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STRATEGIES FOR THE USE OF MODIFIED ATMOSPHERES FOR THE TREATMENT OF GRAIN

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by

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ABSTRACT

This research project covered two different areas of investigation to improve the use of modified atmospheres (MA) as a means for controlling insect and mite pests of grain. The first area was concerned with the testing of various gas generation systems for continuous flow treatments within common grain storage structures. From these a suitable candidate system was selected and further trials were conducted within a range of silos of a specific design type, with integral basal aeration ducts and augers. These gave a series of flow rates which were required for the maintenance of the target 1% oxygen level within each silo. The objective was to develop a reliable application rate schedule for use with nitrogen-based controlled atmosphere generators within any size of silo of this design.

The second area of research was concerned with the effect of the candidate MAs on the condition of the stored grain, and on the pest species themselves under the environmental conditions prevalent in grain stores. The candidates included the juvenile stages of six species of grain beetle;

- *Cryptolestes ferrugineus* (Stephens) (the rust red grain beetle),
- *Oryzaephilus surinamensis* (L.) (the saw-toothed grain beetle),
- *Rhizopertha dominica* (F.) (the lesser grain borer),
- *Sitophilus granarius* (L.) (the granary weevil),
- *S. oryzae* (L.) (the rice weevil),
- *Tribolium castaneum* (Herbst) (the rust red flour beetle);

the diapausing larvae of the warehouse moth,

- *Ephestia elutella* (Hubner),

and five species of mite, including all stages;

- *Acarus chaetoxysilos* Griffiths,
- *A. farris* (Oudemans),

- *A. siro* L.,
- *Lepidoglyphus destructor* (Schrank),
- *Tyrophagus longior* (Gervais).

Three MA generation systems were assessed within various silos: carbon dioxide from 'minitanks', nitrogen generated by pressure-swing adsorption (PSA) and burner gas, generated by burning air in propane to give a nitrogen-rich, low oxygen atmosphere. The burner gas generation system was the least expensive with costs as low as £0.34/tonne of wheat, while the carbon dioxide with an exposure of seven days for the control of the most tolerant pest present, resulted in costs of £14.60/tonne with malt, partly because of the high sorption of the gas by the malt.

Three further trials were carried out using the burner gas generator on silos filled to capacity containing 292 or 1150 tonnes of wheat, or 810 tonnes of malting barley. The gas was introduced into the silo through the aeration duct and a plastic sheet was placed over the surface of the grain to stop the gas escaping into the headspace. A good level of sealing of the auger was necessary to prevent loss of the generated atmosphere. The amount of gas loss which occurred was dependent on changes in wind speed and in temperature. The maintenance flow rates achieved for each silo were used to generate a relationship for the rate required for any silo size. The results showed that 100 tonnes of grain would require 3.4 m^3/h, and 1000 tonnes would require 11.6 m^3/h.

The germination potential of the malting barley was not affected by a three-week exposure to burner gas with 1% oxygen or less. Pre-exposure samples from the 810 tonnes of malting barley gave mean germination potentials of 98.1%. This was not changed after the exposure period spent in the silo's aeration duct, the mean germination potential being 98.3%.

The juvenile stages of the insects were tested in low oxygen and raised carbon dioxide concentrations at 15°C and 70% r.h. *S. granarius* was the most tolerant beetle species and required 38 days exposure in 99.5% carbon dioxide, 60 days in 99.5% nitrogen and 65 days in burner gas with 0.5% oxygen. *T. castaneum*, *C. ferrugineus* and *O. surinamensis* were far less tolerant and required a maximum exposure length of ten days.

E. elutella larvae in diapause were tested at 10°C and 20°C. A higher level of mortality was always achieved at the higher temperature in the same period of time. Larvae were killed after 28 days at 10°C in 90% carbon dioxide, while only 46.5% mortality was achieved in the same time with burner gas, and no mortality had occurred after 38 days with 99% nitrogen, both at the same temperature.

The same conditions were used for the mite tests as for those with the juvenile insect stages. Eggs were the most difficult stage to kill for all species. *T. longior* was the most tolerant species. Burner gas (0.5% oxygen, 10% carbon dioxide and 89.5% nitrogen) was the most effective and gave complete mortality after 16 days, followed by 99.5% carbon dioxide and 0.5% oxygen which gave the same result in 20 days and finally 99.5% nitrogen and 0.5% oxygen which required 22 days exposure. Increasing the oxygen level to 2% in all of these mixtures made the carbon dioxide more effective with an increase in exposure of only six days whereas nitrogen and burner gas required a 12 and a 20-day extension respectively. Even 60% carbon dioxide/40% air mixture was more effective than these two mixtures, requiring a 30-day exposure.

OBJECTIVES

1. To test several MA generation systems for continuous flow treatment and to select the most efficient for trials on different-sized grain silos. The results from these trials would provide the minimum flow rates required to hold the silos at the required 1% oxygen level. This information would be used to the develop a maintenance flow rate relationship which would apply to grain silos of any size.

2. To test the response of pest species to candidate MAs under a number of specific environmental conditions related to control problems in practice.

INTRODUCTION

Modified atmospheres (MA) offer an alternative strategy for insect control in stored grain especially as a replacement for pesticide admixture. This technology involves the alteration of the natural ratio of the atmospheric gases, nitrogen, oxygen and carbon dioxide, to render the atmosphere in stores inimical to pests (Banks and Fields, 1995). MAs are now used in the agricultural and food industries for disinfesting, preserving and protecting raw materials from the time of harvest through to the packaging of finished products (Bell and Armitage, 1992). They can be combined well with the present strategies of drying and cooling and could have an important role in integrated systems for the protection of grain, particularly where residue-free *in situ* treatments are needed (Banks *et al.*, 1991). Sophisticated markets expect that grain should be pesticide-free or with much lower residue levels than those that used to be acceptable. There is also the problem of the development of resistance by target pests (Banks and Fields, 1995). Legislative restrictions also make other alternative treatments, such as fumigation, much more expensive (Banks *et al.*, 1991). Adoption of a nitrogen-based MA treatment at the main grain export terminal in Australia was driven by such problems (Banks, 1994). In fact, the present climate in the UK with the long warm autumns preventing the adequate cooling of stored grain, creates a good opportunity for use of controlled atmospheres which benefit from higher temperatures. At 30°C the exposure period is less than 14 days with 1% oxygen to give control of the pupae, the most tolerant stage of granary weevil, *Sitophilus granarius* (L.) (Bailey and Banks, 1980), whereas a 45-day exposure is necessary to achieve the same result at 15°C (Reichmuth, 1990). It is important to maintain the oxygen level below 1%, though 2% oxygen was given as the upper limit for effective MAs by Bailey (1955).

It is important that the use of MAs does not adversely affect the quality of the stored grain. In this respect a nitrogen-based atmosphere is generally regarded as more effective than carbon dioxide. The use of continuous storage for up to a year did not have a detrimental effect on the germination or end use properties of wheat, rice or barley (Ouye, 1984). Germination potential or germinative energy is an important attribute of malting barley and it is important for the maltsters that at least 95% of the grain present germinates rapidly and at the same time (Duffus and Slaughter, 1980). This ability is much more likely to be lost through climatic conditions of storage such as high temperature which is much more injurious to wheat seeds than MA storage (Fleurat-Lessard *et al.*, 1994). MAs may also be advantageous as they are able to extend storage life of grains at moistures normally considered marginal for safe storage and do not allow fungi to multiply at lower water contents (Banks *et al.*, 1991). However there is a need for the total exclusion of oxygen to ensure the complete inhibition of mycotoxin production (Bell and Armitage, 1992). Once air is restored fungal growth is resumed and mycotoxins are produced as normal.

Various methods are available to generate MAs and various atmospheric gases are used. This project aimed to assess the attributes of each system and then to choose the most ideal generator for the determination of a relationship between maintenance flow rate and silo capacity. Some systems based on carbon dioxide or nitrogen rely on generating gas from liquid and require the delivery of cylinders or the installation of an expensive storage facility. An obvious advantage may be derived by on-site generation. Two examples of these systems pass compressed air through a separating mechanism to remove oxygen and leave pure nitrogen. Pressure swing adsorption (PSA) systems use a molecular sieve of coke whereas an air separation system employs a hollow fibre membrane. They both involve high capital outlay and require a large electrical supply which is not always available at commercial grain stores.

An alternative system produces an oxygen-deficient atmosphere by burning the oxygen in a propane flame. It has the advantage of on-site generation but there is still the initial capital cost. Like the systems employing a separating mechanism, this method relies on a constant supply of the generated atmosphere to the silo to maintain the low oxygen level required. Trials have been conducted for some years (Bell *et al.*, 1991, 1993a, b) with this unit and an alternative system has also been developed in the U.S.A. (Storey, 1980a, b; McGaughey and Akins, 1989). The U.K. system is cheaper to run as it does not require an energy-hungry electric compressor to run the cooling system. The unit only needs a 13 amp supply and therefore is easily used in farm situations. Uncertainties still remain as to the degree of modification required to render an enclosure suitable for MAs, the required gas supply rate and the economics of MA gas supply (Banks and Fields, 1995).

The trials described here represent an investigation into the various alternative methods for MA production and the ease of use and relative economics of each system. After selection of the most suitable system, the possibility of producing a dosing schedule for use with nitrogen-based MAs for bottom-aerated grain silos was investigated. These silos are the type commonly found on farms and at grain stores, and the schedule would provide maintenance rates for any size of silo.

Further work was carried out in the laboratory in support of this field work to discover whether the chosen atmosphere had any adverse effect on the stored grain and to assess the relative efficacies of the different MAs on various pests of stored grain. The germination potential of malting barley, the stored grain with the highest commercial value was assessed after exposure to the burner gas MA. The juvenile stages of *Cryptolestes ferrugineus* (Stephens) (the rust red grain beetle), *Oryzaephilus surinamensis* (L.) (the saw-toothed grain beetle), *Rhizopertha dominica* (F.) (the lesser grain borer), *S. granarius* (the granary weevil), *S. oryzae* (L.) (the rice weevil), and *Tribolium castaneum* (Herbst) (the rust red flour beetle), diapausing larvae *Ephestia elutella* (Hubner) (the warehouse moth) and five mite species, *Acarus chaetoxysilos* Griffiths, *A. farris* (Oudemans), *A. siro* L., *Lepidoglyphus destructor* (Schrank) and *Tyrophagus longior* (Gervais) were all tested in three MAs, namely carbon dioxide, burner gas and nitrogen, at temperatures used for storing grain.

MATERIALS AND METHODS

FIELD TRIALS FOR THE ASSESSMENT OF THE MA GENERATORS

The Sites

Five different-sized grain silos located on different silo complexes were chosen. They were all constructed from curved sections of galvanised steel which were corrugated, except the silo at Stonham Aspall which had smooth sides. The sheets were bolted together with sealant used at the joints and mounted on a concrete base. Each base incorporated a series of aeration ducts, either hexagonal in shape or a set of fingered ducts, fed by a single inlet, and an auger. The latter was fed from a central opening and emerged from the silo base diametrically opposite the aeration duct. The down pipe from the auger to the main conveying system was disconnected. A gas-tight polythene sheet was wrapped around the auger tube and drive motor and taped to give a leak-proof seal. This polythene sheet was then anchored to the side of the silo. The silos used at Wallingford were a sixth and quite different design. They were hexagonal in shape and constructed from concrete. They were inside a building and were integral with its structure. Each silo had a conical bottom connected to the conveying system which was closed off for the trials. Details of all the silos and their locations are given in Table 1 (Pages 65 and 66).

Trial 1: Stonham Aspall

The silo selected was one of eight Bentall 150 tonne capacity silos in a line on a farm site. The silo had been fully loaded but about 30 tonnes had been removed in the month prior to the trial. The grain surface was levelled as the first stage in the preparation of the silo for gas application.

Trials 2 and 4: Wallingford

The silo selected was in the middle of a row of six 260-tonne capacity cells and was adjacent to the outer wall of the building.

Trial 3: Shrewton

Some barley had to be removed from the roof space of the silo to allow easy access. The grain surface was levelled with grain shovels as the first step in the preparation of the silo for dosing. The silo was part of a complex of four rows oriented in a north to south direction, the storage facility of a farmers' co-operative. The silo was the second one from the southerly end of a row of 900 tonne silos. There was a row of 900 tonne silos to the west and one of 2000 tonnes to the east.

Trial 5: Micheldever

This was a large grain store with a capacity of 45,000 tonnes. There were two large floor stores and forty silos. The silo selected for the trial was loaded with 810 tonnes of malting barley. It was situated on the north side of the complex at the end of a row of six similar silos oriented in an east to west direction. There was a band of trees screening it to the north and it was surrounded by silos on all other sides. The malting barley in the silo formed a flattened cone in the roof space.

Trial 6: Hursley

The silo was in the centre of a line of eight of similar capacity oriented in an east to west direction. To the south and separated by a grain conveying system was a row of six 700 tonne silos. On the east was the grain handling buildings and to the west and north were fields. This silo was also very full with 292 tonnes of wheat, forming a flattened cone in the head space.

Trial 7: Linton

This was another large store with a capacity of over 35,000 tonnes all grain being stored in silos. The silo used was on the eastern edge of the store, facing open fields, in a row of four of similar size which was oriented north to south. This silo was also filled to capacity with 1150 tonnes of wheat.

MA Generators

Trials 1 and 3: Nitrogen Generation System

The nitrogen was produced from an air separation unit, a PSA system, which was driven by compressed air. It was based on two beds of molecular-sieve coke. The differing rates of diffusion of oxygen and nitrogen within the micropores of the coke provided the means of separation of these two gases, the oxygen being adsorbed along with the water vapour and the carbon dioxide. For continuous operation there is a switching between the two beds, each alternating in a cycle of adsorption and regeneration with the rise and fall of pressure within each sieve at opposite times to each other. While one bed is pressurised and the nitrogen is passed through to a holding tank, the other is vented off with the oxygen-rich atmosphere passing to waste. Each part of the cycle lasts about a minute. The nitrogen was fed from the holding tank to the silo. The purity of the nitrogen is limited by the flow rate that is required.

The trial at Stonham Aspall used a small experimental unit which was still in the developmental stage. The unit used at Shrewton was a large industrial design working well below its maximum capacity and was capable of producing the necessary flow without raising the oxygen content above 0.3% as recorded by its own meter. The nitrogen was fed from the PSA machines via a 6 mm plastic tube for the first trial and a 2.5 cm copper pipe for the latter. Both these were attached to their respective silo aeration ducts via a coupling in an aluminium plate made to cover each opening. These were held in place by elastic tape and 'G'-clamps.

The compressed air for the smaller PSA unit was provided by a compressor which was able to run off a 440 volt, three phase supply available at the site. However the larger unit used at Shrewton was driven by a 37 kilowatt compressor and there was no electrical supply available to accommodate its power requirement. Power for the compressor was provided by a diesel generator hired for this purpose.

Trial 2: Carbon dioxide

The trial required a special installation to receive the six 'minitanks' which were used to hold the liquid gas. Each was capable of holding 190 kg of liquid carbon dioxide. Each 'minitank' could be isolated which allowed for disconnection once a tank was empty without disrupting the flow of gas to the silo. 2.5 cm diameter copper piping was used to take the carbon dioxide to the silo which had to be modified slightly to receive the dosing line. A threaded spigot was fashioned for the hopper sampling point plate for application of the gas to the bottom of the silo and for the top hatch cover when introduction of gas was switched to the top of the silo. The silo was provided with a vent, to assist during the purge, by running a 2.5 cm diameter hose from the spigot in the hatch to an adjacent window on the top floor.

Trials 4 to 7: Propane burner

A propane-fuelled inert atmosphere generation system was used for these trials. Trial 4 used the latest model of the Aerogenerator (Aerogen Ltd., Alton, Hants). The following three trials employed the original prototype of this machine which had been considerably upgraded at CSL since its original construction. These generation systems burned a calibrated premix of propane and air, with an optimal fuel to air ratio of 1:25 (v/v), in a closed combustion chamber. The resulting mixture of gases (mostly nitrogen, water vapour, carbon dioxide and oxygen) was passed to a condenser where it was cooled. The maximum theoretical output of the two machines was about 20 m^3/h with an atmosphere of less than 1% oxygen.

In the prototype the condenser was water-jacketed and contained many spiral-wound tubes to provide the largest possible surface area for heat exchange. The water which contained 50% ethylene glycol was cooled by an absorption refrigerator driven by waste heat from the combustion process. Most of the water vapour was removed as liquid. It was possible to control the amount of cooling required with a secondary heat exchanger which was fitted in the water-glycol circuit between the refrigerator and the condenser. This was fan-driven with the air blown over radiator vanes. The air was sucked via a large duct from one side of an open-ended box placed over the warm exhaust draft from the refrigerator. Shutters in the sides of the box provided a means of control over the amount of warm air drawn by the fan.

The prototype machine was adjusted to give an output with less than 1% oxygen and a relative humidity of about 50% for each of the flow rates used in the three trials. The cool dry gas mixture was then passed to the treated silo, assisted by a small fan to overcome the back pressure of the silo and the connecting 5 cm flexible hose. The output was connected to the aeration duct of the silo under treatment via a plate which was made to fit the dimensions of each duct opening and a connection to take the hose was fitted in the middle. The output was measured by a flow meter which had to be read manually. This only registered the flow rate rather than being a controller of the rate.

The prototype machine was mounted on a road trailer 3 m x 1.5 m. For weather protection it was covered by a tarpaulin stretched over a frame and was open at the ends to allow a good circulation of air around it. Fuel gas was supplied from two banks of four 47 kg cylinders connected through an automatic change-over valve. The empty cylinders were changed as required to give an uninterrupted supply.

Monitoring the Gas Constituents and Preparations for Atmosphere Application

All gas monitoring of the effluent content and the atmosphere within the silos was carried out using 2 mm nylon sampling lines. These were inserted into the grain and pushed down using metre lengths of rod, which were screwed together. The first section had a slip catch attached which allowed the rods to be drawn up leaving the line at the required depth. The lines were placed in two columns, in the centre and 0.5 m from the silo wall where the roof entrance hatch was positioned. The lines were located at metre intervals from 6m depth to the surface and at the sides on the surface in a cross pattern determined by the hatch's position. A line was also attached to the MA generator's outlet and, in the some of the trials to the aeration duct.

For trials 2 and 4, a different system was used in the concrete cells. Before loading with grain or malt a steel rope supporting gas sampling lines at 2 m intervals (a total of 14) was lowered into the free space, suspended from a steel bar across the top hatch grid. The sampling lines and thermocouples were fed through a small hole in the hatch cover and run via a window to the outside of the building and down to a mobile laboratory housing analytical equipment which was stationed below.

Type T thermocouples were also inserted into the grain to record temperatures throughout the trials. These were attached to rods and pushed into the grain as close to the sampling profiles as possible. In trials 2 and 4, the thermocouples were attached to the steel cable with the gas sampling lines. In all the trials the grain temperature used was recorded at a depth of 4 m except trial 1 where the depth was 2 m in the smaller silo. A further thermocouple attached to the mobile laboratory recorded the ambient temperature. The gas lines and thermocouples were fed out of the silo through the roof hatch, gathered together and passed down to the mobile laboratory where they were attached to the analytical and recording equipment.

A plastic sheet, 70 microns thick, was then placed over the surface of the grain to prevent the MAs leaking into the head space, except in trials 2 and 4. The edges were buried in the grain by the silo walls to improve gas retention further, particularly by providing protection from the wind. This surface sheeting of the grain was not done for trials 2 and 4 as the concrete silos were within a building and were therefore not affected by the wind. However, the upper hatches and plates were sheeted and holes were plugged with plasticine. The shute for incoming grain was sealed with bubble pack and an inflatable bladder.

For trials carried out in the open wind is an important cause of generated atmosphere loss. Wind speed and direction were measured throughout the trials except 2 and 4, by an anemometer (Vector Instruments, Rhyl, Clwyd) which was mounted above the silo, on a walkway or on the silo itself.

For trials 1 to 4, the gas lines for recording the changes in the atmospheres within the silos, fifteen for each trial, were connected to an automatic sampling system. The oxygen content was measured using a microprocessor-controlled HP 5880A gas chromatograph employing a thermal conductivity detector (Hewlett Packard, Bracknell, Berks). There was a lag time between samples to allow the purging of the lines with the fresh sample before the analysis took place. Readings were taken automatically from each line every four hours by using a clock-table programme with the results printed out on a chart recorder. After completion of the trial the oxygen concentrations were adjusted to take account of the 1.1% argon present in the atmosphere. The detection method was unable to differentiate between the two elements. However, the exact quantity was checked by comparing some results with those from a Model 570A paramagnetic oxygen analyser (Servomex Ltd., Crowborough, Sussex). Carbon dioxide levels during trial 2 were compared to those from a Model PA 404 infra-red analyser (Servomex Ltd.).

Trials 5, 6 and 7 used a different sampling and detection system. This could be programmed to move through each of the sampling lines at a pre-set time interval using a Psion Organiser II (Model LZ64). For these trials, the sampler moved to the next line every eight minutes. A complete set of data from each position was completed every two hours. The samples were drawn down the lines using a diaphragm pump and were passed through a bank of instruments contained in the mobile laboratory. A two-minute period was allowed before the data was recorded to allow the readings to stabilise. The instruments were connected in series and consisted of a Series 1400 paramagnetic oxygen analyser, a Series 1400 infra-red carbon dioxide analyser, a Type 1490 infra-red carbon monoxide analyser and a Type 1491 nitrous oxide analyser coupled to a Model 1000D thermal oxidiser (all instruments by Servomex Ltd.). The data from each sample were recorded on a chart in a HR2300 Hybrid recorder (Yokogawa Electric Corporation, Tokyo, Japan). At the same time, recordings were made of the wind conditions and the temperatures.

Assessment of the MA Generation Results

For each trial a record was made from each line and for each environmental condition every six hours. However, for the illustration of the results, only two positions from the centre and two from the side, one at 1 m depth and the other 6 m depth, were used. For the period of the initial purge and at the end of the trial, when the machine was turned off for an assessment of the leak-back rate, a record was made every two hours. The temperature and wind data were used to determine any possible influence they may have had on the gas tightness of the silo.

The efficacy of the flow in lowering oxygen levels was then assessed to produce a suitable purge rate and an achievable maintenance rate for each silo size in relatively calm conditions. The maintenance rate calculation was achieved by taking a mean for the oxygen values at the beginning and at the end of each flow rate period and subtracting the latter from the former. These were calculated after the oxygen in the flow rate had been subtracted to achieve a more representative result. These values were then plotted against flow rate and a regression line fitted. A value for the maintenance rate was identified as the flow rate with no change in the oxygen level.

For trials providing an accurate maintenance flow rate, these values were plotted together and regression analysis was carried out to provide a relationship between any given silo capacity and the maintenance flow rate required. A further comparison was also carried with values from previous work with other MA generators.

MALTING BARLEY GERMINATION ASSESSMENTS

Prior to the start of gassing three samples of the malting barley were taken from the 810 tonne grain bulk, on the surface in the centre, 3 m deep in the centre and 2 m deep at the side of the silo. To test the effect of the MA on the germination of the barley two samples were placed in the aeration duct of the same silo, each in a metal dish, where the grains would be in the most constant 1% or less oxygen atmosphere. This exposure covered the last three weeks of the trial before the machine was turned off. They consisted of a sample from the centre of the grain at 3 m depth and from the side of the silo at 2 m depth.

a) Germination Potential

The technique for testing the germination was that of Henderson (1991). Each of the samples from the silo was reduced using a conical sampler. Four replicates of one hundred grains were placed in four 9 cm petri dishes. Each petri dish contained two white Whatman No. 1 filter papers with 4 ml of water. Each dish was covered with a lid to obtain a good seal. Dishes were placed in a dark cabinet at 18-21°C and were checked once a day for three days and any germinated grains were removed. For the malting industry, germination occurs when the coleorhiza (root sheath) penetrates the husk. The number of grains sprouted after the final day gave the percentage germination of the batch of grain.

b) Water Sensitivity

The same procedure was followed as for a), but 8 ml of water was placed in each petri dish. Only the ventral side of each grain was allowed to touch the paper to avoid drowning the embryo. An assessment of this characteristic is again achieved by a measurement of the percentage germination.

INSECT AND MITE REARING AND TESTING REGIMES

INSECT JUVENILE STAGES

Six species which are all beetle pests of grain and other cereals were tested against various MAs: *C. ferrugineus, O. surinamensis, R. dominica, S. granarius, S. oryzae* and *T. castaneum*. For each species, different strains were used to assess any differences in their tolerance to the MAs (Appendix (Page 121)). The insects used for the tests were from stock cultures. These were maintained by placing 100 mixed age adults in a glass culturing jar (7.5 cm dia. x 14 cm) about one third full of food. Each species was given a particular food mixture:

C. ferrugineus	98 g rolled oats, 48 g wholemeal wheat flour, 10 g brewer's yeast
O. surinamensis	150 g rolled oats
R. dominica	320 g whole wheat
S. granarius	320 g whole wheat
S. oryzae	320 g whole wheat
T. castaneum	190 g wholemeal wheat flour

The stock cultures were reared in controlled environment rooms at 25°C and 70% r.h. except *C. ferrugineus* which was reared at 30°C. The same temperatures were used for the rearing of the cultures to produce the juvenile stages for testing. For these cultures four hundred adults were added to ensure that large numbers of eggs were laid. For all species the exposures were started at the stage of juvenile development that was the most tolerant to the MAs. This meant that the cultures had to be set up in sufficient time before the start which for all the species was the pupal stage. The original adults were removed prior to testing.

For *R. dominica*, *S. granarius* and *S. oryzae,* whose juveniles develop within the wheat grains, the cultures were divided between the containers used for the exposures. For the other species fifty pupae or late fourth instar larvae were counted into each container which were one third full of the fresh culture medium. Three containers were used for each exposure period with a further three used as controls which were kept in the same room as the exposure apparatus. Various different containers were used for the exposures of the insects. These included screw top glass jars (5 cm dia. x 7 cm) which were sealed by nylon mesh held in place by top-less lids, metal mesh tubes (1.5 cm dia. x 100 cm) lined with a tube of nylon mesh and closed with bungs, and glass tubes (2.5 cm dia. x 7.5 cm) sealed with squares of nylon mesh held in place by sections of rubber pipe which were pushed inside the openings of the tubes.

The insects, including the controls, were moved down to the exposure temperature in stages so that they could acclimatise, always keeping 70% r.h. They spent a day at 20°C before moving to 15°C a day before the beginning of the exposure. *C. ferrugineus* spent an extra day at 25°C before the start of this process. At the completion of the longest exposure they were moved back up to their rearing temperatures in similar steps of temperature and duration.

They were then checked weekly for the appearance of adults until emergence from any treated stage was complete. The results were used to calculate the mean emergence for each exposure time as a percentage of the emergence from the control samples for *S. granarius*, *S. oryzae* and *R. dominica*. For the other species the control result was used to correct for mortality that may have arisen from another source. The percentage emergence of the treated was calculated as a proportion of the emergence of the control.

MAs

0.5% Oxygen, 99.5% Nitrogen
0.5% Oxygen, 10% Carbon Dioxide and 89.5% Nitrogen
0.5% Oxygen and 99.5% Carbon Dioxide
2.0% Oxygen and 98% Carbon Dioxide

The gas mixtures were produced using a three channel gas blender (Signal Instrument Co. Ltd., Camberley, Surrey) provided with high purity compressed gases from cylinders. For the high carbon dioxide atmospheres a mixture of this gas and oxygen was used with the latter replaced with compressed air for the 60% carbon dioxide mixture. For the high nitrogen atmospheres this gas was combined with oxygen, and finally for an atmosphere similar to that produced by burning propane in an exothermic gas generator, nitrogen, carbon dioxide and oxygen were used. After the gases were mixed to the required proportions, the gas stream was split into eight with each gas flow limited to 100 ml/min by means of flow control valves. The eight streams were then humidified to 70% by passing the gas over solutions of potassium chloride (Winston and Bates, 1960). Once humidified the gas stream went through the top of the lid and, via a tube, into the bottom of the exposure chamber which consisted of a 5 l desiccator. The exposure containers were placed on a wire mesh layer supported above the bottom of the desiccator.

There was also a vent in the top of the desiccator lid which created a flow-through system as the mixed gas was able to flow out from the desiccator to the atmosphere. This set-up ensured a constant gas mixture within the exposure chamber with no loss through leakage or respiration. The apparatus was kept in a controlled environment room at 15°C and 70% r.h.. Oxygen, carbon dioxide and humidity levels were monitored every day using a Model 570A paramagnetic oxygen analyser (Servomex Ltd.), a Model PA 404 infra-red carbon dioxide analyser (Servomex Ltd.) and a Protimeter DP680 (Protimeter Ltd., Marlow, Bucks) respectively, and were adjusted if required.

At the end of the longest exposure period for each insect the exposure containers and the controls were moved back to their culturing temperatures in similar but opposite steps to those used to acclimatise the insects before exposure. The containers were then checked once a week for any adult emergence. This was continued until all possible emergence had taken place. A comparison was than made with the mean of the exposure results for each time period and those for the controls to see the level of mortality produced by the MAs.

MOTH - *EPHESTIA ELUTELLA*

A) Preparation of Diapausing Larvae and their Testing

(i) Collecting Young Larvae:

Approximately 60 anaesthetised adult moths were placed in a 12 cm plastic sieve and an inverted glass crystallising dish (10 cm dia. x 5.5 cm) were placed over them to prevent escape. A ball of damp cotton wool was used as a source of water to increase fecundity. The sieve was then placed over another glass crystallising dish (12 cm dia. x 6.5 cm) to collect the eggs, and secured in place using tape. The apparatus was then left at 25°C, 60% r.h. for five to seven days which was sufficient time for egg laying and for the emergence of larvae.

A single-haired fine brush was used to transfer two larvae into each of 200 glass tubes (2.5 cm dia. x 7.5 cm) containing approximately 2.5 g moth diet (wheatfeed, de-bittered brewer's yeast and glycerol in the ratio 10:1:2). The tubes were stoppered using squares of nylon mesh material secured with rings of polythene tubing. A minimum of 60 larvae was used for each exposure although, after post-treatment examination, the minimum number of larvae actually tested proved to be 40 because of natural mortality.

(ii) Inducing Diapause:

The larvae were placed in a controlled environment room at 20°C and 60% r.h., with a photoperiod of 12 hours to induce diapause. After approximately ten weeks, the tubes were checked for adult emergence as these individuals would have developed from non-diapausing larvae. When both adults had emerged, the tube was discarded. The remaining larvae were left for a further three to four weeks before testing.

B) Dosing Procedures within the Chamber

i) Carbon Dioxide

Prior to introducing the insects, a vacuum pump was used to reduce the pressure inside the test chamber by the same proportion as the required carbon dioxide level. The pressure was then restored very gradually, using carbon dioxide from a cylinder source, while the contents were stirred using a fan inside the chamber. Carbon dioxide concentrations were measured prior to and during the tests using a Model PA 404 infra-red gas analyser (Servomex Ltd.) and concentrations were maintained using a vent to the atmosphere to allow the introduction of more carbon dioxide to the chamber.

ii) Nitrogen

As with carbon dioxide, a vacuum was used to draw nitrogen into the test chamber from a 'minitank' or cylinder source. The nitrogen concentrations were not measured directly but calculated instead from the oxygen concentrations, measured using a Model 570A paramagnetic oxygen analyser (Servomex Ltd.). Where necessary, gas concentrations were maintained using a vent to the atmosphere to introduce more nitrogen to the chamber.

iii) Simulated burner-gas mixture (1% Oxygen, 12% Carbon dioxide and 87% Nitrogen)

The atmosphere was established by the following steps:

1 The chamber pressure was reduced by 90%.

2 Pressure was restored to atmospheric pressure using nitrogen.

3 The contents were stirred for approximately 30 minutes.

4 The chamber pressure was reduced by 50%.

5 10% of the vacuum was replaced with carbon dioxide.

