monograph. (At: http://www.rrac.info/releases_01.htm)

Prescott, C.V., Buckle, A.P., Hussain, I., Endepols, S. 2007. A standardised BCR-resistance test for all anticoagulant rodenticides. *Int. J. Pest Mgt*. 53(4): 265-272.

Resistance and transgenic crops

Carrière, Y., Dennehy, T.J., Pedersen, B., Haller, S., Ellers-Kirk, C., Antilla, L., Yong Biao, L., Willott, E. & Tabashnik, B.E. 2001 Large scale management of insect resistance to transgenic cotton in Arizona: Can transgenic insecticidal crops be sustained? *J. Econ. Entomol.* 94(2): 315-325. (At: http://esa.publisher.ingentaconnect.com/content/esa/jee/2001/00000094/00000002/art00001)

Ferré, J., Rie, J.V. & MacIntosh, S.C. 2008. Insecticidal genetically modified crops and insecticide resistance management (IRM). *In*: J. Romeis,A. M. Shelton & G. Kennedy. eds. *Integration of insect-resistant genetically modified crops within IPM programmes*. Progress in Biological Control, Vol. 5. Springer, Dordrecht.

Gassmann, A. J., 2012. Field-evolved resistance to Bt maize by western corn rootworm: Predictions form the laboratory and effects in the field. *J. Invert. Path. 110 (2012) 287-293.*

MacIntosh, S.C. 2009. *Managing the risk of insect resistance to transgenic insect control traits*: Practical approaches in local environments, Insecticide Resistance Action Committee, Brussels. (At: http://www.irac-online.org/content/uploads/2009/09/SC-MacIntosh-IRM-manuscript.pdf)

Bates, S.L., Ahao, J., Roush, R.T. & Selton, A.M.. 2005. Insect resistance management in GM crops: past, present and future. *Nature Biotechnology* 23(1): 57-62.

Tabashnik, B.E., van Rensburg, J.B.J. & Carrière, Y. (2009) Field-evolved resistance to Bt crops: definition, theory and data. *Journal of Economic Entomology* 102: 2011-2025 (At: http://docserver.ingentaconnect.com/deliver/connect/esa/00220493/v102n6/s1.pdf?expires=1346048945&id=0000&titleid=10264&checksum=0256E56CD08BF19CE865F2A3A09E4357)

Huang, F., Andow, D.A. & Buschman, L.L. (2011) Success of the high-dose/refuge resistance management strategy after 15 years of *Bt* crops use in North America. *Entomologia Experimentalis et Applicata* 140:1-16 (At: http://onlinelibrary.wiley.com/doi/10.1111/j.1570-7458.2011.01138.x/pdf)

Resistance and disease vectors

Brogdon, W.G. & McAllisterm, J. C. 1998. Insecticide resistance and vector control. *Emerging Infectious Diseases* 4(4): 517-713. (At: http://www.cdc.gov/ncidod/eid/vol4no4/brogdon.htm)

IRAC. 2011. *Prevention and management of insecticide resistance in vectors and pests of public health importance*. 2nd edition. Insecticide Resistance Action Committee (IRAC), Brussels.. (At: http://www.irac-online.org/wp-content/uploads/2009/09/VM-Layout-v2.6_LR.pdf)

Knobler, S.L., Lemon, S. M., Najafi, M., & Burroughs, T. (eds). 2003. *The resistance phenomenon in microbes and infectious disease vectors: Implications for human health and strategies for containment.* Chapter 3 – Vector resistance. National Academies Press, Washington, D.C. (At: http://books.nap.edu/openbook.php?record_id=10651&page=88)

Annex 2 – Examples of actual resistance management plans

Some examples of actual resistance management plans (RMPs) for certain crops or pesticide groups are provided below. The list is not exhaustive and should be considered as indicative only. FAO does not take responsibility for individual RMPs. As stressed in the text, RMPs need to be developed for the specific situation under which the pesticide is used.

Fungicides

General information

- http://pods.dasnr.okstate.edu/docushare/dsweb/Get/Document-2317/F-7663web.pdf
- http://www.croplifeaustralia.org.au/default.asp?V_DOC_ID=1953
- http://www.cottoncrc.org.au/content/Industry/Publications/Pests_and_Beneficials/Insect_Resistance_Management.aspx

Oil Seed Rape

- http://www.pesticides.gov.uk/guidance/industries/pesticides/advisory-groups/Resistance-Action-Groups/frag

Potatoes

- http://www.potatodiseases.org/pdf/Fungicide-Resistance-Management.pdf
- http://www.extension.umn.edu/AgProfessionals/components/CPM/Stevenson_Fungicides.pdf

