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Studies on Prevention of Resistance in *Cryptolestes Ferrugineus*

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Abstract: In this study we have compared the efficacy of different fumigation methods for control of *Cryptolestes ferrugineus* in maize, wheat and paddy, including: the recirculation fumigation with DDVP (by surface application) and AlP (application to surface and at intake), recirculation fumigation under film with DDVP (by intake) and AlP (application at intake and under the surface with small cloth-bag). The experimental results indicate that the last method can control *cryptolestes ferrugineus* effectively in imported wheat and prevent other insect pests at the same time.

Key words: *Cryptolestes ferrugineus*, resistance, prevention

Introduction

Cryptolestes ferrugineus (Stephens) (Coleoptera, Cucujidae Latreille) reproduces 3 – 6 generations per year and is a secondary pest. It over-winters as an adult, surviving on drier grain granule chippings, flour and dust, and its ability to tolerate low temperature is weak. Many individuals have obvious resistance to phosphine and the adult is good at flying. When it present in high numbers, it will cause grain the stack to generate heat and mould growth in part or the entire warehouse. The main damage occurs in cereals, oilseeds, flour and so on, especially in oilseeds and flour.

Resistance to phosphine is becoming broader and graver in stored grain worldwide (Rajendran, S. 1989; Price, L. A. and Mills, K. A. J. 1988; Dyte, C. E., Mills, K. A., and Price, N. R. 1983). Since the 1990s, there have been many reports in succession about serious resistance to *Cryptolestes ferrugineus* in Guang Dong province and Hunan province, China (Yu Wenjiang et al., 2004; Jiang Zhongzhu, Liu Xiaofu et al. 1990; Zhang Xinfu, Chen Jiadong, et al. 1985). In Zhejiang province, we find it is difficult to control *Cryptolestes ferrugineus* with ordinary phosphine dosages and it quickly re-established after fumigation. The National Grain Reserve Jinhua Depot is located in the Jinqu Basin, which experiences high temperatures and high humidity, the average annual temperature is above 22℃ and the mean temperature of grain stack is above 18℃ over half the year. Annual average relative humidity is 70%, and above 70% over 6 months. So relative humidity in the storage is above 65% over half a year. These conditions favour *Cryptolestes ferrugineus* reproduction. *Cryptolestes ferrugineus* is the most difficult pest to control with chemical treatments. It has obvious resistance to phosphine. The resistance factors (Rf) of the *Cryptolestes ferrugineus* is 121.6. There is currently no preventative or chemical control method effective against this species.

In recent years, staff at Jinhua Grain Depot, State Grain Reserves, have explored various methods to control *Cryptolestes ferrugineus*. We investigated the insects, their living habits and the effect of fumigation each year, and have accumulated a large amount of data. We found that fumigation with AlP and DDVP had greatest effect. Based on previous experimental data, we compared different fumigation methods that have been used in maize, wheat and paddy to control insect pests. We adopted two methods: recirculation fumigation with DDVP (by surface application) and AlP (by surface and by intake), the recirculation fumigation under plastic sheet film with DDVP (by intake) and AlP (application at intake and under the surface with small cloth-bag). We compared and analysed the effect of these control methods with the aim of developing an optimal technique to prevent *Cryptolestes ferrugineus* infestation.

1 Materials

1.1 Details of Field Trials are Listed in Table 1

1. *Jinhua Grain Depot, State Grain Reserves. Jinhua City, ZheJiang Province, China* 321018

2. *Zhejiang Branch Co., Central Reserves of Grain Management Co. Hangzhou City, ZheJiang Province, China* 310013

1.2 Materials

1.2.1 Gas Tightness of Storehouse. Half-life when warehouse pressure falls from 500 Pa to 250 Pa: No. 23, 42 sec; No. 35, 40 sec; No. 22, 44 sec; No. 2, 45 sec; No. 9, 43 sec; No. 7, 41 sec; No. 10, 40 sec.

