Lethal and Antifeeding Effects of Carbofuran against the Whitebacked Planthopper, *Sogatella furcifera*(Horvath) (Homoptera: Delphacidae)

LIU Guangjie[1)], R. M. WILKINS

(Department of Agricultural and Environmental Science, The University, Newcastle upon Tyne NE1 7RU, United Kingdom; [1)]Present address: China National Rice Research Institute, Hangzhou 310006, China)

呋喃丹对白背飞虱的致死与拒食作用

刘光杰[1)] R. M. WILKINS

(纽卡索大学农业及环境科学系，英国纽卡索市；[1)]现在地址：中国水稻研究所，杭州 310006)

摘 要：就呋喃丹对白背飞虱雌成虫的致死与拒取食作用进行了研究。采用叶鞘涂抹法、点滴法和根部施药法施用呋喃丹以后，观测白背飞虱的死亡率和取食反应。呋喃丹对白背飞虱的致死中浓度 (LD_{50}) 分别为：0.51 $\mu g/cm^2$(叶鞘涂抹法)，0.86 $\mu g/g$ (点滴法) 和 8.82 $\mu mol/l$ (根部施药法)。其中，点滴呋喃丹对飞虱最有效。当白背飞虱在呋喃丹以 0.016 $\mu g/cm^2$ 涂抹在感虫品种 TN1 稻株叶鞘上和将稻株根部浸入 0.1 $\mu mol/l$ 药液里，以及在每头飞虱上点滴 0.002 ng 后，飞虱在这些处理过的稻株上取食后其蜜露排泄量明显减少。白背飞虱在用呋喃丹处理过 TN1 根部的稻株上的取食行为也采用一种电子装置进行了监测。

关键词：拒食性；呋喃丹；致死作用；白背飞虱；毒理

Abstract: Lethal and antifeeding effects of carbofuran against the adult females of *Sogatella furcifera* were studied by measuring insect mortality and feeding reduction with leafsheath, topical and root-zone applications. The LD_{50} values for leafsheath, topical and root-zone applications were 0.51$\mu g/cm^2$, 0.86$\mu g/g$, and 8.82$\mu mol/l$, respectively. Among the application methods, topical application of carbofuran was most effective against *S. furcifera*. Honeydew excretion reduced significantly when *S. furcifera* fed on the leafsheaths of susceptible TN1 plants treated with carbofuran in 0.016$\mu g/cm^2$ and in 0.1$\mu mol/l$ at root zone, and when insects were treated topically at 0.002ng / female. Feeding behaviour of *S. furcifera* on TN1 plants treated at the root zone was also investigated using an electronic device.

Key words: Antifeeding; Carbofuran; Lethal action; *Sogatella furcifera*; Toxicology

The whitebacked planthopper, *Sogatella furcifera*, is now emerging as a serious pest of rice in several Asian countries, particularly in areas where varieties resistant to the brown planthopper, *Nilaparvata lugens*, have been grown successfully [1]. Both nymphs and adults attack rice plants directly by sucking the phloem sap [2,3] resulting in slow grow, delayed tillering and reduction in grain formation. Various control measures such as chemical, biological and varietal resistance are applied in the management of this pest. The use of insecticides is still the most common method known to bring about immediate reduction in pest populations. The heavy use of some insecticides, however, will cause the resurgence of *S. furcifera* on the treated plants [4,5,6] and contaminate the environ-

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ment. Fujiwara (1971) reported that the insecticide chlordimeform at sublethal levels exhibited antifeeding activity to the phytophagous insects *Pryeria sinica* and *Calopilos miranda* [7]. Since then, entomologists have paid more attention to searching for insect antifeedants, chemicals which reduce and prevent insect feeding. Several insecticides at sublethal levels were reported to possess antifeeding properties against insect pests [8,9,10]. Jeevaratnam (1984) demonstrated that *N. lugens* fed significantly less on susceptible TNl treated with carbofuran at sublethal doses [11].

