We are IntechOpen,
the world's leading publisher of
Open Access books
Built by scientists, for scientists

6,900
Open access books available

185,000
International authors and editors

200M
Downloads

154
Countries delivered to

Our authors are among the
TOP 1%
most cited scientists

12.2%
Contributors from top 500 universities

CLARIVATE ANALYTICS
BOOK CITATION INDEX
INDEXED

WEB OF SCIENCE™
Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

Chapter 18

Biochemical Insecticide Resistance in Tea Pests

Dhiraj Saha

Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/61949

Abstract

Polyphagous insect herbivores encounter numerous toxins (xenobiotics) as they pass through their life cycle; some toxins are produced naturally by the host plants (allelochemicals) and others by humans (insecticides) to manage these insects having pest status. The host plants have evolved defensive mechanisms for protection from herbivory, including chemical repellents and toxins (secondary metabolites). Many classes of insect repellents and toxic substances, such as isoflavonoids, furanocoumarins, terpenoids, alkaloids and cyanogenic glycosides are synthesized in plants. The biosynthetic pathways leading to these allelochemicals are continually evolving to generate new secondary metabolites. Similarly, to control the herbivorous insect pests, numerous chemicals of synthetic origin are used continuously against them. In response, the attacking organisms also evolve mechanisms that enable them to resist the defensive chemicals of their hosts and those toxins of synthetic origin applied for their control. A variety of defence mechanisms, including enzymatic detoxification systems, physiological tolerance and behavioural avoidance, protect insect herbivores from these xenobiotic compounds. Insect pests have evolved the mechanisms to degrade metabolically (enzymatically) or otherwise circumvent the toxic effect of many types of chemicals that we have synthesized as modern insecticides. The extent to which insects can metabolize and thereby degrade these antibiotics or toxins is of considerable importance for their survival in hostile chemical environment. These mechanisms continue to evolve as insects attempt to colonize new plant species or encounter newer molecules of synthetic insecticides. Generally, three main enzymes, general esterases (GEs), glutathione S-transferases (GSTs) and cytochrome P450-mediated monooxygenases (CYPs), are involved in the process of metabolic detoxification of insecticides. During the past 70 years, following the discovery and extensive use of synthetic insecticides, resistance of insects to insecticides has registered the greatest increase and strongest impact. The evolution of resistance to insecticides is an example of evolutionary process. An insecticide is the selection pressure, which results in a very strong but differential fitness of the individual in a population having susceptible and resistant genotypes. The survival and subsequent reproduction of resistant individuals lead to a change in the frequency of alleles conferring resistance in the population over

INTECH
open science | open minds

© 2016 The Author(s). Licensee InTech. This chapter is distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/3.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

time. While selection pressure acts to change allele frequencies within pest populations, the phenotype upon which selection operates is a function of both genotype and the environment. Recent studies in insect detoxifying enzymes have revealed further versatility in the adaptation of insects to their environment by the phenomenon of induction. This is the process in which a chemical stimulus enhances the activity of the detoxification enzyme systems by the production of additional enzymes that metabolize toxic chemical substances. Hence, the influence of environmental factors such as continuous usage of insecticides and the chemical constituents (allelochemicals) of host plants on phytophagous insects can have a great impact to induce the enzymatic detoxification systems of insects, thereby promoting the insecticide resistance mechanisms. While all insects do possess detoxification ability, its magnitude is expected to vary among the species with the nature of its recent environment and feeding ecology. The level and type of detoxifying mechanisms differ greatly, which therefore result in varying toxicity among different developmental stages, species and populations. Variation in detoxifying enzyme activity is responsible in part for the selective toxicity of different insecticides, the development of resistance to insecticides and selective adaptation to host plants. Over-expression of these detoxifying enzymes, capable of metabolizing insecticides, can result in a high level of metabolic tolerance/resistance to synthetic insecticides. Increased expressions of genes encoding the major xenobiotic metabolizing enzymes are the most common cause of insecticide resistance in insects.

Keywords: Insecticide resistance, tea, insect pests, detoxifying enzymes, cytochrome p450, carboxylesterases, glutathione S-transferases, monooxygenases, allelochemical, *Helopeltis theivora*, *Scirtothrips dorsalis*, *Empoasca flavescens*

---

1. Introduction

1.1. Tea, the plantation crop with economic value: Driving the shrub to cup

Tea is produced from young leaves and buds of tea plant, *Camellia sinensis* (L.) O. Kuntze is a native of China, and the Chinese are said to have discovered its use nearly 4,700 years ago. It is believed that Shen Nung, a Chinese emperor who lived some 4,700 years ago, discovered that tea leaves falling into boiling water make a refreshing drink. Tea then became a popular drink in China for both its flavour and medicinal qualities. Eventually, the habit of drinking tea spread throughout Asia and then throughout the world. Currently, tea is the second most preferred and popular drink after water. The word tea had its origin from t'e (pronounced as 'tay') in 'Amoy' dialect while in Cantonese it was called ch'a ('chah'). This is the name by which this wonderful beverage is known in Japan, Iran, Russia, Indonesia, Malaysia, Vietnam and India. Tea was introduced in Japan about AD 800 and was regarded as medicine for about 500 years. Tea was introduced in Europe in the early 17th century with the beginning of trade between Europe and the Southeast Asia.

Now tea is produced in almost every region of the globe. The tea plant is predominantly grown in Asia followed by in Africa and to a very small extent in Europe, South America, Australia and New Zealand. Now, tea is grown in 36 tropical and subtropical countries. Major tea producing countries are India, China, Sri Lanka, Kenya, Japan, Indonesia, Thailand, Bangladesh, Nepal, Vietnam, Turkey and Argentina.

Tea plants are native to East and South Asia and probably originated around the point of confluence of the lands of northeast India, north Burma, southwest China and Tibet. The commercially cultivated tea plants are derived from small-leaved China plants, *C. sinensis*, the Assam plants, *C. assamica* (Masters), and the Cambod plants, *C. assamica* ssp. *lasiocalyx* (Planchon ex Watt) Wight and numerous hybrids among them. Tea plants require warm humid climate, well-distributed rainfall and long sunshine hours. Shoots, comprising two or three tender leaves and a bud, are harvested and processed in factories to manufacture different types of tea.

2. Insect pest occurrences in tea

Being a widespread perennial monoculture crop, tea plantations provide the most congenial microclimate as well as continuous food supply to a number of arthropods. Every part of the tea plant, i.e. leaf, stem, root, flower and seed is subjected to attack by at least one arthropod pest species. According to recent estimates, globally, more than a thousand (1,034 species) arthropods and 82 species of nematodes are associated with tea plantations [1]. Among the insect pests, 32% are from order Lepidoptera, followed by 27% pest species from Hemiptera [2]. In India, only 300 species of arthropods are recorded as pest and about 167 species are from tea-growing Northeast India (Table 1) [3]. The dynamic adaptations of insects have facilitated them to exploit every part of the tea plant and the maximum numbers of pests occur on the foliage.

Among the arthropods that attack tea plant, insect and mite pests are the most damaging [2], causing on average a 5%–55% yield loss [4–6]. Insect pests alone can cause on an average 11%–55% yield loss, if left unrestrained [1].

Tea mosquito bug, thrips, jassids, tea caterpillars (looper, redslug and bunch), aphids, termites, cockchafers and red spider mite are among the various insects and mite pests that cause severe loss in tea production. The damage caused by the tea pests frequently leads to a significant impact on quality and yield, while the degree of pest infestation differs in different tea-growing areas depending on altitude, climate, forest cover and local cultural practices. Tea pests can be broadly classified into the different categories based on their feeding nature, time of occurrence and severity. The major pests are mite pests: red spider mite (*Oligonychus coffeae*), scarlet mite (*Brevipalpus phoenicis*), pink mite (*Acaphylla theae*), purple mite (*Calacarus carinatus*); sucking pests: tea mosquito bug (*Helopeltis theivora*), tea thrips (*Scirtothrips dorsalis* and *Myctrothrips setiventris*), tea jassids (*Empoasca flavescens*), aphids, scale insects; leaf eaters: looper caterpillars (*Hyposidra talaca, H. infixaria, Buzura suppressaria*), red slug caterpillar (*Eterusia magnifica*), bunch caterpillar (*Andraca bipunctata*); and soil-borne pests: termites (*Microcerotermes* sp. *Odontotermes* sp.), cockchafers, nematodes, weevils, cricket [8].

Sl. no.	Name of the pests		Nature of damage	Family: Order
1.	*Andraca bipunctata* Walk.	Bunch caterpillar	Folivores	Bombycidae: Lep.
2.	*Eterusia magnifica* Butl.	Red slug caterpillar	Folivores	Zygaenidae: Lep.
3.	*Buzura suppressaria* Guen.	Looper caterpillar	Folivores	Geometridae: Lep.
4.	*Hyposidra talaca, H. infixaria* Walk.	Black inch worm	Folivores	Geometridae: Lep.
6.	*Lymantria albulunata* Mre.	Sungma caterpillar	Folivores	Lymantridae: Lep.
7.	*Gracillaria theivora* Walsh.	Tea leaf roller	Folivores	Gracilariidae: Lep.
8	*Homona coffearia* Nietner	Tea tortrix	Folivores	Torticidae:Lep.
9.	*Laspeyresia leucostoma* Mayer.	Flushworm	Folivores	Eucosmidae: Lep.
10.	*Holotrichia impressa* Burm.	Cockchafer grubs	Root of young tea	Scarabaeidae: Col.
11.	*Serica assamensis* Brenske	Leaf-eating cockchafer	Leaf eater	Scarabaeidae: Col.
12.	*Asticus chrysochlorus* Wied.	Large green weevil	Leaf eater	Curculionidae: Col.
13.	*Agromyzidae* (Bigot) Meij	Tea leaf miner	Leaf eater	Agromyzidae: Dip.
14.	*Microtermes* spp.	Live wood-eating termite	Stem and root eater	Termitidae: Iso.
15.	*Odontotermes* sp.	Scavenging termite	Stem and root eater	Termitidae: Iso.
16.	*Helopeltis theivora* Waterhouse	Tea mosquito bug	Leaf sucker	Miridae: Hem.
17.	*Empoasca flavescens* Fabr.	Tea greenfly/tea jassid	Leaf sucker	Jassidae: Hem.
18.	*Toxoptera aurantii* Boyer de Fons.	Tea aphid	Leaf sucker	Aphididae: Hem.
19.	*Scirtothrips dorsalis* Hood	Yellow tea thrips	Leaf and bud sucker	Thripidae: Thy.
20.	*Mycterothrips (Teaniothrips) setiventris* Bagnall	Common thrips	Leaf and bud sucker	Thripidae: Thy.
21	*Oligonychus coffeae* Nietner	Red spider mite	Leaf sucker	Tetranychidae: Aca.
22.	*Brevipalpus phoenicis* Geijskes	Scarlet mite	Leaf sucker	Tenuipalpidae: Aca.

Table 1. Major insect and mite pests that occur on tea in India.

IntechOpen

3. Insect pests of tea and management problems

Tea is produced from the young foliage, i.e. young leaves and a bud, and foliage production is increased by seasonal pruning which enhances the leaf cover. The major pests of the crop are those associated with the young foliage. The most important insect pest groups are the folivores (chewing) and sap suckers of the young tender leaves, buds and stems (sucking pest), which damage the most economic part of tea plant that is processed in tea industry for making the tea. These pests cause substantial loss in yield to the tea industry.

In India, different management practices are followed to protect the tea crop against different insect pest groups. Most of the plantations are managed conventionally i.e. using different organosynthetic insecticides, whereas some organic plantations use plant- and animal-based herbal and microbial insecticides. In conventional tea plantations, organo-synthetic insecticides of different functional groups such as organochlorines, organophosphates, synthetic pyrethroids (SPs) and neonicotinoids (NNs) are regularly used throughout the year to control the invasion of different insect pest groups (sucking, folivores and others) [8]. The use of insecticide is cost-effective to planters and a major concern for the environmental degradation due to contamination as well as in resurgence of primary pests [6], outbreak of secondary pests [9], development of insecticide resistance [10, 11], including undesirable residues in made tea [12]. Regular spraying of insecticides leads to the development of higher level of tolerance or resistance to insecticides in many insects [11, 13].

From the early forties onwards, dichlorodiphenyltrichloroethane (DDT) (organochlorine) was regularly used to manage the infestation of *H. theivora,* the major sucking pest in Northeast India [14]. In 1968, endosulfan (cyclodiene: organochlorine) was introduced in the tea plantations of the Dooars region of West Bengal, India, in the form of thiodan 35 EC [15]. Currently, in different conventional tea plantations of tea-growing regions of India, cypermethrin, deltamethrin, quinalphos, monocrotophos, chlorpyriphos, imidacloprid, etc. are extensively used during cropping season to control insect pests [16–18].

Recently, a number of insecticides have been found to be ineffective in controlling the insect and mite pests in different tea-growing regions of India [8]. The development of resistance to different classes of insecticides is one of the causes for persistence and resurgence of insect pests on tea crop [8, 19–21]. A major concern in managing the major insect pests of tea is its high potential to develop resistance rapidly to regularly used insecticides [11]. Continuous and repeated exposure to different classes of insecticides for many years, in addition to their high reproductive potential, short life cycle and numerous annual generations, has limited the management of major pests of tea [11]. Recently, there are reports on the development of resistance to many commonly used synthetic insecticides and consequent failure in controlling many tea pests [10, 22–26]. Such failures are already known in case of organochlorines (OCs), organophosphorus (OP) and synthetic pyrethroid (SP) insecticides and more recently for the newer compound such as neonicotinoids [19–21, 26]. The development of resistance in *H. theivora* populations to different classes of insecticides has been in the range of 1.47–62.99-fold for males and 1.25–62.82-fold for females in Northeast India [19]. Relative toxicity to commonly used insecticides has been observed to vary in *H. theivora* populations from Jorhat, Assam [20], Darjeeling [27], and from sub-Himalayan Dooars region of Northeast India [28]. LC_{50} values of insecticides, when compared with the field dose against *H. theivora* recommended by TRA (Tea Research Association, Tocklai, Assam, India), revealed a pronounced shift in the level of susceptibility of *H. theivora* to all insecticides except acephate [20–21].

For the management of other sucking insects such as yellow tea thrips, *S. dorsalis,* insecticides are also used in conventional tea plantations. In tea ecosystem, control failure and the development of biochemical resistance have been reported in *S.* dorsalis [11, 24]. In India, *S. dorsalis* populations have developed a high degree of resistance to a range of organochlorine (DDT, BHC and endosulfan), organophosphate (acephate, dimethoate, phosalone, methyl-O-demeton and triazophos) and carbamate insecticide (carbaryl) in chili ecosystem [31]. *S. dorsalis* has also developed a high degree of resistance to various insecticides, viz. monocrotophos, acephate, dimethoate, phosalone, carbaryl and triazophos [32]. Recently, several insecticides have been tested on *S. dorsalis* in chili ecosystem in USA and found limited success with chlorfenpyr, spinosad and imidacloprid [33, 34]. The performance of novaluron, abamectin, spiromesifen, cyfluthrin, methiocarb and azadirachtin failed to provide effective control of this pest [35].

Similarly, in another emerging sucking insect pest of tea, tea greenfly, *E. flavescens*, repeated management failure and biochemical insecticide resistance in tea ecosystem from Northeast India have been reported [11, 24]. In China, chemical insecticides including fenvalerate, cyfluthin, cypermethrin and imidacloprid are sprayed to control the leafhoppers as frequently as seven times annually or even more frequently [36–37]. A high level of resistance against many insecticides has been reported in related species, *E. vitis* [38]. The resistance to thiamethoxam was highest and to cyperemethrin was lowest in *E. vitis.* Recently, in Fujian province of China, a regional diversity of resistance to eight insecticides in *E. vitis* has been reported in tea ecosystem with higher resistance level to bifenthrin, acetamiprid, imidacloprid, cartap and chlorfenapyr [39].

A high level of insecticide resistance in folivores, such as black hairy caterpillar, bunch caterpillar, looper pest complex (*Hyposidra talaca, H. infixaria, Buzura suppressaria, Eturesia magnifica*) and in termite of tea ecosystem, has been reported with reduced susceptibility against different insecticides.

Detoxification of insecticides is an important toxicokinetic mechanism for insect pests to tolerate regularly applied insecticides [8, 40–42]. Susceptibility levels against insecticides change mainly due to metabolic detoxification of the insecticides through the induction of some detoxifying enzymes under the stress of different management practices [43–45].

