Improved procedures for measurement of fumigants

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Abstract

The requirement for analysis of volatile chemicals has greatly increased in recent years and, as a result, analytical procedures have improved. Research, in particular by Regulatory Agencies in the United States, such as EPA or NIOSH, has resulted in sensitive detection procedures, suitable for regulatory procedures. The recent principal advances in analysis of 'volatiles' can be readily transferred to analysis of fumigants. These include: use of megabore or capillary columns, in place of packed columns; standard procedures for desorption of fumigants from commodities; use of absorbent tubes ('traps') coupled with either solvent or thermal desorption; and use of headspace chromatography. These techniques are illustrated for grain fumigants, using both literature data and unpublished data from the Stored Grain Research Laboratory. The relevance of improved techniques for measurement of fumigants is discussed. Finally, some critical comments are made on analysis of fumigants in grain.

Introduction

In recent years, great advances have been made in the analysis of 'volatile chemicals', which include solvents, air pollutants, naturally-occurring gases and fumigants. These advances are summarised yearly in the journal, Analytical Chemistry, and also regularly in publications from regulatory agencies in the United States (ACGIH 1983; NIOSH 1985; USEPA 1989). These advances enable use of techniques other than those traditionally used for fumigant analysis in stored products (Heuser 1973; Berck 1975), though some recent advances are outlined by Scudamore (1988). In this paper, the relevance of these advances to analysis and, indeed, use of fumigants is discussed. Examples of fumigant analysis are given from the literature and from unpublished work.

Advances in Analysis of Volatiles in Gas Chromatography

Advances in analysis of volatiles by gas chromatography can be considered under six headings, namely:
1. calibration of standards;
2. detection, including use of specific detectors;
3. separation, including use of capillary and megabore columns;
4. headspace analysis, based on concentration equilibrium between a gaseous and non-gaseous phase;
5. pre-concentration, including trapping techniques and use of membranes; and
6. laboratory accreditation and quality assurance programs.

Calibration of standards

Inaccurate standards are a major source of error in analytical chemistry and the problem is especially severe in gas analyses because of problems in accurately diluting gases. The major recent advance is the provision of calibrated gas standards, at low concentrations, by organisations such as the Environmental Protection Agency. Provision of standards for calibration is an essential part of any quality assurance program (Ratliff 1993). 'Purge and trap' techniques (cf. 5 below) have also been used to obtain a range of concentrations from a primary standard (Rhoderick and Miller 1990). In this technique, different concentrations are obtained by purging gases at measured flow rates over different timed intervals. This enables calibration of machine response to different concentrations of volatile chemicals, including the fumigants methyl bromide and ethylene dibromide.

Nonetheless, standardised official procedures for calibration of gas standards are not yet readily available in one convenient book, though the review by Scudamore (1988) is an excellent step in this direction. Infra-red spectroscopy would seem to be a convenient method of checking concentrations of gas standards, in the same way as ultra-violet spectroscopy is used for liquids and solids, but there is no officially-validated procedure for fumigants.

Detection, including specific detectors

Specific detectors have been used for many years in the analysis of pesticides and fumigants. Table 1 lists the sensitivity of some detectors, with an indication of selectivity. Thus, the flame photometric detector, in the phosphorus mode, is capable of detecting 0.001 ng of phosphorus per second. This means that, if a phosphine peak can be passed through a detector in one second, one can detect 0.001 ng of phosphorus or 0.0011 ng of phosphine. The units of sensitivity (ngG/sec) indicate that sensitivity is related both to detector response and to resolution of the chromatography peak. However, because sensitivity in practice is limited by background 'noise' or interferences, it is often defined in terms of signal-to-noise ratio. For example, 'limits of detection' are typically given as a signal-to-noise ratio of two. In this instance, use of capillary columns (cf. 3 below) greatly reduces noise by better separation of interferences from the peak of interest.

