トルコにおけるアカイエカ（双翅目：カ科）の殺虫剤抵抗性

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Original Article

Insecticide resistance of *Culex pipiens* (Diptera: Culicidae) in Turkey

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Insecticides are the primary means of controlling mosquitoes in Turkey, especially *Culex pipiens* and *Anopheles maculipennis* complex species. To better understand how the resistance status affects the success of vector control programs, we investigated the level of resistance of strains collected from seven localities to 4 larvicides (temephos, fenthion, *Bacillus thuringiensis israilensis* (*Bti*), and *Bacillus sphaericus* (*Bs*)) and 4 adulticides (DDT, malathion, deltamethrin, and permethrin) that are registered (except DDT) and are used heavily in mosquito control programs in Turkey. High levels of resistance were found to temephos in the Birecik, Viransehir, Mersin, Ankara, and Antalya strains, and we found resistance to fenthion in the Birecik, Mersin, Cankiri, and Antalya strains. Resistance levels to other larvicides (*Bti* and *Bs*) and strains (to temephos and fenthion) were less than 10-fold. We determined low mortality rates with the diagnostic dosage of DDT in all strains. Birecik and Viransehir mortality rates were lower than 30%. The diagnostic dosage mortality rates of the other adulticides varied widely (65.8% malathion Antalya, and 97.5% malathion Birecik, and deltamethrin Hatay) but Ankara and Antalya strains appeared to be resistant to all adulticides tested. The other strains were placed in the surveillance category according to the WHO categorization. Diagnostic dosage experiment results revealed that DDT resistance exists in all populations, although this insecticide has not been used for 30 years in Turkey. Our results indicated that *Bti* and *Bs* are very effective control agents for larval control while permethrin and deltamethrin are effective for adult control. © Pesticide Science Society of Japan

Keywords: *Culex pipiens*, insecticide resistance, *Bti*, *Bs*, temephos, fenthion, DDT, permethrin, deltamethrin, malathion.

Introduction

Culex pipiens is an important vector of several human pathogens, such as West Nile virus, Rift Valley Fever virus, and *Bancroftian flariasis*.[12,15,16] This mosquito species is also widespread in the Holarctic region and is considered important in mosquito control programs due to it being a nuisance species among other health-related problems. Although *Cx. pipiens* does not transmit diseases in Turkey, it is the most common mosquito species in urban and rural areas of Turkey. Mosquito control in Turkey is mainly directed against larval stages and five organophosphates, one pyrethroid, three insect growth regulators (IGRs), two biological control agents, and one surface tension agent are currently registered for use against mosquito larvae. Many pyrethroids and organophosphates are currently registered for adult control.[2] These control agents are used in different areas in Turkey and municipal control strategies rely heavily on temephos and *Bti* products. In addition, DDT and malathion were used in mosquito control studies and malaria eradication programs in Turkey during the 1970s. DDT usage was banned after 1980 and malathion usage stopped in the early 1990s. Generally, adult control depends upon active ingredients such as cypermethrin, deltamethrin, and permethrin used in different synergistic combinations. Although these products have been used for many years and numerous field trials and efficiency tests have been conducted, no study dealing with the development of resistance to these insecticides has been undertaken in Turkey.

The emergence of resistance to insecticides is considered one of the main obstacles to effective vector control measures and the prevention of emerging diseases. Resistance development has been reported to chemical larvicides by many authors,[7–9] but these larvicides are still strongly advocated.[21–26] Control of mosquito-borne disease outbreaks relies heavily on the use of insecticides;[3] however, the development of resist-

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Table 1. Description and GPS coordinates of collected populations

Village	City	GPS coordinates
Kundu	Antalya	36 52′N 30 56′E
Akyurt	Ankara	40 08′N 33 04′E
Hacılar	Çankırı	40 20′N 33 52′E
Erdemli	Mersin	36 37′N 34 19′E
Merkez	Hatay	36 11′N 36 08′E
Adacık-Mezra	Birecik	36 56′N 38 00′E
Persiverek	Viranşehir	37 21′N 39 34′E

ance to insecticides can significantly limit the continued application of these control agents.[10] In Turkey, it is necessary to assess the current resistance status of *Cx. pipiens* for effective control programs. The objective of this study was to assess the resistance status and susceptibility of *Cx. pipiens* populations from seven cities in Turkey to four different larvicides and four adulticides.