Generally, three principal enzymes, general esterases (GEs), glutathione S-transferases (GSTs) and cytochrome P450-mediated monooxygenases (CYP450s), are involved in the process of metabolic detoxification of insecticides [41]. Estimation of the activities of these metabolic defence-related detoxifying enzymes gives information on the level of tolerance/resistance of the insect pest population to insecticides and is a useful tool in monitoring the tolerance/resistance to insecticides at population level of the pest. The early detection of metabolic threats related to tolerance/resistance to insecticides in pest specimens is of crucial importance for devising pest control techniques that would minimize the development of tolerant/resistant forms and prevent any undesirable wastage of insecticide, money and manpower.

4. Insecticide resistance mechanisms in insect pests of tea

Insects come across with numerous toxins as they go through their life cycle. Some of these toxins are naturally produced by plants (plant allelochemical) and others by humans (synthetic insecticides). To protect themselves against the natural toxins, insects have evolved various detoxification mechanisms [41] These mechanisms also cross-protect insect pests when they are exposed to synthetic insecticides [25]. Herbivorous insect groups (agricultural pests) are significantly more diverse than their non-herbivorous sister groups [46]. The role of plant in promoting diversification in insects has occurred through co-evolutionary 'defence strategies' among them [47]. This diversification could also have been a result of insects 'tracking' plant phylogenies, with minor chemical changes in plants allowing the evolving populations of insects to change and speciate accordingly, which probably has occurred long after chemical changes in plants [48]. Evolution to herbivory preceded via mixed feeding on reproductive parts or spores, dead tissues of plants and animals and fungi. This progression implies that omnivory preceded generalized herbivory and the evolution of specialization on specific plant taxa was a later accomplishment [49].

Among sucking insect herbivores, the actual food used, i.e. digested whole tissue particularly parenchyma (as in *Helopeltis* sp.), cell content (thrips) and phloem flow (*Empoasca* sp.), influences both the feeding mechanism and feeding behaviour [48]. While the chewing insects (looper caterpillar complex) cause extensive damage, the sucking insects cause modest to barely perceptible damage. However, sucking insects, particularly phloem and digested tissue feeder,impose an additional challenge to the plants as they deplete photosynthates, act as vector of viruses and introduce chemical and protein effectors that alter plant defence mechanisms (signalling) and development [50]. When these attributes are combined with a broad host range, breeding strategies that promote invasiveness, highly evolved feeding strategies, the ability to adapt to a wide range plant habitats and the emergence of insecticide resistance, it is not surprising that sucking insects cause heavy losses in agriculture and horticulture [51].

Insecticide resistance is a genetic change in response to selection pressure of toxicants that impair pest control in the field [52]. Insecticide resistance does not occur unless a structural genetic change occurs that is heritable. Therefore, insecticide resistance is an evolutionary phenomenon that results under the selection pressure of a new toxicant in the environment [8].Thus insecticide resistance is different from insecticide tolerance. Insecticide tolerance is the natural ability of a population to withstand the toxic effect of a particular insecticide. It can develop within one generation as a result of physiological adaptation, i.e. induction of xenobiotic detoxifying enzymes. Hence, variation within a population may include individuals with genetic traits that make them better adapted to survive in exposure to an insecticide. If these individuals survive the insecticide exposure, then the tolerance traits can be passed on to the next generation, thereby enriching the gene pool with those genes. The mechanisms of development of insecticide tolerance can be divided into four levels:

Altered behaviour – avoidance of contact with the insecticide

Development of barrier tissues – reduced penetration of the insecticide though the integument

Enhanced detoxification – higher metabolism of the insecticide

Alteration of receptor – at the target site for the insecticide.

The first level, at which insecticide tolerance can develop, is when the insect encounters an insecticide. An altered behaviour helps the insect to avoid coming into contact with the insecticide. Once the insect comes in contact with the insecticide, a reduced and delayed penetration through the cuticle will reduce the effect of the insecticide at the target site; this is yet another level of resistance. Within the insect's body, the insecticide may be enzymatically metabolized and thereby inactivated. At the third level of resistance mechanism, three systems of xenobiotic detoxification enzymes operate: esterases, glutathione S-transferases and cytochrome P450-dependent monooxygenases. The increased activity of one of these enzyme systems in metabolizing insecticides will result in insecticide tolerance. Alterations at the target site for the insecticide are the last level of insecticide resistance mechanisms. Different classes of insecticides bind to specific target sites and reduced binding at the target site, or increased number of target site molecules may confer insecticide resistance.

4.1. Behavioural resistance

Behavioural resistance mechanisms are the least studied resistance mechanisms in insects, but this is not to say that behavioural resistance is the least significant. Behavioural resistance can be defined as '*evolved behaviours that will reduce an insect's exposure to toxic compounds or that allows an insect to survive in what would otherwise be a toxic and fatal environment*' [53]. Behavioural resistance has been observed in more than 30 species of insects [53]. Avoidance is the first step in the evolution of behavioural resistance [54]. In *H. theivora*, this kind of resistance has been seen [55]. *H. theivora* shows a different egg-laying strategy to avoid insecticide exposure. Even *E. flavescens* avoids exposure to direct sunlight and therefore prefers to stay on the underside of the tea leaves. This behaviour cross-protects it from the direct insecticide exposure in conventional tea plantations during spraying [8]. The same has been found in *S. dorsalis* which resides inside the leaf bud during development and underside of the leaf during adult stage, thereby avoiding direct exposure to insecticides [8].

4.2. Reduced penetration

Reduced penetration of insecticides through barrier tissues of insects is another way in which an insect can modify the effective dose of insecticide at the target site. The mechanism may not prevent the insecticide from eventually entering the insect, but it can reduce the rate at which the insecticide reaches the target site. Reduced penetration has been shown to function as a resistance mechanism to many different insecticides, and, by the nature of this mechanism, cross-resistance is often found [56]. The rate of penetration of insecticides through the insect cuticle or other barriers (peritrophic membrane) depends on the physicochemical properties of the insecticide and the barrier. A reduced penetration contributes to DDT resistance in the tobacco budworm, *Heliothis virescens* F. (Lepidoptera: Noctuidae). DDT-resistant larvae had an altered composition of the cuticle. The protein and lipid contents are greater in the cuticle of resistant larvae and, furthermore, the cuticle of the resistant larvae probably had a higher degree of sclerotization [57]. In *M. domestica*, two resistant strains, with reduced penetration as one of the resistance mechanisms, also showed increased cuticular lipid content; more total lipids, mono-glycerides, fatty acids, sterols and phospholipids were present in the resistant strains compared to a susceptible strain [58]. Reduced penetration has been documented as a resistance mechanism only at the level of the insect cuticle, but any biological membrane may serve as a barrier and thereby give resistance [59]. As a single resistance mechanism it usually only confers low levels (less than threefold) of resistance [59]. Reduced penetration has been shown to function as a resistance mechanism to many different insecticides, including insecticides of the three major classes, OPs, carbamates and pyrethroids. Reduced penetration of OPs through the cuticular barrier has been reported, for example, diazinon in *M. domestica* [60], azinphos-methyl in the pear psylla, *Psylla pyricola* Foester (Hemiptera: Psyllidae) [61] and profenofos in *H. virescens* [62]. However, by slowing the penetration rate of insecticides, this mechanism reduces the risk that the insects' detoxification systems become overloaded, and the dose of insecticide reaches to a lethal level at the target site. In female *H. theivora*, a higher level of body lipid has been found which effectively reduces the penetration of insecticide to the target site [63]. No studies on resistance due to reduced penetration in *E. flavescens* and *S. dorsalis* or any other tea pests have been reported to date. The studies had shown that when different resistance mechanisms are combined in the same individuals, a synergistic effect, resulting in a high level of resistance, may arise [64, 65]. Therefore, even a small degree of reduced penetration can contribute significantly to the overall insecticide resistance of the insect pests.

4.3. Metabolic detoxification

Metabolic detoxification of insecticides is an important toxicokinetic mechanism for insects to tolerate the toxic effects of insecticides. Generally, lipophilic (hydrophobic) insecticides are rapidly detoxified. Organophosphates, organochlorines, carbamates and pyrethroids are lipophilic compounds, and detoxification enzymes transform these insecticides to more hydrophilic and less biologically active compounds so that can be eliminated more easily by excretion. Increased detoxification of insecticides has often been reported in many resistant populations [40]. Three enzyme systems are generally recognized as the major detoxification systems involved in insecticide resistance in insects. These are carboxylesterases, cytochrome P450-dependent monooxygenases and glutathione S-transferases [40, 41].

4.4. Alteration at the target site for insecticide (target site insensitivity)

The biochemical sites for insecticide action differ for different insecticides and are a potential field of research for developing insecticides, which can act specifically or more efficiently on insect biochemical sites compared to mammals. The target site receptor for action of organophosphates, carbamates, organochlorines and pyrethroids is in the nervous system. The enzyme acetylcholinesterases (AChEs) (EC 3.1.1.7) are the target sites for organophosphates and carbamates, and voltage-gated sodium channel of the nerve membrane is the target of pyrethroids and DDT. Neurotoxic insecticides such as cyclodienes (e.g. dieldrin and endosulfan), y-HCH (lindane) and fipronil target gamma-aminobutyric acid (GABA)-receptor [68, 69] and nicotinyl insecticides (imidacloprid and nicotine) target the nicotinic acetylcholine receptor (nACHR) [70]. Alteration at the target site, to less a sensitive target for neurotoxic insecticides, is an important toxicodynamic resistance mechanism in insects [71].

In insects, the potent inhibitors of AChE are organophosphates and carbamates. These compounds inhibit the activity of AChE by forming a stable covalent intermediate, preventing the enzyme to hydrolyse acetylcholine. An accumulation of acetylcholine keeps the ion channel of the receptor permanently open, which eventually kills insect. OPs and carbamates are quasi-irreversible inhibitors of AChE. The organophosphates and carbamates phosphorylate and carbamylate the active site serine of AChE, respectively [72]. Generally, the reactivation time of phosphorylated or carbamylated AChE is long. However, the half-lives of reactivation vary considerably, from minutes to several days, depending on the compound interacting with AChE [73]. Carbamylated AChE generally reactivates faster than phosphorylated AChE. Reduced sensitivity of AChE to inhibition by OPs and carbamates is an important resistance mechanism in insects and is often referred to as altered or insensitive AChE [74]. The presence of insensitive AChE conferring resistance was first noticed in OP-resistant mites, *Tetranychus urticae* Koch (Acari: Tetranycidae) [74] and also found in several insect populations resistant to these compounds [75–77]. Insecticide susceptible and resistant insect pest populations differ in the level of AChE activity [78–80]. A higher level of AChE activity has been reported in *H. theivora* sampled from conventional tea plantations than from organic garden indicating the presence of resistance to insecticides in conventional tea ecosystems [81, 82]. There is no such report on *S. dorsalis* and *E. flavescens* in conventional tea ecosystems to date.

5. Major metabolic detoxifying enzymes in insects

Carboxylesterases, glutathione S-transferases and cytochrome P450-mediated monooxygenases are the three principal enzymes that facilitate the insects to metabolize different kind of toxins. These large enzyme families contain multiple forms with overlapping substrate specificities. Knowledge of insecticide detoxification helps in understanding the mechanism of insecticide resistance, hence the development of a sound resistance management strategy. Detoxification can be divided into phase I (primary) and phase II (secondary) processes (Figure 1).

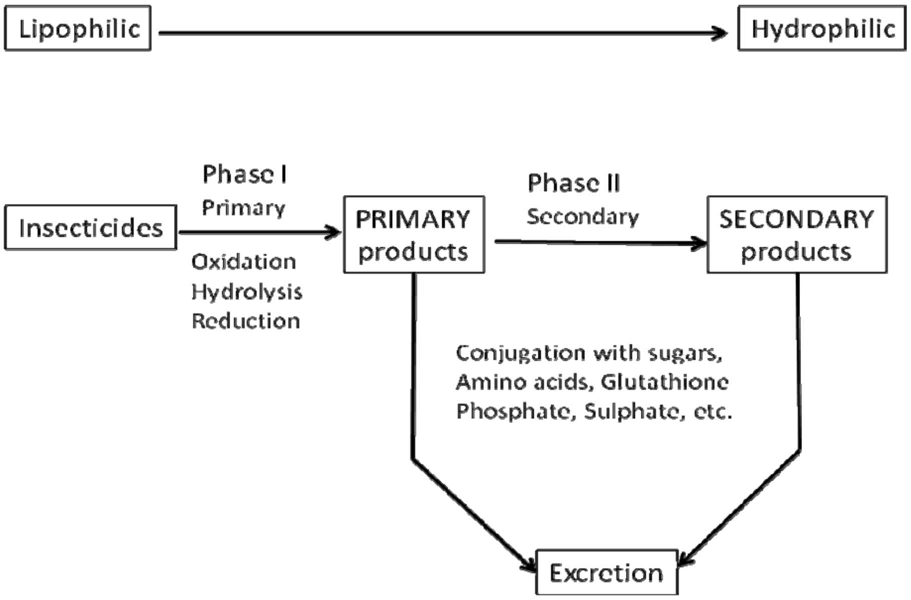
Lipophilic
Hydrophilic
Insecticides
Phase I
Primary
Oxidation
Hydrolysis
Reduction
PRIMARY products
Phase II
Secondary
SECONDARY products
Conjugation with sugars, Amino acids, Glutathione Phosphate, Sulphate, etc.
Excretion

Figure 1. Insecticide detoxification pathways.

Phase I reactions consist of oxidation, hydrolysis and reduction. The phase I metabolites are sometimes polar enough to be excreted but are usually further converted by phase II reactions. In phase II reactions, the polar products are conjugated with a variety of endogenous compounds such as sugars, sulphate, phosphate, amino acids or glutathione and subsequently excreted. Phase I reactions are usually responsible for decreasing the biological activity of toxins, and therefore the enzymes involved are rate limiting with respect to toxicity. The most important function of biotransformation is to decrease the lipophilicity of insecticides, so that they can be excreted quickly [83].

5.1. Phase I reactions

5.1.1. Cytochrome P450 monooxygenases (E.C. 1.14.-.-)

Oxidation is considered the most important among phase I reactions. The oxidative reactions are carried out mainly by a group of enzymes called cytochrome P450 monooxygenases [also known as mixed function oxidases (MFO) or polysubstrate monooxygenases (PSMO), microsomal oxidase, P450 enzymes]. Cytochrome P450, or *CYP* genes, constitutes one of the largest family of genes, with representatives in virtually all living organisms, from bacteria to protists, plants, fungi and animals [84].

In insects, P450 monooxygenases are involved in many processes including roles in the metabolism of plant allelochemicals by herbivores and in detoxification of insecticides. The human genome carries about 57 CYP genes, and insect genomes can carry from 36 CYP genes in the body louse *Pediculus humanus* [85] to 170 in a mosquito [86]. Each P450 protein is the product of a distinct CYP gene, and P450 diversity is the result of successive gene (or genome) duplications followed by sequence divergence [84].

The typically 45–55-kDa P450 proteins are heme-thiolate enzymes. Their essential common feature is the absorbance peak near 450 nm of their Fe^{II}–CO complex for which they are named [87]. P450 enzymes are best known for their monooxygenase role, catalysing the transfer of one atom of molecular oxygen to a substrate and reducing the other to water. The simple stoichiometry commonly describes the monooxygenase or mixed function oxidase reaction of P450:

$$RH+O_2+NADPH+H^+ \rightarrow ROH+H_2O+NADP^+$$

However, oxygen atom transfer is not the only catalytic function of P450 enzymes. They also show activities such as oxidases, reductases, desaturases, isomerases, etc. and collectively are known to catalyse at least 60 chemically distinct reactions (Table 2).