The mass spectrometer is increasingly being used in analysis of volatiles. One obtains the whole spectrum, for purposes of identification, as well as the intensity of 'specific ions'. The machine sensitivity of the mass spectrometer varies with type of chemical, ionisation process and background interference. As an example, HCN and nitrogen have the same m/z values. Nonetheless, specific ion monitoring is very useful in reducing background noise, and thus increasing the signal-to-noise ratio. Currently, sample volume is limited with the mass spectrometer as the detector, principally because of the requirement for capillary columns. However, mass spectrometry is becoming routine in analysis of volatiles principally because it can function as a universal detector. Its

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Table 1.  Sensitivity of some GC detectors.

<table>
<thead>
<tr>
<th>Detector</th>
<th>Sensitivity ng/sec</th>
<th>Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flame Photometric (P)</td>
<td>0.001</td>
<td>P only</td>
</tr>
<tr>
<td>Flame Photometric (S)</td>
<td>0.1</td>
<td>S only&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Thermionic</td>
<td>0.0001</td>
<td>N&amp;P</td>
</tr>
<tr>
<td>Flame Ionisation</td>
<td>0.002</td>
<td>none</td>
</tr>
<tr>
<td>Electron Capture</td>
<td>0.0001</td>
<td>varied&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Photo-ionisation</td>
<td>0.01</td>
<td>varied</td>
</tr>
<tr>
<td>Conductivity (Hall)</td>
<td>0.005</td>
<td>N,S, halides</td>
</tr>
<tr>
<td>Specific ions Mass Spec</td>
<td>0.01</td>
<td>'all'&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Thermal Conductivity</td>
<td>0.3 ng/mL</td>
<td>'all'&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
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</table>

<sup>a</sup>response non-linear but area proportional to concentration squared
<sup>b</sup>linearity limited; easily contaminated
<sup>c</sup>not generally suitable for megabore columns
<sup>d</sup>note the different unit of concentration

use is often coupled with that of a specific detector, when extra sensitivity is required. It is frequently the case that mass spectrometry is used for confirmation of identity, and specific detectors for quantification, although the mass spectrometer can be used for quantitative analysis.

The electron capture detector is usually regarded as 'specific' for compounds containing halogens, or conjugated systems. However, it also responds to oxygen, and not to argon or nitrogen. This is useful because it is difficult to separate argon from oxygen, and argon interference is a problem in analysis of low oxygen atmospheres. Interestingly, there is little detailed information in stored-product literature on techniques for measuring low oxygen concentrations. The response of the electron capture detector to oxygen is shown in Figure 1, together with the response to several fumigants. Thus, the electron capture detector is useful for analysis of low oxygen concentrations, but it becomes saturated at higher oxygen concentrations, where less selective detectors, such as the thermal conductivity detector, can be used at concentration levels where the 'error' from argon is less important.

Specific detectors are, of course, not new but there are several detectors that are 'new' in the sense of being readily available for the first time. These include specific ion monitoring and the photo-ionisation detector. It is not the detectors alone, however, that have increased the limits of detection of volatiles but their integration with other techniques discussed below.

Separation, including use of capillary and megabore columns

Most sensitive methods of determining volatile chemicals now use capillary columns (internal diameter 0.32 mm) or 'megabore' columns (internal diameter 0.53 mm). Capillaries give better resolution but megabores have two advantages for gas analysis. First, they can be readily attached to chromatographs designed for packed columns. Second, because the flow of carrier gas is quite high, typically 10 mL/min, injection volume can be large (e.g. up to 1 mL). It should, however, be noted that some detectors require large quantities of carrier gas, so that replacement of packed columns with megabores requires a make-up gas. This appears to be especially a problem with flame-photometric detectors.

The laboratory of which I am a member routinely uses a GSQ megabore column (J & W 115–3432) for analysis of oxygen, phosphine, carbonyl sulphide, methyl chloride, carbon bisulphide, methyl bromide, and methyl isothiocyanate. The column also separates hydrogen cyanide, though greater sensitivity is obtained on the polar DBwax (J&W 125–7012) or on a column designed for acids, FFAP (J&W 125–3212). A typical chromatograph is shown in Figure 1a, which records the response of an electron-capture detector to oxygen, methyl bromide and carbon bisulphide. The small peaks are methyl isothiocyanate, which was the warning agent in the methyl bromide, and chloropicrin.