The available information on the toxicity of carbofuran to *S. furcifera* is limited. Few studies have investigated antifeeding effect of carbofuran at sublethal levels against *S. furcifera*. Therefore, the work reported here was designed to study lethal and antifeeding effects of carbofuran to *S. furcifera* with leafsheath, topical and root–zone applications, and electronic recording of the feeding behaviour of this pest on treated TNl plants.

Materials and Methods

Carbofuran (2,3–dihydro–2,2–dimethyl–7–benzofuranyl–N–methyl–carbamate) supplied by Bayer Co., Germany with the purity of 99.4% was used. Test insects were reared on fresh TNl plants. Unless otherwise stated, macropters (0~2 day old) of *S. furcifera* and 6–week–old TNl plants were used. All experiments were conducted at 25±2℃ with dark / light period of 12 / 12 h; replicates in given experiment were completely randomized.

1. Mortality Response

Mortality was assessed by counting dead insects at 24 h after treatment. The toxic effect of carbofuran to *S. furcifera* was assessed based on Finney's (1971) probit analysis [12]

Leafsheath Application

A solution (140 μl) of carbofuran in acetone of appropriate strength was applied to a 7 cm^2 surface area of a stem with a small paint brush. The final residues on each stem were 1.106, 0.885, 0.665, 0.443 and 0.221 $\mu g / cm^2$. Acetone only was used as a control. Test plants were left at least 10 min to ensure complete evaporation of the acetone. Fifteen females were introduced into a feeding chamber [13] allowing exposure to the treated stems only.

Topical application

Carbofuran in acetone solution was applied topically with an electronic microapplicator in 0.1μl to the dorsal surface of a test female. The final doses were 2.5, 1.7, 1.2, 0.8 and 0.2 ng / female. Acetone alone was applied to the control. Ten treated insects were introduced into a petri dish and fed by providing two pieces of TNl stems.

Root–zone application

The roots of each plant (no soil) were immersed in approximately 250 ml of aqueous solutions containing carbofuran at 16, 13, 10, 7 and 4 μmol / l. The control was treated with water alone. Ten females were released into each feeding chamber allowing the access to the rice stem and the number of dead insects was counted at 48h after treatment.

2. Antifeeding Response

Honeydew Excretion

The methods of treating test insects were the same as described above. The final doses or concentrations of carbofuran directly / indirectly used to treat test insects were 0.159,0.079 and 0.016$\mu g / cm^2$(leafsheath application), 0.1, 0.02, 0.01 and 0.002ng / female (topical application), and 2, 1.5, 1, 0.5 and 0.1 μmol / l(root–zone application).Three to 5 treated insects were released into a feeding chamber which represented a replicate. Honeydew excretion was collected after 24 h and measured colorimetrically [14]. Reductions in honeydew excreted by treated insects were determined by the differences between honeydew excreted by untreated and treated insects, measuring as antifeeding response.

$$\text{Antifeeding response}(\%) = \frac{B - A}{B} \times 100$$

where, A = quantity of honeydew excreted by carbofuran–treated *S. furcifera* and B = quantity of honeydew excreted by untreated *S. furcifera*.

Antifeeding response of *S. furcifera* on carbofuran was assumed to be the response of a *S. furcifera* population and probit analysis was therefore applied to calculate the median antifeeding dose / concentration (MAD_{50} / MAC_{50}). To compare the 50% responses of *S. furcifera* to carbofuran, the term "efficiency index" was used and calculated as follows:

$$\text{Efficiency index} = \frac{LD_{50}(LC_{50})}{MAD_{50}(MAC_{50})}$$

where the slopes of the log dose—probit lines compared are not similar then this index is not constant, and the comparison is not statistically valid. However, it indicates a level of dosage differences for the two different responses.

Electronic Recording of Feeding Activity

TNl plants were treated with carbofuran in 2μmol / l at root zone. The electronic recording procedure followed by Khan & Saxena [15] and the feeding activities of *S. furcifera* were recorded for 180 min as a replicate, using different plants and insects. Reduction in phloem ingestion was calculated similarly with antifeeding response.

