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Physical Control of Insect Infestations

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1987 BCPC MONO. No. 37 STORED PRODUCTS PEST CONTROL

CONTROL OF STORAGE INSECTS BY PHYSICAL MEANS AND MODIFIED ENVIRONMENTAL CONDITIONS. FEASABILITY AND APPLICATIONS.

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ABSTRACT

The sanitary situation of stocks of foodstuffs and the lack of chemical control possibilities in many storage situations has emphasized the importance of physical methods and of modified environmental conditions in protecting stored products from insect pests. A great number of physical control measures are available for this purpose :

1 - For the disinfestation of grain or derivative products, high temperature fluidized bed has reached a pilot scale.

2 - Protection of stored dried fruits by keeping them inside refrigerated warehouses is used worldwide. Mobile refrigeration units for grain elevators or chilled air ventilation are used to stop the multiplication of insects in grain.

3 - Controlled and modified atmospheres are means of manipulating environmental conditions which have had the greatest development during the past ten years for grain storage.

4 - Non-ionizing electro-magnetic energy, microwaves or high frequency dielectric heating, can be used for dry foodstuffs disinfestation (preventive) and even for packaged goods.

5 - Big irradiation centres with ionizing radiation sources, radioisotopes or accelerated beams of electrons have been built in different parts of the world, principally for grain-insect sterilization.

6 - Mechanical handling or physical force have some applications in flour milling or semolina processing (entoleters, impactors, fluidized bed separators, etc.).

7 - Some marginal means of insect control also have a physical effect such as mineral salt admixture in grain or sound disturbance. It is emphasized that a lot of research is still required before the application of any of these physical techniques for pest control can progress into widespread use.

INTRODUCTION

The storage of grain and other food products must preserve the initial quality and avoid pest infestation at each stage of storage, processing and marketing. The disinfestation procedures now in use, i.e. fumigation or residual insecticide treatments require that grain remains undisturbed for several days in bins and may leave undesirable residues. In addition, these chemical methods of insect control cannot be applied on many stored foodstuffs and their use is limited to grain and empty warehouse disinfestation, at least in most developed countries. In recent years, one well-used fumigant, ethylene dibromide, was suspected of having carcinogenic effects and the residue tolerance level was fixed so low as to prevent its further use. Such preclusions can also affect some contact insecticides, thus highlighting the limited number of compounds available and problems associated with resistance in insect populations. Noxious effects on human health have also been reported with some chemicals (sulphates and sulphites are allergenic in some cases) which reinforces the care that must be taken when using traditional pesticides. The consumer is now aware of the consequences of immoderate use of chemical pesticides in the food chain.

"The shortcomings of agricultural pesticides (usable on grain) has renewed interest in the applications of physics to control insect pests" said Watters (1972) fifteen years ago. This is still true today. Physical means of disinfestation can avoid problems of residues with contact insecticides (and the onset of resistance) and are very useful to deal with insect contamination in processed foodstuffs. In these products, a great part of prevention and control of insect infestation has to rely on physical methods, thus excluding most chemicals.

Much literature is available concerning alternative methods of pest control. I cannot summarize all this work and my paper will be limited to the recent attempts to introduce physical methods for insect control that supplement or replace existing practices.

PHYSICAL METHODS USED FOR INSECT CONTROL IN STORED PRODUCTS TODAY

Thermal shock

A new approach to heat disinfestation of cereals and milling products using hot-air fluidized bed or fluid-lift has been taken in Australia and in France respectively (Dermott & Evans 1978, Evans *et al.* 1983, Fleurat Lessard 1984 & 1985). Pilot scale tests have shown that this process gives complete mortality of insects after a few minutes exposure to air temperature between 90 and 180°C. The temperature of the product during the insect-lethal exposure does not rise above 70°C, and is always followed by a rapid cooling. There is no adverse effect on baking quality of flour extracted from treated wheat or on the technological properties of durum-wheat semolina, provided the air temperature does not exceed 120°C. However, it was recently observed that high air temperatures are more efficient for insect control (Sutherland *et al.* 1986). The cost of heat disinfestation is comparable with that of fumigation. The convenience and uniformity of disinfestation are better than those obtained with insecticides.

The Australian plant for high speed thermal disinfestation of cereal grain has a throughput of 150 tonnes of wheat per hour (Fig. 1).

Refrigeration or fresh air ventilation of cereal grain

The use of moderately low temperatures to control infestations of storage pests was recommended about 20 years ago (Burrell & Burgess 1964, Navarro *et al.* 1969). Recent developments have shown that many grain elevators in mediterranean and subtropical countries can be equipped with refrigeration units to cool grain just after harvest. The temperature is lowered below the development temperature threshold of the main insect species in a few days, and grain temperatures of 10-14°C are maintained. A refrigeration unit is used for several bins in a elevator, each bin being refrigerated in turn for a few days.

In the temperate zone, night temperature, even in summer is sufficiently low to allow cooling with ambient air. Even if moist air is used, the effect of cooling is faster than the moistening of the bottom layer of grain as shown by Berhaut & Lasseran (1985). These authors, in pilot scale experiments run during the past three years, observed that by cooling down to 5°C or below, it is possible to prevent mould growth in grain with 15% moisture content during a one year storage period. The quality of the grain after one year storage with fresh air ventilation (Fig. 2) was slightly better than at harvest time, both for baking properties of wheat and the germinative capacity of barley (Berhaut & Lasseran 1985). The slow rate of grain cooling provided adequate time for the insects to acclimatize and survive but without multiplication. Though these refrigeration or cooling treatments do not kill the insects, they prevent multiplication and, generally if there is an infestation before the refrigeration, adults will try to escape from the grain bulk during cooling. No insecticide is needed and the cost of the treatment can be low (for ambient air ventilation), especially in cool climates.

Controlled and modified atmospheres

Gases like carbon dioxide, nitrogen and carbon monoxide can be used as alternative fumigants to phosphine. Fumigation of grain or other food commodities can be carried out with mixtures of these gases. Disinfestation using controlled-atmosphere can be achieved in a short period of time with pure carbon dioxide. When time is not a constraint and with shelled grain bulked in bins, controlled atmosphere treatment is a means of insect control that has a great potential for large-scale storages (Banks & Annis 1980, Mitsuda & Yamamoto 1980, Calderon & Navarro 1980). It is possible to have underground or underwater storages limiting the cost of the silo construction. There are two main types of treatment : in commercial practice with large airtight bins flushing with a burner gas with less than 1% oxygen and with about 12% carbon dioxide, is competitive with fumigation with only moderate additional expenses for the sealing of the warehouses or the bins (Fig. 3 & Banks 1984). In well sealed storage structures, the commodities could be disinfested with ready-made gas mixtures, generally with a high percentage of carbon dioxide (not below 40% in volume). Controlled atmosphere storage of grain can also be achieved using the natural production of carbon dioxide and the consumption of oxygen by the grain and insects. This method is used in developing countries in underground pits. Nevertheless, insects can survive for a long time before the atmospheric composition inside the pit becomes lethal. Therefore it seems better to stop insect activity using inert gas for the first purge.

Navarro & Calderon (1980) consider that the atmospheric gas composition is only one factor affecting the survival of insects in products under controlled atmospheres. The temperature and moisture of the grain, the leakage rate of the structure and the efficiency of the gas generator are all aspects that must be taken into account and that may affect the choice of treatment (Jay 1980). The problems related to the sorption of CO_2 on grain, the effects on the bins of pressure variations with temperature, and effects of gas composition are still under research. Field applications will undoubtedly need highly trained and competent staff if they are to be successful.

Non-ionizing electro-magnetic energy

Infestation problems encountered in the production of food and fibre materials and in the processing and storage of many of these products can be reduced by heating with high-frequency or microwave energy.

Three potential areas are covered by these non-ionizing, electro-magnetic waves in the field of disinfestation : stored grain insect control, treatment of plant seeds, disinfestation of dry or processed products.

Relative values for the electric field intensities were calculated for wheat and adults of the rice weevil, Sitophilus oryzae (L.) (Nelson & Charity 1972) and the power dissipation ratios for insects and grain were estimated. The most advantageous frequency range for selectively heating the insects was from about 10 to 100 MHz in the range of radio-frequencies or high-frequencies (H.F.). A pilot plant has been developed for grain disinfestation with H.F. dielectric heating (Hafner 1975) after a lot of experimental work at a laboratory scale (Grison & Martouret 1951, Bruel et al. 1960, Boulanger et al. 1969, Nelson 1972). The application of H.F. electric fields to the disinfestation of cereal derived products, dried plants, spices, pulses, biscuits, pet foods, etc, is possible and some ovens are used to treat food products on line before or after packaging (Brown Boveri 1983 & Fig. 4). The product was heated to a temperature level of 65°C before cooling by natural convection or by ventilation.

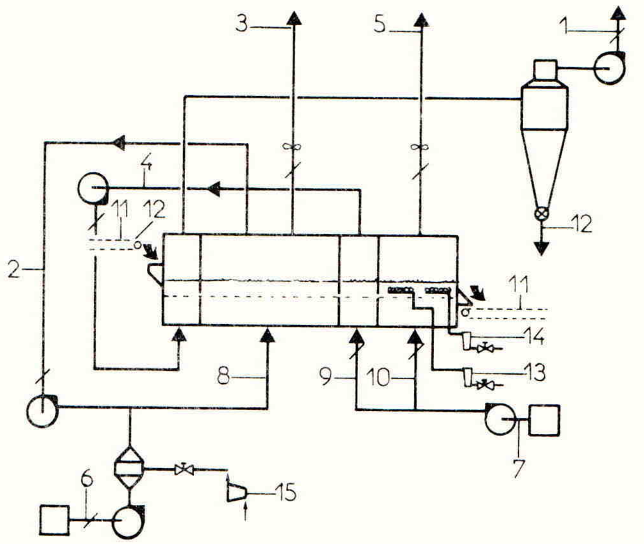
With microwaves, the frequency is in the range of 300-3000 MHz and the effects on the food product are slightly different than for H.F. energy. Liquid water is rapidly extracted from the product and temperatures reach a higher level for the same power intensity (Boulanger et al. 1969, Anglade et al. 1979). When the loss of water is not a problem or is even desirable, microwave heating and vacuum drying can be used (Elias 1979, Tilton & Wardell 1981, Fig. 5). The plant of MacDonnell Douglas in the U.S.A. can either dry grain or disinfest food products at temperatures of more than 100°C. However, for drying corn, the cost of the process has seemed prohibitive to several experts.

The high power requirements of these systems of insect control implies that they cannot be used for grain disinfestation, particularly at a normal flow rate in transit. Yet, with flour and baking derivatives, and other dried vegetables or seeds, H.F. heating is practical, easy to install on a process line, and can offer a complete disinfestation with a high security and automatic control.

Irradiation of stored grain and foodstuffs

Industrial interest for food preservation by ionizing radiation began after the Joint European Committee for Food Irradiation concluded that up to 10 kGy there was no risk of toxicity. Several products can now be treated by irradiation and some others are waiting for approval (Buscarlet, pers. comm.). The development of the process for the control of stored-product insects relies however on basic and applied research in technology, metrology and radiology. Insects can be killed directly with ionizing radiation or only sterilized if a lower dose is employed. The main grain species can survive up to 15 kRad (150 Gy) and Rhyzopertha dominica (F.) and Stegobium paniceum (L.) need about 300 to 400 Gy (Ratti & Cavalloro 1982).

Two different technologies are in competition for the irradiation of stored grain : Gamma facilities using cobalt irradiators and electron accelerators. The radioisotopic sources are generally built at a regional irradiation centre with all the security needed for such a system using the continuous emission of gamma rays. Each source can have



1 - separated air 2 - recirculation of warm air from the heating main chamber 3 & 5 - exhaust air (30%) 4 - warm air for preheating and dedusting chamber 6 - air for combustion in the burner (GPL) 7 - air for the cooling process 8 - air for the heating process 9 - air forced through warm grain for heat recovery 10 - air forced through the evaporative cooling chamber 11 - grain stream 12 - chaff and dust of grain 13 & 14 - cooling water streams for water spraying on the grain 15 - fuel injection.

Fig. 1 - Australian high temperature fluid-bed disinfestation system (plant scheme derived from Evans <u>et al</u>. 1983).

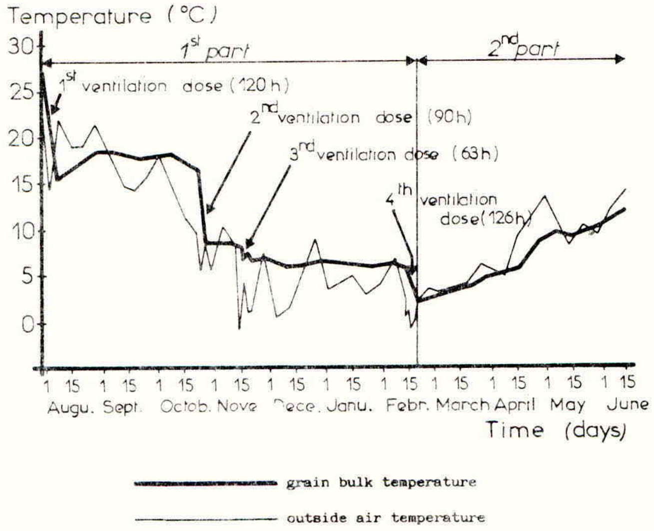


Fig. 2. - Temperature evolution curves of wheat and blown air during the period of storage monitored with the fresh air natural cooling system (after Berhaut & Lasseran 1985).

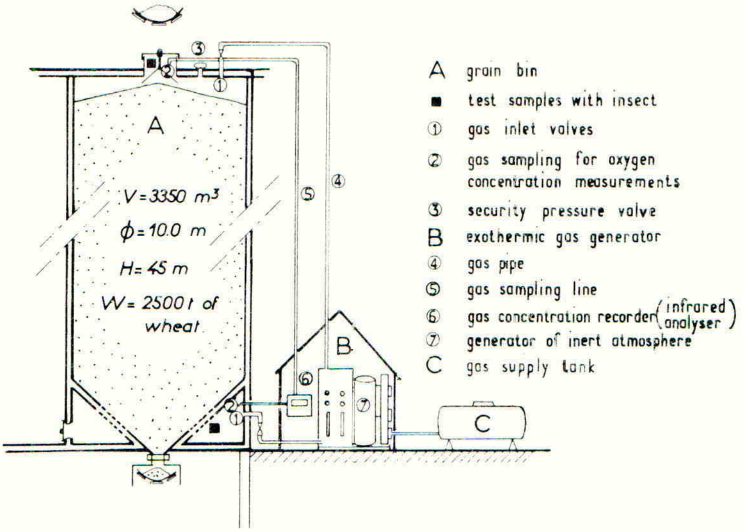
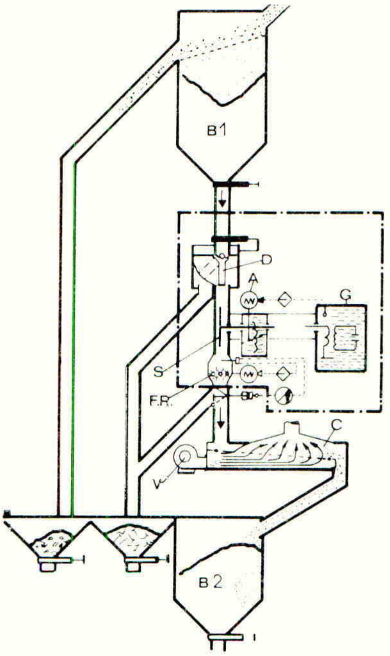


Fig. 3 - Wheat silo equipped with an exothermic inert gas generator (diagram of installation, Fleurat Lessard & Le Torc'h 1985).