6 Atmospheric pressure was restored using nitrogen.

Fine adjustments and maintenance of the atmosphere were achieved either by using a vent to the atmosphere to introduce CO_2 or N_2 as appropriate, or by using a gas blender (Signal Instrument Co. Ltd.) to introduce a gas mixture of the correct proportions. Exact measurements of the gas mixture were checked by using the same analysers as for the juvenile insect stages tests.

All the tests were done in either 400 l or 1700 l steel chambers in controlled environment rooms. For the tests at 10°C, the larvae were taken down to 15°C for at least two days and then left at 10°C for a further two days before testing. After the tests were complete the larvae were returned to 25°C and 70% r.h. and were given a 15-hour photoperiod to break diapause. The tubes were then checked for adult emergence. An assessment of mortality was then made after correcting for any control mortality (Abbott, 1925).

MITES

Production of Mites for Testing

All of the species, *A. chaetoxysilos, A. farris, A. siro*, *L. destructor* and *T. longior*, were reared in 50 ml conical flasks on a heated sterilised medium consisting of de-bittered yeast and wheat germ flakes in a ratio of 3 to 1. A bung of non-absorbent cotton wool was used to seal each flask. The cultures of each species which comprised four flasks were kept in desiccators, one species in each. A relative humidity of 80% in the desiccator was achieved by a solution of potassium hydroxide in the bottom (Solomon, 1951). These desiccators were placed in a room with controlled temperature and humidity conditions at 15°C. The mites were cultured every two weeks with half the culture (1.5 g), discarded and then replaced by an equal quantity of new food. The strains of each mite species used have all been in culture at the laboratory for many years.

For testing, the culture medium was divided up and placed in perspex cells (7 x 7 x 0.3 cm) with a circular hole of 4.3 cm diameter. On one side of the cell a white Whatman filter paper was attached using wood glue. This allowed gas to diffuse into the cell but prevented the mites escaping. Approximately 0.2 g of one week old mite culture was placed within each cell, and a large square cover glass the same size as the cell was held firmly over the top by four bulldog clips.

Mites were exposed to the gas mixtures in the same apparatus as used to test the insect juvenile stages. For each exposure period three replicates were prepared, with ten exposure periods with two days between each of these for each test, plus three cells to act as controls throughout the experiment. All cells were allowed to condition at the exposure conditions within a desiccator for a day prior to exposure. The cells were then placed in their particular exposure chambers while the controls remained in the desiccator. After an exposure they were removed from the chamber and placed in a desiccator separate to the controls at 15°C and 75% r.h. They were then checked for adult mortality after one day and again after one week and then every week until nymphs were detected. If no egg hatch had occurred after three months then the cells were discarded. After this an assessment of the time required to give 100% mortality could be given.

RESULTS

APPLICATION RATES

Trial 1 - Stonham Aspall (120 tonnes of wheat)

Oxygen levels fell rapidly during the first day of the purge and then increased on day 2, when the wind speed exceeded 15 m/s, particularly in the centre on the surface of the grain (Table 2 (Page 67), Figure 1 (Page 81)). By day 6 all points had fallen to between 1 and 2% oxygen but on day 9 levels rose again when the wind again increased to 15 m/s, in spite of increasing the flow to 140 l/min for a day. A detector failure in the gas chromatograph interrupted readings over the second week-end which featured further windy weather. On resumption of monitoring on day 13 levels of oxygen remained high but then fell rapidly as the wind dropped. The following day a serious leak was discovered at the join in the dosing line. Measurements with a flow meter indicated a 10 to 20% loss from this source and the coupling was replaced with a gas tight connector and flow was reduced to 80 l/min (0.4% oxygen in the output) (Table 3 (Page 68)). The trial was disbanded two days later when the grain had to be off-loaded.

Trial 2 - Wallingford (260 tonnes of malt)

Application from the base

Purging at 60-80 kg/h resulted in the movement of carbon dioxide up the silo at the rate of 2 m/h. A reduction in the flow to 20 kg overnight slowed the rate of advance to 1m every 3.5-4 h. A large quantity of carbon dioxide was being sorbed by the malt and as a result the flow was increased to 80 kg/h to complete the purge (Table 4 (Page 68)). However even with this high flow rate the progress of the gas did not increase greatly and the gas ran out before the upper section and the head space had reached the desired minimum 40% carbon dioxide level (Figure 2 (Page 82)). The dosing position was then switched to the head space to see if concentrations could be maintained by a flow of 8 kg/h. There was an initial increase in the head space to over 50% but some points within the malt dropped below 40% and the mean gas concentration in the silo steadily declined. A flow rate between 8 and 20 kg/h appeared necessary to hold the silo at over 60% level.

Application to the head space

The initial flow of 30 kg/h distributed carbon dioxide throughout the silo and all positions increased to over 30% by 17 h. Increasing the flow to 60 kg/h (Table 5 (Page 69)) on the second day had little effect on the build up of the gas, whilst a decrease to 20 kg/h resulted in a slight overall loss in gas levels (Figure 3 (Page 83)). There was noticeable lag between the dose supplied and the concentration attained which demonstrated once again that there was considerable sorption of the carbon dioxide. A sample of 25 kg of the malt sorbed over 80 g of carbon dioxide under laboratory conditions at 20°C. The 250 tonnes of malt in the silo could therefore account for a sorption of up to 800 kg of gas. The temperature of the malt remained constant throughout both these trials (Table 2 (Page 67)).

Trial 3 - Shrewton (900 tonnes of feed barley)

For the initial purge the nitrogen was applied at a rate of 35 m^3/h with a nominal output oxygen level of 0.3%. A more accurate level of the oxygen in the input is shown by the reading from the aeration duct (Figure 4 (Page 84)). The oxygen level here remained constant at the 0.8% level while the PSA equipment was running normally. It took 45 hours (1.9 atmosphere changes) to fully purge the silo (Figure 5 (Page 85)). The flow rate was lowered after 50 hours to 23.5 m^3/h (Table 6 (Page 69)) to try and find the flow rate which would maintain the atmosphere below the 1% level. This was then increased to 29 m^3/h after 101 hours as the oxygen level in the silo had risen well above the 1% level (Figure 5 (Page 85)). This rise was due to a prolonged period of high winds (Figures 6 and 7 (Pages 86 and 87)). From 47 to 75 hours the wind speed was consistently over 20 m/s and there was a twelve hour span within this period when the speed was above 25 m/s, the limit of the instrument's scale. However in the period after the wind speed dropped, and before the increase in flow had occurred, between 80 and 98 hours (Figures 6 and 7 (Pages 86 and 87)), the oxygen content was falling. Therefore, the flow rate of 23.5 m^3/h could have been near the maintenance level.

The oxygen content of the atmosphere had both decreased and increased far more rapidly in the central section of the silo than at the side (Figures 6 and 7 (Pages 86 and 87)). This was more noticeable at the 6.5 m depth indicating that the increase in oxygen level was due to a leakage of the atmosphere from the bottom of the silo. This happened even when the aeration duct had 0.8% oxygen which would seem to indicate that the loss was through the auger outlet. This loss would have been helped by the removal of the excess grain before the trial. The centre of the grain would have been loosened especially above the auger outlet. The gas would move along this region in preference to the more compacted grain surrounding it. The auger was also in the lee of the prevailing wind where a low pressure zone would be formed. This would provide the impetus for the movement of the generated atmosphere from the silo. The walls of the silo had effective sealing, highlighted by the fact that the oxygen level at the side of the silo did not rise above 2% throughout the whole period of high winds. The temperature of the silo contents remained constant throughout the trial (Table 2 (Page 67)) and there were no rapid drops in ambient temperature during the course of the trial.

There was a loss of flow from the PSA unit between 121 hours and 173 hours after the start of the trial. The valves controlling the switching between the filter beds became jammed. Although the machine was restarted it was unable to maintain a flow rate. The exact time of breakdown is not known as the power supply to the mobile laboratory was lost at 121 hours. This was due to moisture in the connections for the power supply. There had been two previous short breaks in the power due to this same problem between 18 and 28 hours and between 80 and 98 hours. The data produced after the final breakdown do not provide any more information on the maintenance flow rate. However, they do show the rate of gas loss from the silo and therefore its relative gas-tightness. This seemed to be relatively uniform throughout the silo and gave a rise of 0.7% oxygen/hour.

The final consideration is the cost of the trial. Over the entire period the generator consumed 4424 litres of diesel fuel giving a total cost of £530.88. As the PSA equipment was not functioning properly for the second half, the consumption of fuel may have been higher as the generator was not working at full load. The hire of the generator for three weeks which included delivery was £1565. Therefore, the total cost of this operation from power supply alone was £2095.88 (ex. VAT). This gives a cost of £2.33 per tonne of grain.

Trial 4 - Wallingford (260 tonnes of malt)

Purging at the rate of 22.5 m^3/h resulted in a much lower than expected replacement of oxygen up the silo and after the first 16-18 h only the bottom half of the silo had dropped below 3% oxygen, and only the bottom quarter to less than 1% (Figure 8 (Page 88)). A check at the top of the silo revealed that there was no flow at the vent, indicating that there was a high loss of gas through the structure of the silo.

It was subsequently found that the output from the Aerogenerator was much lower than predicted. Insertion of a flow meter in the output hose revealed a flow of only 9 m^3/h . Release of the damping valve used to adjust gas pressure permitted a maximum flow of 20 m^3/h with little increase in the oxygen level, but gas output temperature increased significantly indicating that the cooling system was being overloaded. At an output of 9 m^3/h temperatures of the output gas ranged from 5 to 15°C, depending on ambient temperature, and the relative humidity ranged between 80 and 96%. A 20% increase in fuel supply resulted in output gas temperatures increasing to over 24°C.

The temperature in the malt ranged from about 18°C at the surface to about 27°C in the bulk, and cooled very slowly throughout the trial (Table 2 (Page 67)). Temperatures at the bottom were much lower and this meant that the gas input temperatures had to be kept low to avoid condensation at the entrance point in the hopper so the flow was maintained at the 9 m^3/h level throughout the trial. A 1% oxygen level was never achieved throughout the silo. An estimated 220 kg of fuel was consumed at a rate of 15 l/min over the six days of operation.

Trial 5 -Micheldever (810 tonnes of malting barley)

The flow rates used throughout the trial from the CSL propane burner and the gas constituents of the output are shown in Figure 9 (Page 89). For the initial purge a rate of 15.6 m^3/h was used. The oxygen levels dropped rapidly starting with positions in the centre (1.5% oxygen/h) and followed by those at the side (0.6% oxygen/h) which were on the opposite side of the silo to the aeration duct entrance and therefore furthest from the point of gas entry. The atmosphere was moving more slowly laterally, as shown by the simultaneous drop in oxygen at all the side positions even though the gas had to travel further to reach the position on the surface of the grain. The decrease at the centre was followed by a rapid increase in the oxygen level although there was no change in the rates of decrease at the side positions. The increase in the central positions was followed by another decrease and a large oscillating cycle developed over time particularly at the 6 m depth (Figure 10 (Page 90)).

The source of this influx of oxygen was the cool and relatively dense night air pushing into the centre of the silo through the auger pipe. The atmosphere within the silo was very warm relative to the ambient air (Table 2 (Page 67), Figure 11 (Page 91)). This large difference was due to the warm weather after harvest which had given little opportunity for cooling the grain. The lower density of the warm atmosphere within the grain facilitated its displacement. This occurred at night when the temperature difference was greatest. The machine's output entering the base of the silo was ineffective at countering this ingress. Once the auger was sealed after 168 hours this problem was removed. The atmosphere in the silo had dipped below the 3% oxygen level by this time. This extended period at the start meant that an accurate purge time could not be produced.

A further eight flow rates were tried after the purge to find a maintenance rate which would hold the silo at a constant 1% oxygen (Table 7 (Page 70)). These were all run for a sufficient time to replace the atmosphere in the silo twice before the flow was changed. The exception was the step down to 9 m^3/h which was increased again before this period as the oxygen level was already rising above 1%. Generally all the flow rates except 9 m^3/h showed a decrease in oxygen level at the centre of the silo where the level was equal to or approaching the value of the input. This was below the target 1% level throughout the trial except during the purge. The increases in oxygen were at the sides of the silo. This would indicate that below a certain flow level there was ingress into the silo by air from outside. This was not driven by environmental conditions as there were no oxygen increases during the highest winds (Figures 12 and 13 (Pages 92 and 93)) nor when the temperature dropped (Figures 14 and 15 (Pages 94 and 95)) once the auger was sealed.

There were two major increases in oxygen during the trial. At 471 hours the machine became iced-up as the power supply had been cut during an overnight storm. The machine re-started but the secondary heat exchanger did not re-start as it is on a separate manual switch. This meant that there was no moderating influence to curb the efficiency of the cooling system. The second occasion was between 745 and 757 hours when the output oxygen level from the burner increased reaching 7.4%. However, it dropped back just as rapidly and there was no obvious explanation for this temporary malfunction. In both instances the oxygen levels had almost returned to previous levels. In the latter case this rapid recovery was achieved with a flow of only 12 m^3/h. This confirmed that a rapid decrease could be achieved to within 1% of the output oxygen level but after this progress was much reduced.

The pattern of generated atmosphere loss from the silo after the flow from the burner was shut off is shown in Figure 16 (Page 96). A gradual loss of the oxygen from the silo occurred with a distinct difference in the rate of loss between the centre and the sides. At the side there was little time or oxygen level difference between the 1 and 6m deep positions, as for the results of the purge. In the centre there was a time difference between the positions with the 6 m position increasing first and at a faster rate. The increased loss in the centre, especially from the bottom, shows that the main overall gas loss occurred from the base of the silo. The loss from the sides at 915 hours was probably due to the wind increase (Figure 17 (Page 97)). It is harder to understand what caused the increase in the oxygen level at the centre after 969 hours. Even with such differences in gas loss patterns, the overall loss for any position in the silo was between 0.1 and 0.2% oxygen/h.

The results from the oxygen content data show that the maintenance rate for this silo lay between 10.8 and 12 m^3/h. Regression analysis ($R^2 = 0.719$) (Figure 18 (Page 98)) produced the relationship:

$$Y = -0.00266X + 0.03053$$

which gave the result of 11.5 m^3/h.

Trial 6 - Hursley (292 tonnes of wheat)

The purge started with a gas flow rate of 15 m^3/h (Figure 19 (Page 99)). The smaller diameter and lower height of this silo meant that the atmosphere reached the 6 m centre position almost immediately. Analysis of the purge showed that there was a slow decline throughout the silo which was followed by a very rapid drop as shown by the positions illustrated in the Figure 20 (Page 100), with a drop of 2.9%/h for the 1m depth at the centre and 4.4%/h for the positions at the side. These changes had occurred by 14 hours but 1% oxygen was not achieved throughout the silo for a further 52 hours (Figure 20 (Page 100)). This meant that there were 6.9 atmosphere changes before the purge was complete. This was longer than expected and indicated that there was ingress of air from some source. There was no correlation of the rises in oxygen observed during the purge with any change of environmental conditions (Figure 21 (Page 101)).

After the initial purge further flow rate adjustments were performed to find a suitable maintenance rate for the low oxygen atmosphere (Table 8 (Page 71)). This was hampered by occasional increases in wind speed which affected the collection of results under otherwise calm conditions. From 614 to 644 hours the wind averaged 10 m/s and this depleted the atmosphere during the 3 m^3/h flow period (Figures 22 and 23 (Pages 102 and 103)). The rate of flow is important in determining the loss due to wind speed as similar wind conditions (average 8 m/s) during the 9 m^3/h flow did not affect the silo's oxygen content. The main loss of oxygen had been from the 1 m depth positions with the loss higher from the centre positions. The loss from the 6 m position at the silo centre indicated that most probably the auger was the site of ingress.

Ambient temperature was also a factor which had influence on the trial (Figures 24 and 25 (Pages 104 and 105)). The bulk temperature was not as high as in Trial 4 but the ambient temperature dropped below freezing at times during the trial, causing a large temperature differential (Table 2 (Page 67)). There were three periods when this happened. In the first instance between 78 and 138 hours the machine itself was affected, icing up and reducing gas flow to the silo. This loss of flow would have allowed cold, dense atmospheric air to push into the silo which was why the oxygen content within the silo increased, particularly at 1 m depth. The next period was between 174 and 234 hours. Unfortunately the whole of the first period and the start of the second spanned the time interval between visits to the site and so freezing of the machine could not be rectified immediately. The flaps on the box above the refrigerator were closed to increase the amount of warm air blown across the heat exchanger. They were then opened again to prevent over-heating. However, from the increase in oxygen levels the icing up must have occurred again the following night with the temperature dropping to -6.9°C. This produced an increase in oxygen throughout the silo, which was most pronounced at the centre with a delayed and reduced effect at the side. The third period, between 398 and 470 hours with the temperature dropping to as low as -3.7°C, demonstrated that as there was no increase in oxygen at the central 6 m depth sample point, the output flow rate had been not affected by icing-up and therefore the oxygen depletion at 1 m depth was due to the temperature difference alone.

The final phase of the trial assessed the rate at which oxygen increased in the silo after the flow was turned off (Figure 26 (Page 106)). The results show that there was an immediate loss from the 1 m depth at the centre. There appeared to be no obvious reason for this, although without the benefit of the active input of atmosphere the moderate wind speed (8 m/s) may have been the cause (Figure 27 (Page 107)). A rise at 6 m depth in the centre followed by a decrease indicated some circulation of gas within the sheeted bulk. There was, however, no comparable increase anywhere else in the silo which would have been be expected to balance these changes.

The next phase was a gradual rise for all sample points. However, there was an increased rise for central sampling positions starting at the 6 m depth which seemed to coincide with the lowering of the ambient temperature below freezing. The ingress of oxygen must have been through the auger as the lower central positions were the only ones affected although there was a subsequent rise all the way to the surface which would be following the pattern seen during the flow application. The final phase was due to the action of the wind. The sudden wind speed increase which occurred at 838 hours produced an immediate and large increase in oxygen at the 6 m centre and the 1 m side positions which returned to normal atmospheric conditions. Slower responses were seen in the upper central and lower side sections of the silo but these rates of increase in oxygen were increased markedly over their previous rates. This period of high wind showed the limitations of the sealing especially around the silo base and returned the silo's atmosphere to normal within 20 hours. The gradual increase in oxygen prior to the rise in wind speed was very similar throughout the silo and varied from 0.09 to 0.16% oxygen/h with the highest rate at the 6m depth in the centre.

The analysis to produce the maintenance rate for this silo showed that the high winds during the 3 m^3/h flow rate affected the overall result. The equation from regression analysis,

$Y = -0.00374X + 0.02204$ ($R^2 = 0.765$), produced a maintenance flow rate value of 5.89 m^3/h. However, as a flow rate lower than this value had reduced the silo oxygen level, then this must be incorrect. It was also biased by the large drop during the 4.5 m^3/h flow period where the potential for lowering of the oxygen level was large as it was recovering the loss from the 3 m^3/h flow rate. The high value of the latter was due to the increased winds which means that this oxygen level for the silo was not achieved in the desired conditions. Therefore the 4.5 m^3/h value is a better estimate of the maintenance rate.

Trial 7 - Linton (1150 tonnes of wheat)

The output from the burner and its constituents throughout the trial are shown in Figure 28 (Page 108). An oxygen level below 1% was maintained in the output throughout the trial (Table 9 (Page 72)). The first stage involved the purge which used a flow of 19.2 m^3/h. The position 1 m side dropped fastest (1.6% oxygen/h) and last whereas the other positions dropped at 0.7% oxygen/h. The 1% oxygen level was reached after 72 hours (Figure 29 (Page 109)) which meant 2.4 total changes of atmosphere within the silo. During this period the changes in environmental conditions did not have any significant effect on the rate of decrease of the oxygen levels in the silo (Figure 30 (Page 110)).

Further changes in flow rate were made to find a maintenance rate starting with 15.6 m^3/h. The results from these periods (Table 9 (Page 72)) did not show any pattern of increasing oxygen levels with decreasing flow rates. The lowest flow rate tested was 12 m^3/h. After 36 hours which represented 0.75 atmosphere replacements at this flow rate, there was no change in oxygen levels at any point indicating that the maintenance rate was lower than this value. However this value was used as the maintenance rate for this silo size.

The main factor responsible for preventing the collation of any further relevant data was the high level of wind experienced during the trial (Table 2 (Page 67), Figure 31 (Page 111)). As the mean wind speed rose towards 15 m/s the oxygen level at the side of the silo rose rapidly (Figure 32 (Page 112)). The oxygen at the 1 m depth increased to the normal atmospheric level with a trend for this increase in oxygen to be moderated with increasing depth. These were still higher than the values reached at the centre of the silo which were very similar throughout the trial (Figure 33 (Page 113)). Leakage of the generated atmosphere was taking place around the edge of the sheeted grain and through the walls and this depletion affected the atmosphere in the centre. There was no obvious loss from the auger or the aeration duct. Overall it appeared that a mean wind speed around 13 m/s was sufficient to cause this sudden loss in atmosphere. The wind record suffered a failure in the line from the anemometer head which lasted from 408 to 504 hours, missing the start of one period of high winds.

The changes in ambient temperature seemed to have little effect on the silo oxygen content (Figures 34 and 35 (Page 114 and 115)) even though there were some rapid drops in ambient temperature. The maximum difference between the grain and ambient temperature occurred during the final stage of the trial when the machine had stopped. This was not planned and was caused by the seizure of the pump which drove the glycol circuit following two consecutive short power failures after 564 hours. After this the silo was left to return to normal atmospheric conditions. This were not completed by the time the recording equipment was turned off and only the 1 m side position was close to this level (Figure 36 (Page 116)). There were only light winds during this period which did not affect the oxygen levels. However, the decrease in the ambient temperature below freezing may have enhanced the rate of increase in oxygen after this period (Figure 37 (Page 117)). Once this influence was lost with the rise in temperature there was little change in the oxygen levels for the last 50 hours. In all positions the overall gain was 0.1% oxygen/h.

Calculation of the maintenance flow required for any silo capacity

Figure 38 (Page 118) shows the three values produced by the latter three trials in this study. They produce a linear relationship after regression ($R^2 = 0.886$) which is given by the equation:

$$Y = 0.0091X + 2.4767$$

This would mean that 100 tonnes of grain requires a flow rate of 3.4 m^3/h. When these values are combined with those from past work with MA generators (Bell *et al.*, 1991, 1993a, b) it can be seen from Figure 39 (Page 118) that these are all above the present line. The new relationship formed when these are taken into consideration after regression ($R^2 = 0.858$) is:

$$Y = 0.0079X + 3.9387$$

In this form 100 tonnes of grain would require a flow rate of 4.7 m^3/h, and 1000 tonnes would need 11.6 m^3/h..

GERMINATION TESTS

a) Prior to the trial the germination potential of the malt was very high with the same average value from each sampling position of 98.1% with individual samples varying from 95 to 100% (Table 10 (Page 73)). 95% is the lowest level acceptable for a maltster so these values are all good. A greater proportion of the germination took place on the second day with an average from the three samples of 60.1%. There were very few grains left to germinate by the third day (6.5%) though there was a noticeably higher proportion in sample C.

The germination potential was not lost after three weeks under a low oxygen atmosphere with the two averages giving 97 and 99.5% which gives an almost identical mean result to that at the start (Table 11 (Page 74)). The range of individual values improved slightly with values from 96 to 100%. There was still the most germination on day 2 with the average increasing to 71.3% and there was an increase to the proportion germinating on the last day (18.6%), though none of these differences was significant.

b) Sample A from the first set of samples gave a marginal value prior to exposure to burner gas for this factor. A result between 50 and 60% is the critical low limit for this characteristic. The average for the three samples was 66.0% (Table 10 (Page 73)). Most of the germination occurred on the first two days with day 1 having a marginally higher proportion with 52.6% and there was only 7% on day 3.

Water sensitivity had improved by the end of the trial with germination increasing to an average of 80.1% (Table 11 (Page 74)). There was no significant change in the distribution of germination between the three days. This time day two had the higher value at 51.4% and the proportion on day three had dropped to 2.7%.

INSECTS AND MITES

Insect Juvenile Stages

Nitrogen

The first test with this gas assessed the difference in tolerance between the older and younger immature stages of four strains of *S. granarius* to this gas with most cultures being started at the same time except for two of the younger cultures, Lab II and Gain II (Table 12 (Page 75)). There was an apparent difference with Gain I showing greater tolerance. However, as seen with Pres there was a difference in the results from two different sized containers and therefore variability in the results may arise from the numbers of larvae that were in the grains. For Lab and Gain the difference between their two tests was in the age of the cultures. This result did show that the egg stage in the IIs was not more tolerant than the later larval stages. To achieve complete mortality the exposure would have to be extended to the time used for this species in the second test (Table 13 (Page 75)) although the insects were older in that test. A 55-day exposure length killed nearly all the juveniles and indicated no difference in strain tolerance. A 60-day exposure was required to kill all stages.

For *S. oryzae* there was little difference in mortality between the strains and no difference between the exposure times (Table 13 (Page 75)). The 306 strain of *R. dominica* was more tolerant than its counterparts, in spite of its cultures having a younger age range. Tolerance to MAs increases with age. *R. dominica* appeared more tolerant than *S. oryzae* for similar stage juveniles. Both these species would require much longer than 44 days exposure to give complete control.

For the remaining three species there was poor emergence in some of the control replicates particularly for *T. castaneum* Lab (Table 14 (Page 76)). These results affect the accuracy of the emergence results from the exposed juveniles. The reason for this problem was probably the low temperature. In all the strains, whatever the species, there were not as many pupae as required and therefore fourth instar larvae were used as well which are not as tolerant of MAs as pupae. There was little strain difference except CTC of *T. castaneum* which was more tolerant than the other two. *O. surinamensis* was the least tolerant of the three species and would only require a short extension to the five day exposure period for complete control (Table 14 (Page 76)), whereas *C. ferrugineus* obviously needs a period closer to the longest exposure of 8 days tried for *T. castaneum*. This latter species would need an increase in exposure period over this time for complete control.

Burner Gas

A comparison could not be made between strains in *S. granarius* as there was little emergence for the Gain strain in the control (Table 15 (Page 76)). Complete control was achieved by 65 days which was longer than required by nitrogen alone though the significance of this difference is doubtful. For *S. oryzae* and *R. dominica* the results were better than those achieved with nitrogen (Table 13 (Page 75)). For *S. oryzae* the cultures were the same age as for the nitrogen test but a far higher level of control had been achieved in a time period that was shorter than the ones used for nitrogen. In this test the strain RA76 appeared more tolerant of the atmosphere.

The *R. dominica* cultures were much older than those used for the nitrogen test. The emergence was much reduced from those in the nitrogen atmospheres and the reduction was achieved with shorter exposure times.

Carbon Dioxide

For *S. granarius* there was no difference between results with the two oxygen contents (Tables 16 and 17 (Page 77)) though the juveniles used in the test with 0.5% oxygen were older. 38 days were required for both for complete mortality. This is a much shorter time than required for both nitrogen and burner gas.

A similar pattern is seen in *S. oryzae* and *R. dominica*. The former species produced an interesting result as the 2% oxygen mixture gave 100% mortality after 32 days whereas there was survival with 0.5% oxygen. A period longer than 32 days is therefore necessary for the control of both species. Nevertheless carbon dioxide was more effective than the other MAs.

The other three species were only tested with 0.5% oxygen (Table 18 (Page 78)). There does seem to be strain difference for *C. ferrugineus* with Res proving more tolerant. For all species there is a need to extend their exposure times above those tried to achieve complete control as with the nitrogen tests. In a comparison with nitrogen it would appear that there is little difference in effectiveness for *C. ferrugineus* and *O. surinamensis* though carbon dioxide does seem to be better against *T. castaneum.*

Moth - *E. elutella* diapausing larvae

These experiments show the importance of temperature during exposures to MAs. For each of the atmospheres used there was higher mortality for a similar time period under 20°C than under 10°C (Tables 19, 20 and 21 (Pages 78 and 79)). This difference was not as marked for the burner gas mixture as for the other atmospheres.

Carbon dioxide is the most effective of the three atmospheres especially as the highest concentration used still had 4% oxygen present (Table 19 (Page 78)). The two nitrogen mixtures and the 30% carbon dioxide atmosphere did not achieve 100% kill with the exposure periods tested. The burner gas mixture was the least effective though the nitrogen atmosphere did contain 0.5% less oxygen. This reduction was small but appeared to have an appreciable effect as illustrated by the result.

Mites

A similar trend was seen in the tolerance of the species despite the MA mixture used. The most tolerant was always *T. longior* and this was true both for the egg stage (Table 22 (Page 80)) and for the adult stage. Its higher tolerance was more marked in the gas mixtures with 0.5% oxygen levels. It was most tolerant to the nitrogen atmosphere and least to the burner mixture, with carbon dioxide between these two atmospheres. This is a trend seen in the other species except *A. siro* which was most tolerant of 99.5% carbon dioxide. However, when the oxygen content is increased, this trend is altered as there is little difference between the two nitrogen based mixtures in the rapid loss of efficacy, while with carbon dioxide there was very little difference in exposure periods required to give 100% mortality with the increase in oxygen to 2%. The same was true on the change from 98 to 60% even though the oxygen content of the mixture increased to 8%. The only exception was *L. destructor* which had a marked improvement in its survival. An increase in oxygen content in the nitrogen-based atmospheres produced a marked increase in the length of time needed to give 100% mortality for all the species except the cheese mites but their control diminished in comparison to carbon dioxide.

DISCUSSION

The first four trials demonstrated the ease of use of the different generating systems and their relative efficiencies. The PSA machines used in trials 1 and 3 were certainly capable of generating the required atmosphere. However, the first machine did not have enough capacity and lost efficiency through the trial as the coke beds within the pressure vessels retained more oxygen and became less effective in the removal of this gas. The removal of some grain from the silo prior to the test did not help. This meant that the grain was more loosely packed on the side above the auger, near the point of gas entry to the silo, as evidenced by the billowing of the surface sheet directly above this point. As a result there was a difficulty in replacing the oxygen as the incoming gas was not being distributed evenly throughout the silo. Even at flow rates of about 8 m^3/h with 99.5% nitrogen, several days were needed to bring the oxygen levels down to 2%. The problem was also compounded by the lack of a projecting dosing pipe inside the aeration duct hatch plate to take the nitrogen further into the aeration system of the silo.

Trial 3 was similarly beset by technical problems which did not allow a full assessment of the PSA system's full capabilities. The purge was very successful but the only reliable reading from the maintenance flow rate assessment indicated a value below $20m^3/h$. However, the values from trials 5 and 7 indicate that the true level should have been very much below this flow rate. The ability to hold the generated atmosphere may have been affected by the removal of the excess grain in the roof space to allow entry. This would loosen the grain in the centre of the silo as it is removed from the bottom. The compacting of the grain which occurs when the silo is loaded does help to contain the atmosphere within the grain.

