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THE REDUCTION OF CHEMICAL AND MICROBIAL CONTAMINANTS IN WHEAT

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by

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AND D. A. L. SEILER

Final report of a 1 year and 6 months project at the Flour Milling and Baking Research Association, Chorleywood, Hertfordshire, WD3 5SH. The work commenced in January 1990 and was supported by a grant of £45,439 from the Home-Grown Cereals Authority (Project No. 0023/2/89).

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THE REDUCTION OF CHEMICAL AND MICROBIAL CONTAMINANTS IN WHEAT

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ABSTRACT

There is an increasing need for millers to seek ways to ensure that their products are as free as possible from chemical and microbial contaminants. However, the wheat which the millers purchase may contain pesticide residues and microbes which then pass to the flour and other mill products. Currently, although a number of cleaning machines are used in a typical mill screenroom to remove impurities from each batch of wheat milled, only a superficial attempt at cleaning the kernels themselves is made. Therefore, a study was undertaken to investigate alternative procedures for cleaning wheat prior to milling with the aim of lowering or eliminating pesticide residues in bran and microbes in white flour.

On the basis that pesticide residues and microbes in wheat are known to be concentrated in the branny layers of the kernel, wheat samples were subjected to frictional cleaning in a Westrup Laboratory Scarifier type LA-H to remove part of the bran coat. Scouring regimes were chosen on the grounds of practical application in a commercial mill. Thus, only 3-6% of the bran coat was removed and no attempt was made to study wet scouring.

For the pesticide study, wheat samples were treated at the same application rate of 4mg/kg with two formulations (2% dust and 25% emulsifiable concentrate) of pirimiphos-methyl and stored for 1, 3 and 6 months prior to scouring and laboratory milling. Pesticide residue analysis of the mill stocks showed that greater reductions were achieved when the pesticide had been applied as a dust formulation than an emulsifiable concentrate and that with longer storage times it became increasingly difficult to remove the pesticide when applied in the form of an emulsifiable concentrate. However, the scouring regimes employed resulted in only slight overall reductions (17-28%) over and above conventional cleaning.

Wheat samples with naturally occuring microbial loads were subjected to scouring and milling and the mill stocks tested for total viable count, moulds and yeasts and coliform bacteria. Neither scouring regime employed was particularly effective in reducing the microbial load in flour - tenfold reduction being the best result obtained.

Overall, it must be concluded that scouring wheat prior to milling is unlikely to result in sufficiently substantial reductions in either pesticide residue levels or microbial loads to justify the inclusion of such equipment in commercial mills.

INTRODUCTION

The growing significance of food safety issues in recent years has been demonstrated by increased public awareness, greater media coverage and ultimately, the introduction of tighter legislation under the Food Safety Act 1990. This places a greater onus on millers to ensure that their products are as free as possible from chemical and microbial contaminants. However, pesticide residues and microbes if present in flour and other mill products arise from the wheat which the millers purchase and so there is a need to investigate means of reducing or eliminating them from wheat before further processing. This would ideally be achieved in the mill screenroom.

SCREENROOM OPERATION

Wheat arrives at flour mills within a range of moisture contents containing an amount of extraneous matter. It therefore requires cleaning and conditioning (adjustment of moisture content to achieve a uniform degree of mellowness) prior to milling. Both of these operations are carried out in the mill screenroom where a number of specialised machines are employed. A typical screenroom flow would comprise:

A magnet	-	to remove any metal
An intake separator	-	to sift out large and small impurities such as seeds and chaff
A dry stoner	-	to separate out any stones
An aspirator	-	to lift off light impurities
A disc separator	-	to sort according to shape and thereby remove other cereals and seeds.
A damping system	-	for conditioning purposes

In addition the screenroom might employ:

A frictional cleaning machine	-	for removal of any loose skins and adhering dirt from the wheat kernels.

Therefore, a typical screenroom operates by systematically removing impurities first from the wheat sample and finally from the wheat grain itself. It is important to note that only a superficial attempt at cleaning the kernels themselves is made, in contrast with the pearling machines used in the rice milling industry which aim to remove the whole of the bran layer when producing white rice.

LABORATORY CLEANING AND MILLING

Prior to conditioning wheat is cleaned of large and small impurities and dust on a laboratory version of an intake separator. For these experiments the machine used was a Carter Day Dockage Tester. Laboratory milling was performed on a Buhler laboratory mill MLU 202 which mimics the break and reduction systems of commercial mills. Break and reduction flours are blended into a 'straight-run' flour. Bran and offal are mill by-products from the break and reduction systems respectively. The bran and offal may be passed through an impact finisher to produce flour known as finisher flour.

WHEAT SCOURING

A wheat scourer is one example of a frictional cleaning machine. It operates by circulating wheat grains inside an abrasive mesh chamber by means of beaters or brushes - the adjustment of which alter the degree of abrasion. A diagrammatic representation of a horizontal wheat scourer is shown in Figure 1.

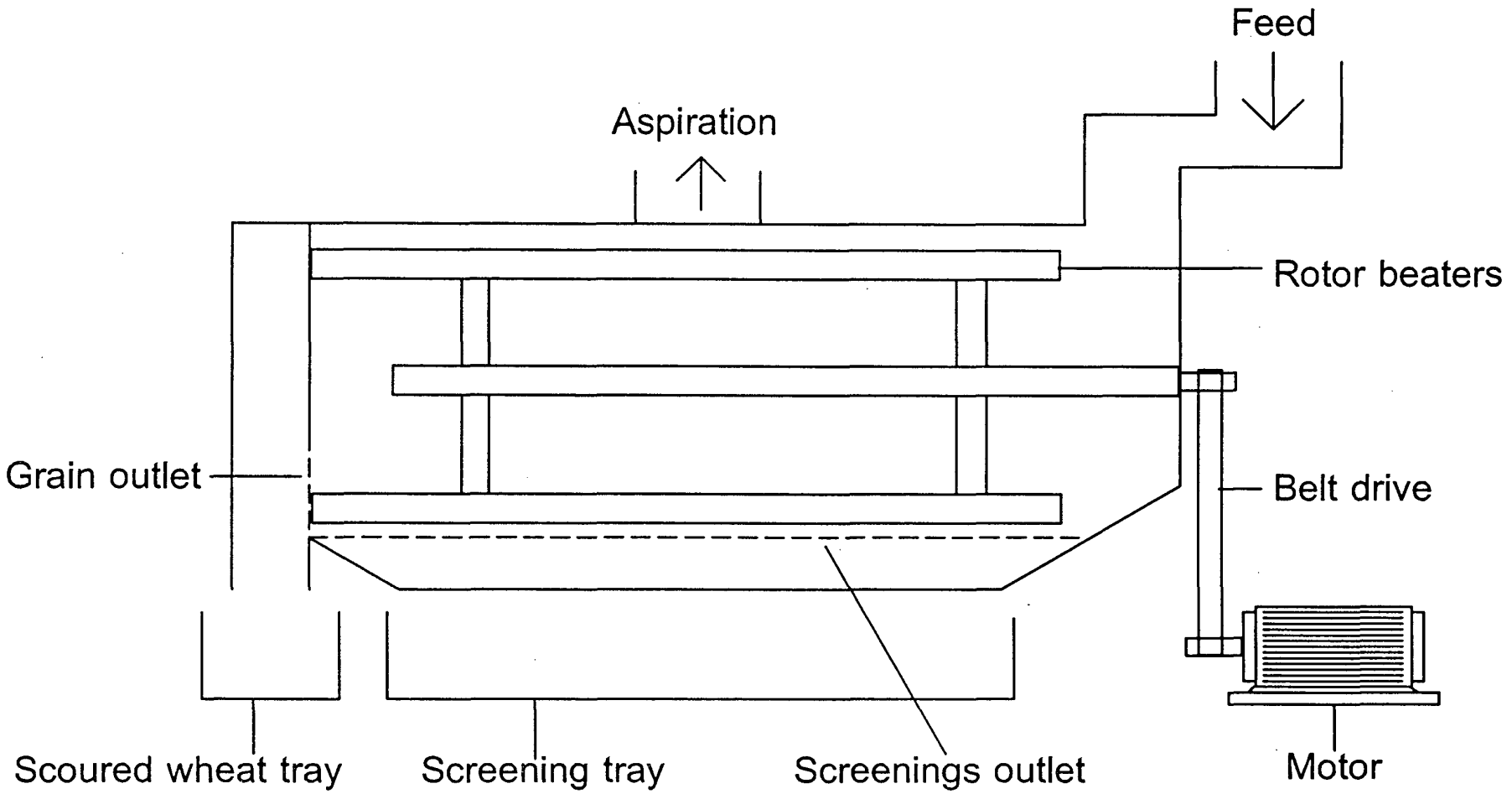
The equipment chosen for this study was a Westrup Laboratory Scarifier type LA-H a commercial-scale version of which is available as an HA polishing/brushing machine.