In Figure 1b the separation of phosphine from hydrogen cyanide using the megabore DBwax is shown. In this case, a thermionic specific detector was used (specific for nitrogen...
and phosphorus). The peak shape for hydrogen cyanide (the second peak) shows less tailing than that for phosphine (the first peak) because the column, DBwax, was selected for optimal detection of hydrogen cyanide. Conversely, the peak shape of phosphine on GSQ is better than that of hydrogen cyanide. Nonetheless, the peak shape of each fumigant in Fig. 1b is acceptable, whereas it would be difficult to obtain good peak shapes for phosphine and hydrogen cyanide on the same packed column. Figure 1c shows separation of methyl isothiocyanate and phenylethyl isothiocyanate on the same column. These chemicals were obtained from unfumigated canola, using a trapping technique (cf. 5).

In Figure 1 the retention times, in minutes, are recorded. It can therefore be seen that peak half-widths of most of the chemicals are 1–2 sec (except for methyl isothiocyanate and chloropicrin in Figure 1a, where, however, the peak width can be narrowed by higher column temperatures). Narrow peak widths are important for two reasons. First, as sensitivity is a function of mass/peak width (Table 1), reducing peak width increases sensitivity. Second, narrow peak width reduces the chance of interference.

The GSQ megabore column enables complete separation of the following fumigants: phosphine, hydrogen cyanide, carbonyl sulphide, carbon bisulphide, ethylene dichloride and chloropicrin, as well as oxygen, possible alteration products such as methyl chloride and hydrogen sulphide, and naturally occurring fumigants such as hydrogen cyanide, methyl isothiocyanate and other isothiocyanates.

**Headspace analysis**

‘Headspace analysis’ involves partitioning of a volatile chemical between a gaseous phase (air or nitrogen) and another phase (typically liquid), thus enabling determination of the volatile chemical in the non-gaseous phase from its concentration in the vapour phase. The method, which has been reviewed (Nunez et al. 1984), has become a standard method for analysis of volatile chemicals in water. Elimination of the use of solvents is one of the great advantages of this method.

Our laboratory has developed a variation of headspace analysis for determination of cyanide residues in grain (Vu and Desmarchelier, unpublished data). In this procedure, grain is placed with water in a sealed container and, after extraction is complete, a standard quality of dilute hydrochloric acid is injected and the cyanide in grain is determined from the concentration of hydrogen cyanide in the headspace. Analytical conditions include a DBwax megabore column and a specific detector (a thermionic detector, specific for nitrogen and phosphorus). This method is much quicker, and more sensitive, than the standard procedure (American Association of Cereal Chemists, 1983) which involves distillation of hydrogen cyanide from a solution of grain in boiling water, and determination of cyanide by argentometric titration.

A method has been developed for headspace analysis of methyl bromide (Greve and Hogendoorn 1979) in which the fumigant is first extracted into acetone, and methyl bromide is determined from the headspace concentration in a sealed container. This method can detect residues down to 10 ppb.

An old method used by Turtle for analysis of hydrogen cyanide, and subsequently modified (American Association of Cereal Chemists 1983), involved refluxing grain in water and trapping the volatile fumigant. In the case of hydrogen cyanide, the trap was aqueous alkali. This method of refluxing grain residues in water has been adapted to headspace analysis (Nunez et al. 1984) by Heikes (1987) and Heikes and Hopper (1986). In this procedure, grain or grain products are refluxed in water under a nitrogen stream, the volatiles are absorbed on a trap (cf 5 below), and taken up in a solvent (solvent desorption), an aliquot of which is injected into the chromatograph. The method detects several halogenated fumigants at detection limits in the range 0.1–6 ng/g (ppb) and carbon bisulphide at a detection limit of 12 ng/g. These detection limits are considerably below former ‘guideline’ maximum residue limits which were set at ‘detection limits’ of 0.1 ppm to 10 ppm. Nonetheless, trapping followed by thermal desorption would result in even greater sensitivity than the solvent desorption used by Heikes. The method, however, is unsuitable for methyl bromide because recoveries of freshly added fumigant are low.