The cost of trial 3 was high but the machine used was of an older design intended to produce a much higher flow rate. Nowadays the machinery required is much more compact, supplied by smaller compressors and thereby reducing the power consumption. A decrease in power requirement would mean that the plant would be able to use the electrical power available on-site rather than using a generator. This would lower the cost of providing the nitrogen which for this trial was £2.33 per tonne of grain.

The problems for the application of carbon dioxide in trial 2 arose mainly from its sorption by the malt, and subsequently further problems caused by incomplete mixing and settling during maintenance flows. However, it did appear that there were cracks within the concrete structure of the silo particularly in the headspace which were allowing the gas to escape into adjacent silos. With these particular conditions it would appear that having purged the bin at 60-80 kg/h from the base for about 9-12 h, or 30-60 kg/h to the head space for 12-24 h, a maintenance flow of about 15 kg/h was able to maintain over 60% of the gas at all positions in the silo. For a recommended treatment period of seven days at 28-29°C, this would require a total of 3,060 kg of carbon dioxide which is double the recommended dosage for a grain bulk of this size in the USA. Dosing to the head space offered the advantage that gas levels increased more evenly throughout the silo to about 60-70%, while dosing to the base produced 90% levels throughout the silo but the grain surface and the head space never reached the required 40% level.

The supply of this gas required a complex installation of pipe work, a vaporiser and pressure regulators for safe delivery and was reliant on bulk deliveries of the liquid gas. The problem encountered with sorption is a well known reversible process, all gas being released on standing in air (Mitsuda *et al.*, 1973). It is a problem with all grains and the amount sorbed varies with temperature and moisture content, with a maximum of 0.42g/kg of gas sorbed in wheat in 24 h at 0°C and 18% m.c. (Cofie-Agblor *et al.*, 1993). Therefore when using this gas it is important to know the sorption capacity of the product at its stored temperature and moisture conditions. The malt tested here sorbed over seven times the amount of carbon dioxide at 28°C as quoted above for wheat at 0°C. The best strategy for the application of the gas for the silo tested was to purge the base for 15 h at 60 m3 followed by a similar flow to the headspace for 10 h. Maintaining a flow of 20 kg/h for the remaining 6 days gave a total requirement of 5100 kg and a cost of £14.60 per tonne of malt.

The Aerogenerator trial was the fourth and last exploratory assessment of MA application systems. However, the output was not sufficient to bring oxygen levels down quickly in the concrete silo, and some points never reached the target 1% level. The loss of output was due to leakage from the combustion chamber itself and a much lower burn could have achieved the same volume of cooled product gas by an adjustment to the output damping valve. These problems with the output did demonstrate the importance of monitoring actual flow rates of product gas. For this system further modifications of the heat exchange system were needed to improve the cooling of the output gas. Although this trial was not entirely successful it did show the potential of this gas generation system with its ease of use and readily available power supply. A previous comparison between costs of a propane burner and carbon dioxide for treating wheat in a welded metal silo had shown that the former system was £7.25/tonne cheaper though this did not take into account the original capital cost of the propane burner (Bell *et al.*, 1993a).

As well as the generation system employed to generate the MA it is important to consider the storage silo itself. The above trials can be separated into two sections by silo construction. Trials 2 and 4 which were carried out in concrete cells at Wallingford, had the advantage of the protection that the surrounding building provided from the vagaries of the weather. The dominance of the environmental conditions over MA operations and the subsequent losses of time under the generated atmosphere were well demonstrated in all the other trials. These losses in time may mean that an extension to the period under the MA will be required. However, laboratory work (Conyers and Bell, in press) has shown that two or three interruptions of 16 h when the oxygen level reached 5% during a continuous exposure in a 1% oxygen atmosphere had no effect on the mortality of adult *C. ferrugineus*, *O. surinamensis* or *S. granarius* (Figures 40 to 42 (Pages 119 to 121)). This means that an overnight loss in atmosphere could be rectified by an increase in flow without an increase in time spent under the atmosphere at the end of the exposure period.

The concrete silo used for both trials at Wallingford should not be faced with this problem of atmosphere loss from changes in the weather. However, the problem was within the structure itself as there were cracks in the concrete particularly near the top and this increased the gas losses for both trials. These trials may well have benefited from sheeting the surface of the malt. Even when storage conditions appear ideal for use with MAs, the need to purge the headspace when this sheet is absent can adversely affect the performance of an atmosphere.

The other five trials were carried out on silos that were not constructed for use with MAs and therefore they were very susceptible to the changes in the weather that can remove the generated atmosphere from a silo very rapidly. Trials 1 and 3 did not produce any results which could be used in the production of the maintenance flow relationship with silo size. Even the final three silos did not have fully comparable conditions. The Micheldever trial had very warm grain which is beneficial for MAs. It would have meant that a relatively short exposure period would have been required if insects had been present as low oxygen atmospheres cause mortality in shorter time periods as temperature increases. However, this is not a good condition for grain in long-term storage and it does create problems for MAs if there is insufficient sealing. An unsealed auger proved to be an ideal route for the ingress of the cooler, relatively more dense atmospheric air driven by a mean 15°C difference between the ambient and the bulk temperature. This influx of air caused an increase in the oxygen levels in the centre of the silo, particularly at night when the temperature difference was 2 or 3°C greater than the day.

For all the purges there was definite pattern to the change in gas concentrations with the oxygen decreasing asymptotically as found by McGaughey and Akins (1989). The drop in oxygen at the steepest section of the curve was much faster for the smaller silo at 2.9 to 4.4% oxygen/h as against the very similar rate of 0.5 to 1.5% oxygen/h for the other two silos. The similarity of the last two was particularly unusual in that there was an appreciable difference in their purge rates. At Micheldever the purge flow rate was only 15.6 m^3/h whereas at Linton it was 19.2 m^3/h. Although no end point for the purge could be calculated for Micheldever it does appear from the results that it would have been close to the 72 hours achieved at Linton. The unusual result was at Hursley where the purge rate was at least three times the flow of the maintenance rate but it still took 76 hours to reach the 1% oxygen level. It has been shown before that it is important to have as high a purge rate as possible for an efficient purge (McGaughey and Akins, 1989, Bell *et al.*, 1993a). The present results do not illustrate this point as, in contrast to Hursley, the Linton result was achieved with a purge rate only 1.6 times its maintenance rate and needed only 2.4 changes of atmosphere. However, it is apparent that the sealing on the Hursley trial could have been improved. As far more time is spent under the maintenance rate and it is important to achieve the 1% oxygen level as quickly as possible to avoid using excessive amounts of fuel during the purge.

The maintenance rates are lower and are affected by any changes in the prevailing weather conditions. Wind speed is the most important factor and determined the flow rate required for maintaining any given oxygen level. However, the size of the grain bulk is also important. At Micheldever with the wind speeds of 8 m/s, flow rates of 11 m^3/h or lower were unable to maintain 1% oxygen. However, in the smaller silo at Hursley a similar wind speed did not affect the oxygen level during a flow rate of 9 m^3/h. This difference is to be expected as with a theoretical doubling of bin size there should be an increase of 1.59 in the flow rate to maintain a similar oxygen level due to the relationship between leakage rates and surface to volume ratios (Bell *et al.*, 1993a). In this example there was not enough increase in the flow for the silo at Micheldever to cope with the extra surface area. There is a trebling in size between Hursley and Micheldever and there is also the expected doubling in the maintenance flow rate as should be required, similar to the findings of Bell *et al.*, (1993a).

The loss in atmosphere due to wind occurred from the side and particularly near the top of the silo. The oxygen level of the whole silo was affected with a time lag before the centre showed an increase to a lower level than at the side. This is different to the pattern of atmosphere loss due to cold. The main factor in this case is the relative difference between the bulk and the ambient temperature. This was at its greatest during the Hursley trial where the difference reached 24.2°C. The freezing temperatures also caused problems for the burner with the losses of flow due to the formation of ice in the outlet. Once the flow was stopped, the modified atmosphere was lost at the bottom in the centre which means that ingress of air was principally through the auger.

The auger inlet to the silo is therefore an area of weakness as far as leakage or ingression of air is concerned. This was demonstrated by the periods of leak back at the end of the trials. At Micheldever the loss of atmosphere was fastest at the 6 m centre position and the same was also true at Hursley where high winds increased the rate of loss in the same area. However, when all three silos are compared they had very low loss rates in calm conditions of approximately 0.1 to 0.2% oxygen/h so they were all sealed to a similar level. These loss rates are lower than those shown for well-sealed silos in Australia (Banks *et al.*, 1991). However, irrespective of the seal it was the changes in environmental conditions that proved to be the main agents for atmosphere loss.

The maintenance rates required as silo size increases represent 0.75, 0.60 and 0.51 changes in atmosphere per day. Therefore, there is support for the theory that maintenance flow rates reduce in proportion to silo size. The values obtained from these trials compared very well with earlier work (Bell *et al.*, 1991, 1993a, b). The value for a 250 tonne silo (Bell *et al.*, 1993b) was extrapolated from the data generated by that trial and was in fact carried out on a similar silo at the same site as the present trial for the 292 tonne silo. Therefore the extrapolated flow rate may have been set too high. However, differences in the effectiveness of sealing of joints in different silos may alter the maintenance rate attainable even in two silos of similar size. The value quoted for the Linton trial was the lowest observed maintenance flow and the true result may be lower than this under more favourable conditions.

The decreasing number of atmosphere changes per day with increasing silo size is reflected in the cost per tonne to carry out the treatment. Taking a treatment purge at maximum flow rate and a four-week run under the estimated maintenance flow for each silo, the expected cost per tonne in terms of propane consumed would be 36p, 24p and 17p for the three sites in ascending order of tonnage, plus a small, constant electricity cost for running the burner. However, these figures may be an over-estimate of the minimum costs per tonne. The flow rates for the Hursley and the Linton trial were both high estimates of the maintenance rates. This is shown by the results of the regression analysis for maintenance rate against silo tonnage. When other values from past trials were included in the analysis, the flow required to hold a 100 tonne silo was 4.7 m^3/h a value higher than that attributed to the 292 tonne silo at Hursley. This result was known to be a high estimate for the maintenance rate of this silo. Therefore it would appear that the relationship obtained from the three most recent trials would provide a more accurate representation. Here it is likely that a flow lower than 3.4 m^3/h could be adequate to maintain an atmosphere in a 100 tonne silo.

These relationships are estimates for calm weather conditions and do not take into account any changes in conditions which might cause a rise in the oxygen level. However, as has already been stated, some interruptions can be withstood without an increase in the exposure times beyond the time normally required to produce 100% mortality in certain grain pest species, although further work will be necessary to define the critical length of such interruptions.

It was important to determine whether the burner gas atmosphere had any effect on the malting barley which is certainly the most valuable UK cereal crop. The brewing industry is trying to avoid the occurrence of any insecticide residues in its products and MAs would provide an alternative treatment strategy provided that they had no adverse effect on any important properties of the grain. There was no change in the grains' germination potential for either the level or the timing. Barley must be able to germinate in a high moisture environment which is used to initiate germination at the start of the malting process. The inability to achieve germination is known as water sensitivity and may occur if water uptake is excessive (Palmer, 1980). The grain can improve and will recover its ability to germinate in these conditions. However, as there was no decrease in the response to these conditions in the current tests, the applied MA was seen to be safe to use with malting barley. In general, storage under atmospheres with reduced concentrations of oxygen slightly improved the retention of quality of grains and therefore the use of these MAs as an alternative insect control technique is quite acceptable (Gras and Bason, 1990).

For MA treatments to become established, it is essential that complete control of any insects present, whatever their stage, can be obtained. In general results have indicated that carbon dioxide atmospheres are usually more toxic than oxygen-deficient ones (Navarro and Donahaye, 1990). This is because carbon dioxide does not rely solely on anoxia to be lethal but also on acidification of the body fluids, due to the formation of carbonic acid, and inhibition of glycolysis (Adler, 1994). This trend was apparent in the tests on juvenile insect stages, warehouse moth and mites. *S. granarius*, one of the most tolerant insects, required 60 days with nitrogen but only 38 days with carbon dioxide to kill the most tolerant pupal stage. These results indicate longer exposure times than those of Reichmuth (1990) who achieved 100% mortality with 99% nitrogen in 45 days and the same result with 90% carbon dioxide and 2% oxygen in 27 days carried out in the same conditions. This difference may be explained by the use of different strains and differences in tolerance to MAs as shown by Adler (1991) working with the same species.

The most tolerant stage of development for MAs occurs where the consumption of oxygen is lowest, often the egg and pupal stages (Reichmuth, 1987). Pupae tend to be the most tolerant stage in the species that develop within the grains. This habitat is itself a low oxygen environment and therefore adapting to such an environment would necessitate the evolution of a low oxygen requirement. Diapause is a state which has an even lower oxygen requirement. This can be seen from the present results of the exposure of *E. elutella* to MAs while in this state. 90% carbon dioxide produced 100% mortality in 14 days at 20°C whereas 99.5% nitrogen only achieved 74.1% in twice the time. The importance of these observations is that this insect must be controlled before it is allowed to develop to its most tolerant phase. Eggs of this species require only 4 days exposure to either 99.5% nitrogen or 97.5% carbon dioxide for 95% mortality (Reichmuth, 1987).

This pattern is completely reversed with the mites as larvae, nymphs and adults are controlled very quickly. There are few published results for the use of MAs against mites and these mainly consider the mobile stages. Navarro *et al.* (1985) showed that the adults of *A. siro* were killed after 3 days in 98% nitrogen and 2% oxygen and that an atmosphere of only 30% carbon dioxide was capable of controlling them in 4 days, with both these tests run at 15°C and 75% r.h. The present test results show that it is the egg stage that needs to be considered for this pest, which is becoming increasingly recognised as a cause of damage within the grain storage system. On occasions where mite species are involved, and the developmental position of the pest population is not known, it is important to continue the exposure to the MA until all eggs would be killed.

The choice of temperature for the test conditions was important as this should represent the conditions that would be found in the storage situation. 15°C is the maximum temperature for the safe storage of grain. It is the minimum developmental temperature for. *S. granarius* (Howe, 1965) and *S. oryzae* (Birch, 1953) and none of the other beetle pests of grain can develop at such a temperature. However, it is not a problem for the mites which can develop below 10°C, and the warehouse moth uses the diapause mechanism to survive periods at much lower temperatures. At low temperature some of the stages of the insects most tolerant to the MAs are affected by the cold alone. This is especially true of *T. castaneum* whose pupae, even at 20°C, though able to achieve adulthood were unable to shed their skins (Howe, 1956). In the present tests the emergence of the individuals of this species held at 15°C though not exposed to the atmospheres, was reduced, particularly in the Lab strain. It is, of course beneficial for control if the most tolerant stage is not present at low temperatures because exposure times can be reduced.

The results from the insect and mite tests suggest that carbon dioxide is the most effective MA. However, when the method of gas generation and ease of field use are considered it is the burner gas system that leads the way, especially economically. It has been demonstrated that with a small amount of additional sealing and the sheeting of the grain surface that a 1% oxygen atmosphere can be held within the common bolted metal silos used for grain storage within the U.K. for a sufficient length of time to control the most tolerant grain pest. The constant flow system compensates for any gas loss so that perfect sealing is not a prerequisite. It is hoped to try this method of MA generation on floor store bulks. This prospect has already been assessed for a small 100-tonne bulk using the CSL version of the propane burner. Oxygen levels of 1% or below were achieved within 48 hours with a flow rate of 14 m^3/h. The atmosphere was then maintained at an adequate level with a flow of $9m^3/h$. This was similar to the flow rate required to hold a 180 tonne bolted metal silo at 1% oxygen. Extra plastic lining for the floor stores walls may be required for better sealing of larger bulks which cannot be completely covered in sheeting. Even so the use of the propane burner for the treatment of the smaller floor store would thus appear to be a viable proposition.

CONCLUSIONS

1. The purging of the silo should take place at the highest flow attainable to lessen the time to reach 1% oxygen.

2. The best fit relationship between maintenance flow rate and bolted metal silo size was $Y = 0.0091X + 2.4767$. This meant 100 tonnes of grain requires a maintenance flow rate of 3.4 m^3/h, and 1000 tonnes requires 11.6 m^3/h.

3. The required changes in atmosphere of the silo per day (atms/d) decreased as the silo size increased: 292 tonnes - 0.75 atms/d, 810 tonnes - 0.60 atms/d,1150 tonnes - 0.51 atms/d.

4. A comparison of the running costs for the three methods of MA generation showed that burner gas is the most economical method of production:
carbon dioxide £14.60/tonne, nitrogen (PSA) £2.33/tonne,
burner gas £0.34/tonne.

5. For carbon dioxide on 260 tonnes of malt, 3 tonnes of gas are required for an exposure of 7 days for control of principal storage pests at 28-29°C.

6. The sealing of all possible areas of gas loss is essential and a gas-proof plastic sheet covering the surface of the grain helps with gas retention during maintenance.

7. The use of an exposure period which produced over 99% mortality for the adults of three important beetle grain pests, *C. ferrugineus, S. granarius* and *O. surinamensis,* meant that there was no increase in mortality after three 16 hour interruptions of 5% oxygen during such an exposure at 20°C and 70% r.h.

8. Malting barley:
a) Germination potential
prior to introduction of burner gas: 98.1%,

after three weeks in the MA: 98.3%.

b) Water sensitivity:
prior to introduction of burner gas: 66%,
after three weeks in the MA: 80.1%.
Both sets of results show that the MA did not affect grain quality.

9. Juvenile stages of beetles showed a marked difference between internal and external grain feeders in tolerance of MAs, the latter being much more susceptible. Carbon dioxide performed much better than burner gas or nitrogen against internal feeders, *S. granarius*, immatures requiring 38 days for control at 15°C in 99.5% carbon dioxide compared with 60 or more days in the other atmospheres.

10. Diapausing larvae of *E. elutella* were not adequately controlled by 4-week exposures to burner gas or nitrogen atmospheres at 20°C but succumbed to carbon dioxide after 2 weeks at 20°C or 4 weeks at 10°C.

11. The egg stage was the most tolerant to MAs in all five species of mite tested. Reducing oxygen to 0.5% in carbon dioxide or nitrogen atmospheres resulted in control of the most tolerant species *T. longior* in 16-22 days at 15°C. With 2% oxygen, carbon dioxide was the most effective atmosphere, achieving control within 26 days.

12. MAs, particularly burner gas, offer a viable means of controlling a grain infestation and also offer the prospect of total storage life protection by continuous on-site production of gas.

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TABLES

Table 1: The dimensions of the silos and their grain contents as used in the seven trials

Location	Stonham Aspall	Wallingford	Shrewton	Wallingford
Date	7/4-23/4/93	16/8-21/8/93	6/12-20/12/93	15/2-19/2/94
Modified Atmosphere	Nitrogen	Carbon dioxide	Nitrogen	Burner Gas
Silo Height (m)	4.5	26	11.4	26
Silo Diameter (m)	6	4.5	11	4.5
Grain	Wheat	Malt	Feed Barley	Malt
Grain Weight (tonnes)	120	260	900	260
Grain Volume (m^3)	99.0	373.8	1103.5	1476.9
Interstitial Volume (m^3)	38.1	143.9	444.3	568.6

Table 1 (Contd.): The dimensions of the silos and their grain contents as used in the seven trials

Location	Micheldever	Hursley	Linton
Date	14/10-28/11/94	15/12-20/1/94	7/2-8/3/95
Modified Atmosphere	Burner Gas	Burner Gas	Burner Gas
Silo Height (m)	10.3	10.9	14.3
Silo Diameter (m)	11	6.6	11
Grain	Malting Barley	Wheat	Wheat
Grain Weight (tonnes)	810	292	1150
Grain Volume (m^3)	1144.4	373.8	1476.9
Interstitial Volume (m^3)	440.6	143.9	568.6

Table 2: Grain temperature and environmental conditions during each of the trials

	Location			
Conditions	Stonham Aspall	Wallingford	Shrewton	Wallingford
Grain Temperature (^{o}C)	8.6 (8.1 - 8.8)	29.6 (29.3 - 29.8)	18.8 (18.1 - 19.2)	25.7 (24.3 - 26.8)
Ambient Temperature (^{o}C)	10.0 (3.0-21.6)	*	*	*
Wind Speed (m/s)	11.1 (2.6 - 22.3)	*	11.1 (1.3 - 25)	*

*: Not recorded

Grain temperature and environmental conditions during each of the trials

		Location	
Conditions	Micheldever	Hursley	Linton
Grain Temperature (^{o}C)	22.7 (22.4 - 22.8)	16.9 (16.2 - 17.7)	9.1 (6.9 - 9.8)
Ambient Temperature (^{o}C)	10.6 (4.3 - 15.5)	4.9 (-6.9 - 12.7)	6.5 (-2.8 -13.2)
Wind Speed (m/s)	3.0 (0.0 - 8.7)	4.0 (0.0 - 17.6)	8.0 (0.0 - 20.0)

Table 3. Stonham Aspall - the results from the application of nitrogen to a silo with 120 tonnes of malting barley

Flow Rate (m^3/h)	% Oxygen in Output	Duration (Hours)	Total nitrogen used (m^3)	Change in silo gas content (atms/day)
0.6	0.8	195	117	0.4
0.8	1.2	20	959	0.5
0.7	0.7	124	87	0.4
0.5	0.4	44	22	0.3

Table 4. Wallingford - the results from the application of carbon dioxide to the base of a silo with 260 tonnes of malt

Flow Rate and application position of carbon dioxide during trial (kg/h)		Duration (Hours)	Total carbon dioxide used (kg)
Base	60	2.3	138
	80	1.4	112
	20	17.6	352
	80	5.1	22
Top	80	0.5	40
	8	17.5	140

Table 5. Wallingford - the results from the application of carbon dioxide to the top of a silo with 260 tonnes of malt

Flow Rate (kg/h)	Duration (Hours)	Total carbon dioxide used (kg)
30	23.3	700
60	5.0	300
10	20.5	205
20	24.8	490

Table 6: Shrewton - the results from the application of nitrogen to a silo with 900 tonnes of feed barley

Flow Rate (m^3/h)	% Oxygen in Output	Duration (Hours)	Total nitrogen used (m^3)	Change in silo gas content (atms/day)
35.0	0.3	50	1750	1.9
23.5	0.3	51	1199	1.3
29.0	0.5	21*	609*	1.6*

* loss of power in mobile laboratory and failure of PSA unit so exact duration at this flow rate unknown

Table 7: Micheldever - The results from the application of burner gas to a silo with 810 tonnes of malting barley

Atmosphere Flow Rate (m^3/h)	Propane Flow Rate (l/min)	Duration of Input (Days)	Total Volume (m^3)	Change in Silo Atmosphere per Day	Mean % Oxygen		
					Input	Mean of Whole Silo	End of each Flow Rate a
15.6	12.5	9	3370	0.85	1.2	b	4.4
14.4	12.0	4	1382	0.78	0.9	1.1	1.9
16.2	14.5	3	1166	0.88	0.6	0.8	0.9
13.2	12.5	3	950	0.72	0.4	c	c
13.8	12.5	2	662	0.75	0.4	1.1	1.1
10.8	10.4	5	1296	0.59	0.5	0.7	1.2
9.0	8.8	2	432	0.49	0.3	1.1	1.4
12.0	12.8	5	1440	0.65	0.2	1.2	1.1
12.6	13.0	4	1210	0.69	0.3	0.5	0.6

a: The mean % oxygen level within the silo when a particular flow rate was altered

b: This flow was not run for sufficient time after the auger was sealed to produce an accurate result

c: The burner stopped running during this flow and there was insufficient time to achieve an accurate result

Table 8: Hursley - The results from the application of burner gas to a silo with 292 tonnes of wheat

Atmosphere Flow Rate (m^3/h)	Propane Flow Rate (l/min)	Duration of Input (Days)	Total Volume (m^3)	Change in Silo Atmosphere per Day	Mean % Oxygen		
					Input	Mean of Whole Silo	End of each Flow Rate *
15.0	12.0	4	1440	2.5	0.5	3.0	0.3
9.0	9.0	11	2376	1.5	1.0	2.3	0.6
6.0	7.5	7	1008	1	0.7	1.2	0.8
3.0	5.8	5	360	0.5	0.3	1.7	3.9
4.5	6.9	6	648	0.75	0.3	2.1	0.8

*: The mean % oxygen level within the silo when a particular flow rate was altered

Table 9: Linton - The results from the application of burner gas to a silo with 1150 tonnes of wheat

Atmosphere Flow Rate (m^3/h)	Propane Flow Rate (l/min)	Duration of Input (Days)	Total Volume (m^3)	Change in Silo Atmosphere per Day	Mean % Oxygen		
					Input	Mean of Whole Silo	End of each Flow Rate *
19.2	17.0	3	1382	0.81	0.2	9.7	0.3
15.6	16.0	9	3370	0.66	0.8	10.0	6.9
14.4	15.0	5	1728	0.61	0.6	6.9	1.3
12.0	12.5	3	864	0.51	0.2	6.3	14.1
13.2	12.8	2	634	0.56	0.7	13.0	9.1

*: The mean % oxygen level within the silo when a particular flow rate was altered

Table 10: The numbers and mean percentages of malting barley grains germinating before the introduction of burner gas

Test		Germination potential				Water sensitivity			
Water added (ml)		4				8			
Sample (Depth)	Replicate	Day 1	2	3	Total	1	2	3	Total
A (Centre 3 m)	1	27	69	1	97	35	17	3	55
	2	33	64	2	99	25	26	5	56
	3	29	68	2	99	40	27	1	68
	4	38	54	5	97	24	24	2	50
	Mean (%)	32.5	65	2.5	98.0	53.8	41.3	4.9	57.3
B (Side 2 m)	1	38	61	1	100	35	23	4	62
	2	38	56	1	95	34	28	6	68
	3	34	63	2	99	26	28	5	59
	4	35	58	5	98	35	25	0	60
	Mean (%)	37.1	60.7	2.2	98.0	52.2	41.8	6.0	62.3
C (Centre Surface)	1	35	47	16	98	48	31	5	84
	2	24	60	12	96	44	25	3	72
	3	33	52	14	99	32	24	2	78
	4	28	55	17	100	38	39	2	79
	Mean (%)	30.5	54.5	15.0	98.3	51.8	38.0	10.2	78.3
	Mean A,B&C(%)	33.4	60.1	6.5	98.1	52.6	40.4	7.0	66.0

Table 11: The numbers and mean percentages of malting barley grains germinating after the introduction of burner gas for three weeks

Test		Germination potential				Water sensitivity			
Water added (ml)		4				8			
Sample (Depth)	Replicate	Day 1	2	3	Total	1	2	3	Total
A	1	10	68	21	99	29	48	7	84
(Centre 3 m)	2	8	81	11	100	38	44	0	82
	3	11	72	16	99	41	41	0	82
	4	12	75	13	100	38	44	1	83
	Mean (%)	10.3	74.4	15.3	99.5	44.5.	53.1	2.4	82.8
B	1	11	69	18	98	33	42	1	76
(Side 2 m)	2	14	75	9	98	41	33	2	76
	3	12	68	16	96	36	41	2	79
	4	2	53	41	96	36	38	4	78
	Mean (%)	10.0	68.1	21.9	97.0	47.2	49.8	3.0	77.3
	Mean A&B(%)	10.1	71.3	18.6	98.3	45.9	51.4	2.7	80.1

Table 12: The mean percentage emergence of adults after juvenile exposure to 0.5% oxygen and 99.5% nitrogen at 15°C

Species	*S. granarius*							
Strain	Lab I	Lab II	Gain I	Gain II	Fors	Fors	Pres	Pres
Age (Weeks)	0-4	0-2	2-4	0-2	0-4	0-4	0-4	0-4
Weight of Grain (g)	20	20	10	10	20	9	20	9
Exposure Time (Days)								
Control (Numbers)	64	63	71	104	240	95	210	74
28	1.6	0	49.3	0	4.6	11.6	26.2	25.7
32	26.6	0	53.5	0	27.5	30.5	26.7	9.5

Table 13: The mean percentage emergence of adults after juvenile exposure to 0.5% oxygen and 99.5% nitrogen at 15°C

Species	*S. granarius*			*S. oryzae*			*R. dominica*		
Strain	Lab	Gain	9104	Lab	RA76	PH3	Lab	915R	306
Age (Weeks)	0-5	0-5	0-3	0-3	0-3	0-3	0-3	0-3	0-2
Weight of Grain (g)	11	11	11	11	11	11	11	11	11
Exposure Time (Days)									
Control (Numbers)	39	59	89	238	173	158	107	98	118
38	-	-	-	38.7	32.4	46.8	62.6	34.7	70.3
44	-	-	-	33.6	34.1	46.8	42.1	49.0	66.1
55	0	1.7	1.1	-	-	-	-	-	-
60	0	0	0	-	-	-	-	-	-

Table 14: The mean percentage emergence of adults after juvenile exposure to 0.5% oxygen and 99.5% nitrogen at 15°C

Species	*C. ferrugineus*		*O. surinamensis*			*T. castaneum*		
Strain	Lab	Res	Lab	MR	Palm	Lab	CTC	BT1
Age (Weeks)	2-4	2-4	3-5	3-5	3-5	5-6	5-6	5-6
Exposure Time (Days)								
Control (Numbers)	33	50	32	36	32	18	42	32
3	91.9	52.0	21.9	5.6	9.4	-	-	-
5	51.5	42.0	3.1	2.8	0	-	-	-
6	-	-	-	-	-	55.6	59.5	28.1
8	-	-	-	-	-	16.7	47.6	28.1

Table 15: The mean percentage emergence of adults after juvenile exposure to 0.5% oxygen, 10% carbon dioxide and 89.5% nitrogen at 15°C

Species	*S. granarius*		*S. oryzae*		*R. dominica*	
Strain	Lab	Gain	Lab	R A76	915(R)	306
Age (Weeks)	3-4	3-4	0-3	0-3	7-9	0-9
Weight of Grain (g)	9	9	9	11	4	4
Exposure Time (Days)						
Control (Numbers)	43	8	218	226	55	111
28	-	-	3.2	18.6	5.5	2.7
32	-	-	3.7	8.0	1.8	1.8
36	-	-	2.8	10.6	3.6	0.9
55	2.3	0	-	-	-	-
60	2.3	0	-	-	-	-
65	0	0	-	-	-	-

Table 16: The mean percentage emergence of adults after juvenile exposure to 2.0% oxygen and 98% carbon dioxide at 15°C

Species	*S. granarius*		*S. oryzae*		*R. dominica*	
Strain	Lab	Gain	Lab	R A76	915(R)	306
Age (Weeks)	3-4	3-4	0-3	0-3	7-9	0-9
Weight of Grain (g)	11	11	11	11	6	6
Exposure Time (Days)						
Control (Numbers)	93	53	267	223	56	70
24	3.2	0	-	-	-	-
28	-	-	0.4	0.4	0	0
32	-	-	0	0	0	1.4
34	1.1	0	-	-	-	-
38	0	0	-	-	-	-

Table 17: The mean percentage emergence of adults emerging after juvenile exposure to 0.5% oxygen and 99.5% carbon dioxide at 15°C

Species	*S. granarius*			*S. oryzae*			*R. dominica*		
Strain	Lab	Gain	9104	Lab	RA76	PH3	Lab	915R	306
Age (Weeks)	0-5	0-5	0-3	0-3	0-3	0-3	0-3	0-3	0-2
Weight of Grain (g)	11	11	11	11	11	11	11	11	11
Exposure Time (Days)									
Control (Numbers)	39	59	89	238	173	158	107	98	118
28	-	-	-	1.3	0.6	4.4	0	1.0	0
32	-	-	-	1.7	0.6	2.5	0	1.0	0
34	0	0	1.1	-	-	-	-	-	-
38	0	0	0	-	-	-	-	-	-

