Cross-resistance and biochemical mechanisms of pyridaben resistance to *Tetranychus urticae* Koch

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Experimental Goals

- Regional variations of susceptibility to pyridaben
- Cross-resistance patterns of pyridaben-resistant *T. urticae*
- Resistance mechanisms to pyridaben

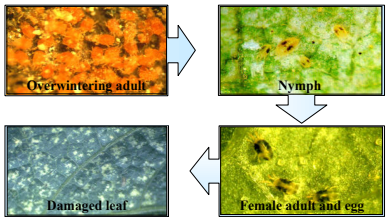
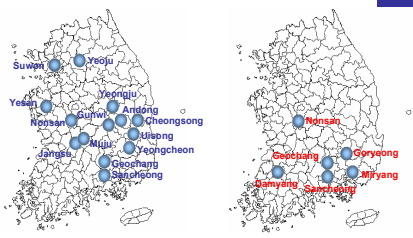


Fig. Life cycle of two spotted spider mite

Exp. 1. Resistant monitoring of *T. urticae* to pyridaben



< Apple orchard > < Strawberry >

Fig. Collecting sites of *T. urticae* on the apple and strawberry

Exp. 1. Resistant monitoring of *T. urticae* to pyridaben

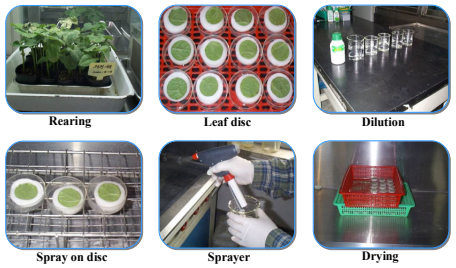


Fig. Bioassay by spray method

Exp. 2. Selection and cross-resistance

- Selection of *T. urticae* by pyridaben
  : A strain of pyridaben resistant *T. urticae* collected from apple orchard in 2002 was further selected with pyridaben 20% WP for 27 generations (PR-27 strain).
- Cross-resistance
  : cross-resistance levels of the PR-27 strain to 16 acaricides were determined by spray method.

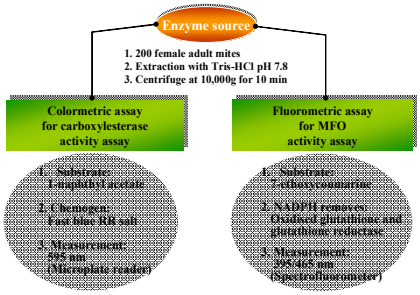
Exp. 3. Mechanism of resistance to pyridaben

1. Synergism tests

- PBO, IBP, TPP, and DMC were used to inhibit detoxification mechanisms by mixed-funtion oxidases(MFO), esterase(EST), DDT dehydrochlorinase
- Synergists were dissolved in acetone and sprayed on to female-infested kidney bean leaf disk 1 hour prior to the acaricide application
- Synergist solutions were prepared at the concentration(ppm)
  : PBO(125), IBP(100), TPP(62.3) for the SUS. Strain
  : PBO(1000), IBP(1000), TPP(100), DMC(1000) for the PR-27 Strain

Exp. 3. Mechanism of resistance to pyridaben

2. Biochemical enzymatic assay



Results

Exp.1. Resistant monitoring of *T. urticae* to pyridaben

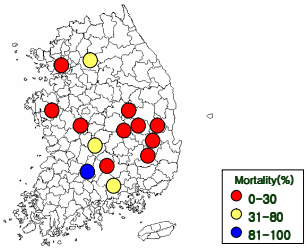


Fig. 1. Susceptibility of *T. urticae* in apple orchard to pyridaben

Exp.1. Resistant monitoring of *T. urticae* to pyridaben

Table. Susceptibilities of *T. urticae* on strawberry

Acaricides	LC_{50}(ppm)						
	GC	KR	MY	NS	DY	SC	SUS
Pyridaben WP	>5000 (300)[a]	3800 (230)	7300 (440)	3600 (220)	>5000 (300)	3800 (240)	17 (1)
FenpyroximateSC	520 (83)	2000 (320)	300 (5)	8.4 (1.3)	470 (74)	34 (5)	6.3 (1)
Fenazaquin EC	1400 (400)	53 (15)	120 (35)	89 (25)	410 (120)	44 (12)	3.5 (1)
TebufenpyradWP	120 (43)	17 (6)	5.0 (2)	14 (5)	68 (25)	9.3 (3)	2.7 (1)

[a] RR : LC_{50} of field strain / LC_{50} of susceptible strain

Exp. 2. Selection and cross-resistance

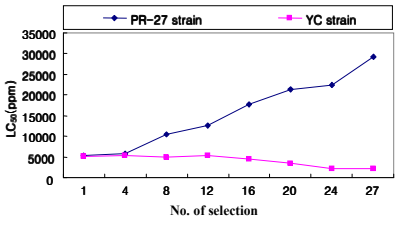


Fig. Resistance development by selection with pyridaben for 27 times (RR of PR-27 strain was 200-fold compared with susceptible stain)

