



## Internal standards: A source of analytical bias for volatile organic analyte determinations

Michael H. Hiatt\*

U.S. Environmental Protection Agency, National Exposure Research Laboratory, Environmental Sciences Division, P.O. Box 93478, Las Vegas, NV 89193-3478, USA

### ARTICLE INFO

#### Article history:

Received 20 August 2010

Received in revised form 2 November 2010

Accepted 29 November 2010

Available online 5 December 2010

#### Keywords:

Volatile organic compounds

Internal standards

Bias

Biota

Soil

Water

Analyses

### ABSTRACT

The use of internal standards in the determination of volatile organic compounds as described in SW-846 Method 8260C introduces a potential for bias in results once the internal standards (ISTDs) are added to a sample for analysis. The bias is relative to the dissimilarity between the analyte and internal standard physical properties that influence how easily analytes are separated from a matrix and concentrated during analysis.

Method 5032 is a vacuum distillation procedure for extracting analytes from a sample for use with Method 8260C. Vacuum distillation is also incorporated within another GC/MS analytical procedure, Method 8261A. Method 8260C/5032 and Method 8261A are experimentally identical, however, Method 8261A uses internal standards differently by relating the recovery of each compound to its boiling point and relative volatility. By processing each analysis (water, soil, and biota) using both Method 8260C and Method 8261A, the two approaches are compared on the basis of analyte bias and the failure rate of the quality controls.

Analytes were grouped by how similar their boiling points and natural log of their relative volatilities (ln RVs) were to their Method 8260C recommended ISTDs. For the most similar analytes, the Method 8260C determinations yielded an average bias less than 10% and a failure to meet calibration criteria less than 7%. However, as the difference between analyte and ISTD became greater the bias increased to over 40% (matrix dependent) and its calibration failure rate approached 70%. In comparison, when the Method 8260C data were reprocessed as Method 8261A determinations, this trend for groupings was minimized with biases increasing from 6% to only 20% and the calibration failure rate went from 0% to 15%.

Published by Elsevier B.V.

### 1. Introduction

Volatile organic compounds (VOCs) make up a major group of compounds routinely monitored as environmental contaminants. RCRA SW-846 Method 8260C is the determinative protocol of choice with a pre-concentration protocol such as headspace (Method 5021) or purge-and-trap (Method 5030C) [1]. There are investigations addressing measurement uncertainty as analytical [2,3], sampling and sub-sampling errors [4], and their comparative importance [5]; however, for VOC determinations there is a potential bias rarely addressed. That is, the dissimilarity between analytes and their ISTDs can result in quite different behavior during analyses.

Method 8260C describes the use of GC/MS for quantification of analytes with guidance for quality control. The method uses internal standards that are added to samples to compensate for changes

in responses between a calibration standard and a sample by simply applying the relative change in internal standard response to the analyte response. This internal standard approach has been part of EPA methods for determining VOCs since being included in Method 624 for VOCs in drinking water [6]. Additional analytes, internal standards, and surrogates have since been added and now RCRA Method 8260C is applied to a wide range of analytes and for a plethora of matrices. Quality controls have been included in the method to insure that the behavior of analytes is uniform (calibration and continuing calibration criteria) and “matrix effects” are minimal (limits for relative response of internal standards and surrogate recoveries). When results deviate from these limitations, the results are considered unreliable and thus qualified as estimated values.

Method 8261A is another SW-846 method that can be used to determine VOCs [7]. Unlike Method 8260C, Method 8261A includes both vacuum distillation pre-concentration and quantitation as a single method. The vacuum distillation pre-concentration procedure described in Method 8261A is also described in Method 5032 for use with Method 8260C. Method 8261A and Methods 8260C/5032 are experimentally identical, except how the internal

\* Corresponding author. Tel.: +1 702 798 2381; fax: +1 702 798 2142.

E-mail address: [hiatt.mike@epa.gov](mailto:hiatt.mike@epa.gov)

standards are used. Rather than relating the response of an analyte to its recommended internal standard, Method 8261A relates the response of an analyte to its boiling point and water-to-air partitioning during distillation (relative volatility). The responses of internal standards are used to solve an algorithm that then is used to determine the recovery of each analyte as a function of the analyte's boiling point and relative volatility.

In two previous studies of Method 8261A, quality control limits were established to ensure that reporting errors were consistent with analytical errors for water [8] and soil [9] and these were used in this study. A critical step to accurately measure contaminants in soil was to ensure that analytes, surrogates, and internal standards were equilibrated with the soil. Attempts to minimize this effect through quick analysis (minimal time for soil and internal standard interactions) can lead to erroneous results [9]. While equilibration of internal standards with sample matrices is not addressed in the method protocols, all analyses used in this study included steps to equilibrate internal standards with the sample matrix, which included mixing internal standards in dry soil. It had been noted that there was a significant difference between Method 8260C/5032 and Method 8261A results when internal standards and analytes were added to soil after dilution with water [9]. This work evaluates the differences when internal standards and analytes are added to dry soil and equilibrated prior to analyses.