Table 18: The mean percentage emergence of adults after juvenile exposure to 0.5% oxygen and 99.5% carbon dioxide at 15°C

Species	*C. ferrugineus*		*O. surinamensis*			*T. castaneum*		
Strain	Lab	Res	Lab	MR	Palm	Lab	CTC	BT1
Age (Weeks)	2-4	2-4	3-5	3-5	3-5	5-6	5-6	5-6
Exposure Time (Days)								
Control (Numbers)	33	50	32	36	32	18	42	32
3	2	84.0	25.0	11.1	31.2	-	-	-
5	33.3	82.0	3.1	0	0	-	-	-
6	-	-	-	-	-	22.2	9.5	28.1
8	-	-	-	-	-	0	2.4	3.1

Table 19: The results after the exposure of diapausing *Ephestia elutella* larvae to carbon dioxide

Temperature (°C)	Carbon dioxide (%)	Last Survival (Days)	Complete mortality (Days)
10	30	40	>40 (4% survived)
	45	28	40
	60	28	40
	90	20	28
20	30	40	>40 (2% survived)
	45	20	28
	60	20	28
	90	10	14

Table 20: The results after the exposure of diapausing *Ephestia elutella* larvae to burner gas mixture (1% oxygen, 12% carbon dioxide and 87% nitrogen)

Temperature (°C)	Exposure (Days)	Mortality (%)
10	10	5.3
	14	7.6
	21	22.9
	28	46.5
20	5	3.4
	7	3.6
	14	7.9
	21	36.4
	28	54.0

Table 21: The results after the exposure of diapausing *Ephestia elutella* larvae to nitrogen (0.5% oxygen and 99.5% nitrogen)

Temperature (°C)	Exposure (Days)	Mortality (%)
10	10	2.0
	14	5.3
	21	5.7
	28	0.0
	38	0.0
20	5	0.0
	7	0.0
	14	15.7
	21	72.4
	28	74.1

Table 22: Number of days required to produce 100% mortality for all stages in 5 species of mite when exposed to different modified atmospheres

	Modified Atmospheres *		
Species	0.5% O_2, 10% CO_2, 89.5% N_2	2% O_2, 10% CO_2, 88% N_2	
A. chaetoxysilos	4	4	
A. farris	4	4	
A. siro	6	36	
L. destructor	2	12	
T. longior	16	36	
	0.5% O_2, 99.5% N_2	2% O_2, 98% N_2	
A. chaetoxysilos	2	2	
A. farris	2	2	
A. siro	8	32	
L. destructor	8	12	
T. longior	22	34	
	0.5% O_2, 99.5% CO_2	2% O_2, 98% CO_2	60% CO_2
A. chaetoxysilos	4	6	2
A. farris	6	4	2
A. siro	12	14	20
L. destructor	2	4	24
T. longior	20	26	30

*: O_2 = Oxygen, CO_2 = Carbon dioxide, N_2 = Nitrogen

FIGURES

Figure 1: Stonham Aspall - Effect of wind speed on the oxygen levels in the silo

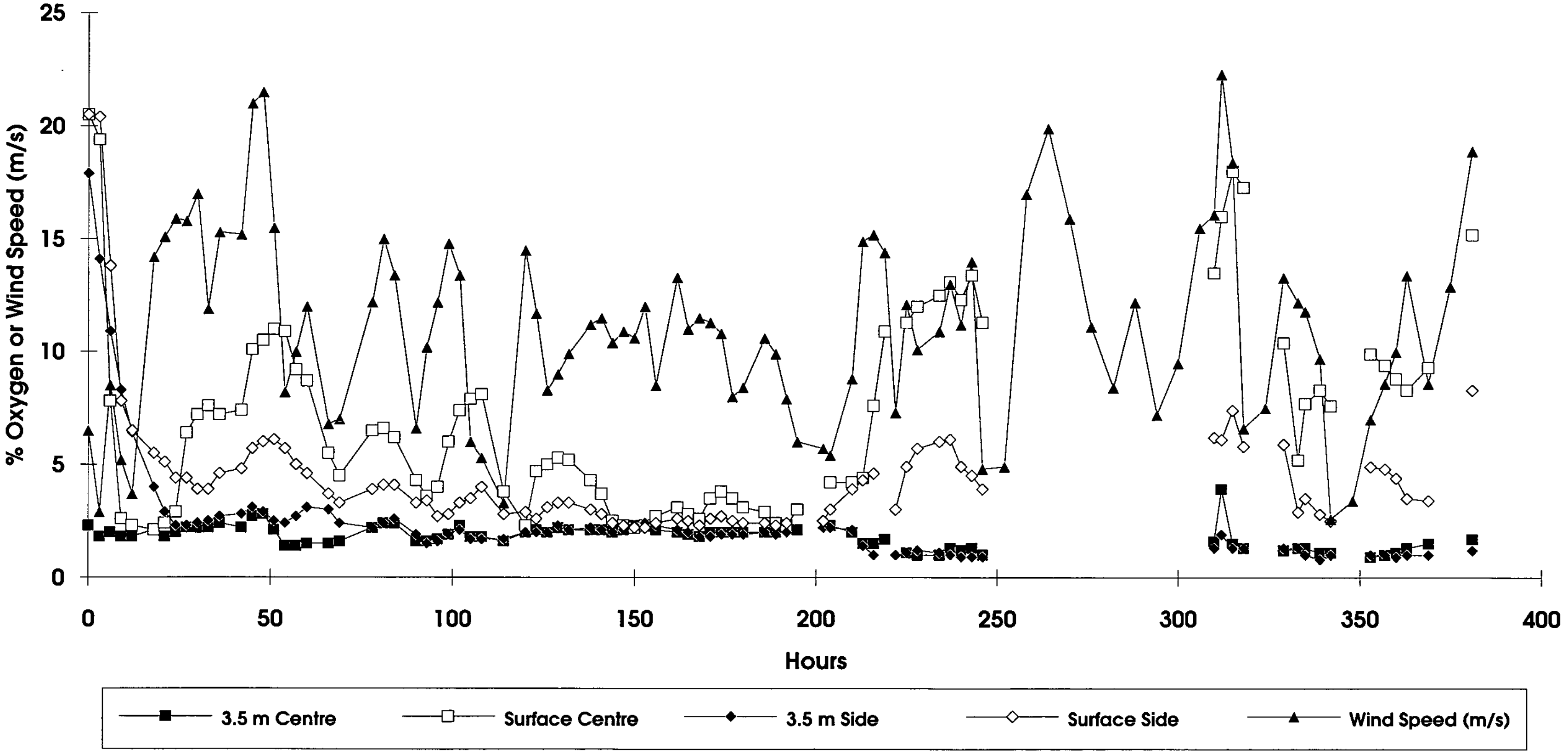


Figure 2: Wallingford - The change in carbon dioxide levels over time with different flow rates applied to the base of the silo

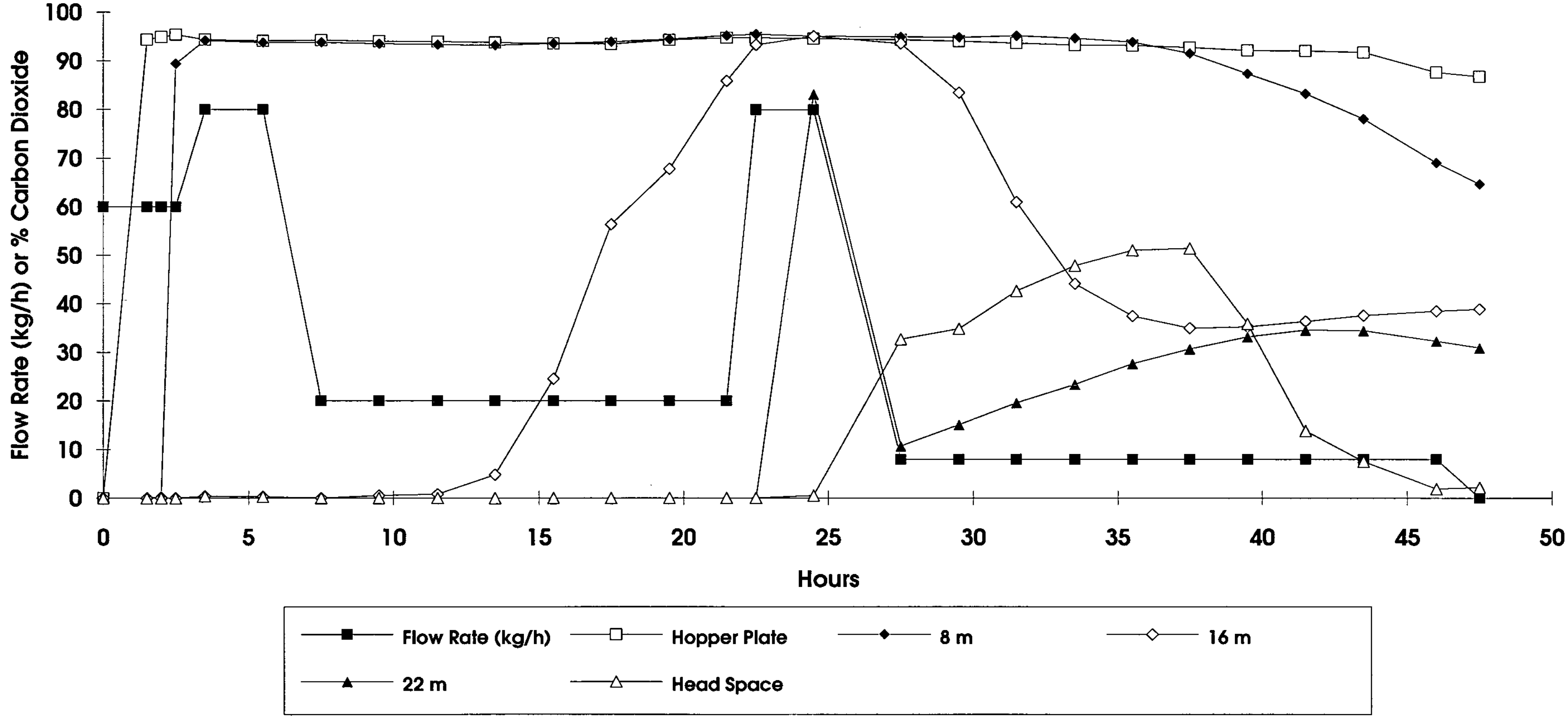


Figure 3: Wallingford - The change in carbon dioxide levels over time with different flow rates applied from the top of the silo

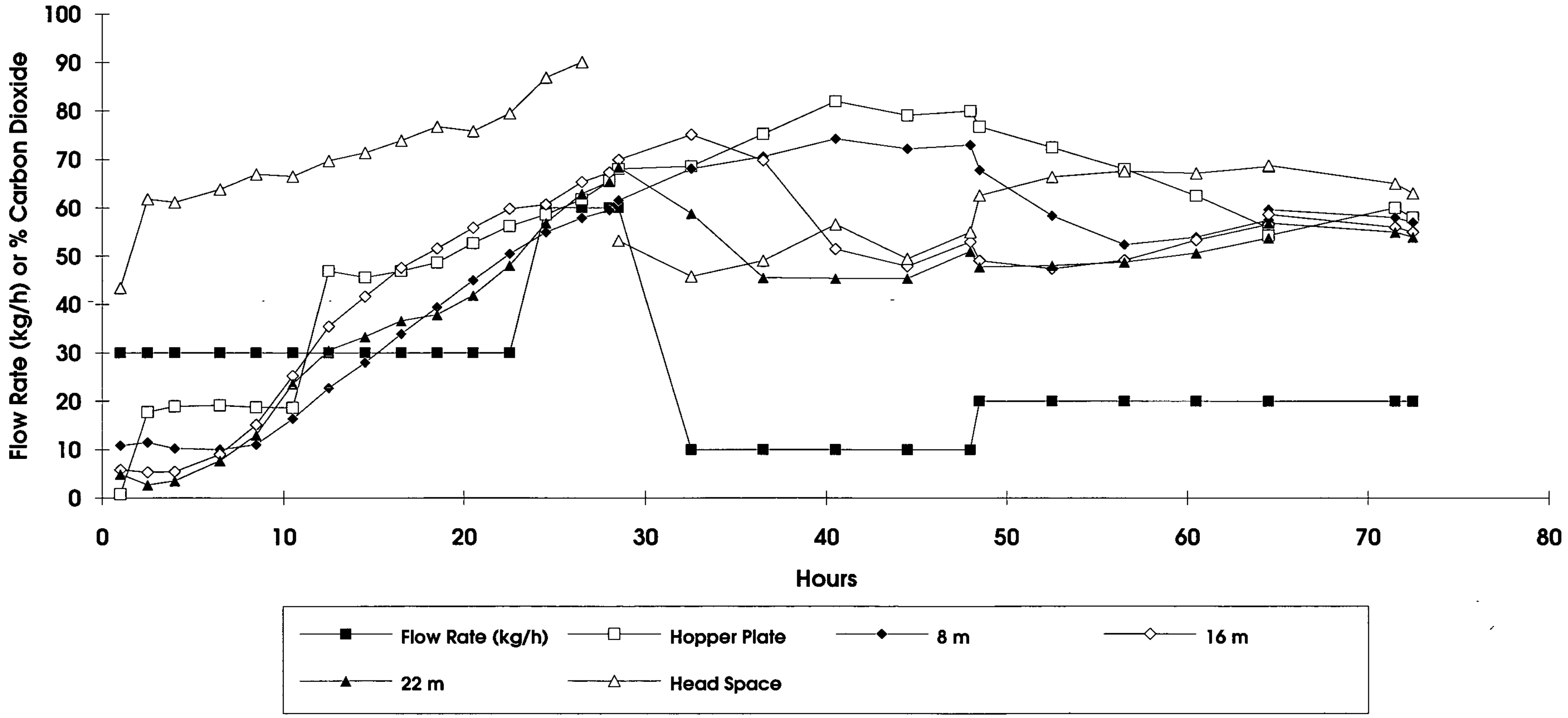


Figure 4: Shrewton - The effect of flow rate on the oxygen level in the aeration duct

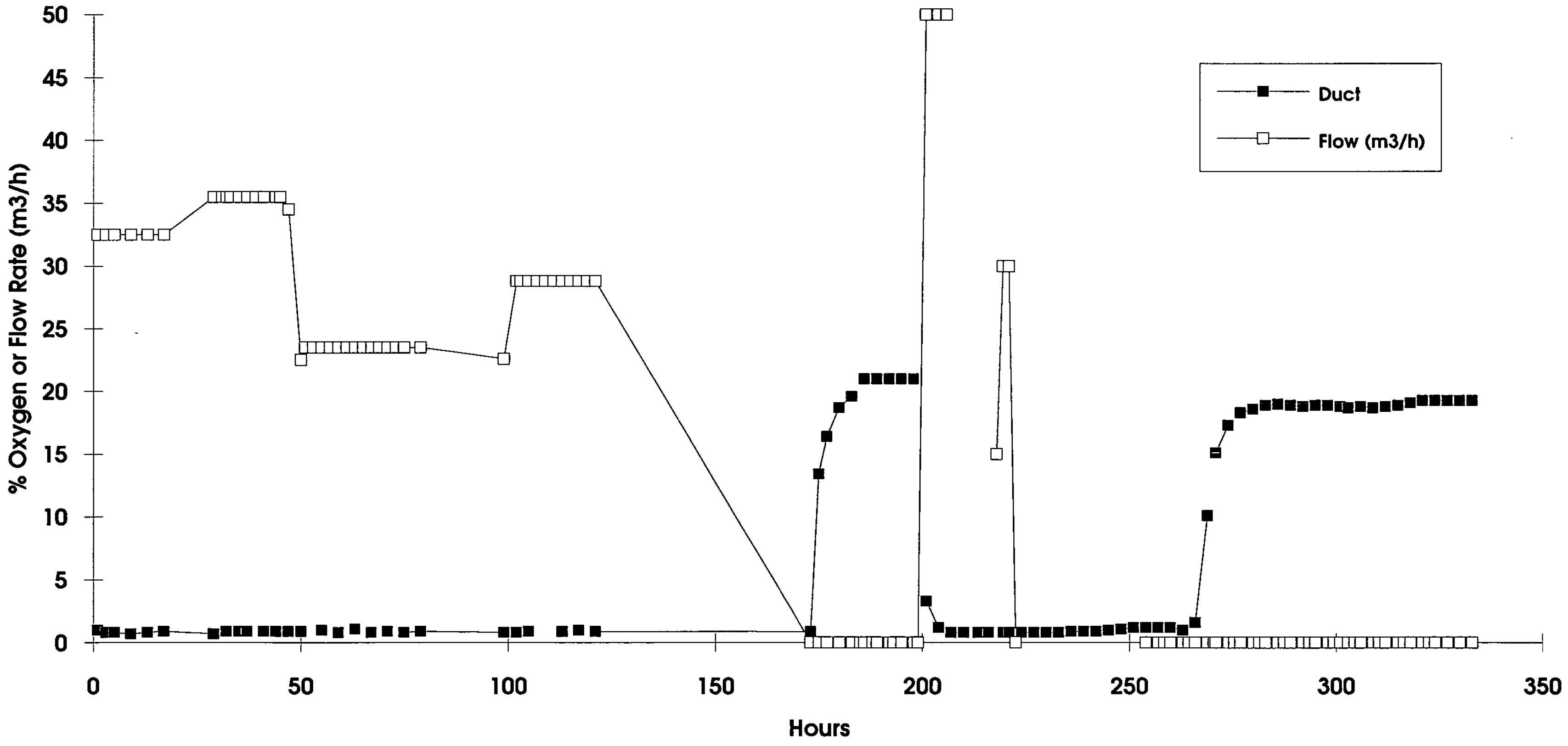


Figure 5: Shrewton - The effect of flow rate on the oxygen level in the silo

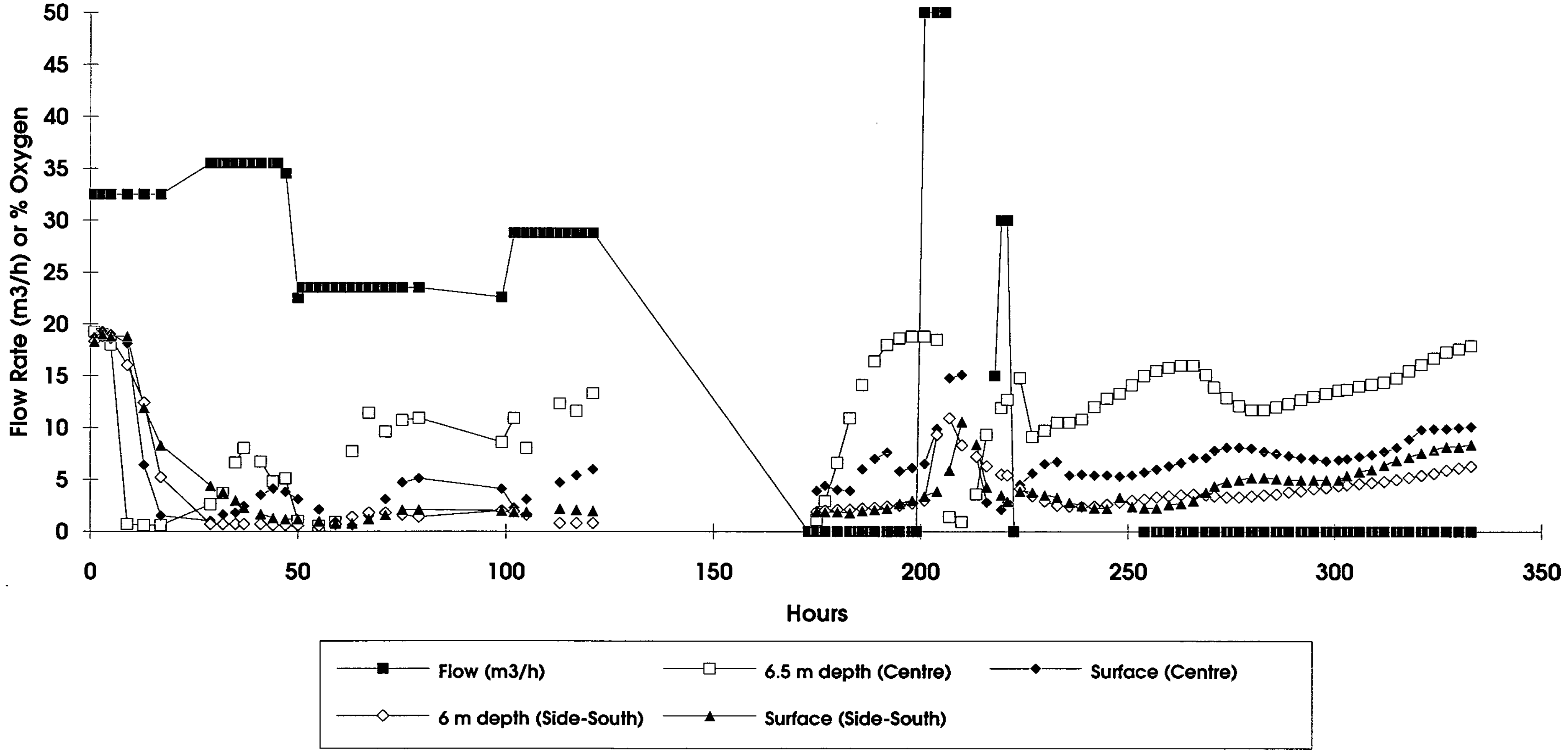


Figure 6: Shrewton - Effect of wind speed on the oxygen level at the centre of the silo

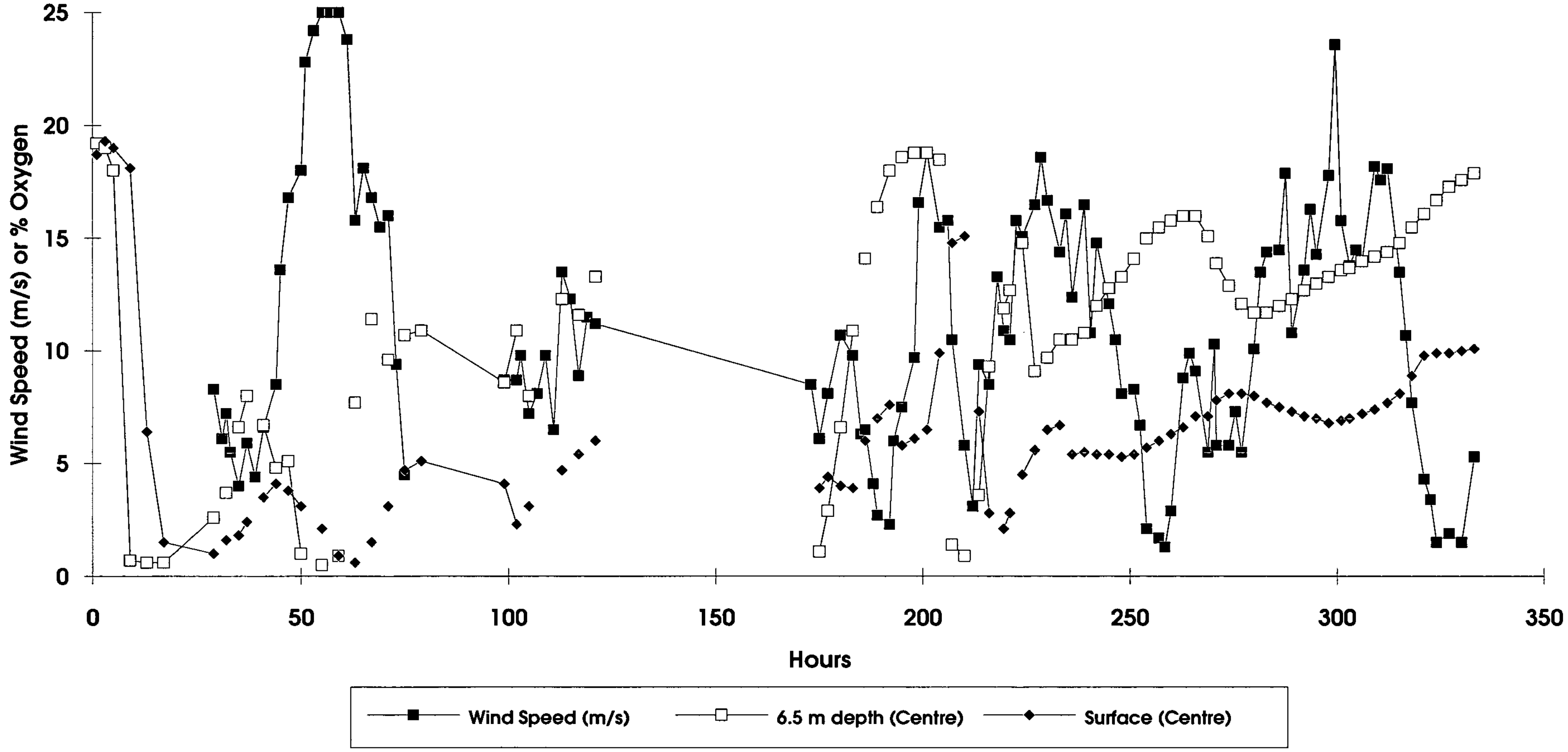


Figure 7: Shrewton - Effect of wind speed on the oxygen levels at the side of the silo

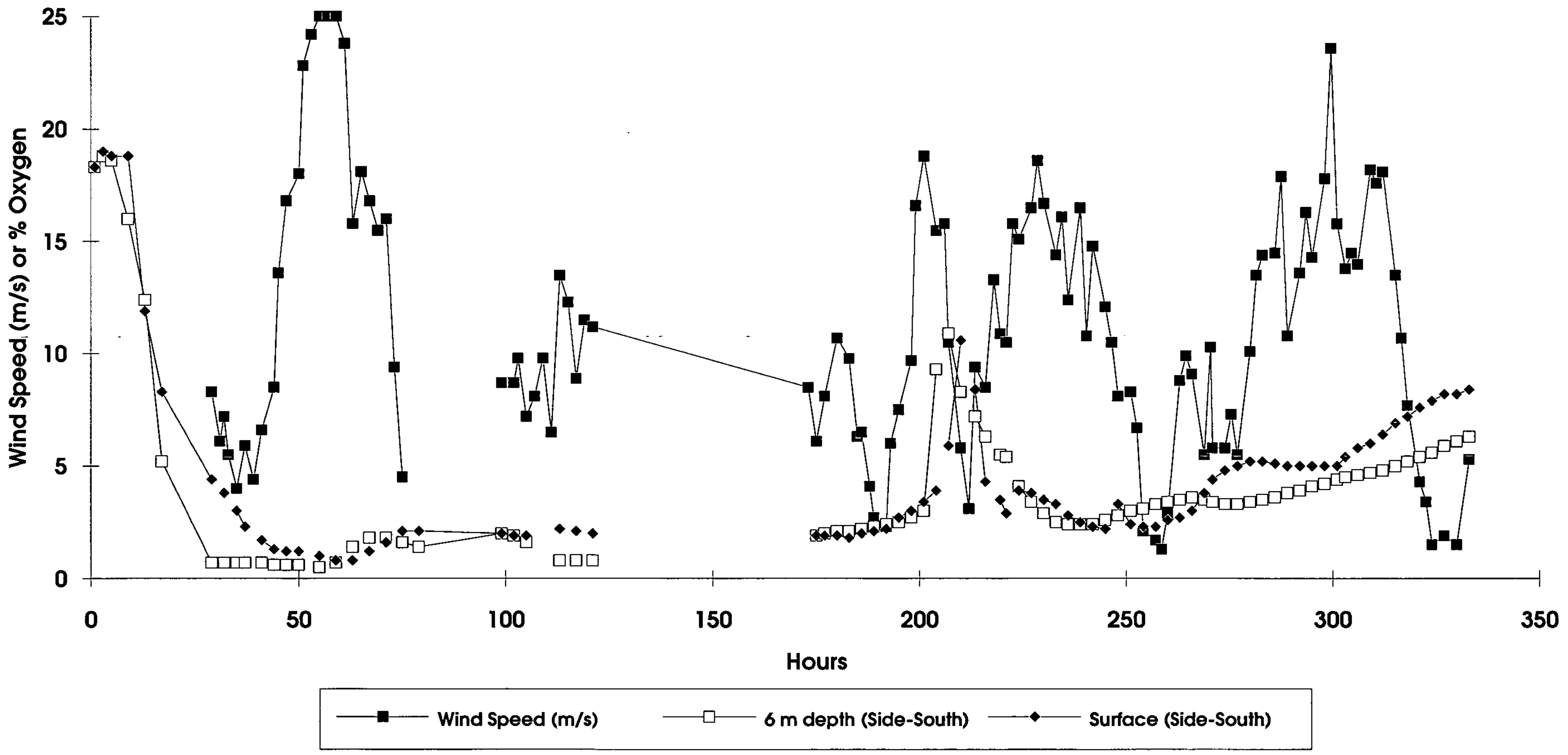


Figure 8: Wallingford - The change in oxygen levels in a silo of malt with different flow rates from the Aerogenerator propane burner.