Materials and Methods

1. *Strains of Culex pipiens*

The study site GPS coordinates and a brief description of the locations are provided in Table 1. Egg rafts, larvae, and adult samples of *Cx. pipiens* were collected from seven different localities and transferred to the laboratory. Sampling site insecticide usage in the last five years is listed in Table 2. Adulticides were also used in different synergistic combinations in all areas.

2. *Laboratory colonization*

Colonies were reared at 26±2°C, 70±5% relative humidity, and 14 : 10 h light : dark conditions. Larvae were fed Tetramin® fish food. Adult females were fed blood from live chickens 2 times per week. Adults had access to 10% sucrose solution from a cotton wick. Containers filled with deionized water were placed inside the cages for female oviposition. Egg cages were transferred to new containers and larvae were reared as described above after hatching. Since we were unable to obtain a susceptible *Cx. pipiens* laboratory strain, we used the *Cx. quinquefasciatus* (sibling species of *Cx. pipiens*) Say laboratory strain, which is known to be susceptible and which has been used in many comparative studies. The *Cx. quinquefasciatus* Say susceptible strain was obtained from IRD-LIN (Montpellier, France) in 2006 and colonized in the laboratory as described above.

3. *Insecticides*

Four different larvicides and four different adulticides were tested. Each larvicide was diluted five times with deionized water. Three larvicides (*Bti* (Vectobac® 12 AS), *Bs* (Vectolex® WDG), and fenthion (Baytex®)) were obtained from the Turkish branch of Bayer Crop Science, and one larvicide (temephos (Abate® 500 EC)) was obtained from KUYAB (Kundu Tourism Investors Association). DDT, permethrin, deltamethrin, and malathion-impregnated papers were obtained from the WHO.

4. *Bioassays*

The susceptibilities of mosquito strains to 4 different larvicides were determined by a bioassay using the methods recommended by the WHO.[22] Bioassays were undertaken by placing groups of 25 early 4th instar larvae in 200 ml deionized water in 250 ml plastic cups with five different concentrations, as recommended on the label. Each bioassay was conducted using three replicates and the tests were replicated on three different days. Mortality was recorded after 24 hr exposure to *Bti*, temephos, and fenthion, and 48 hr exposure to

Table 2. *Culex pipiens* strains used to survey insecticide resistance and the use of insecticides and larval control agents in the last five years

	Insecticide	Birecik	Viransehir	Hatay	Mersin	Cankiri	Ankara	Antalya
Larvicides	Temephos	X	X	X	X		X	X
	Bti	X	X	X	X		X	X
	Diflobenzuron						X	X
	Aqnique						X	X
Adulticides	Cypermethrin	X	X	X	X	X		X
	Alfa Cypermethrin							X
	Cyfluthrin	X	X				X	
	Cyphenothrin						X	
	Deltamethrin	X	X	X		X		
	Permethrin			X	X	X	X	X
	Resmethrin						X	X

Bs since this agent shows larvacidal activity after 48 hours. LC_{50} was determined using probit analysis (EPA probit analysis program Version 1.5). Temephos and *Bti* used in this study were chosen by application frequencies for larvicide tests. Although *Bs* has not been used frequently in many areas, many *Bs* products have recently been registered for larval control in Turkey; therefore, we added this insecticide as a larvicide in this study. Fenthion has been used in larval control programs for a very short time but has been used for agricultural purposes in many areas in Turkey, and is also frequently used in olive production in the Mersin and Birecik regions. We tested fenthion as a larvicide to show if there is an effect of fenthion used in agriculture and to identify its toxicity to mosquito larvae. Pyrethroids were not used in larvicide tests since this group has not been used as a larval control agent in Turkey.

Adult mosquitoes were assayed for susceptibility by diagnostic tests[21] except for the Cankiri and Mersin strains. Susceptibility was determined to DDT, permethrin, deltamethrin, and malathion. One diagnostic concentration was used for all insecticides and a 1 hr application period was used for permethrin, deltamethrin, and malathion and a 2 hr application period was used for DDT. Mortality was assayed 24 hr after application. Insecticide resistance status was evaluated using the classification determined by WHO[23]: 98–100% mortality indicates susceptibility, 80–97% mortality suggests possible resistance requiring confirmation, and <80% mortality suggests resistance.

