Method number:	1010
Version:	2.0
Target concentration:	1.0 ppm (1.8 mg/m^3)
OSHA PEL (TWA):	1.0 ppm (1.8 mg/m^3)
Action Level:	0.5 ppm (0.9 mg/m^3)
Excursion Limit:	5.0 ppm (9 mg/m^3) (15 min)
ACGIH TLV:	1.0 ppm (1.8 mg/m^3)
Procedure:	Samples are collected by drawing workplace air through sampling tubes containing hydrobromic acid coated carbon beads using personal sampling pumps. Samples are extracted with a mixture of water and a 1:1 (v/v) solution of acetonitrile/toluene. Analysis is performed by gas chromatography using an electron capture detector (GC-ECD).
Recommended sampling time and sampling rate:	
TWA:	240 min at 50 mL/min (12 L)
Excursion Limit:	15 min at 50 mL/min (0.75 L)
Reliable quantitation limit:	
TWA:	1.5 ppb (2.6 µg/m^3)
Excursion Limit:	23.5 ppb (42.3 µg/m^3)
Standard error of estimate at the target concentration:	5.3%
Special requirements:	Refrigeration of samples is recommended.
Status of method:	Fully validated method. This method has been subjected to the established evaluation procedures of the Methods Development Team.

March 2007 Yogi Shah

Revised March 2014 Michael Simmons

Methods Development Team
Industrial Hygiene Chemistry Division
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Sandy UT 84070-6406

1. General Discussion

For assistance with accessibility problems in using figures and illustrations presented in this method, please contact OSHA Salt Lake Technical Center (SLTC) at (801) 233-4900. These procedures were designed and tested for internal use by OSHA personnel. Mention of any company name or commercial product does not constitute endorsement by OSHA.

1.1 Background

1.1.1 History

OSHA Method 50[1] for ethylene oxide (EtO) was validated in 1985 and specifies collection of EtO using sampling tubes containing petroleum based charcoal coated with 10% hydrobromic acid (HBr). In this method EtO reacts with HBr to form 2-bromoethanol which is then extracted from the charcoal with dimethylformamide. An aliquot of the extracted 2-bromoethanol is further derivatized with heptafluorobutyrylimidazole (HFBI) and then analyzed by gas chromatography using electron capture detection (GC-ECD). The HFBI derivatization is performed to increase the sensitivity of the method.

OSHA Method 1010 was validated in 2007 when the supplier of HBr coated petroleum charcoal informed SLTC that the charcoal used to prepare the sampler was no longer available. Carbon beads, marketed by SKC, Inc. as Anasorb 747, was proposed for testing as a replacement base medium because it is a synthetic charcoal analogous to petroleum charcoal. Anasorb 747 was coated with 24% HBr, tested, and found to be an acceptable replacement for petroleum charcoal.

With the advancements in gas chromatography (GC) column technology and electron capture detector sensitivity the elimination of the HFBI derivatization was tested. The analysis in this method was performed by capillary column GC-ECD. Analytical results show sufficient sensitivity and precision that the derivatization with HFBI is no longer necessary.

The sampling medium listed in Version 2.0 of this method is the same as Version 1.0. Version 1.0 used methanol as the extraction solvent, and did not include an internal standard. It also did not include a procedure to neutralize or remove the excess acid from the extracted sample, resulting in corrosion of the GC inlet. For Version 2.0, the sample extraction procedure is similar to ASTM Test Method D5578[2], using a 1:1 (v/v) solution of acetonitrile/toluene as the extraction solution. However, instead of using sodium carbonate to neutralize the sample, 0.5 mL of water is added to the sample vial. The water forms a two-phase mixture partitioning the acid into the immiscible aqueous phase. Aliquots from the organic phase of the sample are then injected into the GC for analysis. Version 2.0 also introduces the use of an internal standard and requires the use of matrix matched standards. Matrix matching is performed to ensure that both the samples and standards have equivalent analyte distribution between the two phases.

All of the required validation tests were repeated for Version 2.0. The only validation data kept from Version 1.0 is the interference test using ethylene (Section 4.9).

[1] Cummins, K. Ethylene Oxide (OSHA Method 50), 1985. United States Department of Labor, Occupational Safety and Health Administration Web site. http://www.osha.gov/dts/sltc/methods/organic/org050/org050.html (accessed December 2013).
[2] ASTM Standard D5578, 2004, "Standard Test Method for Determination of Ethylene Oxide in Workplace Atmospheres (HBr Derivatization Method)," ASTM International, West Conshohocken, PA, 1994, DOI: 10.1520/D5578-04, www.astm.org.