Traditionally wheat scourers have been employed to clean loosely adhering dirt and skin from wheat grains. However, their action is such that it provides the opportunity actively to remove part of the wheat bran as well. This action may be employed to reduce or eliminate contaminants which reside largely on the surface of the grain. For example, Bruggemann and Ocker (1991) reported an 80% reduction in lead contamination of wheat by scouring 5-10% of the outer layers of the kernel. The following investigations evaluate the potential benefit to the miller and ultimately the consumer of such a procedure.

REFERENCE

Bruggemann, J. and Ocker, H. D. (1991). Desired and undesired trace elements in cereals and cereal products. ICC 1991 Symposium, June 10-13, Prague, Czechoslovakia.

Figure 1. Diagrammatic representation of a horizontal wheat scourer.



THE REDUCTION OF PESTICIDE RESIDUE LEVELS IN BRAN INTENDED FOR HUMAN CONSUMPTION

OBJECTIVE

To investigate the possibility of reducing pesticide residues in wheat and hence bran intended for human consumption, by a scouring operation prior to milling.

INTRODUCTION

Recently, much attention has been focused on the presence of pesticide residues in cereal foods at the time of consumption and, in particular, on the possibility of elevated residue levels in bran and wholemeal products aimed at health conscious consumers who are following high-fibre diet recommendations (Anon., 1990).

A recent report of the MAFF Working Party on Pesticide Residues in Food (Anon., 1989) drew attention to the need for further research aimed at reducing residues in wheat products, especially bran. However, although application rates are being reduced by adoption of integrated pest management (Osborne, 1991), it is *unrealistic to expect that the need to use pesticides on grain in the UK could be* totally eliminated in the foreseeable future. HGCA Research Review No.12 (Osborne et al, 1988) noted that about 20% of wheat purchased by UK millers contained detectable pesticide residues, albeit well below the maximum residue level in the vast majority of cases, but that during conventional wheat cleaning and milling pesticides are concentrated in the bran and germ fractions of wheat at up to five times the levels found in the whole grain. It was therefore recommended that, although conventional dry cleaning of wheat prior to milling did not result in a reduction in pesticide levels, there was a need to determine whether alternative procedures, which could be incorporated into the screenroom processes, would be more successful.

Since detectable residues in wheat are largely the result of treatment with pesticides during storage (Osborne et al, 1988), a successful cleaning regime must achieve effective residue reductions from grain that has been treated with different formulations of such pesticides and has been stored for different lengths of time. Both these factors were taken into account during the design of the experiment and the choice of the most appropriate pesticide for this work. Pirimiphos-methyl - a fast acting, broad spectrum, organophosphorus insecticide - is the most common pesticide applied to stored grain in the UK at present. It is,

furthermore, a pesistent pesticide, in that its residues do not degrade rapidly during storage (Snelson, 1987). Residues of pirimiphos-methyl are, therefore, more likely to be present in home-grown wheat than those of any other pesticide and, as a consequence, pirimiphos-methyl was considered to be particularly relevant for this work.

Although pirimiphos-methyl residues in wheat grains remain almost entirely on the pericarp (Snelson, 1987), the grain cleaning process in use at a commercial mill did not substantially reduce the levels in the grain. This was because although the material removed as screenings contained residues up to nine times higher than levels in whole grain, the weight of screenings was too small to account for a significant proportion of the pesticide originally present in the grain (Osborne et al, 1988). Two possible approaches to greater removal of the pesticide would be to accelerate its chemical breakdown or to employ a more vigorous physical cleaning method which would remove a greater amount of material as screenings.

Since hydrolysis (decomposition involving water) represents the major breakdown route for pirimiphos-methyl into less toxic metabolites, the most effective cleaning regime for its removal from grain is likely to be wet scouring (washing and wizzing). However, this cleaning technique is no longer used in the UK and its re-introduction would not be practicable on account of the cost of the water utilised, problems associated with the disposal of effluent in accordance with The Rivers (Prevention of Pollution) Act 1961 and the danger of increased microbiological counts due to organisms multiplying in the water.

It was therefore necessary to devise a dry cleaning regime of more vigorous action than a standard piece of screenroom machinery. However, such a regime would be constrained by the commercial availability of the equipment and its suitability for inclusion in to a standard screenroom flow. Of the equipment available in both laboratory and commercial form a wheat scouring machine was considered to be the most appropriate for these purposes. This project therefore concentrates on the development of a suitable dry scouring regime and its efficacy towards the reduction of pesticide residues from bran. It also investigates the effect of pesticide formulation and storage period upon potential residue reductions.

MATERIALS AND METHODS

A quarter tonne batch of 'Mercia' home-grown milling wheat, stated to be pesticide free, was purchased and divided into four equal subsamples, three of which were delivered to ADAS, Slough Laboratory to act as the test samples and a control. The remaining sample was analysed for: protein content (ISO Standard Method 1871/1975), moisture content (ICC Standard No.110/1), Falling Number (ICC Standard No.107), electrophoresis (ICC Standard No.136) and organophosphorus pesticide residues (see p.11). After confirmation of the absence of pesticide residues, the sample was utilised to establish the cleaning regimes to be employed.

Pesticide applications

The organophosphorus pesticide pirimiphos-methyl (product name Actellic, Imperial Chemical Industries plc) was applied as an 25% emulsifiable concentrate (e/c) and a 2% dust formulation at the Slough laboratory. In each case a treatment rate of 4mg/kg was the target.

The e/c formulations were diluted in distilled water and sprayed onto 25kg batches of the grain using a De Vilbis handheld sprayer as the grain was tumbling in a halfbag size concrete mixer (Thomas and Clasper, 1983). After spraying, the grain was tumbled for a further 5 min to ensure even distribution. The dust formulations were weighed out and added to 25kg batches of the grain in the concrete mixer. The opening of the mixer was sealed with polythene and the machine was allowed to run for 5min. Two separate 25kg batches were combined for each treatment and, after blending, the samples were stored at 14.5% m.c. under shelter at ambient conditions in metal dustbins. Subsamples of the wheats were taken with a metal scoop immediately after treatment and also after 1, 3 and 6 months storage.

Wheat cleaning regimes

The initial experimentation to develop the exact cleaning regimes found that a removal of 10% of the wheat pericarp or greater was impractical. This was so for two reasons. Firstly, an undesirable amount of broken grains were produced at higher levels of bran removal and secondly it was considered that an uneconomic yield would result from the further removal of bran. The individual regimes were therefore optimised to minimise the production of broken grains.

Three wheat scouring regimes involving a Westrup laboratory scarifier Type LA-H were developed. In each case a 4kg wheat sample was fed to the scouring section of the scarifier, via the accompanying vibratory feeder (setting 5). 30 seconds were allowed to elapse before the grain outlet was opened to allow the scoured grains out. This procedure, coupled with the adjustment of the outlet to maintain an even flow through the machine, ensured that approximately 1kg of wheat was present in the scouring section during operation, which in turn, ensured a more consistent abrasive action. Scoured bran fragments were collected after use by dismantling the machine and brushing into the appropriate collecting tray. Scoured wheat samples passed out of the scouring chamber and collected in a separate tray.

The three scourer regimes differed only in their treatment of the wheat samples prior to abrasion. Samples were either untreated (dry), predamped (an addition of 1% water 5 min prior to scouring), or moistened (to 15.5% m.c. 20 min prior to scouring). Water addition was achieved by shaking the wheat samples with appropriate quantities in sealed plastic pots.

All scoured samples and a separate control were passed through a CarterDay Dockage Tester prior to conditioning for milling. Samples of the screened material taken from the Tester were combined with the relevant bran fragments from the scourer prior to analysis for pesticide residues.

Laboratory milling

Scoured and cleaned wheat samples were conditioned to 15.5% moisture content prior to milling by shaking for 5 min with the required quantity of water in a sealed plastic container. The moisture was added in the late afternoon the day before milling so that samples received a lying time of between 16 and 24 hours. A capacitance moisture meter (Sinar Agritec, Weybridge, Surrey) was used to determine the moisture content of the samples before and after conditioning.

4 kg samples of conditioned wheat were milled into flour, bran and offal fractions in a Buhler MLU 202 laboratory mill which was clothed and had the rolls set according to EC Regulation 1628/77. A Buhler laboratory impact finisher MLU 302 was used to re-treat the bran and offals twice (Robinson and Stewart, 1980). The flour from finishing the bran and offal was blended with the straight run flour in a KEK-Gardner 8LD C/2 double cone mixer and passed through a Russell-Finex 14550 redresser fitted with a 300μm screen. Milling was carried out under

controlled temperature at 20°C and relative humidity of 65% (Hook et al, 1984). Extraction rate was calculated on a total products basis, with the requirement that 98.5% of the feed was recovered from the mill. In order to guard against the time of day effect associated with laboratory milling (Hook et al, 1982) and the possibility of further degradation of the pesticide between milling and analysis, the milling of the wheats from each regime was replicated and the replicates were milled in a random order.