RESULTS

1. *Bioassays*

Laboratory bioassays with *Bti* against 4th instar larvae of *Cx. pipiens* showed high mortality for all tested groups after 24 hr exposure. LC_{50} values ranged from 0.061 to 0.706 ITU/m^2. Cankiri and Antalya strains showed high sensitivity to *Bti* and the LC_{50} value of the Antalya strain was nearly four-fold lower than the control strain. LC_{50} values of Hatay and Viransehir strains were nearly three-fold higher than the control group (Table 3). LC_{50} values of all other strains were similar to those of the control group.

Tests with *Bs* yielded similar results to *Bti* but the toxicity of *Bs* seemed to be higher than *Bti* by RR_{50}. LC_{50} values ranged from 32.11 to 767.83 ITU/m^2. Cankiri (32.11) and Antalya (53.52) strains showed very high sensitivity to *Bs*. The LC_{50} value for the Cankiri strain was nearly eight-fold lower

Table 3. Toxicity (LC_{50}) and resistance ratio (RR_{50}) of four larvicides against *Cx. pipiens* from seven different sampling areas

Strains		*Bti* (ITU/m^2)	*Bs* (ITU/m^2)	Temephos (ml/m^2)	Fenthion (ml/m^2)
Birecik	LC_{50}	0.397 (0.111–1.434)	264.56 (200.75–348.50)	0.11 (0.091–0.133)	1.024
	Slope±SE	2.07±0.35	2.19±0.26	2.17±0.18	2.34±0.26
	RR_{50}	1.69	1.14	27.5	17.35
Viransehir	LC_{50}	0.688 (0.508–0.928)	767.83 (535.85–1088.68)	0.105 (0.077–0.143)	0.477 (0.332–0.687)
	Slope±SE	2.36±0.33	1.72±0.22	2.18±0.29	1.63±0.20
	RR_{50}	2.94	3.33	26.25	8.08
Hatay	LC_{50}	0.706 (0.353–1.409)	490.52 (365.69–652.09)	0.034 (0.028–0.040)	0.28 (0.229–0.343)
	Slope±SE	1.72±0.23	2.05±0.25	2.70±0.24	1.99±0.20
	RR_{50}	3.01	2.12	8.5	4.74
Mersin	LC_{50}	0.262 (0.153–0.451)	269.44 (214.74–337.96)	0.252 (0.211–0.300)	4.19 (3.41–5.26)
	Slope±SE	1.84±0.21	2.12±0.20	2.02±0.14	2.40±0.28
	RR_{50}	1.11	1.16	63	71.01
Cankiri	LC_{50}	0.224 (0.177–0.290)	32.11 (24.03–42.87)	0.008 (0.006–0.011)	0.625 (0.502–0.779)
	Slope±SE	3.69±0.64	2.56±0.36	2.32±0.31	2.12±0.19
	RR_{50}	0.95	0.13	2	10.59
Ankara	LC_{50}	0.333 (0.145–0.736)	314.78 (268.82–368.57)	0.067 (0.024–0.188)	0.867 (0.733–1.027)
	Slope±SE	1.59±0.23	2.14±0.14	2.01±0.40	1.91±0.12
	RR_{50}	1.42	1.36	16.75	14.69
Antalya	LC_{50}	0.061 (0.034–0.105)	53.52 (9.79–299.62)	0.075 (0.023–0.245)	0.529 (0.454–0.618)
	Slope±SE	1.55±0.19	1.65±0.29	2.06±0.45	2.26±0.15
	RR_{50}	0.26	0.23	18.75	8.96
Cx. quinquefasciatus	LC_{50}	0.234 (0.160–0.340)	230.35 (185.25–287.54)	0.004 (0.003–0.004)	0.059 (0.045–0.076)
	Slope±SE	1.17±0.11	3.56±0.47	3.11±0.44	2.48±0.31
	RR_{50}	1	1	1	1