The Association of Official Analytical Chemists has, for many years, called for a multi-residue determination of fumigants. There are a number of multi-residue methods that have been tested. These include purge and trap procedures (e.g. Heikes 1987) and solvent extraction (e.g. Daft 1987). In 1988 (Schmidt et al. 1988) the Association of Official Analytical Chemists recommended further work on the method of Daft (1987). The method involves extraction into acetone/water, back-extraction into iso-octane and determination by gas chromatography. This procedure enables detection of several fumigants down to parts per billion levels, but is unsatisfactory for methyl bromide and has not been tested on phosphine. However, a protocol for more detailed evaluation was considered acceptable by the Association but, as of 1992 ‘no further work has been accomplished toward initiating a collaborative study, although the Associate Referee plans to do so as time permits’ (Sawyer 1992). The method of Daft (1987) uses essentially old technology, which enables it to be conveniently used by pesticide laboratories, but it suffers the problems of all methods using solvent extraction, including disposal problems and possibilities of contamination and interference.

**Pre-concentration**

Volatile chemicals can be concentrated on ‘traps’. These are either chemical traps (e.g. charcoal, Tenax) or cold traps. Volatiles can be removed from chemical traps with a small quantity of solvents (solvent desorption) or with heat (thermal desorption). Recent advances involve direct coupling of a trap to a chromatograph, so that all the trapped material is eluted as one peak. [The intermediate step of ‘thermal focusing’ is also used whereby material is transferred from a larger to a smaller trap, and then onto the gas chromatograph, e.g. Burger et al. (1991) and Pankow et al. (1988). ‘Thermal focusing’ may employ several traps outside the gas chromatograph, or traps may be part of a multi-column system inside the chromatograph.]

Tangeman (1986) used such techniques to determine concentrations of carbonyl sulphide and carbon bisulphide in laboratory air. Gases were trapped on a tube containing Tenax at low temperatures and the tube was transferred to the injection port of the chromatograph. The detection limit of carbonyl sulphide was 1.2 ng/m³ (3 parts per trillion). This, incidentally, was less than one thousandth of the concentration in the laboratory air. Kallio and Shibamoto (1988) determined methyl bromide and chloropicrin at similar sensitivities, after trapping the gases on a cold capillary column and then rapidly heating the column, which was directly attached to the gas chromatograph.

Membranes have been used to concentrate volatile gases (Blanchard and Hardy 1985). The method avoids the problems associated with traps and proved useful for such fumigants as acrylonitrile, carbon tetrachloride and various halogenated alkanes.
Quality assurance programs

Laboratory accreditation and quality assurance programs have long been a part of the analysis of pesticides (e.g. Ratliff 1993, NATA 1992). Written procedures, interlaboratory trials, etc., are a regular part of the work of pesticide laboratories. The advantages of participation in interlaboratory trials is obvious from a wide literature and also obvious to anyone who has participated in such trials. How this might be done is discussed later in this paper.

Overview

Detectors are capable of determining chemicals at less than 0.001 ng/sec, columns can give resolution of a few seconds, and trapping techniques enable all the volatiles in large quantities of air to be focused into one concentrated injection. All these techniques use standard, purchasable, items. It appears that detection limits of 1 ng/g (1 ppb) on food and 1 ng/ml in air (1 ppt) are readily achievable, and even lower limits of detection seem possible.

As part of a general environmental program, regulatory authorities in the United States have validated procedures that detect parts per billion levels in foodstuffs, and parts per trillion levels in air. These are limits of detection much more sensitive than those published from stored-product laboratories. They are also below the 'guideline' levels (i.e. detection limits) of 100–1000 ppb that form part of food regulations. These detection limits have implications for the regulation of fumigants in foodstuffs. It is possible that the current 'residue-free' status of fumigants will not remain intact (presuming, for the sake of the argument, that it currently exists).