The first insect P450 cloned and sequenced was CYP6A1 from *Musca domestica* in 1989 [88]. The P450 gene complement (CYPome) size of an insect genome is not a definite number [89]. Insects can survive with small CYPomes even in toxic environments. The human body louse *Pediculus humanus*, with 36 CYP genes, is known to become highly resistant to many classes of insecticides [85], and the honeybee, with just 46 CYP genes [90], is not more sensitive than

Reaction catalysed	P450
Oxidase activity O_2 to H_2O, H_2O_2, O_2^{-}	CYP6A1 (and probably most P450 enzymes)
Aliphatic hydroxylation C–H hydroxylation	CYP4C7, CYP6A1, CYP6A2, CYP6A8, CYP6G1, CYP6M2, CYP6CM1vQ, CYP9T2, CYP12A1, CYP18A1, CYP302A1 CYP306A1, CYP312A1, CYP314A1, CYP315A1
O-dealkylation	CYP6A1, CYP6D1, CYP6A5, CYP6B4, CYP6B17, CYP6B21, CYP6G1, CYP6Z2, CYP6CM1vQ, CYP9A12, CYP9A14, CYP12A1, CYP321A1,
Dehalogenation	CYP6G1
Epoxidation	CYP6A1, CYP6A2, CYP6B8, CYP6B27, CYP6AB3, CYP6 CYP6AB11, CYP9E1, CYP12A1, CYP15A1, CYP321A1
Aromatic hydroxylation	CYP6D1, CYP6G1, CYP6M2
Heteroatom oxidation and dealkylation Phosphorothioate ester oxidation	CYP6A1, CYP6A2, CYP6D1, CYP12A1
N-dealkylation	CYP6A5, CYP12A1
N-oxidation	+(Nicotine)
S-oxidation	+(Phorate)
Aldehyde oxidation	CYP18A1
Complex and atypical reactions	
Cyanogenic glucoside biosynthesis:	CYP405A2
Val/Ile to oximes	CYP332A3
Oximes to cyanohydrins	CYP6M2
Aryl ether cleavage	+?(Sterols, ecdysteroid)
Carbon–carbon cleavage	CYP4G1
Decarbonylation with C–C cleavage	+(Defensive steroids)
Aromatization	–
Reduction	–
Endoperoxide isomerisation	

Table 2. Enzymatic reactions catalysed by insect P450 enzymes (adapted from Feyereisen, 2005).

other species in a comparison to the toxicity of 62 insecticides [91]. The main driver of CYPome evolution is of course gene duplication, followed by divergence (by neofunctionalization or subfunctionalization) or death (pseudogenization or deletion) [84].

5.1.2. Carboxylesterases (EC 3.1.1.1)

Carboxylesterase or esterase is a collective term for the enzymes that hydrolyse carboxylic esters [92]. Classification of these enzymes is difficult because of their overlapping substrate

IntechOpen

IntechOpen

specificity [93]. However, the esterase classification of Aldridge [94] is generally recognized. According to that classification, esterases inhibited by paraoxon in a progressive and temperature-dependent manner are called B-esterases and those which are not inhibited are A-esterases [94]. Some A-esterases can hydrolyse OPs, through an acylated cysteine in their active site, and are termed phosphoric triester hydrolases (EC 3.1.8.) [95, 96]. The term carboxylesterase is now mainly attributed to B-esterases [95, 96]. These enzymes have an active site serine residue, hence the terms B-esterase and serine hydrolase are synonymous. Insecticides such as organophosphates, carbamates, pyrethroids and some juvenoids, which contain ester linkages, are susceptible to hydrolysis. Esterases are hydrolases that split ester compounds by the addition of water to yield an acid and alcohol.

Esterases that metabolize organophosphates can be divided into three groups: A-esterases which are not inhibited by organophosphates but hydrolyse them; B-esterases, which are susceptible to organophosphate inhibition; and C-esterases which are uninhibited by organophosphates and do not degrade them [41].

There are two types of esterases that are important in metabolizing insecticides, namely, carboxylesterases and phosphatases (also called phosphorotriester hydrolases or phosphortriesterases). Carboxyl esterases, which are B-esterases, play a significant role in degrading organophosphates, carbamates, pyrethroids, and some juvenoids in insects. The best example is malathion hydrolysis, which yields both α- and β-monoacids and ethanol [41]. Phosphatases are A-esterases that detoxify many organophosphorous insecticides especially phosphates in insects. In houseflies, paraoxon can be hydrolysed to diethyl phosphoric acid and *p*-nitrophenol. Phosphatases also hydrolyse the alkyl groups of organophosphates. Paraoxon is hydrolysed by the enzyme in houseflies. Several amides containing organophosphorous insecticides such as dimethoate and acephate have been shown to be hydrolysed by carboxylamidases to their corresponding carboxylic acid derivatives [41].

5.2. Phase II reactions

Phase I reactions with xenobiotics result in the addition of functional groups such as hydroxyl, carboxyl and epoxide. These phase I products can further undergo conjugation reactions with endogenous molecules. These conjugations are called phase II reactions. The endogenous molecules include sugars, amino acids, glutathione, phosphate and sulphate. Conjugation products are usually more polar, less toxic and more readily excreted than their parent compounds. Thus, the process with only a few exceptions results in detoxifications.

Three types of conjugation reactions occur in insects. Type I requires an activated conjugating agent that then combines with the substrate to form the conjugated product. Type II involves the activation of the substrate to form an activated donor that then combines with an endogenous molecule to yield a conjugated product. In Type III, conjugation can proceed directly between the substrate and the conjugating agent without involving activation. Thus, Type I and II require information of high-energy intermediates before the conjugation reactions proceed. The chemical groups required for Type I are –OH, NH_2, COOH and SH (glucose conjugation, sulphate conjugation and phosphate conjugation); for Type II COOH (amino acid conjugation); and for Type III, halogens, alkenes, NO2, epoxides, ethers and esters (glutathione conjugation).

5.2.1. Glutathione S-transferases (EC 2.5.1.18.)

Glutathione conjugations are performed by a group of multifunctional enzymes known as glutathione S-transferases and are involved in detoxification mechanisms of many molecules. GSTs are involved in the transport of physiologically important lipophilic compounds. These enzymes catalyse reactions in which the sulphur atom of glutathione provides electron for nucleophilic attack on a second electrophilic substrate; the latter can be endogenous natural substrates such as epoxides, organic hydroperoxides, or activated alkenals resulting from oxidative metabolism. These enzymes catalyse the conjugation of reduced glutathione (GSH) with electrophilic substrates. Glutathione S-transferases perform a variety of reactions including:

The S-alkylation of GSH by alkyl halides and related compounds.

The replacement of labile aryl halogen or nitro groups by GSH.

The replacement of labile arakyl halogen and ester groups by GSH.

The addition of GSH to various epoxides.

The addition of GSH to α-, β-unsaturated compounds including aldehydes, ketones, lactones, nitriles and nitro compounds.

The *O*-alkyl and *O*-aryl conjugation of phosphorothiotes and phosphates with GSH.

The glutathione conjugate is subsequently transformed to mercapturic acid through the stepwise loss of glutamic acid and glycine to a cysteine conjugate, which is finally acetylated before excretion. Because of their broad substrate specificities, glutathione S-transferases are responsible for the detoxification of numerous xenobiotics [97]. More than 40 GST genes have been identified in insects [98, 99]. Mammalian GSTs have been classified into eight cytosolic classes (alpha, mu, pi, theta, sigma, zeta, kappa and omega) and a microsomal class on the basis of their amino acid sequence, immunological properties and substrate specificity. Each class shares 40% or higher amino acid in common. The classification of insect GST is not clear. The majority of insect GSTs do not belong to mammalian classes. Insect glutathione S-transferases consist of two subunits (homodimers and heterodimers) of molecular weight between 19 and 35 kD. Two classes of insect GSTs (Class I and Class II) were reported [100], which have been referred to as the Delta and Sigma class, respectively. Recently, a new class of insect GSTs, referred to as Epsilon has been described in several species of insects including *Anopheles gambiae* [101]. Purified cytosolic and microsomal glutathione S-transferase isozymes from fall armyworm larvae possessed cumenehydrperoxide peroxidase [102]. A Delta class GST purified from German cockroaches also showed high peroxidase activity [103]. The name of each GST is composed of the initials of the species scientific name, followed by the acronym GST, a capital letter to designate the class name and an Arabic number for the individual protein, such as AgGSTD2.

Glutathione S-transferases are important in the metabolism of organophosphorous insecticides resulting in detoxification [99, 104]. For example, methyl parathion is dealkylated by glutathione S-tranferases to form desmethyl parathion and methyl glutathione [41]. On the contrary, parathion can be de-arylated by glutathione S-transferases to produce diethyl phosphorothioic acid and S-(*p*-nitrophenyl) glutathione [41]. Interestingly, a glutathione S-transferase isozyme from the housefly exhibits DDT-dehydrochlorinase activity, showing that DDT-dehydrochlorinase (DDTase) is one of the glutathione S-tranferases [104]. DDT-dehydrochlorinase converts DDT to DDE, resulting in detoxification [41].

6. Detoxifying enzymes and insecticide metabolism

The metabolism of insecticides by P450 enzymes is very often a key factor in determining toxicity to insects and to non-target species. The importance of monooxygenases in insecticide resistance became evident in the early 1960s, when it was shown that resistance to carbaryl could be abolished by the P450 inhibitor sesame [106]. Additional evidence of monooxygenase-based resistance quickly amassed [107, 108]. Monooxygenase-mediated detoxification is frequently found as a major mechanism of resistance, and unlike target site resistance, detoxification has the potential to confer cross-resistance to toxins independent of their target sites [109, 110]. Most cases of monooxygenase-mediated resistance result from an increase in detoxification (Table 3). However, in cases where the parent insecticide must undergo monooxygenase-mediated bioactivation, as is the case for many organophosphates, it is also possible that resistance could be achieved through decreased activation [111]. Although this has been reported once, it does not appear to be a common mechanism of resistance. This may explain why esterases are relatively more common than monooxygenases in resistance to some organophosphates [110, 112]. The classical example is probably the metabolism of phosphorothioate insecticides. In many cases, the active ingredients of organophosphorus insecticides are phosphorothioate (P=S) compounds (also known as phosphorothionates), whereas the molecule active at the acetylcholinesterase target site is the corresponding phosphate (P=O) (Figure 2).

P450 enzymes that metabolize OPs can metabolize other insecticides as well, and this sometimes leads to potentially useful interactions. Thus, enhanced detoxification of dicofol in spider mites can lead to enhanced chlorpyriphos activation, and hence negative cross-resistance [113]. Similarly, permethrin resistance in horn flies is suppressible by piperonyl butoxide, and negatively related to diazinon toxicity [114]. In *H. armigera* populations from West Africa, triazophos shows negative cross-resistance with pyrethroids, and in this case, the synergism shown by the OP towards the pyrethroid appears due to an enhanced activation to the oxon form [115]. Organophosphorus compounds (disulfoton and fenthion) are also activated by thioether oxidation (formation of sulphoxide and sulfone). The metabolism of pyrethroid by P450 enzymes is well studied in insects. Hydroxylations and further metabolism make pyrethroid metabolism and has been noticed for the single enantiomer of deltamethrin (Figure 3).

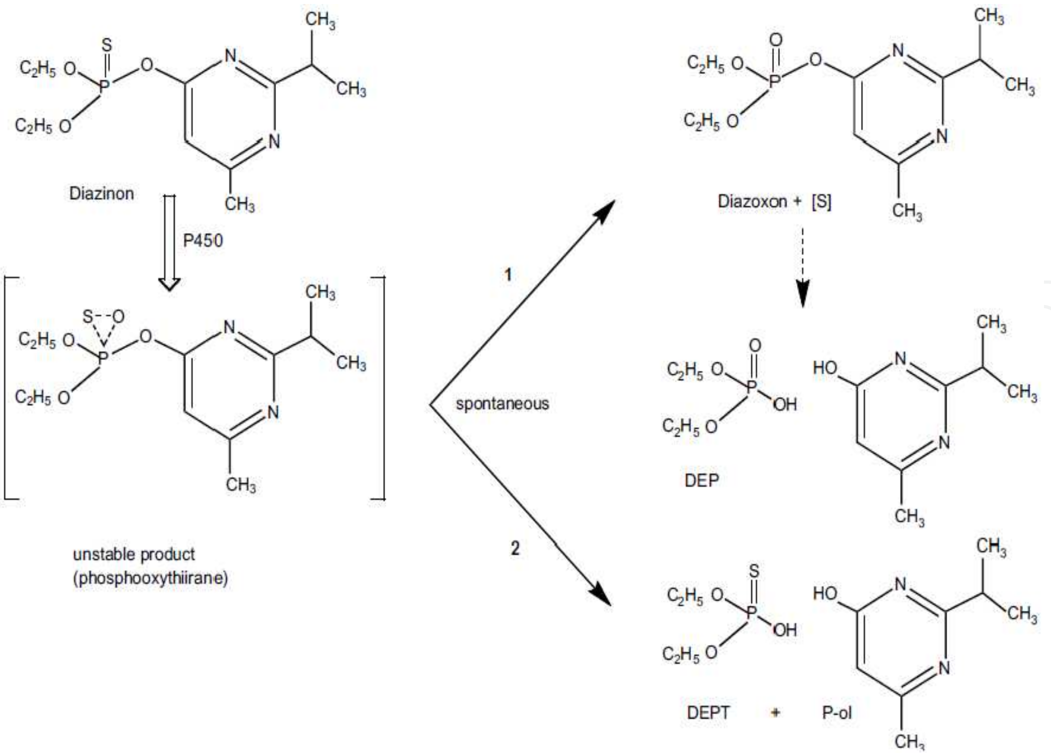
Diazinon
P450
unstable product
(phosphooxythiirane)
1
spontaneous
2
Diazoxon + [S]
DEP
DEPT
+
P-ol

Figure 2. Metabolism of diazinon by cytochrome P450. Following an insertion of oxygen into the substrate, a reactive intermediate collapses (1) by desulphuration or (2) by cleavage of the ester linkage. DEP, diethylphosphate; DEPT, diethylphosphorothioate; P-ol, 2-isopropoxy-4-methyl-6-hydroxypyrimidine; [S], reactive form of sulphur released during the reaction. Adopted from Feyereisen, 2012.

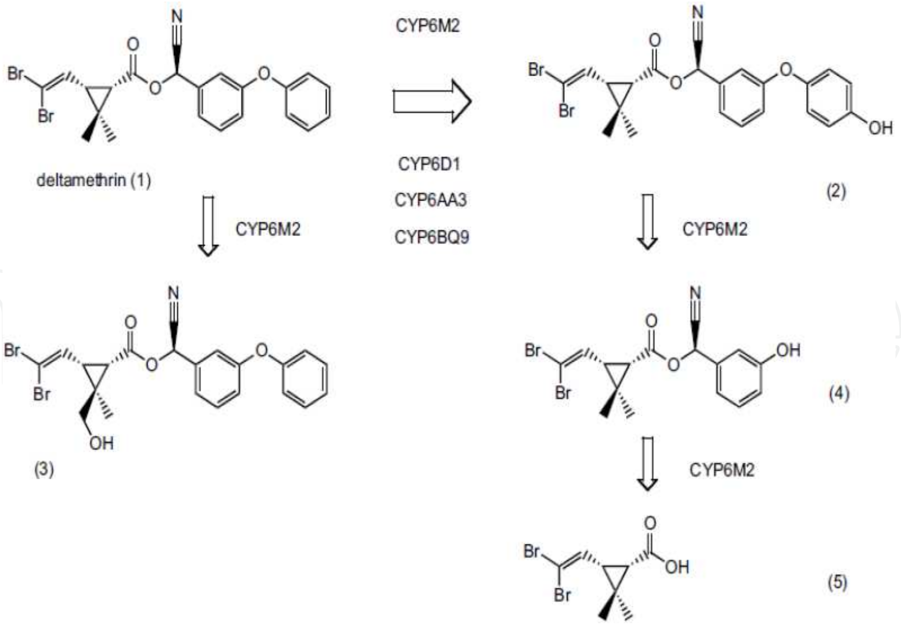
deltamethrin (1)
CYP6M2
CYP6D1
CYP6AA3
CYP6BQ9
CYP6M2
(2)
CYP6M2
(3)
(4)
CYP6M2
(5)

Figure 3. Metabolism of deltamethrin by insect P450 enzymes: (1) deltamethrin; (2) 4′ hydroxydeltamethrin; (3) *trans*-hydroxymethyl-deltamethrin; (4) cyano (3-hydroxyphenyl) methyl deltamethrate; (5) deltamethric acid. Adopted from Feyereisen, 2012.

The currently banned cyclodiene insecticides aldrin, heptachlor and isodrin are epoxidized by P450 enzymes to the environmentally stable toxic epoxides dieldrin, heptachlor epoxide and endrin, respectively [116]. Recombinant CYP6A1, -A2, -A8, -B8 and -B27; CYP12A1; and CYP321A1 can catalyse these epoxidations. Examples of pro-insecticide metabolism include the activation of chlorfenapyr by N-dealkylation [117] and diafenthiuron by S-oxidation [118]. In each case, the insect P450-dependent activation is a key in the selective toxicity of these pro-insecticides that target mitochondrial respiration. Recombinant housefly CYP6A1 catalyses the activation of chlorfenapyr (Figure 4).

