THE DETECTION OF INSECTS IN GRAIN USING CONVENTIONAL SAMPLING SPEARS

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Abstract

A sampling strategy for bulk grain, using two different devices to collect samples, was tested using dead insects and dyed grain as markers. The markers were added to bulk grain at the rates of 1/5kg, 1/kg and 5/kg and samples were withdrawn using a gravity spear and a compartmented spear. The tests were performed in both France and England, following the same protocol. No difference was shown between the two types of spear and the results from the two counties were broadly similar,excepting differences in the quality of the batches of grain used. Despite the collection and examination of a much larger mass of grain than would be normal in this type of sampling, detection of either dead insects or dyed grain was unreliable at the two lower levels of inclusion. Only when added at 5/kg, could insects and dyed grains be detected with reliability and the total population be estimated.

INTRODUCTION

Traditionally, insect populations in bulk grain have been detected and estimated by withdrawing and examining samples. The same technique is also used to determine general quality parameters of grain such as moisture content, fine material and protein content. Despite the obvious importance of obtaining representative values and the commercial implications attached to the results, limited research appears to have been carried out on the topic.

Although many workers must have had reservations about the effectiveness and reliability of grain sampling to detect and quantify insects in bulk grain, few practical alternative methods were available. However, the recent developments in insect detection, such as acoustic methods (Feurat-Lessard and

Andrieu, 1986) and, more particularly, trapping techniques (Loshiavo, 1975), have begun to offer viable alternatives to sampling. Unfortunately, trapping techniques are limited mostly to static bulks of grain and there is still a need to establish the quality of grain during transit. Storekeepers, exporters and commercial processors have to make rapid judgements on loads of grain as they are delivered to the silo, port or mill. Collection and examination of samples is still the most widely practised method.

Although new approaches to pest detection in bulk grain appear to offer far more sensitive methods of detecting insects, some means of calibrating the new methods has to be found. Up to now, workers have either related trap catches to total pest populations at the beginning and end of experiments or to the results obtained by the removal and examination of samples (Cogan and Wakefield, 1987). This latter approach has practical merit as it relates trap catch to conventional methodology. However, the lack of data on the efficiency of collection and examination of sample, is a serious drawback.

Against this background it was decided to investigate the effectiveness of sampling methods. Problems associated with grain storage and trading are world-wide, so an international project was set up between the U.K. and France. This paper reports the first phase of the work in which conventional spear sampling was assessed in both countries, using similar methodology.

MATERIALS AND METHODS

i) Grain

Milling quality wheat was obtained from commercial sources. The specification for the grain required that it contained less than 1% fine material and had a moisture content in the range of 13.5 - 15%. On delivery, samples of the grain were examined for infestation.

ii) Containers for the grain

The tests at Slough, England, used three metal bins measuring 3x3x3 m and each containing about 20 tonnes of grain. At Bordeaux, France the grain was held in three cylindrical plastic bins measuring 1.65m in diameter and 2.5m deep. These were filled to a depth of about 1.7 - 2 m with about 2.5 tonnes of grain in each bin. In both countries, there was access to the top of the bins for sampling.

iii) Sampling spears

Two types of spear were used; a single compartment gravity spear that collected 200 - 250g samples and a 1.6m long, multi-compartmented spear. The gravity spear remained closed when pushed into the grain and did not open until withdrawal began. Therefore, it is likely that this spear collected a sample from a discrete part of the grain bulk. Although nominally the same, there was a slight difference between the the spears used in

France and England. The compartmented spear used in England had 11 compartments and collected a sample of about 300g but the French spear had only 8 compartments and collected a sample of less than 200g. With both spears, the compartments were closed as the spear was inserted and were then opened by turning the handle, thus collecting a series of samples from different depths in the grain.

Both types of spear are in widespread use for experimental purposes and in commerce.

iv) Sampling markers

Dead insects were used as markers for the experiment. It was decided that the mobility associated with live insects would increase the difficulties in obtaining a representative method of estimating populations. Therefore, dead adult *Sitophilus granarius* and *Oryzaephilus surinamensis* were used. The insects were produced in mass cultures and killed by freezing at -18°C for 48 hours. In addition, some grain taken from the main bulks was dyed with a green colour and the dyed grains were also used as markers.