1.1.2 Toxic effects (This section is for information only and should not be taken as the basis of OSHA policy.)

The basis for OSHA's ethylene oxide standard, 29 CFR 1910.1047, was a determination by OSHA "based on animal and human data, that exposure to EtO presents a carcinogenic, mutagenic, genotoxic, reproductive, neurologic and sensitization hazard to workers". This standard establishes an 8-hour time-weighted average (TWA) permissible exposure limit for occupational exposure to EtO of 1 ppm, an 8-hour TWA action level of 0.5 ppm, and a 15 min excursion limit of 5 ppm.[3]

Acute short-term exposure to EtO causes "nausea, headache, weakness, vomiting, drowsiness, incoordination, and irritation of the eyes, nose, throat, and lungs". Skin contact with EtO solutions can cause "severe dermatitis, blisters, edema, burns, and frostbite". Chronic exposure can cause "skin sensitization, numbing of the sense of smell, and susceptibility for respiratory infection".[4]

1.1.3 Workplace exposure

Most EtO is used in the production of other chemicals including ethylene glycol, polyethylene terphthalate polyester, nonionic surface active agents, glycol ethers, ethanolamines, and choline. Less than 1% of the annual U.S. production is used for sterilization in the health care and medical products industries.[5]

The National Institute for Occupational Safety and Health estimated that between 1981 and 1983 270,000 U.S. workers were potentially exposed to ethylene oxide. It was estimated that of this number, 22% were exposed to the gas and 78% to materials containing ethylene oxide.[6]

1.1.4 Physical properties and other descriptive information[7]

synonyms:	Dihydrooxirene; dimethylene oxide; 1,2-epoxyethane; ethane oxide; oxane; oxirane; alpha, beta-oxidoethane; ETO; EtO; EO
IMIS[8]:	1190
CAS number:	75-21-8
boiling point:	10.8 °C
melting point:	-111 °C
specific gravity:	0.871
molecular weight:	44.05
appearance:	colorless gas
vapor density:	1.5 (air = 1)
solubility:	soluble in water
vapor pressure:	146 torr at 20 °C
flammable limit:	3-100% by volume
flash point:	2% (w/w) in water -2 °C (closed cup)

[3] Occupational Exposure to Ethylene Oxide; Final Standard (Federal Register # 49:25734), 1984. U.S. Department of Labor, Occupational Safety and Health Administration Web site. http://www.osha.gov/pls/oshaweb/owadisp.show_document?p_table=FEDERAL_REGISTER&p_id=12438 (accessed December 2013).

[4] American Conference of Governmental Industrial Hygienists, Inc. *Documentation of the Threshold Limit Values and Biological Exposure Indices*, 7th ed.; Cincinnati, OH, 2001; Vol. 2, pp. Ethylene Oxide – 1 through Ethylene Oxide – 14.

[5] American Conference of Governmental Industrial Hygienists, Inc. *Documentation of the Threshold Limit Values and Biological Exposure Indices*, 7th ed.; Cincinnati, OH, 2001; Vol. 2, pp. Ethylene Oxide – 1 through Ethylene Oxide – 14.

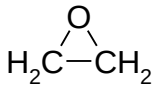
[6] International Agency for Research on Cancer, *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, Some Industrial Chemicals*, Vol. 60, Ethylene Oxide, IARC: Lyon, France, 1994, p 78.

[7] American Conference of Governmental Industrial Hygienists, Inc. *Documentation of the Threshold Limit Values and Biological Exposure Indices*, 7th ed.; Cincinnati, OH, 2001; Vol. 2, pp. Ethylene Oxide – 1 through Ethylene Oxide – 14.

[8] Ethylene Oxide (Chemical Sampling Information), 2012. United States Department of Labor, Occupational Safety and Health Administration Web site. http://www.osha.gov/dts/chemicalsampling/data/CH_240450.html (accessed December 2013).

molecular formula: C_2H_4O

structural formula:



This method was validated according to the OSHA SLTC "Validation Guidelines for Air Sampling Methods Utilizing Chromatographic Analysis"[9]. The Guidelines define analytical parameters, specify required laboratory tests, statistical calculations, and acceptance criteria. The analyte air concentrations throughout this method are based on the recommended sampling and analytical parameters. Air concentrations listed in ppm are referenced to 25 °C and 760 mmHg (101.3 kPa).

2. Sampling Procedure

All safety practices that apply to the work area being sampled should be followed. The sampling equipment should be attached to the worker in such a manner that it will not interfere with work performance or safety.

2.1 Apparatus

Samples are collected with 7-cm × 4-mm i.d. × 6-mm o.d. glass sampling tubes packed with two sections of HBr coated Anasorb 747 carbon beads (HBr-CB). The front section contains 100 mg and the back section contains 50 mg of HBr-CB. The sections are held in place and separated with glass wool plugs. For this evaluation, commercially prepared sampling tubes were obtained from SKC, Inc. (catalog no. 226-178).

A sampling tube holder is required to protect the worker from the sharp end of the glass sampling tube.

Samples are collected using a personal sampling pump calibrated to within ±5% of the recommended flow rate with the sampling device in-line.

2.2 Reagents

None required

2.3 Technique

Immediately before sampling, break off the ends of the flame-sealed tube to provide an opening approximately half the internal diameter of the tube. Wear eye protection when breaking the ends of the tube. Use sampling tube holders to minimize the hazard to the worker from the broken ends of the tubes. All tubes should be from the same lot.

The smaller section of adsorbent in the sampling tube is used as a back-up and is positioned nearest the sampling pump. Attach the tube holder to the sampling pump so that the adsorbent tube is in an approximately vertical position with the inlet facing down in the worker's breathing

[9] Eide, M.; Simmons, M.; Hendricks, W. Validation Guidelines for Air Sampling Methods Utilizing Chromatographic Analysis, 2010. United States Department of Labor, Occupational Safety & Health Administration Web site. http://www.osha.gov/dts/sltc/methods/chromguide/chromguide.pdf (accessed December 2013).

zone during sampling. Position the sampling pump, tube holder, and tubing so they do not impede work performance or safety.

