## EFFECT OF AIRTIGHT STORAGE ON INSECT PESTS OF STORED PRODUCTS

N.J. BURRELL

### 1 INTRODUCTION

Many workers have shown that removal of oxygen by respiration causes insects to die in a sealed container (Dendy and Elkington, 1920). Thus over a period of 14 days at temperatures of  $29^{\circ}$  to  $30^{\circ}$ C, Bailey (1955, 1956, 1957, 1965) found a concentration of oxygen below about 2%, killed six important species of grain infesting beetles. Since, however, the susceptibilities of the various adult and immature stages were not equally affected, it seems likely that when a mixed population of insects is sealed into an airtight bin, some species will be eliminated sooner than others and some might even survive. In practice also there is some risk of oxygen diffusing into containers and permitting some survival (Oxley and Wickenden, 1963). In any event, low temperatures prolong insect life and so delay the process of disinfestation.

The test described here was set up at short notice, when a farm infestation was discovered, to find if butyl-rubber airtight grain storage bins could be used in practical farm conditions in the U.K. to destroy an existing infestation in grain.

## 2 METHODS

## 2.1 Farm store

The investigation was carried out between March and July using 40 tonnes of heavily infested barley of 15.5-16% moisture content from a farm store in which a temperature of 42°C was recorded and in consequence seeds were sprouting on the surface in the centre of the bulk and some 0.5 tonnes was visibly mouldy. The badly damaged barley was destroyed and the rest was transferred into two airtight bins. The five abundant species of insects in the barley bulk were <u>Oryzaephilus surinamensis</u> L., <u>Sitophilus granarius</u> L., <u>Cryptolestes ferrugineus</u> Steph., Ahasverus advena Waltl. and <u>Typhaea stercorea</u> L.

## 2.2 Airtight bins

The two airtight bins, each consisted of a cylinder of welded-steel mesh and a cylindrical lining bag made of black butyl-rubber 0.75 mm thick which could be sealed at the top. These were erected in the open about 7 m from the farm store. Bin 1 was 4 m in diameter and 2.7 m high; Bin 2, was 3.4 m diameter and 3.4 m high. Twenty tonnes of barley, mainly from the heated core in the farm store including some of the damp barley from near the surface, was placed in Bin 1 and 20 tonnes from the cooler periphery of the original farm bulk were placed in Bin 2. The grain was moved by shovel onto a rubber belt elevator into the bins.

The bins had already been used for storage and were slightly damaged and dirty. All holes found were patched but 3 weeks later when Bin 1 was inspected at the end of the test, prior to the final sampling, three further slits, 2-4 mm long, were found in the top sheet and were repaired after sampling the grain.

## 2.3 Temperatures

Three thermocouples were placed in a vertical row up the centre of the bins at distances of 1.0, 1.8 and 2.6 m above floor level and temperatures were measured during each weekly visit to the farm.

## 2.4 Gas sampling

Two rigid pvc air sampling tubes of 5 mm internal diameter were placed in each bin during loading, one at the centre and one in the small air space below the top sheet. The inner end of each tube was guarded by wire mesh to prevent blockage and the outer end was sealed with rubber tubing closed by a screw clip.

Air samples were drawn from the sealed bins, initially at daily intervals and later at weekly intervals, by a **dou**ble-action pump and stored in 30 ml metal cylinders at a pressure of 10-20 bars, for later analysis at the laboratory using a modified Haldane apparatus.

## 2.5 Sampling for insects and moisture content

Immediately after loading, before the bins were sealed, barley samples were taken from selected locations with a 200 g gravity-filled sampling spear. The sampling points were spaced vertically at intervals of 0.3 m below 7 points evenly spaced over the upper surface in Bin 1 and below 6 in the narrower Bin 2 yielding 56 samples from Bin 1 and 63 from Bin 2. Some samples were omitted from the sampling pattern where there was a possibility of displacing thermocouples or gas sampling tubes. Every sample was sieved to remove all free-living stages of insects but because the insects were so numerous, and counting was carried out under difficult conditions on the farm premises, only adults were counted. At the end of storage, because there were so many insects initially, the number of sampling points in the bulk was reduced by increasing the vertical spacing to 0.6 m. However, inspection of the bins before opening revealed three small punctures in Bin 1 so a sample was taken from the surface grain close to each puncture and further 1<sup>4</sup> surface samples were taken from each bin. These

56

samples were sieved to remove free living and dead adults and then all the samples from each depth from Bin 2 were collected together in lots of about 1.2 kg and kept in a room at 25°C. At intervals of 2-13 days thereafter, the barley samples were sieved and adults were removed, recorded and destroyed but all other debris sieved off was replaced.