Pesticide residue analysis

20g samples of wheat and flour, and 10g samples of bran and offal were taken and ground if necessary in a Glen-Creston laboratory mill prior to analysis. Samples were then transferred to maceration flasks for extraction with acetone-methanol (1:1). After blending twice for 2 min in the flasks, the resultant supernatants were decanted off and centrifuged for 5 min at 2500 rpm. The supernatants were then transferred to 1l separating funnels for partitioning with dichloromethane as described by Bottomley and Baker (1984). The extracts produced were rotary evaporated to dryness and recovered by rinsing into 10ml graduated stoppered tubes with cyclohexane-ethyl acetate (1:1). The extracts were then made up to 10ml and transferred to foil-topped glass vials. Clean-up was carried out by gel permeation chromatography (GPC) as devised by Chamberlain (1990). An automated system consisting of a Gilson 232/401 automatic sample processor and injector, a Gilson 305 piston pump and a column, 450 mm x 25 mm i.d., packed with Bio-Beads SX3 was used. The resulting extracts were rotary evaporated to dryness, and recovered by rinsing into 5ml graduated stoppered tubes with hexane and made up to 5ml for analysis.

The solvents used were all HPLC grade, except for the hexane which was pesticide grade, and dichloromethane which was AnalaR.

Quantitative determinations were made by gas chromatography (GC) using a flame photometric detector (FPD) and a glass column, 5 ft x 3 mm i.d., packed with 5% OV17 on Gas Chrom Q (80-100 mesh). The GC operated at oven temperature of 215°C, injection temperature of 220°C and detection temperature of 250°C.

Analyses were randomised and carried out in blind duplicate with a series of pesticide standards (1-10 ng/μl).

RESULTS

Wheat analysis

The laboratory analyses of the original wheat sample and the residues resulting from the pesticide applications are shown in Tables 1 and 2 respectively.

Cleaning procedures

The percentage of screenings produced by the scourer and dockage tester are shown in Table 3. The levels of pesticide residue associated with these fractions are shown in Tables 4-6.

Laboratory milling

The distribution of the wheat stocks upon laboratory milling is shown in Tables 7-9. The level of pesticide residue associated with each fraction is, in turn, indicated in Tables 10-12.

Pesticide analysis

Pesticide recoveries from the clean-up column were in the range 94-104%, whilst those recovered by the GC after spiking the sample at the beginning of the extraction with acetone-methanol, were within the range 86-89%.

DISCUSSION

Wheat cleaning

The amount of pericarp removed by the scourer during cleaning (between 3-6% when expressed as a percentage of the bran received upon milling) was similar for each of the three regimes, irrespective of the mode of pesticide application and the length of storage period (Table 3). A similar picture emerged for the removal of screenings through the dockage tester, although use of the scourer under any regime created more screenings than the dockage tester alone - primarily because of the increased production of broken grains that separated out in the screenings. Predamping or moistening the wheat prior to scouring was

found to have little effect on the removal of pericarp, or the production of screenings and hence broken grains.

The pesticide residue levels detected in the screenings from the scourers regimes and the dockage tester control samples did not differ greatly (Tables 4-7). A statistical analysis of the data demonstrated no significant difference between the dry scouring regime and conventional cleaning after 3 months storage. It would appear therefore, that the scourer regimes had achieved very little in terms of residue removal. However, it should be remembered that the scourer regimes removed more screenings than the dockage tester alone, and therefore, direct comparisons of residue levels are inappropriate. To clarify the situation two graphical representations have been made (Figs 1 & 2). These express the residue levels associated with the screenings, as a percentage of the total pesticide residue recovered for each cleaning regime. (This was achieved by first multipying the residue level detected by the weight of fraction produced and then calculating the percentages as appropriate). There are two points to note from these Figures: firstly, that similar results emerge for the two modes of pesticide application, and secondly, that different results emerge for the individual cleaning regimes. It would appear therefore, that each regime removes subtly different wheat screenings and that furthermore, this removal is not affected by the mode of pesticide application.

Distribution of pesticide upon laboratory milling

The laboratory milling procedure adopted resulted in the production of three mill stocks: flour, bran and offal. These stocks essentially comprise: the wheat endosperm, large pieces of the pericarp and the remainder of the pericarp together with the wheat embryo respectively. Their distribution upon milling was found to be not greatly affected by the pesticide formulation, the length of storage period or the cleaning regime employed (Tables 7-9). In fact, the small differences that did occur can probably be attributed to the conditioning of the wheat samples prior to milling. There were, however, large differences in the residue levels associated with the various mill stocks.

The distribution of pesticide on laboratory milling is known not to be uniform as most of the residue associates with the bran and offal fractions and only a small portion separates out with the flour (Demarchelier et al, 1980; Cerna et al, 1978; Bengston et al, 1980). This is primarily because there is only a limited redistribution of pesticide within the grain during storage (Rowlands 1967; 1971;

1976). A similar overall picture was produced in this study (Tables 10-12). Furthermore, the overall decline in residue levels during storage, for instance compare Tables 10 and 12, was similar to that previously reported (Bengston et al, 1980; Mensah et al, 1979). It should be noted however, that this apparent decline in residue levels may actually represent a decline in the proportion of 'free' residue as it is thought that an increasing amount becomes inaccessible to extraction or 'bound' (Sampson, 1986; Leahy and Curl, 1982; Rowlands, 1981).

The efficacy of the individual scourer cleaning regimes in reducing residue levels from bran is shown in Figs 3 and 4. Three points arise from these Figures. Firstly, that the individual scourer regimes were found to be similarly effective for a particular mode of pesticide application; secondly, as might be expected, the scourer regimes removed residues more effectively if the pesticide had been applied as a dust formulation; and thirdly, that it became more difficult to remove e/c residues after prolonged storage, presumably because of the tendency for residues to become 'bound'. These points were confirmed by a statistical analysis of the data which found: that the only significant reductions occurred between the three scourer regimes and conventional cleaning; that these differences occurred at the 0.1% level except for the the e/c formulation after 3 months storage which was not significantly different; and that the final residue levels of each regime were higher for the e/c treatment than the dust formulation after 6 months storage (5% level). This last point is illustrated by referring to Figs 5 and 6 which show an increase in the percentage of recovered pesticide associating with the bran fractions during storage, particularly for the e/c treatment. However, from Figs 3, 4, 5 and 6 it is apparent that the actual levels of residue reduction brought about by scouring are small compared to that achieved by conventional cleaning alone. For instance, a comparison of the mean data from Tables 10-12 shows that reductions of 17-28% of the dust formulation and 0-26% of the e/c treatment (compared to the control sample) are all that are achieved.

CONCLUSIONS

1. Greater reductions in residue levels were observed when the pesticide had been applied as a dust formulation than when in the form of an emulsifiable concentrate.

2. With longer storage times it became increasingly difficult to remove the pesticide when applied in the form of an emulsifiable concentrate.

3. The three scourer regimes employed achieved only slight overall bran residue reductions compared to conventional cleaning.

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Table 1: Characteristics of original wheat sample.

Moisture content	14.3%
Protein content	11.6%
Hagberg Falling Number	351 sec
Varietal purity by electrophoresis (14 grain)	14 Mercia
Pesticide residue (organophosphorus)	none detected. (i.e. < 0.1mg/kg)

Table 2: Treatment level of pesticide applications.

Treatment regime	Residue detected (mg/kg)
pirimiphos-methyl ec	3.45
pirimiphos-methyl dust	3.35

Table 3: Percentage of dirty wheat removed as screenings for each cleaning regime.

Cleaning regime		Scourer			Dockage Tester		
		i	*ii*	*iii*	*i*	*ii*	*iii*
Dockage Tester only:	1mo.		n/a		0.6	0.6	0.6
	3mo.		n/a			0.7	0.7
	6mo.		n/a			0.7	0.7
Scarifier + 1% m.c:	1mo.	0.6	0.6	0.6	1.2	1.1	1.6
(5min)	3mo.		0.6	0.7		1.8	2.3
	6mo.		0.8	0.7		2.1	1.4
Scarifier + 1% m.c:	1mo.	0.5	0.7	0.9	1.8	1.7	2.2
(20min)	3mo.		0.8	0.9		2.7	2.9
	6mo.		0.8	0.7		2.5	1.9
Scarifier dry:	1mo.	0.6	0.7	0.9	1.6	1.7	2.0
	3mo.		0.7	0.7		2.4	2.5
	6mo.		0.8	0.9		2.4	3.3

i = Control untreated wheat
ii = pirimiphosmethyl e/c
iii = pirimiphosmethyl dust

mo. = month
n/a = not applicable

For each of the following tables a coding system applies:

A: CarterDay dockage cleaned wheat only.
B: Scoured wheat, predamped with 1% m.c. 5 min. prior to cleaning.
C: Scoured wheat, moistened to 15.5% m.c., 20min. prior to cleaning.
D: Scoured wheat, no conditioning.

Table 4: Effect of cleaning regimes upon pesticide residues associated with wheat after one month of storage.