Draw the air to be sampled directly into the inlet of the tube holder. The air being sampled is not to be passed through any hose or tubing before entering the sampling tube.

Sample for up to 240 min at 50 mL/min (12 L) to collect TWA (long term) samples.

Sample for 15 min at 50 mL/min (0.75 L) to collect excursion limit (short-term) samples.

After sampling for the appropriate time, remove the adsorbent tube and seal it with plastic end caps. Seal each sample end-to-end with a Form OSHA-21 as soon as possible.

Submit at least one blank sample with each set of samples. Handle the blank sample in the same manner as the other samples except draw no air through it.

Record sample air volume (liters), sampling time (minutes), and sampling rate (mL/min) for each sample, along with any potential interference on the Form OSHA-91A.

Submit the samples to the laboratory for analysis as soon as possible after sampling. If a delay is unavoidable, store the samples at refrigerator temperature (~4 °C). Ship any bulk samples separate from the air samples.

3. Analytical Procedure

Adhere to the rules set down in your laboratory's Chemical Hygiene Plan[10] (for instance: OSHA SLTC adheres to the rules set down in the OSHA SLTC Chemical Hygiene Plan). Avoid skin contact and inhalation of all chemicals and review all appropriate MSDSs before beginning the analytical procedure. Follow all applicable quality assurance practices established in your laboratory's internal quality system (for instance: OSHA SLTC follows the quality assurance practices established in the OSHA SLTC Quality Assurance Manual).

3.1 Apparatus

GC equipped with an ECD. An Agilent Model 7890 GC equipped with an automatic sample injector was used in this validation. An Agilent ChemStation was used to control the GC.

A GC column capable of separating 2-bromoethanol from the extraction solvent and internal standard. A Restek Rtx-Volatiles, 30-m × 0.25-mm i.d., (1-µm df) capillary column (catalog no. 10900) was used in this validation.

GC inlet liner. A Restek Sky 4.0 mm ID Low Pressure Drop Precision Inlet Liner w/wool (catalog no. 23309.1) was used in this validation.

In-line GC gas traps. In this validation an Agilent Big Universal Trap (catalog no. RMSHY-2) was used for purifying hydrogen gas. An Agilent Oxygen/Moisture Trap (catalog no. OT3-2) was used for purifying nitrogen gas.

An electronic integrator or other suitable means of measuring GC detector response. Waters Empower 3 Data System was used in this validation.

Water purifier. A Barnstead NANOpure Diamond system was used to produce 18.0 MΩ-cm DI water in this validation.

[10] Occupational Exposure to Hazardous Chemicals in Laboratories. *Code of Federal Regulations*, Part 1910.1450, Title 29, 2003.

Amber glass vials with PTFE-lined caps. Two-milliliter vials were used in this validation.

Dispenser or pipettes capable of delivering 1.0 and 0.5 mL of extracting solvent and water to prepare standards and samples.

Class A volumetric flasks. Two-milliliter and other convenient sizes for preparing standards.

Calibrated 10-µL syringe for preparing standards.

A mechanical shaker. An Eberbach mechanical shaker was used to extract the EtO derivative from HBr-CB in this evaluation.

3.2 Reagents

Toluene, [CAS no. 108-88-3], HPLC grade. The toluene used in this evaluation was 99.9% (lot no. 042461) purchased from Fisher Scientific (Pittsburg, PA).

Acetonitrile, [CAS no. 75-05-8], CHROMASOLV Plus grade. The acetonitrile used in this evaluation was ≥99.9% (lot no. 05735JH) purchased from Sigma Aldrich (Milwaukee, WI).

DI water, 18.0 MΩ-cm.

1-Bromo-4-fluorobenzene (ISTD), [CAS no. 460-00-4]. The 1-bromo-4-fluorobenzene used in this evaluation was 99% (lot no. 01127COV) purchased from Aldrich (Milwaukee, WI).

2-Bromoethanol, [CAS no. 540-51-2]. The 2-bromoethanol used in this evaluation was 97% (lot no. A0281954) purchased from Acros Organics (Morris Plains, NJ).

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Extraction solvent. Extraction solvent used in this validation consisted of 0.075 µL/mL of 1-bromo-4-fluorobenzene in a 1:1 (v/v) solution of acetonitrile/toluene. The 1-bromo-4-fluorobenzene was added as an internal standard. The extraction efficiency is affected by the extraction solvent, the internal standard, the sampling medium, and the technique used to extract the samples. Other reagents and techniques than described in this method can be used provided they are tested as specified in the validation guidelines.[11]

3.3 Standard preparation

Prepare a concentrated stock standard of 2-bromoethanol in the extraction solvent. Prepare matrix matched working standards by injecting microliter amounts of the concentrated stock standard into a 2-mL vial containing 100 mg of HBr-CB, 1 mL of extraction solvent, and 0.5 mL of 18.0 MΩ-cm DI water. Inject the stock standard into the water. Both the extraction solvent and water should be dispensed from the same dispensers used to extract the samples.