1.2.2 PH_3 monitoring instruments: HL −210 type detecting instrument (Beijing Xinghualaoke Trade Ltd).

1.2.3 Grain temperature monitoring system: GSM grain electron temperature measureing system (Shaanxi Academy of Food Science and Edible Oil).

1.2.4 Recirculation Fumigation System: Stationary recirculation fumigation system (Beijing China Grain Science & Technology).

1.2.5 Sampling in Bulk: Grain multifunctional sampling system 1 500 kW, (Chengdu Grain Storage Research Institute).

1.2.6 Gas-proof material: adopt 0.16 mm PVC film

1.2.7 Fumigation chemicals 56% ALP; 和 85% ALP (Jiangsushuangling Chemical Industry Ltd.); DDVP (Nan Tong City, Jiangsu Province product)

2 Methods

2.1 Sealing Methods

2.1.1 Storehouse Sealing

In all experimental storehouses, the method of "two layers of non-woven fabric and three thickness of glue" was adopted to seal between the wall and precast floor slab and between the precast floor slabs. All doors, windows, gable axials and aeration ducting were sealed gas-tight using 0.16 mm PVC film.

2.1.2 Surface Sealing

The surface of grain mass was covered with 20 cm thick wraps that were filled with bran, and then used to seal 0.16 mm PVC film.

2.2 Fumigation Methods

2.2.1 Determining the Dosages

Factors taken into account to calculate dosage included: storage location, design and volume, insect density, type and resistance levels, climatic conditions and grain variety and sorption potential. The AlP dosages applied were: in the storehouse No. 23, No. 22, No. 9 : 150, 150 and 154 gm/tonne, respectively. DDVP dosage was 4 kg for each. In the warehouse No. 35, No. 2, No. 7, No. 10, ALP dosage were 152, 153, 153, 155 g/tonne respectively, and DDVP dosages were 6, 4, 4 and 4 kg, respectively.

2.2.2 Application Method

The insecticides were applied manually. AlP: in storehouses No. 23, No. 22 and No. 9, was applied to the surface on a plastic tray, and to the air-vents in two sacks. In the warehouse No. 35, No. 2 , No. 7 and No. 10, a small cloth-bag was put on the surface and two sacks containing AlP were placed in the air-vents. When the phosphine concentrationwas less than 200 ppm, more AlP was applied. In warehouses No. 23, No. 22 and No. 9, DDVP was sprayed on old sesame sacks which were placed on the grain surface. In warehouses No. 35 , No. 2 , No. 7 and No. 10, DDVP was sprayed on old sesame sacks which were placed on intake in the augur. Fumigation methods, application methods, and dosages are detailed in Table 2 for all warehouses.

2.2.3 Recirculation and measuring phosphine Concentration

Recirculation and concentration monitoring methods were carried out according to the recommendations of "Aluminum phosphide circular current fumigation operation standards". We used these guidelines as general principle but also considered the reality of the fumigation at the same time. After applying the chemicals, recirculation was continued for 48 h. The concentration was determined at 8 o'clock in the morning each day, and afterwards phosphine was recirculated for 2 – 3 hours in each 24 hours. After the recirculation was ended, the fumigant concentration was checked again. Concentration was monitored until the gas was cleared and AlP residue removed. There were 5 check points (a centre and 4 others around it), and each point distributed in three tiers. Gas dispersal was expressed by standard square difference (S); S value is getting smaller as approaching 0, explaining the data fluctuation less, concentration distribution homogeneity increases more.

2.2.4 Evaluation of the Fumigation Effect

Efficacy of the fumigation was measured as "time interval for no insect detection". Methods used to determine efficacy included: (1) As soon as ventilation period ended, examine grain for insect mortality. (2) Take random 1kg samples from the grain and incubate these in isolation in the laboratory at 25℃ for 30 days—this will indicate survival of immature stages. (3) Monitor grain stack for insects.