Cleaning Regime / sample.	Residue of pirimiphosmethyl (mg/kg)	
	e/c treatment	dust treatment
Dirty wheat	2.5	3.0
A: Cleaned wheat	2.8	2.4
screenings	20.3	16.9
B: Cleaned wheat	2.5	2.5
screenings	23.0	23.4
C: Cleaned wheat	2.5	2.5
screenings	21.0	21.0
D: Cleaned wheat	2.4	2.3
screenings	23.3	21.0

NB: All results are expressed as the mean of two replicates.

Table 5: Effect of cleaning regimes upon pesticide residue levels associated with wheat after three months of storage.

Cleaning Regime / sample	Residue of pirimiphosmethyl (mg/kg)	
	e/c treatment	dust treatment
Dirty wheat	3.0	2.8
A: Cleaned wheat	2.2	2.4
screenings	17.1	18.4
B: Cleaned wheat	2.7	2.2
screenings	14.5	10.8
C: Cleaned wheat	2.4	2.8
screenings	13.7	11.6
D: Cleaned wheat	2.4	2.3
screenings	15.8	15.1

NB: All results are expressed as the mean of two replicates.

Table 6: Removal of pesticide residue during cleaning operation after six months of storage.

Cleaning Regime / sample	Residue of pirimiphosmethyl (mg/kg)	
	e/c treatment	dust treatment
Dirty wheat	2.0	1.7
A: Cleaned wheat	2.4	1.8
screenings	7.3	7.8
B: Cleaned wheat	2.4	1.5
screenings	10.1	12.6
C: Cleaned wheat	1.6	1.9
screenings	9.2	11.0
D: Cleaned wheat	1.6	1.7
screenings	11.9	9.7

NB: All results are expressed as the mean of two replicates.

Table 7: Distribution of wheat stocks upon laboratory milling after one month of storage.

Cleaning Regime	% of stock separating as Flour, Bran, Offal.								
	Control			pirimiphos e/c			pirimiphos dust		
	F	B	O	F	B	O	F	B	O
A:	74.3	16.4	9.3	74.1	16.1	9.8	74.1	16.6	9.3
B:	74.7	16.5	8.8	73.8	15.5	10.7	74.7	15.4	9.9
C:	74.3	15.9	9.8	74.4	15.9	9.7	74.5	15.7	9.8
D:	74.6	16.1	9.3	75.0	14.6	10.4	75.0	15.4	9.6

Table 8: Distribution of wheat stocks upon laboratory milling after three months of storage.

Cleaning Regime	% of stock separating as Flour, Bran, Offal.					
	pirimiphos e/c			pirimiphos dust		
	F	B	O	F	B	O
A:	75.4	16.8	7.8	75.9	16.9	7.2
B:	75.8	16.2	8.0	76.1	15.5	8.4
C:	76.3	16.1	7.6	77.1	15.8	7.1
D:	75.9	15.5	8.6	76.2	16.3	7.5

Table 9: Distribution of wheat stocks upon laboratory milling after six months of storage.

Cleaning Regime	% of stock separating as Flour, Bran, Offal.					
	pirimiphos e/c			pirimiphos dust		
	F	B	O	F	B	O
A:	74.8	18.6	6.6	74.8	18.8	6.4
B:	75.0	18.0	7.0	75.2	17.1	7.7
C:	75.1	19.0	5.9	75.5	18.0	6.5
D:	75.6	18.1	6.3	75.8	17.4	6.8

Table 10: Distribution of pesticide residue in the milled fractions after one month of storage.

Cleaning Regime		Residue of pirimiphos-methyl (mg/kg)	
		e/c treatment	dust treatment
A:	flour	1.63	0.91
	bran	12.90	12.89
	offal	10.40	10.78
B:	flour	0.63	0.76
	bran	9.58	9.30
	offal	9.65	9.82
C:	flour	2.08	0.73
	bran	9.66	9.92
	offal	10.51	8.65
D:	flour	1.46	0.76
	bran	9.50	10.72
	offal	9.56	12.40

Table 11: Distribution of pesticide residue in the milled fractions after three months of storage.

Cleaning Regime		Residue of pirimiphosmethyl (mg/kg)	
		e/c treatment	dust treatment
A:	flour	0.79	0.91
	bran	9.23	11.54
	offal	8.42	9.96
B:	flour	0.81	0.81
	bran	10.07	9.30
	offal	8.00	9.85
C:	flour	0.73	0.79
	bran	8.51	8.47
	offal	9.01	10.29
D:	flour	0.73	0.91
	bran	8.84	9.10
	offal	9.51	9.56

Table 12: Distribution of pesticide residues in the milled fractions after six months of storage.

Cleaning Regime		Residue of pirimiphosmethyl (mg/kg)	
		e/c treatment	dust treatment
A:	flour	0.69	0.64
	bran	7.80	7.88
	offal	9.38	8.79
B:	flour	0.61	0.62
	bran	7.16	5.95
	offal	7.96	8.86
C:	flour	0.63	0.64
	bran	7.25	6.02
	offal	7.37	7.93
D:	flour	0.62	0.66
	bran	6.74	6.52
	offal	7.84	8.63

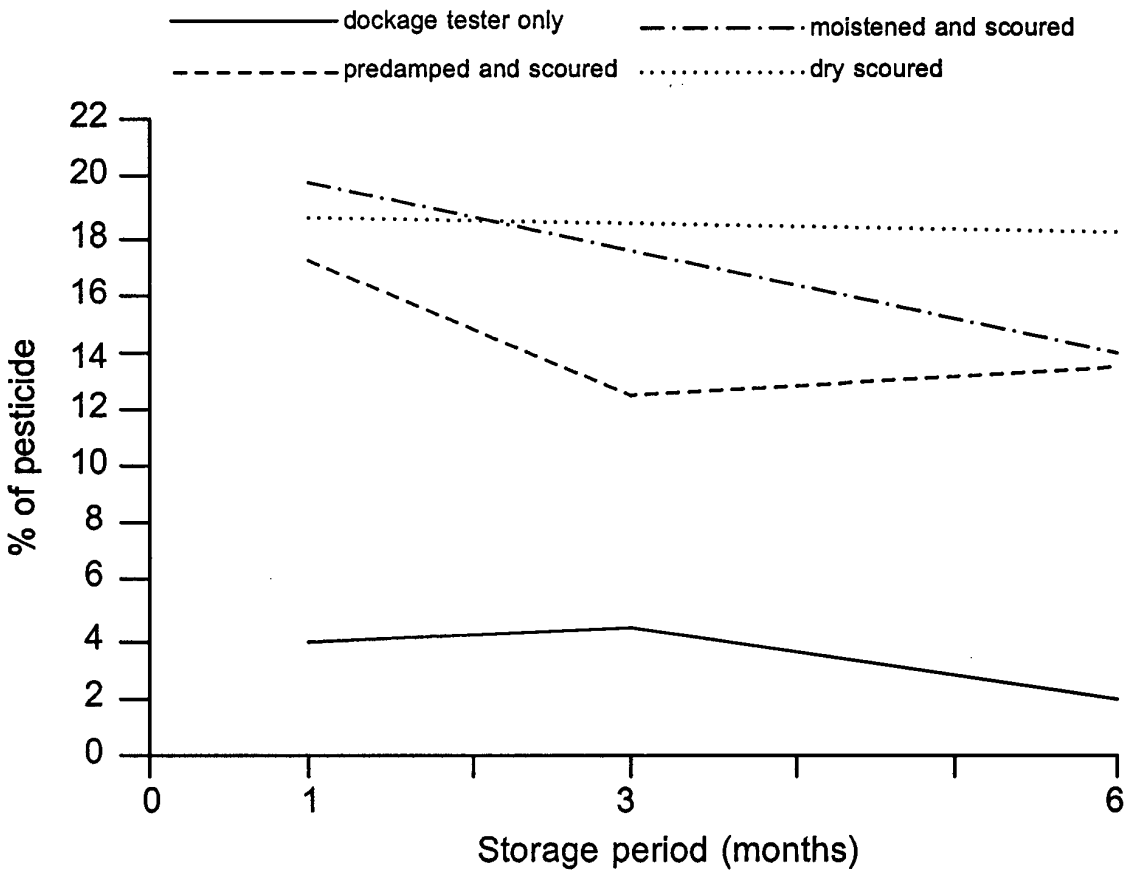

Figure 1 The percentage of total pesticide recovered associated with the wheat screenings - pirimiphos methyl e/c application