A - adaptator of impedence
B1 - wheat bin for feeding the plant
B2 - disinfested-product bin
C - cooling system (fluid-bed)
D - derivative security system for metal particles
F.R. - product flow rate regulation
G - H.F. generator
V - ventilator
S - scanner.

Fig. 4. Pilot plant scheme for the disinfestation of grain and derivate products with high frequency electric fields (document Brown Boveri 1975)

1 - Wet grain in
2 - Product vacuum lock
3 - Drying chamber
4 - Waveguide
5 - Vacuum pump
6 - Condenser
7 - Dry grain out
8 - Product vacuum lock
9 - Power supply

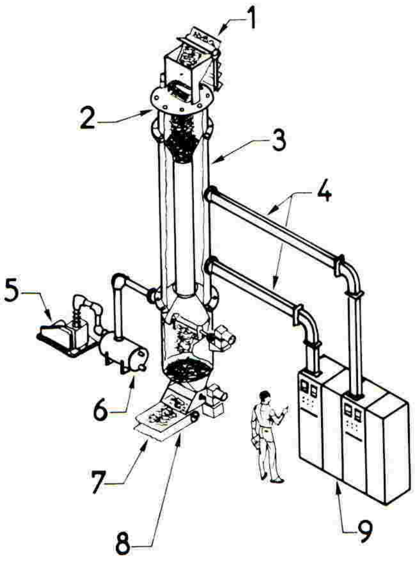


Fig. 5 - Combination of microwaves and vacuum for drying or disinfesting the cereals (after Elias 1979).

a capacity of several MCi and is principally used for disinfestation of fruits, and occasionally for spices or other processed food products on pallets. The technology of the electron accelerators has been improved more recently for industrial applications. The Soviet plant in Odessa harbour, designed only for grain irradiation, was set up in 1980. The linear accelerator of 1.4 MeV energy has a power of 20 kW. Grain is passed down a channel at a speed up to 6 m/s and spread in a layer of about 25 mm thick and 500 mm wide. It is treated with 200 Gy and complete kill of insects is obtained about one week after the irradiation (the survivors are sterilized). The flow rate of grain is 200 t/h in this plant. The reported cost is $0.1 U.S. per ton of wheat for the power input. Some accelerators were recently built for industrial or medical purposes in different countries but the disinfestation of grain remains a marginal use for these plants; the presence of living adults after a treatment renders the comparison with the other means of disinfestation very difficult. The plant requires a heavy investment which can exceed £1 million ($1.5 million approximately).

Mechanical handling, air separators and entoleters

Impact machines associated with sieves or air separators are extensively used in flour mills to prevent infestation in wheat and flour. The standards for the filth content of the flour must be satisfied and new systems of grain cleaning in flour mills are based on the separation of broken or infested kernels by sifters. Air separation is preferred to the old washing technique for cleaning the grain before milling. The devices operating on whole grain are named "impactors", concentrators, sterilators, etc. Entoleters are used only on the flour, and are less important for the control of infestation than the first separation of grain. However, the mill and the flour warehouse must be protected against reinfestation. In some situations where old buildings cause difficulties with a general fumigation, entoleters represent the only method to kill the insects that might be present in the finished flour. Entoleters can also be used for grain at a low speed (1400 rpm) before the milling process, (Banks, pers. com.).

Other marginal physical means of insect control in grain

Miscellaneous, physical or mechanical means were used in the past for insect disinfestation of foodstuffs.

With heat sensitive products, keeping them frozen over several days, at temperatures below minus 18°C is sometimes used. This is very suitable with high quality foods such as dried milk for babies or for high value products such as tobacco.

The use of sound energy has also been investigated although attempts to apply it on an industrial scale have met with failure, even with amplitudes of the order of 130 dB (Andrieu et al. 1978).

The use of partial or high vacuum on stored product insects has not proved very effective. Insects have a physiologically resistance to vacuum and the spiracles can be closed completely over several days, without any major adverse effect on reproduction following treatment.

The admixture of inert, hygroscopic or abrasive powders to grain is used in developing countries. Insects are sensitive to the loss of body water and this type of product, mineral salts for the main, increases the rate of water loss. This cannot be balanced by the insect which is killed by dessication. Some new means of relative humidity control in storage premises can also be used against the common insect pests in foodstuffs which cannot survive a long exposure to less than 50% R.H.

CONCLUSIONS

There is renewed interest in the application of physical control means against stored-product insect pests. The public concern about the hazards associated with the use of agricultural pesticides has intensified the research for alternative methods of control. The theoretical basis for such control measures were worked out many years ago but new developments and new technology make them even more appropriate.

The building of new plants which make use of the best technology available in a country have confirmed the effectiveness of some of the procedures such as accelerated-electron irradiation, controlled atmosphere storage or high frequency powered waves, for insect control in foodstuffs. However, when costs are taken into account each situation must be considered separately.

None of these methods used alone can, in all circumstances, give equal efficiency as insecticides or fumigants which have a much wider range of applications. Each physical method of control must be integrated carefully into pest control programs. Nevertheless, in the case of processed foodstuffs a great part of sanitation and product protection can be based on appropriate physical methods. A greater utilization of these will improve the quality of stored food products from the standpoint of freedom from infestation and lack of insecticide residues.

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CONTROLLING INSECTS BY COOLING GRAIN

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ABSTRACT

Recent research on ambient aeration has concentrated on methods of achieving temperatures near freezing from autumn to spring and the effects of these and humidity interactions on insect infestations. Infested bins without aeration heated due to insect activity whereas those which were aerated with a fan controlled by a time clock or a manually adjusted thermostat did not. However, low levels of three insect species survived in the latter throughout a 2 year experiment. There was a considerable improvement in the technique when a differential thermostat was used to control the fan. Lower temperatures were achieved more rapidly in the bulks and consequently there was a complete kill of *Oryzaephilus surinamensis* and *Sitophilus granarius*. Temperatures at the grain surface were not greatly affected and *Oryzaephilus surinamensis* survived there. Since this latter species appeared to migrate from aerated bins, surface or peripheral insecticide treatments would seem appropriate for infested, aerated bins. In the laboratory, dampening, cooling, drying and a high airflow appeared to cause it to disperse widely.

INTRODUCTION

Problems of moisture translocation

The main barrier to the successful storage of many commodities, even if they are relatively dry, is that they are harvested in autumn, when the weather is still warm. As the surface of the grain bulk cools in the winter, the moisture from the warm air rising from beneath by convection tends to condense there, increasing the moisture content (m.c.), allowing fungal and mite growth and permitting the grain to germinate. Before the advent of aeration as a means to cool grain it was traditional to turn the grain between bins to even out temperature gradients, but the cooling that occurred during this process was inadequate. Most storage thus depended on natural cooling and as the rate of cooling is a function of bulk size, managing larger bulks was a problem.

Cooling grain by aeration

The use of aeration for cooling grain was first developed to reduce problems of moisture translocation (e.g. Holman 1960) but its potential for producing low temperatures to combat arthropod pests has also been considered to some extent.

In Britain grain cooling is most economically carried out by the passage of air at the relatively low rate of about 10 m^3h^{-1} $tonne^{-1}$ through bulks of grain. Higher airflows may cool the grain more quickly but higher back pressures result, requiring larger fans, thus using more energy, which in turn costs more. The deposition of moisture from cold winter air also becomes a problem with higher airflows. It is a common misconception that aeration, as defined here, that is, "the passage of ambient air at relatively low flow rate" will also dry grain. It will not, since this technique does not use enough air to remove a significant amount of moisture. Using the same logic, it is also difficult to raise the overall m.c. of a bulk significantly.

THE CONTROL OF INSECTS USING AERATION

Recommendations

In Britain, Burges and Burrell (1964) made the first contribution to the use of aeration to control insect infestations. They defined 17°C as being the safe temperature for uninfested grain, based on Eastham and Seagrove's (1947) estimate that *Sitophilus granarius* L. would take more than 100 days to complete its life cycle at this temperature. However, they also suggested that temperatures down to 5°C were necessary, depending on the existing infestations and how long it takes to load the bin and cool it. This latter recommendation appears to have been overlooked so it is worth reproducing here (Table 1.)

TABLE 1

Temperature (°C) to which the warmest point in a grain store should be initially lowered to protect against insects (Burges and Burrell 1964).

Initial nos. of *S. granarius* per kg.	Infestation source (e.g.)	Loading and aeration completed in : (wks)		
		1	4	8
0.004	Fabric of stores, sacks, driers, conveyers, vehicles	17°C	17°C	17°C
0.04		17°C	17°C	12°C
0.44	Store previously infested	17°C	12°C	12°C
4.40	Infested bulks nearby	12°C	5°C	5°C

Today, because grain is stored for longer periods, the period of aeration needs to continue until temperatures of 5°C or 12°C are attained, if insects are to be prevented from breeding.

Large scale observations

Physical studies concerned with system design, air volume and temperature relationships, have determined the efficacy of the technique for lowering grain temperatures. Biological studies have also been useful in defining lower temperatures at which breeding ceases, and those which will kill pests. However, in order to determine the efficacy of aeration as a technique for preventing infestation, reducing or killing existing populations of pests, it is necessary to carry out large-scale practical tests under the fluctuating conditions which occur in grain stores. Considerable practical problems are associated with such tests and costs can be large. Convincing comparisons generally require an untreated bin (control) in which an infestation is allowed to build up. With grain being so valuable, this is not always easy to arrange. The detection of small numbers of insects in large bins by conventional methods is difficult and labour intensive. The apparent absence of insects from a cooled bin proves little since it may only reflect the scarcity of pests in that particular store.

Despite the problems of large scale tests, Burrell (1967) published details of 2 studies in which infested bins were cooled. In one, started in August, two 8 tonne bins were infested with 1 S. granarius L. per 4.4 kg and the bins were cooled from 18 to 3°C in 5 days, using refrigerated aeration. The results illustrated the difficulties inherent in such tests. In 5 monthly samplings, 64 kg of grain was sieved from 211 samples and produced only 1 insect. At the end of the test in March, 3 tonnes from each bin were sieved and only 7 live and 32 dead S. granarius were found of the 3,400 introduced. The missing insects must have either escaped from the bin or remained undetected in the unsampled 5 tonnes of grain in each bin.

In the other study, a 140 tonne bin was infested with O. surinamensis L. and cooling commenced in December. As the bin cooled insect numbers fell and proportionately the number of dead rose. Nevertheless, the insect density in some samples increased, suggesting some redistribution of pests was occurring (Table 2). At the end of the test the number of live and dead insects recovered was less than the initial numbers, suggesting that sampling was either missing pockets of infestation or some insects were leaving the bin.

TABLE 2.

Change in temperature and numbers of O. surinamensis before (16.12) and after cooling a heavily infested bin (n = 16) (Burrell 1967).

Date	16.12	8.2	28.2
No of live and dead/kg	292	178	185
% dead	-	34	41
max T °C	34.9	20.8	15.9
mean T °C	26.2	12.1	10.9
Samples increasing in insect density		5	8

In another test (Armitage & Burrell 1978), two 40 tonne bins infested with O. surinamensis were cooled using portable aeration spears. Numbers of live insects fell by about half in both bins after cooling (Table 3). Once again, despite the overall fall in insect numbers, the population in 24 out of 130 and 18 out of 59 samples from each bin respectively appeared to increase, suggesting some degree of re-distribution. Large numbers of insects were blown out of the bins by the aeration fans. However, these must have moved very close to the duct to experience air velocities sufficiently high to dislodge them (Armitage 1981).

Large scale experiments

Most of the preceding tests were large scale observations made when opportunity allowed examination of existing infestions. These were followed by large scale experiments making aerated/unaerated comparisons and using some replication.

The first of these experiments was started in 1974 (Armitage & Stables 1984). Four 30 tonne bins of wheat were set up and into the centre of each was placed a core of grain infested with O. surinamensis.

TABLE 3.

Temperatures and insect density before and after cooling 2 bins infested with *O. surinamensis*. (Armitage and Burrell 1978).

	Bin 1				Bin 2			
	Initial		Final		Initial		Final	
	max	mean	max	mean	max	mean	max	mean
Temperature °C	21.0	14.5	9.5	6.5	29.5	17.0	14.5	11.5
Insects/kg	990	165	475	73	2415	305	2500	145

TABLE 4.

A summary of changes in infested aerated (a) and unaerated (u) bins of wheat after storage (Armitage and Stables 1984).

Bin	I(a)	II(u)	III(a)	IV(u)
Length of test (months)	22	22	22	8
Time until heating (months)	-	14	-	3
Time below 5°C (months)	6	0	6	0
Total samples	45	45	45	14
No. infested	6	16	5	14
S. granarius/kg	1.4	6.1	0.2	7
O. surinamensis/kg	1.9	1.6	1.1	28
C. ferrugineus/kg	0.1	2.2	0.2	56

After a period in which the insects were allowed to establish, two of the bins were aerated and two were left unaerated.

The peak initial mean number of *O. surinamensis*/kg in Bins II-IV was 2, 2.7 and 4.7 respectively but in aerated Bin I it was 17/kg. Both unaerated bins heated, the aerated ones did not. However, *O. surinamensis* survived 2 winters in the aerated bins (as did the other species that were subsequently detected (Table 4), although numbers in the most heavily infested were reduced to one eighth. Unfortunately there was no way of telling how many insects in the aerated bin had invaded from the infested unaerated bins nearby, so the enigma of the survival of low levels of insects in aerated bins remained.

Heating of cooled grain by insects

In Burrell's (1967) account of aerating grain infested with *O. surinamensis* he mentioned that grain cooled from 37 to 15°C containing 125-450 insects/kg increased in temperature by 3°C in a fortnight in February when aeration was discontinued. In December, grain in the same position warmed from 24 to 37°C in 3 days when the fan was switched off. In Armitage and Stables' (1984) account an area of one bin containing 675 *S. granarius*/kg increased from 11-13°C in January to 18-21°C in less than a month. Thus, it is not sufficient to cool grain to a given temperature and then discontinue aeration.

