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HYDROGEN CYANIDE AS AN IMMEDIATE ALTERNATIVE TO METHYL BROMIDE FOR STRUCTURAL FUMIGATIONS

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ABSTRACT

Flour mill fumigations are currently carried out with methyl bromide (MB). By 2005, this gas will be phased out in developed countries because it has been listed under the Montreal Protocol as a substance that depletes the ozone layer. Hydrogen cyanide (HCN) was used as the fumigant of choice for mills in the first part of the last century, and was then replaced by MB. Consequently, it should now be re-evaluated as a potential alternative to MB. However, knowledge about HCN's efficacy as a fumigant is outdated and does not conform to the notion of concentration time (Ct) product. Therefore, the objective of this study was to determine the biological efficacy of HCN in terms of Ct product for the different stages of three of the most prevalent species infesting mills and food manufacturing premises, the confused flour beetle *Tribolium confusum,* the red flour beetle *Tribolium castaneum,* and the Indianmeal moth, *Plodia interpunctella* In addition, the granary weevil *Sitophilus granarius* was studied in the adult stage only, since this is the only stage present in mills. Two series of trials were carried out: one consisted of, dose-trials to define the relationship between Ct product and insect mortality without presence of a rearing medium; the other consisted of trials in which inserts were placed at different depths in flour to measure HCN efficacy in penetrating residual heaps of flour. Our findings on the different stages of each species (eggs, young larvae, old larvae, pupae and adults) show that the Ct product required to obtain a LD_{90} ranged from 1 to 4 g·h·m^{-3}, according to developmental stage, this being very low when compared to MB. Nevertheless, in contrast to MB, large differences were found between the species as shown in the high tolerance of granary weevils to HCN. The poor penetration of HCN into flour is due to significant sorption. Consequently, to be effective against *T. confusum* adults buried in flour, a Ct product as high as 60 and 100 g·h·m^{-3} is required to kill this species at depths of 10 and 15 cm, though it does not kill *S. granarius*. In practice, the Ct product needed to kill all stages of these major mill and food factory pests should be around 10 g·h·m^{-3}, though to obtain HCN penetration and kill insects at a depth of about 10 cm in flour heaps, the prevailing Ct products should be around 60 g·h·m^{-3}. This implies that the corresponding initial dose should be 5 g m^{-3}, taking into account the presence of minor leaks.

INTRODUCTION

Good sanitation of flour mills and other industrial food manufacturing premises is carried out for the most part through thorough cleaning and by monitoring the insect population, but also by chemical means using, contact insecticides and fumigants. Contact insecticidal treatments consist of surface spraying or space aerosol applications, but because the efficacy of these treatments is somewhat limited, complementary fumigations with a total curative action are often used to disinfest the sites. In some countries such as France, Germany, and Switzerland, two fumigants are authorized namely: methyl bromide (MB), which is currently the most commonly used fumigant, and hydrogen cyanide (HCN), which had been used since 1886 (Cotton, 1963), and was largely used as the fumigant of choice for flour mills in the first part of the last century. Phosphine (PH_3) is not suitable due to its corrosive characteristics and the prolonged exposure times required. History reveals a process of constant reappraisal. In the 1970's, MB replaced HCN for the good reasons of better penetration, lower cost and ease of cylinder refill. The phase-out of MB means that these advantages are no longer valid and HCN may now be envisaged for use again with renewed interest.

However, knowledge on the efficacy of HCN as a fumigant is outdated and existing data cannot be correlated with the present concept of the Ct product (Bond, 1984). Therefore, the objective of this study was to determine the insecticidal efficacy of HCN in terms of Ct product for the different developmental stages of three of the most prevalent species in the food industry and in flour mills: the confused flour beetle *Tribolium confusum* (J. du Val), the red flour beetle *Tribolium castaneum* (Herbst), and the Indianmeal moth *Plodia interpunctella* (Hübner). In addition, the adult stage of the granary weevil *Sitophilus granarius* (L.) was studied, this being the only stage present in mills. Two series of trials were carried out. In the first, dose-experiments were undertaken to define the relationship between the concentration and time (Ct product) required to obtain mortality of the insects without the presence of a rearing medium (flour), while in the second series of trials, an evaluation was made of the penetration of HCN into flour, and its efficacy at different depths of farinaceous material that may be encountered in places where residual cleaning has not been effectively carried out.