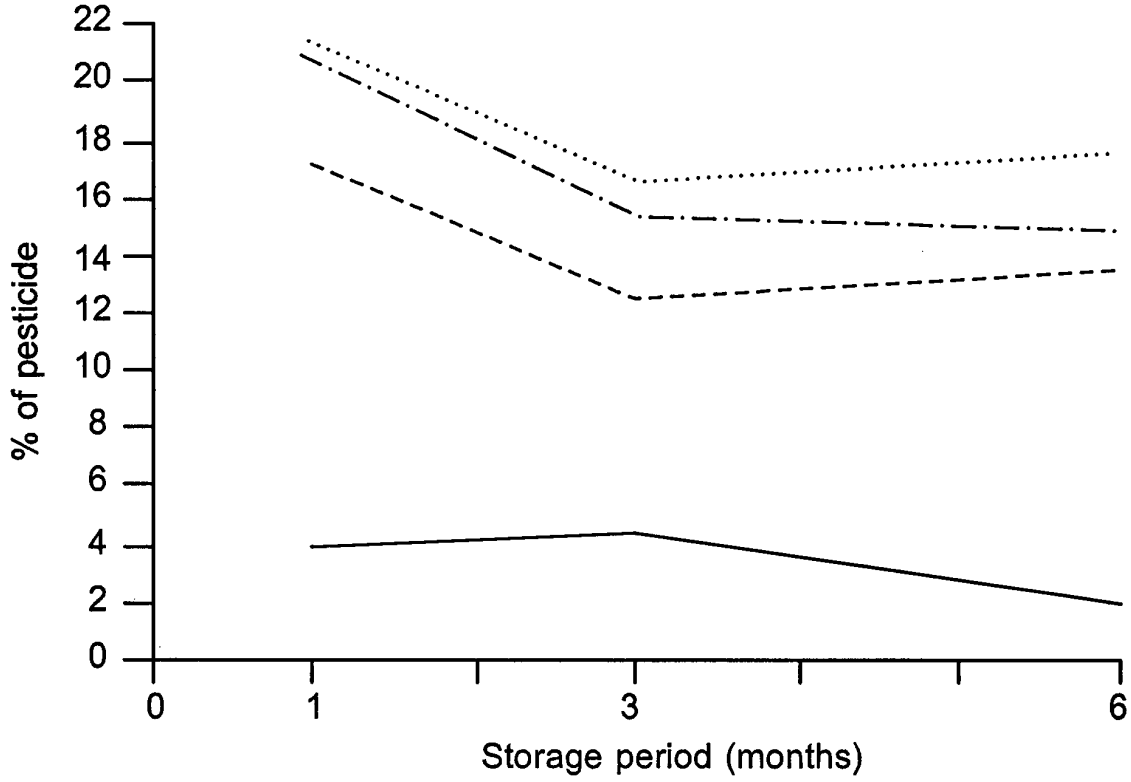

Figure 2. The percentage of total pesticide recovered associated with the wheat screenings - pirimiphos methyl dust application

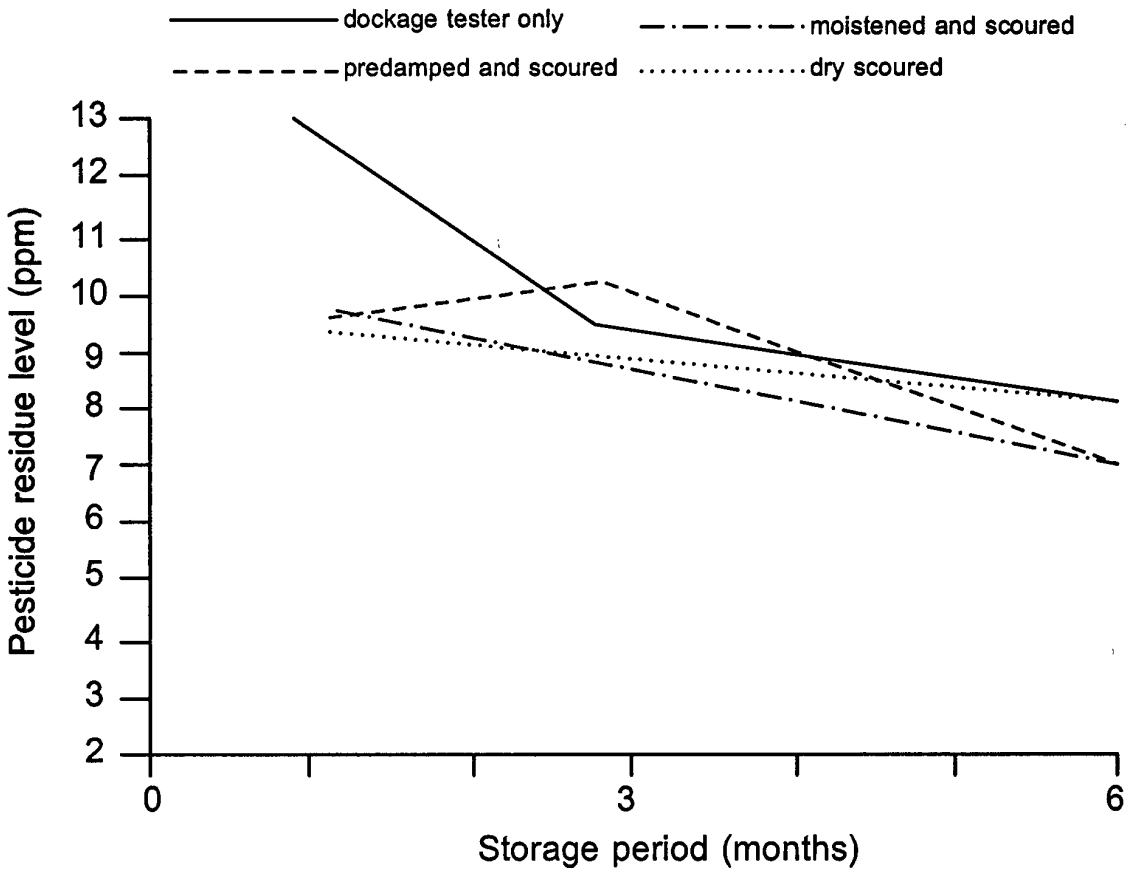

Figure 3. The pesticide residue level associated with the bran fraction after milling - primiphos methyl e/c application

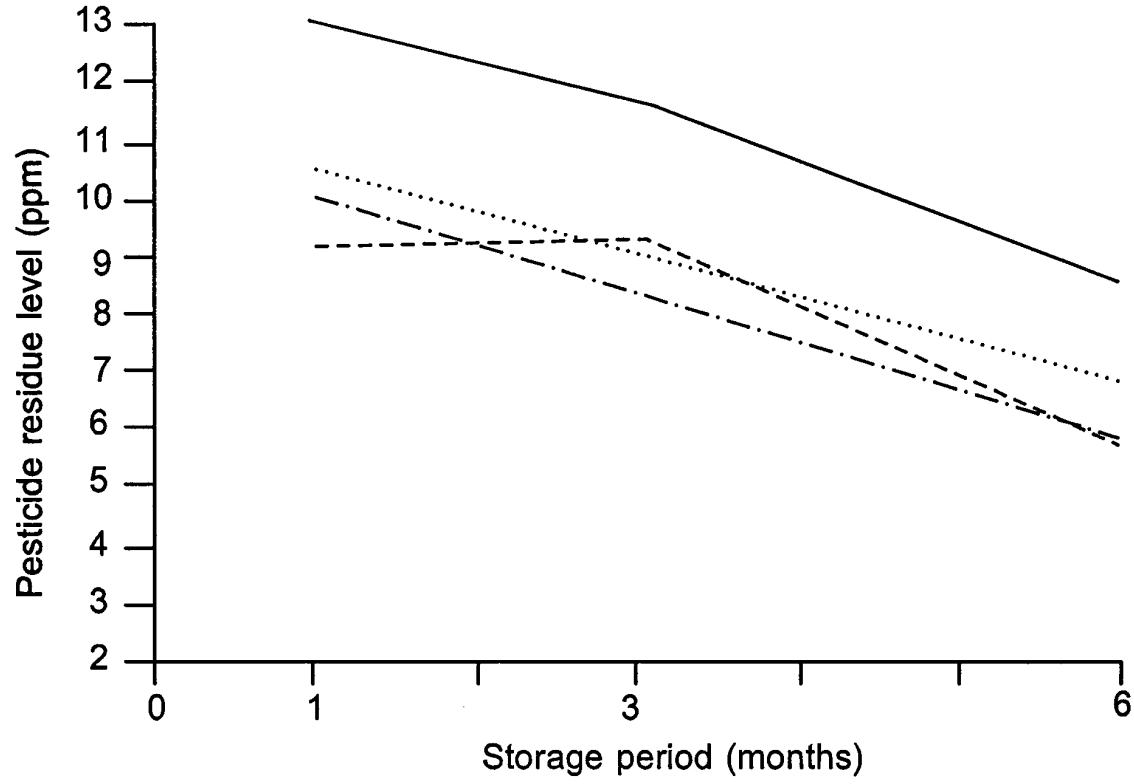

Figure 4. The pesticide residue level associated with the bran fraction after milling - pirimiphos methly dust application