The movement of insects in response to aeration
In the first large scale observations reported above the numbers of insects in aerated bins appeared to fall, but this could not always be accounted for by insect deaths. Similarly, while insect densities in some samples fell, in others increases occurred. Therefore, measurements were made to examine if more insects were escaping from the aerated than the unaerated bins in the large scale experiment started in 1974 (Armitage & Stables 1984).

TABLE 5.

Mean daily numbers of O. surinamensis trapped in grease bands (n = 5) around aerated (a) and unaerated (u) bins expressed as a percentage of those estimated by sampling (n = 14) to exist in the bins.

Bin	I (a)	II (u)	III (a)	IV (u)
No.	277	56	78	33
%	0.3	0.5	0.5	0.1

When comparison was made in December 1974 (Table 5), when the aerated bins were being cooled from 19-20°C to 11-12°C the raw data indicated that higher numbers were escaping from aerated than unaerated bins. However, escapes were proportionate to the density of the population; most insects were caught around the bins with the heaviest infestation. More insects continued to be caught around aerated than unaerated bins in 1976, when bait bags were used to catch them and more were caught in June than earlier or later in the year.

Although S. granarius and Cryptolestes ferrugineus (Steph.) were also present in the bins, few were trapped. This disinclination of S. granarius to move from the middle of the bins was also borne out by its distribution within the bin. Only rarely was it found at the surface while in contrast, O. surinamensis were often found there.

The problem of insect movement in response to aeration was further studied in the laboratory by comparing the distribution of insects in aerated and unaerated tubes 1 wk after approximately 100 adults had been introduced into the centre (Armitage et al. 1983). A high airflow or cooling or dampening or drying encouraged the dispersion of O. surinamensis. The example shown below demonstrates how O. surinamensis disperses more at the lower temperature than S. granarius which tended to stay in the middle of the tube (Table 6) although in this instance there was no difference between aerated and unaerated tubes.

Killing insects using aeration
The inherent inefficiency and inaccuracy of sampling large bulks of grain to find free-roaming insects and the uncertainty of inter-bin movements of insects means that, in order to determine if low temperatures in aerated bins kill insects, a more rigorous experimental approach must be applied. Accordingly the following regime was adopted.

TABLE 6.

Mean per cent of insects at the centre and ends of aerated (a) and unaerated (u) tubes (n = 3) of grain in an experiment to determine the effect of air movement (Armitage *et al*. 1983).

	Centre				Ends			
	O. surinamensis		*S. granarius*		*O. surinamensis*		*S. granarius*	
	a	u	a	u	a	u	a	u
10°C	4	7	53	53	88	81	29	17
25°C	10	18	66	51	63	55	6	20

Firstly, the expected length of survival of the insects at low temperature was deduced from the literature. The inter-author variation is very wide (Table 7) but explicable in terms of the conditioning of the pests. Normally, if insects are held at low temperatures above their freezing point they will survive for longer periods if the r.h. is high, or if they have been subjected to a gradual aclimatisation programme (Ushatinskaya 1954), or if they are receiving nourishment; for instance if the food is present as broken rather than intact grain (Granovsky and Mills 1982). In addition it is quite likely that there will be strain differences within species. (David *et al*. 1977).

The range of results obtained from the literature showed the importance of testing survival under practical large scale conditions and this was done in 3 successive years between 1982-5. Insects were caged and placed at the surface and centre of two aerated bins - one controlled manually, the other with an automatic differential thermostat. The *S. granarius* died out every year in both manually operated and automatically operated bins. Half the *O. surinamensis* survived at the bin surface but, in two out of three years they died at the centre of both aerated bins.

Parallel laboratory studies examined the survival of the two species after acclimatisation, which consisted of dropping the temperature 1°C per day, a rate similar to that occurring in October during the early stages of aeration. Tables 8 and 9 compare the time taken for half of the insects of both species to die in either the large scale or laboratory scale tests, estimated from lines drawn by eye through plots of percent mortality against time. Only at the centre of the 'differential' bins did the two species die quicker than at 6°C in the laboratory tests. For instance, in 1984-5, half the *O. surinamensis* there died in 114 days, which in laboratory tests occurred between +2 and 4°C.

The problem of surface infestations

The early large-scale aeration trials suggested that aeration could not normally prevent low level infestations surviving in aerated grain, despite some mortality and the migration of some species in and out of the bins. The latest test suggested some of the problems may lie in the surface grain which is subject to temperature and moisture fluctuations, and where mites can exist, even in dry grain. Elder and Ghally (1983) considered the surface of aerated bins to be the most likely place for insects to breed because it was the warmest, and Mathlein (1961) recorded

TABLE 7

The range in survival time (days) of O. surinamensis (O.s) and S. granarius (S.g.) at low temperature.

Temperature °C±1	10	5	3.5	2	0	-2	-5	-7
O. s.	48-209[a]	52[j]		9[b]	6.9[i]	25[c]	20[c]	
	60[b]				0.3[j]		15[c]	
	86[j]						2.5-5[i]	
S. g.	165->385[a]	105[c]	110[f]	38[g]	73[f]	40[c]	40-45[c]	28-33[c]
		178[d]		14-29[h]	85[c]	90[c]	20-75[k]	4[g]
		21>66[e]			30-100[k]	23[g]		

estimated from:

a Evans 1983
b Thomas & Shepherd 1940
c Mathlein 1961a, 1961b
d Granovsky & Mills 1984
e David et al 1977
f Cotton 1956
g Robinson 1956
h Solomon and Adamson 1955
i Obretenchev 1983
j Nawrot 1972
k Ushatinskaya 1954

TABLE 8.

The time taken (days) for 50% of insects to die at the surface(s) and centre (c) of two aerated bins in 3 years. (n = 6)

	S. granarius					O. surinamensis				
		Manual		Differential			Manual		Differential	
Year	Control (11°C)	S	C	S	C	Control (15°C)	S	C	S	C
82-3	*162	-	51	-	41	*210	*174	81	*170	66
83-4	>210	134	72	130	40	210	181	148	181	58
84-5	>203	116	96	91	72	197	>203	168	196	114

* extrapolated

enormous numbers of O. surinamensis at the surface of an aerated bin in March at temperatures of 5-10°C.

Aeration in other climates

In Israel (eg. Navarro et al. 1969) and Australia, aeration has been used against insects successfully for two decades, either with or without refrigeration. Lowest temperatures are expected to be in the region of 15°C, and although insects in these climates may even increase during aerated storage, the reduction in numbers, compared with those estimated to exist without cooling, was considered a great success. Aeration in warmer climes is also used to slow the breakdown of pesticides and may thus act to retard the development of resistant populations.

TABLE 9

The time taken (days) for 50% of insects to die at 70% r.h. and a range of low temperatures. (including acclimation time). (n = 6)

Temperature (°C)	+6	+4	+2	0	-2	-4	-6
S. granarius	86	77	75	74	61	48	60
O. surinamensis	139	118	108	95	60	47	42

IN CONCLUSION

This paper has outlined the achievements of practical large-scale observations and experiments in developing the use of cooling for the control of insect pests in grain. It is now possible to recommend cooling to 0-5°C by November and expect this to occur, providing correctly designed aeration systems are employed. It is also reasonable to expect this temperature to be maintained until Spring. The movement of beetles with (or without) aeration remains an enigma. Those moving within aerated bins might be controlled by boundary treatments of insecticides and further research may enable us to exploit the phenomenom in control strategies. The question of the response of mite populations to cooling regimes also needs to be taken into consideration.

It is clear that the potential of physical control by aeration has not yet been fully exhausted. It can present an alternative strategy to combat pesticide resistance, so limiting the amount of toxic chemical in the environment, and is irreplaceable for the prevention of moisture movement.

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APPLICATION OF MODIFIED ATMOSPHERES FOR CONTROLLING STORED GRAIN INSECTS

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ABSTRACT

Modified atmospheres (MAs) were investigated as a substitute for conventional stored grain insect control methods. The response of the four developmental stages of *Sitophilus oryzae* (L.), *Oryzaephilus surinamensis* (L.) and *Tribolium castaneum* (Herbst) to different MAs at temperatures from 15° to 32°C was studied. The pupae of all three species, plus the larvae of *S. oryzae*, were less susceptible than eggs and adults to the tested MAs. Exposure to 60 % CO_2 in air for 120 h at 27°C was sufficient to cause 100% mortality of all stages of *O. surinamensis*, but not of *S. oryzae* or *T. castaneum*.

A field trial was conducted to determine the feasibility of the method in a bin containing 6 881 tonnes of wheat, and the effect of the treatment on a natural population of stored product insects was studied. Mortality was correlated with the location of the insects in regard to temperature and movement of the CO_2 front from the bottom to the top of the treated bin.

INTRODUCTION

Grain in a postharvest situation is very close to the consumer and great care is taken by regulatory agencies when a new chemical treatment is submitted for approval. Most pesticide companies realize this and do not consider it worthwile to obtain registration for their products for postharvest use. When to this situation is added the residue problems (Berck 1974, Danse *et al*. 1984), the development of insect resistance (Bond 1983, Champ and Dyte 1976), health hazards, annual losses and the continued surveillance by regulatory agencies of the currently used pesticides, it is obvious that the control strategy for insect pests in post-harvest situations is in serious trouble.

An alternative technique available for insect control in postharvest situations is the application of modified atmospheres (MAs). This technique involves changing the proportions of the normal atmospheric constituents of the store, O_2, N_2 and CO_2, to create an atmosphere lethal to insects. The makeup of this MA must be maintained within set limits for an adequate length of time to achieve insect control.

The MA method of grain storage has great potential and it is remarkable that the exact limits of dosage and mortality for stored-product pests have been so inadequately defined. Work has concentrated largely on the adult stage, which appears to differ in susceptibility from the larvae, pupae and eggs which so far, have largely been ignored (Bailey and Banks 1980).

The time required to obtain a certain level of insect mortality upon exposure to a given atmospheric gas composition is dependent on the temperature of the environment (Jay 1984) and the moisture content of the commodity (Navarro 1978, Navarro and Calderon 1974). This effect is shown to be similar for different insect species, which differ in their susceptibility to a given gas concentration. In practice, these data should be considered very carefully. It is clear that at a given gas composition, much longer exposures will be needed to obtain effective control when low temperatures prevail in the grain bulk. Furthermore, since differences in temperature exist in the various parts of the grain bulk, the lowest temperature recorded should be that which determines the length of the exposure time required for the treatment. In the use of MA treatments, the most resistant stage of the most resistant insect species found in the grain bulk to be treated should be the determining consideration.

This paper presents the effects of different mixtures of the normal constituents of the atmosphere on different life stages of three major storage grain pests at several temperatures, and results of a field trial using CO_2 for the control of storage insects.

MATERIALS AND METHODS

Effects of modified atmospheres on insects

Sitophilus oryzae (L.) were reared on wheat, Oryzaephilus surinamensis (L.) on oat meal, and Tribolium castaneum (Herbst) on wheat feed mixed with 5% brewer's yeast at 26°C and 60-70% relative humidity (r.h.). For tests carried out with S. oryzae, wheat internally infested with immature insects was blended and then 25 g of this blend was exposed to the test atmospheres. The gas mixtures were obtained by preparing each mixture in a 3-liter container supplied with gases from pressurized cylinders of O_2, CO_2 and N_2, or from cylinders containing mixtures of CO_2 and air or low O_2 in N_2. These mixtures were passed into test chambers of 100 ml at a rate of 15 ml/min or into test chambers of 4 liters at a rate of 30 ml/min, after they had been conditioned to 55% r.h. by purging them through gas washing bottles containing appropriate solutions (Navarro and Donahaye 1972, Jay 1984).

The effectiveness of the treatment for immature S. oryzae was determined by dividing the total number of adult insects that emerged after the treatment by the total number that emerged in the controls and converting this to percent reduction in emergence (RIE) of adults. For all other insect species and their development stages, mortality counts were corrected by comparison with the relevant untreated groups (Abbott 1925). Each exposure, MA composition, and temperature was replicated three or four times, and the results are reported as means of each treatement.

Field application of CO_2

A field study was carried out in a 11 280 m^3 welded steel bin partially filled with 6 881 tonnes of soft red winter wheat. Gas sampling lines and thermocouples for temperature measurement were installed at different locations in the grain bulk. Gas samples for CO_2 concentration measurements were taken about 10 times a day for the duration of the trial. Grain samples of c. 900 ml were taken before and after treatment with a Prob-A-Vac$^{(R)}$ vacuum system at six sites in the bin and from depths of 1.1 to 9.1 m in the bulk. A total of 68 samples of grain were taken before treatment and 72 samples after treatment. The temperature of the grain near the top of the bulk ranged from 18° to 20°C and from 24° to 33°C in the center of the bulk. A series of constant pressure tests was carried out to determine the degree of gastightness of the bin. A comparative analysis based on this information revealed that the estimated air ingress rate was approximately 1% per day. The bin was purged from the bottom for 32.5 h and from the top for an additional 6.5 h at a rate of 426.7 kg CO_2/h (237.3 m^3 CO_2/h).

RESULTS AND DISCUSSION

Effects on *Sitophilus oryzae*

The results of (*a*) percent RIE of adults following exposure of immature stages and (*b*) mortality of adults, obtained at four temperatures and two atmospheres, are presented in Table 1. At 15°C the only stage affected by the tested MAs was the adult. The increase in temperature in the test environment resulted in a more rapid kill of adults and immature stages. At 32°C all of the tested MAs produced 100% adult mortality in less than 24 h.

TABLE 1

Percent reduction in emergence (RIE) of adult *Sitophilus oryzae* from eggs, larvae and pupae, and mortality of adults, exposed to two modified atmospheres at four different temperatures.

Atmosphere (%)			Temp.	Exposure time	% RIE of adults from treated -		% Mortality of treated adults
O_2	CO_2	N_2	(°C)	(h)	Eggs	Larvae & pupae	
1	0	99	15	24	0	7.2	27
				120	29.0	17.6	100
			21	24	0	3.2	57
				120	89.8	28.7	100
			27	24	53.3	15.3	96.6
				120	100	30.9	100
			32	24	43.9	16.6	100
				120	100	67.5	100
8	60	32	15	24	7.2	0	74
				120	36.2	35.5	100
			21	24	0	17.9	100
				120	100	91.6	100
			27	24	35.4	20.6	100
				120	100	98.5	100
			32	24	74.6	30.2	100
				120	100	98.5	100

However, at all the experimental temperatures none of the MAs produced 100% RIE of the treated larvae and pupae after 120 h exposure. These results show that the adults were the most sensitive stage, followed in decreasing order by eggs, larvae and pupae. Storey (1975) investigated the influence of the atmosphere (1% O_2; 8.5 - 11.5 CO_2; the balance being principally N_2) produced by an exothermic inert-atmosphere generator on different developmental stages of *S. oryzae* when exposed at 21° and 27°C. At both temperatures the earlier and later stages were more susceptible and the intermediate stages were less susceptible to the tested MAs. At 27°C, 95% mortality of the 4th larval instar through early pupal development occurred after about 10 days for *S. oryzae*. In the present study an attempt was made to increase the influence of the tested MAs by exposing the insects at 32°C. However, 120 h exposure even at 32°C was not sufficient to obtain complete control for larvae and pupae (Table 1). Between the two tested MAs, the mixture containing 60% CO_2 in air was the more promising, especially against the larvae and pupae.