Table 4. Percentage mortality of *Cx. pipiens* adults from five different sampling areas exposed to the diagnostic doses of DDT, permethrin, deltamethrin and malathion

Strains		DDT 4%	Permethrin 0.75%	Deltamethrin 0.05%	Malathion 5%
Birecik	No. Dead	30	114	115	117
	Mortality (%)	25	95	95.8	97.5
Viranşehir	No. Dead	31	102	114	107
	Mortality (%)	25.8	85	95	89.1
Hatay	No. Dead	72	110	117	106
	Mortality (%)	60	91.6	97.5	88.3
Ankara	No. Dead	73	92	96	79
	Mortality (%)	60.8	76.6	80	65.8
Antalya	No. Dead	70	94	91	81
	Mortality (%)	58.3	78.3	75.8	67.5

All experiments were performed using 8 replicates and each replicate comprised 15 female mosquitoes

than the control group and the Antalya strain showed five-fold lower values than the control group. Results for the Viransehir and Hatay strains were similar to that of *Bti*. All populations were RR_{50} lower than *Bti* except Mersin strain but Mersin strain RR_{50} was nearly identical for *Bti* and *Bs*.

All strains showed varying degrees of resistance to temephos and fenthion when compared to the control group. Median lethal concentrations of temephos ranged from 0.008 (Cankiri) to 0.252 (Mersin) ml/m^2. The Cankiri strain showed high sensitivity to temephos but two-fold resistance when compared to the control group. The Mersin strain showed low sensitivity to temephos (0.252 ml/m^2) compared with the other groups and 63-fold resistance. The other strains' resistance ratios ranged from 8.5 (Hatay) to 27.5 (Birecik). Fenthion was registered for larval control and used for a short time period but was later banned by the Ministry of Health. Although fenthion is not commonly used for mosquito control in Turkey, all strains showed resistance to this insecticide. Fenthion LC_{50} values ranged from 0.28 (Hatay) to 4.19 ml/m^2 (Mersin). The strains showed resistance spectra ranging from 4.73-fold (Hatay) to 71.01-fold (Mersin) (Table 3). The results were similar to temephos but resistance ratios were lower for all strains except Cankiri and Mersin. Resistance levels in the Cankiri strain were low for all insecticides; however, *Bti* and *Bs* are frequently used in control programs in Ankara and Antalya; hence, these strains showed high levels of sensitivity to these control agents.

According to the diagnostic tests, all strains were resistant to DDT (mortality rates under 80%) and showed low mortality with this insecticide. Birecik and Viransehir strains showed very low mortality with this insecticide (25% and 25.8%, respectively) (Table 4). Hatay, Birecik and Viransehir strains were placed in the resistance surveillance category for permethrin, deltamethrin, and malathion according to the WHO (mortality rates between 80% and 97%). Ankara and Antalya strains were determined to be in the resistant category for all insecticides tested.

Discussion

Temephos and *Bti* products are mostly used as larvicides around the world.[16,30,31] As in most other countries, these larvicides have been used in many areas in Turkey. Other larvicides, such as fenthion, fenitrothion, and malathion, have not been used frequently in mosquito control programs in Turkey in the last decade. The IGR product (Diflobenzuron) and surface films (Monomolecular film) are also used for larvicidal applications, but not frequently. Adult control programs depend heavily on pyrethroid and organophosphate insecticides. The most frequently used control products include cypermethrin, cyphenothrin, deltamethrin, permethrin, and pyrethroid+synergist combinations.[2]

This study revealed that the larvae of all strains were highly sensitive to *Bti*. LC_{50} values in Antalya and Cankiri strains were lower than the control group. Although *Bti* products have been used only sporadically in vector control applications, they were still effective for controlling *Cx. pipiens* in all tested areas; however, some strains showed 2–3-fold resistance to *Bti*, which was expected, since the toxicity of biological larvicides can vary among laboratories[8–17] and strains. All populations showed high sensitivity to *Bti*, but this result should be treated with caution. Although there was 3-fold resistance to *Bti* in Birecik and Viransehir, *Bti* applications in these areas are routinely conducted once or twice a year. *Bti* briquettes have also been used for larval control in these areas in the last two or three years. These results also revealed that Birecik and Viransehir strains do not mix rapidly, leading to localized resistant populations. Although Paul *et al.*[16] reported 33-fold resistance to *Bti* in the Syracuse *Cx. pipiens* strain, Wirth *et al.*[26] reported 10-fold susceptibility in the Cyprus *Cx. pipiens* strain.