Bin 1 was opened for insect sampling after 3 weeks and then closed for gas sampling for another 5 weeks until the grain in the bin was removed for feeding to cattle. Bin 2 was kept closed for gas sampling for as long as possible and sampled for insects after 15 weeks immediately before the grain was removed for cattle food.

## 2.6 Removal of oxygen from Bin 2

Five weeks after sealing, Bin 2 still had an oxygen concentration near 10%. Therefore, the bin was opened sufficiently for 100 kg of damp barley at 23% moisture content to be added on a polythene sheet and the bin was rapidly resealed.

## 3 RESULTS

## 3.1 Temperatures

Immediately after loading, the temperatures in the centre of Bin 1 varied from  $16.5^{\circ}$  to  $35^{\circ}$ C and those in Bin 2, from  $12^{\circ}$  to  $16^{\circ}$ C. The maximum temperature recorded in Bin 1 reached  $40^{\circ}$ C during the first day but fell to  $35^{\circ}$ C during the first week and to  $23^{\circ}$ C after three weeks when the bin was opened and sampled. No temperature below  $15^{\circ}$ C was recorded in this bin during these three weeks but the minimum temperature recorded reached  $13^{\circ}$ C by the time emptying was started.

Grain temperatures in Bin 2 scarcely changed during storage.

## 3.2 Gas analysis

The two oxygen concentrations obtained from each bin on each visit never differed by more than 1% so the means are presented in Fig. 1.

In Bin 1 the oxygen concentration fell to 4% in 2 days and reached a minimum of 0.3% after about 10 days (Fig. 1 upper). After 3 weeks, when the bin was opened for 6 hours for grain sampling, the oxygen concentration rose to nearly 15% but when the bin was resealed it began to fall steadily though more slowly than before. This is evidence that the bin still contained respiring organisms, possibly micro-organisms, in the patches of damp grain transferred from around the farm hot spot into the test bins.

The oxygen level in Bin 2 had fallen only to 10% in about 4.5 weeks and even when damp grain was added, it fell only slightly faster but it reached a minimum of 1% after 2 months and remained below 5% until the bin was opened after 15 weeks.



Fig. 1. Diagram showing changes in gas concentration with time and sampling occasions in airtight Bin 1 (upper) and Bin 2 (lower).

## 3.3 Insect numbers

Approximately 5000 live adult insects were removed from each bin in the initial samples indicating a population of about 8 million adults per bin. The initial numbers for each species were similar in both bins.

## Bin 1

The initial 56 samples contained a mean of 91 adults per sample (range 5-392) with all species spread fairly evenly throughout the bin. There were only 33 live adults in the final 38 samples and of these 26 were in the 13 samples from the upper surface, 17 being in the three samples taken close to the visible small punctures. This represents a survival of only 0.6%. The majority of these survivors were <u>C. ferrugineus</u>, which, unlike the other species present, had a high rate of survival near the punctures (Table 1.)

## Bin 2

The initial 63 samples each contained a mean of 78 live adults (range 16-255) again fairly evenly distributed though slightly more abundant in the top half of the bin (Table 2.) The final 51 samples contained a total of 10 live adults of which 7 were found in 21 samples from the upper surface although no obvious leaks were found. This represents a survival of only 0.2%. However, 13 <u>S. granarius</u>, five <u>C. ferrugineus</u> and one <u>O. surinamensis</u> emerged from the grain samples at  $25^{\circ}$ C (Table 3) and had survived exposure to concentrations of 1-4% oxygen and 15% CO<sub>2</sub> maintained during the last 8 weeks of the test. No A. advena or T. stercorea were found alive after airtight storage.