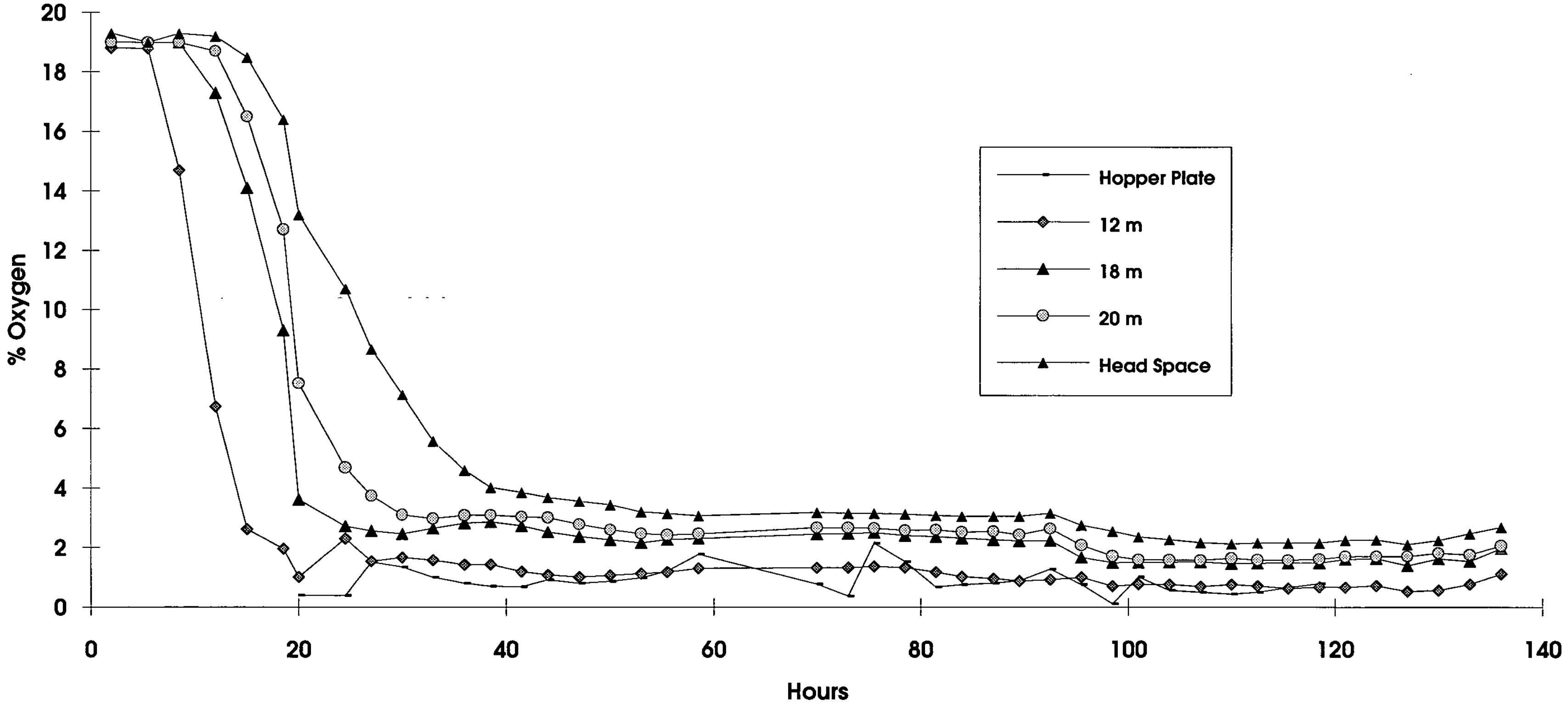


Figure 9: Micheldever - Output flow rate and contents

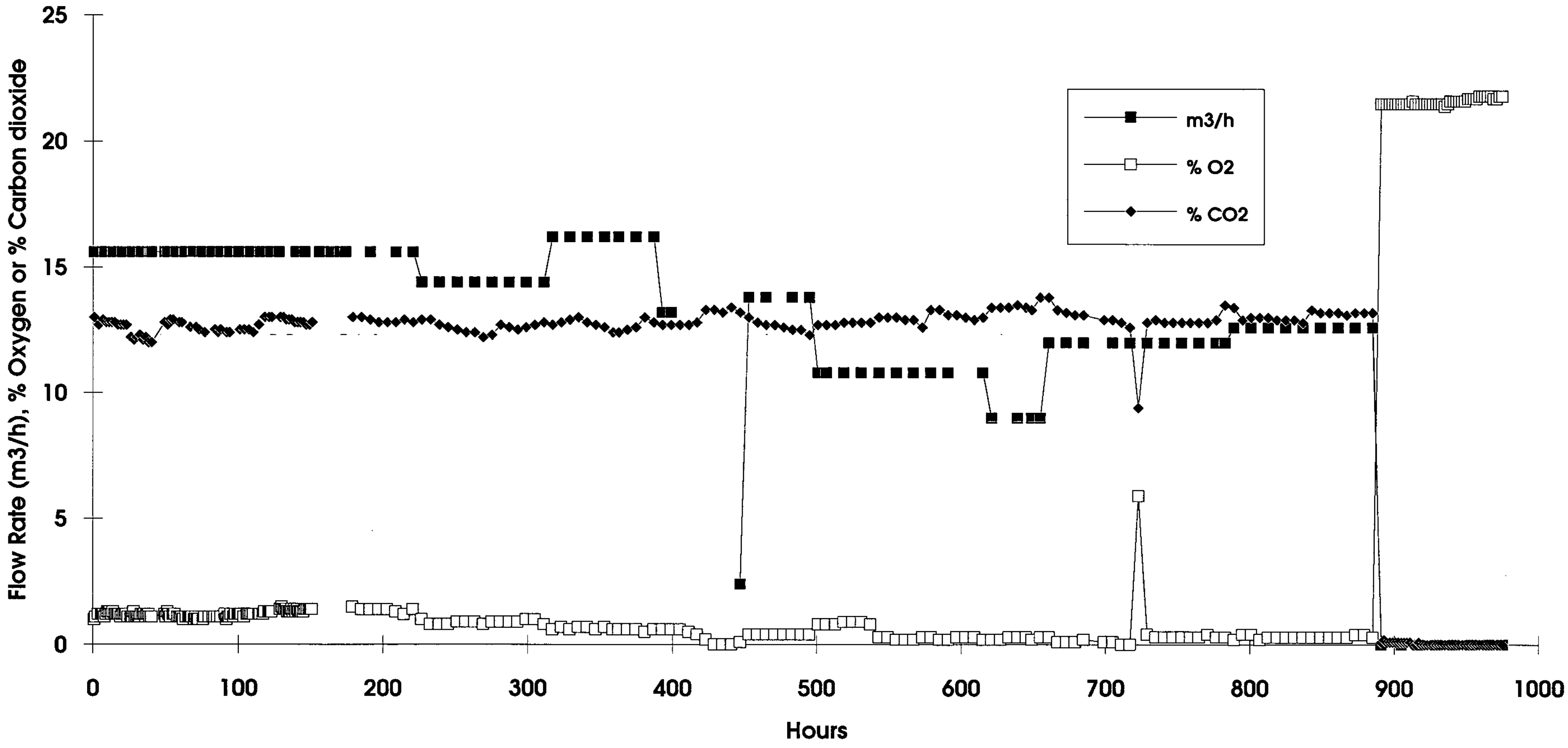


Figure 10: Micheldever - Silo oxygen contents during the purge

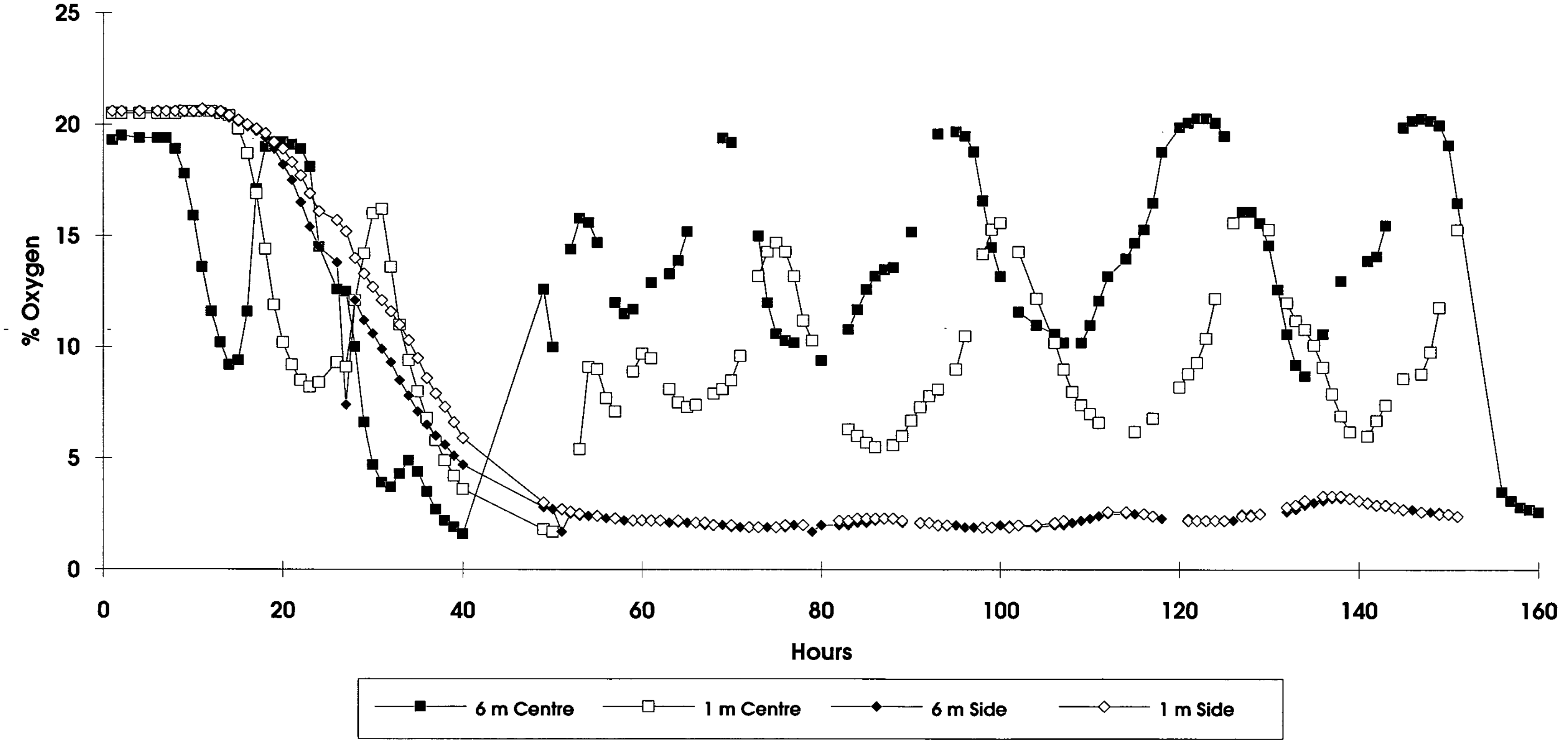


Figure 11: Micheldever - Environmental conditions during the purge

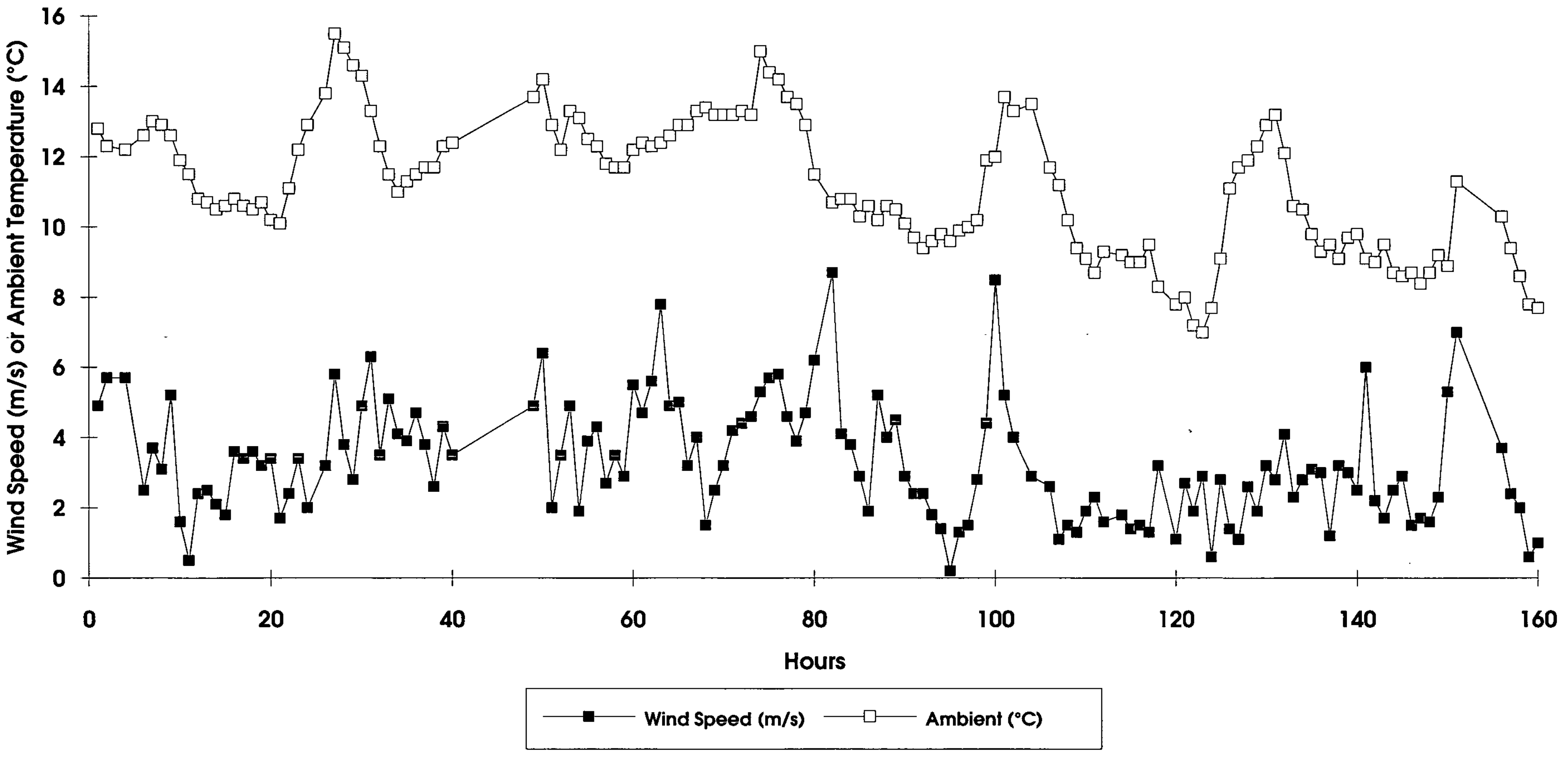


Figure 12: Micheldever - Effect of wind speed on the oxygen content at the centre of the silo

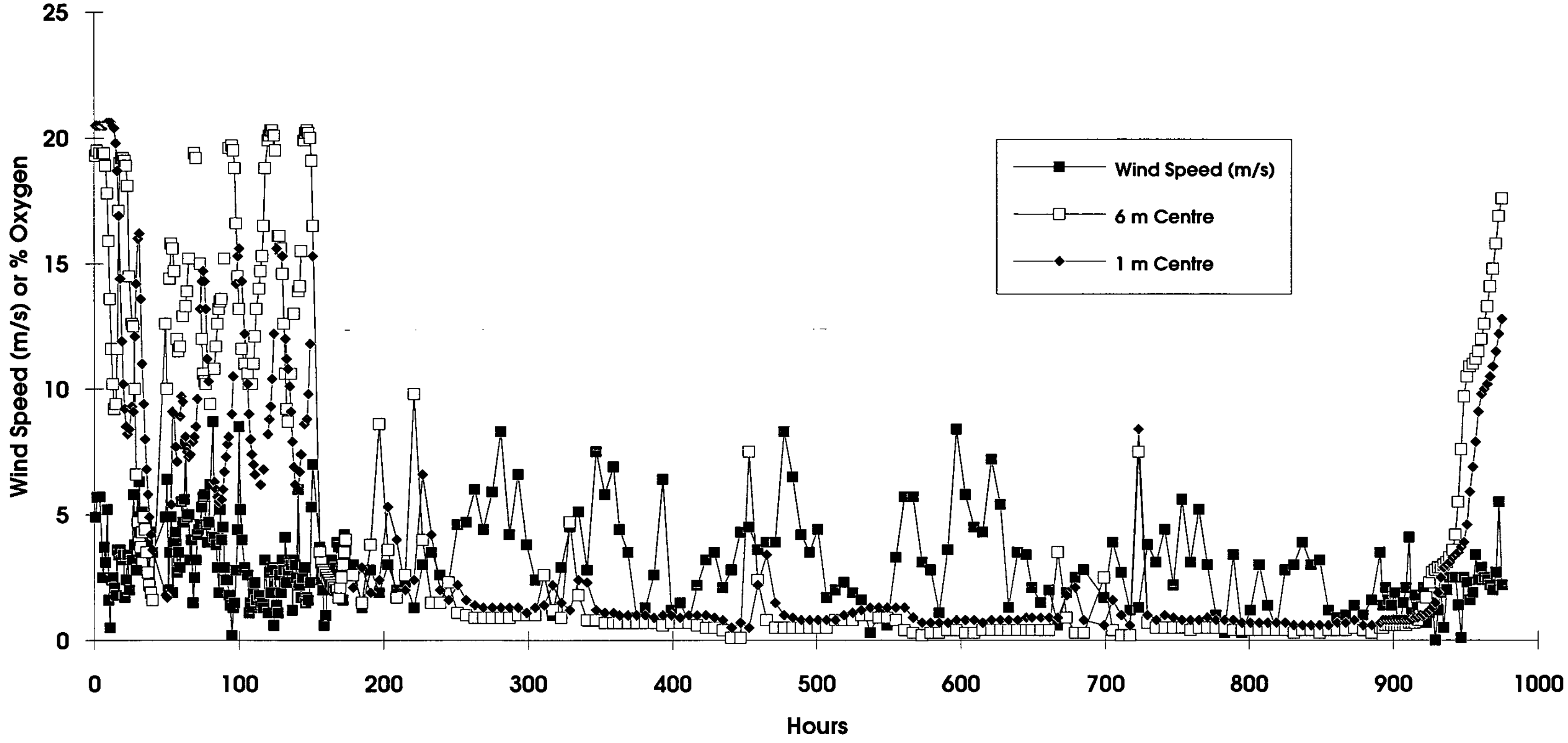


Figure 13: Micheldever - Effect of wind speed on the oxygen content at the side of the silo

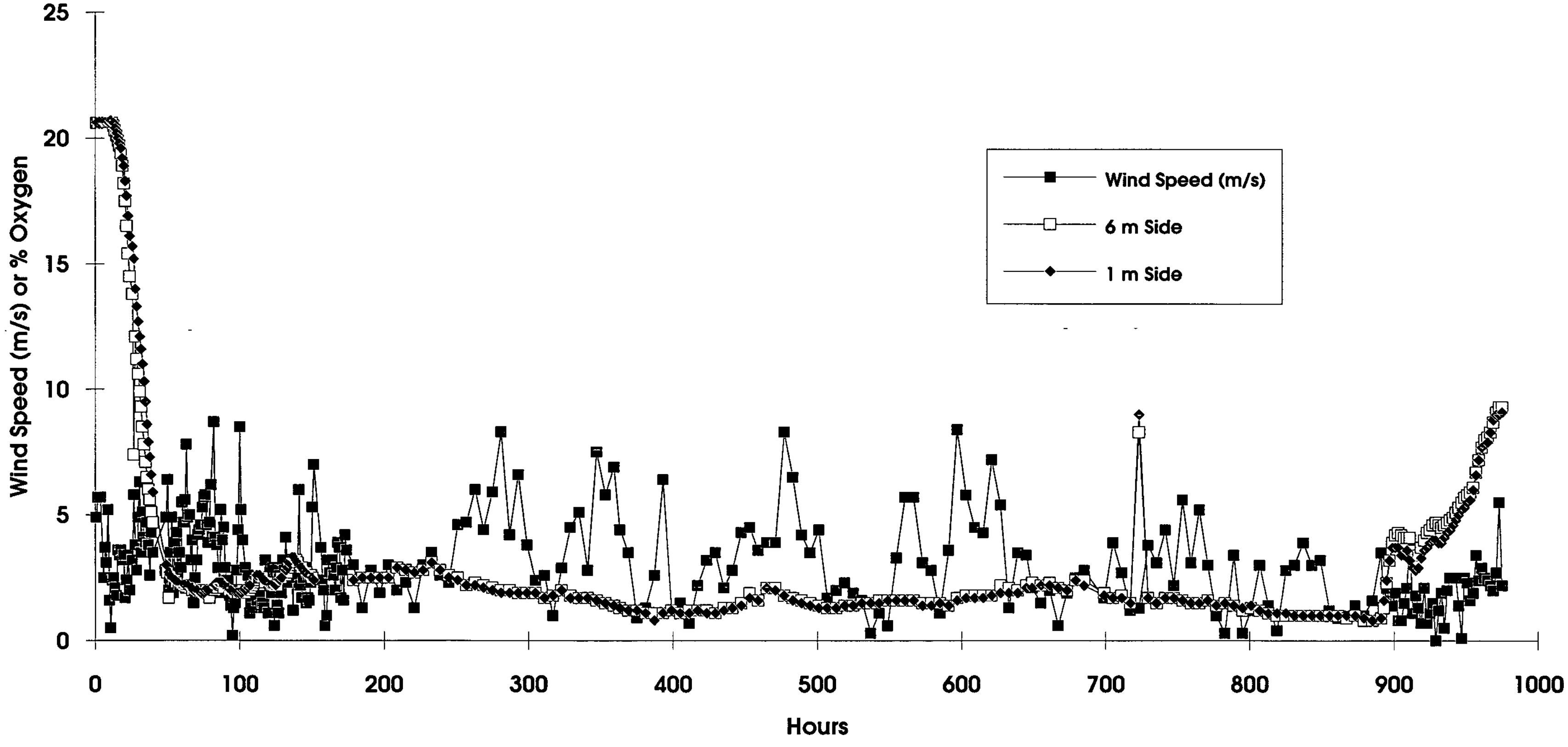


Figure 14: Micheldever - Effect of ambient temperature on the oxygen content in the centre of the silo

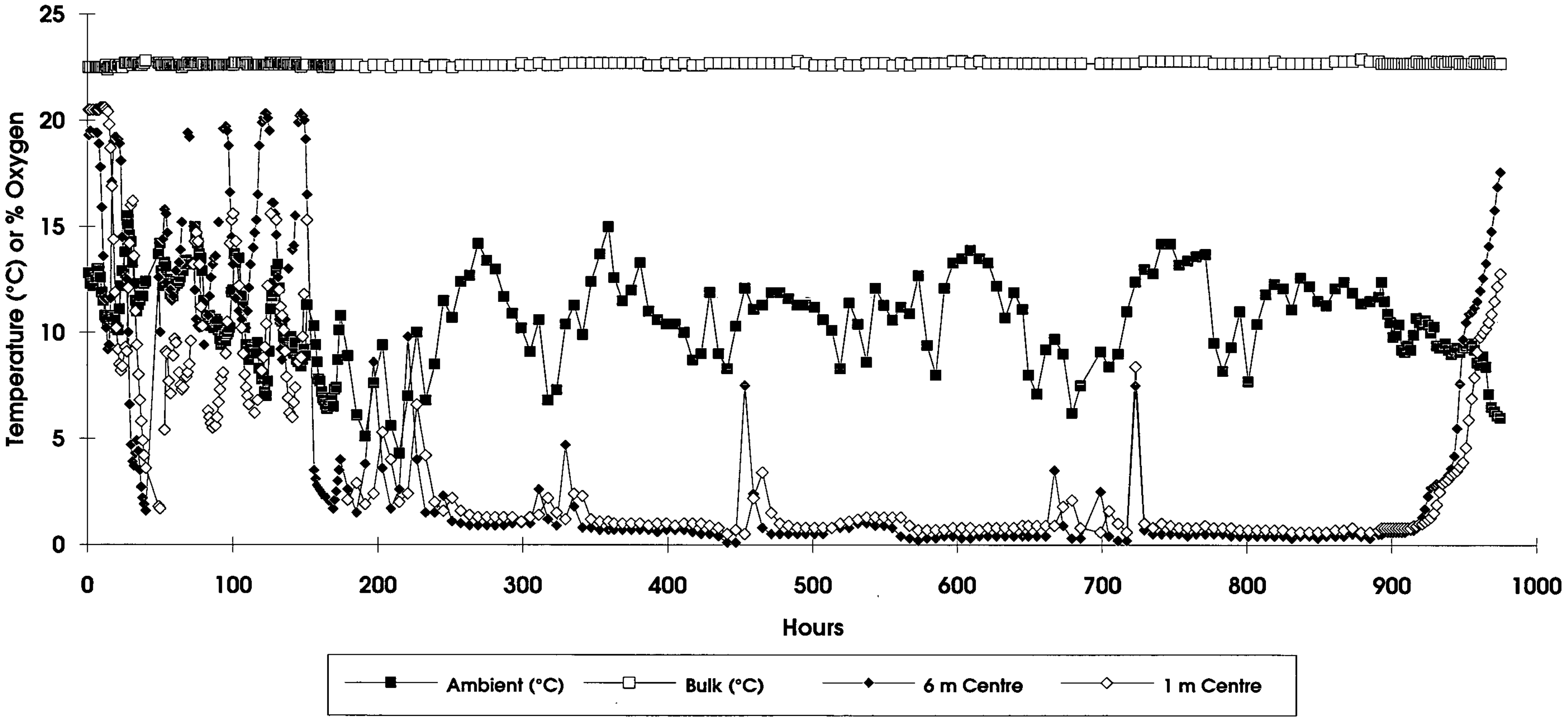


Figure 15: Micheldever - Effect of ambient temperature on the oxygen content at the side of the silo

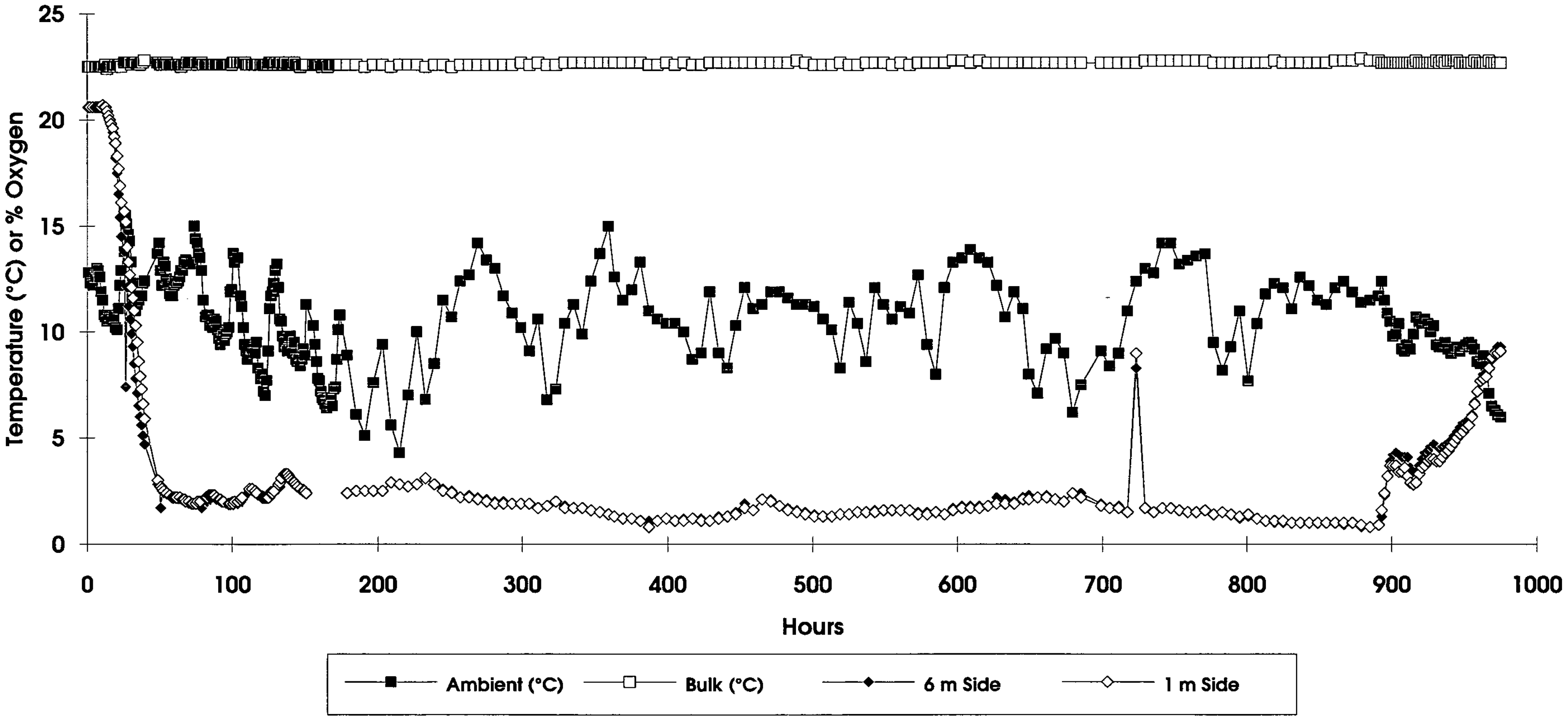


Figure 16: Micheldever - Change in the silo oxygen content after the flow was stopped