For example:

Prepare a concentrated stock standard by injecting 7.0 µL of 2-bromoethanol into a 1-mL volumetric flask containing about 0.5 mL of extraction solvent and then diluting to the mark with the extraction solvent.

$$(7.0\ \mu L \times 1.76\ mg/\mu L \times 0.97\ (purity) \times 44.05/124.97) / 1.00\ mL = 4.21\ mg/mL\ as\ EtO$$

[11] Eide, M.; Simmons, M.; Hendricks, W. Validation Guidelines for Air Sampling Methods Utilizing Chromatographic Analysis, 2010. United States Department of Labor, Occupational Safety & Health Administration Web site. http://www.osha.gov/dts/sltc/methods/chromguide/chromguide.pdf (accessed December 2013).

[Molecular weight of EtO is 44.05. Molecular weight of 2-bromoethanol is 124.97 and the density is 1.76 mg/µL.]

The stock standard will remain stable for three months if stored in a sealed glass vial and placed in a freezer when not in use.

Prepare a two times the target level working standard (2 ppm) by spiking 10.0 µL of the stock standard into a 2-mL vial containing 100 mg of HBr-CB taken from the front section of a sampling tube, 1 mL of extraction solvent, and 0.5 mL of 18.0 MΩ-cm DI water. Inject the stock standard into the water.

(10.0 µL × 4.21 mg/mL × 1 mL/1000 µL × 1000 µg/mg) = 42.1 µg/sample as EtO

Place the working standard vials on a shaker and shake for 60 min.

Note: only aliquots from the organic phase of the working standards are injected into the GC. This is done by adjusting the sample depth of the autosampler (Section 3.5). The water is added to extract the acid from the organic phase.

Bracket sample concentrations with standard concentrations. If sample concentrations fall outside the range of prepared standards, prepare and analyze additional standards to confirm instrument response.

3.4 Sample preparation

Remove the plastic end caps from the sample tube and carefully transfer the front section of HBr-CB to a 2-mL vial. Transfer the back-up section of HBr-CB to a separate 2-mL vial. Discard the glass tube along with the glass wool plugs.

Add 1.0 mL of the extraction solvent, followed by 0.5 mL of 18.0 MΩ-cm DI water to each vial and immediately seal with PTFE-lined caps.

Place the sample vials on a shaker and shake for 60 min.

Note: only aliquots from the organic phase of the sample are injected into the GC. This is done by adjusting the sample depth of the autosampler (Section 3.5). The water is added to extract the acid from the organic phase.

3.5 Analysis

3.5.1 Analytical conditions

<u>GC column conditions</u>	
column:	Restek Rtx-Volatiles, 30-m × 0.25-mm i.d., (1-µm df), (Restek catalog no. 10900, or equivalent)
flow:	1.2 mL/min (hydrogen)
column mode:	constant flow
initial average velocity:	36.7 cm/sec
<u>GC oven conditions</u>	
oven temperature:	70 °C (hold 6.5 min), ramp to 105 °C at 5 °C/min (hold 0 min), ramp to 250 °C at 35 °C/min (hold 1 min)
run time:	18.6 min

GC autosampler conditions

injection volume:	1 µL
sample depth:	**10 mm**

GC inlet conditions

liner:	Restek Sky 4.0 mm ID Low Pressure Drop Precision Inlet Liner w/wool (Restek catalog no. 23309.1, or equivalent)
temperature:	250 °C
split ratio (flow):	100:1 (120 mL/min)
septum purge:	3 mL/min (hydrogen)
total flow:	124.2 mL/min

GC detector conditions

temperature:	280 °C
const col + makeup flow:	60 mL/min (nitrogen)

GC analog out

zero:	0
range:	4

retention times

2-bromoethanol:	6.3 min
ISTD:	13.5 min

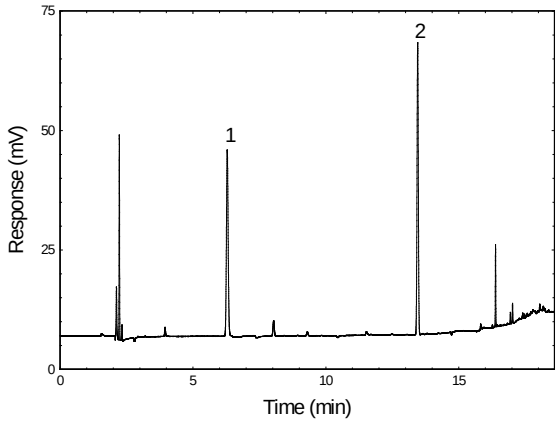

Figure 3.5.1. Chromatogram obtained at the target concentration with the recommended analytical conditions (1: 2-bromoethanol; 2: ISTD).

3.5.2 An internal standard calibration method is used. A calibration curve can be constructed by plotting ISTD-corrected response of standard injections versus micrograms of analyte per sample. Bracket the samples with freshly prepared analytical standards over a range of concentrations.