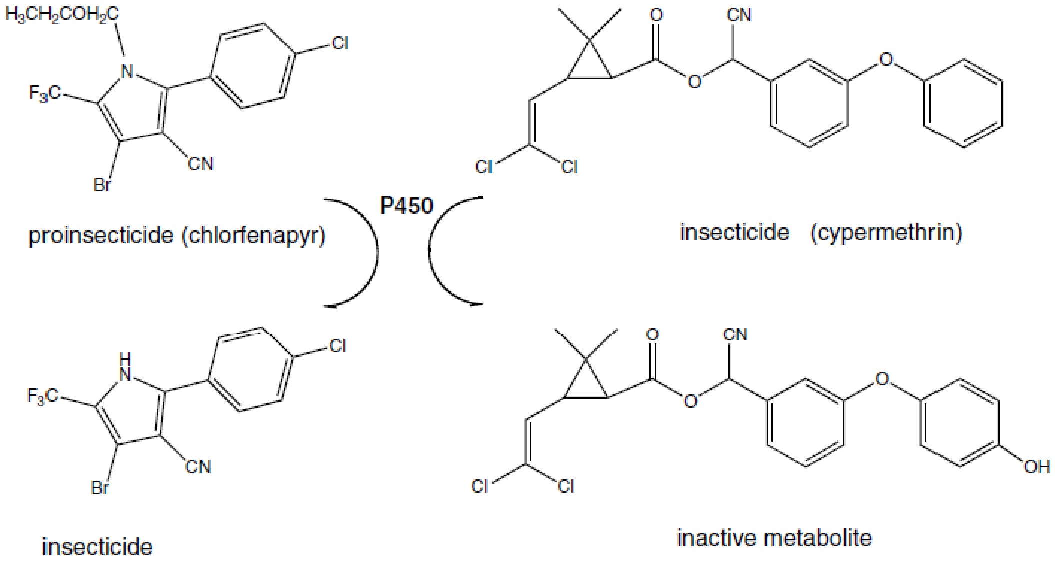

Figure 4. Chlorfenapyr and cypermethrin metabolism. The same P450 in *Heliothis virescens* probably activates the pyrrole and inactivates the pyrethroid, resulting in negative cross-resistance. Adopted from Feyereisen, 2012.

In *H. virescens*, the toxicity of chlorfenapyr is negatively correlated with cypermethrin toxicity [119]. The metabolism of imidacloprid is also of interest, particularly in relation to resistance. Piperonylbutoxide can synergize the toxicity of imidacloprid, and two P450 enzymes, CYP6G1 of *D. melanogaster* and CYP-6CM1vQ of *Bemisia tabaci*, have been shown to metabolize this neonicotinoid [120, 121] (Figure 5). Hydroxylations at the 4 and 5 positions can lead to the olefinic metabolite or to the dihydroxylated metabolite. In the whitefly, the 5-hydroxy metabolite is not toxic, but the 4-hydroxy metabolite is as toxic as the parent compound, so region selectivity may be of importance.

Despite the continuous use of insecticides, there are repeated failures in controlling the sucking insect pest species in recent years [11, 21] in different conventional tea plantations of Terai, the Dooars and Darjeeling foothill regions. Such a failure occurs due to changes in the susceptibility level of the pest species to the applied insecticides. Susceptibility level changes mainly due to metabolic detoxification of the insecticides through higher level of activity of some insecticide detoxifying enzymes under the stress of different management practices [8, 10, 11, 22]. In another mirid pest, *Lygus lineolaris,* metabolic resistance to insecticides due to enhanced level of activity has been reported by many authors [177, 178].

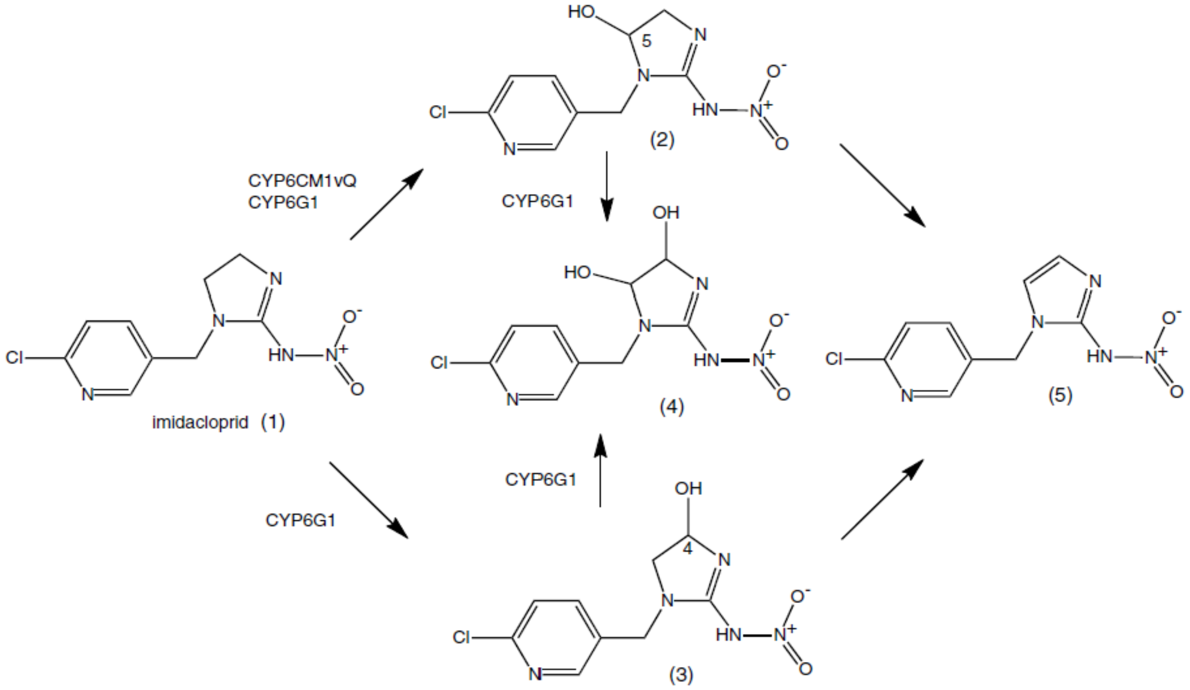

Figure 5. Metabolism of imidacloprid by insect P450 enzymes: (1) imidacloprid; (2) 5-hydroxyimidacloprid; (3) 4-hydroxyimidacloprid; (4) dihydroxyimidacloprid; (5) Non-enzymatically derived dehydroimidacloprid. Adopted from Feyereisen, 2005.

Species	P450 Over-expressed	Resistance pattern
Musca domestica	CYP6A1	OP, carbametes [122]
		IGR [123, 124]
	CYP6A5v2, CYP6A36	Pyrethroids [125–127]
	CYP6D1, CYP6D3	Pyrethroids [128–130]
	CYP6D1	Pyrethroids [131]
	CYP6D1, CYP6D3v2	Pyrethroids [132]
	CYP6A24	Pyrethroids [133]
	CYP12A1	Pyrethroids [134]
Drosophila melanogaster	CYP6A2	DDT, malathion [135–137]
	CYP6A8	Malathion [137, 138]
		Malathion [137]
		DDT [139]
	CYP6G1	DDT [140, 141]
		DDT [142]
		Lufenuron, propoxur [141]
		Imidacloprid [141]
		Imidacloprid [142]
		DDT [143]

IntechOpen

Species	P450 Over-expressed	Resistance pattern
		Diazinon [144]
	CYP12D1/2	DDT [143]
		DDT [139]
	CYP12A4	Lufenuron [145]
Drosophila simultans	CYP6G1	DDT, imidacloprid, Malathion [139]
Anopheles gambiae	CYP6Z1	Pyrethroids [146]
	CYP325A3	Pyrethroids [147]
	CYP6M2, CYP6P3	Pyrethroids [148]
	CYP6P3	Permethrin [149]
	CYP6M2, CYP6Z2	Permethrin [150]
	CYP4C27, CYP4H15	DDT [151]
	CYP6Z1,2, CYP12F1, CYP314A1	DDT [147]
A. stephensi	CYP325C1	Pyrethroids [152]
A. funetus	CYP6P4, CYP6P9	Pyrethroids [153, 154]
Aedesaegypti	CYP9J10,27,32	Pyrethroids [155]
Culex pipien squinquefasciatus	CYP9M10	Permethrin [156]
	CYP6F1	Permethrin [157]
	CYP4H34, CYP6Z10, CYP9M10	Permethrin [158]
	CYP4H21, H22, H23, CYP4J4, CYP4J6	Deltamethrin [159]
Heliothis virescens	CYP9A1	Thiodicarb [160]
Helicoverpa zea	CYP6B8,B9	Cypermethrin [161]
H. armigera	CYP4G8	Pyrethroids [162]
	CYP6B7	Pyrethroids [163]
	CYP6B7, CYP9A12, CYP9A14	Pyrethroids [164]
	CYP4S1, CYP337B1	Fenvalerate [165]
	CYP4L5,11, CYP4M6,7, CYP6AE11, CYP9A14, CYP332A1, CYP337B1	Deltamethrin [166]
Plutella xylostella	CYP6BG1	Cypermethrin [167]
	CYP4M20	Cypermethrin [168]
Lygus lineolaris	CYP6X1	Permethrin [169]
Bemisia tabaci	CYP6CM1vQ	Imidacloprid [170]
Nilaparvata lugens	CYP6ER1	Imidacloprid [171]
Myzus persicae	CYP6CY3	Neonicotinoids [172]
Diabrotica virgifera	CYP4	Me-parathion [173]
Tribolium castaneum	CYP6BQ8,9,10, CYP436B1, B2	Deltamethrin [174]
Blattella germanica	P450MA,	Chlorpyriphos [175]
	CYP4G19	Pyrethroids [176]

Table 3. Over-expressed CYP genes in insecticide-resistant strains.

IntechOpen

IntechOpen

In Western Flower Thrips, *Frankliniella occidentalis*, metabolic detoxification of insecticides has been reported by many authors [179]. In *Bemisia tabaci*, metabolic resistance due to enhanced activity of insecticide resistance-related enzymes has also been reported [180].

7. Host allelochemicals, induction of detoxifying enzymes and insecticide resistance

IntechOpen

Understanding the diversity of insect responses to chemical pressures (plant allelochemicals and insecticides) in their local ecological context represents a key challenge in developing sustainable pest control strategies. Plants and insects have had co-existing relationships for a long time. Insects were suppressed either by other insects or toxins or by plant defence mechanisms in order to create a balance between the insect pest population and host. Each plant species has a unique set of defence traits ranging from morphological to phytochemical parameters that have behavioural and physiological ramifications for a potential herbivore consumer [181, 182]. Therefore, the resistance mechanisms evolved by insects to deal with the chemical defences of plants are similar to those mechanisms that have evolved to resist synthetic insecticides. The chemical structure of some synthetic insecticides is comparable to that of some plant-produced compounds (e.g. pyrethroids and nicotinoids). Insect resistance to plant allelochemicals interferes with their resistance to synthetic insecticides [183]. From the evolutionary perspective, despite the key role of the chemical 'arms race' in driving the co-evolution of plants and insects, much research has focused so far on describing the diversity of plant chemicals and their effects on herbivores. Hence, the understanding of insecticide resistance mechanisms as well as taking into account other ecological parameters is important in predicting the spread of insecticide resistance in natural pest populations and in choosing the optimum strategy for managing insect pest populations. Less is known about the multiple mechanisms evolved by insects to overcome these chemical defences (Table 4). These mechanisms include contact and ingestion avoidance, excretion, sequestration, degradation of the toxin and target site mutation.

IntechOpen

Biotransformation of plant toxins is one of the major weapons that insects have evolved in their co-evolutionary arms race with plants [204]. To date, metabolic resistance to plant chemicals has been identified not only in herbivorous insects [194] but also in detrivorous insects such as mosquito larvae feeding plant debris [205]. Metabolic resistance often results from the overproduction of 'detoxification enzymes' that can metabolize plant xenobiotics (allelochemicals). This mechanism is often associated with phenotypic plasticity, as the production of detoxification enzymes is usually induced by the presence of plant xenobiotics in the diet of the insect.

Induction of insect detoxifying enzyme activities by plant allelochemicals is a clear manifestation of biochemical phenotypic plasticity and has been documented in several instances [206, 207]. Many of the theories and some of the experiments implicitly or explicitly deal with the insect's ability to metabolize plant secondary substances by P450 and other enzymes. In those studies, a 'higher activity of midgut microsomal oxidase enzymes in polyphagous than in

Plant allelochemicals	**Target (mechanism of effect)**	**Resistance mechanisms**	**Species**
Alkaloids	Neuroreceptors (inhibition), ion channels (antagonists), nucleic acids (disruption of DNA synthesis), feeding (deterrent owing to bitterness), enzymes (inhibition)	Modification of nicotine synthesis by salivary glucose oxidase	*Helicoverpa zea* (Lep.) [184]
Cardenolides	Nervous system (depressing activity); Na^+, K^+-ATPase (specific inhibitor)	Canal trenching behaviour, target site mutation	*Danaus plexippus* (Lep.) [185] *Chrysochus* sp. (Col.) [186]
Cyanogenic glycosides	Electron transport (inhibition of mitochondrial cytochrome oxidase)	Ingestion avoidance, sequestration and detoxification	*Schistocerca americana* (Ort.), *Hypera brunneipennis* (Col), *Zygaena* sp. (Lep.), *Clossiana euphrosyne* (Lep.), *Heliconius sara* (Lep.) [187]
Glucosinolates	Respiration (inhibition)	Detoxification by GSTs, detoxification by glucosinolate sulphatase, formation of nitriles instead of isothiocyanate detoxification by P450s, detoxification by N-oxidation and sequestration	*Myzus persicae* (Hem.) [188] *Plutella xylostella* (Lep.) [189] *Pieris rapae* (Lep.) [190] *Drosophila melanogaster* [191] *Estigmene acrea* (Lep.) [192] *Tyria jacobaeae* (Lep.) [193]
Flavonoids and phenolic acids	Respiration (inhibition), growth (inhibition)	Ingestion avoidance, decrease of toxin levels in gall tissue, glycosylation by UDP-glycosyl transferase, sequestration and/or excretion	*Manduca sexta* (Lep.) [194] *Potania* sp. (Hym) [195] *Bombyx mori* (Lep.) [196]
Iridoid glycosides	Feeding (deterrent owing to bitterness), nucleic acids (inhibition of DNA polymerase), proteins (denaturant and cross-linking activities)	Sequestration	*Longitarsus* sp. (Col.) [197]
Coumarins and furanocoumarins	Nucleic acids (photoactivie DNA bonding), pro-oxidant activity	Detoxification by P450s, detoxification by GSTs	*Papilio polyxenes* (Lep.) [198] *Depressaria pastinacella* [199] *Spodoptera frugiperda* (Lep.) [200]
Protease inhibitors	Digestive system (inhibition of protease)	Over-expression of insensitive protease	*Callosobruchus maculatus* (Col.) [201]
Terpinoids	Nervous system (inhibition of acetylcholine esterases); feeding (deterrent owing to physical barrier and bitterness); growth and development inhibitor (pheromone analogue)	Repression of genes involved in biosynthetic pathways	*Spodoptera exigua* (Lep.) [202]
Tannins	Feeding (complexation of salivary and gut proteins); pro-oxidant activity	Synthesis of anti-oxidant compounds	*Orgyia leucostigma* (Lep.) [203]

Lep., Lepidoptera; Col., Coleoptera; Ort., Orthoptera; Hym., Hymenoptera; Dip., Diptera; Hem., Hemiptera.

Table 4. Plant allelochemicals and associated resistance mechanisms in insects.

monophagous species indicates that the natural function of these enzymes is to detoxify natural insecticides present in the larval food plants'. The estimation of aldrin epoxidation in gut homogenates of last instar larvae from 35 species of Lepidoptera showed that polyphagous species had on average a 15 times higher activity than monophagous species. This trend was seen in sucking insects as well. A 20-fold lower aldrin epoxidase activity was found in the oleander aphid *Aphis nerii* (specialist feeder on two plant families, Asclepiadaceae and Apocynaceae) when compared to the potato aphid *Myzus euphorbiae* or to the green peach aphid *Myzus persicae* (both are generalists found on 30–72 plant families) [208]. A similar type of observation was made for other detoxification enzymes. In mites, predatory mite has a five times lower aldrin epoxidase activity than its herbivorous prey [209]. The toxicity of the natural phototoxin α-terthienyl is inversely proportional to the level of its metabolism in Lepidoptera and is related to diet breadth. Metabolism is highest in *Ostrinia nubilalis*, which feeds on numerous phototoxic Asteraceae; lower in *Helicoverpa virescens*, which has a broad diet, including some Asteraceae that are non-phototoxic; and lowest in *Manduca sexta*, a specialist of Solanaceae [210].

