Donahaye, E.J., Navarro, S. and Leesch J.G. [Eds.] (2001) Proc. Int. Conf. Controlled Atmosphere and Fumigation in Stored Products, Fresno, CA. 29 Oct. - 3 Nov. 2000, Executive Printing Services, Clovis, CA, U.S.A pp. 781-787

TIME TO KILL PHOSPHINE RESISTANT *RHYZOPERTHA DOMINICA* USING A CONCENTRATION PROFILE TYPICAL OF FIELD EXPOSURE

P.C. ANNIS* AND HELEN A. DOWSETT
Stored Grain Research Laboratory, CSIRO Entomology, Canberra, ACT 2601, Australia
*[*e-mail: peter.annis@ento.csiro.au]*

ABSTRACT

A considerable number of studies have been carried out on the response of stored product pests to phosphine (PH_3). These usually involve constant concentrations for defined exposure periods. These studies show that the concentration by time product required to obtain a defined level of kill varies considerably with concentration (or exposure time). This kind of study is useful in setting dosage regimes for methods of application where a more or less constant concentration is maintained. However, in commercial PH_3 treatments (using PH_3 generated in situ from metal phosphide) the concentration builds up to a maximum over 1 to 3 days and then decays over the remaining exposure period. Many factors affect the concentration profile including the nature of the phosphide preparation, leakage, sorption, temperature and humidity. This makes defining the required dosage regime a complex process. The laboratory exposures reported here mimic various field concentrations. The results are used to judge the adequacy of particular regimes by observing the level of kill at various times through the exposure period. The response to the concentration by time product (Ct) at each time end point is then compared to that predicted with a constant concentration exposures. The relationship between (Ct) obtained in these two manners is discussed.

INTRODUCTION

Fumigation with phosphine (PH_3) is one of the major methods of insect control in stored products. Most PH_3 fumigation is carried out using a metal (usually aluminium) phosphide preparation that reacts with moisture to produce PH_3. This gives a concentration profile during the fumigation that consists of build up to a maximum and decay from that maximum. The actual shape of the profile depends on a range of parameters including: distribution, sealing, temperature, nature of the preparation, and sorption. The variation of average PH_3 concentration is to a large extent predictable especially in a more or less sealed structure (Annis and Banks, 1993). The precise effect of this concentration profile on insect survival is largely unknown (Reichmuth, 1985).

A considerable number of studies have been carried out assessing the effects of constant concentrations for defined exposure periods, Fig. 1 (Annis these

proceedings). On the basis of these observations it is obvious that the concentration by time product (Ct) required to kill insects varies considerably with concentration (or exposure time), Fig. 2. This data is useful in setting dosage regimes in treatments where a more or less constant concentration is maintained, however, the data has been used widely as a basis for phosphide use where the concentration is far from constant. The treatment rates derived from this data have been widely used and are normally successful although the extrapolation from fixed concentration to a varying one remains questionable.