Effects on *Tribolium castaneum*

The mortality of four developmental stages of *T. castaneum* exposed to two MAs at three different temperatures is shown in Table 2. At 15°C complete mortality was achieved for larvae and adults exposed to the 60% CO_2 atmosphere for 120 h. Although mortality studies on the effect of 60% CO_2 on eggs are not complete, the most sensitive stage to this atmosphere appears to be the adult. At 32°C complete mortality was obtained within 120 h for all developmental stages exposed to either atmospheric composition.

TABLE 2

Mortality of the four developmental stages of *Tribolium castaneum* exposed to two modified atmospheres at three different temperatures.

Atmosphere (%)			Temp. (°C)	Exposure time (h)	% Mortality			
O_2	CO_2	N_2			Eggs	Larvae	Pupae	Adults
1	0	99	15	24	20.8	1.5	1.1	1.0
				120	97.7	42.3	—*	22.4
			21	24	28.7	11.4	—*	0.0
				120	100	99.0	39.5	69.1
			32	24	88.8	55.1	40.5	100
				120	100	100	100	100
8	60	32	15	24	—*	1.3	0.9	23.8
				120	—*	100	28.1	100
			21	24	—*	6.3	11.6	100
				120	—*	100	99.5	100
			32	24	60.2	29.8	72.8	99.5
				120	100	100	100	100

* Data not available.

AliNiazee (1971) showed that an exposure at 26.7°C and 38% r.h., to 100% CO_2 produced complete mortality of *T. castaneum* eggs, larvae, pupae and adults within 60, 48, 60 and 18 h, respectively. However, in the same work it was reported that at 15.6°C 100% mortality was obtained for the larval stage exposed to 100% CO_2 within 84 h. This result may be compared with the results shown in Table 2, whereby at 15°C a 60% CO_2 atmosphere caused complete mortality of larvae and adults within 120 h. For the same temperature AliNiazee (1971) reported that 168 h was required when pupae were exposed to 100% CO_2.

Effects on *Oryzaephilus surinamensis*

Mortality of the four developmental stages of *O. surinamensis* exposed to three CO_2 concentrations at four different temperatures is shown in Table 3. At 15°C complete mortality was obtained for the larval and the adult stages exposed for 120 h, whereas the eggs and pupae showed a lower sensitivity to the tested CO_2 concentrations. At 21°C, although pupae were

TABLE 3

Mortality of the four developmental stages of *Oryzaephilus surinamensis* exposed to three CO_2 concentrations at four different temperatures.

Atmosphere (%)			Temp. (°C)	Exposure time (h)	% Mortality			
O_2	CO_2	N_2			Eggs	Larvae	Pupae	Adults
8	60	32	15	24	56	42.6	30.7	16.1
				120	84	100	47.8	100
			21	24	55	92.1	36.9	76.2
				120	100	100	95.4	100
			27	24	81	51.8	19.8	69.5
				96	100	100	100	100
			32	24	100	56.8	56.5	26.7
				96	100	100	100	99.0
5	75	20	15	24	41	37.0	31.3	13.2
				120	90	100	61.5	100
			21	24	57	96.6	23.5	76.0
				120	100	100	88.0	100
			27	24	90	95.6	33.4	94.2
				72	100	100	100	100
			32	24	100	100	86.1	100
				48	100	100	98.9	100
2	90	8	15	24	58	29.0	39.3	23.6
				120	99	100	60.1	100
			21	24	63	85.1	41.2	88.2
				120	100	100	89.8	100
			27	24	78	100	23.7	100
				72	100	100	100	100
			32	24	100	100	98.7	100
				48	100	100	100	100

again relatively insensitive, a 120 h exposure was sufficient for complete mortality of eggs even at 60% CO_2 level. At 27°C, a 96 h exposure to 60% CO_2 was sufficient to obtain complete mortality of all developmental stages of the insect (Table 3). At 32°C, all developmental stages were effectively controlled after 48 h at a CO_2 concentration of 90%.

Little information on the effect of CO_2 on O. surinamensis is available. Storey (1980) has shown that adults of this species when exposed to 1% O_2 and 9.0-9.5% CO_2 with the balance N_2, suffer 95% mortality after 47 h exposure at 15°C. The high sensitivity of this species to atmospheres containing less than 1% O_2 with the balance as N_2, at relative humidities ranging from 9% to 98%, was investigated by Jay et al. (1971). The results obtained in the present work demonstrate that the pupal stage of O. surinamensis is the most resistant to CO_2 atmospheres. However, at 27°C, a 72 h exposure to 75% CO_2 would result in complete mortality of all developmental stages of this species.

Field experiment

Table 4 shows the mean number of dead and live insects found in the grain samples taken from different locations before and after the CO_2 treatment, in the silo containing 6 881 tonnes of wheat. In this trial a 5 day treatment of CO_2 was applied even though data from laboratory studies indicate that a 7 to 10 day treatment would be necessary to obtain complete control. Due to technical reasons, after an average CO_2 concentration of 60-90% had been maintained for 5 days, the bin was aerated and the CO_2 concentration declined to a level which prevented continued effectiveness against the insects. The results indicate that although the percentage of live Sitophilus spp. after the treatment was reduced to 0.9, that of Tribolium spp. was 4.4%.

TABLE 4

Mean total number of insects (dead and live) per sample and percent live insect species found in the wheat bulk of 6 881 tonnes before and after treatment with CO_2.

	Before treatment		After treatment	
Insect species	Number of insects per sample	% Live	Number of insects per sample	% Live
Sitophilus spp.	8.8	58.8	14.2	0.9
Cryptolestes spp.	2.4	96.3	2.4	1.1
Tribolium spp.	0.3	65.0	0.3	4.4
Rhyzopertha dominica	0.4	66.7	0.2	0.0

The grain bulk temperature in this trial was not homogeneous. When samples taken from the 18°-20°C temperature region were compared with those from the 24°-33°C region in the grain bulk, it was evident that mortality of Sitophilus spp. was higher (99.9% vs 79.2%) in the samples taken from the latter region. From this trial the importance of the temperature and the treatment time needed for successful control when using CO_2 in actual field situations is evident.

A comparative analysis to determine the degree of gastightness was performed based on information obtained from the constant pressure test (Banks and Annis 1984). The air ingress into the structure was estimated to be approximately 1% per day which would result in a drop in the CO_2 concentration of approximately 0.75% per day. Based on gas concentration measurements, the resultant decay in CO_2 concentration after the termination of the maintenance phase was found to be 0.47% per day. An attempt was made to seal all apparent leaks found in the bin structure, but apparently some small cracks were not detected. This indicates that the level of gastightness of a silo used for the application of MAs requires careful consideration. The size and location of any leaks, the atmospheric temperature and the barometric pressure may affect the actual CO_2 decay rate.

The initial purge of CO_2 was through the main aeration duct located at the bottom of the bin, and was carried out for 32.5 h. During this period, a high CO_2 concentration was formed at the bottom. Gas measurements indicated that CO_2 gradients expanded progressively toward the upper layer of the bulk. A large proportion of the bin volume at the bottom had a CO_2 concentration of 80-100%, but the CO_2 concentration remained at 0-20% in the top layer, around the peak of the roof, after 32.5 h of purge. Then, the CO_2 supply was connected to the top line and a high purge flow was continued for an additional 6.5 h. This caused the CO_2 concentration in the upper layers of the bin to rise to 40-60%, but there was still an area between this layer and the high CO_2 concentrations below it that contained only 20-40% CO_2. These low CO_2 concentrations recorded close to the surface of the bulk apparently also contributed to the survival of insects. For effective insect control, after the bin is purged with CO_2, it has been found necessary to recirculate the mixture in order to obtain an even CO_2 concentration (Wilson et al. 1980). The results of our trial demonstrate that recirculation of the CO_2-air mixture should also be considered.

CONCLUSIONS

From tests carried out with three stored product insect species, the immature stages were found to be the most tolerant to the tested MAs. However, a 120 h exposure at 27°C was sufficient to cause 100% mortality of O. surinamensis. The study demonstrated that MAs have the potential to replace conventional residue-producing chemicals for the control of storage insects. However, additional work is necessary in the areas of the economic feasibility of the method, sealing techniques of the bins, and recirculation requirements, especially for CO_2 treatments. There is also a need for further basic bioassays aiming at demonstrating the relationships among exposure time, temperature, and the MAs necessary to obtain adequate levels of control of different stored product insect species.

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EFFECT OF GRAIN MOISTURE CONTENT ON THE ESTABLISHMENT AND MAINTENANCE OF A LOW OXYGEN ATMOSPHERE CONTAINING CARBON DIOXIDE

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ABSTRACT

An atmosphere resembling that produced by the combustion of liquid paraffin gas fuels, containing about 1% oxygen (O_2) and up to 15% carbon dioxide with the balance as nitrogen, was generated by a gas blending apparatus fed by gas cylinders. Wheat of different moisture content (m.c.) held in $19m^3$ capacity welded steel bins was purged with the atmosphere at the rate of about 5 l/min for 3 days and then at about 1 l/min for 4-7 days. Oxygen concentrations in the grain were lowered to the blender output level after little more than one replacement of the interstitial space, especially at higher grain moisture contents. Carbon dioxide levels took much longer to approach the blender output level. In grain of over 14% m.c. at 18°C, and over 15% at 13°C, O_2 levels carried on falling throughout the purge until at all points they were less than the blender output level. When bins were sealed to a standard permitting a 50% decay of an applied pressure of 500Pa in about 7 min, the low O_2 atmosphere was maintained at 13°C after cessation of purging.

Results are discussed in relation to larger scale trials, insect control and current requirements for grain storage.

INTRODUCTION

The use of modified atmospheres as a pesticide-free method of disinfesting bulk commodities is currently generating much interest worldwide (Bailey & Banks 1980, Jay 1984, Navarro et al. 1985, Fleurat Lessard 1987, Reichmuth 1987). Many economies in developing and developed countries alike rely heavily on the ability to export insect-free consignments of agricultural produce, and currently can only achieve this standard by the use of pesticides. The increased international awareness of potential hazards arising from the presence of pesticide residues in foodstuffs, coupled with a complementary emphasis on the need to protect pest control company operators from contact with, or exposure to toxic chemicals, is stimulating much research into alternative methods of pest control. The use of modified atmospheres is an attractive proposition because in many ways the technology is already available, the effects are if anything beneficial to the preservation of the stored material, particularly in terms of the germination of seed embryos and suppression of mycotoxin production (Paster et al. 1983), and the costs of treatment, although higher than for most pesticide formulations, are low in comparison to the losses incurred through pests and spoilage, or, more recently, the enhanced value that can be realized for commodities free from pesticide residues.

Currently there are two major approaches to stored product pest control using modified atmospheres, firstly, on site generation of gas, and secondly, bulk transport of liquified gas to the store. Carbon dioxide (CO_2) and nitrogen (N_2) are available in bulk supply, and also in cylinders. For smaller storage structures (up to 100 t of grain), a cylinder based supply may be cheaper than reliance on bulk containers. For very large storage facilities, the provision of on-site storage tanks for the gas would be an investment enabling delivery by road-hauled bulk containers at any time preceding the treatment, and would reduce or avoid demurrage charges. For all sizes of structure CO_2 offers many advantages over N_2 because a 90% replacement atmosphere in air remains effective until leakage exceeds 50-60%, whereas N_2 has to achieve a 100% atmosphere replacement and can only tolerate a 5% leak back of air (1% O_2) if full efficacy is to be retained.

On-site gas generation methods include the use of air compressors and molecular sieves to separate N_2 from air, or combustion of a hydrocarbon fuel to produce a low O_2 atmosphere containing some CO_2. This latter method has been developed in the USA and has been the major focus of recent research into modified atmospheres at the Slough Laboratory and elsewhere. For storage facilities of moderate to large size it offers an economic method of treatment with costs not very much greater than pesticide admixture, provided that a reasonable degree of sealing can be obtained. The atmosphere is more toxic than N_2 alone, provided that the CO_2 level can be raised well above 10%. Combustion of propane yields about 13%, and of butane about 15% CO_2. The tests described here investigate the effects of grain moisture content (m.c.) on the establishment and maintenance of a simulated exhaust atmosphere, produced from gas cylinders, comprising about 1% O_2 and 15% CO_2 in N_2.

METHODS

Experiments were conducted in three bins of welded steel construction of $19m^3$ capacity. Starting with wheat of about 14.5% m.c., 12t lots were prepared at m.c. ranging from 12.7 to 16.5%, adding water as necessary or drying using heat assisted fans. Prior to loading of the prepared grain in a bin, sampling lines were inserted at metre intervals from the hopper upwards and an additional line was attached to an apical valve fitted to provide a vent. Three thermocouples were attached to the sample line bundle at 2m intervals to monitor temperatures. After loading the bins a $3m^3$ headspace remained. Hatches, hoppers and plates were closed and the bin was sealed using tapes and adhesives. Leaving the 1.5mm bore apical vent open the bin was then subjected to a pressure test by applying 1000-Pa positive pressure (4" water gauge) with a 30 l/min output air pump, and monitoring pressure decay to below 250 Pa with an inclined manometer. Bins were tested for leaks by the soap bubble method and all were sealed to a minimum standard of at least 5 minutes for a 50% decay of pressure.

The experimental atmosphere was provided by using a 5 l/min output 'Signal' gas blender connected to cylinder supplies of air, N_2 and CO_2. An output line from the blender was tapped into the base of each bin and the gas composition was monitored using a 'Servomex' O_2 analyser and a 'GOW-MAC' CO_2 monitor connected in series to a side arm of the output line. The blender was adjusted to provide atmosphere containing 14.5-15% CO_2 and 0.9-1.1% O_2. Prior to starting the purge, test insects in batches of 100 on food in wire gauze cages were inserted into the grain from the observation hatch at depths of 2m, 1m and surface using metal rods and threaded cage holders. Controls were inserted into grain in a neighbouring bin. Species tested were laboratory reared adults of Sitophilus granarius (L.), S. oryzae (L.), Cryptolestes ferrugineus (Stephens) and Oryzaephilus surinamensis (L.). After allowing 2 or 3 days for acclimatisation of insects, the resealed bin was connected to the blender output to start the purge, and measurements were taken at intervals through the sampling lines to estimate gas levels within the bin. After three days of purging, by which time a total atmosphere replacement had been achieved, the bin was connected to a 1 l/min output blender to maintain the atmosphere. The application of gas was stopped altogether after a further 4 days in the summer trials at 18°C, and after a further 7 days in the autumn when grain temperatures had fallen to 13°C. Insects were removed the following day in the summer, while in the autumn the leakage of air back into the bin was monitored for a further 4 days before opening the hatch. Mortality was assessed after 7 and 14 days incubation at 25°C in the laboratory.