In addition to insecticides, insect carboxylesterases also metabolize many glycosides. B-glucosidase enzyme is active towards a variety of glucosides in fall armyworms, corn earworms, cabbage loopers and valvet bean caterpillars. The *p*-nitrophenyl β-D-glucoside, 4-methyl umbelliferyl β-D-glucoside, D (+)-cellobiose, D-amygdalin and helicon were preferred substrates whereas sinigrin, phloridzin, α-solanine, tomatine and linamarin were poor substrates for these insects and many other insects reported to date [211]. β-Glycosidases have been shown to play important roles in the survival of certain phytophagous insects [211]. The ability of peach tree borer, *Synanthedon exitiosa,* larvae to survive well on prunasin-containing peach tree is because they can metabolize cyanogenic glycosides through β-glucosidase and detoxify the released cyanide by β-cyanoalanine synthase, thereby allowing them to utilize peach trees [212]. Another example is the larvae of the tiger swallowtail, *Papilio glaucus,* which feeds on quacking aspen, which contains various phenolics glycosides (e.g. salicortin). These larvae hydrolyse the glycosides by β-glucosidase and detoxify the released phenolics aglycone by a highly active esterase, thereby allowing them to survive on aspen [213, 214].

Glutathione S-transferases are also involved in the metabolism of many toxic plant allelochemicals. These plant allelochemicals may be of many diverse groups including α, β-unsaturated carbonyl compounds (e.g. *trans*-cinnamaldehyde, trans-2-hexanal), isothiocyanates (e.g. allylisothiocyanate, benzyl isothiocyanate) and organothiocyanates (e.g. benzyl thiocyanate) as have been documented in some instances [41]. The glutathione S-transferase activities are lower in the specialist insects than in the generalists. In the crucifer-adapted cabbage looper for the metabolism of isothiocyanates (plant allelochemical), the activity of this enzyme wasfound to be two- to sixfold higher than that in the fall armyworm [215, 216]. These findings strongly advocate that glutathione S-transferases play an important role in developing resistance towards plant allelochemicals in phytophagous lepidopteran insects [215–217]. Many plant allelochemicals are potent inhibitors of glutathione S-transferases in many insects [218]. Many flavonoids, other phenols and α-, β-unsaturated carbonyl compounds are also found to be potent inhibitors of the enzymes.

H. theivora, E. flavescens and *S. dorsalis* all are polyphagous in nature. *H. theivora* known to feed on at least sixteen different plant families reported till date [11]. Similarly, *E flavescens* is also polyphagous [11]. *S. dorsalis* has been documented to attack more than 150 hosts from at least 40 different plant families [219]. Hence, these pests are exposed to a wide variety of plant allelochemicals of diverse groups having the potential to induce the activity of these resistance-related enzymes. A higher level of detoxifying enzyme activity in *H. theivora* has been reported when reared on two alternative hosts, i.e. *Mikania micrantha* (Asteraceae) and *Psidium guajava* (Myrtaceae), than on tea [220]. Over the four hundred million years of co-evolution with plants, phytophagous insects have developed diverse resistance mechanisms to cope with plant chemical defences. Because insects face a geographical mosaic of chemical environments, from non-toxic to highly toxic plants, the costs associated with resistance traits vary with the probability of encountering a toxin. Moreover, other selection pressures, such as the presence or absence of competitors and predators, can also influence the costs and selection of particular resistance traits. Thus, the complexity of the local community composition is a key factor in maintaining the diversity of adaptive mechanisms to plant xenobiotics. These mechanisms are more plastic and complex compared with those involved in resistance to insecticides, perhaps because environments in which insecticides are heavily used also tend to have communities of low diversity and complexity. However, because some detoxification enzymes are involved in plant toxins and insecticides metabolism, cross-resistance mechanisms can be predicted to be observed under specific environmental conditions. Deciphering the impact of allelochemicals in cross-resistance mechanisms with insecticides at a local scale, and comparing the molecular and evolutionary mechanisms of resistance to phytotoxins and synthetic insecticides, represent promising areas of research for developing long-term sustainable insect control strategies for the effective management of pest concern [220].

8. Genetics and insecticide resistance in tea pests

Earlier, common visible markers including mophometrices, eye colour, body spots or bands and hairs or spines, wing venation were used as phenotypic markers in studying the pattern of dispersal, mating behaviour, population variability and inheritance of genetic traits in insects [221, 222]. Although the phenotypic markers are found at all times of life span of the organism and can be readily used for studies in field conditions, they suffer from many practical limitations. The major drawback is that these visible phenotypes are relatively infrequent and often hard to score. Because the phenotype markers are rare, use of these markers in mapping a trait is difficult. For all such difficulties and with the concurrent advancement in biochemical methodologies, protein markers then became more popular. Protein markers made a significant contribution in the early periods when DNA technologies were not so much advanced, as it is now [223]. A diverse range of novel molecular (DNA) markers are now available for entomological investigations. Currently, both DNA and protein markers have revolutionized the biological sciences and have enhanced many fields of insect study, especially agricultural entomology [224].

Insecticide resistance is the result of an increase in the ability of individuals of an insect species to survive insecticide application and is an important example of man-driven evolution [225]. Alleles conferring resistance may arise and spread in populations and to other populations with variable success, depending on factors such as selective forces, genetic variability, gene flow, population size and environmental conditions [220]. Studies that map the population structure of pest insects, as well as the potential for gene flow between populations, are needed to understand the development of resistance and prevention of its spread [226, 227]. Development of resistance is often rapid in isolated populations that have been treated by insecticides [228]. The rate of development of insecticide resistance may, however, be influenced by gene flow between treated and untreated populations by maintaining the frequency of resistance alleles at a low level [229]. Contaminant exposure was a poor predictor of population structure and the level of gene flow was a better predictor of relatedness [230]. Gene flow may balance divergence by opposing the effect of selection pressures [229]. Population genetic patterns should therefore be investigated with reference to geographical variability, as well as selection pressure. Detoxification resistance occurs when enhanced levels or modified activities of biotransformation enzymes prevent the insecticide from acting on its site of action because the metabolites produced have little or no activity compared with the original substance [231]. These changes may be due to mutations resulting in a protein with slightly different properties or altered expression.

IntechOpen

As chemical control is frequently used to avoid economic damage, the sucking insects have been subjected to major selection pressure. Insecticides will probably continue to be the main control method in the near future and therefore it is important to study the structure of sucking insect population and change in insecticide susceptibility. There are several techniques for estimating the genetic diversity such as randomly amplified polymorphic DNA analysis, microsatellites, minisatellites, restriction fragment length polymorphism analysis and amplified fragment length polymorphism (AFLP) analysis. DNA markers are also suitable for use with small amounts of insect material and can be used with stored, dry or old samples. Some have complex multi-locus banding patterns, which may be of a non-Mendelian nature (e.g. randomly amplified polymorphic DNAs (RAPDs)). They have an expanding range of applications, many involving intra- and interspecific discriminations.

IntechOpen

Acknowledgements

The author expresses his sincere thanks to the Head, Department of Zoology, University of North Bengal, for providing laboratory space. The author also expresses sincere appreciation to those scientists and authors based on whose concept, hypothesis and work this chapter has been developed. The author also expresses his thanks to the University of North Bengal for providing uninterrupted local area network that has immensely helped in searching and collecting information. The author also expresses his sincere thanks to InTech Open Access Publishing Group, editor of the book '*Insecticide Resistance*' Prof. Stanislav Trdan (University of Ljubljana, Biotechnical Faculty, Dept. of Agronomy, Chair for Phytomedicine, Agricultural

Engineering, Crop Production, Pasture and Grassland Management, Slovenia) and Ms Sndra Bakic (InTech Europe, Rijeka, Croatia).

Author details

Dhiraj Saha*

Address all correspondence to: dhirajsaha_nbu@rediffmail.com; dhirajento.nbu@gmail.com

Insect Biochemistry and Molecular Biology Laboratory, Department of Zoology, University of North Bengal, Raja Rammohunpur, P.O.- North Bengal University, Siliguri, District - Darjeeling, West Bengal, India

IntechOpen

References

[1] Chen Z. M. and Chen X. F. (1989) An analysis of world tea fauna. *Journal of Tea Science* 9: 73–88.

[2] Muraleedharan N. (1992) Pest control in Asia. In Tea: Cultivation to Consumption. Wilson, K.C. and Clifford, M.N. (edited by N. Muraleedharan). Chapmann & Hall, London, pp. 375–412.

[3] Das G. M. (1965) Pests of tea in North East India and their control. Memorandom No. 27, pp. 169–173, Tocklai Experimental Station, Tea Research Association, Jorhat, Assam.

[4] Muraleedharan N. (2007) Tea insects: ecology and control, pp. 672–674. In Encyclopedia of Pest Management. CRC Press, London.

[5] Rattan P. S. (1992) Pest and disease control in Africa, pp. 331–352. In Tea: Cultivation to Consumption (edited by K. C. Wilson and M. N. Clifford). Chapman & Hall, London.

[6] Sivapalan P. (1999) Pest management in tea, pp. 625–646. In Global Advances in Tea Science (edited by N. K. Jain). Aravali Books, New Delhi.

[7] Hazarika L. K., Bhuyan M. and Hazarika B. N. (2009) Insect pests of tea and their management. *Annual Review of Entomology* 54: 267–284

[8] Saha D. and Mukhopadhyay A. (2013) Insecticide resistance mechanisms in three sucking insect pests of tea with reference to North East India - an appraisal. *International Journal of Tropical Insect Science* 33(1): 46–70.

[9] Cranham J. E. (1966) Tea pests and their control. *Annual Review of Entomology* 11: 491–514.

[10] Saha D., Roy S. and Mukhopadhyay A. (2012) Insecticide susceptibility and activity of major detoxifying enzymes in female Helopeltis theivora Waterhouse (Heteroptera: Miridae) from sub-Himalayan tea plantations of North Bengal, India. *International Journal of Tropical Insect Science* 32: 85–93.

[11] Saha, D. (2014) Assessment of Population Variability at Subcellular Level of Some Common Sucking Tea Pests from Darjeeling Hill and its Adjoining Plain. A Ph.D. Thesis. University of North Bengal, Sliguri-734013, District - Darjeeling, West Bengal, India.

[12] Chaudhuri T. C. (1999) Pesticide residues in tea, pp. 369–378. In Global Advances in Tea Science (edited by N. K. Jain). Aravali Books, New Delhi.

[13] Komagata O., Kasai S. and Tomita T. (2010) Overexpression of cytochrome P450 genes in pyrethroid- resistant Culex quinquefasciatus. *Insect Biochemistry and Molecular Biology* 40: 146–152.

[14] Das G. M. (1963) Some important pests of tea. *Two and a Bud* 10: 4–8.

[15] Mukerjea T. D. (1968) Thiodan: a broad spectrum new insecticide. *Two and a Bud* 15: 6–10.

[16] Barbora B. C. and Biswas A. K. (1996) Use pattern of pesticides in tea estates of North East India. *Two and a Bud* 47: 19–21.

[17] Sannigrahi S. and Talukdar T. (2003) Pesticide use patterns in Dooars tea industry. *Two and a Bud* 50: 35–38.

[18] Gurusubramanian G., Rahman A., Sarmah M., Roy S. and Bora S. (2008) Pesticide usage pattern in tea ecosystem, their retrospects and alternative measures. *Journal of Environmental Biology* 29: 813–826.

[19] Gurusubramanian G. and Bora S. (2007) Relative toxicity of some commonly used insecticides against adults of Helopeltis theivora Waterhouse (Miridae: Hemiptera) collected from Jorhat area tea plantations, South Assam, India. *Resistant Pest Management Newsletter* 17: 8–12.

[20] Gurusubramanian G. and Bora S. (2008) Insecticidal resistance to tea mosquito bug, Helopeltis theivora Waterhouse (Miridae: Heteroptera) in North East India. *Journal of Environmental Research Development* 2: 560–567.

[21] Gurusubramanian G., Senthilkumar N., Bora S., Roy S. and Mukhopadhyay A. (2008) Change in susceptibility in male Helopeltis theivora Waterhouse (Jorhat population, Assam, India) to different classes of insecticides. *Resistant Pest Management Newsletter* 18: 36–39.

[22] Saha D., Mukhopadhyay A. and Mahadur M. (2010) Variation in detoxifying enzymes of Assam thrips, Scirtothrips dorsalis Hood (Thysanoptera: Thripidae) from

organically and insecticide managed tea plantations. North Bengal University. *Journal of Animal Science* 4: 45–52.

[23] Saha D., Mukhopadhyay A. and Mahadur M. (2012) Detoxifying enzymes activity in tea greenfly, Empoasca flavescens Fabricius (Homoptera: Jassidae) from sub Himalayan tea plantations, pp. 257–266. In Proceedings of National Symposium on Biodiversity Status and Conservation Strategies with special Reference to North East India (edited by R. Varatharajan). Centre for Advanced Studies, Department of Life Sciences, Manipur University, Manipur, India.

[24] Saha D., Roy S. and Mukhopadhyay A. (2012) Seasonal incidence and enzyme-based susceptibility to synthetic insecticides in two upcoming sucking insect pests of tea. *Phytoparasitica* 40: 105–115.

[25] Saha D., Mukhopadhyay A. and Bahadur M. (2012) Variation in the activity of three principal detoxifying enzymes in major sucking pest of tea, Helopeltis theivora Waterhouse (Heteroptera: Miridae) from sub- Himalayan tea plantations of West Bengal, India. *Proceedings of Zoological Society*. (July-Dec 2013) 66(2): 92–99.

[26] Kumar, B., Saha, D. and Mukhopadhyay, A. (2014) Enhancement of resistant ratio vis-a-vis defence-enzymes activity in tea mosquito bug, *Helopeltis theivora* Waterhouse (Hemiptera: Miridae) selected through exposure to sub-lethal dose of monocrotophos. *Proceedings of Zoological Society*. DOI: 10.1007/s12595-014-0124-5 (Published online on 11th September, 2014)

[27] Bora S., Rahman A., Sarmah M. and Gurusubramanian G. (2007) Relative toxicity of pyrethroid and nonpyrethroid insecticides against male and female tea mosquito bug (Darjeeling strain). *Journal of Entomological Research* 37: 37–41.

[28] Roy S., Gurusubramanian G. and Mukhopadhyay A. (2008) Insecticide persistence and residual toxicity monitoring in tea mosquito bug, *Helopeltis theivora* Waterhouse (Hemiptera: Heteroptera: Miridae). *Resistant Pest Management Newsletter* 17: 9–14.

[29] Rahman A., Sarmah M., Phukan A. K., Roy S., Sannigrahi S., Borthakur M. and Gurusubramanian G. (2005) Approaches for the management of tea mosquito bug, Helopeltis theivora Waterhouse (Miridae: Heteroptera), pp. 146–161. In Proceedings of the 34th Tocklai Conference 'Strategies for Quality in the Digital Era' (edited by A. K. Barooah, M. Borthakur and J. N. Kalita). Tocklai Experimental Station, TRA, Jorhat, Assam.

[30] Roy S. (2010) Evaluation of the levels of insecticide susceptibility of *Helopeltis theivora* Waterhouse (Heteroptera: Miridae) and development of an efficacious strategy for management of the pest in Dooars tea plantation of North Bengal, PhD thesis, University of North Bengal, India.

[31] Reddy G. P. V., Prasad V. D. and Rao R. S. (1992) Relative resistance in chilli thrips, Scirtothrips dorsalis (Hood) populations in Andhra Pradesh to some conventional insecticides. *Indian Journal of Plant Protection* 20: 218–222.

[32] Vanisree K., Upendhar S. and Rajasekhar P. (2011) Toxicity of certain novel insecticides against chilli thrips, Scirtothrips Dorsalis (Hood). *Resistant Pest Management Newsletter* 21: 17–21.

[33] Seal D. R., Ciomperlik M., Richards M. L. and Klassen W. (2006) Comparative effectiveness of chemical insecticides against the chilli thrips, Scirtothrips dorsalis Hood (Thysanoptera: Thripidae), on pepper and their compatibility with natural enemies. *Crop Protection* 25: 949–955.

[34] Seal D. R., Klassen W. and Sabines C. (2007) Management of chilli thrips, Scirtothrips dorsalis (Thysanoptera: Thripidae): effectiveness of neonicotinoids and spinosyns and ineffectiveness of pyrethroids. *Proceedings of the Caribbean Food Crops Society* 43: 39–48.

[35] Dogˇramaci M., Arthurs S. P., Chen J., McKenzie C., Irrizary F. and Osborne L. (2011) Management of chilli thrips Scirtothrips dorsalis (Thysanoptera: Thripidae) on peppers by *Amblyseius swirskii* (Acari: Phytoseiidae) and *Orius insidiosus* (Hemiptera: Anthocoridae). *Biological Control* 59: 340–347.

[36] Shi C. H., Shang J. N. and Chen Y. F. (2001) Dynamic residues of imidacloprid in tea products and its application to control of tea insect pests, pp. 165–166. In Integrated Pest Management in Relation to Safe Agricultural Products (edited by Y. J. Yu and Q. H. Zhang). China Agriculture Press, Beijing.