MATERIALS AND METHODS

Hydrogen cyanide production in the laboratory

Hydrogen cyanide was produced by allowing a solution of diluted sulfuric acid (H_2SO_4) to react with a solution of potassium cyanide (KCN) according to the following reaction:

$$2\ KCN + H_2SO_4 \longrightarrow K_2SO_4 + 2\ HCN$$

A 5 mL volume of potassium cyanide at 100 g/L was put in a 1-L gas-production jar. A syringe containing 2 mL of sulfuric acid solution was injected into the upper part of the jar allowing the acid to drain directly into the jar and into contact with the KCN. A 100 mbar absolute vacuum was created previously in the jar for safety, in order to prevent a build-up of excess pressure. This reaction is highly exothermal and is complete, since there is excess acid. Homogenization of the HCN concentration was obtained during return of the jar to atmospheric pressure.

Concentration measurements.

The method used for HCN concentration measurement was gas chromatography with a flame ionization detector. HCN concentration in the gas-production jar was measured in order to determine the volume of gas mixture to add to exposure flasks in order to obtain a given dose. The same analyses were carried out in the exposure flasks in order to measure changes in HCN concentration over time. The calibration was carried out using a reference gas of 8,900 ppm HCN compressed in a cylinder.

For each of the insect species, the efficacy of HCN was tested at the egg, larval, pupal and adult stages of development except for *S. granarius* where only the adult was investigated. Insect breeding was carried out in air conditioned rooms at 27±1°C, and 70±10% r.h., with a photoperiod of 16 hours/day and 8 hours/night for the two species of *Tribolium* and *Plodia interpunctella,* and without a photoperiod for *S. granarius*.

Fumigant toxicity was studied by exposing the different species at their different stages to a range of doses. The fixed parameters were temperature at 20°C and exposure period at 1 h for the Ct experiments and 24 h for experiments on gas penetration into flour. In each case, the control batches were subjected to the same temperature conditions.

Fumigation of insects without flour

The batches of insects were placed in gastight exposure flasks of 2-L capacity. Having calculated the HCN dose to be applied, the corresponding gas volume was withdrawn from the gas-production jar using a Hamilton gas syringe and injected directly into the exposure flasks containing the insects. Three gas concentration measurements were performed during the treatment at 1 min, 20 min and 1 h after gas application. After the fumigant had been aired from the exposure flask, the insects were placed in petri dishes containing wheat flour for flour beetles and wheat for the granary weevil. The dishes were then put in a rearing room, and mortality was counted 7 and 14 d after treatment.

Fumigation of insects in flour

The fumigations were carried out in gastight 3-L cylindrical glass jars, 40 cm high and 10 cm in diameter equipped with four septa at different heights. These jars were filled with wheat flour to a height of 20 and 30 cm respectively in the first and second series of experiments.

Fumigation in sealed cylindrical jars: Two 24-hour trials at 20°C were carried out with two different HCN doses, 5 g m^{-3} and 6 g m^{-3}. These doses are close to those applied during mill fumigations and correspond to a Ct product of 12.6 g·h·m^{-3} and of 14.2 g·h·m^{-3}. Batches of adult *T. confusum* and *S. granarius* wrapped in paper envelopes were placed at 4, 8, 12, 16 and 20 cm depths within the flour. The fumigant was applied in the space above the flour. A slight vacuum was created in the jars to facilitate injection of the gas. Gas concentrations were measured throughout the treatment at three levels: in the air above the flour; 2 cm into the flour; and at the bottom of the jar (20 cm from the surface). At the end of the exposure period, the jars were opened to allow the fumigant to dissipate and the insects were then placed in petri dishes for mortality counts 7 and 14 d after treatment.