Bs bioassay results were similar to those for *Bti*. LC_{50} values for Antalya and Cankiri strains were lower than the control group and values were higher than the control group in Birecik, Hatay, Viransehir, and Ankara strains. *Bs* products are not commonly used in Turkey and this agent was only used for a short period in Ankara and Antalya. These results may be explained by the low or non-usage of *Bs* products in all areas; however, the results of Viransehir and Hatay strains cannot be fully explained by the usage profile. These results are similar to those reported previously for *Bs*. Paul *et al.*[16] showed a low level of *Bs* resistance in the Albany strain, which had not come in contact with *Bs* previously. Although *Bs* showed higher toxicity in the five test areas, this microbial insecticide showed a toxic effect after 48 hr. If urgent control operations are needed in these study areas, the late toxicity effect of this larvicide may be problematic.

Temephos is an effective control agent and is used in most countries around the world;[30] however, the WHO[24] recommends treating drinking water with temephos at dosages not exceeding 1 mg active ingredient per liter (1 ppm). Temephos is primarily a larval control agent in Turkey, as in most other countries. Many studies have shown low or high levels of resistance to this insecticide. Bonning *et al.*[6] showed resistance to temephos in 1982 in Lucca, Italy. Resistance to organophosphate compounds had increased and there was also evidence of resistance to propoxur and bendiocarp in the same area in 1984. Bisset *et al.*[5] showed different degrees of resistance to three larvicides (malathion, propoxur, and temephos) in three different areas. Our results showed that five strains had high degrees of resistance and the larval habitats of three strains were near drinking water pits. The temephos usage profile and frequency in all areas need to be reevaluated according to our results and WHO recommendations. Fenthion is not commonly used as a larval control agent in Turkey but is frequently used for agricultural purposes. Olive farmers in particular apply this insecticide for pest management. Three strains (Viransehir, Hatay, and Antalya) showed moderate resistance while the other four strains showed high resistance to this insecticide. These results cannot be fully explained by the insecticide's application profile. Mersin and Birecik strains gave very unusual results when compared with other results. These areas are two of the most important olive agricultural plantation areas in Turkey outside the Aegean region. The high resistance is possibly related to high agricultural insecticide application. Indeed, the development of resistance in *An. sacharovi* in Adana was a consequence of agricultural pesticide use.[13] Akiner *et al.*[1] also indicated that, in their two study areas, the high resistance of *Musca domestica* might be related to the agricultural insecticide usage profile. Resistance ratios of other strains showed that fenthion is potentially used for larval control in some areas; however, Sun[19] reported 24.8-fold resistance in *Cx. pipiens* to this insecticide after continuous application for 15 generations. Our results also showed that resistance ranged from 4.74 (Hatay) to 71.01 (Mersin) and attention needs to be given to the usage of this insecticide in all areas.

All *Cx. pipiens* strains showed high or moderate resistance to chemical larvicides but were very sensitive to biological larvicides. The results revealed that these mosquito control agents are still very effective in Turkey when compared to other countries. Zayed *et al.*[28] showed different degrees of fenitrothion and temephos resistance in *Cx. pipiens* in Egypt. Zayed *et al.*[29] also reported that malathion, temephos and chlorpyrifos are very effective whereas strong resistance has arisen toward fenitrothion, bromophos, and fenthion in Egypt. Paul *et al.*[16] reported different degrees of resistance to *Bs* and *Bti* in Syracuse strains and Wirt *et al.*[26] showed up to 10-fold variability in *Bti* susceptibility in *Cx. pipiens* from Cyprus.