[37] Tao T., Hu K. M., She Y. P., Zhu Q. Z. and Luo Z. (1996) Studies on the integrated management techniques of smaller green leafhopper in tea plantation of south Yunnan. *Acta Phytophysiologica Sinica* 23: 310–314.

[38] Nian-wu W., Jin-han X. U., Zheng C., Hui W., Ling-ling Z. and Xiong G. (2004) Resistance level of Empoasca vitis (Gothe) in different tea plantations [J]. *Journal of Tea Science*, 2-.

[39] Jia-Xiang Z., Jian-Wei F. U., Qing-Quan S. U., Jian-Yu L. I. and Zhi-Xiong Z. (2009) The regional diversity of resistance of Tea Green Leafhopper (Gothe), to insecticides in Fujian Province. *Journal of Tea Science* 2-.

[40] Soderland D. M. and Bloomquist J. R. (1990) Molecular mechanisms of insecticide resistance, pp. 58–96. In Pesticide Resistance in Arthropods (edited by R. T. Roush and B. E. Tabashnik). Chapman and Hall, New York.

[41] Yu S. J. (2008) The Toxicology and Biochemistry of Insecticides. CRC Press, Florida. 296 p.

[42] Wu S., Yang Y., Yuan G., Campbell P. M., Teese M. G., Russell R. J., Oakeshott J. G. and Wu Y. (2011) Overexpressed esterases in a fenvalerate resistant strain of the cotton bollworm, *Helicoverpa armigera*. *Insect Biochemistry and Molecular Biology* 41: 14–21.

[43] Buès R., Bouvier J. C. and Boudinhon L. (2005) Insecticide resistance and mechanisms of resistance to selected strains of *Helicoverpa armigera* (Lepidoptera: Noctuidae) in the south of France. *Crop Protection* 24: 814–820.

[44] Cao C.W., Zhang J. Gao X.W. Liang P. and Guo H.L. (2008) Overexpression of carboxylesterase gene associated with organophosphorous insecticide resistance in cotton aphids, *Aphis gossypii* (Glover). *Pesticide Biochemistry and Physiology* 90: 175–180.

[45] Cao C.W., Zhang J., Gao X.W., Liang P. and Guo H.L. (2008) Differential mRNA expression levels and gene sequences of carboxylesterase in both deltamethrin resistant and susceptible strains of the cotton aphid, *Aphis gossypii. Insect Science* 15: 209–216.

[46] Mitter C., Farrell B. D. and Futuyma D. J. (1991) Phylogenetic studies of insect/plant interactions: Insights into the genesis of diversity. *Trends in Ecology and Evolution* 6: 290–293.

[47] Ehrlich P. R. and Raven P. H. (1964) Butterflies and plants: a study in coevolution. *Evolution* 18: 586–608.

[48] Bernays E. A. (1998) Evolution of feeding behavior in insect herbivores: success seen as different ways to eat without being eaten. *Bioscience* 48: 35–44.

[49] Dethier V. G. (1954) Evolution of feeding preferences in phytophagous insects. *Evolution* 8: 33–54.

[50] Kaloshian I. and Walling L. L. (2005) Hemipterans as plant pathogens. *Annual Review of Plant Biology* 43: 491–521.

[51] Goggin F. L. (2007) Plant–aphid interactions: molecular and ecological perspectives. *Current Opinion in Plant Biology* 10: 399–408.

[52] Sawicki, R. M. (1987) Definition, detection and documentation of insecticide resistance, pp.105-112. In Combating resistance to xenobiotics: Biological and chemical approaches (edited by E. Hoorwood). Chichester, England,

[53] Sparks T. C., Lockwood J. A., Byford R. L., Graves J. B. and Leonard B. R. (1989) The role of behavior in insecticide resistance. *Journal of Pesticide Science* 26: 383–399.

[54] Brattsten L. B. (1988) Potential role of plant allelochemicals in the development of insecticide resistance, pp. 313–348. In Novel Aspects of Insect–Plant Interactions (edited by P. Barbosa and D. K. Letourneau). John Willey & Sons, New York.

[55] Roy S. and Mukhopadhyay A. (2011) Insecticide-Induced Change in Egg-Laying Strategy of *Helopeltis theivora* (Hemiptera: Miridae) on Tea Shoot (*Camellia sinensis*). *Proceedings of Zoological Society* 64 (1): 54–56.

[56] Price N. R. (1991) Insect resistance to insecticides: mechanisms and diagnosis. *Comparative Biochemistry and Physiology* 100C, 319–326.

[57] Vinson S. B. and Law P. K. (1971) Cuticular composition and DDT resistance in the tobacco budworm. *Journal of Economic Entomology* 64: 1387–1390.

[58] Patil V. L. and Guthrie F. E. (1979) Cuticular lipids of two resistant and a susceptible strain of houseflies. *Pesticide Science* 10: 399–406.

[59] Scott J. G. (1990) Investigating mechanisms of insecticide resistance: methods, strategies, and pitfalls, pp. 39–57. In Pesticide Resistance in Arthropods (edited by R. T. Roush and B. E. Tabashnik). Chapman and Hall, New York.

[60] Forgash A. J., Cook B. J. and Riley R. C. (1962) Mechanisms of resistance in diazinon-selected multi resistant *Musca domestica*. *Journal of Mechanisms of Economic Entomology* 55: 544–551.

[61] Van de Baan H. E. and Croft B. A. (1991) Resistance to insecticides in winter- and summer-forms of pear psylla, *Psylla pyricola*. *Pesticide Science* 32: 225–233.

[62] Kanga L. H. B. and Plapp F. W. J. (1995) Target-site insensitivity as the mechanism of resistance to organophosphorus, carbamate, and cyclodiene insecticides in tobacco budworm adults. *Journal of Economic Entomology* 88: 1150–1157.

[63] Roy S., Gurusubramanian G. and Mukhopadhyay A. (2008) Variation in endosulfan susceptibility and body lipid content of *Helopeltis theivora* Waterhouse (Heteroptera: Miridae) in relation to the use pattern of the insecticide, in sub-Himalayan Dooars tea plantations. *Journal of Plantation Crops* 36: 388–392.

[64] Martinez-Torres D., Foster S. P., Field L. M., Devonshire A. L. and Williamson M. S. (1999) A sodium channel point mutation is associated with resistance to DDT and pyrethroid insecticides in the peach-potato aphid, Myzus persicae (Sulzer) (Hemiptera: Aphididae). *Insect Molecular Biology* 8: 339–346.

[65] Mutero A., Pralavorio M., Bride J. M. and Fournier D. (1994) Resistance-associated point mutations in insecticide-insensitive acetylcholinesterase. *Proceedings of the National Academy of Sciences USA* 91: 5922–5926.

[66] Brown T. M. and Brogdon W. G. (1987) Improved detection of insecticide resistance through conventional and molecular techniques. *Annual Review of Entomology* 32: 145–162.

[67] Scharf M. E., Neal J. J. and Bennett W. G. (1998) Changes of insecticide resistance levels and detoxication enzymes, following insecticide selection in the German cockroach, *Blattella germanica* (L.). *Pesticide Biochemistry and Physiology* 59: 67–79.

[68] Casida J. E. (1993) Insecticide action at the GABA-gated chloride channel: recognition, progress, and prospects. *Archives of Insect Biochemistry and Physiology* 22: 13–23.

[69] Narahashi, T. (1996) Neuronal ion channels as the target sites of insecticides. *Pharmacology and Toxicology* 79: 1–14.

[70] Sattelle D. B. (1985) Acetylcholine receptors, pp. 395–434. In Comprehensive Insect Physiology, Biochemistry, and Pharmacology (edited by G. A. Kerkut and L. I. Gilbert). Vol. 11. Pergamon, Oxford.

[71] Byrne F.J. and Toscano N. C. (2001) An Insensitive Acetylcholinesterase Confers Resistance to Methomyl in the Beet Armyworm *Spodoptera exigua* (Lepidoptera: Noctuidae) *Journal of Economic Entomology* 94(2): 524–528.

[72] Eldefrawi A. T. (1985) Acetylcholinesterases and anticholinesterases, pp. 115–130. In Comprehensive Insect Physiology, Biochemistry, and Pharmacology (edited by G. A. Kerkut and L. I. Gilbert). Vol. 12. Pergamon Press, Oxford.

[73] Aldridge W. N. and Reiner E. (1972). Enzyme inhibitors as substrates. North-Holland Publishing Co., Amsterdam.

[74] Smissaert, H. R. (1964) Cholinesterase inhibition in spider mite susceptible and resistant to organophosphate. *Science* 143: 129–131.

[75] Fournier D. and Mutero A. (1994) Modification of acetylcholinesterase as a mechanism of resistance to insecticides. *Comparative Biochemistry and Physiology* 108C, 19–31.

[76] Hama H. (1983) Resistance to insecticides due to reduced sensitivity of acetylcholinesterase, pp. 299–331. In Pest Resistance to Pesticides (edited by G. P. Georghiou and T. Saito). Plenum Press, New York.

[77] Oppenoorth F. J. (1985) Biochemistry and genetics of insecticide resistance, pp. 731–773. In Comparative Insect Physiology, Biochemistry and Pharmacology (edited by G. A. Kerkut and L. I. Gilbert). Vol. 12. Pergamon Press, Oxford.

[78] Guedes R. N. C., Zhu K. Y., Kambhampati S. and Dover B. A. (1997) An altered acetylcholinesterase conferring negative cross-insensitivity to different insecticidal inhibitors in organophosphate-resistant lesser grain borer, *Rhyzopertha dominica*. *Pesticide Biochemistry and Physiology* 58: 55–62.

[79] Harold J. A. and Ottea J. A. (1997) Toxicological significance of enzyme activities in profenofos resistant tobacco budworms. *Heliothis virescens* (F.). *Pesticide Biochemistry and Physiology* 58: 23–33.

[80] Levitin E. and Cohen E. (1998) The involvement of ace-tylcholinesterase in resistance of the California red scale, *Aonidiella aurantii* to organophosphorus pesticides. *Entomologia Experimentalis et Appllicata* 88: 115–121.

[81] Sarker M. and Mukhopadhyay A. (2006a) Studies on salivary and midgut enzymes of a major sucking pest of tea, Helopeltis theivora (Heteroptera: Miridae) from Darjeeling plains, India. *Journal of the Entomological Research Society* 8: 27–36.

[82] Sarker M. and Mukhopadhyay A. (2006b) Studies on some enzymes related to insecticide resistance in Helopeltis theivora Waterhouse (Insecta: Heteroptera: Miridae) from Darjeeling foothills and plains. *Journal of Plantation Crops* 34: 423–428.

[83] Li X., Schuler M. A. and Berenbaum M. R. (2007) Molecular mechanisms of metabolic resistance to synthetic and natural xenobiotics. *Annual Review of Entomology* 52: 231–253.

[84] Feyereisen R. (2012) Insect CYP genes and P450 enzymes, pp. 236–316. In Insect Molecular Biology and Biochemistry (edited by L. I. Gilbert). Academic Press, London.

[85] Lee S. H., Kang J. S., Min J. S., Yoon K. S., Strycharz J. P., Johnson, R., Mittapalli, O., Margam, V.M., Sun, W., Li, H.M., Xie, J., Wu, J,, Kirkness, E.F., Berenbaum, M.R., Pittendrigh, B.R. and Clark, J.M. (2010) Decreased detoxification genes and genome size make the human body louse an efficient model to study xenobiotic metabolism. *Insect Molecular Biology* 19: 599–615.

[86] Arensburger P., Megy K., Waterhouse R. M., Abrudan J., Amedeo P., Antelo, B., Bartholomay L., Bidwell S., Caler E., Camara F., Campbell C.L., Campbell K.S., Casola C., Castro M.T., Chandramouliswaran I., Chapman S.B., Christley S., Costas J., Eisenstadt E., Feschotte C., Frasher-Liggett C., Guigo R., Haas B., Hammond M., Hansson B.S., Hemingway J. Hill S.R., Howarth C., Ignell R., Kennedy R.C., Kodira C.D., Lobo N.F., Mao C., Mayhew G., Michel K., Mori A., Liu N., Naveira H., Nene V., Nguyen N., Pearson M.D., Pritham E.J., Puiu D., Qi Y., Ranson H., Ribeiro J.M. Roberston H.M., Severson D.W., Shumway M., Stanke M., Strausberg R.L., Sun C., Sutton G., Tu Z.J., Tubio J.M., Unger M.F., Van Lan Dingham D.L., Vilella A.J., White O., White J.R., Wondji C.S., Wortman J., Zdobnov E.M., Birren B., Christensen B.M., Collins F.H., Cornel A., Dimopoulos G., Hannick L.I., Higgs S., Lanzaro G.C., Lawson D., Lee N.H., Muskavitch M.A., Raikhel A.S., Atkinson P.W., (2010) Sequencing of *Culex quinquefasciatus* establishes a platform for mosquito comparative genomics. *Science* 330: 86–88.

[87] Omura T. and Sato R. (1964) The carbon monoxide-binding pigment of liver microsomes I. Evidence for its hemoprotein nature. *Journal of Biological Chemistry* 239: 2370–2378.

[88] Feyereisen R., Koener J. F., Farnsworth D. E. and Nebert D. W. (1989) Isolation and sequence of cDNA encoding a cytochrome P-450 from an insecticide-resistant strain of the house fly, *Musca domestica*. *Proceedings of the National Academy of Science USA* 86: 1465–1469.

[89] Feyereisen R. (2011) Arthropod CYPomes illustrate the tempo and mode in P450 evolution. *Biochimica et Biophysica Acta* 1814: 19–28.

[90] Claudianos C., Ranson H., Johnson R. M., Biswas S., Schuler M. A., Berenbaum M. A., Feyereisen R. and Oakeshott J. G. (2005) A deficit of detoxification enzymes: pesticide sensitivity and environmental response in the honeybee. Insect Molecular Biology 15, 615–636.

[91] Hardstone M. C. and Scott J. G. (2010) Is *Apis mellifera* more sensitive to insecticides than other insects? *Pest Management Science* 66: 1171–1180.

[92] Hemingway J. and Karunaratne S. H. P. P. (1998) Mosquito carboxylesterases: a review of the molecular biology and biochemistry of a major insecticide resistance mechanism. *Medical and Veterinary Entomology* 12: 1–12.

[93] Heymann E. and Jakoby W.B. (1980) Carboxylesterases and amidases, pp. 291–323. In Enzymatic basis of detoxication (edited by E. Heymann and W. B. Jokoby). Academic Press, New York,.

[94] Aldridge W.N. (1993) The esterases: perspectives and problems. *Chemico-Biological Interactions* 87: 5–13.

[95] Reiner E. (1993) Recommendations of the IUBMB nomenclature committee – comments concerning classification and nomenclature of esterases hydrolysing organophosphorus compounds. *Chemico-Biological Interactions* 87: 15, 16.

[96] Walker C.H. (1993) The classification of esterases which hydrolyse organophosphates–recent developments. *Chemico-Biological Interactions* 87: 17–24.

[97] Yu S.J. (1996) Insect glutathione *S*-transferases. *Zoological Studies* 35: 9–19.

[98] Ranson H., Claudianos C., Ortelli F., Abgrall C., Hemingway J., Sharakhova M.V., Unger M.F., Collins F.H. and Feyereisen R. (2002) Evolution of supergene families associated with insecticide resistance. *Science* 298: 179–181.

[99] Ranson, H. and Hemingway, J. (2005) Mosquito glutathione transferases. Review. *Methods in Enzymology* 401: 226–241.

[100] Fournier D., Bride J.B., Poirie M., Berge J.P. and Plapp F.W. Jr. (1992) Insect Glutathione S-transferases: Biochemical characteristics of the major forms from house flies susceptible and resistant to insecticides. *Journal of Biological Chemistry* 267: 1840–1845.

[101] Ranson H., Rossiter L., Ortelli F., Jensen B., Wang X., Roth C.W., Collins F.H. and Hemingway J. (2001) Identification of a novel class of insect glutathione S-transferases involved in DDT resistance in the malaria vector, *Anopheles gambiae*. *Biochemical Journal* 359: 295–304.

[102] Yu S.J. (2002) Biochemical characteristics of microsomal and cytosolic glutathione S-transferases in larvae of the fall armyworm, *Spodoptera frugiperda* (J. E. Smith). *Pesticide Biochemistry and Physiology* 72: 100–110.

[103] Ma B. and Chang F.N. (2007) Purification and cloning of a delta class glutathione s-transferase displaying high peroxidase activity isolated from German cockroach, *Blattella germanica*. *FEBS Journal* 274: 1793–1803.