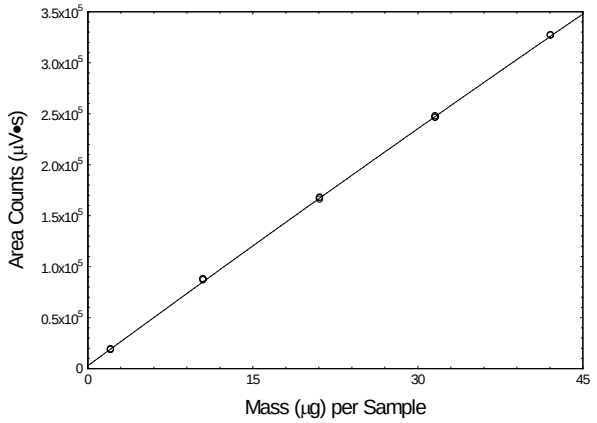

Figure 3.5.2. Calibration curve for EtO ($y = -5.5x^2 + 7916x + 2826$).

3.6 Interferences

3.6.1 Any compound that produces an ECD response, and has a similar retention time as the analyte or internal standard, is a potential interference. If any potential interferences were reported, they should be considered before samples are extracted. Generally, chromatographic conditions can be altered to separate interferences from the analyte.

3.6.2 When necessary, the identity of an analyte peak can be confirmed with additional analytical data or procedures (Section 4.10).

3.7 Calculations

The amount of EtO per sample is obtained from the appropriate calibration curve in terms of micrograms per sample, uncorrected for extraction efficiency. The back section is analyzed primarily to determine the extent of sampler saturation. If any analyte is found on the back section, it is added to the amount on the front section. If more than 20% of the total amount is found on the back section, report that the sampler may have been saturated on the Form OSHA-91B. This total amount is then corrected by subtracting the total amount (if any) found on the blank. The air concentration is calculated using the following formulas.

$$C_M = \frac{M}{VE_E}$$

where C_M is concentration by weight (mg/m^3)
M is micrograms per sample
V is liters of air sampled
E_E is extraction efficiency in decimal form

$$C_V = \frac{V_M C_M}{M_r}$$

where C_V is concentration by volume (ppm)
V_M is 24.46 (molar volume at NTP)
C_M is concentration by weight (mg/m^3)
M_r is molecular weight = 44.05

4. Method Validation

General instruction for the laboratory validation of OSHA sampling and analytical methods that employ chromatographic analysis is presented in "Validation Guidelines for Air Sampling Methods Utilizing Chromatographic Analysis"[12]. These Guidelines detail required validation tests, show examples of statistical calculations, list validation acceptance criteria, and define analytical parameters. Air concentrations listed in ppm are referenced to 25 °C and 760 mmHg (101.3 kPa).

4.1 Detection limit of the analytical procedure (DLAP)

The DLAP is measured as the mass of analyte introduced onto the chromatographic column. Ten analytical standards were prepared with approximately equal descending increments of analyte with the highest standard containing 84.2 ng/mL. This is the concentration that would produce a peak approximately 10 times the response of a blank at or near the chromatographic retention time of the analyte. These standards and the blank were analyzed with the recommended analytical parameters (1-µL injection with a 100:1 split). The data obtained were used to determine the required parameters (standard error of estimate and slope) for the calculation of the DLAP. Values of 993.3 and 31.9 were obtained for the slope and standard error of estimate respectively. The DLAP was calculated to be 0.096 pg.

Table 4.1
Detection Limit of the Analytical Procedure

concentration (ng/sample)	mass on column (pg)	area counts (µV·s)
0	0	0
8.4	0.084	0
16.8	0.168	101
25.3	0.253	133
33.7	0.337	244
42.1	0.421	340
50.5	0.505	441
59.0	0.590	502
67.4	0.674	607
75.8	0.758	709
84.2	0.842	783

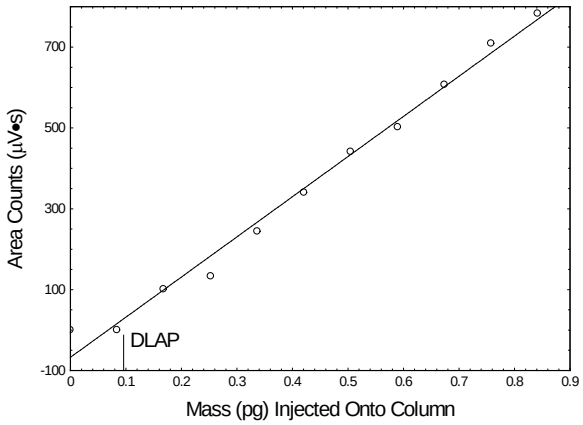

Figure 4.1. Plot of data to determine the DLAP (y = 993.3x - 67.4).

4.2 Detection limit of the overall procedure (DLOP) and reliable quantitation limit (RQL)

The DLOP is measured as mass per sample and expressed as equivalent air concentrations based on the recommended sampling parameters. Ten samplers were spiked with approximately equal descending increments of analyte, such that the highest sampler loading was 84.2 ng/sample. This is the amount spiked on a sampler that would produce a peak approximately 10 times the response of a sample blank at or near the chromatographic retention time of the analyte. These spiked samplers and the sample blank were analyzed with the recommended analytical parameters, and the data obtained used to calculate the required parameters (standard error of estimate and the slope) for the calculation of the DLOP. Values of 10.2 and 32.4 were obtained for the slope and standard error of estimate respectively. The DLOP was calculated to be 9.5 ng/sample (0.44 ppb or 0.79 µg/m^3).

