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Efficacy and Limitations of Phosphine "Spot-Fumigation" against Five Coleoptera Species of Stored Product Pests in Wheat in a Grain Store – Short Note

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Abstract

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Field validation of spot-fumigation with phosphine (PH_3) applied by a subcontracted pest-control company in a farm grain store infested by pests was conducted. Inside and outside of the fumigated grain spot, containers with adults of 5 species of coleopteran stored-product pests were regularly spaced. The beetle pests were the internally feeding *Sitophilus granarius* and *Rhyzopertha dominica,* and the externally feeding *Tribolium castaneum, Oryzaephilus surinamensis,* and *Cryptolestes ferrugineus*. A 100% mortality of all pest species inside the fumigated spot under the sheet used for the application was stated. The efficacy (maximal) sharply declined with the increasing distance from the fumigated spot: 50% mortality was observed at 5 m from the spot, 38% mortality at 10 m, and a mortality of 23% was observed at 15 m. Mortality was different among the species, and the most sensitive was *O. surinamensis,* whereas the most tolerant were *S. granarius*, *R. dominica,* and *T. castaneum.* Although an efficient fumigation within the spot was found, the efficacy was low in the grains surrounding the spot. The practical implications of the findings are discussed.

Keywords: stores; stored grain; pests; protection; insecticides; fumigation; PH_3; disporsal

The infestation of stored grain by pests leads to decrease of seed germination (Stejskal *et al.* 2014) and to contamination of the finished cereal products with allergenic arthropod filth and fragments (Stejskal & Hubert 2008; Trematerra *et al.* 2011). Insecticides are among the most commonly implemented tactics to keep pest populations suppressed under tolerable thresholds (Stejskal 2003) until the grain has cooled down to a safe storage temperature after harvest. Because of negative environmental effects, entire groups of pesticides with the most efficient active ingredients were deregistered in the past decade (e.g. dichlorvos and methyl bromide in EU). Increased pest resistance endangers the remaining insecticide groups. Because prospects are limited for the development and registration of new insecticide formulations, current research has focused on pest protection with the "old" insecticides and on establishing and validating practices to "slowdown" the evolution of resistance. Phosphine (PH_3) is a major pesticide fumigant for the control of stored grain pests, which is also endangered by increased resistance of these pests. For example, Opit *et al.* (2012) discovered high levels of resistance in several strains of major storage pests to PH_3 in the USA. The misuses, over-uses, and inadequate fumigation techniques (e.g. exposures at high temperatures in poorly sealed enclosures) likely contributed to the rapid evolution of pest resistance to PH_3 in many countries (Bell 2000; Phillips *et al.* 2001). Various PH_3 fumigation procedures were analysed to identify the risk factors that affect efficacy and resistance. This resulted in technological developments in air-tightness and fumigant circulation in industrial

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stores (elevators, silos, bins, ships, etc.). In addition to standard and mainstream procedures, minor and sometimes controversial fumigation procedures should be included in the research focus because they may be commonly used in practice. One of the methods is called "quick stored products disinfestation by PH_3 before processing" (Ducom *et al.* 2004). Another understudied minor technique used in grain stores and flour mills is called "spot-fumigation" (Monro 1969; Bond 1984). In the FAO fumigation manual, Bond (1984) described spot-fumigation as follows: "Treatment of localised areas in a grain mass is often a useful technique for dealing with incipient infestations. These spots are usually recognised and defined by a local rise in temperature. Liquid-type fumigants applied through tubes or aluminium phosphide (AlP) tablets are the best materials to use. In this type of work, the tendency is to under-dose". Ignatowicz (2010) stated that "spot treatments are zone oriented, (and) they are as small as possible because size is money". Fumigation is expensive, and as an alternative to whole store fumigation, the localised targeted treatment of the infested spots would save operational costs. Spot-fumigations are also more environmentally friendly than whole store/mill fumigations (e.g. Phillips *et al.* 2001; Aulicky *et al.* 2015) since they leave less air residues. Spot-fumigation may be attractive for farmers not only in underdeveloped countries but also in developed ones subjected to economic crises. Although spot-fumigation may be economically attractive, the use is associated with the risk of under dosing (Bond 1984) and the occurrence of sublethal doses of PH_3, which leads to the evolution of resistance. However, we were not able to find published information on the efficacy of spot treatment in a grain store. Therefore, we conducted a first-time practical field validation of the spot-fumigation of PH_3 executed by a subcontracted pest-control company in a farm grain store infested by pests. We focused on the efficacy of PH_3 not only within the fumigated spot, but also in the surrounding areas because pests tend to migrate intensively in the grain. Sinclair and Alder (1984) documented that pests migrate in and outside stored grain mass even when populations are low.