Results

1. Toxic and Antifeeding Response

The mortality and antifeeding probit regression equations together with the calculated $LD_{50}(LC_{50})$ and $MAD_{50}(MAC_{50})$ values are given in Table 1. Compared with antifeeding, the insect's mortality increased drastically with the increasing dose (concentration) used. Among 3 application methods, topical application had highest efficiency index, indicating that it was most effective against *S. furcifera*. The insect excreted significantly ($P<0.05$) less honeydew when carbofuran was applied on leafsheath at 0.016μg / cm^2, topically treated at 0.002ng / female and applied at root zone in 0.1μmol / l (Table 2).

2. Electronic Recording of Feeding Activity

During the 180min feeding period, *S. furcifera* made more probes, and had longer salivation and shorter phloem ingestion periods on treated TNl than on untreated (Table 3). The frequency of phloem ingestion durations falling in the range of 20~ 130 min was much lower on treated TN1 than on untreated TNl (Fig.1). Moreover, there were more than 60% of short (<10 min / probe) phloem ingestion durations on treated TN1 in a contrast of less than 40% on untreated TNl.

Discussion

The results reported here illustrate that carbofuran was most toxic against *S. furcifera* in topical application. Compared with 0.46 and 0.32μg / g, the topical LD_{50} values of carbofuran against *S. furcifera* reported respectively by Heinrichs *et al.* [16] and Fabellar and Heinrichs [17], the topical LD_{50} value (0.86 μg / g) in the present study was

Table 1. Calculated probit regressions, $LD_{50}(LC_{50})$ and $MAD_{50}(MAC_{50})$ values for responses of *S. furcifera*[1)] treated with carbofuran

Treatment	Response	Regression equation[2)]	LD_{50} or MAD_{50}	Fiducial limits	Efficiency index
Leaf sheath (μg / cm^2)	Mortality	y = 2.81+3.11x	0.51	0.44—0.58	
	Antifeeding	y = 4.38+0.60x	0.11	0.05—0.23	4.8
Topical (μg / g)	Mortality	y = 1.96+2.86x	0.86	0.65—1.13	
	Antifeeding	y = 4.17+0.85x	6.72	4.48—9.70	128.9
Root zone (μmol / l)	Mortality	y = 2.19+2.97x	8.82	7.45—10.45	
	Antifeeding	y = 3.99+1.05x	0.90	0.64—1.27	9.8

Note: 1) Mean female body weight = 1.34mg. 2) y indicates probit and x log dose (concentration); units are μg / cm^2 in leafsheath, μg / g in topical and μmol / l in root zone.

Table 2. Antifeeding effect of carbofuran against *S. furcifera*[1)]

Treatment method	Concentration	Quantity of amino acids in honeydew[2)] (μg dry weight / female per 24h)	Antifeeding response (%)
Leafsheath(μg / cm^2)	0.159	28.6a	55.2
	0.079	33.7a	47.3
	0.016	43.8 b	31.5
	0.0	63.9 c	
Topical(ng / female)	0.1	23.3a	77.9
	0.02	34.5a	67.3
	0.01	54.1 b	48.8
	0.002	77.9 c	26.2
	0.0	105.6 d	
Root zone(μmol / l)	2.0	18.5a	67.4
	1.5	23.0 b	59.5
	1.0	30.9 c	45.6
	0.5	36.2 d	36.3
	0.1	46.9 e	17.4
	0.0	56.8 f	

Note: 1)Within a treatment method, means followed by the same letters are not significantly different at P=0.05 by Duncan's (1951) multiple range test(DMRT). No mortality was observed with any concentration (i.e., less than 5% mortality). 2) Average of 3 replicates; Quantity of amino acids in honeydew measured colormetrically using glutamic acid as standard.