3 Results and Discussion

3.1 Integrated Fumigation Effect on *Cryptolestes Ferrugineus* and Other Insect Pest

After ventilation, we inspected grain in the seven warehouses. The mortality of *Cryptolestes ferrugineus* was 100% without exception. Random samples of grain were removed for isolated incubation but no insects were detected after 30 days. In follow-up inspections, *Cryptolestes ferrugineus* were only found in warehouses No. 22 and No. 23, where the time interval without insects was 88 days and 80 days, respectively. Insects were not detected in any other warehouse. A few mites were detected in some samples from warehouse No. 35, No. 10 , No. 7, No. 9, at 4 months after ventilation (1/kg、1/kg 、2/kg 、3/kgrespectively). Up to the present, *Cryptolestes ferrugineus* and acaridae have not beenfound in warehouse No. 2. To sum up, efficacy of the treatment in each of the warehouses was as follows: No. 2 > No. 35, No. 10 > No. 7 > No. 9 > No. 22, No. 23. Detailed results are shown in Table 3.

3.2 PH_3 Concentrations over Time

PH_3 concentrations at each check point are shown in Table 4. Phosphine distributed comparatively equality in No. 10 warehouse because of three "非" shape PVC circular pipes (with 6% orifice coefficient) placed under the grain mass surfaces at a depth of 60 cm, which facilitated homogeneous dispersal of the gas. But from the mortalities, the infection of distribution homogeneity was less. Phosphine concentration was equal to or greater than 300 ppm for 20, 16, and 22 days in No. 35, No. 2, No. 22 storages, respectively; PH_3 concentration was equal to or greater than 200 ppm for 28, 25 and 23 days in No. 2, No. 35, No. 10 storage, respectively; and, PH_3 concentration was equal to or greater than 100 ppm for 38 and 37 days in No. 22 and No. 9 storages, respectively. Prevention of infestation was achieved in storages No. 2, No. 35, No. 10, where phosphine concentration was maintained at equal to or greater than 200 ppm for a long time, whereas the effect was relatively poor in No. 22 and No. 23 because of loss of fumigant from the warehouse. These results demonstrate that complete control of resistant *Cryptolestes ferrugineus* can be achieved if the phosphine concentration is maintained at equal to or greater than 200 ppm for a long time.

3.3 The application Technique and Recirculation

Our trials indicate that effective control of grain insect pests can be achieved by recirculating fumigant using the following methods: by the aluminum phosphide film with small cloth-bag and by the combination of application of phosphine and DDVP at intake. Investigating the possible cause, we conclude that air tightness was crucial, which was directly correlated with PH_3 concentration and lowering the CT value. Gas tightness was obviously improved by covering the grain with PVC film and when the time to half pressure decay from −500Pa to −250Pa was more than 90 sec. The CT value was higher, so the effect of fumigation was better.

3.4 Effect of the Different Grain Types on Insect Control

Most even dispersal of PH_3 occurred in the storehouse containing wheat, dispersal was lowest in the paddy stored storehouse and it was between in the corn storehouse. When adopting recirculation fumigation under film, the effectiveness of fumigation was best in wheat, the corn and paddy took second place. The possible cause was the different sorption properties of grain types, which caused the PH_3 concentration discrepancy. Previous research (Jiang Liguo et al. 1999) indicated that sorption of PH_3 capability in turn was paddy > corn > wheat, it is compatible with our result.

3.5 Effect of Applying AlP and DDVP Together

The mortality of insect pests was 100% in 7 warehouses, the control effect was obvious especially to *Cryptolestes ferrugineus*. The possible cause may be that phosphine acts on insect energy metabolism system, insecticidal effectiveness is strong, penetration and diffusion of gases is good, sorbs less, desorbs rapidly, yet some insect has produced strong resistance to phosphine. DDVP acts on insect nervous system, it can induce insect movement from the grain mass. The effect is rapid, the "knock down" activity is strong, especially to insects of Coleoptera and Lepidoptera. The shortcoming is that volatilization and diffusion is slow, permeation is weak (Shen Zonghai, 1997). The current results indicate that fumigation with phosphine and DDVP together can complement each other, had a synergistic effect, and enhanced the insecticidal effect.