RESULTS

The summer trials

The m.c. of grain set up in the three bins was 12.7, 14.5 and 16.5%. Away from the surface and periphery, mean grain temperatures in the bins were 18.0 ± 0.5°C. Discounting fluctuations at the surface or in the headspace, the temperatures recorded varied by less than 1°C over the

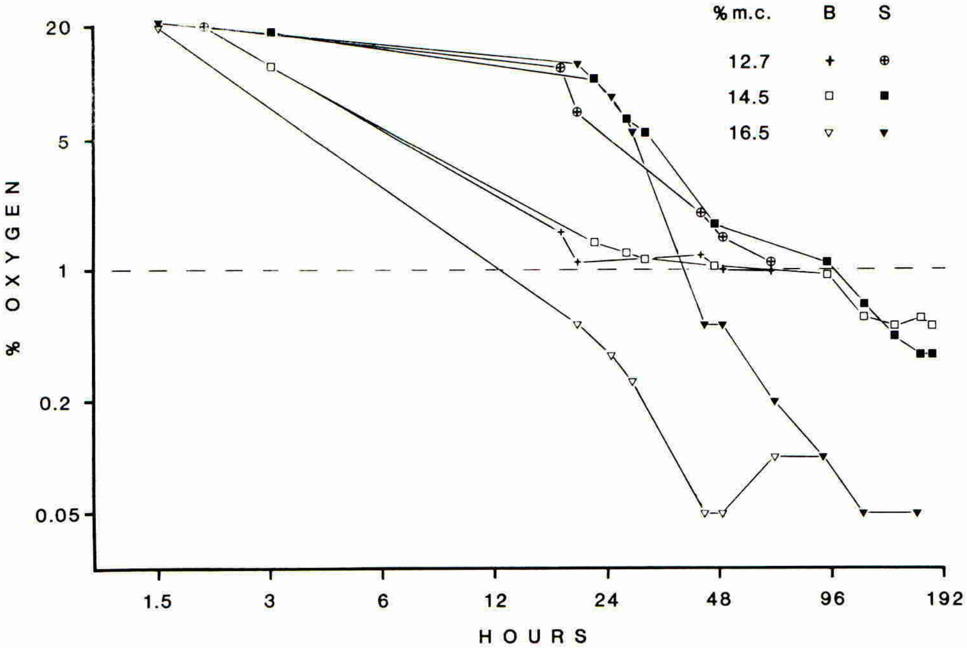

Fig.1. Oxygen levels near the grain surface (S) and at 2m depth (B, base of bin above hopper zone) in grain of different m.c. at 18°C.

treatment time in any particular bin. Purging with a gas mixture containing 15.0 ± 0.5% CO_2 and 1.0 ± 0.1% O_2 at the rate of 4.7 l/min achieved a 90% replacement of the atmosphere in each bin within 43h (Figs 1 and 2). This represented about 1.2 replacements of the total air space within the loaded bins.

A further day of purging was required to bring O_2 levels down to the gas blender output level except in grain at the highest m.c. (Fig. 1), and at this time the gas flow to the bin was reduced to about 0.9 l/min by switching to a lower output gas blender. In the bin containing 16.5% m.c. grain, O_2 levels continued to fall and by the 5th day were less than 0.1% at most points, indicating microbiological activity. By this time O_2 levels in the 14.5% m.c. grain also fell to below that of the blender output so that 1 week after the start of purging, O_2 levels had fallen to 0.35% at some points. Leakage into gas sampling lines prevented monitoring of the purge in the bin of lowest m.c. beyond the third day. Up to this time the profile of gas levels at all points within the bin closely resembled that for grain at 14.5% m.c.

The CO_2 concentrations, whether near the grain surface or near the base, lagged behind the blender output for some days after O_2 levels had stabilized (Fig. 2), and for grain at 14.5 m.c. were still 0.5-1% behind after 1 week of purging, by which time 2½ atmosphere changes had occurred. There was no survival of adults of the four test species exposed in any of the three bins.

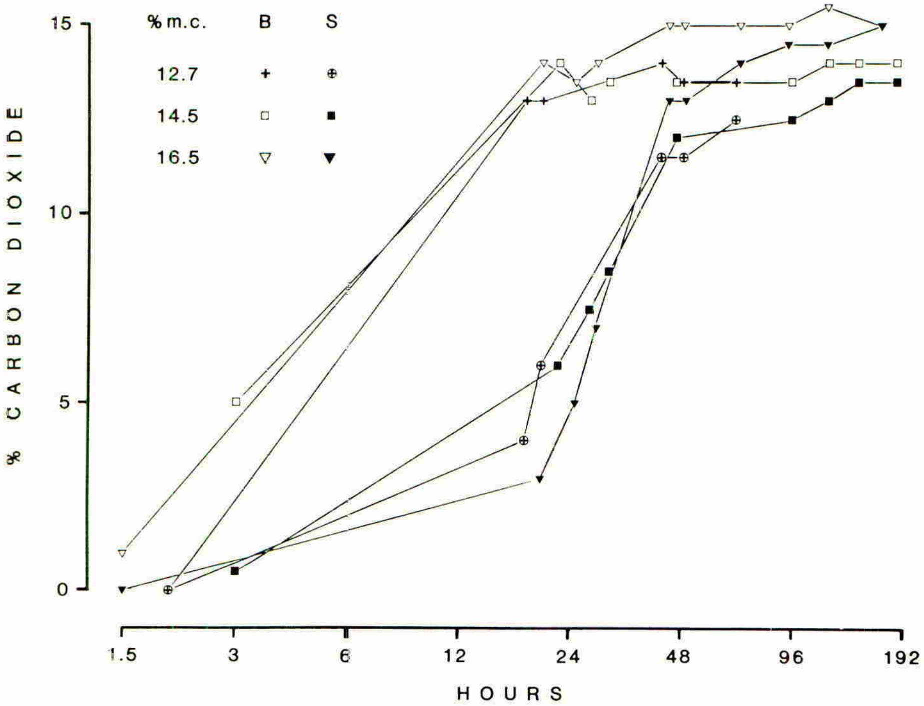

Fig. 2. Carbon dioxide levels near the grain surface (S) and at depth 2m (B, base of bin above hopper zone) in grain of different m.c. at 18°C.

The autumn trials

During this series, grain of m.c. 13.9, 14.7 and 15.4% was loaded into the three bins at 13.0 ± 0.5°C. After insertion of test insects and replacing hatches, pressure tests revealed half lives of 7 ± 1 min between 1000 and 250Pa. Purging with an atmosphere containing 0.95 to 1.1% O_2 and 14.5 to 15.0% CO_2 in N_2 at the rate of 4.7 l/min again achieved a 90% replacement of the atmosphere well within 48h (Figs 3 and 4). Again the CO_2 level lagged behind O_2 in approaching the blender output level at all points. Indeed, only in the grain at 15.4% m.c. was the end point reached within 10 days (3½ atmosphere changes), while apart from minor fluctuations O_2 levels at the grain surface approached the blender output by the third day (Fig. 3). As at 18°C, O_2 levels in the bin with grain of the highest m.c. continued to fall after reaching the output level and by the end of the purge were less than 0.6% at all points other than the one nearest the gas inlet at the hopper bottom. No fall below 1% O_2 was apparent in the 14.7% m.c. grain within the 10-day purge time at 13°C.

Where profiles of gas measurements from the centre of the bins and from the headspace were compared for grain of 14.5 to 14.7% m.c. at 13 and 18°C, a close similarity was observed over the first 4 days (Fig. 5). Thereafter a difference became apparent as O_2 levels continued to fall, and CO_2 levels approached the blender output, at the higher temperature.

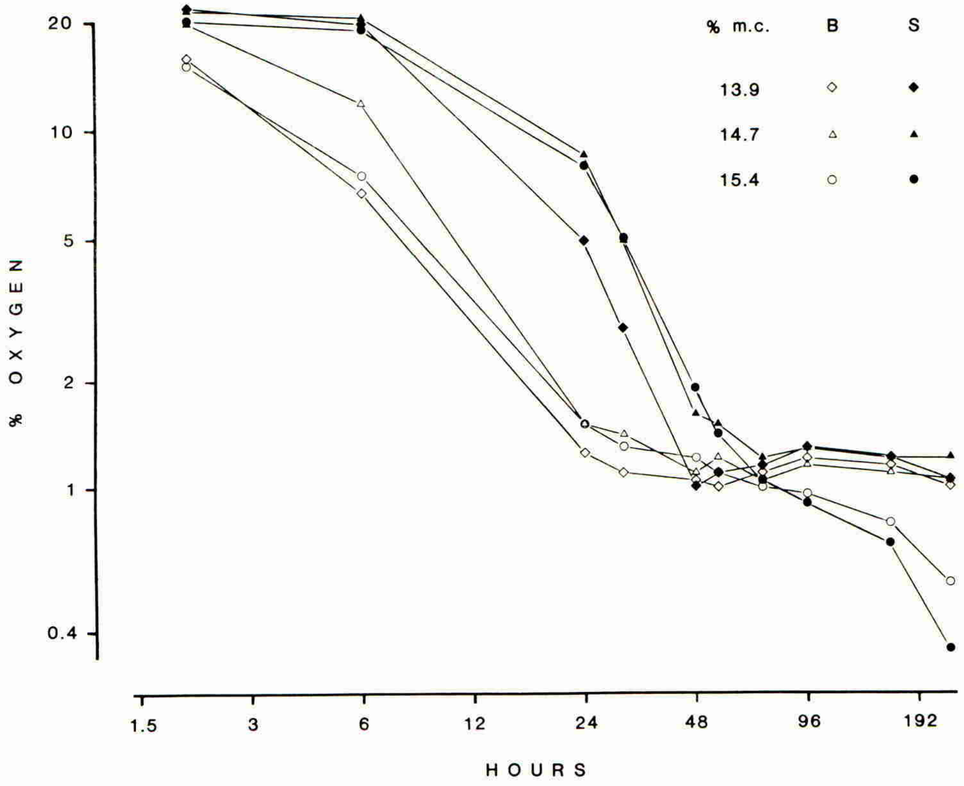

Fig. 3. Oxygen levels near the grain surface (S) and at 2m depth (B, base of bin above hopper zone) in grain of different m.c. at 13°C.

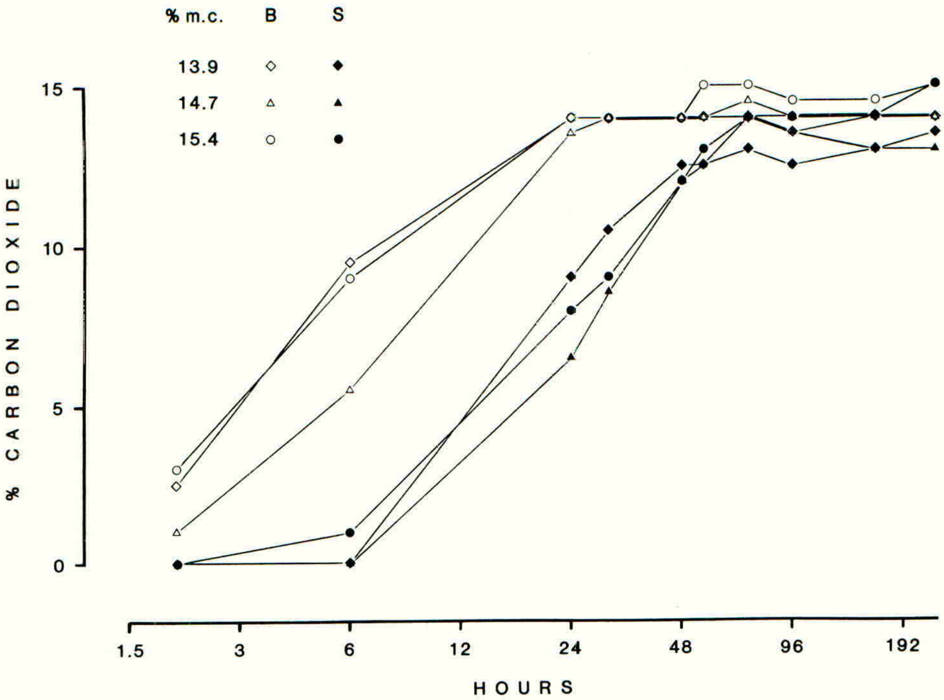

Fig. 4. Carbon dioxide levels near the grain surface (S) and at depth 2m (B, base of bin above hopper zone) in grain of different m.c. at 13°C.

TABLE 1

Mortality (%) of adult <u>Cryptolestes ferrugineus</u> (2 cages of 100 ± 2 insects per position) exposed for 14 days to a low O_2 raised CO_2 atmosphere in grain of differing moisture content at 13 ± 1°C.

Sample position	Grain moisture content (%)		
	13.9	14.7	15.4
Grain surface	33.7	78.5	92.3
1m depth	81.5	79.9	99.0
2m depth	92.0	95.6	99.5
Control	10.3	8.0	14.2

When purging was completed, continued monitoring of gas levels in grain at 13°C revealed the slow leakage of air back into the bins. However, in the bin containing grain at 15.4% m.c., this leakage appeared to proceed much slower than in the other bins and it was clear that O_2 was still being removed from the atmosphere. By the fourth day of monitoring the O_2 level, which had risen from 0.35 to 0.9% by 30h after ceasing the purge, had again fallen to 0.65% in the headspace (Fig. 6). Survivals were recorded of adult <u>C. ferrugineus</u> from all of the bins but the level varied according to the m.c. of the grain and the depth to which cages were inserted (Table 1). Lowest survivals were recorded from the 2m deep samples exposed in the bin containing grain at 15.4% m.c. No survival of adult S. granarius occurred in any of the bins.