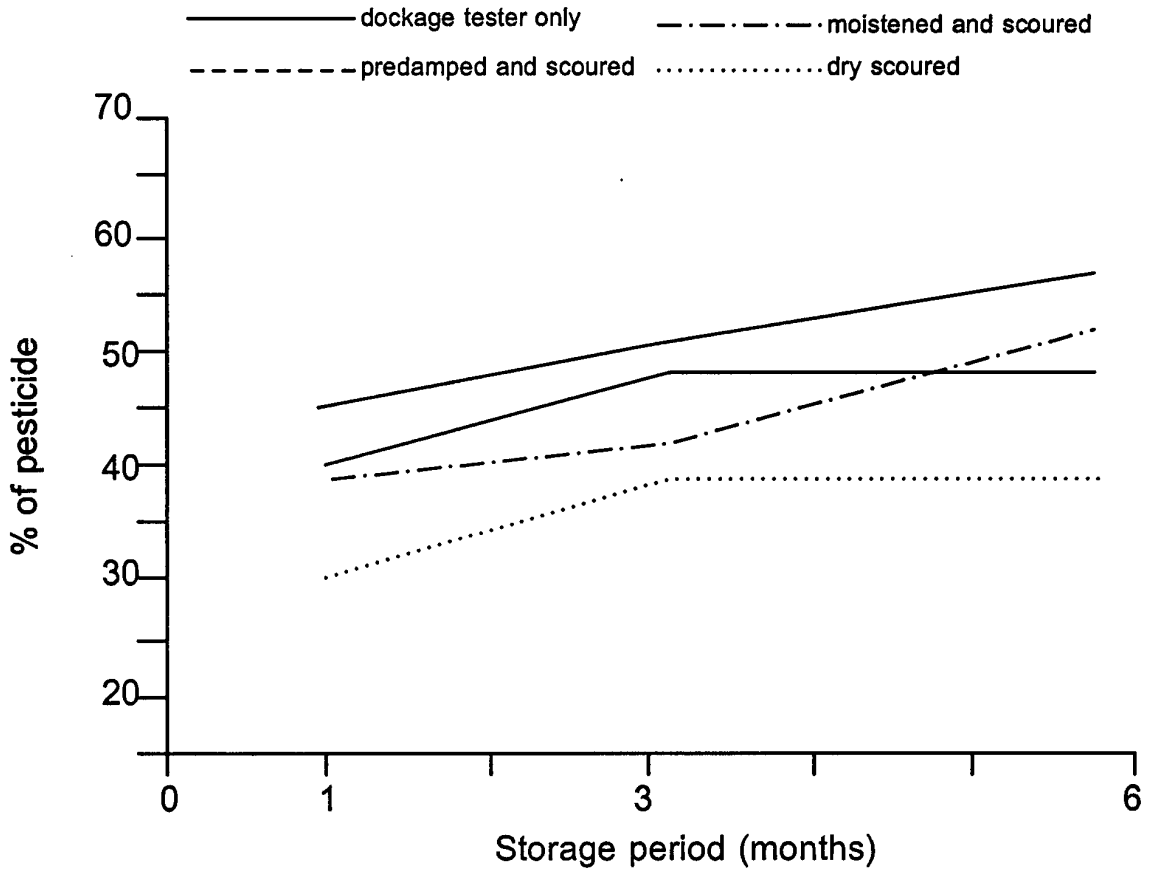

Figure 5. The percentage of recovered pesticide residue level associated with the bran fraction after milling - pirimiphos methlyl e/c application

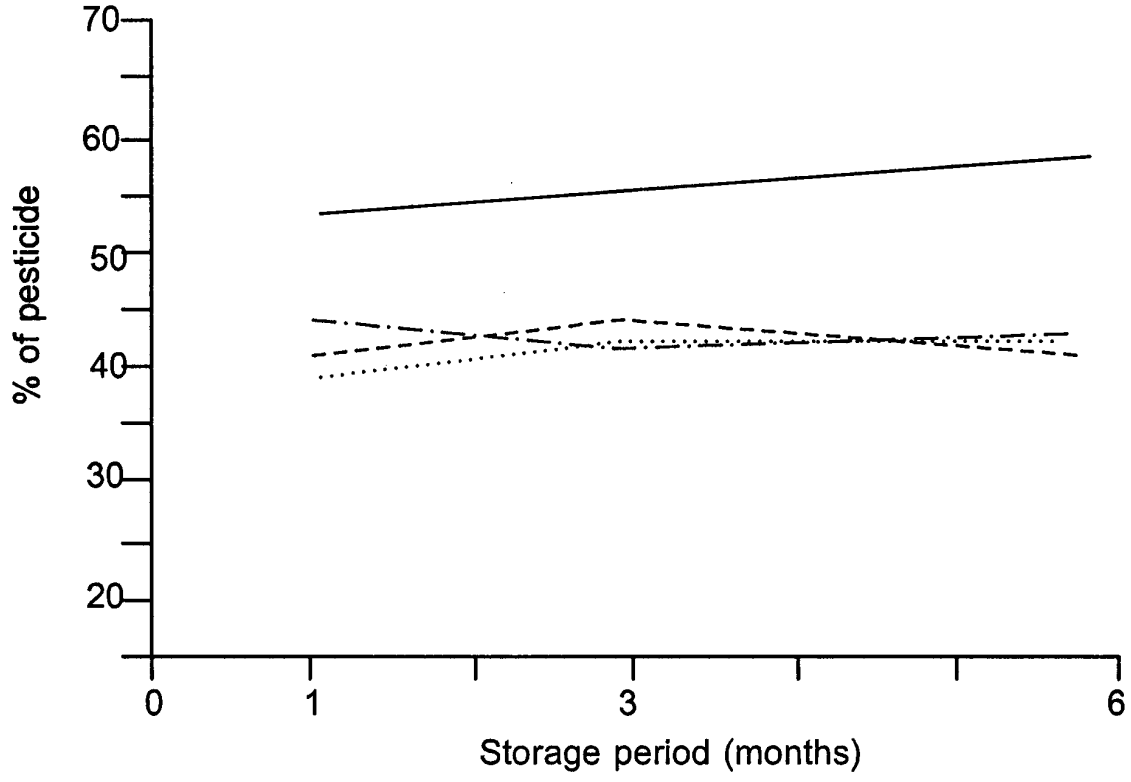

Figure 6. The percentage of recovered pesticide residue level associated with the bran faction after milling - pirimiphos methyl dust application

THE REDUCTION OF MICROBIOLOGICAL COUNTS IN WHEAT AND FLOUR

OBJECTIVE

To investigate the efficacy of a scouring operation prior to milling for reducing the number of microorganisms present in wheat and hence flour.

INTRODUCTION

In recent years there has been an increasing demand for the miller to supply special flours conforming to set microbiological specifications. These special flours are required for canned foods, where numbers of thermophilic bacteria are critical; for lightly processed or unprocessed baby foods, where food poisoning bacteria must be absent and the general level of contamination must be low; and for a wide variety of other foods such as refrigerated dough products, pie pastry, sausages, and pasta products where low counts of bacteria and moulds are required.

Most of the microorganisms in flour samples are present on wheat grains prior to milling. Furthermore, most of these microbes are associated with the outer surface of the grain - especially the grain beard (Seiler, 1986). Therefore, there is potentially an opportunity to reduce the microbial load of wheat and hence flour via a cleaning operation in the mill screenroom prior to milling. Such an operation could assist the miller in producing "low count" flours.

Realistically there are two ways in which a miller might reduce the microbial content of wheat grains, either physically or chemically - however, for the reasons mentioned earlier (page 8) it might be undesirable to use a chemical treatment for grain. Therefore, any cleaning operation devised, should preferably be based on a physical separation of a portion of the pericarp from the intact endosperm. A grain scourer is well suited for such a separation. Although largely employed to dislodge adhering dirt from intact wheat grains, the abrading operation of the scourer lends itself well to the removal of wheat pericarp as well. This project concentrates on the suitability and efficacy of a scouring process for reducing microbial loads.

MATERIALS AND METHODS

A 40kg batch of Galahad home-grown milling wheat was obtained from an ADAS Secondary Trial and cleaned through a CarterDay Dockage Tester prior to scouring. Samples of the screened materials taken from the Dockage Tester were combined and mixed before microbiological testing.

Wheat scouring regimes

During the development of the scouring regimes, and from previous experience with the pesticide work, it became apparent that an excessive amount of broken grain was produced when removing large amounts of pericarp using the scourer fitted with a wire mesh chamber. To overcome this problem an additional alternative scouring regime was employed. The performance of the two regimes was compared.

Both wheat scouring regimes involved the same Westrup laboratory Scarifier Type LA-H that was used for the pesticide work. Regime One employed a steel mesh drum (No.12), metal beaters and wheat samples of 3kg size. This regime utilised three scouring times of 5, 10, and 15 minutes. Regime Two employed a carborundrum drum, nylon-brushes, wheat samples of 1kg, and scouring times of 3 and 10 minutes. The scouring times of both regimes were chosen to represent visibly distinct stages in the removal of the pericarp. Regime Two was repeated using duplicate subsamples (as Run 1 and 2) in order to enable a statistical interpretation of the results to be made.