Fumigation in open cylindrical jars in a 1m^3 fumigation chamber: The experimental design was the same as above, but the glass jars remained open and were placed inside a 1 m^3 fumigation chamber. This allowed a relatively constant HCN concentration to be achieved, reproducing what might occur in a flour mill or food factory. The insects were placed at depths of 5, 10, 15, 20 and 30 cm in the flour. The fumigant dose was applied directly into the chamber by the reaction of a H_2SO_4 solution with a solution of KCN. Two dosages were used: 2.5 and 5 g m^{-3} of HCN. During the treatment, fumigant concentrations in the air space were measured.

RESULTS

The results were subjected to probit analysis (Finney, 1977) using SAS statistical software. The findings on adult susceptibility were compared with susceptibilities to MB obtained through our own experimentation or with results given in the scientific literature**.**

Fumigation of insects without flour

Plodia interpunctella: Due to a lack of eggs of different ages, only the results of two-day-old eggs were analyzed. The Ct products giving LD_{90} (Table 1 and Fig. 1) ranged from 1.0 g·h·m^{-3} for young larvae to 1.2 g·h·m^{-3} for old larvae, with 2.6 g·h·m^{-3} and 2.0 g·h·m^{-3} for pupae and eggs respectively.

In comparison, the results of Bell (1976), showed that for two-day-old and older eggs, which was the stage most resistant to MB under the same temperature conditions, it was necessary to apply MB at a Ct product of 16 g·h·m^{-3} to obtain a mortality of 50%, whereas we found that only 1.7 g·h□h^{-3} (or 10 times less) of HCN was sufficient.

TABLE 1
Ct product necessary to obtain LD_{50} and LD_{90} for a 1-hour HCN exposure of different stages of *Plodia interpunctella* at 20°C

Life stage	LD 50 g·hm^{-3}	50% Fiducial limits	LD 90 g·hm^{-3}	90% Fiducial limits	Slope ln (Ct)± SD
Pupae	1.6	1.4 – 1.8	2.6	2.2 – 3.4	2.7 ± 0.5
Old larvae	0.7	0.6 – 0.8	1.2	1.1 – 1.5	2.5 ± 0.3
Young larvae	0.8	0.6 – 0.9	1.1	1.0 – 1.6	3.4 ± 0.9
2-day-old eggs	1.7	1.6 – 1.8	2.0	1.9 – 2.3	7.0 ± 1.0

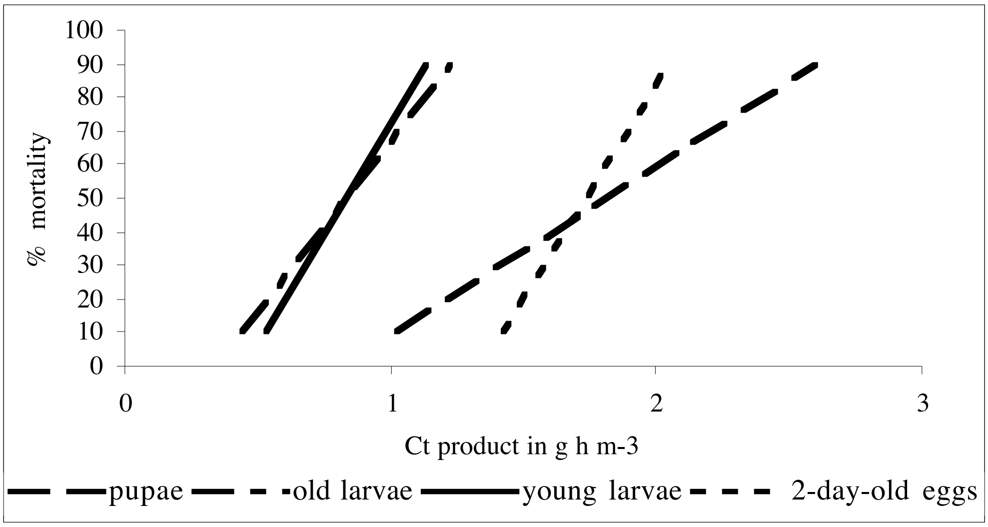

Fig. 1. Probit mortality curves of 1 hour exposures of HCN on the life stages of *Plodia interpunctella* at 20°C.