Table 3. The electronically–recorded events in 180 min feeding of *S. furcifera* on TNl plants treated with aqueous carbofuran solution of 2μmol / l at root zone

Treatment	Electronically recorded events[1)]			
	Probes (no)	Salivation (min)	Phloem ingestion (min)	Non–feeding (min)
Treated TNl	11.0 a	29.6 a	84.3 a	66.1 a
TNl	6.4 b	11.1 b	151.7 b	17.2 b

Note: 1) Average of 7 replicate, each replicate using a new plant and new insect. In a column, means followed by the same letters are not significantly different at P = 0.05 by DMRT.

obviously high. Similarly, Nagata [18] and Fabellar and Heinrichs [17] obtained different topical LD_{50} values of 0.32 and 0.76μg / g for propoxur, and 0.55 and 1.04μg / g for carbaryl, respectively. This kind of phenomenon could be due to the variations in insect morphs, body weight and age of test insects or possibly droplet size of test solution. So far, there are few reports on the LD_{50}(LC_{50}) values of carbofuran against *S. furcifera* with leafsheath and root–zone applications. In leafsheath application, it is probable that the mortality of *S. furcifera* was caused by the presence of carbofuran which permeated into the vascular bundles of rice plants. More than 80% mortality was recorded after a day when *S. furcifera* infested on TNl plants sprayed with carbofuran 12F at 0.25kg (a.i.) per hectare [19]. The mortality on treated TNl plants resulted from the translocation and persistence in the

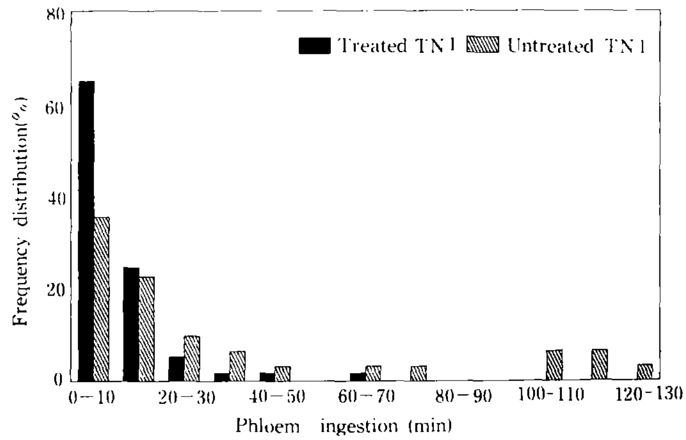

Fig.1. Frequency distribution of 88 individual phloem ingestion durations (57 for treated TNl and 31 for untreated TNl in seven replicates) of *S. furcifera* in 180 min feeding on TNl plants immersed into aqueous carbofuran solution of 2 μmol / l

treated plants. Siddaramappa *et al.* demonstrated that most of the carbofuran absorbed by rice was accumulated in the leaves and in the stem region [20].

An insect's feeding can be inhibited even on most preferred host plants containing an optimal combination of feeding stimuli when applied with effective feeding deterrents [21]. The present investigation shows that carbofuran, an insecticide widely used in the paddy field, also depresses feeding of *S. furcifera* at sublethal levels. The insect fed significantly less on susceptible TNl plants treated with carbofuran in all three application methods. The finding was supported by electronic recording of the feeding behaviour of *S. furcifera* treated indirectly with carbofuran. The insect became restless, probed more and had shorter phloem ingestion periods (< 80min / probe) on the treated TNl plants than on the untreated ones. The preliminary tests indicated that reductions in honeydew excretion were caused by treating with carbofuran at sublethal levels rather than other factors such as CO_2 anaesthetization of, and acetone applied on, test insects [22]. Also, Jeevaratnam (1984) reported that honeydew production by *N. lugens* reduced significantly on TN1 plants painted with carbofuran solution in acetone at 0.07μmol / cm^2 and applied at 0.005% at the root zone [11]. Moreover, Meisner *et al.* demonstrated that larval weight gain of the Egyptian cotton leafworm, ***Spodoptera littoralis***, was significantly lower on alfalfa–treated with 0.3, 0.2, and 0.1% (a. i.) of pronamide [23] Therefore, it could be concluded that *S. furcifera* feeding was inhibited by carbofuran which was translocated into rice phloem and depressed feeding of the pest. However, effective delivery systems and appropriate field testing are required to evaluate any potential of carbofuran as an antifeedant in an IPM approach to *S. furcifera* control.

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