With both regimes, wheat samples were fed to the scouring section of the scarifier via the accompanying vibratory feeder (setting 6), and the required scouring time was allowed to elapse before the grain outlet was opened to allow the scoured grains out. After the Regime One scouring operations the bran fragments were collected by dismantling the machine and brushing into the appropriate collecting tray, whilst the scoured wheat samples passed out of the scouring chamber and collected in a separate tray. With Regime Two, scoured bran fragments and wheat were both collected in the same tray. Therefore, it was necessary to achieve a separation after scouring. Manual sieving via a 1.0mm sieve was employed. This had the effect of removing most of the bran particles from the scoured wheat sample. Further bran particles attached to the wheat grains were removed using compressed air.

The scarifier was thoroughly cleaned after each scouring operation with the use of brushes, vacuum aspiration and compressed air. Practical considerations prevented the use of aseptic techniques throughout the experimentation.

Samples of scoured wheat and bran particles were collected as appropriate for microbiological testing.

Laboratory milling

Scoured and unscoured wheat samples were conditioned to 15.0% moisture content prior to milling by shaking with the required quantity of water in a sealed plastic container. The moisture was added in the late afternoon the day before milling so that samples received a lying time of between 16 and 24 hours. A capacitance moisture meter (Sinar Agritec, Weybridge Surrey) was used to determine the moisture content of the samples before and after conditioning.

Subsamples of conditioned wheat were removed immediately prior to milling (for method see p.10) and the three milling fractions: bran, offal and flour (straight run + two finisher flours) were also taken for microbiological testing.

Microbiological analysis

Outline of tests: Initial experimentation involving the Regime One technique was devised to follow the distribution of microbial counts throughout the scouring, conditioning and milling processses. This was achieved by testing subsamples of each of the fractions produced, i.e. scoured bran and wheat, conditioned wheat and flour, bran, and offal. Having established the distribution of counts throughout the process using the Regime One technique, it was possible to concentrate on the wheat and flour fractions only with the Regime Two technique. Regime Two was repeated on two separate occasions using duplicate subsamples in order to permit a statistical interpretation to be made of this data.

Microbiological testing: Samples (40g) were presoaked (wheat for 60min, flour; bran and offal for 30min) in 360ml of sterile saline peptone water. After soaking, each sample was mixed for 5 minutes using a stomacher. The primary dilution was allowed to settle for 5 minutes before removing the supernatant to prepare the secondary dilutions.

With samples from Scarifier Regime One 0.1ml of the dilutions were spread plated in duplicate onto cysteine electrolyte deficient agar (CLED), oxytetracycline glucose yeast extract agar (OGYE) and violet red bile agar (VRB) media to test for Total Viable Count (TVC), Moulds and Yeasts, and Coliform bacteria respectively. An overlay of VRB medium was made to the VRB spread plates.

Dilutions from samples from Scarifier Regime Two were also plated in duplicate, using a spread plate method with the VRB medium but a spiral plating method was used with CLED and OGYE.

The VRB and CLED plates were incubated at 30°C for 1 and 4 days respectively. The OGYE plates were incubated at 25°C for 4 days.

After the required incubation period, the number of colonies present on each plate were counted and the count per g of sample calculated.

RESULTS

Laboratory milling

The performance of the scoured wheat samples upon laboratory milling is shown in Table 1 (Regime One) and Table 2 (Regime Two).

Microbiological analysis

The effects of Regime One scouring on the counts of bacteria and mould in the wheat and milled fractions are shown in Tables 3 and 4 respectively. The results are also presented as histograms in Figure 1 for total viable count and Figure 2 for moulds. Table 5 shows the effect of Regime Two scouring on the bacteria and mould counts of wheat and flour. A graphical representation of these data is presented in Figure 3 for total viable count and Figure 4 for moulds and yeasts.

No results are included for the coliform counts since the numbers present in the wheat used were too low to enable differences to be detected between scouring treatments.

Statistical analysis

A statistical interpretation of the data from Regime Two is provided in Table 6 (Run 1) and Table 7 (Run 2). Least significant differences (LSD) are given to assess the statistical significance of the differences between means. Any pair of means is significantly different ($p < 0.05$) if they differ by more than this figure.

DISCUSSION

Wheat milling

Tables 1 and 2 outline the milling performance of the scoured and unscoured wheat samples from the two regimes. It is evident from these data that with increasing scouring time a greater percentage of scoured fragments are removed. As would be expected this action has the effect of increasing flour extraction rates from the remaining wheat kernel upon milling. However, when it is considered that up to 9.9% (Regime Two) and 15.2% (Regime One) of the total wheat kernel was removed in the form of scoured fragments, it is interesting to note that the flour extraction rates increased by only 7.4% and 3% respectively. These results suggest that the Regime One technique in particular was not selective in its removal of bran from the wheat kernel.

Regime One scouring procedure

From Table 3 it is evident that passing the wheat as received through the Carter Day Dockage Tester was effective in removing screenings with a very high bacterial count and leaving a cleaned wheat with a count of bacteria reduced by about tenfold. Surprisingly, the mould count was hardly changed as a result of this treatment.

Scouring caused a reduction in the counts of bacteria and moulds in the wheat but there is little evidence to suggest that this increased with scouring time. The reduction is small and scarcely enough to give optimism that flour counts will be reduced to a significant extent.

The total viable counts in the scoured wheat after conditioning were rather higher than in the wheat before conditioning suggesting that some multiplication had occurred during the conditioning period. It will be noted (Figure 1) that the increase in count with the scoured wheat is greater than with the wheat which

was not scoured. This may suggest that water uptake and hence multiplication of bacteria was more rapid when the wheat was scoured. The counts in the conditioned wheats are similar and no difference in the level of bacteria in the flour might be anticipated. Unlike bacteria there is no indication of multiplication of moulds in the conditioned wheat. A slight reduction in numbers of moulds occurred with the longer scouring times.

Despite the fact that the conditioned wheats gave similar counts, a trend can be observed for the numbers of bacteria and moulds to be lower in all the milling fractions (i.e. bran, offal and flour) derived from the scoured wheats (Table 4, Figure 2). The effect was particularly noticeable with the wheats scoured for 5 and 10 min. With flour, the bacterial count was reduced by about 6 fold and the mould count by over 10 fold as a result of these treatments.

Regime Two scouring procedure

In general, there was good agreement between counts carried out on the duplicate samples (Table 5). Taking all treatments into account the coefficient of variation for bacterial counts was <6.5% in Run 1 and <17.0% in Run 2 (Tables 7 and 8). Replication tended to be less good with mould and yeast counts especially with flour where the variation was 17.2% in Run 1 and 44.2% in Run 2. Even so, the situation is satisfactory.

Rather different results were obtained on the two Runs. In Run 1, with bacteria, there was little difference between the counts in scoured and unscoured wheat or flour (see Figure 3). The same is true for mould and yeasts in wheat but there is a small reduction with scouring time in the case of flour (see Figure 4). As in the Regime One test, there is an increase in total viable count in the wheat after conditioning.

in Run 2, a reduction in counts of bacteria and moulds/yeasts occurred with the unscoured wheat, conditioned wheat and flour with scouring time. The effect was small but significant in most cases.

Neither scouring regime was found to be particularly effective in reducing the microbial load in flour - a finding in agreement with Eyles et al (1989). The greatest reductions in count were obtained using the Regime One scouring procedure but the best that was achieved was a tenfold reduction which is only of marginal commercial interest. The discrepancy in results between the

replicated samples in the two Regime Two tests does not give confidence that this level of reduction would be achieved on all occasions.

It is possible that rather more consistent reductions in microbial numbers might be achieved by scouring under conditions of continuous production. In the tests described, the Buhler mill was cleaned to remove residual flour before feeding in the wheat samples. Nevertheless, it seems inevitable that some higher count material was present in the mill which could alter the counts in the flour from the test wheats. It is difficult to estimate how this batch system would compare to a commercial continuous production system - for whilst a continuous production system might be more likely to self clean, it would probably develop dead areas which could serve as reservoirs for further contamination of the mill. Thus, it would be wrong to damn the usefulness of a scouring procedure on the basis of these tests.

It is not easy to see how a laboratory procedure can be adapted to give a better picture of what is likely to happen in the commercial situation. Changes could be made to optimise conditioning time and scarifier performance but it is not possible to create the continuous flow of wheat which would exist at a mill. It is recommended that any future tests to evaluate the worth of scouring procedure should be carried out at a commercial mill.

CONCLUSION

It is unlikely that the commercial scale version of the wheat scouring machine under study when incorporated in the screenroom flow could achieve commercially useful microbial count reduction in resultant milled flours.

ACKNOWLEDGEMENTS

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Seiler, D.A.L. (1986). The microbial content of wheat and flour. International Biodeterioration Supp., 22, 35-40.

Table 1: Milling performance of Regime One wheat samples.

Wheat Sample	Extraction Rate on products (%) Straight run			(+2FF)	Scoured fragments (%)
	Flour	Bran	Offal	Flour	
Control (unscoured)	69.3	23.5	7.20	74.7	---
Scoured 5 minutes	71.0	21.9	7.10	76.6	4.70
Scoured 10 minutes	71.0	21.4	7.60	76.8	6.30
Scoured 15 minutes	72.2	20.5	7.30	77.7	15.2

+2FF = with two finisher flours.