All analyses and calibrations were performed prior to implementing quality controls. Therefore, this study provides insight into the costs associated with those controls and if their implementation improves the accuracy of determinations. This work is generally applicable to other pre-concentration methods used with Method 8260C as water-to-air partitioning and boiling point would be major physical properties impacting recovery of volatiles. However, additional properties (not a major consideration during vacuum distillations) such as diffusion or partitioning between a trap phase and vapor phase need to be evaluated.

## 2. Experimental

### 2.1. Evaluation parameters

One point for comparison of Methods 8260C and 8261A use of internal standards was how their different determinations might deviate from known values for an analysis. This deviation was measured as absolute bias to eliminate cancellation of positive bias with any negative bias results. The magnitude of an average absolute bias for a collection of analytes is an indication how accurate determining an amount of analyte from the collection might be.

Another comparison of the differing internal standard approaches was how well analyses meet quality controls (QC criteria). The quality controls used for this evaluation were the standard deviation of generated calibration curves, comparison of a daily standard to the calibration curve (continuing calibration standard), the response of internal standards in samples to their standard response and the recovery of surrogates. The calibration criteria include both the limit for standard deviation of a calibration curve for an analyte and how much it's continuing calibration standard could deviate from its calibration (20% for Method 8260C and 40% for Method 8261A). The limits in how much the response of internal standards in a sample analysis can deviate from its calibration response (relative response) are provided in Table 1. The limits for recoveries of the various surrogate compounds in any sample are also provided in Table 1. When any of the above criteria was not met for an analyte determination the result is classified as not passing quality control (Fail QC).

For the vacuum distillation methods, Method 8261A and Method 5032, it was shown that the primary properties relating to

the recovery of compounds were boiling point and relative volatility [11]. In that work a few compounds were used as reference points to relate the ease of vacuum distilling compounds from water to partition coefficients. Because of the importance found for these properties, boiling points and relative volatilities were used to describe the physical differences between an analyte and the corresponding Method 8260C internal standard. Rather than look at each analyte individually, analyte results were grouped relative to how similar they are to their respective 8260C internal standard. Group 1 includes all analytes that have boiling points within 10 °C of their respective ISTD and the natural logarithm of a relative volatility (ln RV) within one of its ISTD. Group 2 includes analytes that have boiling points within 20 °C of its ISTD and ln RV within two of its ISTDs and not in Group 1, Group 3 includes the analytes that have boiling points within 50 °C and within three ln RV of the ISTD and not in Group 1 or 2, and Group 4 includes the remaining compounds.

### 2.2. Quantitation

Method 8260C internal standards and surrogates were as recommended in Method 8260C (Table 1). The compound tetrahydrofuran-*d*<sub>8</sub> was added as a surrogate for the more polar compounds. Reprocessing of the raw data generated for the previous Method 8261A studies [8,9] was possible as the recommended internal standards for Method 8260C were included in the Method 8261A studies with the exception that 1,2-dichlorobenzene-*d*<sub>4</sub> was substituted for 1,4-dichlorobenzene-*d*<sub>4</sub>. All raw data used to create Method 8261A calibration curves, continuing calibration and samples were reprocessed as per Method 8260C.

The internal standards and surrogates used with Method 8261A are as previously published [9]. The software used to perform calibration and quantitation was SMCReporter 4.2 available from EPA's web pages [10]. Limits for the internal standards and surrogate recoveries as described in the previous studies are presented in Table 1.

### 2.3. Samples

There were three sources of analyses used in this study. The water analyses had been generated in a previous study [8] investigating the reporting error associated with Method 8261A and these analyses were re-quantified as per Method 8260C/5032. The soil analyses were generated in study [9] reporting error associated with Method 8261A analyses of soil and these analyses were re-quantified as per Method 8260C/5032. The biota results were performed as part of this work and the instrumentation used described in Supplementary Information.

Table 2 lists the matrices studied. Water samples were modified with NaCl (0.1, 0.3, and 1 g), glycerin (0.1, 0.3, and 1 g), detergent (0.05, 0.1 and 0.2 mL), and peanut oil (0.1, and 0.3 g); 5 mL of the modified water samples was analyzed.

Soil samples consisted of acid-washed sand and three top soils (NV, GA, and OR). As minimal efforts to introduce internal standards were shown to have a potential to understate amounts of VOCs in soil samples [9], only the analyses in that study where the introduction of compounds to soil was rigorous to ensure interaction with the matrix were used. The preparation of soil samples included the additions of internal standards, surrogates, and analytes being added to 5 g dry soil then connected to the distiller (sealed) for overnight equilibration. 5 mL water and a magnetic stir bar were added the following day and the mixture stirred with use of a magnetic stirrer prior to distillation.

Biota samples were analyzed for this study to demonstrate severe matrix effects. These samples included 5 g grass, 2.5 g pine needles, 2.5 g rosemary, and 5 g muscle (tuna and shrimp). Fresh

Download English Version:

<https://daneshyari.com/en/article/10547781>

Download Persian Version:

<https://daneshyari.com/article/10547781>

[Daneshyari.com](https://daneshyari.com)