MATERIAL AND METHODS

Store, grain, spot fumigation. The studied store was a hangar-type flat store (length 50 m, width 18 m, height 7 m) that accommodated 2100 t of wheat with the layer 4 m in height. According to the farmer, only a local infestation of pests occurred in the stored grain, and he decided to fumigate that part of the grain before selling it. A subcontracted licensed pest control company fumigated the grain. Thus, the situation was a unique opportunity to evaluate the efficacy of spot-fumigation in a real-world routine application. The owner of the grain store allowed bioassays with the pests. However, the pest-control company did not allow measurements of the PH_3 concentrations during the fumigation because of safety concerns. In the absence of direct measurements of PH_3 concentration and distribution, the alternative method proposed and used by Arthur (2008) and Campbell *et al.* (2014) was followed. Bioassay containers with sensitive laboratory insects were used for mapping the distribution of insecticide

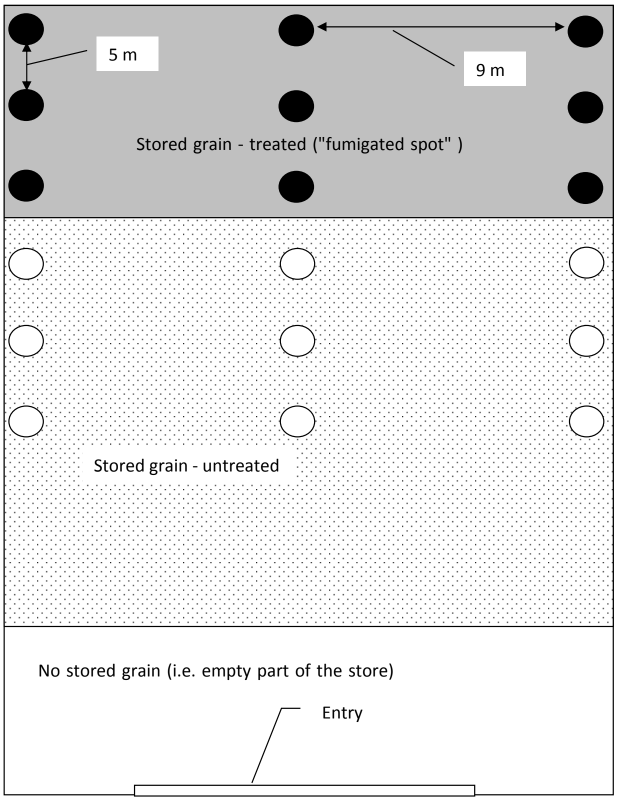

Figure 1. Layout of the flat grain store with sampling locations. One sample (circle) position contained 5 containers with pests located at 0.1 m depth of grain and 5 containers at 1 m depth of grain

Black circle – position of sample located inside fumigated spot; White circle – position of sample outside fumigated spot, Grey zone – fumigated spot of grain mass (560 t) under sheet; Dotted zone – grain mass (1540 t) neighbouring the fumigated spot

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in the store with a regular grid, and pest mortality was used as indirect measure of insecticide spatial distribution.