[12] Eide, M.; Simmons, M.; Hendricks, W. Validation Guidelines for Air Sampling Methods Utilizing Chromatographic Analysis, 2010. United States Department of Labor, Occupational Safety & Health Administration Web site. http://www.osha.gov/dts/sltc/methods/chromguide/chromguide.pdf (accessed December 2013).

Table 4.2
Detection Limit of the Overall Procedure

mass per sample (ng)	area counts (μV·s)
0	0
8.4	74
16.8	199
25.3	247
33.7	359
42.1	493
50.5	512
59.0	578
67.4	733
75.8	807
84.2	823

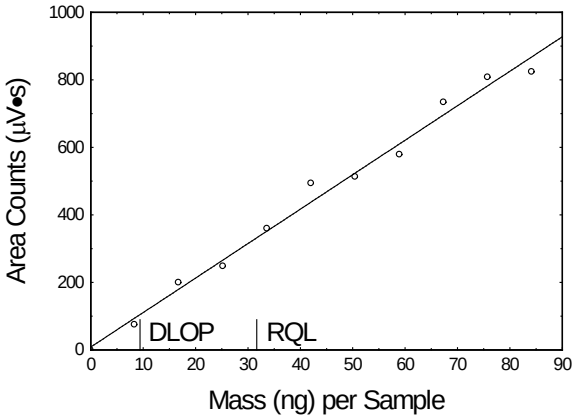

Figure 4.2.1. Plot of data to determine the DLOP/RQL (y = 10.2x + 8.6).

The RQL is considered the lower limit for precise quantitative measurements. It is determined from the regression line parameters that were obtained for the calculation of DLOP providing 75% to 125% of the analyte is recovered. The RQL is 31.7 ng/sample (1.5 ppb or 2.6 μg/m^3). Recovery at this concentration is 98.3%.

When excursion limit samples are collected, the air concentration equivalent to the reliable quantitation limit becomes larger. For example, the reliable quantitation limit is 23.5 ppb (42.3 μg/m^3) for EtO when 0.75 L is sampled.

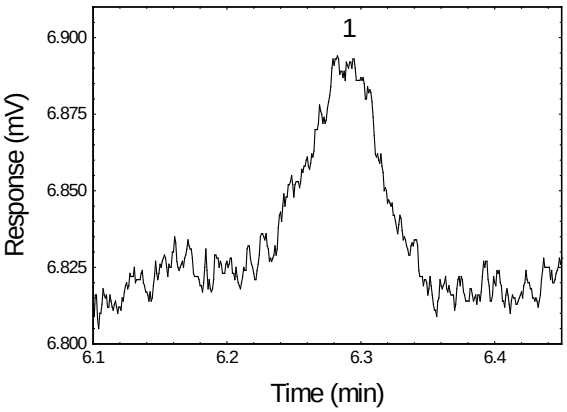

Figure 4.2.2. Chromatogram of the RQL (1: 2-bromoethanol).

4.3 Precision of the analytical method

The precision of the analytical method was measured as the mass equivalent to the standard error of estimate determined from the quadratic regression of data points from standards over a range that covers 0.1 to 2 times the target concentration. A calibration curve was constructed and shown in Section 3.5.2 from the three injections of five standards. The standard error of estimate was -0.18 μg.

Table 4.3
Instrument Calibration

×target concn	0.1×	0.5×	1.0×	1.5×	2.0×
(μg/sample)	2.1	10.5	21.1	31.6	42.1
area counts (μV·s)	18418	87722	165636	247626	327019
	18075	87339	167696	246244	326534
	18924	86572	167123	245838	326819

4.4 Storage stability test

Storage samples for ethylene oxide were prepared by sampling a dynamically generated controlled test atmosphere using the recommended sampling parameters. The concentration of EtO in the test atmosphere was the target concentration (1 ppm or 1.8 mg/m^3), and the relative humidity was 80% at 21 °C. Thirty-six storage samples were prepared. Six samples were analyzed on the day of generation. Fifteen samples were stored at reduced temperature (4 °C) and the other fifteen were stored in a closed drawer at ambient temperature (about 21 °C). At 2-5 day intervals, three samples were selected from each of the two storage sets and analyzed. Sample results are not corrected for extraction efficiency.

Table 4.4.1
Ambient Storage Test for EtO

time (days)	ambient storage recovery (%)		
0	94.7	95.0	95.5
3	93.3	93.3	92.3
7	88.1	88.6	90.2
10	87.9	91.7	95.3
14	92.3	92.6	91.9
17	89.3	89.5	92.9

Table 4.4.2
Refrigerated Storage Test for EtO

time (days)	refrigerated storage recovery (%)		
0	94.4	97.6	94.7
4	97.6	97.4	94.6
7	96.0	94.7	95.0
10	93.3	90.9	91.9
14	95.8	94.8	93.0
17	93.2	94.8	93.4

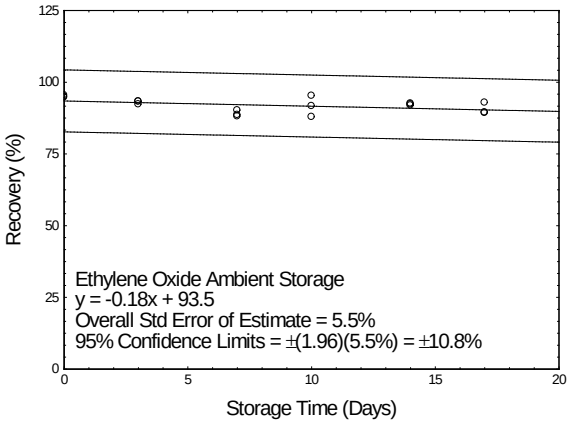

Figure 4.4.1. Ambient storage for EtO.