Tribolium confusum: Table 2 and Figure 2 show that the LD_{90} of the pupae and eggs of *T. confusum* were close to 4 g·h□h^{-3}, with 1 g·h□h^{-3} for young larvae. A comparison with MB fumigation for the adult stage shows that for MB a Ct product of 35.7 g·h·m^{-3} is necessary to obtain 90% mortality, or almost 28 times more than with HCN.

TABLE 2
Ct products necessary to obtain LD_{50} and LD_{90} for a 1-hour HCN exposure of different stages of *Tribolium confusum* at 20°C and a comparison with sensitivity of adults to MB

Life stage	HCN					MB		
	LD_{50} g·h m^{-3}	50% Fiducial limits	LD_{90} g·h m^{-3}	90% Fiducial limits	Slope ln (Ct) ± SD	LD_{90} g·h m^{-3}	90% Fiducial limits	Slope ln (Ct) ± SD
Adult	1.0	0.9-1.0	1.3	1.0-1.4	4.4±0.5	35.7	31.5-55.4	5.1±1.6
Pupa	2.1	2.6-ND	4.3	ND	1.9±5.2			
Old larva	0.8	0.5-1.12	1.9	1.3-5.7	1.6±0.4			
Young larva	0.6	0.2-0.7	1.0	0.8-2.4	2.1±0.7			
Egg	0.6	0.5-0.7	3.6	2.5-6.4	0.7±0.1			

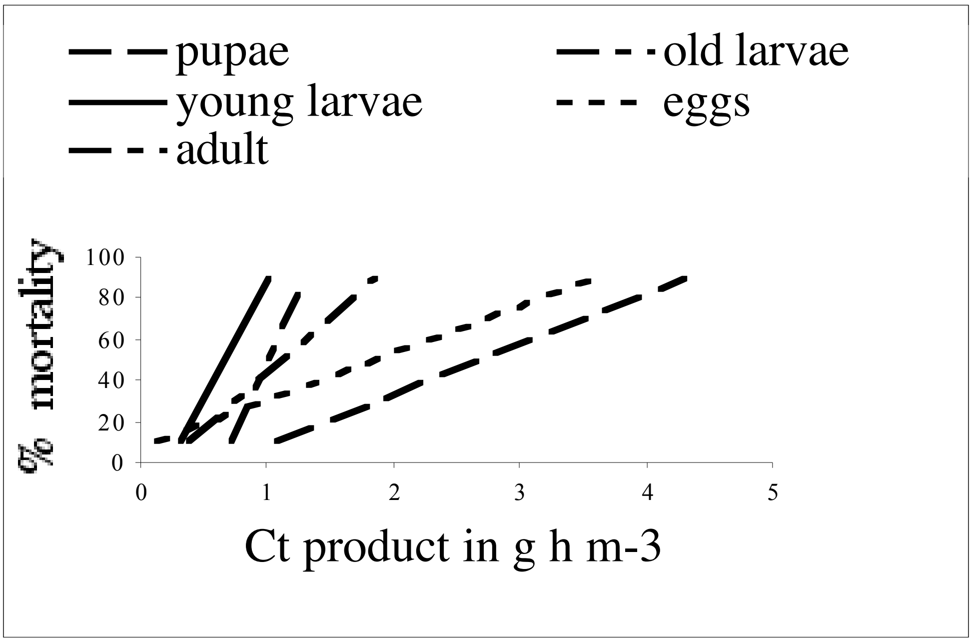

Fig. 2. Probit curves of HCN action on Confused flour beetles for a 1-hour treatment at 20ºC.