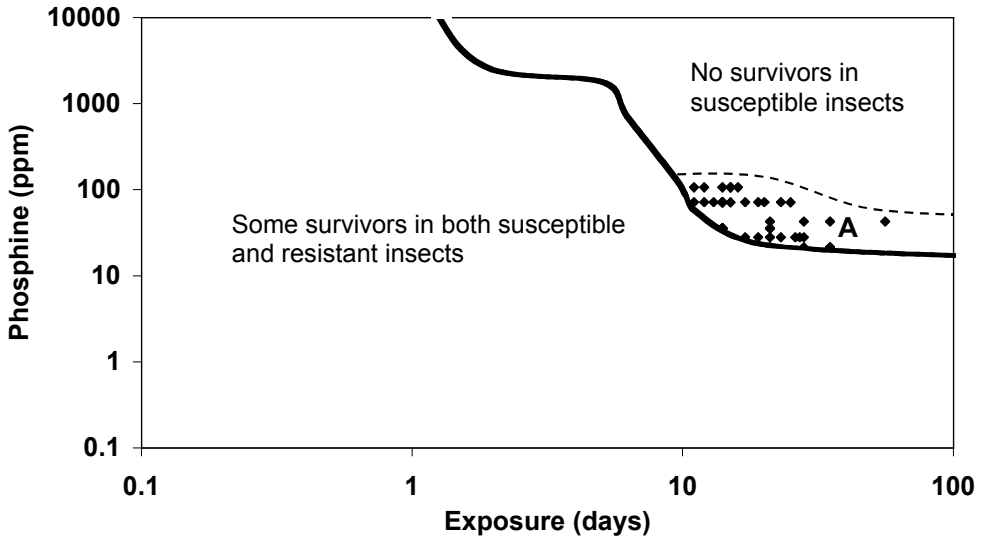

Fig. 1. Summary of collected phosphine mortality data. Data from any stage of 49 species of insects associated with stored products and exposures at >15ºC, but excludes data from diapausing Trogoderma spp. The region marked **A** is a range of concentration/time combination that are no longer assured of giving control due largely to resistant *Rhyzopertha dominica*, are individual observations with some survival.

The recent occurrence in Australia of populations of *Rhyzopertha dominica* that are significantly resistant to low constant concentrations of PH_3 (Collins these proceedings) has meant that Australian recommendations for phosphide application rates may also require re-assessment.

The usable exposure range lost to constant concentration of PH_3 treatment due to the development of strong resistance in *R. dominica* is indicated in Fig. 1. The study reported in this paper was designed to provide a quick answer to the question, "Does this loss of efficacy at constant low PH_3 concentration make the current phosphide application rates obsolete?"

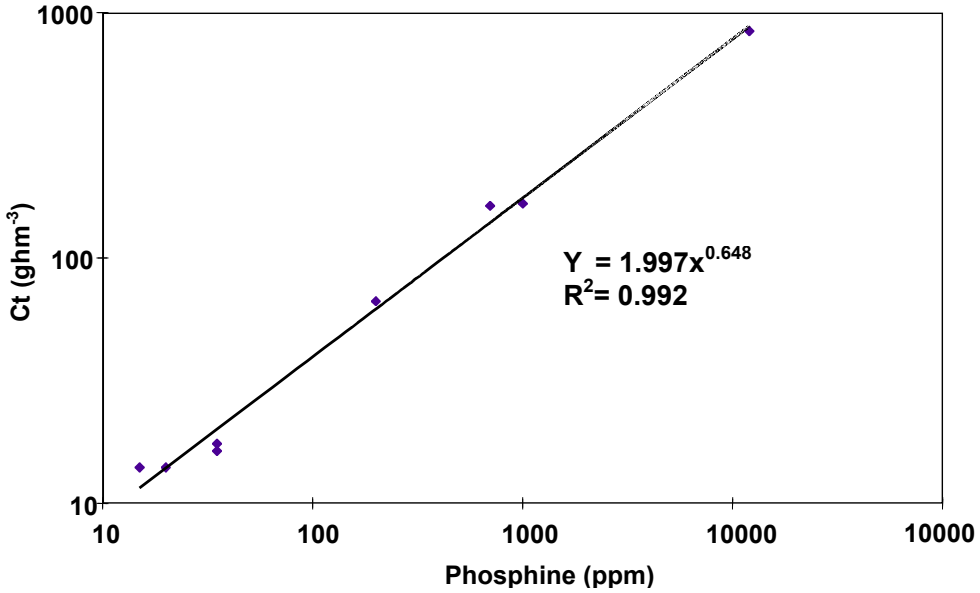

Fig. 2. Concentration by time product (Ct) required for complete kill of non-tolerant species over a range of phosphine concentrations. Data derived from approximate survival/no survival boundary in Fig. 1.

METHOD

Cultures of mixed stages of a resistant strain of *R. dominica*, from Millmerran in Queensland, were exposed to a simulated PH_3 concentration profile that is consistent with a well executed fumigation using aluminium phosphide preparations (Fig. 3). This profile was produced by the controlled dilution of PH_3 in a specially designed apparatus (Fig. 4). Relative humidity and temperature of the exposed cultures were kept at 65% r.h. and 25ºC respectively. Cultures were removed from the apparatus at daily intervals from 1 to 10 days. After exposure, the whole cultures (both tests and controls) were kept at 65% r.h. and 25ºC and sieved weekly for 10 weeks. The cumulative number of emergent adults was recorded.