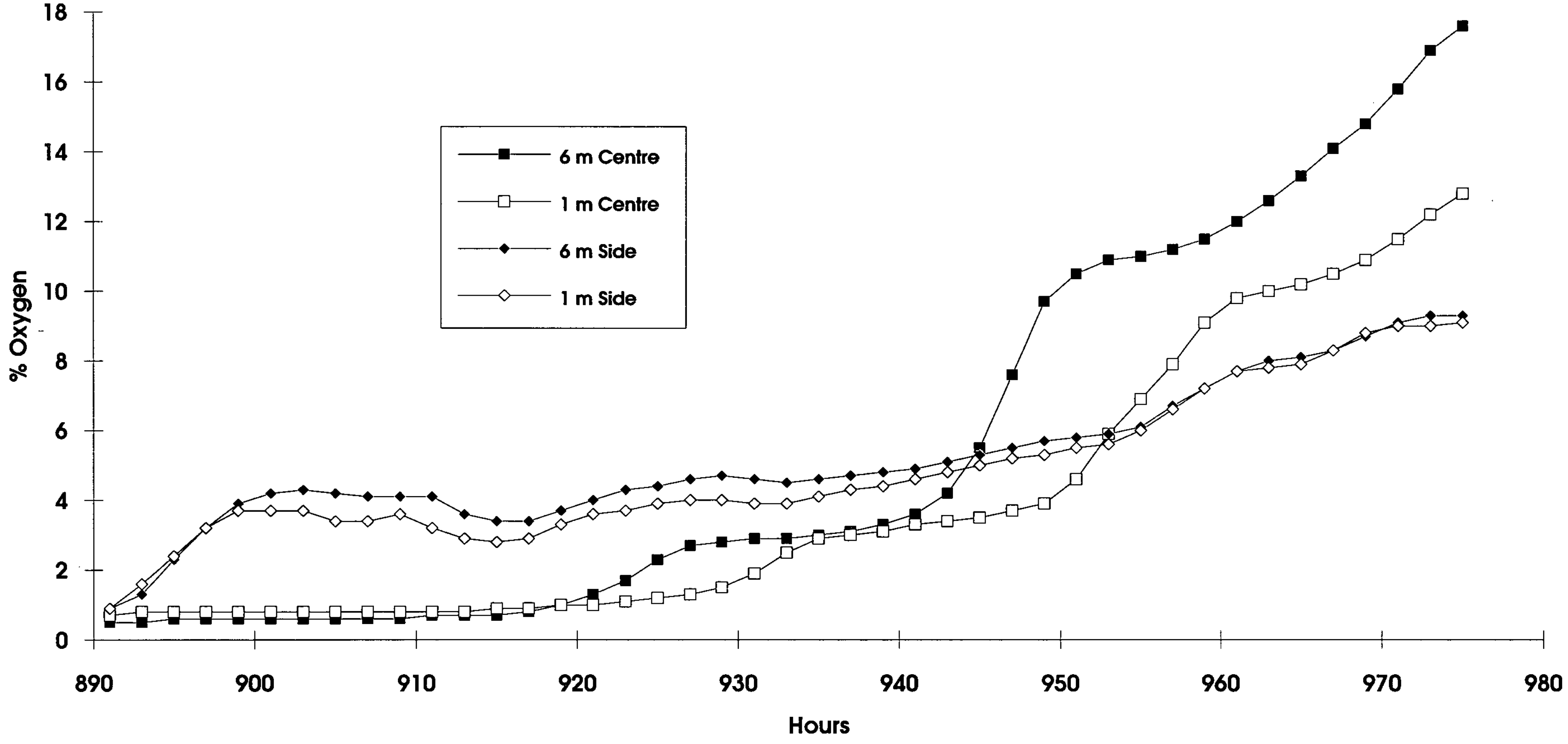


Figure 17: Micheldever - Environmental conditions after the flow was stopped

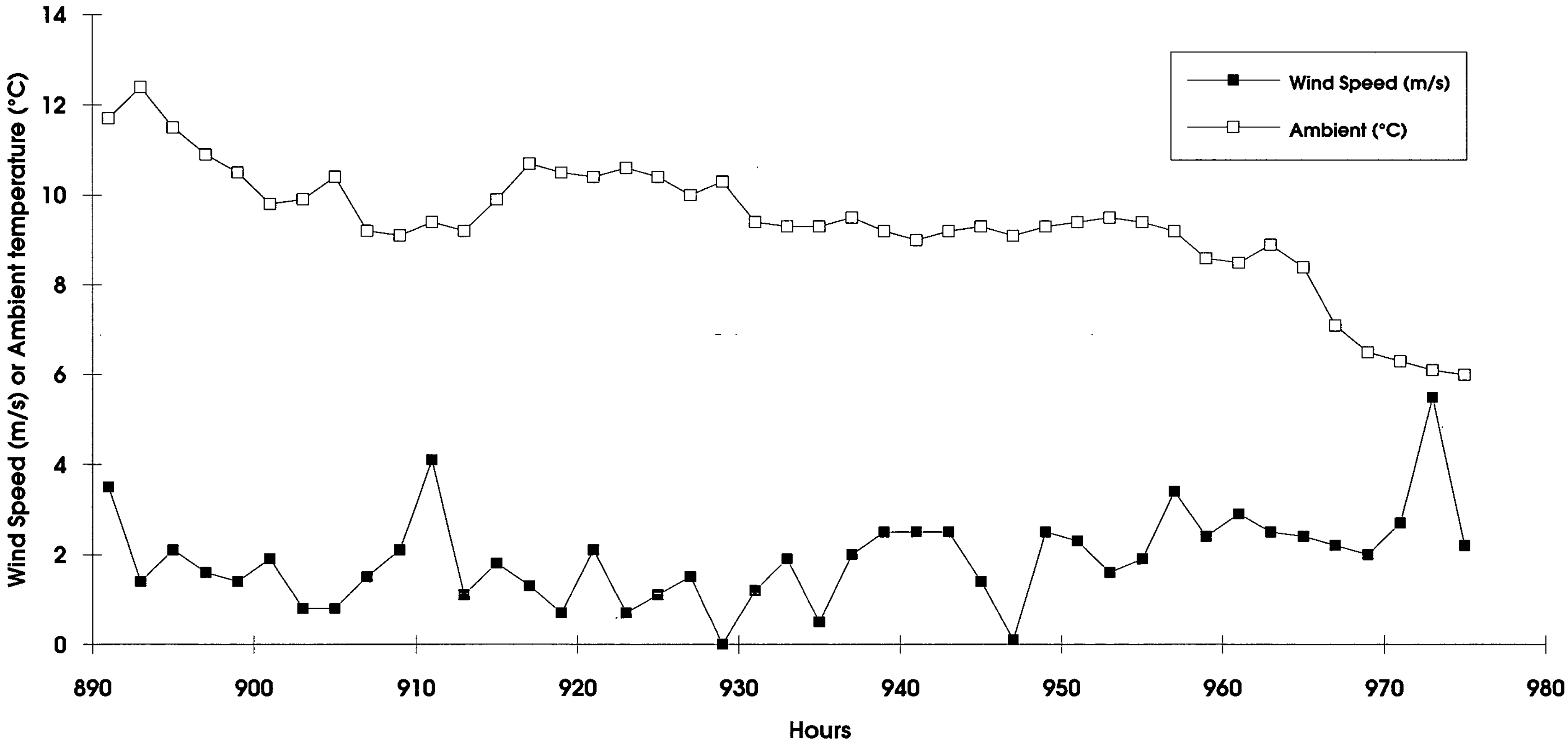


Figure 18: Micheldever - Change in the mean silo oxygen content due to flow rate

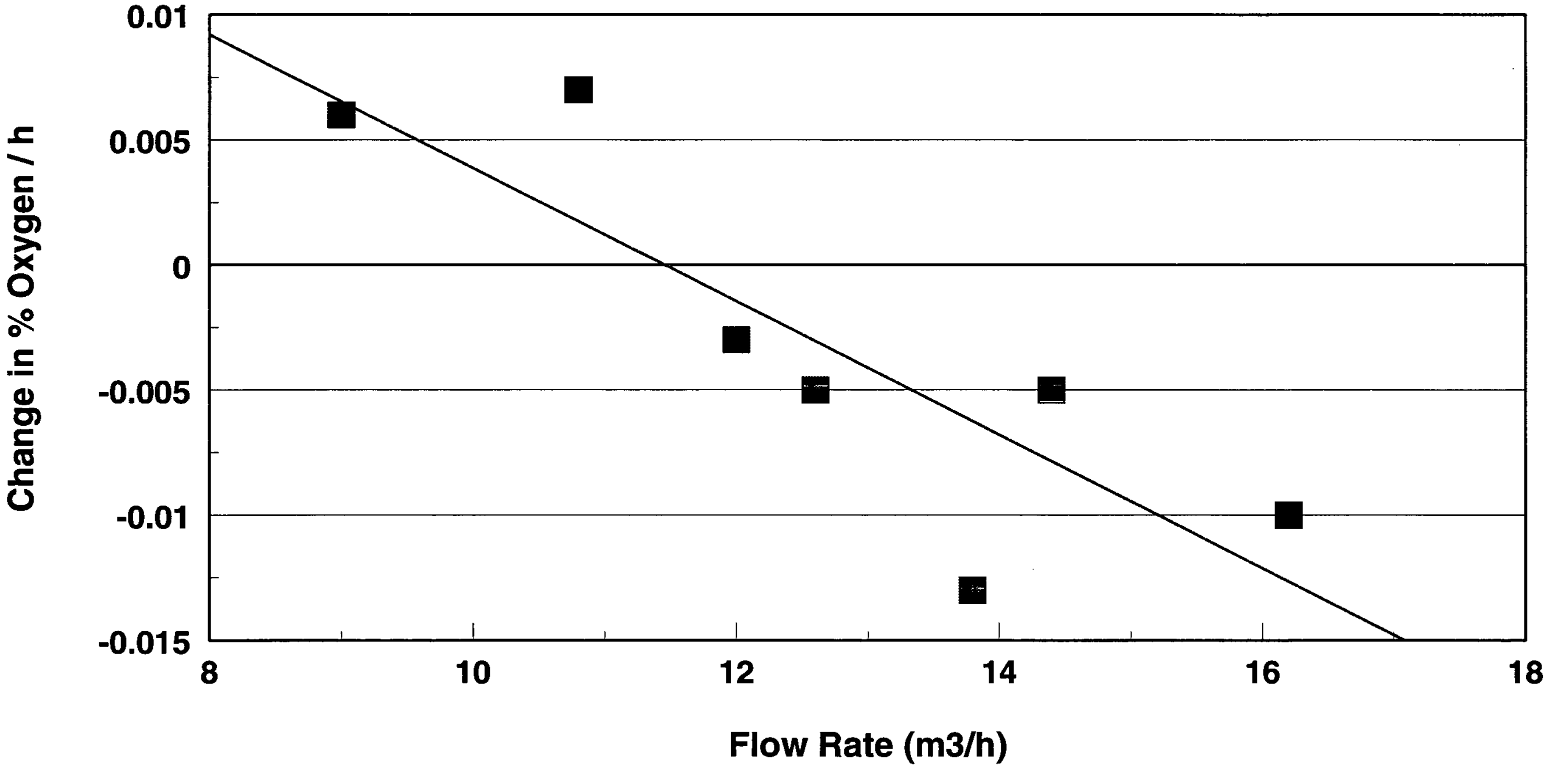


Figure 19: Hursley - Output flow rate and contents

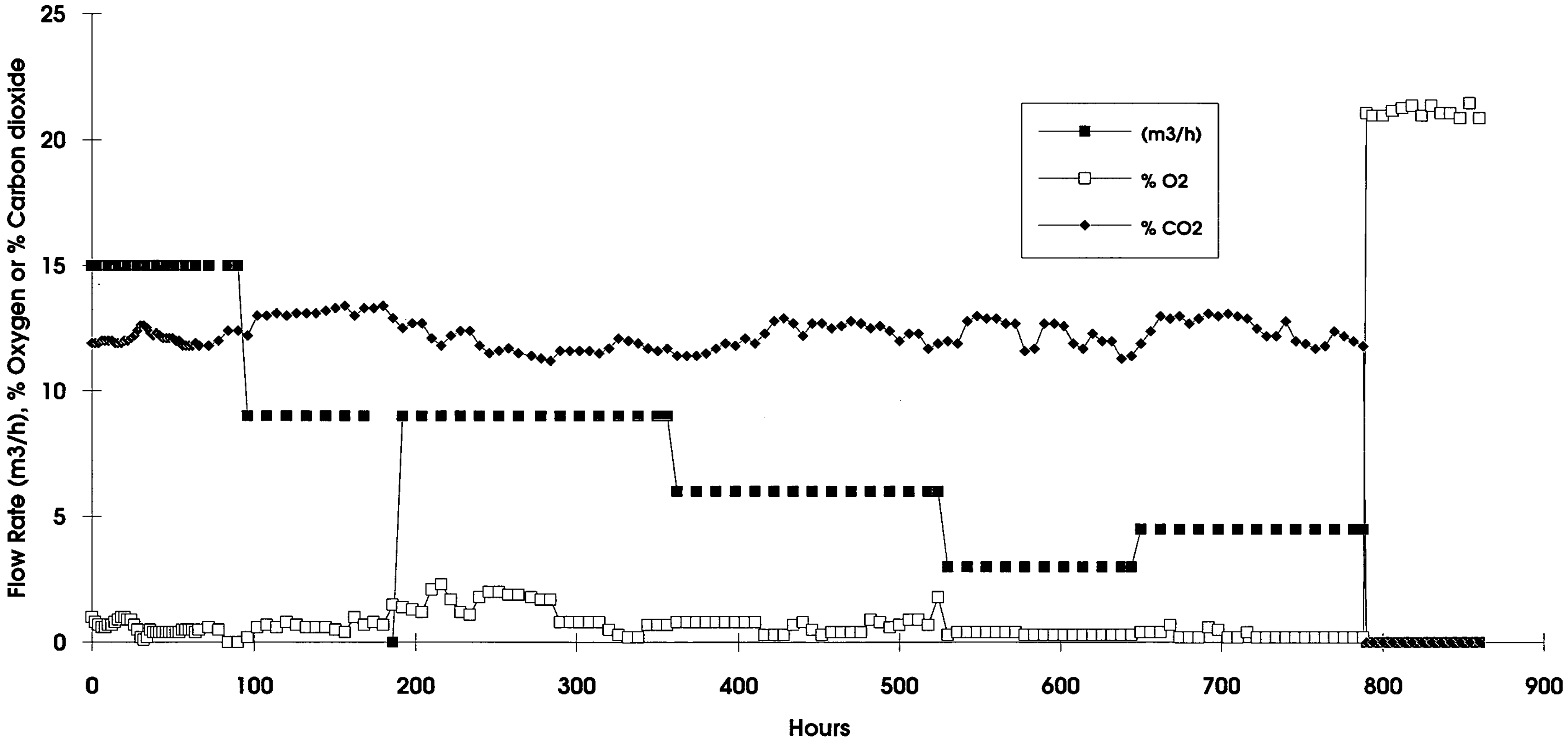


Figure 20: Hursley - Silo oxygen contents during the purge

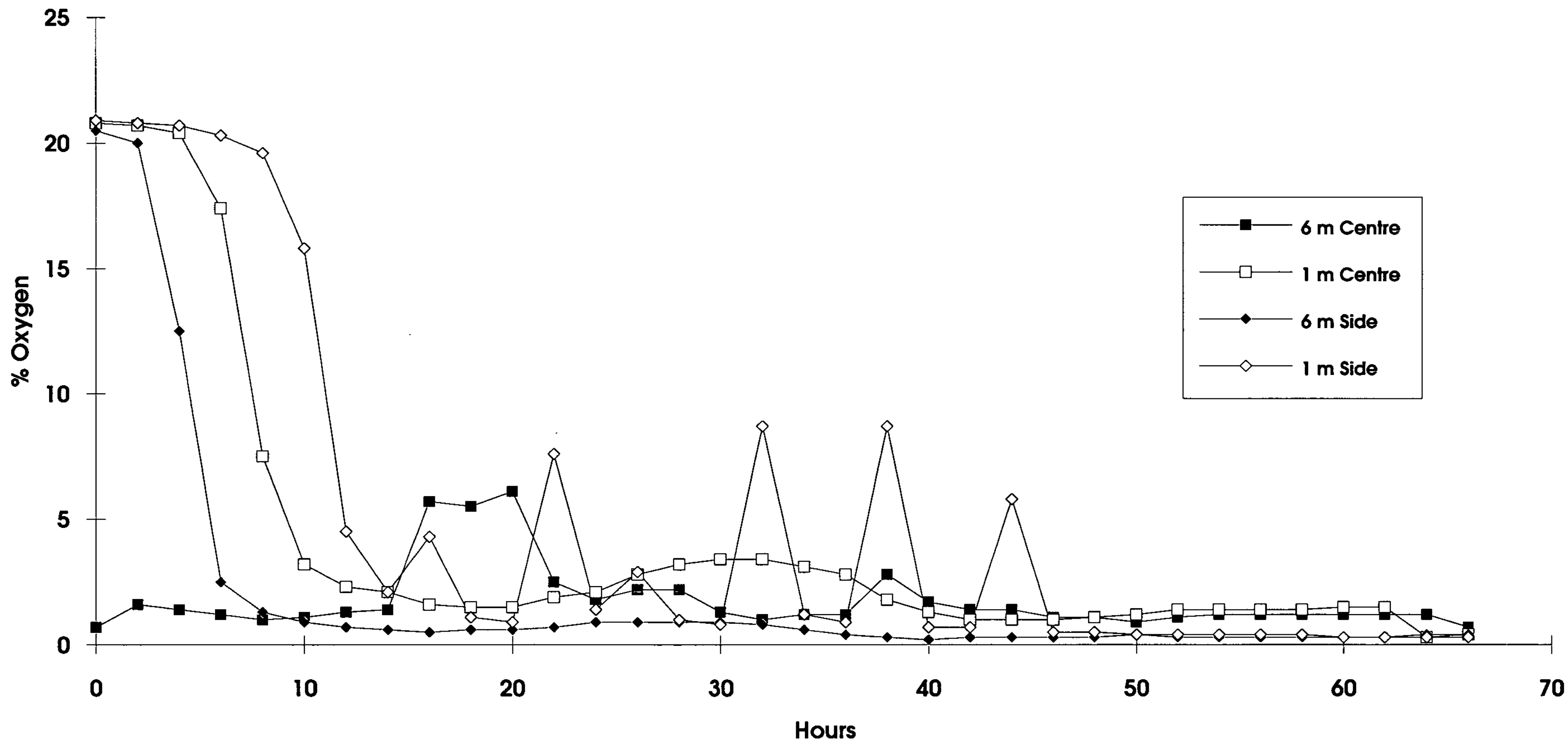


Figure 21: Hursley - Environmental conditions during the purge

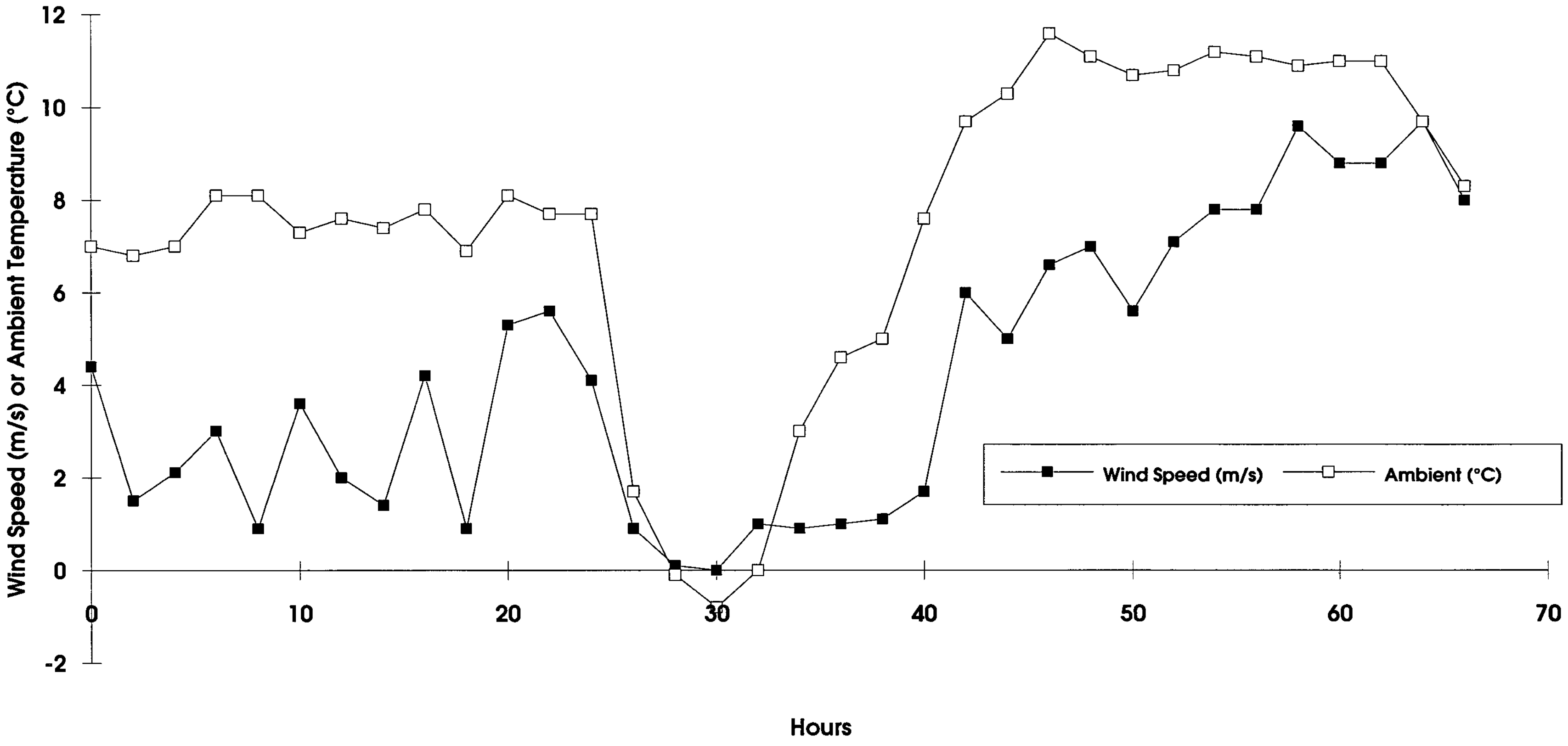


Figure 22: Hursley - Effect of wind speed on the oxygen content at the centre of the silo

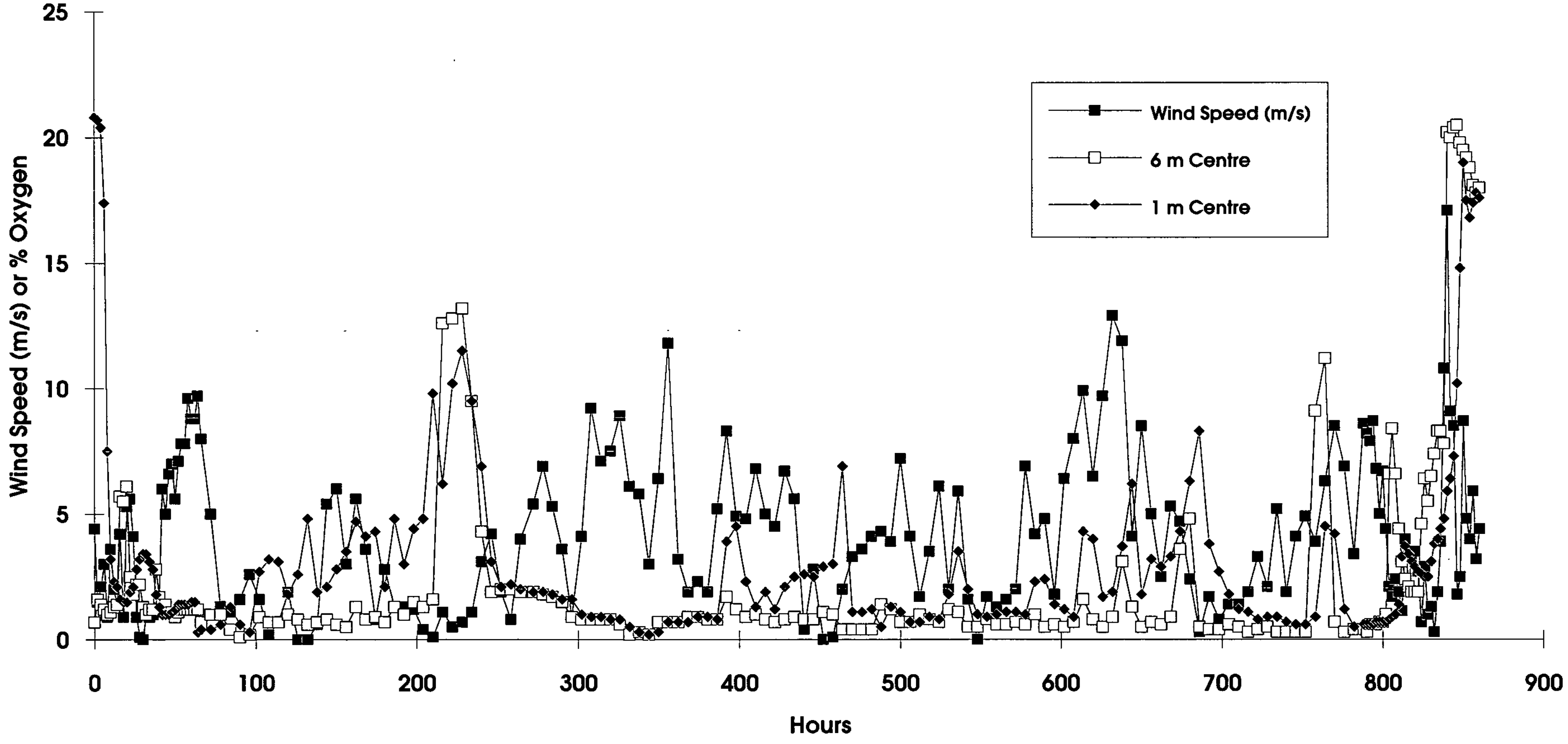


Figure 23: Hursley - Effect of wind speed on the oxygen content at the side of the silo

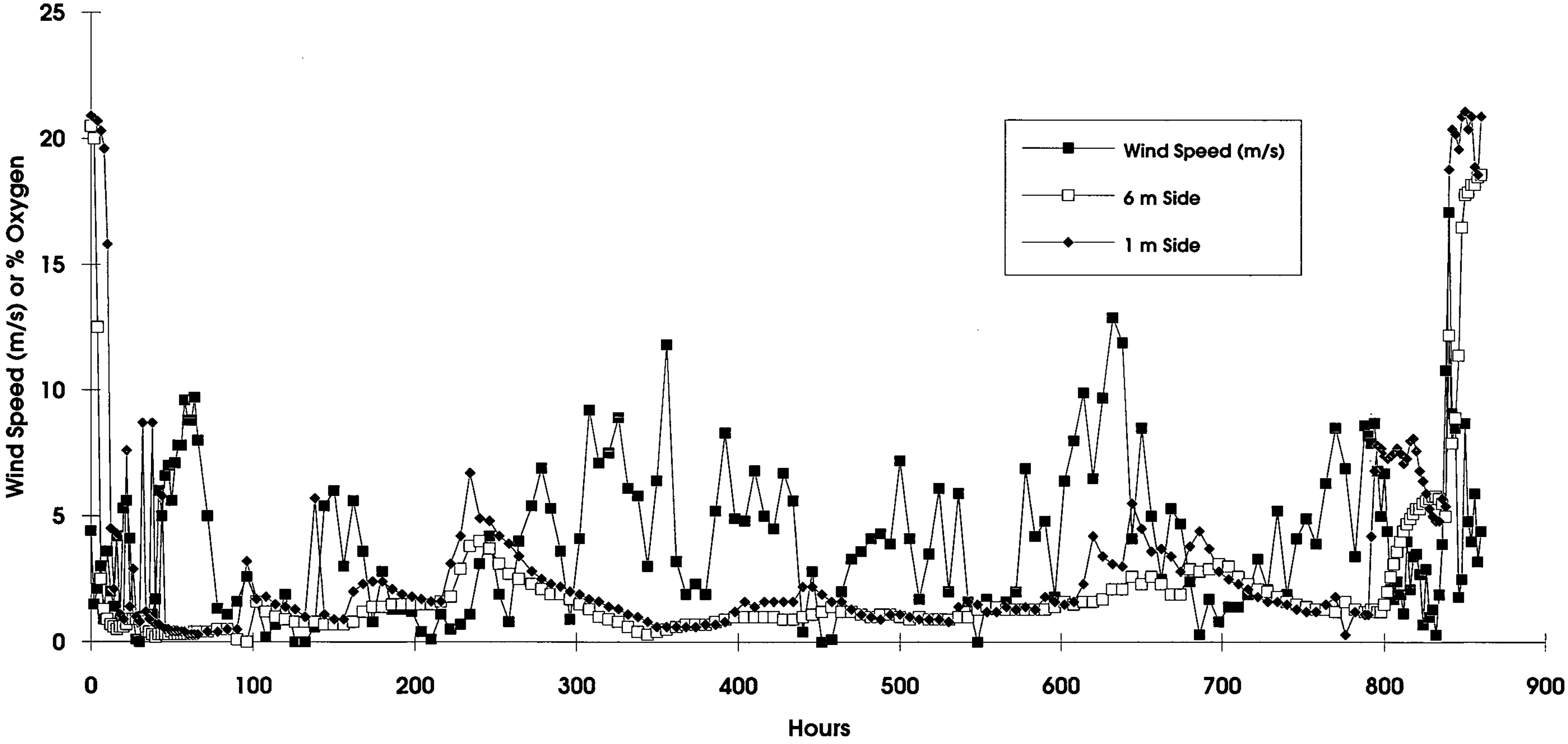


Figure 24: Hursley - Effect of ambient temperature on the oxygen content in the centre of the silo

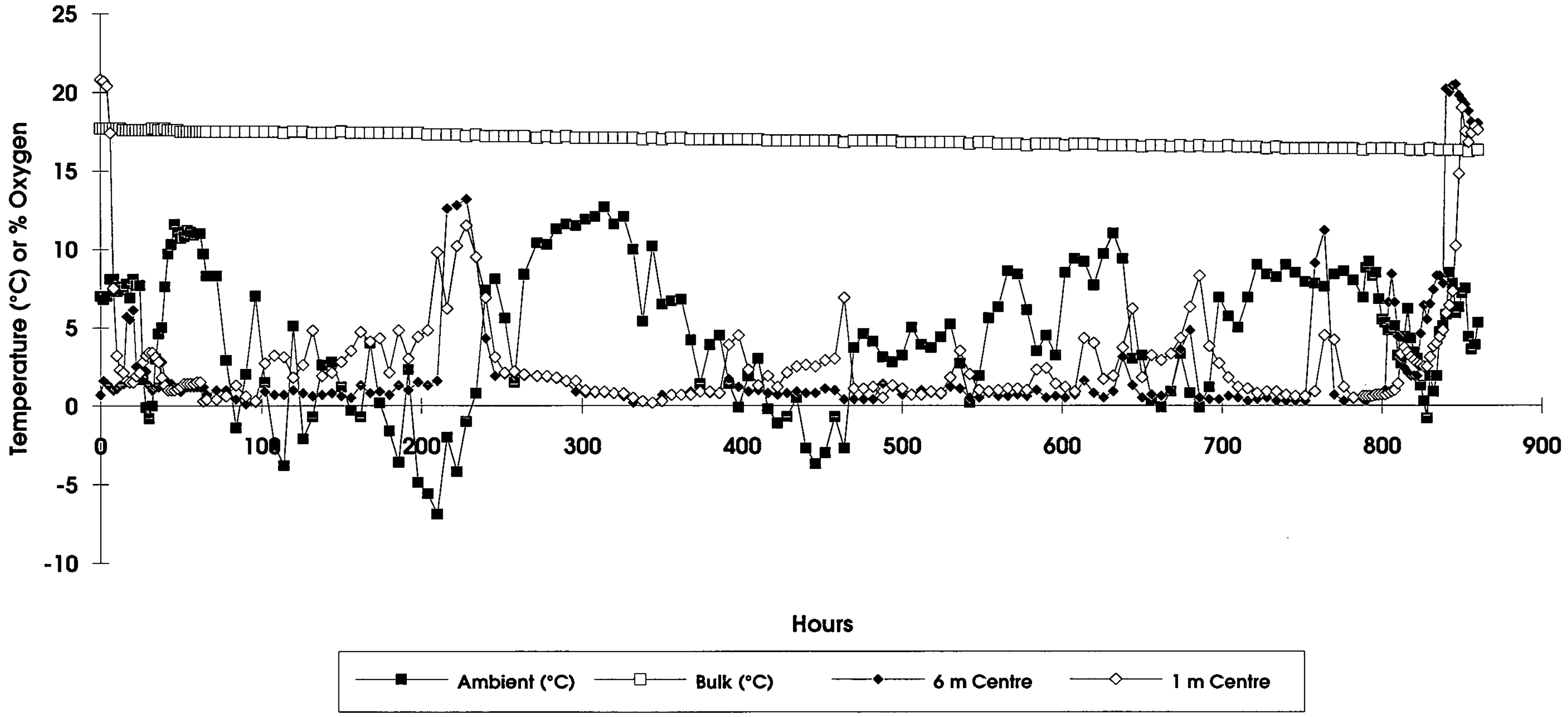


Figure 25: Hursley - Effect of ambient temperature on the oxygen content at the side of the silo