Fumigation procedure. Figure 1 shows the layout of the grain store and where the fumigation spot created a rectangular zone in the store. Fifteen pellets of PH_3 per 1 t of wheat were applied evenly 1–2 m into the profile using a special hollow metal rod applicator. Of the 2100 t of stored wheat grain, 560 t were treated and 1540 t remained untreated. After application, a plastic sheet covered the fumigated spot (Figure 1, the grey zone), and the grain was exposed to PH_3 for 10 days.

Bioassay and data evaluation. Five species of stored product pests were tested and represented the most important beetle pests in Czech flat stores (Stejskal *et al.* 2003). Internally feeding pests included the grain weevil (*Sitophilus granarius)* and the lesser grain borer (*Rhyzopertha dominica).* Externally feeding pests included the red flour beetle (*Tribolium castaneum*), the saw-toothed grain beetle (*Oryzaephilus surinamensis*), and the rust-red grain beetle (*Cryptolestes ferrugineus*). The beetles for the bioassays were from cultures of insecticide-sensitive strains kept at the Crop Research Institute, Prague. Twenty adults were enclosed in each plastic container, and the containers were made from plastic tubes with both sides covered with textile mesh (UHELON, polyamid, 9.5 × 139; Silk and Progress s.r.o., Brněnec, Czech Republic) to enable gas entrance and to prevent insects from escaping. One day before fumigation, the containers were inserted into the grain mass at two heights (0.1 m subsurface of the grain mass, and 1.0 m deep inside the grain mass). A regular grid both inside (Figure 1, black circles) and outside (Figure 1, white circles) the fumigated spot was created by the location of the containers in the grain mass. The next day (after 24 h), after removal of the sheet and ventilation of the grain store, the containers were transferred to the laboratory and assessed for pest mortality and knockdown. Ten-paired temperature and grain humidity measurements were collected, 5 samples from the subsurface (0.1 m) and 5 from the 1 m depth. The average temperature and humidity were 16.08 ± 0.10°C and 55.5 ± 0.18% for the subsurface layer and 20.6 ± 0.02°C and 55.1 ± 0.11% for the 1 m depth layer, respectively. The bioassay mortality data were not transformed and were evaluated (ANOVA and Tukey's HSD test) with the software STATISTICA CZ 7.0 (StatSoft, Inc., Tulsa, USA).

RESULTS

Inside the fumigated spot under the plastic sheet (Figure 1, black circles in the grey area), the mortality of all tested species was 100% irrespective of the location or depth in the grain (i.e. either 0.1 m or 1 m). By contrast, a differential rate of survival and low mortality was found (Table 1) in the zone neighbouring the fumigated spot (Figure 1, white circles in dotted area). Statistical analysis showed that the effect of species was significant at $P = 0.03$ ($F = 3.49$, $df = 2.83$). The most sensitive species to treatment outside the fumigated spot was *O. surina-*

Table 1. Efficacy of PH_3 (average percentage mortality + SE) at various distances (0–15 m) from the PH_3 fumigation spot on 5 species of pests located 1 m below and near (0.1 m) the grain surface

	0 m	5 m	10 m	15 m	Control
Located 1 m below					
Sitophilus granarius	100.00 ± 0.00	11.67 ± 3.33	3.33 ± 3.33	1.67 ± 1.67	0.00 ± 0.00
Rhizopertha. dominica	100.00 ± 0.00	1.67 ± 1.67	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Tribolium castaneum	100.00 ± 0.00	5.00 ± 2.89	1.67 ± 1.67	1.67 ± 1.67	0.00 ± 0.00
Oryzaephilus surinamensis	100.00 ± 0.00	50.00 ± 8.66	38.33 ± 10.14	10.00 ± 2.89	6.67 ± 1.67
Cryptolestes ferrugineus	100.00 ± 0.00	23.33 ± 13.02	10.00 ± 2.89	3.33 ± 3.33	0.00 ± 0.00
Located near 0.1 m					
Sitophilus. granarius	100.00 ± 0.00	10.00 ± 5.77	6.67 ± 3.33	1.67 ± 1.67	1.67 ± 1.67
Rhizopertha. dominica	100.00 ± 0.00	3.33 ± 1.67	1.67 ± 1.67	0.00 ± 0.00	1.67 ± 1.67
Tribolium castaneum	100.00 ± 0.00	1.67 ± 1.67	1.67 ± 1.67	1.67 ± 1.67	0.00 ± 0.00
Oryzaephilus surinamensis	100.00 ± 0.00	41.67 ± 19.65	28.33 ± 6.67	23.33 ± 11.67	6.67 ± 3.33
Cryptolestes ferrugineus	100.00 ± 0.00	33.33 ± 25.87	3.33 ± 3.33	3.33 ± 1.67	0.00 ± 0.00