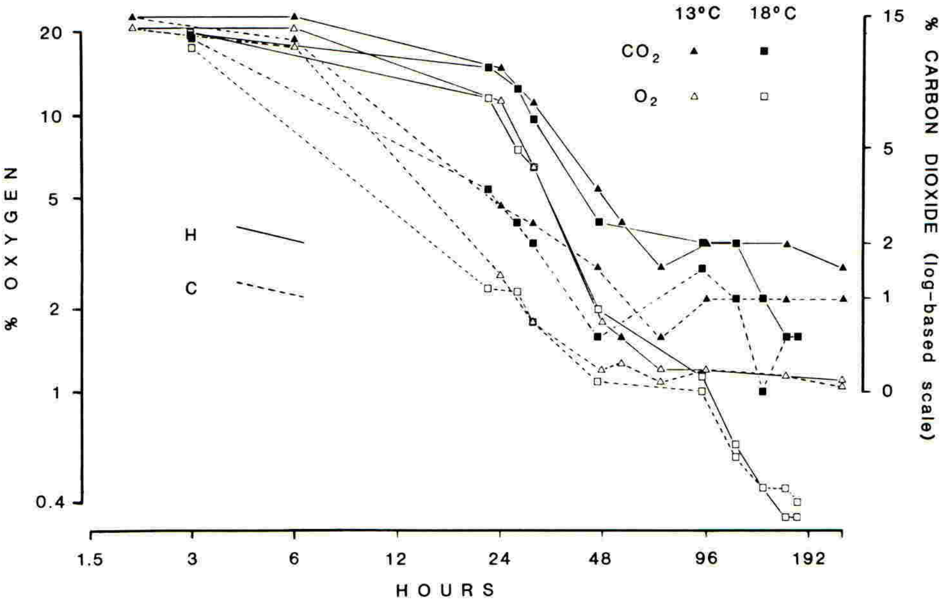

Fig. 5. Levels of CO_2 and O_2 in bins containing wheat at 14.6 ± 0.1% m.c. purged at 13 and 18°C, sampling from the centre (C) of the grain bulk and the headspace (H). CO_2 scale is the difference between the blender output and the measured CO_2 concentration at H or C.

DISCUSSION

It is evident from the results that gas produced by blending air, N_2 and CO_2 in the ratio 1:16:3 (1% O_2, 84% N_2 and 15% CO_2), and fed in to the grain bins at the low rate of 5l/min, replaced the air in the bins with very little mixing of gas across the purge front. The very small difference in density between air (14.4) and the simulated exhaust gas (15.2) was apparently sufficient to allow the incoming gas to push out the original atmosphere. Such an effect is not normally experienced in purging structures with N_2 alone.

Normally purge rates are much higher than those described here and it is usual to attempt to purge a structure within a working day. Recent trials conducted in the USA by C L Storey, at which the author was present, investigated the purging of 300t rice silos with a mobile exothermic gas generator utilizing propane as a fuel and producing an atmosphere containing about 12-13% CO_2. In one silo, purging with a $60m^3/h$ gas flow rate achieved a complete atmosphere replacement in the grain and headspace in about 6h, indicating again that very little mixing between air and applied atmosphere had occurred during the purge. This silo was much less gas-tight than the present bins, an applied pressure of 200Pa decaying to 100Pa in about 1 min. The present findings indicate that low output gas generators may be used to purge relatively gas-tight bins quite successfully.

At both 13 and 18°C considerable sorption of CO_2 occurred in all bins. True to the principle that physical sorption is negatively correlated with temperature, more gas was removed at 13°C than at 18°C, as

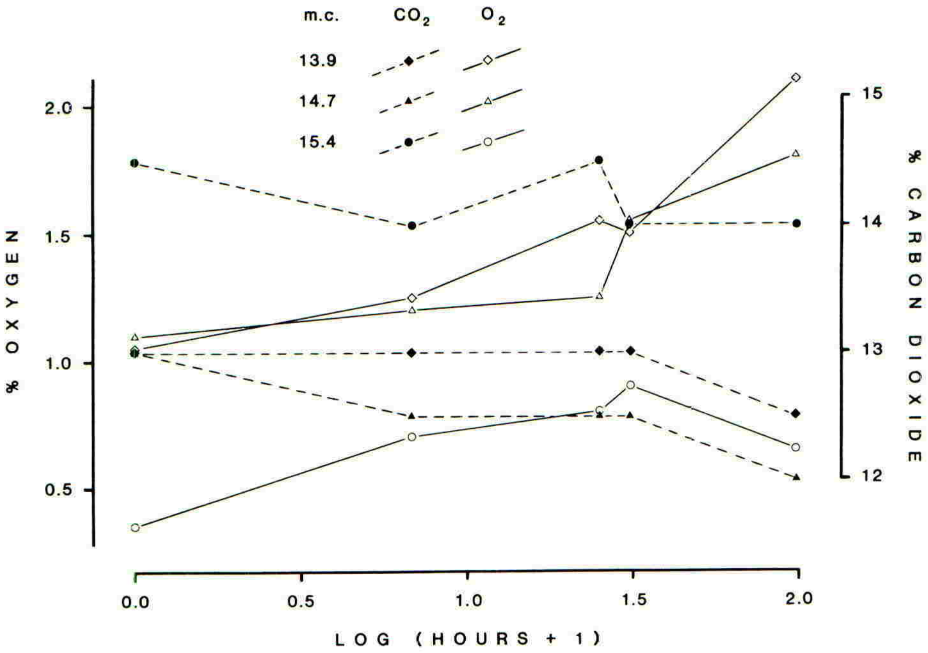

Fig. 6. Headspace CO_2 and O_2 levels in the bins after stopping purge.

judged by the time required for gas output from the bins to approach the input levels. With grain at 16.5% m.c. at 18°C, 7 days were required for equilibrium whereas 10 days were required at 13°C for grain of 15.4% m.c. At about 14.6% m.c., CO_2 levels were mostly within 0.5% of the blender output by the sixth day of purging at 18°C, but had not approached within 1% by the 10th day of purging at 13°C (Fig. 5). Increased m.c. is known to increase sorption, but in the case of CO_2 any such effect was apparently overridden by the effect of temperature and possibly by the production of the gas by microorganisms on the grain.

Oxygen levels in the grain fell to less than the 1% level of the incoming gas with m.c. of 16.5 or 14.5% at 18°C (Fig. 1) or 15.4% at 13°C (Fig. 3). This removal of O_2 was dependent on temperature as well as grain m.c. and at 18°C was evident on the first day of purging in grain of m.c. 16.5%. In this bin O_2 levels fell to 0.05% at some points within 48h. In grain of 15.4% m.c. at 13°C, O_2 levels fell more slowly than in grain of 14.5% m.c. at 18°C. Raising the temperature is known to increase microbiological activity in reducing O_2 levels (Qianyu 1984) even though raised CO_2 and very low O_2 levels reduce growth (Paster et al. 1983). An examination of the grain at 15.4% m.c. revealed the presence of yeasts and Penicillium spp.

From a practical standpoint, the continued removal of O_2 in a purged bin of grain offers the useful prospect of prolonging the life of the applied insecticidal atmosphere without further use of fuel. The present trials were conducted in bins which were able to meet the relatively stringent standard for the gas-tightness of retaining structures recommended in Australia for the use of CO_2 in a single application to disinfest bulk grain, namely an applied positive pressure of 500Pa decaying to 250Pa in not less than 5 min (Banks & Annis 1977 1980). At the relatively low temperature of 13°C it was clear that with grain of 15.4% m.c. O_2 levels were kept below 1% for 4 days after cessation of purging (Fig. 6) and there was every prospect that the low O_2 level could be held for a much longer period. As a result the mortality of adult _C. ferrugineus_, already higher than in the bins with grain of lower m.c. (Table 1), could easily have been increased to 100% by extending the exposure by a few days. Immature stages of grain pests normally require much higher temperatures for development and of the common species only _S. granarius_ is known to develop below 15°C. To control immature stages of this species, very long exposures of up to 8 weeks duration may be required (Spratt, E.C., personal communication). Much shorter exposure times are required for control of insect stages with modified atmospheres as temperatures are increased. In a well-sealed structure the depletion of O_2 by moister grain can keep pace with incoming O_2 from air leaks, particularly at higher temperatures, and control may be achieved by purging with a volume of gas amounting to as little as two changes of the atmosphere in a well sealed bin. A slight reduction in the standard of sealing will result in a more rapid ingress of O_2 and will have the effect of increasing the grain m.c. necessary for O_2 levels to be held down at each temperature. For any structure best results will be obtained at higher temperatures.

At present wheat and barley going into government intervention storage in the UK is required to be of m.c. not exceeding 15%, and, depending on the time of year, cooled to 20°C or below. For trading purposes the Grain and Feed Trade Association (GAFTA) will deal with consignments up to 16% m.c. There is thus some overlap between the current commercially acceptable standards of grain conditioning and those parameters which would offer the prospect of enhancing control using a low O_2 atmosphere. Currently there is a need for suitable gas generators to become available for widespread use in farm grain bins, but high output machines for larger scale treatments are already manufactured overseas. More work is required on the grain temperatures and m.c. which have the effect of keeping O_2 levels low under a range of standards of gas tightness, and on the minimum levels of flow of combusted gas necessary to maintain atmospheres in the less gas-tight structures.

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1987 BCPC MONO. No. 37 STORED PRODUCTS PEST CONTROL

CONTROL OF INSECTS IN CONFECTIONERY WALNUTS USING MICROWAVES

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ABSTRACT

Insects were killed by direct exposure to microwave energy but it was likely that much of the lethal effect came from heat conducted from the interior of the generator unit. However, insects mixed in with walnuts were readily killed and heating infested samples to 75°C followed by cooling to ambient, killed all adult Tribolium castaneum and Oryzaephilus surinamensis and the larvae of Ephestia cautella. The effects of maintaining nuts at elevated temperatures following microwave heating was also assessed. Longer exposures to 55°C killed all the above insects in samples of walnuts as well as the larvae of Plodia interpunctella. Samples of nuts containing all stages of all four species were disinfested by 15 minutes at 60°C. Organoleptic tests indicated that such heating did not adversely affect the flavour of the nuts.

In order to develop a practical technique the cooling characteristics of walnuts in their original packaging of 12.5 kg boxes were assessed. The boxes of nuts were heated to temperatures likely to give complete control of insects and then allowed to cool. The differences between the centre and periphery were found to be sufficiently great to give a risk of insect survival in the outer regions whilst subjecting nuts at the centre to excessively long exposure to heat. An alternative method of heating nuts in a thin layer on a conveyor belt was therefore considered more satisfactory.

INTRODUCTION

Radio frequency energy such as microwaves can be used to raise the temperature of a wide range of products. This has many potential uses but the disinfestation of foodstuffs is particularly attractive and has been the subject of much research. Nelson (1973) reviewed the technical aspects of microwave heating to disinfest food and suggested that the effectiveness of the method might depend on being able to heat selectively the pests rather than the substrate. He further suggested that improvements could come from adjusting the frequency of the radiation used to maximise the heating effects for any specific pest.

Several workers have assessed the technique under practical conditions with varying degrees of success. Kirkpatrick (1974) found that cowpea weevils, Callosobruchus maculatus infesting peas were not controlled by temperatures which seriously reduced germination. He also reported that the same temperature produced by infrared radiation was more effective against insects. Hurlock et al. (1979) were able to control several species of stored product insects infesting wheat. However, they were less successful when the insects were infesting cocoa where exposure to the dose that killed all insects in wheat allowed 20% of those infesting cocoa to survive. These workers also supplemented the microwave heating with hot air to extend the period at elevated temperature which improved the effectiveness of the technique.

The use of microwaves to disinfest food clearly has a number of advantages, particularly with food for human consumption. The treatment leaves no residues and lends itself to commercial requirements for an "instant" kill method that can be applied during a production process. However, there are a number of disadvantages that must be overcome before the technique can be used in practice. For example, few workers have reported consistently obtaining 100% kill by microwave heating for a wide range of insects, yet in the processed food industry, no survival of pests can be tolerated. The need for complete kill is made more difficult by likely restrictions on maximum temperatures because of flavour changes in the product. The possibility of exploiting selective heating of the pests rather than the substrate seems impractical. In the U.K. the use of microwaves is strictly controlled and only two frequencies, 896 and 2540 MHz, are permitted for heating. Neither of these frequencies are close to the likely optima for heating insects (Nelson 1973). In any case, the small size of insects must limit the amount of microwave energy that they absorb in relation to the substrates. Their large surface area to volume ratio is also likely to allow them to dissipate heat readily.

It is clear that before microwave heating can be used as a commercial tool to disinfest foodstuffs some careful development work is needed to ensure that the method will give complete kill of all stages of all likely pests under practical conditions. This paper reports the work carried out to assess the feasibility of disinfesting confectionery quality walnuts by microwave heating and to provide guidance on a practical control technique. It does not provide a detailed account of the effects of microwave heating on insects but concentrates on the steps required to develop a method that can be used under commercial conditions.

The work was carried out in three stages. Firstly, some preliminary checks were made to establish the basic parameters of the technique. Following the satisfactory completion of these tests more detailed studies were undertaken to establish the effects of various temperature/time combinations on pests. Finally, some consideration was given to the practical aspects of adapting the method to a commercial production line.

EXPERIMENTAL WORK

Materials

Insects:- A pest intelligence data base held at the Slough Laboratory (Anon 1983) was used to establish the most likely pests of stored walnuts. Records of infestation in walnuts imported into the U.K. over the last 10 years, suggested that the Saw-toothed Grain Beetle (*Oryzaephilus surinamensis*), the Rust Red Flour Beetle (*Tribolium castaneum*), the Tropical Warehouse Moth (*Ephestia cautella*) and the

Indian Meal Moth (Plodia interpunctella) were the most common pests. These species, obtained from standard laboratory cultures, were used for the work.

Walnuts:- Shelled Indian walnut halves were supplied in 12.5 kg cardboard boxes. The contents of several boxes were mixed together to provide a single, uniform batch. The moisture content of a sample of nuts from this batch was found to be 4.4% as determined by an oven method (BS 4289/3 1978, 103°C for 3 hours then checked hourly until constant weight).

Containers:- The nuts were exposed to microwaves in polypropylene boxes, measuring 200 x 96 x 60 mm. These could be sealed with snap-on lids as required. Samples of infested nuts were equilibrated to room temperature before and after exposure in plastic bags or wide-necked glass jars.

Microwave generators:- Two sources of microwaves were used during the experiment. An industrial microwave generator was used to provide radiation at 896 MHz. This unit used a conveyor belt to move samples through a wave guide which contained and focussed the microwaves. A laboratory unit working at 2450 MHz was also used.

With both generators the following formula was used to calculate the period of exposure needed to give a specific temperature rise:-

$$\text{Power (watts)} = \frac{\text{Mass of sample} \times 4.2 \times (T_2 - T_1)}{\text{Time (seconds)}}$$

T_1 = Original temperature °C
T_2 = Final temperature °C

Methods

Preliminary investigation:- An attempt was made to assess the effect of microwave heating on T. castaneum by exposing the insects directly to microwaves in the laboratory unit. Batches of 50 insects were placed on a layer of filter paper at the bottom of a 500 ml beaker and exposed to a microwave field of about 600 watts for 30, 60 or 90 seconds. The beaker was insulated from direct contact with the unit by a further pad of filter paper. After treatment the insects were transferred to a constant 25°C and 70% r.h. for 24 hours before an assessment of mortality was made.