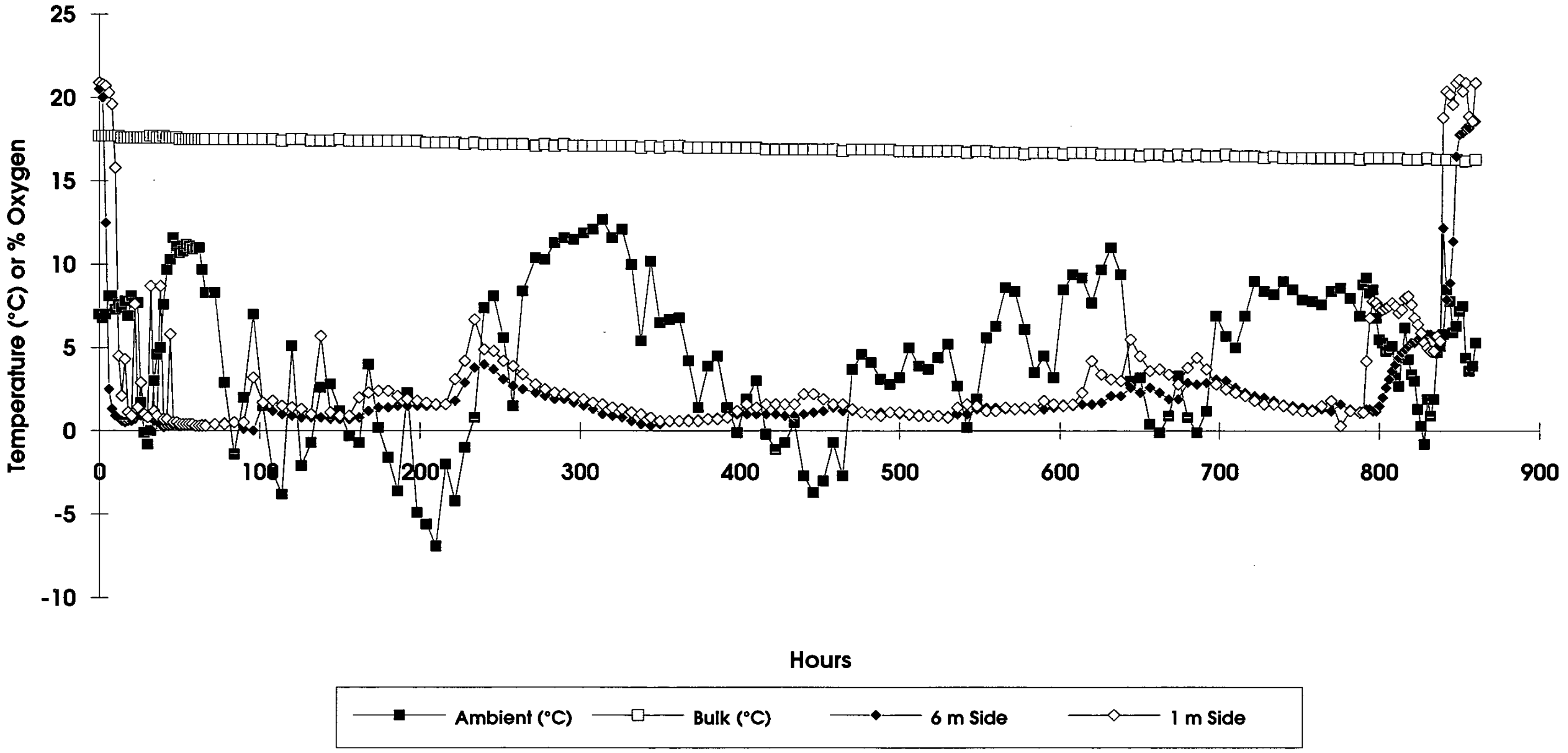


Figure 26: Hursley - Change in the silo oxygen content after the flow was stopped

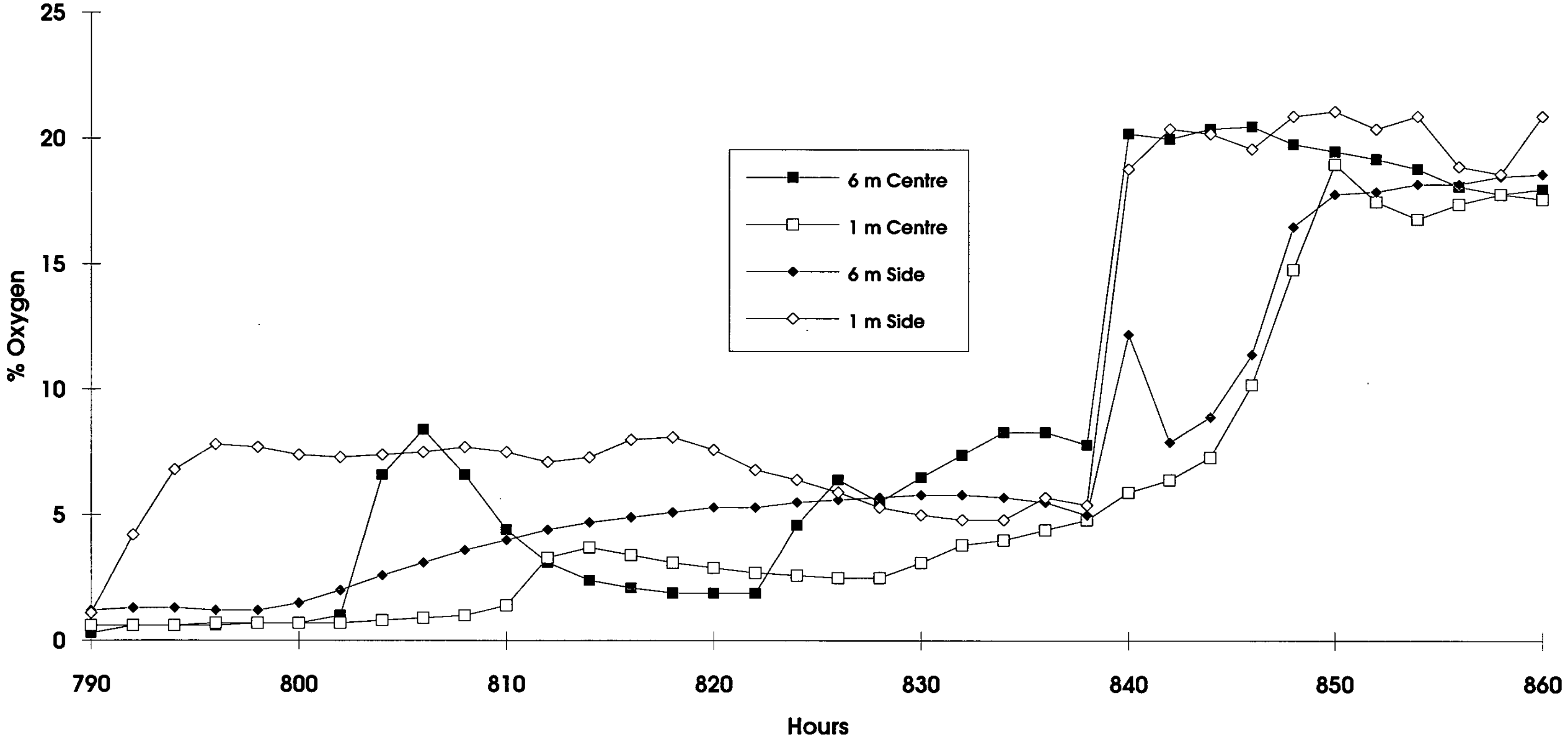


Figure 27: Hursley - Environmental conditions after the flow was stopped

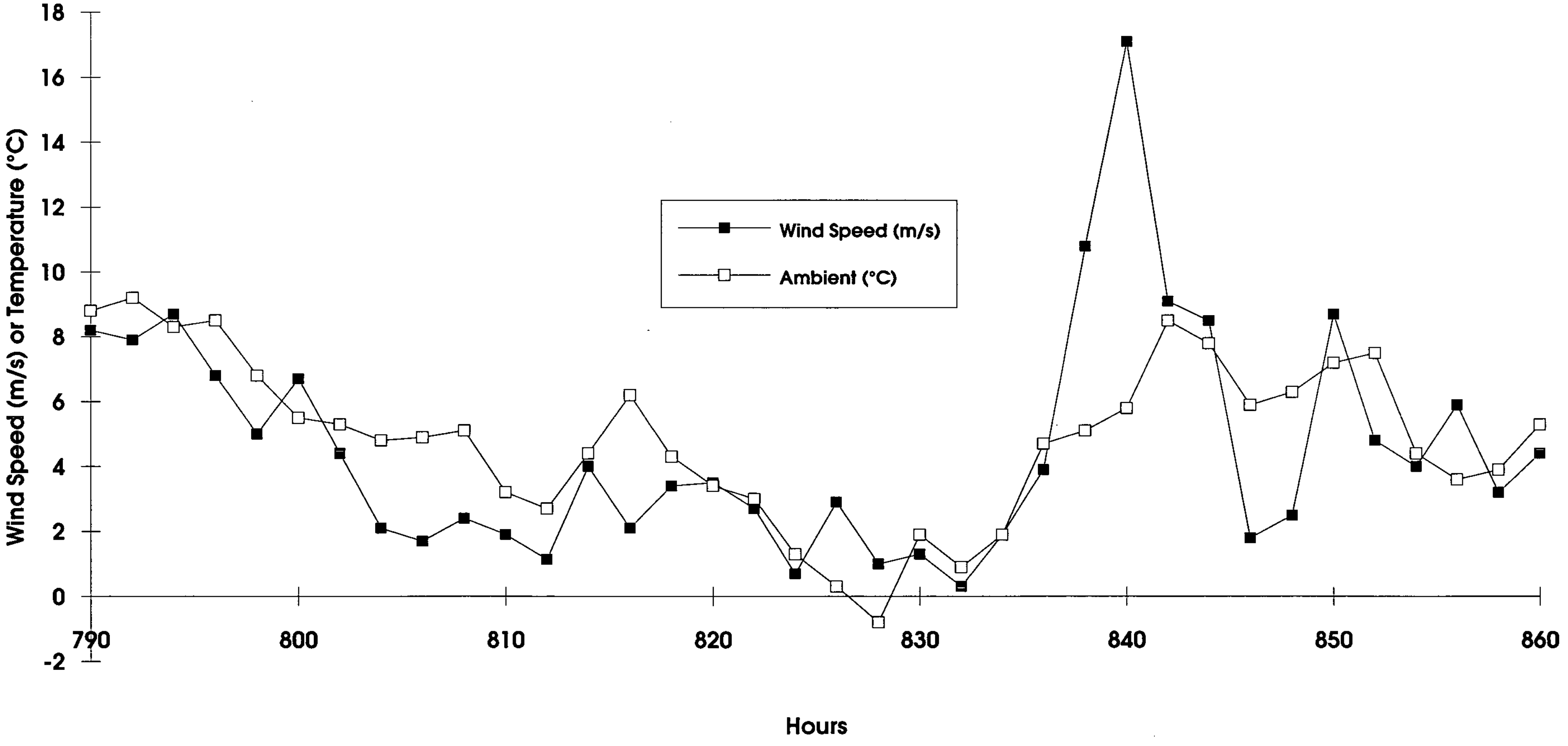


Figure 28: Linton - Output flow rate and contents

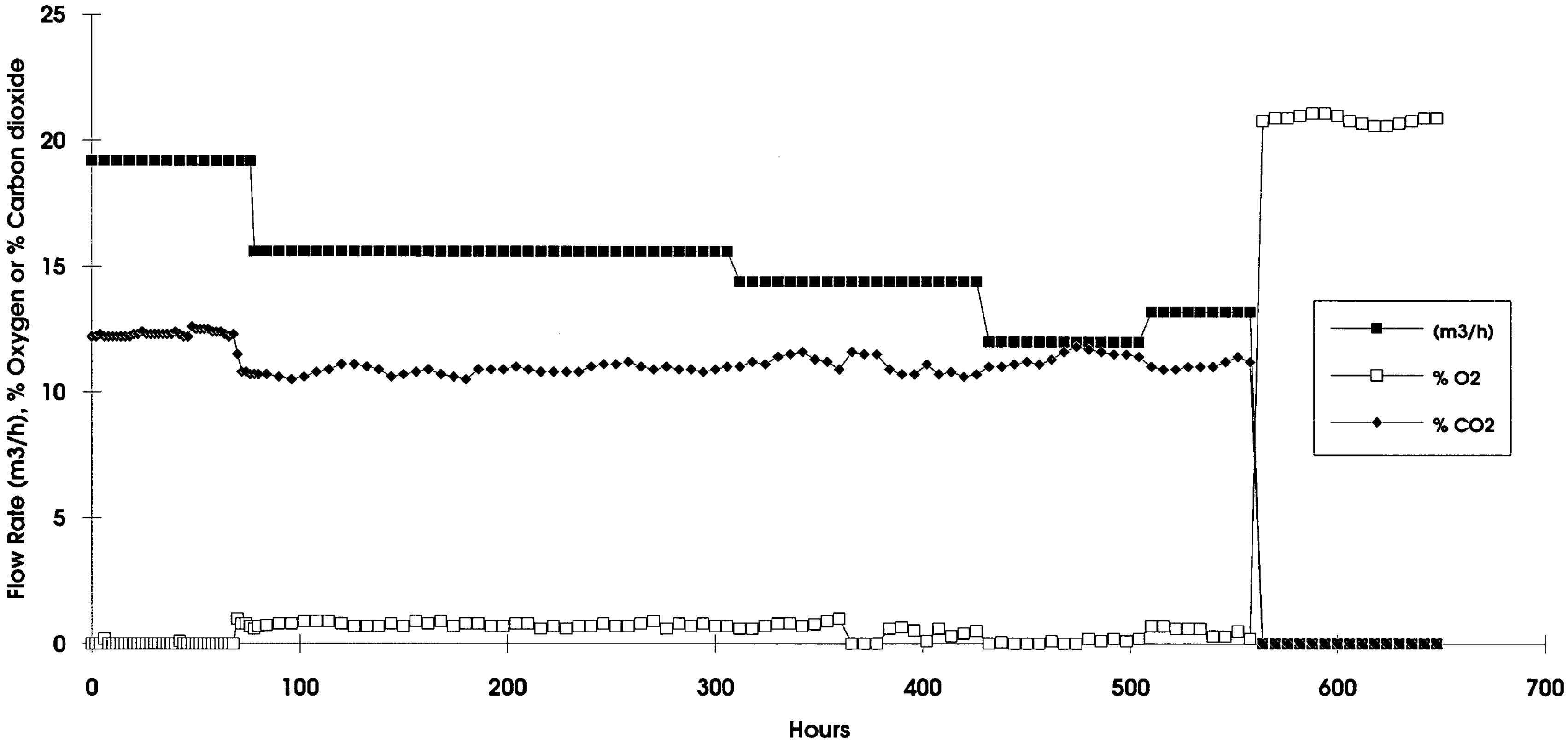


Figure 29: Linton - Silo oxygen content during the purge

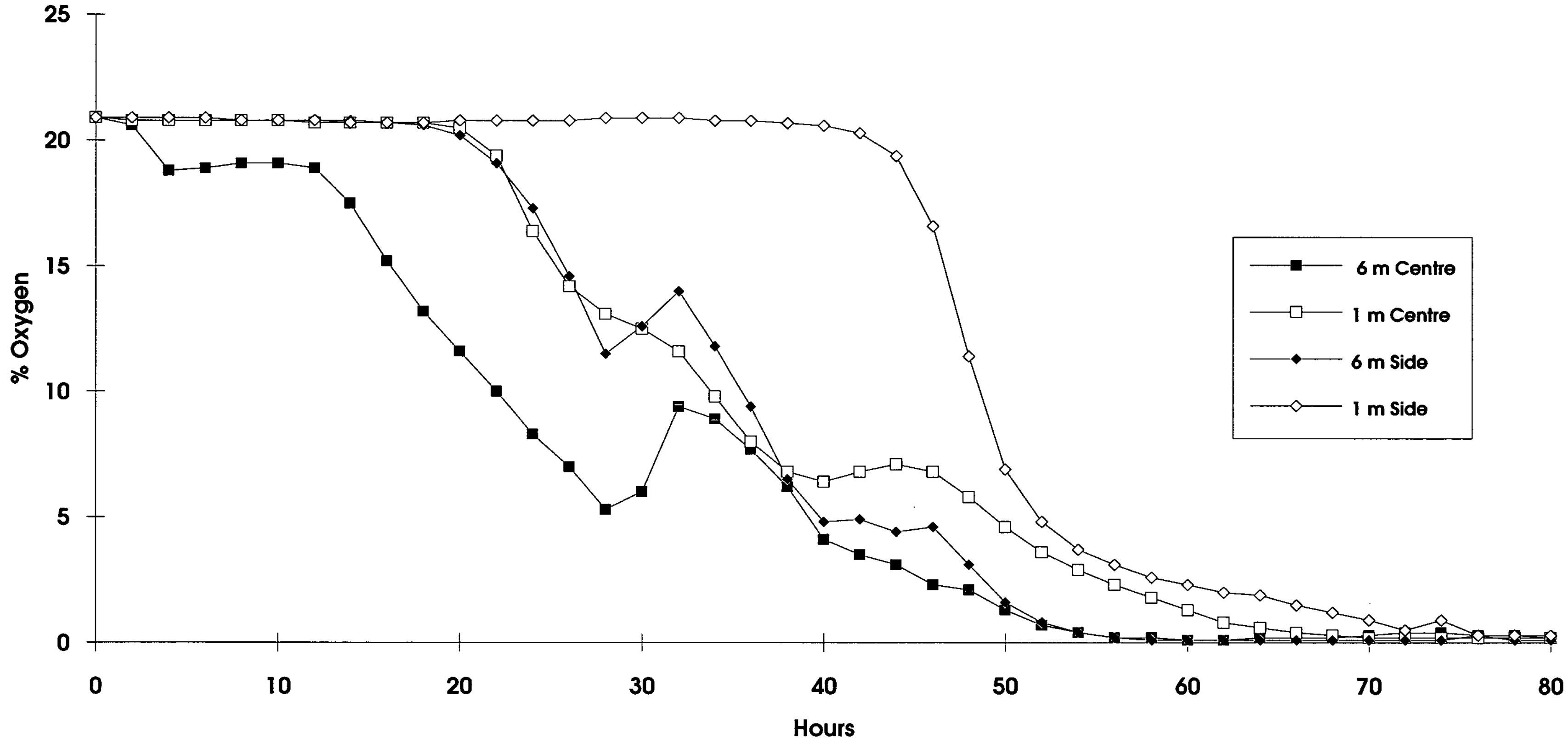


Figure 30: Linton - Environmental conditions during the purge

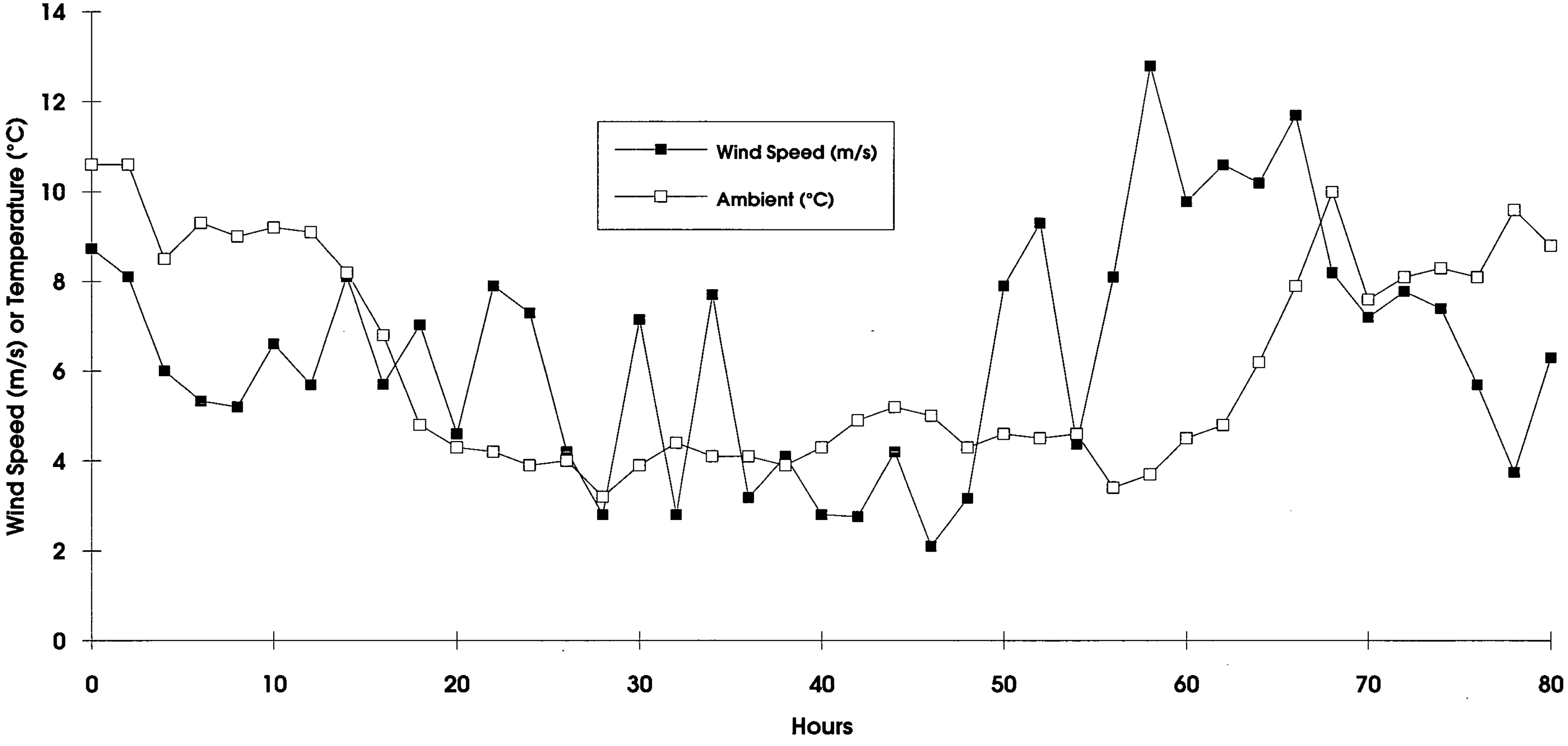


Figure 31: Linton - Wind speed during the trial

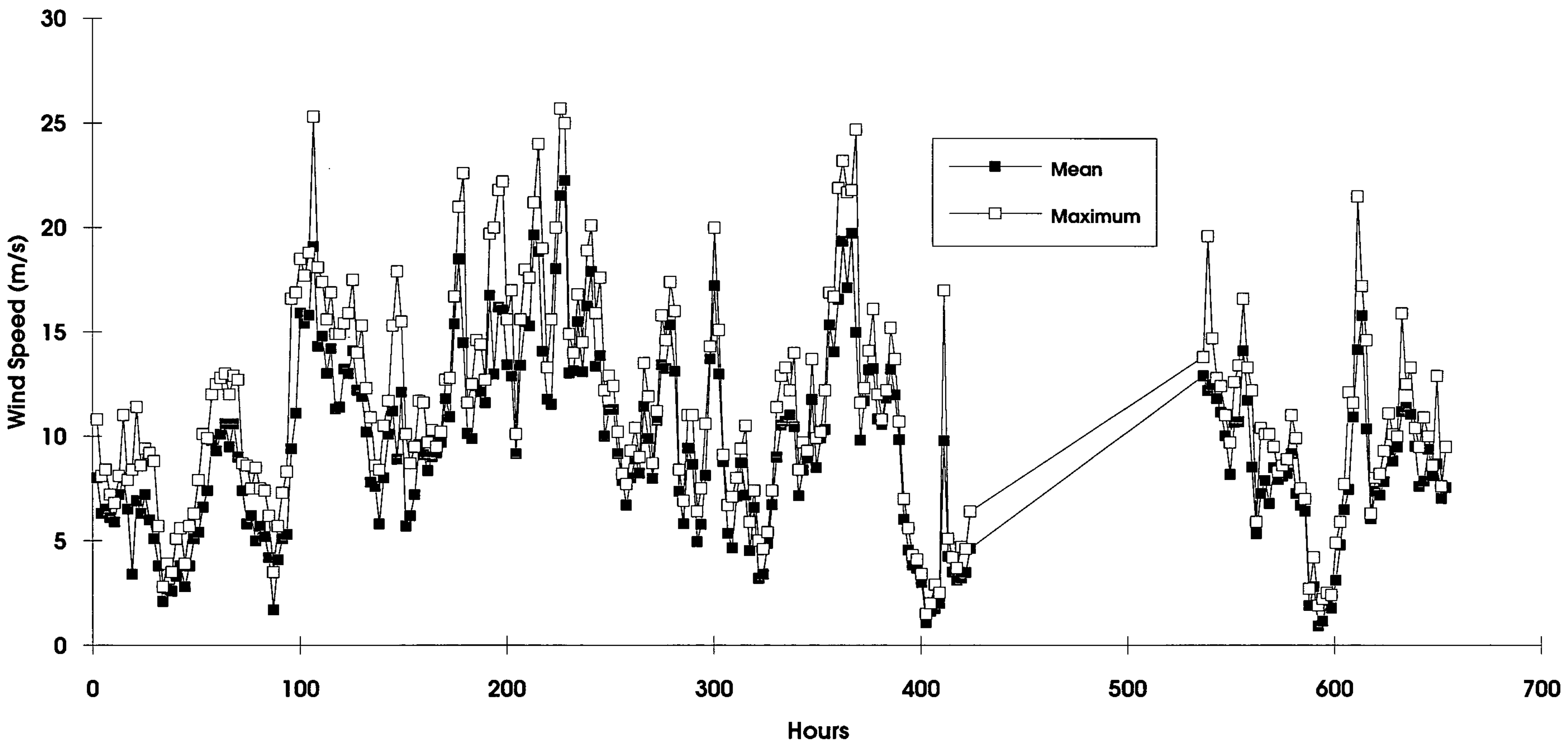


Figure 32: Linton - Effect of wind speed on the oxygen contents at the side of the silo

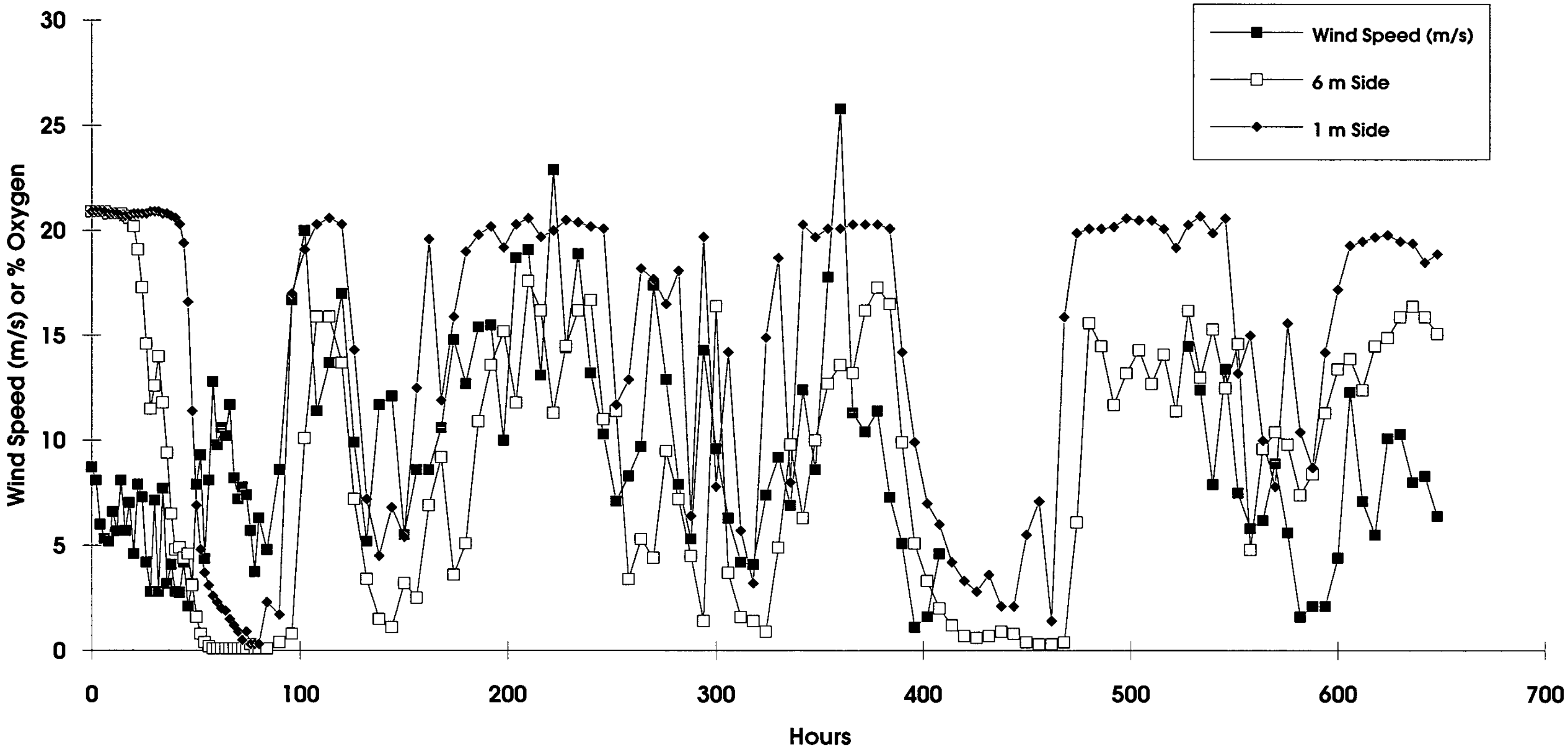


Figure 33: Linton - Effect of wind speed on the oxygen content in the centre of the silo

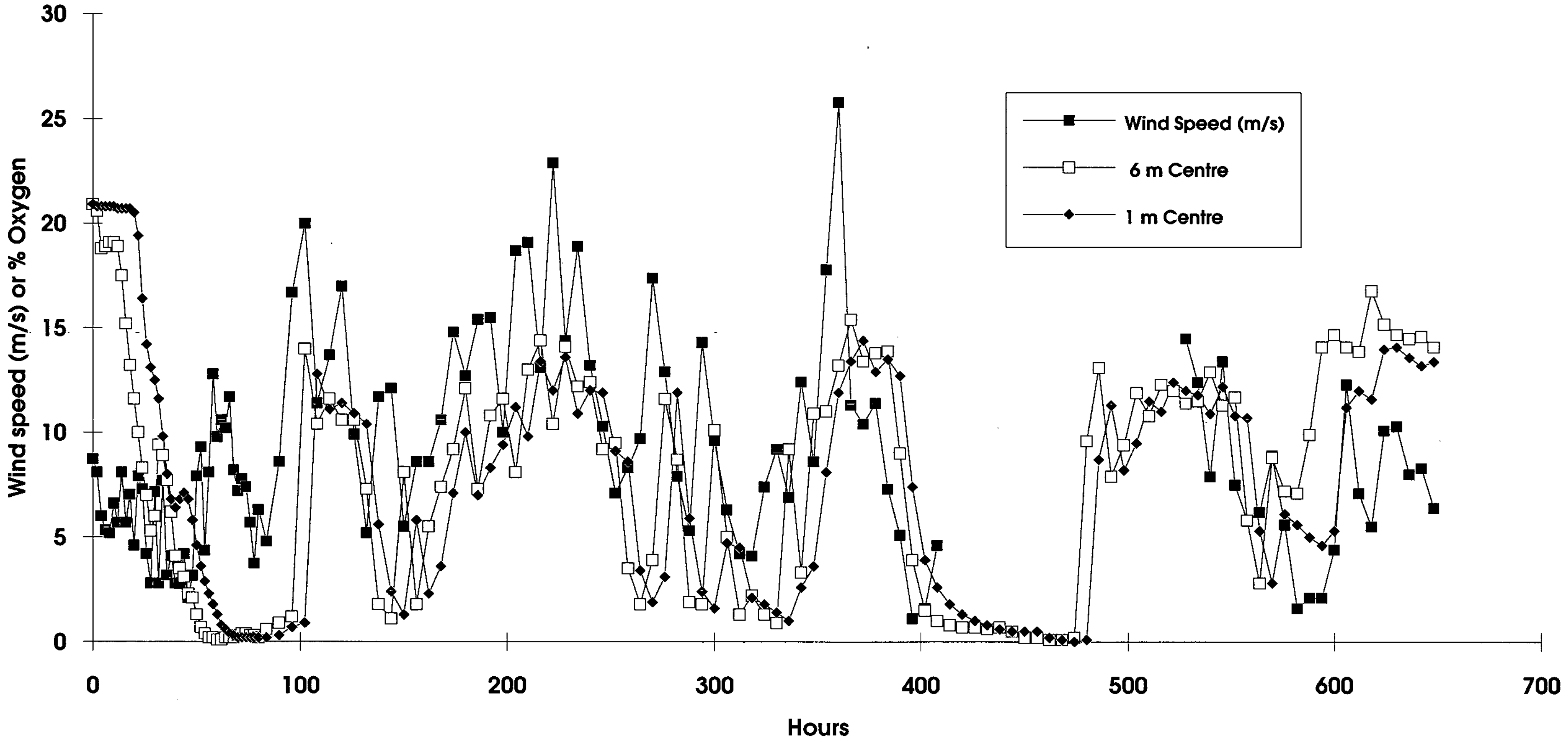


Figure 34: Linton - Effect of ambient temperature on the oxygen content in the centre of the silo