In Turkey, carbamates and pyrethroid insecticides are used in huge proportions in agriculture but mostly aerosol formulations of pyrethroids are used for mosquito control by private bodies and municipalities. Kasap *et al.*[13] described this situation and different resistance spectra for different *An. sacharovi* populations in Turkey. They showed that the kdr-type gene is responsible for pyrethroid resistance in *An. sacharovi*. All strains were found to be resistant to DDT. Birecik, Hatay, and Viransehir strains were placed in the surveillance category for the pyrethroid group. Ankara and Antalya strains were resistance to DDT, pyrethroids, and malathion, but DDT mortality rates were lower than in other strains.

Pyrethroid resistance is explained by two major mechanisms: increased detoxification and changes in target site insensitivity. Many studies have reported pyrethroid cross resistance resulting from field selection with DDT and implicating a knock-down mechanism,[11] although some reports of larval DDT resistance in *Ae. aegypti* were linked to increased metabolic levels.[14] Generally metabolic detoxification is associated with changes in monooxigenase activity[4] but elevated glutathione S transferases (GSTs) levels and nonspecific esterases have also been reported to confer pyrethroid resistance.[18–20] The permethrin resistance of Antalya and Ankara strains may be explained by the usage profile, but this did not occur for deltamethrin in the same areas. This may explain the stronger cross resistance than the insecticide usage profile. Birecik and Viransehir strains showed high resistance to DDT but did not show resistance to permethrin or deltamethrin. This makes it difficult to describe the overall results in these areas; therefore, a more detailed study is required to understand (biochemically and molecularly) the overall situation.

Similar to other countries, insecticide resistance causes severe problems for vector control programs as well as resulting in financial losses and environmental contamination. In this study, we recorded moderate to high levels of resistance to all tested chemical larvicides while resistance levels to biological larvicides were low or zero. In light of this result, it is suggested that the application of biological insecticides in circulation with chemical insecticides would be the most effective strategy for vector control programs in Turkey. When the re-

sults are evaluated for adult control programs, resistance to DDT is still high despite the lack of use of this insecticide for over 30 years. This result also puts all pyrethroid-based insecticides at risk. Insecticide resistance to the discriminating dosage of the three insecticides tested in this study was high in both Ankara and Antalya regions, where management programs are continually implemented. The remaining regions can be classified as regions where monitoring programs should be set up in accordance with WHO standards. More detailed studies are needed to establish new and more effective eco-friendly insecticide usage profiles that will meet both health and agricultural concerns. Monitoring programs to effectively monitor frequency shifts of resistant individuals in a population due to various factors should be set up in all study regions. As a first step, susceptibility and effectiveness tests should be conducted. This study is important as it points out that the genetic mechanisms causing resistance to DDT are still active, as shown by the high level of resistance to this insecticide. Although this study sheds important light on the resistance level and status of several important insecticide classes, more detailed biochemical and genetic analyses are needed.

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英文編掲載報文・短報等の要旨

解　説

無脊椎動物のニコチン性アセチルコリン受容体－農学，獣医学そして人間健康科学において重要な化学製品と医薬品のターゲット

David B. Sattelle

幾つかの線虫や昆虫ゲノムの解析につれて，遺伝子ファミリーの分子生物学的並びに機能的多様性およびそれらの機能的役割が加速的に理解されるようになってきた．ニコチン性アセチルコリン受容体（nAChR）遺伝子ファミリーは，高速なシナプス伝達を介在し，殺虫剤として，また人体の抗寄生虫薬のターゲットとして注目されてきた．遺伝子モデル生物の自由生活線虫 *Caenorhabditis elegans*，そして寄生線虫の *Brugia malayii* のゲノムはすでに解読された．ショウジョウバエ *Drosophila melanogaster* に続き，マラリア媒介種である *Anopheles gambiae*，貯蔵農産物害虫である *Tribolium castaneum*，農業生産上極めて有益な蜜蜂 *Apis mellifera*，その昆虫など，多くの生物のゲノムが入手できるようになってきた．いくつかの線虫 nAChR ファミリーは，多くのサブユニットアイソフォームで知られている大 nAChR ファミリーに属しているが，昆虫の nAChR ファミリーはよりコンパクトである．しかし，昆虫の nAChR ファミリーにおいても，選択的スプライシングや RNA エディティングによって同程度の分子生物学的多様性が確認されている．L タイプ線虫レバミソール感受性 nAChR および N タイプニコチン感受性 nAChR を構成するサブユニットはすでに解析され，機能的に発現された．また，イミダクロプリド感受性 nAChR も昆虫の α サブユニットを含むハイブリッド受容体を用いて発現された．これらの研究は，受容体ターゲットに基づいた新規動物用健康薬品や農業用薬剤のスクリーニングの有望性を提示している．