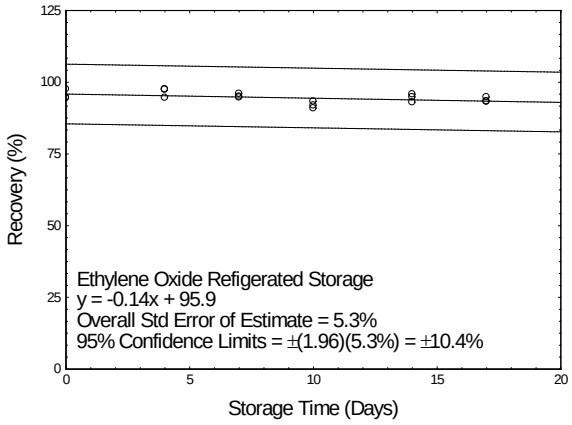

Figure 4.4.2. Refrigerated storage for EtO.

A storage test of samples collected from a low humidity atmosphere was also performed by sampling a dynamically generated controlled test atmosphere using the recommended sampling parameters. The concentration of EtO in the test atmosphere was two times the target concentration (2.0 ppm or 3.6 mg/m^3), and the relative humidity was 13% at 21 °C. Eighteen storage samples were prepared. Three samples were analyzed on the day of generation. Fifteen samples were stored in a closed drawer at ambient temperature (about 21 °C). At 2-5 day intervals, three samples were selected from the storage set and analyzed. Sample results are not corrected for extraction efficiency.

Table 4.4.3
Ambient Storage Test for EtO
(Low Humidity)

time (days)	ambient storage recovery (%)		
0	94.6	90.0	95.6
3	92.1	92.8	92.7
7	87.8	90.9	90.4
10	92.8	93.2	97.8
14	91.0	90.4	89.1
17	87.1	85.5	84.2

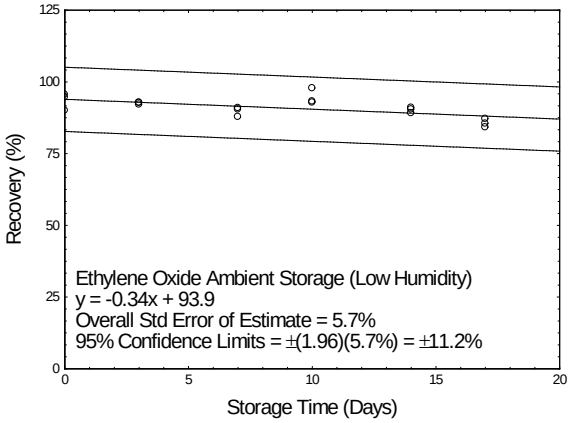

Figure 4.4.3. Low humidity ambient storage for EtO.

Low humidity storage is not normally performed, but was for this validation because of a decrease in recovery of 2-bromoethanol in an earlier OSHA method for EtO when sampling low humidity atmospheres[13]. A significant loss of analyte was not detected when samples were collected and stored on HBr-CB after sampling a low humidity atmosphere.

4.5 Precision (overall procedure)

The precision of the overall procedure at the 95% confidence level is obtained by multiplying the overall standard error of estimate by 1.96 (the z-statistic from the standard normal distribution at the 95% confidence level). Ninety-five percent confidence intervals are drawn about the regression lines in the storage stability figures shown in Section 4.4.

The precision of the overall procedure at the 95% confidence level for the reduced temperature (4 °C) 17-day storage test (at the target concentration) is ±10.4%. It was obtained from the overall standard error of estimate (5.3%) of the data shown in Figure 4.4.2. It contains an additional 5% for sampling pump error.

The recovery of EtO from samples used in a 17-day storage test remained above 93.5% when the samples were stored at 4 °C.

4.6 Reproducibility

Six samples were prepared by sampling a dynamically generated controlled test atmosphere similar to that used in the collection of the storage samples. The concentration of EtO in the test atmosphere was the target concentration (0.952 ppm or 1.71 mg/m^3), and the relative humidity was 81% at 22.1 °C. The samples were submitted to the OSHA Salt Lake Technical Center for analysis. The samples were analyzed after being stored for twelve days at 4 °C. Sample results were corrected for extraction efficiency. No sample result for EtO had a deviation greater than the precision of the overall procedure determined in Section 4.5.

Table 4.6
Reproducibility Data for EtO

theoretical (µg/sample)	recovered (µg/sample)	recovery (%)	deviation (%)
20.6	21.6	104.9	4.9
20.5	20.8	101.5	1.5
20.1	20.1	100.0	0.0
20.3	21.1	103.9	3.9
20.1	20.9	104.0	4.0
19.7	20.4	103.6	3.6

[13] Cummins, K. Ethylene Oxide (OSHA Method 50), 1985. Unites States Department of Labor, Occupational Safety and Health Administration Web site. http://www.osha.gov/dts/sltc/methods/organic/org050/org050.html (accessed 2013).