4 Conclusion

4.1 Fumigation with PH_3 only for long term, additionally grain is mainly exchanged from other province in ZheJiang Province, so as to the insect pest is introduced along with the grain. So the *Cryptolestes ferrugineus* appear strong resistance and its source is broad. These bring difficulties to insect control in stored grain.

4.2 This investigation indicates the optimal control method: recirculation fumigation under film, mixing AlP (the combination of application at intake and under the surface with small cloth-bags) and DDVP (intake application). The DDVP dosage is 1.3 g/m^3 and the PH_3 effective concentration is equal to or greater than 200 ppm for more than 28 days.

4.3 Because sorption capability of grain types varies, the effect of fumigation is different. From the experiment, when adopting recirculation fumigation under film with intake and under surface application, the effectiveness of fumigation is best in the wheat stored storehouse, the corn and paddy take second place.

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References

[1] Dyte, C. E., Mills, K. A., and Price, N. R. 1983. Proceedings 6th British Pest Control Controll Conference, Cambridge. Session 7, Paper 19

[2] Price, L. A. and Mills, K. A. J. 1988. Stored Prod. Res. 24, 51-59

[3] Rajendran, S. 1989. Pesticide Research Journal. 12, 111-115

[4] Zhang Xinfu, Chen Jiadong, et al. 1985. The preliminary investigation on phosphine resistant of cryptolestes ferrugineus and control suggestion. Grain Storage. 2, 10-14

[5] Jiang Zhongzhu, Liu Xiaofu et al. 1990. Test report on phosphine resistant of three Crypfolestes adults. Journal of the Chinese Cereals and Oils Association. 2, 35-42

[6] Yu Wenjiang et al., 2004. Integrated analyzing the control cryptolestes ferrugineus. LIANG YOU CANGCHU KEJI TONGXUN. 6, 31-33

[7] Jiang Liguo et al. 1999. Comparison of effect of multifactors on grain adsorbing Ph_3. Grain Storage. 4, 18-22

Table 1. Details of trials

NO.	Type	Volume (ton)	Type	Quantity (ton)	Storage date	Moisture content (%)	Test weight (g/L)	Foreign matter (%)	Brown rice recovery (%)	Average temperature (℃)	Species and density			
											Angoumoisgrain moth (per kg of grain)	Sitophilus zeamais (per kg of grain)	Cryptolestes Ferrugineu (per kg of grain)	Acaroid mites (per kg of grain)
23	Large warehouse	2720	corn	2627	2007.04	13.0	719	1.0	/	17.4	/	8	8	mass
35	Large warehouse	4070	corn	3800	2007.06	13.2	720	0.6	/	19.8	2	3	8	mass
22	Large warehouse	2720	import wheat	2750	2004.11	10.4	803	0.7	/	14.1	/	/	12	mass
2	Large warehouse	2720	import wheat	2700	2004.10	11.3	816	0.8	/	16.5	/	/	12	mass
9	Large warehouse	2720	early paddy	2047	2005.12	12.8	/	1.0	75.0	16.5	/	/	14	mass
7	Large warehouse	2720	early paddy	1946	2005.10	12.4	/	1.0	76.0	19.9	/	/	14	mass
10	Large warehouse	2720	early paddy	2012	2007.04	13.5	/	1.0	76.4	25.0	/	7	7	mass