[104] Che-Mendoza A., Penilla R.P. and Rodríguez D.A. (2009) Insecticide resistance and glutathione S-transferases in mosquitoes: A review. *African Journal of Biotechnology* 8 (8): 1386–1397

[105] Clark A.G. and Shamaan N.A. (1984) Evidence that DDT-dehydrochlorinase from the house fly is a glutathione S-transferase. *Pesticide Biochemistry and Physiology* 22: 249–261.

[106] Eldefrawi M.E., Miskus R. and Sutcher V. (1960) Methylenedioxyphenyl derivatives as synergists for carbamate insecticides on susceptible, DDT-and parathion-resistant house flies. *Journal of Economic Entomology* 53: 231–234.

[107] Georghiou G. P. and Metcalf R. L. (1961) The adsorption and metabolism of 3-isopropylphenyl-n-methylcarbamate by susceptible and carbamate selected strains of house flies. *Journal of Economic Entomology* 54(2): 231–233.

[108] Schonbrod R.D., Phillco W.W. and Terriere L.C. (1965) Hydroxylation as a factor in resistance in house flies and blow flies. *Journal of Economic Entomology* 58(1): 74–77.

[109] Agosin M. (1985) Role of microsomal oxidations in insecticide degradation, pp. 647–712. In Comparative Insect Physiology, Biochemistry and Pharmacology (edited by L. I. Gilbert and G. A. Kerkut). Vol. 12. Pergamon, Oxford.

[110] Scott J. G. (1991) Insecticide resistance in insects, pp. 663–677. In Handbook of Pest Management (edited by D. Pimental). CRC Press, Boca Raton.

[111] Konno T. E. H. and Dauterman W. C. (1989) Studies on methyl parathion resistance in Heliothis virescens. *Pesticide Biochemistry and Physiology* 33: 189–199.

[112] Oppenoorth F. J. (1985) Biochemistry and genetics of insecticide resistance, pp. 731–773. In Comparative insect physiology, biochemistry and pharmacology (edited by G. A. Kerkut and L. I. Gilbert). Vol. 12. Pergamon Press Oxford, UK.

[113] Hatano R., Scott J. G. and Dennehy T. J. (1992) Enhanced activation is the mechanism of negative crossresistance to chlorpyrifos in the dicofol-IR strain of *Tetranychus urticae* (Acari: Tetranychidae). *Journal of Economic Entomology* 85: 1088–1091.

[114] Cilek J. E., Dahlman D. L. and Knapp F.W. (1995) Possible mechanism of diazinon negative cross-resistance in pyrethroid-resistant horn flies (Diptera: Muscidae). *Journal of Economic Entomology* 88: 520–524.

[115] Martin T., Chandre F., Ochou O. G., Vaissayre M. and Fournier D. (2003) Oxidases responsible for resistance to pyrethroids sensitize *Helicoverpa armigera* (Hubner) to triazophos in West Africa. *Insect Biochemistry and Molecular Biology* 33: 883–887.

[116] Drabek J. and Neumann R. (1985) Proinsecticides, pp. 35–86. In Insecticides (edited by D. H. Huston and T. R. Roberts). Wiley, London.

[117] Black B. C., Hollingworth R. M., Ahammadsahib K. I., Kukel C. D. and Donovan S. (1994) Insecticidal action and mitochondrial uncoupling activity of AC303, 630 and related halogenated pyrroles. *Pesticide Biochemistry and Physiology* 50: 115–128.

[118] Kayser H. and Eilinger P. (2001) Metabolism of diafenthiuron by microsomal oxidation: Procide activation and inactivation as mechanisms contributing to selectivity. *Pest Management Science* 57: 975–980.

[119] Pimprale S. S., Besco C. L., Bryson P. K. and Brown T. M. (1997) Increased susceptibility of pyrethroid-resistant tobacco budworm (Lepidoptera: Noctuidae) to chlorfenapyr. *Journal of Economic Entomology* 90: 49–54.

[120] Joussen N., Heckel D. G., Haas M., Schuphan I. and Schmidt B. (2008) Metabolism of imidacloprid and DDT by P450 CYP6G1 expressed in cell cultures of *Nicotiana tabacum* suggests detoxification of these insecticides in Cyp6g1-overexpressing strains of *Drosophila melanogaster*, leading to resistance. *Pest Management Science* 64: 65–73.

[121] Karunker I., Morou E., Nikou D., Nauen R., Sertchook R., Stevenson B. J., Paine M. J. I., Morin S. and Vontas J. (2009) Structural model and functional characterization of the *Bemisia tabaci* CYP6CM1vQ, a cytochrome P450 associated with high levels of imidacloprid resistance. *Insect Biochemistry and Molecular Biology* 39: 697–706.

[122] Feyereisen R., Koener J. F., Farnsworth D. E. and Nebert D. W. (1989) Isolation and sequence of cDNA encoding a cytochrome P-450 from an insecticide-resistant strain of the house fly, *Musca domestica*. *Proceedings of the National Academy of Science USA* 86: 1465–1469.

[123] Cariño F., Koener J. F., Plapp F. W., Jr. and Feyereisen R. (1992) Expression of the cytochrome P450 gene CYP6A1 in the housefly, *Musca domestica*. *ACS Symposium Series* 505: 31–40.

[124] Sabourault C., Guzov V.M., Koener J.F., Claudianos C., Plapp F.W. and Feyereisen R., (2001) Overproduction of a P450 that metabolizes diazinon is linked to a loss-of-function in the chromosome 2 ali-esterase (Mda E7) gene in resistant house flies. Insect Molecular Biology 10: 609–618

[125] Zhu F., Liu N. (2008a) Differential expression of CYP6A5 and CYP6A5v2 in pyrethroid resistant house flies, *Musca domestica*. Archives of Insect Biochemistry and Physiology 67: 107–119.

[126] Zhu F., Feng J., Zhang L., Liu N. (2008b) Characterization of two novel cytochrome P450 genes in insecticide-resistant house-flies. Insect Molecular Biology 17: 27–37.

[127] Zhu F., Li T., Zhang L., Liu N. (2008c) Co-up-regulation of three P450 genes in response to permethrin exposure in permethrin resistant house flies, Musca domestica. BMC Physiology 8: 18.

[128] Liu N. and Scott J. G. (1996) Genetic analysis of factors controlling high-level expression of cytochrome P450, CYP6D1, cytochrome b5, P450 reductase, and monooxygenase activities in LPR house flies, *Musca domestica*. *Biochemical Genetics* 34: 133–148.

[129] Kasai S. and Scott J. G. (2001a) Cytochrome P450s CYP6D3 and CYP6D1 are part of a P450 gene cluster on autosome 1 in the house fly. *Insect Molecular Biology* 10: 191–196.

[130] Kasai S. and Scott J. G. (2001b) Expression and regulation of CYP6D3 in the house fly, *Musca domestica* (L.). *Insect Biochemistry and Molecular Biology* 32: 1–8.

[131] Kasai S. and Scott J. G. (2000) Overexpression of cytochrome P450 CYP6D1 is associated with monooxygenase-mediated pyrethroid resistance in house flies from Georgia. *Pesticide Biochemistry and Physiology* 68: 34–41.

[132] Kamiya E., Yamakawa M., Shono T. and Kono Y. (2001) Molecular cloning, nucleotide sequences and gene expression of new cytochrome P450s (CYP6A24, CYP6D3v2) from the pyrethroid resistant housefly, *Musca domestica* L. (Diptera: Muscidae). *Applied Entomology and Zoology* 36: 225–229.

[133] Shono T., Kasai S., Kamiya E., Kono Y. and Scott J. G. (2002) Genetics and mechanisms of permethrin resistance in the YPER strain of house fly. *Pesticide Biochemistry and Physiology* 73: 27–36.

[134] Guzov V. M., Unnithan G. C., Chernogolov A. A. and Feyereisen R. (1998) CYP12A1, a mitochondrial cytochrome P450 from the house fly. *Archives of Biochemistry and Biophysics* 359: 231–240.

[135] Waters L. C., Zelhof A. C., Shaw B. J. and Ch'ang L. Y. (1992) Possible involvement of the long terminal repeat of transposable element 17.6 in regulating expression of an insecticide resistance-associated P450 gene in *Drosophila*. *Proceedings of the National Academy of Science USA* 89: 4855–4859.

[136] Maitra S., Dombrowski S. M., Waters L. C. and Ganguly R. (1996) Three second chromosome-linked clustered Cyp6 genes show differential constitutive and barbital-induced expression in DDT-resistant and susceptible strains of *Drosophila melanogaster*. *Gene* 180: 165–171.

[137] Maitra S., Dombrowski S. M., Basu M., Raustol O., Waters L. C. and Ganguly R. (2000) Factors on the third chromosome affect the level of cyp6a2 and cyp6a8 expression in *Drosophila melanogaster*. *Gene* 248: 147–156.

[138] Maitra S., Price C. and Ganguly R. (2002) *Cyp6a8* of *Drosophila melanogaster*: Gene structure, and sequence and functional analysis of the upstream DNA. *Insect Biochemistry and Molecular Biology* 32: 859–870.

[139] Le Goff G., Boundy S., Daborn P. J., Yen J. L., Sofer L. Lind R., Sabourault C., Madi-Ravazzi L. and ffrench-Constant R.H. (2003) Microarray analysis of cytochrome P450 mediated insecticide resistance in *Drosophila*. *Insect Biochemistry and Molecular Biology* 33: 701–708.

[140] Daborn P., Boundy S., Yen J., Pittendrigh B. and ffrench-Constant R. (2001) DDT resistance in *Drosophila* correlates with *Cyp6g1* over-expression and confers crossresistance to the neonicotinoid imidacloprid. *Molecular Genetics and Genomics* 266: 556–563.

[141] Daborn P. J., Yen J. L., Bogwitz M. R., Le Goff G., Feil E., Jeffers S., Tijet N., Perry T., Heckel D., Batterham P., Feyereisen R., Wilson T. G. and ffrench-Constant R. H.

(2002) A single P450 allele associated with insecticide resistance in *Drosophila*. *Science* 297: 2253–2256.

[142] Catania F., Kauer M. O., Daborn P. J., Yen J. L., ffrench-Constant R. H. and Schlotterer C. (2004) World-wide survey of an Accord insertion and its association with DDT resistance in *Drosophila melanogaster*. *Molecular Ecology* 13: 2491– 2504.

[143] Brandt A., Scharf M., Pedra J. H., Holmes G., Dean A., Kreitman M. and Pittendrigh B. R. (2002) Differential expression and induction of two *Drosophila* cytochrome P450 genes near the Rst (2) DDT locus. *Insect Molecular Biology* 11: 337–341.

[144] Pyke F. M., Bogwitz M. R., Perry T., Monk A., Batterham P. and McKenzie J. A. (2004) The genetic basis of resistance to diazinon in natural populations of *Drosophila melanogaster*. *Genetica* 121: 13–24.

[145] Bogwitz M. R., Chung H., Magoc L., Rigby S., Wong W., O'Keefe M., McKenzie J.A., Batterham P. and Daborn P. J. (2005) *Cyp12a4* confers lufenuron resistance in a natural population of *Drosophila melanogaster*. *Proceedings of the National Academy of Science USA* 102: 12807–12812.

[146] Nikou D., Ranson H. and Hemingway J. (2003) An adult specific CYP6 P450 gene is overexpressed in a pyrethroids resistant strain of the malaria vector, *Anopheles gambiae*. *Gene* 318: 91–102.

[147] David J. P., Strode C., Vontas J., Nikou D., Vaughan A., Pignatelli P.M., Louis C., Hemingway J. and Ranson H. (2005) The *Anopheles gambiae* detoxification chip: A highly specific microarray to study metabolic-based insecticide resistance in malaria vectors. *Proceedings of the National Academy of Science USA* 102: 4080–4084.

[148] Djouaka R. F., Bakare A. A., Coulibaly O. N., Akogbeto M. C., Ranson H., Hemingway J. and Strode C. (2008) Expression of the cytochrome P450s, CYP6P3 and CYP6M2 are significantly elevated in multiple pyrethroid resistant populations of *Anopheles gambiae* s.s. from Southern Benin and Nigeria. *BMC Genomics* 9: 538.

[149] Muller P., Warr E., Stevenson B. J., Pignatelli P. M., Morgan J. C., Steven A., Yawson A. E., Mitchel S. N., Ranson H., Hemingway J. Paine M. J. I. Donnelly M. J. (2008) Field-caught Permithrin-Resistant *Anopheles gambiae* overexpress CYP6P3, a P450 that metabolises Pyrethroids. *PLoS Genetics* 4 (11): e1000286.

[150] Muller P., Donnelly M. J. and Ranson H. (2007) Transcription profiling of a recently colonised pyrethroid resistant *Anopheles gambiae* strain from Ghana. *BMC Genomics* 8: 36.

[151] Vontas J., Blass C., Koutsos A. C., David J. P., Kafatos F. C., Louis C., Hemingway J. Christophides G. K. and Ranson H. (2005) Gene expression in insecticide resistant and susceptible *Anopheles gambiae* strains constitutively or after insecticide exposure. *Insect Molecular Biology* 14: 509–521.

[152] Vontas J., David J. P., Nikou D., Hemingway J., Christophides G. K., Louis C. and Ranson H. (2007) Transcriptional analysis of insecticide resistance in *Anopheles ste-*

phensi using cross-species microarray hybridization. *Insect Molecular Biology* 16: 315–324.

[153] Amenya D. A., Naguran R., Lo T. C., Ranson H., Spillings B. L., Wood O.R., Brooke B. D., Coetzee M. and Koekemoer L. L. (2008) Over expression of a cytochrome P450 (CYP6P9) in major African malaria vector, *Anopheles funestus*, resistant to pyrethroids. *Insect Molecular Biology* 17: 19–25.

[154] Wondji C. S., Irving H., Morgan, J., Lobo N. F., Collins F. H., Hunt R.H., Coetzee M., Hemingway J. and Ranson H. (2009) Two duplicated P450 genes are associated with pyrethroid resistance in *Anopheles funestus*, a major malaria vector. *Genome Research* 19: 452–459.

[155] Strode C., Steen K., Ortelli F. and Ranson H. (2006) Differential expression of the detoxification genes in the different life stages of the malaria vector *Anopheles gambiae*. *Insect Molecular Biology* 15: 523–530.

[156] Hardstone M. C., Komagata O., Kasai S., Tomita T. and Scott J. G. (2010) Use of isogenic strains indicates CYP9M10 is linked to permethrin resistance in *Culex pipiens quinquefasciatus*. *Insect Molecular Biology* 19: 717–726.

[157] Kasai S., Weerashinghe I. S., Shono T. and Yamakawa M. (2000) Molecular cloning, nucleotide sequence and gene expression of a cytochrome P450 (CYP6F1) from the pyrethroid-resistant mosquito, *Culex quinquefasciatus* Say. *Insect Biochemistry and Molecular Biology* 30: 163–171.

[158] Komagata O., Kasai S. and Tomita T. (2010) Overexpression of cytochrome P450 genes in pyrethroid-resistant *Culex quinquefasciatus*. *Insect Biochemistry and Molecular Biology* 40: 146–152.

[159] Shen B., Dong H. Q., Tian H. S., Ma L., Li X. L., Wu G. L. and Zhu C. L. (2003) Cytochrome P450 genes expressed in the deltamethrin- susceptible and -resistant strains of *Culex pipiens pallens*. *Pesticide Biochemistry and Physiology* 75: 19–26.

[160] Rose R. L., Goh D., Thompson D. M., Verma K. D., Heckel D. G., Gahan L. J., Roe R. M. and Hodgson E. (1997) Cytochrome P450 (CYP) 9A1 in *Heliothis virescens*: The first member of a new CYP family. *Insect Biochemistry and Molecular Biology* 27: 605–615.

[161] Hopkins B. W., Longnecker M. T. and Pietrantonio P. V. (2010) Transcriptional overexpression of CYP6B8/CYP6B28 and CYP6B9 is a mechanism associated with cypermethrin survivorship in field-collected *Helicoverpa zea* (Lepidoptera: Noctuidae) moths. *Pest Management Science* 67: 21–25.

[162] Pittendrigh B., Aronstein K., Zinkovsky E., Andreev O., Campbell B., Daly J., Trowell S. and ffrench-Constant R. H. (1997) Cytochrome P450 genes from *Helicoverpa armigera*: Expression in a pyrethroid-susceptible and -resistant strain. *Insect Biochemistry and Molecular Biology* 27: 507–512.

[163] Ranasinghe C. and Hobbs A. A. (1998) Isolation and characterization of two cytochrome P450 cDNA clones for CYP6B6 and CYP6B7 from *Helicoverpa armigera* (Hub-

ner): Possible involvement of CYP6B7 in pyrethroid resistance. *Insect Biochemistry and Molecular Biology* 28: 571–580.