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報　文

鉄ポルフィリン触媒によるペルメトリン，フェンバレレートの酸化

福島雅雄，片木敏行

環境中での農薬の微量代謝分解物の化学構造を明らかにするため，効率良く代謝分解物を収集する様々な手段が講じられているが，本研究ではピレスロイド系殺虫剤の鉄ポルフィリンを触媒とする酸化手法への適用性を調べた．シクロプロピル基の 1 位を ^{14}C で標識したトランスペルメトリン，およびクロロフェニル基あるいはフェノキシフェニル基を ^{14}C で標識したフェンバレレートとそれらの部分構造であるジクロロビニル菊酸，フェノキシベンジルアルコールについてポルフィリン環の β 位，メソ位の置換基あるいは軸配位子が異なる 6 種の鉄ポルフィリンを触媒とし，過酸化水素およびメタクロロ過安息香酸を酸化剤とする酸化反応を行った．エーテルおよびエステル結合の開裂や芳香環の水酸化などの反応が認められ，一部は動植物や昆虫における代謝物と一致した．ポルフィリン環の β 位やメソ位に電子吸引基を導入すると反応率が上昇し，軸配位子が弱い電子供与基の場合には酸化反応性が上昇することが示唆された．

病原糸状菌 *Sclerotium rolfsii* ITCC 5226 および炭腐病原菌 *Macrophomina phaseolina* ITCC 0482 に対するニームオイル，ニコチン酸およびオオウイキョウの α,β-不飽和カルボニル化合物との混合製剤による相乗的カビ防止効果

Archna Rani, Sapna Jain, Prem Dureja

食中毒法 Food Poisoning Method を用いて，様々な濃度のニームオイル（インドセンダン *Azadirachta indica*）（N1），ニコチン酸（N2），そしてオオウイキョウ（N3）を，α,β-不飽和カルボニル化合物（1a–1i）と組み合わせることによって，インビトロで計 90 製剤の病原性糸状菌 *Sclerotium rolfsii* ITCC 5226 株および炭腐病原菌 *Macrophomina phaseolina* ITCC 0482 株に対するカビ防止効果を検討した．*S. rolfsii* に対して，製剤 3 : 1, 1a−N2（−）が最も高い効果を示し，66 ppm で 1a の活性を 2.7 倍増大させた．一方，製剤 3 : 1, 1f−N3 は，1f 単独の場合に比べ，*M. phaseolina* に対する活性が 5 倍も増えた．他の多くの製剤も，かび防止活性の増加に著しい相乗効果を示した．本研究は，合成製品と天然製品を組合せることによって，病原性真菌を制御する新規な手法の可能性を示唆した．

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トルコにおけるアカイエカ（双翅目：カ科）の殺虫剤抵抗性

M. M. Akiner, F. M. Simsek, S. S. Caglar

トルコにおいて，殺虫剤の使用が，蚊を，特にアカイエカ（*Culex pipiens*）とハマダラカ（*Anopheles maculipennis*）群を，駆除する主な方法となっている．殺虫剤抵抗性がど