Despite the efforts to insulate the insects from the effects of conducted heat, much of the heat they received probably came by conduction from the beaker or by radiation from the interior of the unit. Ninety seconds exposure was needed to give 100% mortality, by which time the beaker was very hot to the touch. This approach was abandoned as being unlikely to yield results that would be useful in a practical assessment. Further work was concentrated on the effects of heating on insects mixed with nuts.

The accuracy and repeatability of the heating achieved by the laboratory microwave generator was tested. Three-hundred gram batches of walnuts in polypropylene boxes were heated to temperatures calculated to be 60, 70 or 80°C. Five replicates at each temperature were used and the temperature of the nuts was measured immediately after heating using a thermistor probe. The mean temperatures achieved and the standard deviations were:- 61.66 ± 1.4, 71.03 ± 1.72 and 79.17 ± 2.34°C. These were close to the intended temperature and some of the variation may have been caused by changes in ambient temperature that occurred during the trial.

A further preliminary test was carried out in which infested nuts were exposed to microwaves using an industrial microwave generator. Three-hundred gram samples of nuts were infested with 10 E. cautella larvae and 25 T. castaneum and O. surinamensis adults 24 hours before being exposed to microwaves. The infested nuts were placed in sealed plastic boxes and 5 replicates were heated to 50, 60, 75 or 90°C, the temperature being controlled by adjusting the power setting. The samples took about two minutes to pass through the unit, although they were not exposed to microwaves for all of this period. Five replicates were left unheated as controls. After exposure all samples were transferred to plastic bags and allowed to cool to ambient (about 25°C). Measurement of the temperature of the nuts shortly after removal from the microwave generator suggested that the samples had been heated to within 2-5°C of the calculated temperature. Twenty four hours after heating, the insects in the samples were examined and the mortality of the insects was assessed. The results are given in Table 1 and show that a temperature of 75°C or more gave complete kill of insects. The E. cautella larvae appeared to be more susceptible than the two species of beetle.

TABLE 1. The mean percent mortality of insects infesting walnuts heated by a commercial microwave generator to a range of temperatures and then allowed to cool to ambient.

Temperature °C	Species T. castaneum	O. surinamensis	E. cautella
50	67.2 ± 21.2	84.2 ± 15.1	95.6 ± 8.8
60	82.8 ± 11.8	94.6 ± 3.8	100
75	100	100	100
90	100	100	100
Control	1.2 ± 1.0	5.4 ± 6.1	11.2 ± 9.8

Uninfested nuts were also heated in the same way to provide samples for organoleptic assessment by a professional taste panel. The results indicated that temperatures of up to 75°C seemed to have little adverse effect on taste.

Following these encouraging results it was decided that further work to develop a practical control strategy was justified. However, work was concentrated on assessing the effects of heating nuts rather than heating insects in isolation. Likely commercial microwave heating processes almost certainly will involve passing nuts through a generator on a conveyor belt so that there is obvious scope for extending the time that nuts spend at a given temperature. Therefore, a series of experiments was set up to examine the effects of various temperature/time combinations on infested nuts.

Experimental trials:- Twenty-five adult O. surinamensis or T. castaneum, or ten E. cautella or P. interpunctella larvae were added to 300 g batches of walnuts. The following day the infested nuts were heated to 50, 55 or 60°C (taking between 63 and 84 seconds) in a laboratory microwave generator. The nuts were then transferred immediately to a hot-air oven at the appropriate temperature and held for 15, 30 or 60 minutes. On removal they were cooled to ambient as quickly as possible by a forced air draft and transferred to wide-necked glass jars and left at ambient conditions overnight. The nuts were then tipped out onto a tray and the condition of the insects was assessed. Five replicate batches of nuts were used for each species/temperature/time combination. Batches of infested nuts were also left unheated to act as controls.

In addition to the infested nuts, 5 x 300 g batches of nuts without insects were heated to each of the following temperature/time combinations:- 50, 55, 60, 70 and 80°C for 15, 30 or 60 minutes. These nuts provided samples for organoleptic assessment.

Since all stages of an insect's life-cycle can be present under practical conditions the effect of heating on established infestations in nuts was assessed. Twenty-five adults of each of the species were added to 300 g batches of nuts in wide-necked jars. The samples were incubated for 10 weeks at 25°C and 70% r.h. All species bred readily on the nuts and had completed their life-cycle within the period of incubation. The nuts and insects were transferred to plastic boxes and heated as previously described to 60°C for 15 minutes. The nuts were then cooled to ambient, returned to the jars and held at 25°C and 70% r.h., before being examined for live insects 1 and 28 days later. All insects found during the assessment were removed. Controls were treated in the same way except that they were not heated.

Practical considerations:- The method by which the nuts could be exposed to the microwaves was considered. The initial tests had shown that nuts could be heated satisfactorily in small boxes passed through an industrial microwave generator. The nuts are normally supplied in 12.5 kg cardboard boxes and treatment of the unopened boxes would have a number of practical advantages. Industrial generators are available that could heat such boxes but the effectiveness of the technique would depend on temperature gradients within the box, particularly when cooling. All the nuts would need to reach a temperature/time combination that would kill all pests but would also need to cool sufficiently rapidly to prevent flavour changes. Therefore, a series of tests were set up to check the cooling characteristics of 12.5 kg boxes of nuts. Temperature probes were placed at 15 points throughout the box, with one additional point being used to record ambient temperature. The box was heated in a hot-air oven to 60 or 80°C and allowed to cool to 20°C. During cooling temperatures were recorded at 15 minute intervals using a data logger.

RESULTS

Preliminary investigations:- The results of the initial tests, given earlier, can be summarised as follows:-

i) Walnuts can be heated readily by microwaves.
ii) The heating can be controlled accurately.
iii) The insects used in these trials were killed by short exposure to 75°C.
iv) The flavour of the nuts was not affected by short exposure to 75°C.

Experimental trials:- The results of the various temperature/time combinations are given in Table 2. All exposure periods at temperatures above 55°C gave complete kill of all three species. At 50°C all three exposure periods killed all the larvae of both species of moth and 60 minutes exposure at 50°C gave 100% kill of *O. surinamensis* and 99.2% kill of *T. castaneum*. However, there was appreciable survival of both species of beetle after 15 and 30 minutes exposure to this temperature.

TABLE 2. The effect of various temperature/time combinations on insects infesting walnuts. The figures are means of the percentage mortality. The samples of infested nuts were heated to the required temperature in a laboratory microwave generator and maintained at that temperature in a hot air oven.

Temperature °C	Time in minutes	Species			
		T. *castaneum*	*O.* *surinamensis*	*E.* *cautella*	*P.* *interpuntella*
50	15	51.2 ± 8.5	72.6 ± 15.0	100	100
	30	74.7 ± 14.9	83.3 ± 13.8	100	100
	60	99.2 ± 1.7	100	100	100
Control		0	0.8 ± 1.6	2.85 ± 5.7	0
55	15	100	100	100	100
	30	100	100	100	100
	60	100	100	100	100
Control		0	0.8 ± 1.6	0	0
60	15	100	100	100	100
	30	100	100	100	100
	60	100	100	100	100
Control		0	0	0	3.3 ± 6.6

The results of the organoleptic tests were not completely clear as some of the control samples scored lower than treated nuts. However, it was apparent that heating to 60°C for 1 hour or 75°C for up to 30 minutes had no discernible effect on flavour.

The samples of nuts in which infestation had been allowed to develop were all heavily infested at the time of treatment and, because the second generation had begun to develop, all stages of the life-cycle were likely to have been present. The results of counts 1 and 28 days after heating are given in Table 3. No live insects were found in any sample. Both species of moth were assessed in the same samples and no differentiation was made between the species. Numbers of adult moths were not recorded as this stage is relatively short-lived and the samples contained many dead adults before heating. However some live adults were present in the samples before treatment but were all dead at the time of the first assessment. The *O. surinamensis* larvae were not counted but had any survived the treatment they would have emerged as adults by the 28 day count. This is clearly demonstrated by the controls where a mean of 127 adults had emerged between the 1 and 28 day counts.

TABLE 3. The mean number of live insects (5 replicates) found after heating walnuts with an established infestation to 60°C for 15 minutes.

				Species		
	T. castaneum			*O. surinamensis*	Moths*	
	a	p	l	a	p	l
				24 h after heating		
Control	35.4	34.6	84.2	171.2	34.6	27.5
	± 8.5	± 7.3	± 9.2	± 42.5	± 26.8	± 18.3
Heated	0	0	0	0	0	0
				4 weeks after heating		
Control	22.6	1.4	40.6	127.0	7.0	112.6
	± 4.3	± 0.8	± 9.7	± 41.3	± 9.9	± 87.3
Heated	0	0	0	0	0	0

a = adults p = pupae l = larvae

*samples originally infested with mixed *E. cautella* and *P. interpunctella*

Practical considerations:- Cooling curves for various points in the boxes were established. The outer layer of nuts cooled to below 50°C, a temperature at which insect survival becomes likely, within 15 minutes. However, the centre of the box took more than 8 hours to cool to this temperature.

DISCUSSION

This work shows that microwave heating can be used to kill a range of likely pests infesting walnuts. A short exposure to 75°C gave complete control of the insects tested. However, lower temperatures maintained for longer periods were also effective.

There was no indication that insects heated more rapidly or to higher temperatures than the substrate. Tests on the insects alone were unsatisfactory because no suitable method of insulating them from other sources of heat produced within the generator could be found. Much of the microwave energy was absorbed by the walls of the unit or the beaker containing the insects. A similar amount of available energy was able to raise the temperature of 300 g of nuts from about 20°C to 70°C.

Longer exposure to lower temperatures also gave complete kill of the 4 species of insect. Less than 1% of the T. castaneum adults survived 60 minutes exposure to 50°C but even this indicates the considerable tolerance of these insects to heat. The moth larvae were more susceptible than either of the beetle species and were all killed by 15 minutes at 50°C. All exposure periods at 55°C gave 100% kill of all species and 15 minutes at this temperature would appear to be the lowest temperature/time combination to give complete control. However, in practice, an adequate safety margin must be incorporated into a control program to take account of any variations in the temperature of the product. Therefore, a recommendation that nuts be heated to 60°C and held at that temperature for 15 minutes would appear to be appropriate.

This work shows that microwave heating could be exploited to disinfest walnuts on a commercial scale. The process is well suited to a continuous flow on a production line. However the problems of temperature variation and unequal cooling mean that the process is more likely to be satisfactory if the nuts are heated in a thin layer rather than in the 12.5 kg boxes. The temperature/time combinations (60°C for 15 minutes) needed to ensure all pests are killed do not seem to adversely affect the flavour of the nuts.

ACKNOWLEDGEMENTS

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7.

Summaries of Poster Presentations

RODENT CONTROL IN STORED PRODUCTS

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Rats and mice inflict losses in a wide range of stored commodities. In most countries the pest complex comprises one or more of the cosmopolitan species, Rattus norvegicus, Rattus rattus and Mus musculus, as well as members of the indigenous rodent fauna.

The most obvious loss inflicted by rodents is the consumption of the commodities in store. However, to gain access to this food, rodents often damage the fabric of buildings and packaging materials. The costs of making good this damage often exceeds the value of direct food losses. The pests also foul with droppings, urine and hairs much of the stored produce they do not eat, and the cost of either cleaning or rejecting contaminated foodstuffs must be included in the toll they exact.

Rodent control in stores calls for an integrated approach. A high standard of house-keeping does much to reduce infestation levels and aid inspection. A wide range of proofing measures may be applied to exclude rodents from stores. Usually, however, there is a need to apply rodenticides, mainly as baits, at some stage of an integrated programme.

Among the target species mentioned above, R. rattus and M. musculus are relatively tolerant to the warfarin-type anticoagulants and populations of all three species resistant to these compounds now exist in many temperate and a few tropical countries. The second-generation anticoagulants, therefore, are widely used against rodents infesting stored products.

Among the second generation anticoagulants (difenacoum, brodifacoum, bromadiolone and flocoumafen), brodifacoum is the most potent, having a low LD50 to all three cosmopolitan species. Complete kills in one-day feeding tests support the claim to 'single-feed' action. This high potency is a prerequisite in most stored products situations where there is often abundant alternative food.

The results of a series of replicated field trials conducted on farms in the UK, encompassing a wide range of storage facilities and stored commodities showed that brodifacoum baits eradicated rat infestations more quickly, used less bait and required fewer site visits than the other compounds tested.

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DELTAMETHRIN FOR GRAIN PROTECTION

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Abstract :

Deltamethrin is an insecticide belonging to the pyrethrinoîds family. It is used for the cereals and legumes protection at a dosage varying from 0.25 to 0.5 ppm + PB or a minimum at 1 ppm without PB.

According to the equipment used for the cereals protection, 3 different formulations are commercialized :

- K.OTHRINE GRAIN ULV 6 g/l of deltamethrin + 60 g/l of PB. It is used by fogging at the bottom of the elevators by the means of apparatus at the dosage of 4.2 to 8.4 litres for 100 tons of grain.

- K.OTHRINE GRAIN EC 25 PB containing 25 g/l of deltamethrin + 250 g/l of PB. It must be sprayed out on the grain conveyed by a conveyor belt.

- K.OTHRINE GRAIN 2 PP containing 2 g deltamethrin/kg of dust. It is dusted at 500 g/ton.

Deltamethrin is effective at these above dosages against R. dominica, T. granarius, and Callosobruchus maculatus and Chinensis.

Stable to humidity and temperature, Deltamethrin has an half life varying from 9 to 25 months.

DIFFERENT EFFECTS OF SOME ANTIOXIDANTS ON THE PRODUCTION OF AFLATOXINS ON STARCHY AND OILY SEEDS

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Our previous works have demonstrated that butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and cysteamine (CYS) were capable of inhibiting, at different extent and without affecting the fungal growth, the output of aflatoxins and their congeners induced by Aspergillus parasiticus (NRRL 2999) and its mutant strains, namely NOR-1 (producing norsolorinic acid + averantin + aflatoxins), AV-1 (producing averufin), AVN-1 (producing averantin) , VER-1 (producing versicolorin) and A. flavus (ATCC 32591) (producing sterigmatocystin + O-methylsterigmatocystin). On the contrary other antioxidants , such as Vitamin E, Vitamin C, reduced glutathione and cysteine, in the same cultural conditions, significantly increased the production of mycotoxins.