Tree fruit

- http://tfpg.cas.psu.edu/56.htm

Herbicides

General information

- FAO – Management of herbicide-resistant weed populations:

  ftp://ftp.fao.org/docrep/fao/011/a1422e/a1422e00.pdf
- http://www.croplifeaustralia.org.au/default.asp?V_DOC_ID=1954
- http://www.dpi.qld.gov.au/cps/rde/xchg/dpi/hs.xsl/26_4240_ENA_HTML.htm
- http://www.dpi.qld.gov.au/cps/rde/xchg/dpi/hs.xsl/26_4239_ENA_HTML.htm
- http://www.croplifeaustralia.org.au/default.asp?V_DOC_ID=1854,
- http://www.croplifeaustralia.org.au/files/resistancemanagemen/herbicides/2010%20Herbicde%20Resistance%20Management%20Strategies.pdf

Cotton

- http://cottoninfo.ucdavis.edu/Production_Guidelines/
- http://www.cotton.org/tech/pest/upload/07CIweedresistbulletin.pdf

Maize

- http://www.nwnyteam.org/Corn%20Congress%20Presentations/Herbicide%20Resistance%20Management%20Sstrategies.pdf

Transgenic maize

- http://text.lsuagcenter.com/NR/rdonlyres/FC8C9299-F8CA-4F99-869D-f3EB0FB0B5502/45400/pub2963herbicideresistancecotton2008HIGHRES.pdf

Insecticides

General information

- http://www.croplifeaustralia.org.au/default.asp?V_DOC_ID=1955

Cotton

- http://www.cottoncrc.org.au/industry/Publications/Pests_and_Beneficials/Insect_Resistance_Management

Brassica vegetables

- http://www.sardi.sa.gov.au/__data/assets/pdf_file/0005/91616/irm_flyer_sept_2008.pdf

Greenhouses

- www.entomology.umn.edu/cues/4015/ppts/greenhouseRM.ppt

Oil seed rape

- http://www.irac-online.org/news/updated-monitoring-and-irm-guidelines-in-oilseed-2
- http://www.pesticides.gov.uk/guidance/industries/pesticides/advisory-groups/Resistance-Action-Groups/frag

Ornamentals

- http://solutionsforyourlife.ufl.edu/hot_topics/agriculture/whiteflies.html#resistance

Mixed crops (cotton, melons and vegetables)

- http://www.cals.arizona.edu/pubs/insects/az1319.pdf

Potatoes

- http://www.nationalpotatocouncil.org/NPC/p_documents/document_280607084102.pdf
- http://www.hort.uconn.edu/IPM/veg/htms/cpbipm.htm
- http://www.pesticides.gov.uk/guidance/industries/pesticides/advisory-groups/Resistance-Action-Groups/irag

Row crops

- http://www.pesticides.gov.uk/guidance/industries/pesticides/advisory-groups/Resistance-Action-Groups/irag

Strawberries

- http://www.ipmcenters.org/pmsp/pdf/CASTRAWBERRY.PDF

Annex 3 – Expert groups

Expert groups on resistance

International – Resistance Action Committees (RACs)

Expert groups of CropLife International composed of experts from pesticide industry.

- Fungicide Resistance Action Committee (FRAC): http://www.frac.info/frac/index.htm
- Herbicide Resistance Action Committee (HRAC): http://www.hracglobal.com/
- Insecticide Resistance Action Committee (IRAC): http://www.irac-online.org/
- Rodenticide Resistance Action Committee (RRAC): http://www.rrac.info/

UK – Resistance Action Groups

UK-based Resistance Action Groups composed of experts from pesticide industry and independent organizations.

- http://www.pesticides.gov.uk/guidance/industries/pesticides/advisory-groups/Resistance-Action-Groups

Australia – Resistance Management Review Groups

Expert groups of CropLife Australia composed of experts from pesticide industry

- http://www.croplifeaustralia.org.au/default.asp?V_DOC_ID=1952

Expert groups – Commodity and other groups

- Entomological Society of America – Resistance information available at: http://www.entsoc.org/Search/default.aspx (Enter *resistance* in search box)
- Weed Science Society of America – Resistance information available at: http://www.wssa.net/00Search/search.php?zoom_query=herbicide+resistance
- European Weed Society – Resistance information available at: http://www.ewrs.org/herbicide_resistance.asp
- National Cotton Council – Resistance information available at: http://www.cotton.org/search.cfm (Enter *insecticide resistance* or *herbicide resistance* in the search box).
- WERA060: Management of Pesticide Resistance (from WERA60) http://nimss.umd.edu/homepages/home.cfm?trackID=9616