The dead insects and dyed grains were added to the grain bulks as the grain was moved between containers by means of augers, conveyers and elevators. At Slough the grain was moved at a rate of about 10 tonnes/h and a mixture of dyed grain and insects was added to the flow of grain at the point where it was discharged from the conveying system into a bin. In practice, it did not prove possible to add the markers in a continuous flow, so appropriately sized batches were added at 10 minute intervals. At Bordeaux a similar procedure was followed but the conveying rate was 4 tonnes/h.

The experiments were repeated three times using different rates of inclusion for the markers. These rates were 1/5kg of grain, 1/kg and 5/kg, for each species and for the dyed grain. The same bulk of grain was used throughout the experiment so the rates of addition were cumulative. However, it must be born in mind that the process of conveying is likely to have damaged the dead insects and many may have been broken up to the point where they were not noticed during the assessment of samples.

v) Collection of samples

Samples were withdrawn from three points from each bin: diagonally across the corners of the square bins used at Slough and along the diameter of the bins at Bordeaux. In both cases, one point was at the centre. The precise methods of collecting samples varied between the two types of spear. With the gravity spear, samples were collected at depths of 0.1, 0.6 and 1.1m from each point. Three replicate lots of samples were collected from each bin for each level of inclusion of markers. The multi-compartmented spear was inserted to its maximum depth, opened, filled and then closed, before withdrawal. The grain in the compartments was then bulked to give a single sample from each sampling point. Once again, this procedure was replicated three times per sample point for each level of inclusion. In France the replicate samples for each bin were mixed to give a single lot of grain for examination. In England, each sample was examined separately.

vi) Assessment of samples

The numbers of dead insects, dyed grain and the amount of fine material in each sample or composite sample was determined. Each sample was shaken 30 times over a 2mm mesh to remove insects and fine material. The sievings were weighed and examined for insects. The Slough results separated the insect species but at Bordeaux they were combined. The sieved grain was then spread in a mono-grain layer on a tray and examined for dyed kernels.

RESULTS

The large number of individual samples collected and examined during the experiment (71 samples/inclusion rate for the gravity spear) required some condensation, analysis and simplification before presentation. Variations occurred between the methodology used by Slough and Bordeaux, and these limit the number of comparisons that can be made between the results obtained in the two countries. However, the general trends shown by the results in both countries can be compared. For the purposes of this paper, variation between replicate bins, which was in any case small, is ignored.

Data from all samples for each inclusion rate and sampling spear were expresed as means. The results, together with standard deviations are given in Table 1.

The numbers of insects in grain are often expressed in terms of numbers per kg. Therefore, the numbers detected per kg of total insects and dyed grains have been calculated from the sampling data. The results from both sites were combined and are given in Table 2. The level of insects and dyed grains detected per kg, when expressed as mean values for all samples collected at both sites, appear to give good general correlation with the levels of inclusion. However, these figures were achieved by using data from a large number of samples from relatively small quantities of grain. The results from Slough, where all samples were kept separate, offer the opportunity for further analysis in terms of calculating the number of samples that contained 1 or more insects or dyed grains. The results of these calculations are given in Table 3.

DISCUSSION AND CONCLUSIONS

The small standard deviations for the weights of sample shows that the methods of collection were consistent and reliable. The differences in average weights of samples between Slough and Bordeaux can be explained by the French and English wheat having different specific weights. The differences between the weights of fines can also be attributed to differences between the two batches of grain. However, it should be noted that the standard

Table 1. Numbers of insects and dyed grains recovered from grain artificially contaminated with *O. surinamensis* and *S. granarius*, and dyed grains. The figures in parentheses are standard deviations.