Table 2: Milling performance of Regime Two wheat samples.

Wheat Sample		Extraction Rate on products (%) Straight run			(+2FF)	Scoured fragments (%)
		Flour	Bran	Offal	Flour	
Control	(Run 1)	70.7	24.2	5.10	76.0	---
	(Run 2)	70.9	24.1	5.00	76.0	---
3 min	(Run 1)	73.5	19.8	6.70	79.0	2.9
	(Run 2)	73.6	20.8	5.60	77.8	2.8
10 min	(Run 1)	77.7	14.6	7.70	83.4	9.3
	(Run 2)	77.3	15.4	7.30	83.4	9.9

+2FF - with two finisher flours.

Table 3: Effect of Regime One scouring procedure on the microbial count in wheat and scoured fractions.

Sample	Scouring time (min)	Count / g of sample	
		TVC	Mould Count
Wheat as received	--	435000	96000
Screenings (CarterDay)	--	4500000	595000
Cleaned Wheat (control)	--	56000	101000
Scoured Bran	5	305000	64500
Scoured Bran	10	615000	54000
Scoured Bran	15	1670000	33500
Wheat Scoured	5	25000	24500
Wheat Scoured	10	12000	25000
Wheat Scoured	15	16000	14000
Conditioned Wheat	0	64000	51000
Conditioned Wheat	5	88500	51000
Condtioned Wheat	10	45500	14000
Condtioned Wheat	15	53000	23000

Table 4: Effect of Regime One scouring procedure on the microbial count in wheat milled fractions.

Sample	Scouring time (min)	Count / g of sample	
		TVC	Mould Count
Buhler Bran	0	90000	24000
Buhler Bran	5	24000	10000
Buhler Bran	10	23500	2000
Buhler Bran	15	36500	3000
Buhler Offal	0	269000	175000
Buhler Offal	5	19500	34000
Buhler Offal	10	71500	63500
Buhler Offal	15	143000	94500
Buhler Flour	0	20600 20000	26500 22500
Buhler Flour	5	4050 4400	1850 1950
Buhler Flour	10	4750 3700	1250 1300
Buhler Flour	15	11600 13500	10500 14500

Table 5: Effect of Regime Two scouring on the microbial count of wheat and flour.

Wheat Sample	Scouring time (min)	Count / g of sample			
		TVC		Mould + Yeast	
		Run 1	Run 2	Run 1	Run 2
Control wheat	0	37500	113637	21600	28000
		39000	101515	27200	27600
Scoured wheat	3	28100	22800	10000	23400
		28200	32800	9100	22100
Scoured wheat	10	22353	3000	9000	11300
		19312	2900	8400	6000
Conditioned Wheat	0	75758	77273	24900	17900
		71212	81819	28900	17300
Conditioned Wheat	3	31818	62121	9400	5900
		35454	59091	7700	7000
Conditioned Wheat	10	36364	59091	16300	9400
		39000	57576	16400	8000
Buhler Flour	0	5800	9700	1300	1500
		5900	7400	1400	1000
Buhler Flour	3	9400	3900	1000	400
		10200	4000	1200	200
Buhler Flour	10	5000	3500	700	600
		5400	4000	500	300

Table 6: Statistical evaluation of counts in Run 1.

Sample	Mean Log Count / g of sample					
	Total Viable Count			Mould + Yeast Count		
	Wheat	Condt	Flour	Wheat	Condt	Flour
Control	4.583	4.866	3.767	4.385	4.429	3.130
Scoured 3 min	4.450	4.526	3.991	3.979	3.929	3.039
Scoured 10 min	4.318	4.576	3.716	3.939	4.214	2.772
Standard devation	0.027	0.025	0.020	0.046	0.044	0.069
LSD (5% Level)	0.086	0.081	0.064	0.146	0.141	0.219
C. Variation (%)	6.410	5.930	4.710	11.20	10.70	17.20

Table 7: Statistical evaluation of counts in Run 2.

Sample	Mean Log Count / g of sample					
	Total Viable Count			Mould + Yeast Count		
	Wheat	Condt	Flour	Wheat	Condt	Flour
Control	5.031	4.900	3.928	4.444	4.246	3.088
Scoured 3 min	4.437	4.782	3.597	4.357	3.808	2.477
Scoured 10 min	3.470	4.766	3.573	3.916	3.938	2.628
Standard Devation	0.068	0.014	0.054	0.113	0.042	0.159
LSD (5% Level)	0.216	0.045	0.171	0.359	0.134	0.508
C. Variation (%)	16.90	3.280	13.20	29.70	10.20	44.20

KEY FOR TABLES 6-7: Condt = Conditioned Wheat

LSD = Least Significant Difference

C. Variation = Coefficient of Variation

Figure 1. The effect of Regime One scouring and conditioning on the Total Viable Counts of wheat and milled flour.

Figure 2. The effect of Regime One scouring and conditioning on the mould counts of wheat and milled flour.

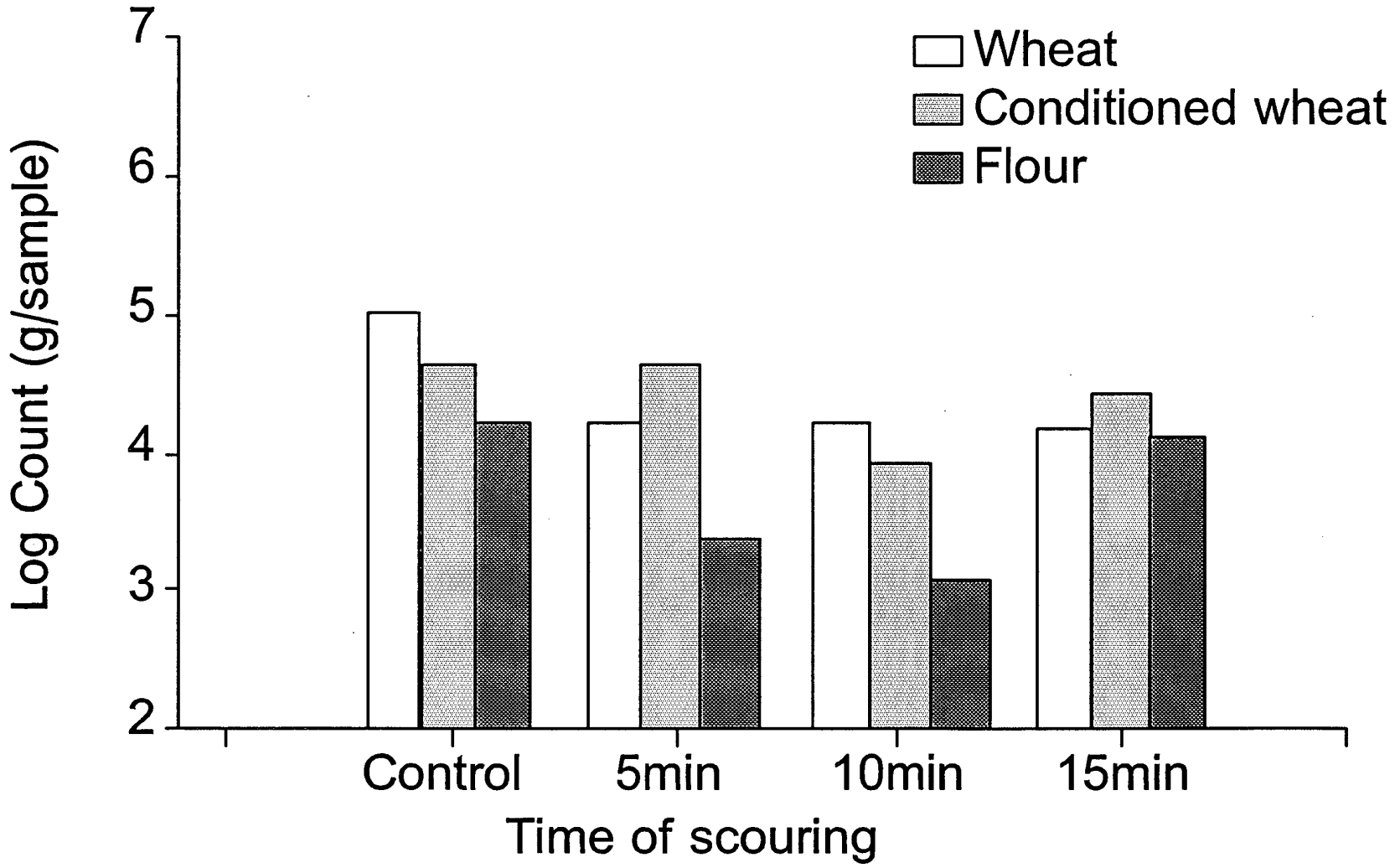

Figure 3. The effect of Regime Two scouring and conditioning on the Total Viable Counts of wheat and milled flour.

Figure 4. The effect of Regime Two scouring and conditioning on the Mould and Yeast count of wheat and milled flour.