Tribolium castaneum: The results of exposure of *T. castaneum* to HCN are comparable to those obtained for *T. confusum* except for the pupal stage where an

LD_{90} of 6.2 g·h m^{-3}, was obtained for *T. castaneum*, though this difference is not statistically significant (Table 3 and Fig. 3). A comparison with MB fumigation of the adult stage under the same conditions indicates that for MB it is necessary to apply a Ct product of 25.7 g·h m^{-3}, compared to 0.8 g·h m^{-3} for HCN.

TABLE 3
Ct product necessary to obtain LD_{50} and LD_{90} for a 1-hour HCN exposure time on different stages of *Tribolium castaneum* at 20ºC and a comparison with MB exposure of adults

Life stage	HCN					MB		
	LD_{50} g·h m^{-3}	50% Fiducial limits	LD_{90} g·h m^{-3}	90% Fiducial limits	Slope ln (Ct) ± SD	LD_{90} g·h m^{-3}	90% Fiducial limits	Slope ln (Ct) ± SD
Adult	0.8	0-0.9	1.2	1.0-1.5	3.4±0.5	25.7	22.4-33.6	3.1±0.6
Pupa	2.3	1.7-5.0	6.2	3.4-35.3	1.3±0.3			
Old larva	0.3	0.0-0.6	2.0	1.1-57.6	0.7±0.2			
Young larva	0.6	0.5-0.6	0.8	0.7-0.9	4.4±0.7			
Egg	0.6	0.6-0.8	1.3	0.9-8.9	1.0±0.5			

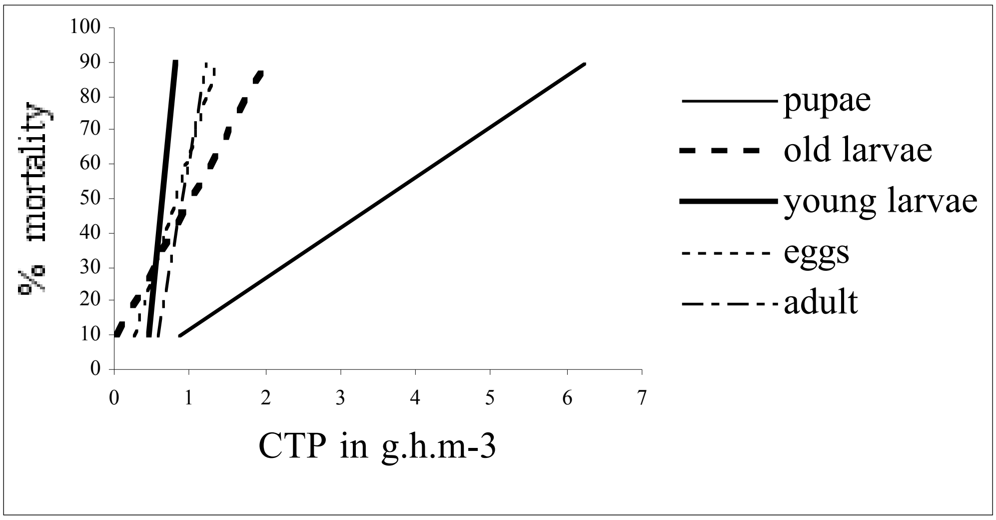

Fig. 3. Probit curves of HCN action on Red flour beetles for a 1-hour treatment at 20ºC.

Sitophilus granarius: The results given in Table 4 and fig. 4 indicate that *S. granarius* is more tolerant to HCN than to MB, the probit slope being around 2 for

HCN treatments and 4.8 for MB. The Ct product for a mortality of LD_{90} was 36 g·h·m^{-3} with HCN, whereas for MB it was 15.7 g·h·m^{-3}.