Exp. 2. Selection and cross-resistance

Table. Cross-resistance of pyridaben resistance strain to various acaricides

Acaricide	PR-27		YC		RR
	LC_{50}(ppm)	Slope±SE	LC_{50}(ppm)	Slope ±SE	
Monocrotophos	147.0	2.5±0.3	86.0	3.2±0.2	1.7
Pyraclofos	14.0	2.1±0.2	26.0	2.6±0.3	0.5
Bifenthrin	3.5	0.9±0.8	11.0	2.9±0.3	0.3
Fenpropathrin	8.6	1.5±0.2	5.3	1.4±0.3	1.6
Azocyclotin	900	2.7±1.7	590	1.9±0.2	1.5
Abamectin	0.2	3.7±0.9	0.2	1.7±0.2	1
Acequinocyl	13.0	0.8±0.2	14.0	1.2±1.6	0.9
Bifenazate	2.1	1.7±1.6	3.5	1.8±0.2	0.6
Chlorfenapyr	4.5	2.7±0.2	2.6	3.6±0.4	1.7
Fenbutatin oxide	12	1.8±0.2	12	2.0±0.3	1
Pyraclofos	14	2.1±0.2	16	2.6±0.3	0.5
Fenazaquin	>2000	-	300	2.4±0.9	>7.8
Fenpyroximate	>1600	-	130	1.2±0.3	>12
Tebufenpyrad	>1600	-	180	1.0±0.1	>8.7
Chlorfenapyr.Pyridaben	12	1.2±1.5	19	2.8±0.3	0.6
Pyridaben	>26000	-	2100	1.0±0.8	>13

* RR : LC_{50} of PR-27 / LC_{50} of Yeongcheon strain

Exp. 3. Mechanism of resistance

Table. Toxicity of pyridaben with synergist to susceptible strain

Treatment	LC_{50}				SR
	n	ppm	Slope±SE	95% CL	
Pyridaben only	540	130	1.4±0.1	110-160	1
+PBO (1:1)	560	32	1.3±0.6	21-42	4.2
+IBP (1:1)	630	43	1.9±0.1	28-70	3.1
+TPP (1:1)	450	140	1.0±0.1	110-180	1.0

SR = LC50 values of pridaben without synergist / LC50 values of pyridaben with synergist

Table. Toxicity of pyridaben with synergists to pyridaben resistant strain

Treatment	LC_{50}				SR
	n	ppm	Slope±SE	95%CL	
Pyridaben only	720	4900	1.9±0.2	4100~5800	1
+PBO (1:1)	540	13000	0.6±0.1	7300~42000	0.4
+IBP (1:1)	548	4000	1.0±0.1	3000~5400	1.2
+TPP (1:1)	645	10000	1.7±0.3	7700~13000	0.5
+DMC (1:1)	650	15000	1.3±0.3	11000~30000	0.3

SR = LC_{50} values of pridaben without synergist / LC_{50} values of pyridaben with synergist

Exp. 3. Mechanism of resistance

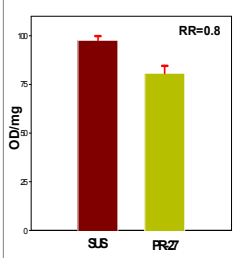


Fig. Carboxylesterase activities in susceptible and PR-27 strains of *T. urticae*

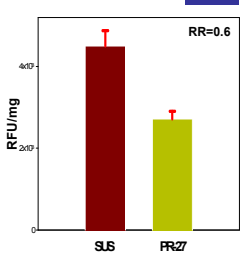


Fig. Fluorometric MFO activities in susceptible and PR-27 strains of *T. urticae*

Summery

1. Field populations of *T. urticae* collected from apple trees and strawberries appeared to be highly resistant to pyridaben
2. The PR-27 strain was extremely resistant to pyridaben (RR: 86).
3. The strain exhibited positive cross-resistance to fenpyroximate (RR >12), fenazaquin (RR >7.8) and tebufenpyrad(RR >8.7). The PR-27 strain showed low level of cross-resistance to fenpropathrin abamectin, monocrotophos, dicofol, chlorfenapyr, azocyclotin and fenbutatin oxide.
4. Synergist experiments with specific enzyme inhibitors revealed that piperonyl butoxide(PBO) had the low effect on the efficacy of pyridaben. Also there is no significant difference in mixed function oxidase(MFO) activities between the S and PR-27 strains(RR 0.6).
5. Iprobenfos(IBP) being used as specific inhibitor of carboxylesterase showed low inhibition to PR-27 mite and esterase activity of resistant strain was lower than that of susceptible one(RR 0.7).