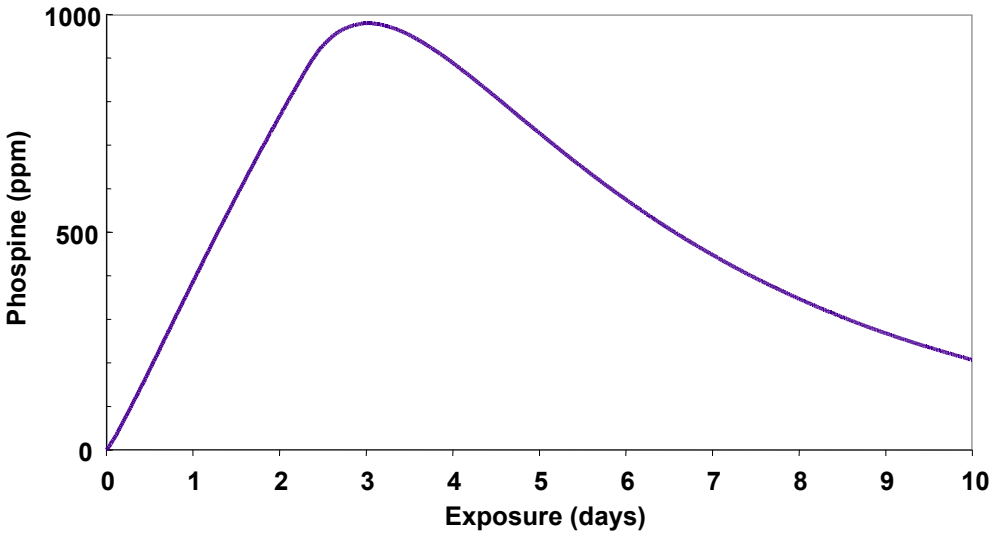

Fig. 3. Phosphine concentration profile used in this study. Equivalent to the average concentration for an aluminium phosphide treatment with: a moderately sorptive product; a full storage; 1.5 g m^{-3} application rate of phosphine, gas interchange rates of 0.25 day^{-1}; grain at 12% mc and 25ºC.

RESULTS

There was a progressive decrease in survival from day 1 to day 4. No adult insects emerged with any exposure longer than 5 days, and there was a sharp cut-off in survivors between 4 and 5 days treatment (Table 1). By 6 days the cumulated Ct was 136 g·h□m^{-3} and the average concentration to that time was 614 ppm.

The pattern of emergence (peak emergence after 4 weeks) suggested that survival at marginal exposures (4 and 5 days) most probably occurred during the earlier developmental stages rather than adults, pupae, or late larvae.

DISCUSSION

The primary purpose of this work was to investigate the applicability of current Australian phosphide application rates to PH_3 resistant *R. dominica*. The results clearly show that properly conducted aluminium phosphide fumigation in a sealed storage at the Australian phosphide label dose of 1.5 g□m^{-3} for 10 days is more than adequate for control of this strain. This is significant because this strain is highly resistant, on an exposure time basis, to lower PH_3 concentrations (>15□days and <120 ppm). This resistance is such that with a constant concentration of <120 ppm PH_3 will not control this strain.