TABLE 4
Comparison of the action of HCN and MB on adult *Sitophilus granarius* at 20ºC, for different exposure times

Exposure time	HCN			MB		
	LD90 g·h m^{-3}	90% Fiducial limits	Slope ln (CT) ± SD	LD90 g·h m^{-3}	90% Fiducial limits	Slope ln (CT) ± SD
1 h at 20ºC	-	-	-	15.7	13.8 – 20.4	4.8 ± 0.9
2 h at 20ºC	36.2	27.3 – 59.9	1.8 ± 0.2			

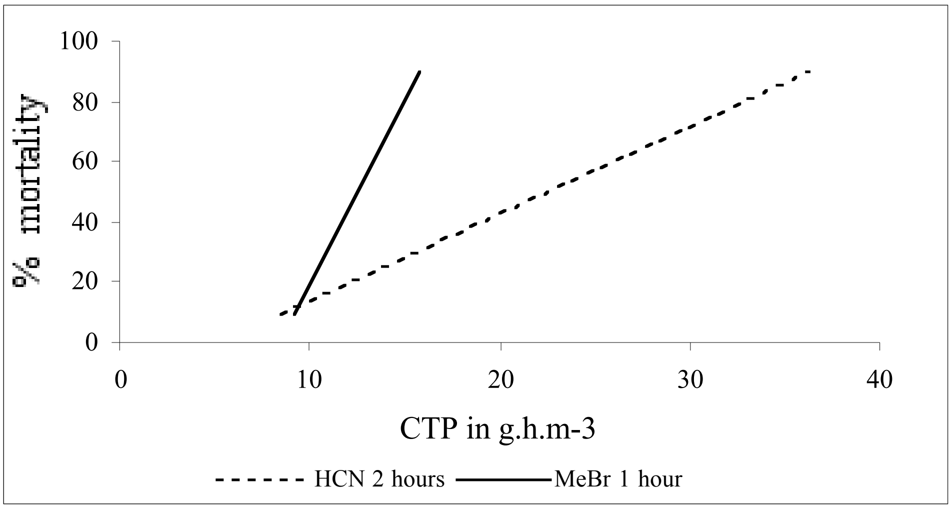

Fig. 4. Comparison of HCN and MB efficacy on adult Granary weevils at different exposure times at 20ºC.

Our findings show that HCN is very effective against these major flour mill and food industry insects, since it only requires a Ct product of around 5 g·h·m^{-3} to obtain an LD_{90}, except for *S. granarius* which only reproduces in grains and not in milled products. Even those stages that are usually difficult to kill, namely the eggs and pupae do not require a high Ct product.

Fumigation of insects in flour

HCN penetration: From Fig. 5, it can be seen that HCN was strongly sorbed into flour. In fact, the HCN concentration measured above the flour decreased from 6.0 to 2.8 g·m^{-3} in 4 min and stabilized within 6 h. Within 24 h, the three concentration measurement points above and within the flour reached an equilibrium whose value oscillated between 0.25 and 0.35 g·m^{-3}.

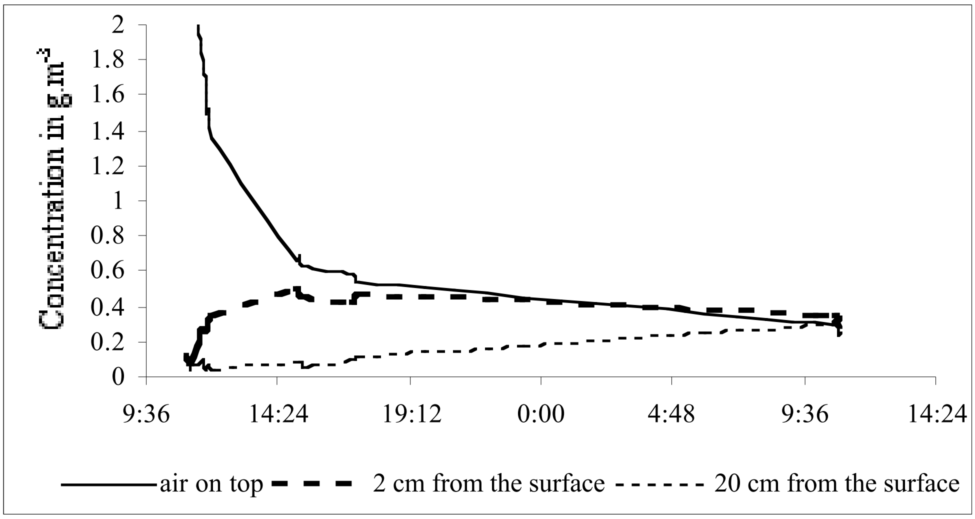

Fig. 5. HCN concentration measurements at different points in a cylindrical glass jar.