Following these results, we have tested the effect of different concentrations (from 0.01% to 0.1% w/v) of BHA, BHT, Vitamin E and Vitamin C (suspended in 0.05% alkylamide betaine in order to obtain a uniform distribution) on sunflower, maize and wheat seeds properly moistened and inoculated with 10^5 conidia of the above Aspergillus strains. Analysis of mycotoxins was performed by HPLC on RP-18 column. Fungal growth was measured by GC on SP 2330 column, as increased concentrations of hexosamine (glucosamine plus galactosamine) present in the chitin of fungal wall. The stability of each antioxidant was determined, both in presence and in absence of fungi, by HPLC on RP-18 column. As "in vitro" only BHA and BHT significantly reduced($p< 0.01$) mycotoxin production on wheat, maize and sunflower seeds. Their action, which takes place essentially by preventing mould growth, was more marked on starchy than on sunflower seeds($p < 0.05$).

This effect might be ascribed to:

1)the higher level of oxidase enzymes(which are capable of oxidizing antioxidants) on the hull of sunflower as compared to starchy seeds;

2)the higher concentration of lipids(which support fungal growth by acting as carbon source) on the surface of sunflower as compared to starchy seeds.

This work was supported by European Community Grant TSD. A.111.I.(S).

ACID TREATMENT OF GRAIN - A RISK FOR GROWTH OF ASPERGILLUS FLAVUS/PARASITICUS AND AFLATOXIN FORMATION

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SUMMARY

In 1986 extensive growth of A. flavus/parasiticus and aflatoxin formation was frequently demonstrated in a survey of grain from 146 Swedish farms using acid preservation. Aflatoxins were found in grain samples from 41 % and 35 % of the farms using 70 % and 85 % mixtures of formic acid, respectively. In grain treated with propionic acid, aflatoxins were found in samples from 5 % of farms using this acid. Strains of A. flavus/parasiticus isolated from acid treated grain were used to further study the risk for aflatoxin formation in connection with acid preservation.

When cultivated on aflatoxin producing agar positive reactions (i.e. blue green fluorescence) were more common (56 %) among strains originating from formic acid treated grain than among strains originating from grain treated with propionic acid (4 %).

Spores and mycelium from 113 strains of A. flavus/parasiticus were cultivated on Czapek Dox agar containing formic acid or propionic acid in concentrations of 0.1 %, 0.3 % or 0.5 % (by weight) in order to study differences in acid torerance among the strains. Growth from spore inoculations were observed at 0.1 % and 0.3 % formic acid and 0.1 % propionic acid while growth from mycelium inoculations only occurred at 0.1 % formic or propionic acid. A higher tolerance towards formic acid was noticed for spores from strains of A. flavus/parasiticus isolated from grain treated with formic acid compared to those isolated from grain treated with propionic acid.

Moist barley (75 % d.m.), untreated, treated with 0.5 % formic acid or 0.3 % propionic acid was inoculated with 11 strains of A. flavus/parasiticus. Preliminary results from samples taken over a five month period indicate that both growth and toxin production from the inoculated strains is favoured by the presence of formic acid, while propionic acid seems to suppress both growth of A. flavus/parasiticus and aflatoxin formation.

THE USE OF BEHAVIOUR MODIFYING CHEMICALS IN MONITORING INSECT PESTS OF STORED PRODUCTS

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ABSTRACT

The ability to detect insect pests at low population levels is becoming increasingly important in integrated pest management strategies for Stored Product Pests. Sex and aggregation pheromones, plus food attractants in some cases, are being used in a variety of trap designs to monitor both moth and beetle pests of bulk and processed grain. Sex pheromone based trapping devices have been shown to give distinct advantages in warehouse moth surveillance. Improved detection and significant reductions in infestation levels have been achieved in many industries through the use of such systems. Other benefits derived from their use include the more accurate timing of control applications and substantial savings in material and labour costs. The use of behaviour modifying chemicals in trapping devices for beetle pests of stored products is not as advanced as that for moths and, as a consequence, the widespread use of such devices has not been achieved. Nevertheless, products are available which can be used to monitor beetle pests in bulk grain (pitfall and probe traps) and in processed food (bait bags and Storgard traps). These devices have been accepted by the end users to varying degrees usually dependent on their reliability, trapping efficiency and ease of use. Much development work is in progress to improve on these devices with the eventual hope of integrating them successfully into pest management strategies for stored product insects.

A RECIRCULATORY FUMIGATION TECHNIQUE FOR STACKED COMMODITIES

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INTRODUCTION

Imported groundnut kernels destined for human consumption often need to be fumigated with methyl bromide (MeBr) during the winter in unheated warehouses. Conventional dockside and warehouse stack fumigations use vaporised MeBr under gas-proof polythene-type sheeting.

Collaborative trials between ADAS (Wildlife and Storage Biology) and Northern Fumigation Services Ltd identified the adverse effects of low temperature fumigation as:

- Excessive layering of the heavier-than-air fumigant
- Excessive over-dosing of the lower sacks in the stack
- Very high bromide residues in the lower sacks
- Considerable under-dosing of the upper sacks in the stack
- Very uneven concentration-time products (CTPs)

In order to achieve satisfactory CTPs to control the infestation the dosage of MeBr must be high enough to cater for the relative scarcity of fumigant at the top of the stack which results in high concentrations of gas around the base of the stack and gross contamination of the nuts.

METHODS

Further collaborative trials were designed to overcome these problems by achieving rapid thorough mixing of fumigant and air by means of simple recirculation beneath the gas-proof sheet.

The recirculation system consisted of a large diameter corrugated and perforated plastic land drainage pipe laid at the base of the stack, to collect the heavy fumigant, which was then redistributed via an electrically powered radial fan through the original heavy duty polythene delivery pipe, branched manifold and perforated layflat tubing across the top of the stack. The recirculatory fan was connected up immediately after the initial gassing, and was run for a total of 3 hours. Gas concentrations were measured using nylon sampling line and a thermal conductivity meter.

RESULTS

The conventional, non-recirculatory fumigation resulted in MeBr concentration ratios from the bottom to the top of the stack 3 hours after gassing of 3:1, and 5 hours after gassing 2.5:1. With the recirculatory method in use, mean concentration ratios were 2:1 after 30 minutes, and 1.4:1 after 3 hours. There was almost complete mixing of the gas and air after 5 hours.

CTP means in the standard trial ranged from 1300mg h/l at the base to 620mg h/l at the top of the stack ($65g/m^3$; 0-4°C) and CTP means in the recirculatory fumigation ranged from 365mg h/l at the base to 300mg h/l at the top of the stack ($50g/m^3$; 3-9°C).

The equipment used was cheap, and simple to operate, and further refinements of the technique should be pursued to the benefit of the food industry and the consumer.

THE EFFECT OF TWO AMMONIUM PROPIONATE FORMULATIONS ON GROWTH IN VITRO OF ASPERGILLUS SPECIES FROM HAY

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SUMMARY

Normally, hay must be dried to a water content of less than 20% before baling to prevent microbial growth, associated heating and loss of nutrients. To shorten the field drying period, chemical preservatives are often added to moist hay. These preservatives are mainly based on organic acids and their salts: ammonium salts of propionic acid are particularly widely used for this purpose.

Screening of potential preservatives for control of micro-organisms in vitro has often been carried out without considering the effects of water availability (water activity, a_w). Of particular interest are the *Aspergillus* species, because of their ability to grow in intermediate moisture content hays. Many important *Aspergillus* spp. involved in spoilage of agricultural products, particularly members of the *Aspergillus glaucus* group, *A.niger* and *A.versicolor* grow optimally at 0.90-0.95 a_w (= 29-35% water content). This poster compares the effect of two widely used chemicals in proprietary formulation, fully neutralized (AP) and half-neutralized (H-AP) ammonium propionate on the growth of *Aspergillus* spp. at different levels of water availability.

The efficacy of the two chemicals against growth of *Aspergillus* spp. was tested on 1% malt extract agar at different levels of water availability in the range 0.995-0.85 a_w (= 40-25% water content of hay). In general, *Aspergillus flavus*, *A.fumigatus*, *A.nidulans*, *A.niger* and *A.versicolor* were more sensitive to the H-AP in the range 0.995-0.95 a_w than to AP. At 0.90 and 0.85 a_w growth of most species occurred in the absence of chemicals but only of a few species in 27 mm/l AP. Of the *Aspergillus glaucus* group members tested *A.sejunctus* was most tolerant, growing in 27 mm/l H-AP and 54 mm/l AP at 0.85 a_w. The number of days for growth initiation in 27 mm/l H-AP was greater than with AP at different levels of water availability.

The complex interaction between substrate water availability, temperature and microbial growth should be appreciated when screening potential preservatives for agricultural products.

ELECTRONIC RODENT DETERRENTS : DO THEY WORK?

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SUMMARY

Over the years Rentokil's Research and Development Division has been asked to evaluate several electronic rodent deterrents. These have fallen into three classes : ultrasonic, electro-magnetic and vibrational. All are supposed to repel rodents or prevent them from feeding and reproducing. This poster looks at a "typical" example of each of them.

The ultrasonic device was tested against a population of mice confined to two rooms connected by an opening 30 cm square. Both rooms provided adequate food, water and harbourage. The machine was placed in one of them. Food consumption in each room was recorded for four weeks before treatment and for five weeks with the machine on. Total consumption declined but there was no change in feeding locality. Mice still fed in areas directly exposed to ultrasound.

The electro-magnetic and vibrational devices were tested against rats in an 18 x 18m outdoor pen connected by tunnels to other smaller pens and part of a nearby building. All areas provided food, water and harbourage. The machines were erected in one corner of the main pen. Food consumption from different parts of the site was recorded for a four week pre-treatment period and then for four or eight weeks with the machines on. The electro-magnetic device may have caused a small decline in total food consumption but did not repel animals from feeding sites nearer to the machine. The vibrational device made little difference either to total consumption or to feeding locality preference.

These results indicate that electronic devices of the kinds tested are unlikely to be effective for controlling rodent pests.

TRAPS FOR DETECTING STORED PRODUCT INSECTS: ARE THEY RELIABLE?

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OBJECTIVES

Inspection of food processing premises for the presence of stored product insects (SPI) is time consuming and therefore costly. Insect traps offered the hope that SPI could be reliably detected, and inspection concentrated on infested areas. Before selecting trap types for use by Rentokil inspectors we needed to know how close traps had to be to infestations to capture insects, whether capture rate reflected infestation level, and whether traps were simple to use.

METHODS

The traps evaluated were :- (1) Bait trap - foodstuffs in mesh envelope (MAFF design), (2) Storgard trap - harbourage, food oil, *Tribolium* aggregation pheromone, (3) Moth funnel trap - funnel with *Ephestia* female sex pheromone and dichlorvos strip.

Trials were carried out by experienced inspectors in 12 food processing premises. At 1-4 week intervals trap catches were recorded. Levels of infestation and distances of traps from nearest infestations were noted.

RESULTS

Bait and Storgard traps captured mainly *Tribolium confusum* and *Ptinus tectus*. Even when placed on top of infestations some traps failed to capture insects. Trapping rates did not reliably reflect infestation levels. Removal of all captured insects from traps was difficult. Traps sometimes captured insects before infestations were located by inspection.

Moth funnel traps captured mainly *Ephestia kuehniella*. 80% of the traps within 5m of infestations always captured moths. Capture rates reflected infestation levels. Traps often captured moths before infestations were located by inspection.

CONCLUSIONS

Bait and Storgard traps are not reliable enough for routine use by experienced inspectors. They might be used to check apparently insect-free areas, and would be helpful to non-specialist inspectors.

Moth funnel traps are a good inspection aid and may be used to detect established moth infestations, to monitor infestation levels, and to give early warning of incoming infestations.

A SURVEY OF THE INCIDENCE OF BOOKLICE IN THE DOMESTIC KITCHEN ENVIRONMENT.

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INTRODUCTION

Over the past 20 years the tropical booklouse Liposcelis bostrychophilus has developed as a major cause of consumer complaints in several sectors of the food industry, particularly those concerned with flour. A major question is; are booklice common household inhabitants and so able to get into food once it has been put in domestic kitchen cupboards, or do they contaminate food before it is bought by the general public? This survey was designed to establish how common booklice are in kitchens and to see if there are any links between the presence of booklice and features of the household environment, food storage conditions or the buying patterns of the occupants.

METHODS

The survey asked participants to place 5 small "pads", which were attractive to booklice, amongst their stores of flour and other dried foods during October 1986 and to complete a questionnaire. The returned pads were examined for evidence of booklice and other arthropods. The questionnaire was designed to identify aspects of the domestic situaton which may correlate with the presence or absence of booklice, such as environmental features, storage methods used and shopping characteristics.

RESULTS

Everyone on the payroll list of King´s College, London (some 1600 people) and a further 200 friends and contacts were invited to take part in the survey. Of these 1800 requests, 541 (c.30%) households responded by returning the questionnaires and "pads". Seventy eight (14.4%) of the respondents returned "pads" which contained Liposcelis eggs, nymphs or adults. There was no statistically significant geographical bias to these positive results. An analysis of the questionnaire data showed no correlation between the incidence of booklice and any environmental characteristics of households (the presence, absence or combination of condensation problems, central heating or double glazing),or the ways in which dry foods were stored or where they were kept, or with any particular flour brand, the rate of its replenishment or with any specific retail outlet.

The replies to the questionnaire also illustrated the way people respond to finding booklice in food. Assuming that all complaints to shops are passed on, then the manufacturer will only get to hear of about 50% and the local Environmental Health Offices only about 3% of booklouse related complaints in food.

CONCLUSIONS

Liposcelis bostrychophilus is not a common household pest and there is no evidence that differing conditions in houses affect their suitability as booklouse habitats. The lack of any correlations in the data strongly suggest that booklice appear in houses on a random basis and this does not rule out the possibility that the booklice are introduced to houses in food products.

THE ROLE OF MATING STIMULI AND SEED AVAILABILITY IN DETERMINING THE EGG MATURATION RATE OF THE BEAN WEEVIL CALLOSOBRUCHUS MACULATUS.

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Often the reproductive output of an insect is assessed from the total number of eggs laid or from egg laying rates. However, these measurements may be misleading because many insects do not lay all the eggs that they mature. Egg maturation and oviposition rates may therefore be separate measures of egg production influenced by different factors. We investigated the relative importance of seed availability and presence of mates in controlling egg maturation and oviposition rates in the bean weevil Callosobruchus maculatus (F.) (Coleoptera: Bruchidae).

Four experimental treatments were used: One group of female beetles was given access to both males and seeds; a second was given neither of these, and the remaining two groups were given either males or seeds but not both. On each day for the next five days individuals from each group were dissected and the number of mature eggs counted. An additional group of virgin females was dissected within thirty minutes of emergence from the seeds as adults.

Females dissected on day 0 held about six eggs in their ovarioles, indicating that egg maturation is initiated before emergence. However, only females, given access to both males and seeds continued to mature eggs at a high rate (c. 12 eggs per day). Females that were given mates but not seeds were the only group which laid appreciable numbers of eggs at sites other than on seeds. Therefore mating may 'prime' the female for egg laying, but mechanical stimuli emanating from ovarioles bulging with eggs inhibit further maturation of eggs.