Table 2. Fumigation method, application way and dosage

NO.	Fumigation method	ALP application way and dosage		DDVP application way and dosage	
		application way (first application + second application) (kg)	Dosage of each ton grain (tons/kg)	application way (kg)	Dosage (g/m^3)
23	Interval application, recirculation	surface (9 + 4.5) + intake (4 + 1)	150	surface (4)	0.8
35	Interval application, recirculation under film	under film with small cloth – bag (12 + 7) + intake (4.5 + 1.5)	152	intake (6)	1.1
22	Interval application, recirculation	surface (9.3 + 4.7) + intake (3 + 1.3)	150	surface (4)	0.8
2	Interval application, recirculation under film	under film with small cloth – bag (8.6 + 4.7) + intake (3 + 1.3)	153	intake (4)	1.3
9	Interval application, recirculation	surface (6 + 3.3) + intake (2.7 + 1.3)	154	surface (4)	1.1
7	Interval application, recirculation under film	under film with small cloth – bag (6.1 + 3.3) + intake (2 + 1.3)	153	intake (4)	0.8
10	Interval application, recirculation under film	under film with small cloth – bag (5.6 + 2.7) + intake (3.4 + 1.3), three "非 – shape" PVC circular pipes with 6% orifice coefficient were placed under grain mass surfaces 60 cm	155	intake (4)	1.1

Annotate: ALP dosage convert into powder

Table 3. Fumigation effectiveness

NO	ifferent PH_3 concentration maintaining time			otal exposure time (day)	PH_3 concentration while deflating (ppm)	The temperature where insects happened during fumigation		Mortality (%)	Insects species and density (per kg of grain)		None insects intervals (day)	Integrated effect evaluation of fumigation
	100	300	200			Max (℃)	Min (℃)		Cultivated for 30d	After the deflation		
23	9	19	35	40	47	31.0	25.5	100	None	1 cryptolestes ferrugineus, after 81d	80	*
35	20	25	36	40	56	33.4	29.2	100	None	1 acaroid mites, after 100d	No main insects	****
22	13	20	38	42	48	27.0	25.0	100	None	1 cryptolestes ferrugineus, after 89 d	88	*
2	16	28	32	35	53	27.7	25.0	100	None	None	up to the present	*****
9	11	20	37	41	42	29.8	27.2	100	None	3 acaroid mites, after 93 d	No main insects	**

NO	ifferent PH_3 concentration maintaining time			otal exposure time (day)	PH_3 concentration while deflating (ppm)	The temperature where insects happened during fumigation		Mortality (%)	Insects species and density (per kg of grain)		None insects intervals (day)	Integrated effect evaluation of fumigation
	100	300	200			Max (℃)	Min (℃)		Cultivated for 30d	After the deflation		
7	10	22	35	38	52	30.6	26.3	100	None	2 acaroid mites, after 79 d	No main insects	***
10	11	23	36	42	50	35.0	29.7	100	None	1 acaroid mites, after 104 d	No main insects	****

Table 4. The change of PH_3 concentration in experimental warehouse every 3d

No.		23	35	22	2	9	7	10
First application time		2007.6.19	2007.7.19	2007.8.6	2007.8.21	2007.7.24	2007.8.10	2007.6.22
1^{st}	southeast	380	600	360	550	496	570	560
	northeast	406	580	410	318	486	278	530
	northwest	520	600	>1000	458	291	420	535
	southwest	492	570	945	324	450	488	540
	center	452	610	300	460	272	765	425
	S	58	16	340	99	109	181	53
4th	southeast	456	450	645	>1000	414	446	332
	northeast	326	410	540	890	390	368	328
	northwest	418	460	>1000	890	368	318	318
	southwest	398	408	>1000	895	412	402	312
	center	438	470	384	>1000	350	590	300
	S	50	29	277	59	28	104	13
7th	southeast	368	493	660	680	330	440	312
	northeast	238	472	484	600	334	260	308
	northwest	300	492	925	710	338	380	305
	southwest	354	470	334	585	320	380	300
	center	356	418	384	615	314	286	290
	S	54	30	240	54	10	74	8
10th	southeast	346	382	570	535	286	325	218
	northeast	216	377	416	386	296	162	214
	northwest	278	380	452	476	294	283	204
	southwest	318	378	370	324	280	280	199
	center	316	340	410	376	242	186	210
	S	50	18	76	85	22	70	8
13th	southeast	258	296	328	428	144	210	158
	northeast	127	303	292	326	240	152	148
	northwest	154	294	316	382	179	182	138
	southwest	276	304	336	254	154	179	139
	center	266	268	360	306	160	166	150
	S	70	15	25	68	38	22	8