	Sample wt. g	Fines g	Insects/sample O. sur.	S. gran.	Total	Dyed grains /sample
			Inclusion rate of 1/5kg			
Gravity spear						
Slough	208 (8.5)	0.4 (0.2)	0.03 (0.2)	0	0.03 (0.2)	0
Bordeaux	226 (5.9)	2.9 (1.1)			0.02 (0.1)	0.03 (0.2)
Compartmented spear						
Slough	259 (20.6)	0.3 (0.2)	0.04 (0.2)	0	0.04 (0.19)	0.04 (0.19)
Bordeaux	592 (33.3)	12.1 (18.6)			0.06 (0.24)	0.06 (0.24)
			Inclusion rate of 1/kg			
Gravity spear						
Slough	223 (18.9)	1.5 (0.3)	0.1 (0.3)	0.1 (0.3)	0.2 (0.3)	0.03 (0.6)
Bordeaux	227 (9.5)	2.77 (1.2)			0.4 (0.9)	0.3 (0.6)
Compartmented spear						
Slough	273 (28.2)	1.2 (0.2)	0.2 (0.4)	0.2 (0.4)	0.3 (0.4)	0.1 (0.5)
Bordeaux	586 (27.8)	6.6 (1.8)			1.2 (1.7)	0.8 (1.5)
			Inclusion rate of 5/kg			
Gravity spear						
Slough	239 (23.5)	1.0 (0.4)	0.1 (0.5)	0.3 (0.3)	0.4 (0.5)	0.7 (2.1)
Bordeaux	225 (10.8)	3.0 (1.2)			2.7 (3.6)	0.7 (2.1)
Compartmented spear						
Slough	285 (14.5)	1.2 (0.7)	0	0.4 (0.8)	0.4 (0.8)	0.8 (0.9)
Bordeaux	576 (21.5)	8.5 (2.6)			6.0 (6.7)	2.0 (2.5)

Table 2. The numbers of insects and dyed grains found per kg in samples removed from wheat to which insects and dyed grains had been added at three inclusion rates.

	Inclusion rate	Insects/kg	Dyed grains/kg
<u>Compartmented spear</u> (54 samples)	1/5kg	0.1	0.1
	1/kg	1.5	0.9
	5/kg	6.0	3.2
<u>Gravity spear</u> (162 samples)	1/5kg	0.1	0.1
	1/kg	1.5	0.9
	5/kg	5.8	3.0

Table 3. The number of grain samples containing one or more dead insects or dyed grains.

	Inclusion rate	No. samples with 1 or more insects	No. samples with 1 or more dyed grains
<u>Gravity spear</u> (81 samples)			
	1/5kg	1	0
	1/kg	21	6
	5/kg	33	44
<u>Compartmented spear</u> (27 samples)			
	1/5kg	1	1
	1/kg	8	3
	5/kg	15	15

deviations for fine material were large at both sites. These results must cast some doubt on the validity of any sampling regime to give an accurate estimation of this important commercial quality criterium.

The inclusion rate of 1/5kg was too low for reliable detection of either dyed grains or insects, despite the removal and examination of more than 4kg of grain from 20 tonnes at Slough or 2.5 tonnes at Bordeaux. Results at Slough were little better at the inclusion rate of 1/kg, although at Bordeaux, the the mean results with the compartmented spear suggest some chance of detecting insects or dyed grains with a single sample. However, the mean result must be cosidered in the light of the large standard deviation. Conventional sampling theory suggests that at the inclusion rate of 1/kg, a sample mass of 3kg should ensure detection (Fleurat-Lessard, 1988). However, this theory is based on the assumption that the insects have a random and even distribution, whereas they are almost certainly clumped rather than evenly distributed. At the highest inclusion rate, the likelihood of detecting insects and dyed grains appeared good but the standard deviations were still large and precluded any comparisons between the effectiveness of the two types of spear. The large mean number of insects detected by Bordeaux using the compartmented spear was caused by two samples that each contained 10 or more insects. This was the only evidence of grossly uneven distribution found throughout the trial.

No significant difference could be detected using the Slough data between the two species of insects at any level of inclusion. In general, taking account of the standard deviations, the detection rates of dyed grains and combined insects was little different. However, combining the two species meant that there were twice as many insects as dyed grains and this could have been expected to be reflected in the ease of detection. Therefore, there is some indication that the dead insects were more difficult to detect than dyed grains.

The individual samplings, in which 9 samples were removed from each bin with the gravity spear and 3 samples with the compartmented spear, gave very poor estimates of the level of inclusion. However, if all the sample data were combined for both sites, the prediction of the level of inclusion of dyed grains became good, although for insects the value was always low. Similarly, the chance of detection was directly related to the level of inclusion and the number of samples, or perhaps the weight of grain, examined. The Slough data for individual samples shows that the probability of a sample containing an insect or a dyed grain was very low at the inclusion rate of 1/5kg and that eighty or more samples would have to be taken with the gravity spear to detect the insects (about 18kg of grain). At 1/kg two thirds of the samples still contained no markers, suggesting that multiple samples would still be needed to ensure detection. If a comparison is made between these levels of inclusion and practical infestations, the problems of using spear sampling to detect pests for a pest management programme are clear. At 1/5kg, the population increase likely during a single generation would bring the level of infestation to a totally unacceptable level. Even at the highest level of inclusion only half the samples contained markers so that a single spear sample taken from a truck load of grain has only a 50% chance of detecting infestation as high as 5/kg.

