

Pome Fruits—Chemical Control

Porphyric Insecticides

Larry J. Gut

Washington State University Tree Fruit Research and Extension Center, Wenatchee, WA

Keywords: porphyric insecticide, *Trichoplusia ni*

The term porphyric insecticides has been proposed to define a new class of insecticides which act by modulating the porphyrin-heme biosynthetic pathway in insects. When used alone or in combination with δ -aminolevulinic acid (ALA), they induce the accumulation of protoporphyrin IX (Proto) in darkness. Treated insects are killed very rapidly upon exposure to light by the photodynamic action of the accumulated tetrapyrroles. Larval death is associated with loss of body fluids, followed by rapid desiccation. The uncontrolled Proto biosynthesis and accumulation probably cause death of the treated insects via singlet oxygen formation or by inducing the premature release of the highly reactive superoxide (O_2^-) and hydroxy ($\bullet OH$) radicals.

Several analogs of 1,10-Phenanthroline (OPh) and 2,2'-Dipyridyl (DPy) exhibited potent porphyric insecticidal activity against larvae of *Trichoplusia ni*. Evaluation of 18 OPh and DPy based chemicals revealed that death of *T. ni* larvae was positively correlated with Proto accumulation in darkness (Tables 1 and 2). The highly significant enhancement of conversion of exogenous ALA to Proto by OPh, 5-nitro-OPh (5NOPh), 5-chloro-OPh (5ClOPh), 4,7-dimethyl-OPh (47DMOPh), 3,4,7,8-tetramethyl-OPh (3478TMOPh), DPy, and terpyridine (TPy) was accompanied by greater than 90% larval mortality within the first two hours of exposure to light. Some modulators (e.g., 5NOPh, 47DMOPh; DPy) also induced the accumulation of Proto in the absence of exogenous ALA. However, concentrations of Proto under these conditions were at least 4-fold less than concentrations in the presence of exogenous ALA.

An additional 141 N-heterocyclic chemicals were evaluated for porphyric insecticidal activity. Each was a structural analog of one of seven parent molecules or templates (Pyridinium ion, 2-Oxypyridine, Pyrrole, 8-Hydroxyquinoline, Nicotinamide, Nicotinic acid, Picolinic acid). A majority of the chemicals exhibited low levels of insecticidal activity. Mortalities of less than 30% after 3 days exposure to light were recorded for 119 of the 141 treatments. The 24 relatively active modulators were primarily based on the 8-hydroxyquinoline, 2-oxypyridine, pyridinium ion or pyrrole templates.

Evaluation of active and inactive analogs of each template revealed that death of *T. ni* larvae was positively correlated with Proto accumulation in darkness. The less active picolinic acid, nicotinic acid, and nicotinamide based modulators were relatively poor enhancers of Proto accumulation.

In contrast, Proto contents greater than 10 nmoles/100 mg protein were generated by several analogs of the four active templates and were accompanied by mortalities of 50% or greater after 3 days exposure to light (Table 3). Moreover, the highly enhanced conversion of

exogenous ALA to Proto by Isocarbostyryl (ICBS), 1,1-Diethyl-2,4-cyanine iodide (11D24CyI), 8-Hydroxyquinoline-5-sulfonic acid (8OHQUIN5SA), and 8-Hydroxy-7-(4-sulfo-1-naphthylazo)-5-quinoline sulfonic acid (8OHSN5QUINSA) was accompanied by significantly high mortality within the first two hours of exposure to light. The induced accumulation of Proto in the absence of exogenous ALA by 11D24CyI also resulted in significant mortality within a few hours of exposure to light.

The highest levels of mortality after 3 and 6 days exposure to light were recorded for treatment with 1,1-Diethyl-4,4-carbocyanine iodide (11D44CbCyI), alone or in combination with ALA (Table 2). However, 11D44CbCyI did not induce high levels of Proto accumulation and enhancement of Proto accumulation appeared to be less pronounced than was indicated by the high mortality. To determine if the high insecticidal activity was not entirely photodynamically related we evaluated its effectiveness under three light regimes: a) 17-h dark followed by 7-h light for three 24-h periods, b) continuous dark for 72-h, and c) continuous light for 72-h. Levels of Proto accumulation were determined at 17-h and at 24-h. Light appeared to be a requirement for achieving significant kill of *T. ni* larvae. Continuous exposure of larvae to dark was accompanied by low levels of mortality. The highest levels of mortality were recorded for treatment with the modulator alone or in combination with ALA and continuous exposure to light. Recorded levels of Proto content were not closely associated with insecticidal activity. Moderate levels of mortality occurred following treatment with both 11D44CbCyI and ALA + the modulator, but Proto content was much lower in the former treatment. Moreover, proto accumulations after 17-h and 24-h exposure to light were very low.

Table 1. Effects of ALA and substituted 1,10-Phenanthrolines on proto accumulation and larval mortality in *Trichoplusia ni*. ALA and modulators were incorporated into *T. ni* diet to final concentrations of 4mM and 3mM, respectively. Proto content is expressed as nmoles per 100 mg of protein.