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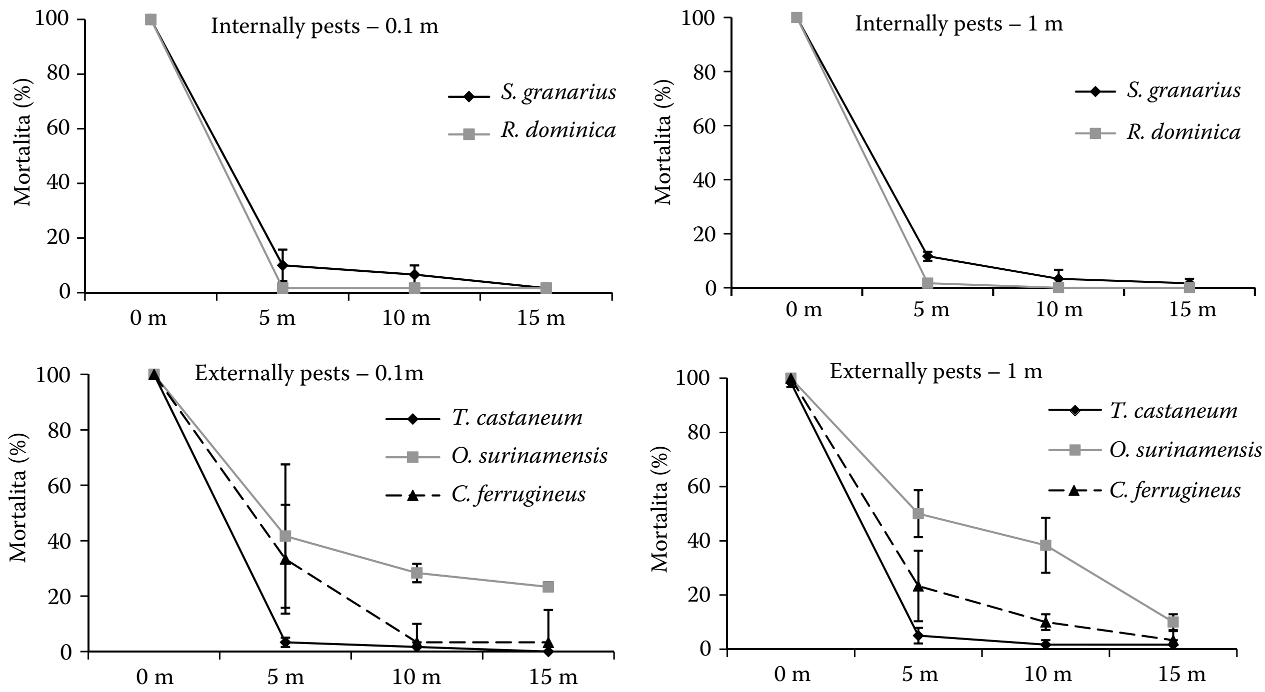