## TABLE 1

Number of live adult insects from Bin 1 samples

A. Before	airtight	storage	_					
Samples		Insect	0.s	C.f	<u>A.a</u>	S.g	T.s	
Position	Number	Numbers	s per samp	le				
Bulk	Index <th< td=""></th<>							
		Min	11	0	0	0	0	
		Mean	70	18	5	1	l	
Top surface	7	Max	101	10	2	3	3	
		Min	0	0	0	0	0	
		Mean	53	3	1	2	1	
Total	56	Total	3824	902	249	86	57	
B. After	airtight s	storage f	for 3 week	5				
Bulk	20	Max	0	2	0	l	0	
		Min	0	0	0	0	0	
		Mean	0	0.2	0	0.1	0	
Top surface	18	Max	1	7	0	1	0	
-		Min	0	0	0	0	0	
		Mean	0.1	1.2	0	0.1	0	
Total	38	Total	2	27	0	4	0	

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A. Before	airtight	storage						_	
Samples		Insect	0.ș	C.f	A.a	S.g	T.8		
Position	Number	Numbers	per sample	•					
Bulk	58	Max Min Mean	232 13 60	45 0 12	14 0 2	4 0 0.8	10 0 1.3		
Top surface	5	Max Min Mean	143 30 71	18 1 9	10 0 4	1 0 0.2	3 0 1.4		
Total	63	Total	3876	768	165	45	83		

Numbers of live adult insects from Bin 2 samples

B. After airtight storage for  $3\frac{1}{2}$  months

Bulk	30	Max	1		0	0	0	
	-	Min	0	0	0	0	0	
		Mean	0.03	0.1	0	0	·0	
Top surface	21	Max	2	3	0	0	0	
-		Min	0	0	0	0	0	
		Mean	0.14	0.19	0	0	0	
Total	51	Total	4	6	0	0	0	

# TABLE 3

Breeding out tests at 25°C on bulk samples from Bin 2

Depth (m)	0	0.6	1.2	1.8	2.4	3.0		
Insect	Numbers per sample						Total	
0.s	0	0	0	0	0	1	1	
C.f	3	0	0	1	0	1	5	
S.g	Ō	3	8	1	1	0	13	

TABLE 2

#### 4. DISCUSSION

Although insufficient oxygen leaked into the small airspace at the top of Bin 1 to raise the concentration above that in the middle of the bin it was enough to permit the survival of a few adult insects for nearly 3 weeks, indicating that higher concentrations than those measured existed close to the punctures. The mortality of <u>C. ferrugineus</u> adults reached over 97% in the bulk but most of the survivors of this species were found in the vicinity of these leaks. Only 2 live <u>O. surinamensis</u> and 4 <u>S. granarius</u> were found after storage so mortality was almost complete. However there were sufficient survivors, particularly of <u>C. ferrugineus</u> to recolonise the bulk if oxygen and temperature later became suitable. The Bin 1 test was completed in March, exposing the insects around the periphery to low ambient temperatures which may have promoted their survival by reducing their activity.

Because oxygen remained above 5% for 2 months in Bin 2, final sampling was delayed till summer and in this bin,only 6 adult <u>C. ferrugineus</u> and 4 adult <u>O. surinamensis</u> were found alive in 51 final samples. Despite the higher kill of adults in Bin 2, the breeding out tests showed that survival of the three granivorous species had occurred in the bulk.

The damp grain at 23% moisture content placed at the top of Bin 2 to absorb oxygen and speed disinfestation was estimated to be sufficient to produce an anaerobic atmosphere in the bin within a week under the prevailing temperature conditions assuming a dry matter loss near 0.01% per day (Burrell, 1974, pp. 424-7) and assuming that gaseous diffusion was not a limiting factor. Because deoxygenation occurred more slowly than expected, a further test was set up at the laboratory in a similar butyl-rubber bin, 3.4 m diameter, holding 10 tonnes of uninfested wheat 1.5 m deep at 15.3% moisture content. In this test 60 kg of grain wetted to 30% moisture, with an estimated dry matter loss near 0.1% per day, was spread in a layer 2.5 cm deep on a plastic sheet covering 35% of the upper grain surface and the bin was then sealed. The atmosphere at the top of the bin reached 2% oxygen in less than 2 days but took 8 days to reach the same oxygen concentration at the centre of the bin only 0.75 m below the top.

## 5 CONCLUSIONS

The procedure of sealing infested grain into an airtight bin to kill the insects when their own respiration has removed oxygen from the bin can be satisfactory for a heavy infestation in warm grain but is likely to prove uneconomic for light infestations or cool grain because of the long storage period required. Even when the conditions provide a sufficiently low concentration of oxygen it appears, in practice, that sufficient insects are likely to survive to provide an inoculum for reinfestation if the grain is removed from the bin.

The method, therefore, has some limitations and the replacement of oxygen by flushing out the air with  $CO_2$ , described in later papers, may prove more satisfactory. However, the use of fungal respiration or fermentation to achieve anaerobic conditions shows considerable promise, seems highly cost effective and merits further investigation.

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