のように蚊の防除プログラムの正否に影響するかをよりよく理解するため，4種類の殺幼虫剤（テメホス，フェンチオン，*Bacillus thuringiensis israilensis*（*Bti*）および*Bacillus sphaericus*（*Bs*））と4種類の殺成虫剤（DDT，マラチオン，デルタメトリンおよびペルメトリン）に対する7ケ所から採集された蚊種の殺虫剤抵抗性を調べた．殺成虫剤は，DDTを除き登録されており，いずれもトルコの蚊防除プログラムで非常によく使われている．Birecik種，Viransehir種，Mersin種，Ankara種およびAntalya種に，高いテメホス抵抗性が見つかった．また，Birecik種，Mersin種，Cankiri種およびAntalya種にフェンチオン抵抗性が見つかった．ほかの殺幼虫剤（*Bti*と*Bs*）に対する抵抗性またはテメホスとフェンチオンに対する抵抗性種の抵抗性は10倍以下であった．すべての種において，DDTの診断用量に対する死亡率が低かった．Birecik種とViransehir種の死亡率は30%以下であった．その他の殺成虫剤の診断用量死亡率は大きく変動（マラチオンはAntalya種で65.8%，Birecik種で97.5%；デルタメトリンはHatay種で97.5%）したが，Ankara種とAntalya種は使用したすべての殺成虫剤に対して抵抗性を示した．その他の種はWHOの分類で要監視種目に分類されている．診断用量実験の結果では，トルコで過去30年間DDTは使用されていなかったにも関わらず，すべての群種にDDT抵抗性が存在することを明らかにした．我々の結果から，*Bti*と*Bs*が幼虫の駆除に有効であること，ペルメトリンとデルタメトリンは成虫の駆除に有効であることが示された．

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LC/TOF-MSを用いた農産物中の酸性農薬等の多成分スクリーニング分析法

秋山由美，松岡智郁，三橋隆夫

LC/TOF-MSを用いて，農産物中に残留する酸性農薬等95種を迅速にスクリーニング分析する方法を開発した．酸性農薬等は，GC/MS分析において妨害となる脂肪酸を除去するために用いられるPSAに吸着するため，多成分一斉分析法の対象に加えることができなかった．そこで，多成分分析法において，PSA精製前の抽出液の一部を分取し，溶媒置換したものをLC/TOF-MSで測定した．精密質量を測定することにより，分子イオンおよびフラグメントイオンを共存成分から分離し，選択的に検出することができた．6種類の農産物について，添加濃度0.1 μg/gで回収実験を行った結果，平均回収率は49~127%，RSDは20%未満であり，定量限界値は0.01~0.02 μg/gであった．農産物140検体のモニタリング検査に本法を適用したところ，2,4-Dがレモンおよびオレンジから，フルアジホップが未成熟いんげんから，ジクロルプロップがりんごから，それぞれ0.02~0.03 μg/gで検出されたが，いずれも基準値未満であった．本分析法は，農薬520種を対象とした既存の多成分一斉分析法と組み合わせて，酸性農薬等95種に対しても，高感度かつ迅速なスクリーニング分析を可能にした．

光異性化および代謝分解を考慮した水田における農薬挙動予測モデル（PADDY）の開発：除草剤ピリミノバックメチルへの適用

稲生圭哉，水谷浩之，與語靖洋，池田光政

水稲用除草剤ピリミノバックメチルを対象とし，水田における光異性化および代謝分解を考慮した挙動予測が行えるようにPADDYモデルの改良を行った．田面水中におけるピリミノバックメチル*E*体と*Z*体間の異性化は，紫外線（B領域）により進行する可逆的反応として表現した．また，土壌中における主要な代謝分解物の生成・消失を逐次の一次反応として表現した．改良したモデルの検証を行うため，水田ライシメータや水田圃場におけるピリミノバックメチルおよび主要な代謝分解物の消長を実測した．改良型PADDYモデルは，*E*体と*Z*体間の光異性化反応を考慮することにより，田面水および土壌中におけるピリミノバックメチルの挙動を精度よく予測することができた．また，主要な代謝分解物についても予測できることが示された．

短報

殺虫剤毒性の評価における新規高速大量スクリーニング法（HTS法）とボトルアッセイ法の比較

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殺虫剤の毒性は，一般的にWHOテスト法やボトルアッセイ法で評価されている．最近，高速大量スクリーニング法（HTS法）が開発され，蚊の挙動や死滅に対する殺虫剤効果の評価に使われるようになってきた．我々は，HTS法とボトルアッセイ法を比較して，ネッタイシマカ*Aedes aegypti*に対する殺虫剤の毒性を評価した．二つの方法は等価であることが分かった．異なった主な点は：1）HTS法よりボトルアッセイ法において蚊の反応時間は短かった；2）蚊のノックダウンと死滅はより低い用量で発生した．これらの結果は，各種媒介生物群の挙動反応に対する様々な化学物質の効果評価手法の指針となるであろう．

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