For several decades, workers in stored-product laboratories were the leading experts in analysis of fumigants. Without wishing to offend any individual, this is no longer the case as 'fumigant analysis' may now be considered to be a sub-set of the general topic of 'analysis of volatiles', where regulatory agencies in the USA are at the forefront in the development, or at least in the publication, of sensitive methods of analysis.

General Critique of Fumigant Analysis

Although much work has been done on fumigant analysis, there is still no general regulatory method validated by bodies such as the (International) Association for Official Analytical Chemistry, and particularly none that uses modern techniques. From the point of view of stored products, it is unfortunate that multi-residue procedures such as those of Heikes (1987) or Daft (1987) give poor recoveries of methyl bromide and do not include data on phosphine. Perhaps it is the responsibility of workers in stored-products to ensure that commonly-used fumigants are included in multi-residue methods of analysis.

Analysis of fumigant residues, like analysis of most insecticide residues, faces the fundamental problem of 'aged' or 'weathered' residues. That is, if one analyses residues of a chemical some time after application, one does not know the 'true' answer. In addition, recovery of freshly added or 'spiked' residues is insufficient evidence that the method works on 'aged' residues. In homogeneous media, such as air or water, the problem of aging is not important, as the mixture remains homogeneous. The problem is important in heterogeneous media, such as soils or grains, where chemicals migrate from sites with low binding energy to sites from which it is difficult to dislodge the chemical. General techniques to solve this problem have long been in use (e.g. Sandall 1959) and long applied to grain protectants (e.g. Desmarchelier et al. 1977). Such techniques require the use of several regimes which vary extraction conditions, and the 'true' answer is that obtained from several different procedures, provided no other procedure gives higher results. Gunther (1962) has claimed that results from procedures that rely on recovery of 'spiked' samples for validation are 'illusory'. On this criterion (which I personally fully support) most residue determination of fumigants, and perhaps all residue determination of fumigants, are 'illusory', i.e. the validation is satisfied by the procedures used merely describe the results obtained by the procedures, and not necessarily the 'true' value.

If one accepts the criteria of Sandall, Gunther and others, validation of methods of analysis of aged residues of fumigants requires obtaining the same answer by several different techniques. This is a methodology we employ at SGRL, but it is very time-consuming. It is also very rare that different methods give the same answer, to within the degree of precision claimed by each method.

Validation of analytical procedures by adhering to protocols such as those of Sandall (1959) is important to remove systematic errors of analysis. However, it is still possible for analytical errors to occur with valid analytical procedures. To detect, and ultimately to reduce, such 'random' errors, interlaboratory trials are standard practice in laboratories which determine pesticides. The advantages of such trials are well published (e.g. IUPAC 1978). I know of no such interlaboratory trials for grain fumigants, and certainly none in stored-product laboratories. It would certainly be useful to conduct such trials, which would either detect significant variations between laboratories or, in the event that we all obtained the same answer, give credence to results from different laboratories. A collaborative study on fumigant residues would be difficult, but not impossible. For a start, gaseous samples can be reliably maintained, as they are in samples certified by such agencies as the U.S. Environmental Protection Agency. Second, such gaseous samples could be used for 'spiked', i.e. freshly-added, samples in fumigant analysis. Third, it would be possible to transport 'aged' residues in sealed containers, ideally at low temperature, for analysis on a pre-determined date.

In summary, I believe that we have marvellous technology available for fumigant analysis, but that we have failed to take the necessary steps to validate our results. With currently available technology, it is feasible, and almost routine, to be able to detect 1 ppb on food and 1 ppt in air. However, if we accept the arguments of Sandall (1959) and Gunther (1962), and validation of their arguments for protectants on grain (Desmarchelier et al. 1977), we must doubt whether our procedures have been shown to give the 'true' answer and, in the absence of inter-laboratory check programs, whether existing procedures are accurately used.

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References