[164] Yang Y., Chen S., Wu S., Yue L. and Wu Y. (2006) Constitutive overexpression of multiple cytochrome P450 genes associated with pyrethroid resistance in *Helicoverpa armigera*. *Journal of Economic Entomology* 99: 1784–1789.

[165] Wee C. W., Lee S. F., Robin C. and Heckel D. G. (2008) Identification of candidate genes for fenvalerate resistance in *Helicoverpa armigera* using cDNA-AFLP. *Insect Molecular Biology* 17: 351–360.

[166] Brun-Barale A., Hema O., Martin T., Suraporn S., Audant P., Sezutsu H. and Feyereisen R. (2010) Multiple P450 genes overexpressed in deltamethrin- resistant strains of *Helicoverpa armigera*. *Pest Management Science* 66: 900–909.

[167] Bautista M. A., Tanaka T. and Miyata T. (2007) Identification of permethrin-inducible cytochrome P450s from the diamondback moth, *Plutella xylostella* (L.), and the possibility of involvement in permethrin resistance. *Pesticide Biochemistry and Physiology* 87: 85–93.

[168] Baek J. H., Clark J. M. and Lee S. H. (2010) Cross-strain comparison of cypermethrin-induced cytochrome P450 transcription under different induction conditions in diamondback moth. Pesticide Biochemistry and Physiology 96: 43–50.

[169] Zhu Y. C. and Snodgrass G. L. (2003) Cytochrome P450 CYP6X1 cDNAs and mRNA expression levels in three strains of the tarnished plant bug *Lygus lineolaris* (Heteroptera: Miridae) having different susceptibilities to pyrethroids insecticide. *Insect Molecular Biology* 12: 39–49.

[170] Karunker I., Benting J., Lueke B., Ponge T., Nauen R., Roditakis E., Vontas J., Gorman K., Denholm I. and Morin S. (2008) Over-expression of cytochrome P450 CYP6CM1 is associated with high resistance to imidacloprid in the B and Q biotypes of *Bemisia tabaci* (Hemiptera: Aleyrodidae). *Insect Biochemistry and Molecular Biology* 38: 634–644.

[171] Bass C., Carvalho R. A., Oliphant L., Puinean A. M., Field L. M., Nauen R., Williamson M. S., Moores, G. and Gorman, K. (2011) Overexpression of a cytoichrome P450 monooxygenase, CYP6ER1, is associated with resistance to imidacloprid in the brown plant hopper, *Nilapatvata lugens*. *Insect Molecular Biology* 20(6): 763–773.

[172] Puinean A. M., Foster S. P., Oliphant L., Denholm I., Field L. M. Millar N. S., Williamson M. S. and Bass C. (2010) Amplification of a cytochrome P450 gene is associated with resistance to neonicotinoid insecticides in the aphid *Myzus persicae*. *PLoS Genetics* 6(6): e1000999. doi:10.1371/journal.pgen.1000999.

[173] Scharf M. E., Parimi S., Meinke L. J., Chandler L. D. and Siegfried B. D. (2001) Expression and induction of three family 4 cytochrome P450 (CYP4)* genes identified from insecticide-resistant and -susceptible western corn rootworms, *Diabrotica virgifera virgifera*. *Insect Molecular Biology* 10: 139–146.

[174] Zhu F., Parthasarathy R., Bai H., Woithe K., Kaussmann M., Nauen R., Harrison D. A. and Palli S. R. (2010) A brain-specific cytochrome P450 responsible for the majority of deltamethrin resistance in the QTC279 strain of *Tribolium castaneum*. *Proceedings of the National Academy of Sciences USA* 107: 8557–8562.

[175] Scharf M. E., Lee C. Y., Neal J. J. and Bennett G. W. (1999) Cytochrome P450 MA expression in insecticide-resistant German cockroaches (Dictyoptera: Blattellidae). *Journal of Economic Entomology* 92: 788–793.

[176] Pridgeon J. W., Zhang L. and Liu N. (2003) Overexpression of CYP4G19 associated with a pyrethroid-resistant strain of the German cockroach, *Blattella germanica* (L.). *Gene* 314: 157–163.

[177] Zhu Y. C., Snodgrass G. L. and Ming Shun Chen M. S. (2004) Enhanced esterase gene expression and activity in a malathion-resistant strain of the tarnished plant bug, Lygus lineolaris. *Insect Biochemistry and Molecular Biology* 34: 1175–1186.

[178] Zhu Y. C., West S., Snodgrass G. and Luttrell R. (2011) Variability in Resistance-related Enzyme Activities in Field Populations of the Tarnished Plant Bug, *Lygus lineolaris*. *Pesticide Biochemistry and Physiology* 99: 265–273.

[179] Ferrari J. A., Morse J. G., Georghiou G. P., Sun Y (1993) Elevated esterase activity and acetylcholinesterase insensitivity in citrus thrips (Thysanoptera: Thripsidae) populations from the San Joaquin Valley of California. *Journal of Economic Entomology* 86: 1645–1650.

[180] Vassiliou V., Emmanouilidou M., Perrakis A., Morou E., Vontas J., Tsagkarakou A. and Roditakis E. (2011) Insecticide resistance mechanisms in tea pests 69 Insecticide resistance in *Bemisia tabaci* from Cyprus. *Insect Science* 18 30–39.

[181] Wink M. and Waterman P. (1999) Chemotaxonomy in relation to molecular phylogeny of plants. *Annual Plant Reviews* 2, 300–341

[182] Wink M. (2003) Evolution of secondary metabolites from an ecological and molecular phylogenetic perspective. *Phytochemistry* 64: 3–19.

[183] Yu S. J. and Ing R. T. (1984) Microsomal biphenyl hydroxylase of fall armyworm larvae and its induction by allelochemicals and host plants. *Comparative Biochemistry and Physiology* 78C: 145–152.

[184] Musser R. O., Hum-Musser S. M., Eichenseer H., Peiffer M., Ervin G., Murphy J. B. and Felton G. W. (2002) Herbivory: caterpillar saliva beats plant defences – a new weapon emerges in the evolutionary arms race between plants and herbivores. *Nature* 416: 599–600.

[185] Helmus M. R. and Dussourd D. E. (2005) Glues or poisons: which triggers vein cutting by monarch caterpillars? *Chemoecology* 15: 45–49

[186] Labeyrie E. and Dobler S. (2004) Molecular adaptation of *Chrysochus* leaf beetles to toxic compounds in their food plants. *Molecular Biology and Evolution* 21: 218–221.

[187] Zagrobelny M., Bak S., Rasmussen A. V., Jørgensen B., Naumann C. M. and Møller B. L. (2004) Cyanogenic glucosides and plant–insect interactions. *Phytochemistry* 65: 293–306.

[188] Francis F., Vanhaelen N. and Haubruge E. (2005) Glutathione S-transferases in the adaptation to plant secondary metabolites in the Myzus persicae aphid. *Archives of Insect Biochemistry and Physiology* 58: 166–174.

[189] Ratzka A., Vogel H., Kliebenstein D. J. Mitchell-Olds T. and Kroymann J. (2002) Disarming the mustard oil bomb. *Proceedings of the National Academy of Science USA* 99: 11223–11228.

[190] Wittstock U., Agerbirk N., Stauber E. J., Olsen C. E., Hippler M., Mitchell-Olds T., Gershenzon J. and Vogel H. (2004) Successful herbivore attack due to metabolic diversion of a plant chemical defense. *Proceedings of the National Academy of Science USA* 101: 4859–4864.

[191] Fogleman J.C. (2000) Response of Drosophila melanogaster to selection for P450-mediated resistance to isoquinoline alkaloids. *Chemico-Biological Interactions* 125: 93–105.

[192] Hartmann T., Theuring C., Beuerle T., Klewer N., Schulz S., Singer M. S. and Bernays E. A. (2005) Specific recognition, detoxification and metabolism of pyrrolizidine alkaloids by the polyphagous arctiid Estigmene acrea. *Insect Biochemistry and Molecular Biology* 35: 391–411.

[193] Naumann C., Hartmann T. and Dietrich Ober D. (2002) Evolutionary recruitment of a flavin dependent monooxygenase for the detoxification of host plant acquired pyrrolizidine alkaloids in the alkaloid-defended arctiid moth Tyria jacobaeae. *Proceedings of the National Academy of Science USA* 99: 6085–6090.

[194] Glendinning J. I. (2002) How do herbivorous insects cope with noxious secondary plant compounds in their diet? *Entomologia Experimentalis et Applicata* 104: 15–25.

[195] Nyman T. and Julkunen-Tiitto R. (2000) Manipulation of the phenolic chemistry of willows by gall-inducing sawflies. *Proceedings of the National Academy of Science USA* 97: 13184–13187.

[196] Luque T., Okano K. and O'Reilly D. R. (2002) Characterization of a novel silkworm (*Bombyx mori*) phenol UDP-glucosyltransferase. *European Journal of Biochemistry* 269: 819–825.

[197] Willinger G. and Dobler S. (2001) Selective sequestration of iridoid glycosides from their host plants in Longitarsus flea beetles. *Biochemical Systematics and Ecology* 29: 335–346.

[198] Petersen R. A., Zangerl A. R., Berenbaum, M. R. and Schuler M. A. (2001) Expression of CYP6B1 and CYP6B3 cytochrome P450 monooxygenases and furanocoumarin metabolism in different tissues of *Papilio polyxenes* (Lepidoptera: Papilionidae). *Insect Biochemistry and Molecular Biology* 31: 679–690.

[199] Nitao J. K., Berhow M., Duval S. M., Weisleder, D., Vaughn S. F., Zangerl A. and Berenbaum M. R. (2003) Characterization of furanocoumarin metabolites in parsnip webworm, *Depressaria pastinacella*. *Journal of Chemical Ecology* 29: 671–682.

[200] Yu S. J. (2002) Biochemical characteristics of microsomal and cytosolic glutathione S-transferases in larvae of the fall armyworm, *Spodoptera frugiperda* (J. E. Smith). *Pesticide Biochemistry and Physiology* 72: 100–110.

[201] Moon J., J., Salzman R. A., Ahn J. E., Koiwa H. and Zhu-Salzman K. (2004) Transcriptional regulation in cowpea bruchid guts during adaptation to a plant defence protease inhibitor. *Insect Molecular Biology* 13: 283–291.

[202] Bede J. C., Musser R. O., Gary W. Felton G. W. and Korth K. L. (2006) Caterpillar herbivory and salivary enzymes decrease transcript levels of *Medicago truncatula* genes encoding early enzymes in terpenoid biosynthesis. *Plant Molecular Biology* 60: 519–531.

[203] Barbehenn R. V., Poopat U. and Spencer B. (2003) Semiquinone and ascorbyl radicals in the gut fluids of caterpillars measured with EPR spectrometry. Insect Biochemistry and Molecular Biology 33: 125–130

[204] Berenbaum M. R. (2002) Postgenomic chemical ecology: from genetic code to ecological interactions. Journal of Chemical Ecology 28(5): 873–896.

[205] Meyran J. C., David J. C., Rey J. P., Cuany D. Bride A. and Amichot J. M. (2002). The biochemical basis of dietary polyphenols detoxification by aquatic detritivorous Arthropoda. *Recent Research Developments in Analytical Biochemistry* 2: 185–199.

[206] Fraenkel G. S. (1959) The raison d'etre of secondary plant substances. *Science* 129: 1466–1470.

[207] Krieger R. I., Feeny P. P. and Wilkinson C. F. (1971) Detoxication enzymes in the guts of caterpillars: an evolutionary answer to plant defenses? *Science* 172: 579–581.

[208] Mullin C. A. (1986) Adaptive divergence of chewing and sucking arthropods to plant allelochemicals, pp. 175–209. In Molecular Aspects of Insect-Plant Associations (edited by L. B. Brattsten and S. Ahmad). Plenum Press, New York.

[209] Mullin C. A., Croft B. A., Strickler K., Matsumura F. and Miller J. R. (1982) Detoxification enzyme differences between a herbivorous and predatory mite. *Science* 217: 1270, 1271.

[210] Iyenger S., Arnason J. T., Philogene B. J. R., Werstiuk N. H. and Morand P. (1990) Comparative metabolism of the photoxic allelochemical α-terthienyl in three species of lepidopterans. *Pesticide Biochemistry and Physiology* 37(2): 154–164.

[211] Yu S. J. (1983) Induction of detoxifying enzymes by allelochemicals and host plants in the fall armyworm. *Pesticide Biochemistry and Physiology* 19: 330–336.

[212] Reilly C. C., Gentry C. R. and McKay J. R. (1987) Biochemical evidence for resistance of root stocks to the peach tree borer and species separation of peach tree borer and lesser peach tree borer (Lepidoptera: Sesiidae) on peach trees. *Journal of Economic Entomology* 80: 338–343.

[213] Lindroth R. L. (1989) Host plant alteration of detoxication activity in Papilio glaucus glaucus. *Entomologia Experimentalis et Applicata* 50: 29–35.

[214] Lindroth R. L., Scriber J. M. and Hsia M. T. S. (1988) Chemical ecology of the tiger swallowtail: mediation of host use by phenolic glycosides. *Ecology* 69, 814–822.

[215] Wadleigh R. W. and Yu S. J. (1987) Glutathione transferase activity of fall armyworm larvae toward a, b-unsaturated carbonyl allelochemicals and its induction by allelochemicals. *Insect Biochemistry* 17: 759–764.

[216] Wadleigh R. W. and Yu S. J. (1988) Detoxification of isocyanate allelochemicals by glutathione transferase in three lepidopterous species. *Journal of Chemical Ecology* 14: 1279–1288.

[217] Yu S. J. (1989) Purification and characterization of glutathione transferases from five phytophagous Lepidoptera. *Pesticide Biochemistry and Physiology* 35: 97–105.

[218] Yu S. J. and Abo-Elghar G. E. 2000. Allelochemicals as inhibitors of glutathione S-transferase in the fall armyworm. *Pesticide Biochemistry and Physiology* 68: 173–183.

[219] Mound L. A. and Palmer G. M. (1981) Identification, distribution and host plants of the pest species of Scirtothrips (Thysanoptera: Thripidae). *Bulletin of Entomological Research* 71: 467–479.

[220] Saha D., Mukhopadhyay A. and Bahadur M. (2012) Effect of host plants on fitness traits and detoxifying enzymes activity of major sucking insect pest of tea, *Helopeltis theivora* (Heteroptera: Miridae). *Phytoparasitica* 40: 433–444.

[221] Bartlett A. C., Wilson N. M. and Mattix E. B. (1968) The fate of genetic markers in populations of boll weevils. *Journal of Economic Entomology* 61: 808–812.

[222] Bartlett A. C. and Butler G. D. Jr. (1975) Genetic control of the cabbage looper by a recessive lethal mutation. *Journal of Economic Entomology* 68: 331–335.

[223] Loxdale H. D. and Lushai G. (1998) Molecular Markers in Entomology. *Bulletin of Entomological Research* 88: 577–600.

[224] Loxdale H. D., Brookes C. P. and De Barro, P. J. (1996) Application of novel molecular markers (DNA) in agricultural entomology. In The Ecology of Agricultural Pests (edited by W. O. C. Symondson and J. E. Liddell). Chapman and Hall, London.

[225] Daly J. C. (1993) Ecology and genetics of insecticide resistance in *Helicoverpa armigera:* interactions between selection and gene flow. *Genetica* 90: 217–226.

[226] Roush R. T. and Daly J. C. (1990) The role of population genetics in resistance research and management, pp. 97–152. In Pesticide Resistance in Arthropods (edited by R. T. Roush and B. E. Tabashnik). Chapman & Hall, New York.

[227] Labbe P., Lenormand T. and Raymond M. (2005) On the world wide spread of an insect resistance gene: a role for local selection. *Journal of Evolutionary Biology* 18: 1471–1484.

[228] Denholm I., Sawicki R. M. and Farnham A. W. (1985) Factors affecting the resistance to insecticides in houseflies, *Musca domestica.* IV: the population biology of the flies on animal farms in south-eastern England and its implications for the management of resistance. *Bulletin of Entomological Research* 75: 143–158.

[229] Lenormand T. (2002) Gene flow and the limits to natural selection. Trends in Ecology and Evolution 17: 183–189.

[230] Whitehead A., Anderson S. L., Kuivila K. M., Roach J. L. and May B. (2003) Genetic variation among interconnected populations of Catostomus occidentalis: implications for distinguishing impacts of contaminants from biogeographical structuring. Molecular Ecology 12: 2817–2833.

[231] Brogdon W. G. and McAlliester J. C. (1998) Insecticide resistance and vector control. Emerging Infectious Diseases 4: 605–613.

IntechOpen

IntechOpen