Treatment	Proto content ¹	Modulator alone			Proto content	Modulator + ALA		
		% larval mortality after the indicated length of time in the greenhouse				% larval mortality after the indicated length of time in the greenhouse		
		2 hours	1 day	3 days		2 hours	1 day	3 days
Control	1.52a	0.0a	0.0a	0.0a	2.99a	0.0a	0.0a	5.0ab
29DM47DPOPhDS	0.81a	0.0a	0.0a	0.0a	1.97a	0.0a	0.0a	0.0a
PHTN	1.03a	0.0a	0.0a	0.0a	3.87a	0.0a	1.7a	10.0ab
29DM47DPOPh	1.22a	0.0a	0.0a	0.0a	1.62a	0.0a	0.0a	6.7ab
47DPOPh	1.34a	0.0a	0.0a	0.0a	8.64a	6.7a	10.0a	26.7bc
5CIOPh	2.04a	0.0a	0.0a	10.0ab	133.85c	93.3b	97.7c	96.7d
3478TMOPh	5.06a	0.0a	1.7a	8.3ab	117.02c	91.7b	95.0c	95.0d
OPh	8.53a	1.7a	1.7a	5.0ab	207.00d	98.3c	100.0c	100.0d
47DMOPh	15.93a	0.0a	3.3a	8.3ab	131.46c	90.0b	96.7c	96.7d
5NOPh	50.37b	3.3a	25.0b	36.7c	190.14d	98.3c	98.3c	98.3d
Regression coefficient ²						0.95	0.94	0.91

¹Means followed by the same letter within a column are not significantly different at the 5% level of significance.

²Linear correlation of Proto content and larval mortality at the 5% level of significance.

Table 2. Effects of ALA and substituted 2,2'-Dipyridyls on proto accumulation and larval mortality in *Trichoplusia ni*. ALA and modulators were incorporated into *T. ni* diet to final concentrations of 4mM and 3mM, respectively. Proto content is expressed as nmoles per 100 mg of protein.

Treatment	Modulator alone				Modulator + ALA			
	Proto content ¹	% larval mortality after the indicated length of time in the greenhouse			Proto content	% larval mortality after the indicated length of time in the greenhouse		
		2 hours	1 day	3 days		2 hours	1 day	3 days
Control	0.85a	0.0a	0.0a	0.0a	2.88a	0.0a	0.0a	1.7a
22'DTh5NP	0.73a	0.0a	0.0a	0.0a	1.75a	0.0a	0.0a	1.7a
BQ	0.93a	0.0a	0.0a	0.0a	1.95a	0.0a	0.0a	0.0a
DPyDS	0.96a	0.0a	0.0a	0.0a	1.55a	0.0a	0.0a	0.0a
22'DThPNO	0.70a	0.0a	0.0a	0.0a	10.46a	18.3ab	41.7b	46.7c
44'DPDPy	1.22a	0.0a	0.0a	1.7a	2.29a	0.0a	0.0a	0.0a
Ph2PyKO	1.48a	0.0a	0.0a	1.7a	21.87a	20.0ab	21.7a	28.3b
44'DMDPy	1.87a	0.0a	0.0a	0.0a	18.07a	10.0ab	13.3a	23.3ab
TPy	6.22a	1.7a	5.0a	10.0a	181.10b	96.7c	96.7c	98.3d
DPy	15.00a	25.0b	46.7b	48.3c	176.33b	100.0c	100.0d	100.0d
Regression coefficient ²						0.99	0.94	0.93

¹Means followed by the same letter within a column are not significantly different at the 5% level of significance.

²Linear correlation of Proto content and larval mortality at the 5% level of significance.

Table 3. Effects of ALA and insecticidally active 8-Hydroxyquinolines, Pyridinium ions, 2-Oxypyridines, and Pyrroles on proto accumulation and larval mortality in *Trichoplusia ni*. ALA and modulators were incorporated into *T. ni* diet to final concentrations of 4mM and 3mM, respectively. Proto content is expressed as nmoles per 100 mg of protein.

Treatment ¹	Modulator alone				Modulator + ALA			
	Proto content ²	% larval mortality after the indicated length of time in the greenhouse			Proto content	% larval mortality after the indicated length of time in the greenhouse		
		2 hours	1 day	3 days		2 hours	1 day	3 days
Control	1.35	0.0	0.0	1.7	2.15	0.0	0.0	0.0
8OHSN5QUINSA	1.79	0.0	0.0	0.0	41.77	51.7	66.7	71.7
8OHQUIN5SA	4.53	0.0	0.0	0.0	32.11	43.3	70.0	78.3
EM4567THYDRO	0.92	0.0	0.0	0.0	10.01	10.0	18.3	53.3
ICBS	1.17	0.0	1.7	1.7	16.34	66.7	76.7	80.0
11D44CbCyI	3.45	0.0	71.7	96.7	21.92	25.0	61.7	88.3
BMACRN	5.64	0.0	0.0	10.0	10.91	0.0	26.7	53.3
11D24CyI	58.11	36.7	55.0	71.7	86.99	66.7	78.3	83.3

¹8-Hydroxyquinolines=8OHSN5QUINSA, 8OHQUIN5SA; Pyridinium ions=BMACRN, 11D44CbCyI, 11D24CyI; 2-Oxypyridines=ICBS; Pyrroles=EM4567THYDRO.