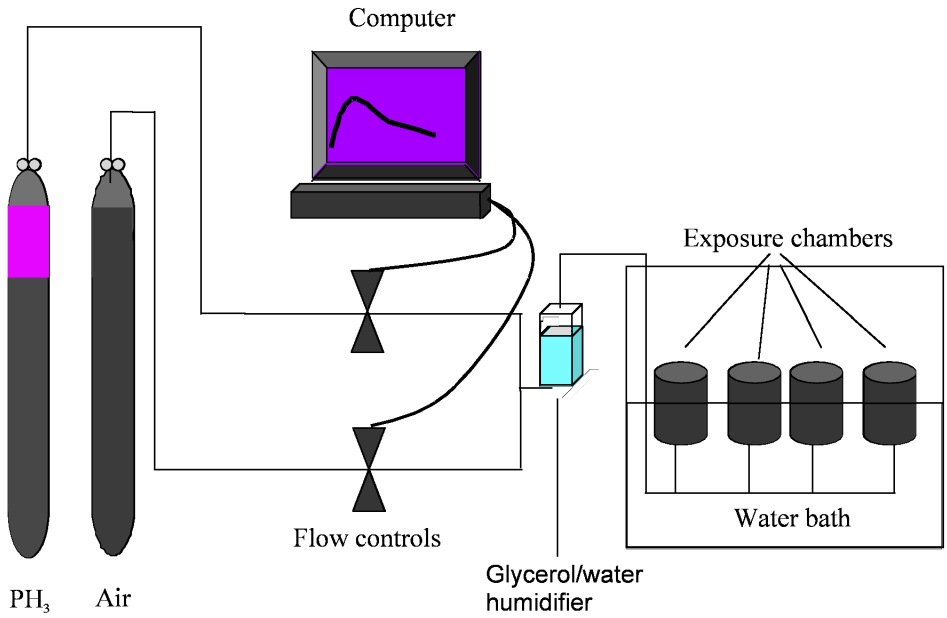

Fig. 4. A laboratory apparatus designed to expose insects to a phosphine concentration profile typical of that observed in phosphide fumigations.

TABLE 1
Combined results of replicate exposure to whole cultures of *Rhyzopertha dominica* to the typical field concentration profile shown in Fig. 2

Exposure (days)	Cumulative number of live adults from treatments	Number of live adults from equivalent controls	Average phosphine concentration to this time (ppm)	Ct (g·h□n^{-3})
1	296	380	175	6.2
2	167	380	367	25.6
3	>200	566	520	55.8
4	258	566	596	87.4
5	1	566	617	114.4
6	0	180	614	136.3
7	0	180	589	152.9
8	0	180	558	166.3
9	0	180	527	176.5
10	0	180	497	184.4

The results also give some insight to the use of Ct as a measure of effective dose where the concentration varies with time. Complete mortality was obtained with an accumulated Ct of between 114 and 136 g·h□m^{-3} (Table 1). This is equivalent to the Ct required with a constant concentration of 513–614 ppm (Fig.□2). This value contrasts with other possible ways of estimating the Ct required (Table 2) using a combination of the data in Fig. 2 and the tested concentration profile in Fig. 3.

TABLE 2
Comparison of true Ct (by integration of concentration by time curve) and Ct accumulated at time of complete mortality, calculated on a range of different assumptions about the effective concentration

Basis of calculation	Assumed value of concentration	Calculated Ct (g·h m^{-3})	Ct required at assumed concentration
True values		136	
Applied dose	1.5g m^{-3} (storage volume)	216	184
Highest concentration	980 ppm	197	173
Concentration at time of complete kill	650 ppm	130	133
Minimum conc. giving mortality in this strain, i.e. most effective Ct (Fig 2)	120 ppm	24	44
Average concentration	614 ppm	123	128

The current study is based on a single concentration profile. However, it is clear from the results that Ct calculated from applied dose, end concentration, or maximum concentration give poor estimates of the Ct required to give a high level of kill in an aluminium phosphide treatment.

CONCLUSIONS

Although the current study is based on one concentration profile, the following limited conclusions are possible:

When fumigation is carried out in a sealed storage for the required time, the standard phosphide dosage used in Australia (1.5g□m^{-3}) is adequate for the most resistant insects so far found in Australia.

Neither the end concentration nor the maximum concentration gives a good estimate of the required Ct for disinfestation by an aluminium phosphide treatment.

Other profiles representing different fumigation regimes are required before more understanding is possible.

ACKNOWLEDGEMENTS

Pat Collins of QDPI first identified resistance in, and supplied, the *R. dominica* used in this study. Financial support from the Participants to the Stored Grain Research Laboratory is appreciated.

REFERENCES

Annis, P.C. and Banks H.J. (1993) A predictive model for phosphine concentration in grain storage structures. In: *Proc. Int. Conf. on Controlled Atmosphere and Fumigation in Stored Products*, (Edited by Donahaye, E.J., Navarro, S. and Varnava, A.), 21-26 April 1996, Printco Ltd., Nicosia, Cyprus, 299-312.

Annis, P.C. (2001) Phosphine dosage regimes required for high mortality: a data-base approach, (these proceedings).

Collins, P.J. and Daglish, G.J. (2001) Combating the evolution of strong resistance to phosphine in Australia, (these proceedings).

Reichmuth, Ch. (1985) Response of the granary weevil *Sitophilus granarius* (L.) to gradually increasing and decreasing phosphine concentrations. *Anz. Schädlingskde., Planzenschutz, Umweltschutz,* **58,** 10-16.