Figure 3. Decline of PH_3 efficacy (expressed as % pest mortality) with the increasing distance (5, 10, and 15 m) outside the fumigated spot (0 m represent average mortality of 100% inside the fumigated spot) for internally feeding pests (grain weevil – *Sitophilus granarius*, lesser grain borer – *Rhyzopertha dominica*) and externally feeding pests (red flour beetle – *Tribolium castaneum*; saw-toothed grain beetle – *Oryzaephilus surinamensis*, rust-red grain beetle – *Cryptolestes ferrugineus*) located at two depths inside the grain (0.1 m and 1 m)

mensis, whereas the most tolerant were *S. granarius, R. dominica,* and *T. castaneum.* The effect of distance was also significant at $P = 0.01$ ($F = 586.61$, $df = 4.83$). Figure 2 demonstrate clearly that PH_3 fumigation efficacy declined sharply with increased distance from the fumigation spot (the maximalmortality was 50%, at 10 m mortality was 38.3%, and at 15 m mortality was 23.3%). The mortality pattern was the same for both depths. On average, the PH_3 efficacy decreased more rapidly for internally than for externally feeding pests. The average values must be considered carefully because the externally feeding *T. castaneum* was also pesticide tolerant.

DISCUSSION

Both farmers and researchers (e.g. Sinha & Wallace 1966) have long noticed that stored grain pests do not occur uniformly but are found in aggregations in which the population buildup is extremely rapid because of the production of heat (Mani *et al.* 2001; Athanassiou *et al.* 2011). To treat such local zones, spot-fumigation techniques were proposed (Bond 1994) but were not validated with data under practical conditions. In this field case study, we found that spot-fumigation was 100% efficient on the sensitive strains of pests in the fumigated grain spot. Ducom *et al.* (2007) found that in the absence of highly resistant strains (if present, the treatment may fail), PH_3 required only a few hours to kill the active and visible stages, even at low temperatures and low concentrations. With these findings (Ducom *et al.* 2007), our results of high PH_3 efficacy inside the fumigated spot were not surprising. The more interesting result was the finding that the efficacy of PH_3 decreased dramatically even within the short distance of 5 m from the fumigated spot. Barker (1974b) found that although PH_3 spread effectively through the grain mass (with a velocity of 0.49 cm/min), it created a pronounced concentration gradient from the point of introduction to the point of leakage in the top of the treated steel drum. According to Barker (1974a), the PH_3 leakage rate from the treated grain is 0.049 g/cm^2/s. Even without leakage, the gas concentration is diluted when it enters the untreated grain mass because of intergranular air. Air space represents a significant proportion (36–49% accord-

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ing to Barker 1974a) of the stored wheat volume. A further decrease in PH_3 concentration is caused by commodity absorption (Reed & Pan 2000). Generally, the combination of these three factors was responsible for the decrease in concentration with the increase in distance from the point of gas introduction into grain. Because a rapid decrease in the efficacy of PH_3 occurred at a short spatial scale outside the fumigated spot in our field study, we postulate that the method of spot fumigation can cause sufficient mortality only in situations where (*i*) it is possible to precisely locate all the pest aggregations and where (*ii*) pest emigration/dispersal from the infested spots into the areas surrounding the fumigated spot cannot occur. The fulfillment of these two conditions is the weakest point for the safe and practical use of this method. The evidence is accumulating that it is not easy to precisely detect all pest locations either because of low efficacy of traps (e.g. Stejskal 1995) or differential sensitivity of various detection methods and sampling programs (e.g. Stejskal *et al.* 2008; Athanassiou *et al.* 2011; Jian *et al.* 2014).

CONCLUSION

Spot-fumigation can work well inside the treated spot under conditions similar to those described by Ducom *et al*. (2007) in their operational methods "Quick stored products disinfestation by PH_3 before processing". These conditions include a sensitive (non-resistant) pest strain, sensitive stadia, sheeting, proper exposure time, and high grain temperatures. However, because of inherent uncertainty of precise detection (Jian *et al*. 2014) and targeting of all pest spots in the grain mass (especially at low pest populations), the method is probably not very robust. Mistargeting will result in untreated spots with pests, and infestation resurgence can be expected. Another potential problem associated with this method is that the emigrated (Sinclair & Alder 1984) and undetected pests in the vicinity of fumigated spots are treated inefficiently, and such exposure to low or sublethal gas doses poses a risk (Phillips *et al*. 2001) that the most sensitive individuals from the population are eliminated, which will lead to an increase in PH_3 resistance.

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