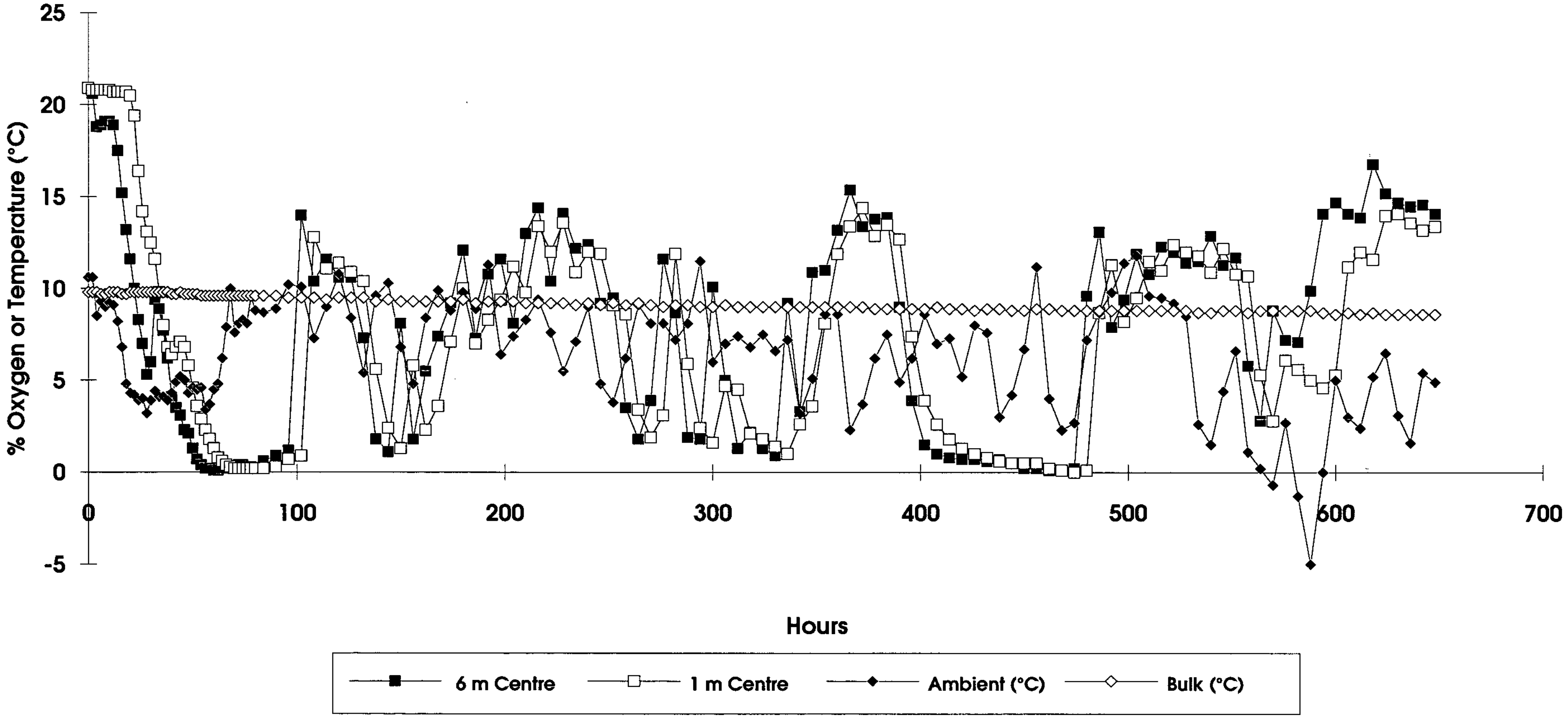


Figure 35: Linton - Effect of ambient temperature on the oxygen content at the side of the silo

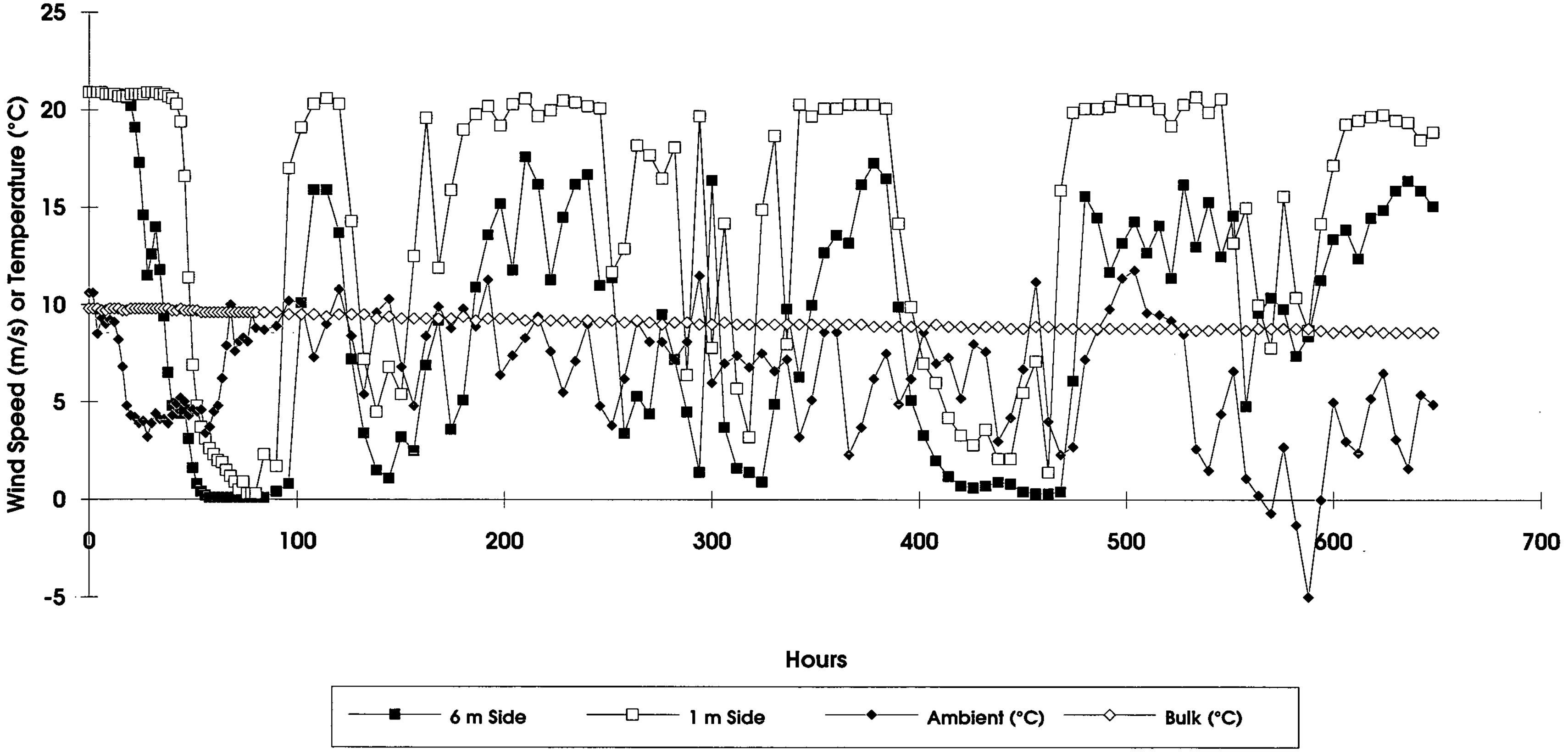


Figure 36: Linton - Change in the silo oxygen levels after the flow was stopped

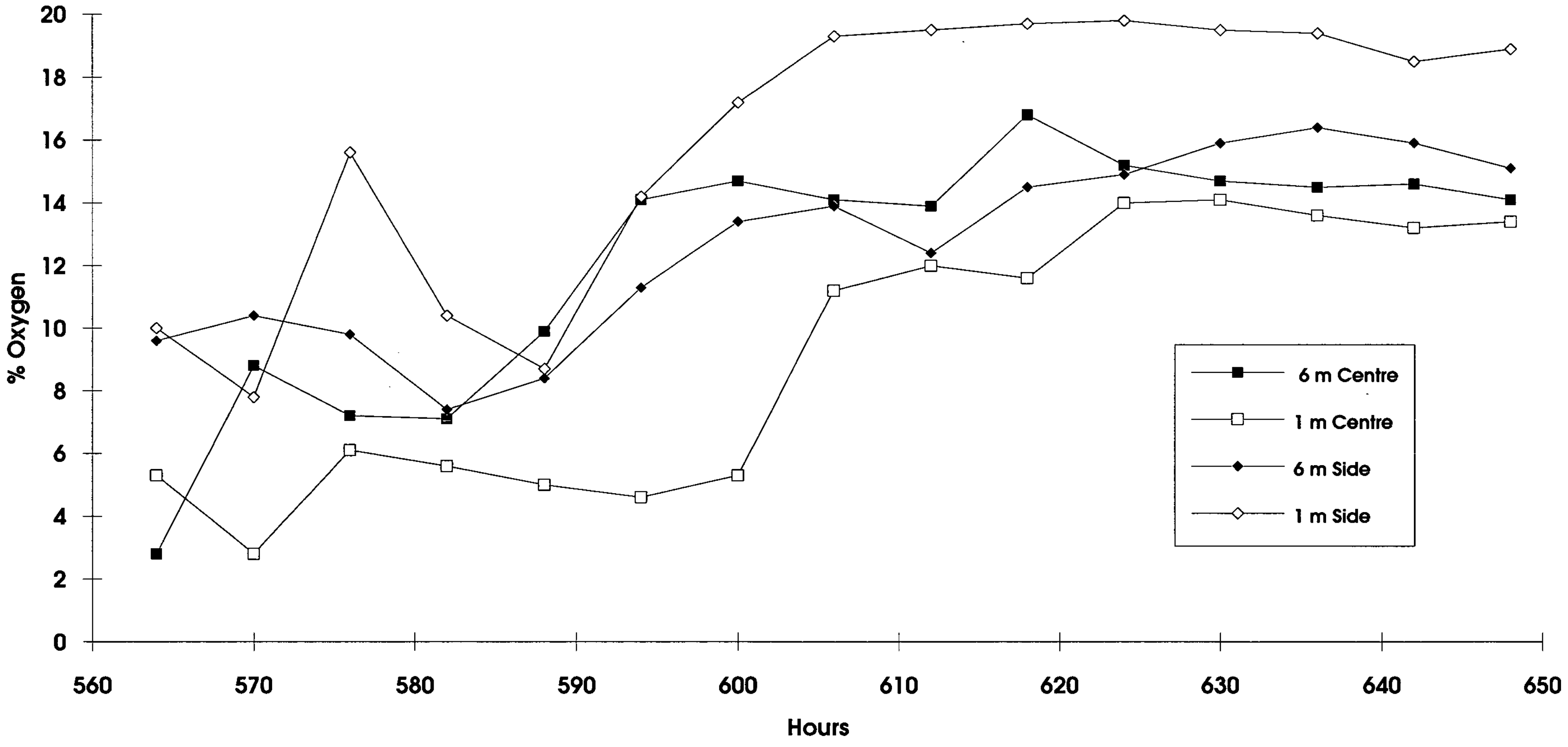


Figure 37: Linton - Environmental conditions after the flow was stopped

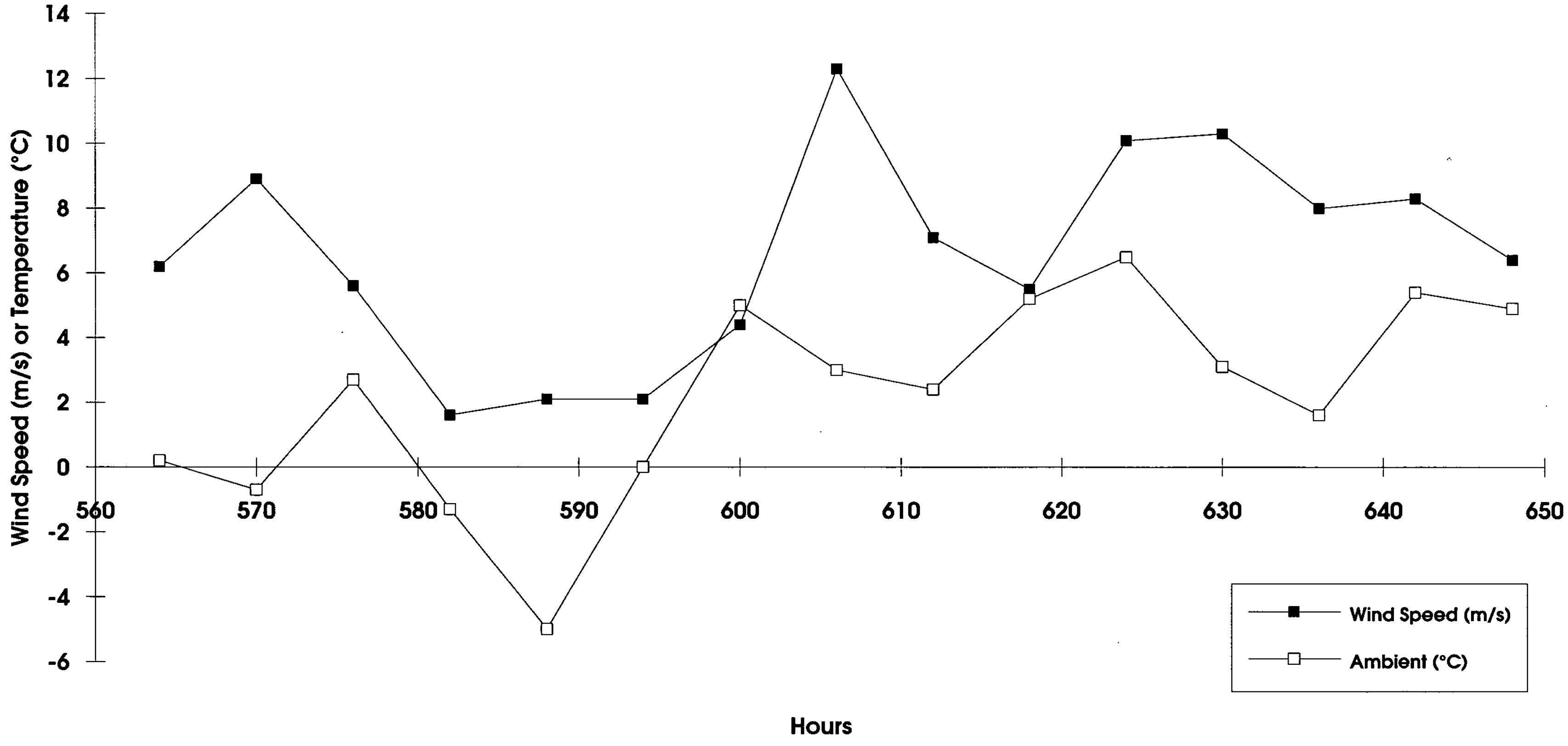


Figure 38: Relationship between silo capacity and flow rate

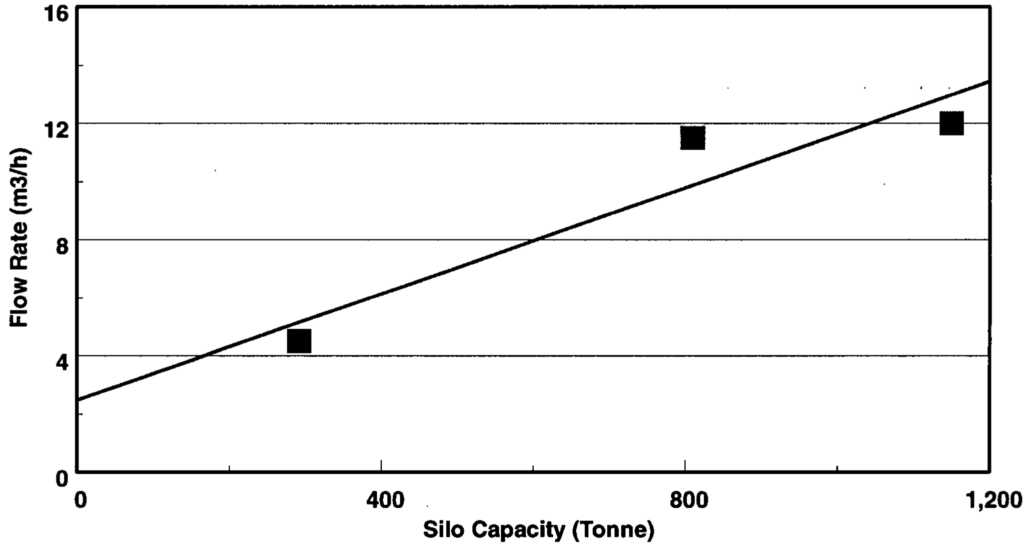


Figure 39: Relationship between silo capacity and flow rate with maintenance rates from other published results

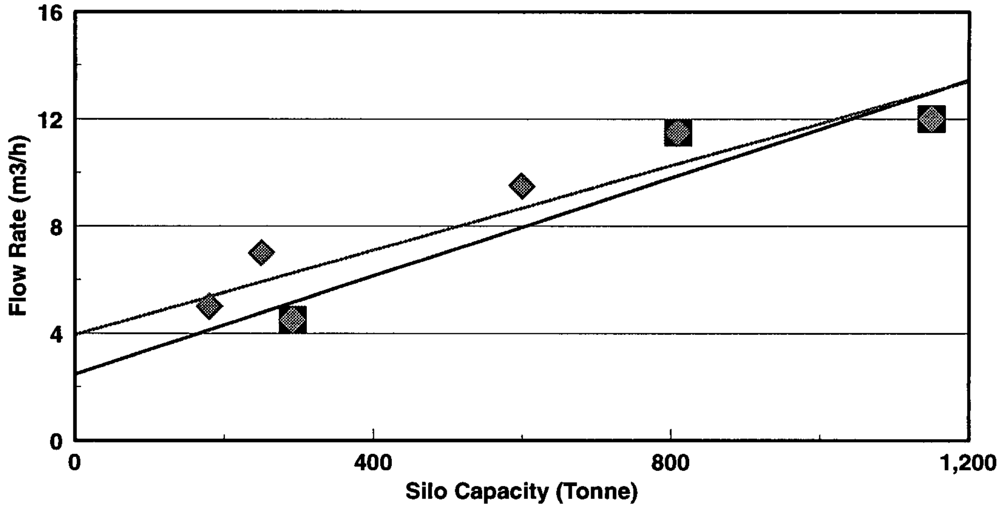


Figure 40: The effect on the mortality of adult *Cryptolestes ferrugineus* of interrupting 1% oxygen exposures with 16 hour periods of elevated oxygen levels

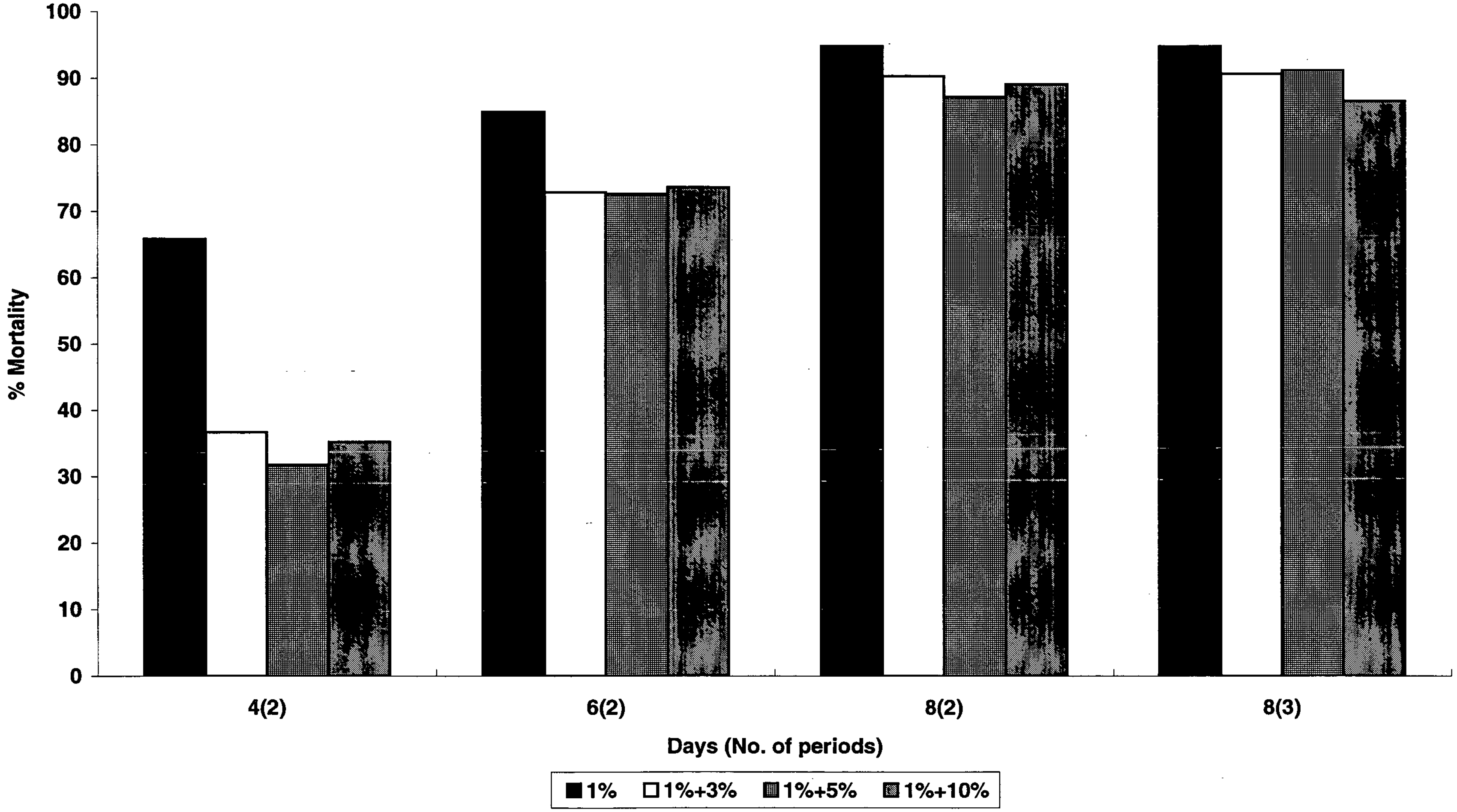


Figure 41: The effect on the mortality of 2 strains of adult *Sitophilus granarius* of interrupting 1% oxygen exposures with 16 hour periods of elevated oxygen levels

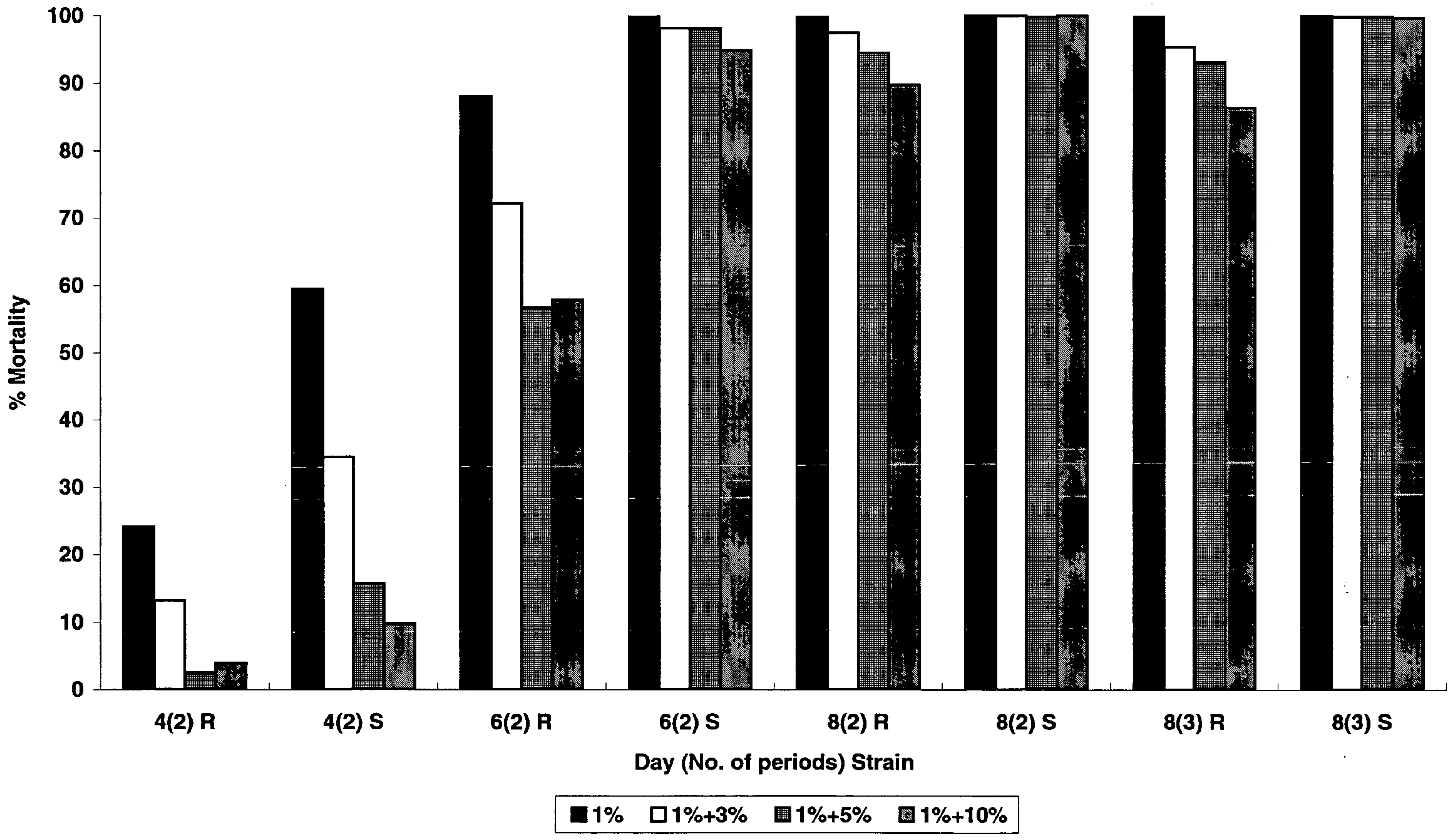
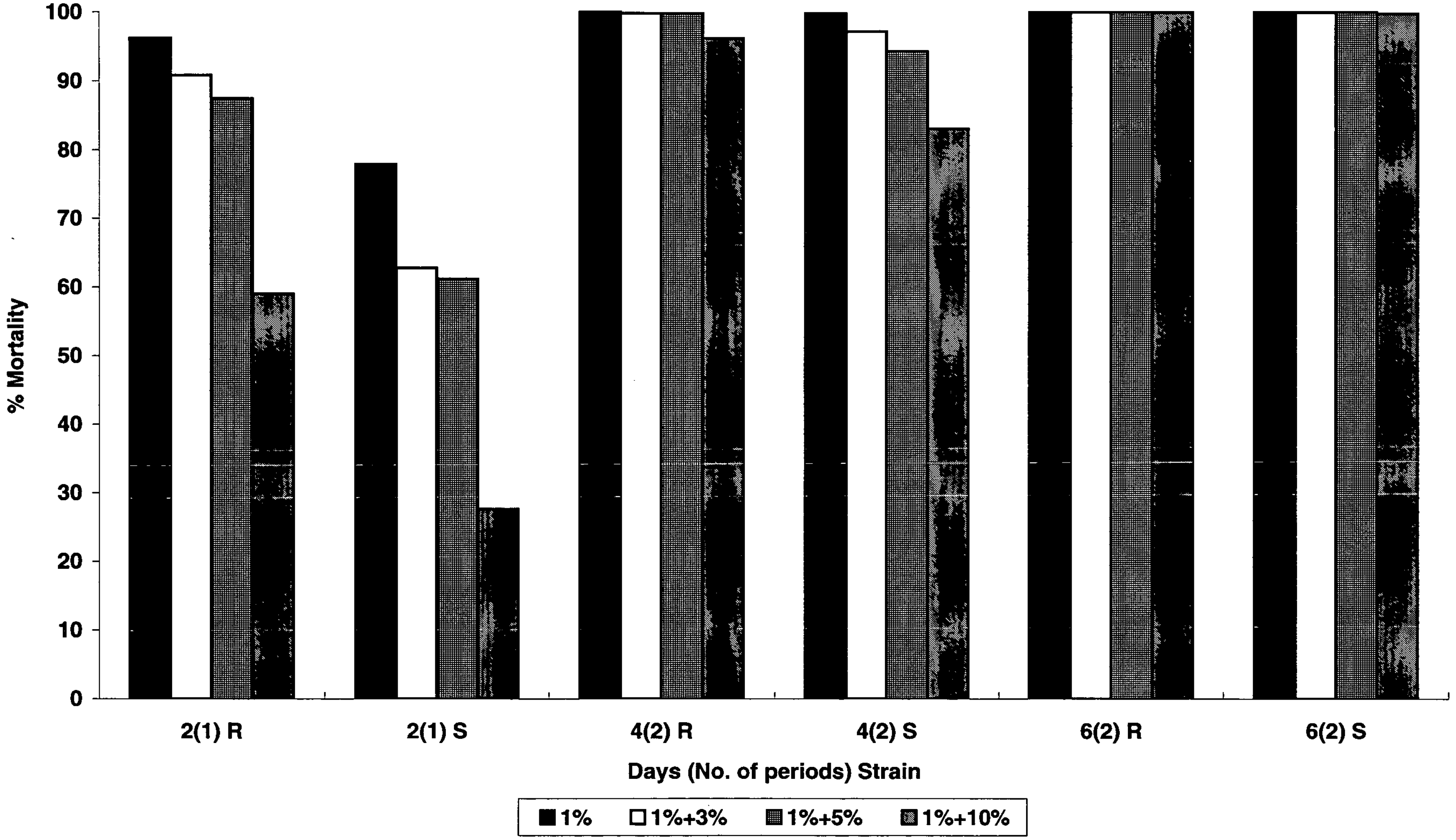


Figure 42: The effect on the mortality of 2 strains of adult *Oryzaephilus surinamensis* of interrupting 1% oxygen exposures with 16 hour periods of elevated oxygen levels



APPENDIX

Strains of species used for the juvenile stages exposure tests

C. ferrugineus	Lab	Laboratory standard
	Res	Strain selected for Phosphine resistance
O. surinamensis	Lab	Laboratory standard
	MR	Malathion-resistant
	Palm	Phosphine-resistant strain from Palmital, Brazil
R. dominica	Lab	Laboratory standard
	915R	Malathion-resistant
	BR2	Phosphine-resistant
S. granarius	Lab	Laboratory standard
	Gain	Malathion-resistant
	9104	Strain collected at Langwathby with Phosphine tolerance
	Fors	Strain collected at Forstal in the Information Gathering Exercise
	Pres	Strain collected at Preston in the Information Gathering Exercise
S. oryzae	Lab	Laboratory standard
	RA76	Malathion-resistant
	1102	Methyl Bromide-resistant
T. castaneum	Lab	Laboratory standard strain
	CTC	Malathion-resistant
	BT1	Phosphine-resistant from Bangladesh