Fumigation in sealed cylindrical glass jars: Under the experimental conditions, the highest Ct product was found at a depth of 4 cm, where only 32% of *S. granarius* were killed, as against 100% mortality of *T. confusum*. Beyond that depth, only partial kills were obtained.

TABLE 5

Comparison of adult *Sitophilus granarius* and *Tribolium confusum* mortalities at different depths of flour fumigated with HCN for 24 h with a Ct product of 12.6 and 14.2 g·h m^{-3} at 20ºC

		S. granarius mortality (%)		*T. confusum* mortality (%)	
Dosage in g m^{-3}		5	6	5	6
Ct product in g·h m^{-3}		12.6	14.2	12.6	14.2
Depth in cm	4	28	32	66	100
	8	8	14	24	63

	12	2	2	6	35
	16	0	6	6	12
	20	0	2	10	6
Control		2	2	0	0

Cylindrical glass jar treatment in a 1 m^3 chamber: The results given in Table 6 show that the initial dosages giving Ct products of 62 g·h·m^{-3} and 105 g·h·m^{-3} were not sufficient to control *S. granarius* even at a depth of 5 cm within the flour. However, for *T. confusum*, complete control was achieved up to depths of 10 and 15 cm respectively.

TABLE 6

Comparison of adult *Sitophilus granarius* and *Tribolium confusum* mortalities at different depths in flour fumigated with HCN in a 1 m^3 chamber for 24 h at Ct products of 62 and 105 g.h m^{-3} at 20ºC

		S. granarius mortality %		*T. confusum* mortality %	
Dosage in g m^{-3}		2.5	5	2.5	5
Ct product in g.h m^{-3}		62	105	62	105
Depth in cm	0	0	1	2	5
	5	**26**	**84**	100	100
	10	19	68	**100**	100
	15	15	32	76	**100**
	20	10	22	37	80
	30	2	8	7	33

These findings reveal that fumigation with HCN of flour mills and in the food industry, presents a constraint not found in the use of MB. Because of the poor penetration of HCN into flour where many insects reside, as much residual flour and farinaceous material as possible should be removed before the fumigation is carried out.

DISCUSSION

HCN is a good alternative to MB for structural fumigations. Some major insect pests of flour mills and food manufacturing premises, such as the flour beetles *T. confusum* and *T. castaneum*, and the Indianmeal moth *P. interpunctella* are very sensitive to this gas.

Our findings on the sensitivities of the eggs, young larvae, old larvae, pupae and adults of each species to HCN show that the Ct product required to obtain an LD_{90}

varies according to species and stage, from 1 to 4 g·h·m^{-3}. This is very low in comparison with corresponding Ct product values for MB. However, in contrast to MB, large differences were found between the species in sensitivity to HCN as was shown in the high tolerance of *S. granarius* to HCN.

The poor penetration of HCN into flour is due to considerable sorption. As a result, Ct products as high as 60 and 100 g·h·m^{-3} are required to be effective against *T. confusum* buried at depths of 10 and 15 cm in flour respectively, while *S. granarius* adults are not controlled at these Ct products.

In practice, the Ct products needed to kill any stages of the major mill pests should be around 10 g·h·m^{-3}, but to obtain HCN penetration to a depth of about 10 cm in flour-heaps in order to kill residual infestations, the Ct product should be around 60 g·h·m^{-3}. Consequently, the corresponding initial dose could be 5 g·m^{-3} taking into account the presence of minor leaks.

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