The difficulties of detecting and estimating dead insects and dyed grains experienced during these experiments are almost certainly related to the amount of grain collected and examined and the distribution of the markers within the bulks of grain. Every effort was made during the addition of the markers to use a regular and consistent method and only very limited evidence of clumped distribution was obtained. However, although the distribution of the dead insects and dyed grains may have been random it was almost certainly not even. Thus, large volumes of grain within the bulks, particularly at the lower levels of inclusion, would not have contained markers. Therefore, chances of detection and accuracy of prediction are improved by increasing the numbers of samples and/or the amount of grain that is examined. However, a realistic view, in the light of the results from this work, would suggest that using conventional spears, the detection and estimation of populations at or below 1/kg may be impossible.

The methods used in these experiments are in general, comparable with standard recommendations for grain sampling such as the ISO method or USDA recommendation for export grain. However, the ISO method recommends that 1kg of grain is examined from 20 - 30 tonne lots and the USDA recommendations are for substantially lower sampling rates. The results from the work reported here would suggest that the neither method is likely to consistently detect insects unless the level of infestation exceeds 5/kg.

It is reasonable to question the validity of results obtained with dead insects in relation to live populations that have the ability to move through the grain and, perhaps be attracted or repelled from certain parts of the bulk. However, in practice, unless the reasons for the movement of the insects can be anticipated, there must be a large element of luck in the likelihood of a sample being withdrawn from a local concentration of pests. Clearly, the temperature and moisture content need to be taken into account when sampling a static bulk of grain but this may not be applicable when sampling grain recently loaded on a vehicle.

The conclusions arising from the work so far have important implications for both research on insect populations and for commercial transactions. It is clear that the results of intake sampling at mills, silos, etc can only be considered to give a definitive answer if insects are found. Failure to detect pests, using for example the ISO method, is merely an indication that the infestation may be present at a level below 5/kg. The inability to detect and estimate low levels of infestation by spear sampling, unless large numbers of samples are examined, must create serious problems for workers attempting to calibrate trapping methods. It may be impossible to relate trap catch data to insect populations in terms of numbers/kg and may be more realistic to devise some system of risk factor.

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LA DETECTION DES INSECTES DANS DU GRAIN PAR L'UTILISATION DES SONDES D'ECHANTILLONNAGE CLASSIQUE.

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Résumé

On a étudié l'efficience et la répétabilité de deux types de sondes d'échantillonnage largement répandues et servant à estimer la qualité du grain, en apportant une attention particulière à la détection des insectes. Un lot de blé de bonne qualité a été contaminé par des adultes morts et des grains colorés à des densités diverses. Des échantillons ont été récoltés à l'aide des deux types de sondes. On a mesuré la densité d'insectes, de grains colorés et d'impuretés fines, puis les résultats ont été analysés statistiquement. Ce travail a été accompli dans deux lots de 3 et 20 tonnes de grain, et toutes les expériences ont été répétées. L'expérience a été réalisée à la fois en France et en Angleterre.

Lorsqu'on a corrigé les résultats en fonction du poids de grain enlevé, il n'a pas été constaté de variations entre les résultats obtenus avec les deux types de sondes, à l'exception de la sonde à compartiments avec la laquelle on a enregistré une plus grande quantité d'impuretés. Aucune des deux sondes n'a pu détecter d'insectes ou de grains colorés à des taux d'incorporation (mélange) allant jusqu'à 1/kg, malgré les pressions d'échantillonnage qui étaient jusqu'à dix fois plus élevées que la normale. Une détection fiable a pu être obtenue à des taux d'incorporation de 5/kg, mais même ce niveau élevé de contamination n'a pu être quantifié avec précision.

Ces résultats ont une implication technique très importante, et justifient d'autres recherches.