4.7 Sampler capacity

For TWA sampling the sampling capacity of the front section of an HBr-CB sampling tube was tested by sampling a test atmosphere containing twice the PEL of EtO (1.95 ppm or 3.51 mg/m^3) with a relative humidity of 82% at 22 °C. Sampling trains were prepared by removing the back sections of three sampling tubes and connecting each front section tube to a complete HBr-CB sampling tube. Sampling was begun and the back tubes were changed after sampling for 4 and 6 hours at 50 mL/min. Breakthrough, defined as 5% of the analyte passing through the front section of the tube, did not occur. This test was repeated with a relative humidity of 20% for 4 hours and no analyte was detected in the back tubes.

Table 4.7.1
Breakthrough of EtO From the Front Section of Sampler at 4 and 6 Hours at 50 mL/min (82% Relative Humidity)

test no.	air vol (L)	sampling time (min)	downstream conc (mg/m^3)	break-through (%)
1	12.1	240	0.076	2.22
	18.2	360	0.093	2.70
2	12.2	240	0.085	2.45
	18.3	360	0.107	3.09
3	12.1	240	0.056	1.63
	18.2	360	0.074	2.13

For TWA sampling the sampling capacity of the front section of an HBr-CB sampling tube was also tested by sampling a test atmosphere containing twice the PEL of EtO (1.98 ppm or 3.57 mg/m^3) with a relative humidity of 81% at 22 °C. Sampling trains were prepared by removing the back sections of three sampling tubes and connecting each front section tube to a complete HBr-CB sampling tube. Sampling was begun and the back tubes were changed after sampling for 8 and 10 hours at 50 mL/min. Breakthrough was not observed.

Table 4.7.2
Breakthrough of EtO From the Front Section of Sampler at 8 and 10 Hours at 50 mL/min (81% Relative Humidity)

test no.	air vol (L)	sampling time (min)	downstream conc (mg/m^3)	break-through (%)
1	24.6	480	0.070	1.93
	30.7	600	0.085	2.36
2	24.6	480	0.093	2.60
	30.7	600	0.114	3.16
3	24.4	480	0.107	2.97
	30.5	600	0.129	3.59

For excursion sampling the sampling capacity of the front section of an HBr-CB sampling tube was tested by sampling a test atmosphere containing 5 ppm of EtO (9.0 mg/m^3) with a relative humidity of 80% at 21 °C. Sampling trains were prepared by removing the back sections of three sampling tubes and connecting each front section tube to a complete HBr-CB sampling tube. Sampling was performed for 15 min at 200 mL/min. Breakthrough was observed. This test was repeated with a relative humidity of 20% and no analyte was detected in the back tube.

Table 4.7.3
Breakthrough of EtO From the Front Section of Sampler at 200 mL/min for 15 min (80% Relative Humidity)

test no.	air vol (L)	sampling time (min)	downstream conc (mg/m^3)	break-through (%)
1	2.92	15	1.26	14.0
2	3.00	15	1.32	14.8
3	2.94	15	1.31	14.6

For excursion sampling the sampling capacity of the front section of an HBr-CB sampling tube was further tested by sampling a test atmosphere containing 5 ppm of EtO (9.0 mg/m^3) with a relative humidity of 80% at 21 °C. Sampling trains were prepared by removing the back sections of three sampling tubes and connecting each front section tube to a complete HBr-CB sampling tube. Sampling was performed for 15 min at 50 mL/min. No breakthrough was detected.

Table 4.7.4
Breakthrough of EtO From the Front Section of Sampler at 50 mL/min for 15 min (80% Relative Humidity)

test no.	air vol (L)	sampling time (min)	downstream conc (mg/m^3)	break-through (%)
1	0.77	15	0.00	0.00
2	0.77	15	0.00	0.00
3	0.75	15	0.00	0.00

In summary, for TWA sampling at a sampling rate of 50 mL/min, a breakthrough of over 5% of the upstream concentration was never exceeded even after 10 hours of sampling; therefore, sampler capacity was never exceeded. The recommended sampling time is 4 hours at 50 mL/min. For excursion sampling, at a sampling rate of 50 mL/min for 15 min, no breakthrough was observed. The recommended sampling time is 15 min at 50 mL/min.

4.8 Extraction efficiency and stability of extracted samples

The extraction efficiency is affected by the extraction solvent, the internal standard, the sampling medium, and the technique used to extract the samples. Other reagents and techniques than described in this method can be used provided they are tested as specified in the validation guidelines.[14]

Extraction efficiency

The extraction efficiency of EtO was determined by liquid-spiking the front section of four samplers at each concentration level with 2-bromoethanol. These samples were stored overnight at ambient temperature and then analyzed. The overall mean extraction efficiency, over the working range of 0.1 to 2 times the target concentration, was 96.7%. The extraction efficiency at the RQL was 100.4%. The presence of water had no significant effect on extraction efficiency. The extraction efficiency of the 50 mg back section (B section) of the HBr-CB was also tested. At the target concentration the difference in extraction efficiency between the 100 mg front section and the 50 mg back section was 3.5%. The extraction efficiencies for the RQL, wet, and back section are not included in the overall mean. Wet media were prepared by drawing humid air (80% relative humidity at 21 °C) at 50 mL/min for 240 min. The data obtained are shown in Table 4.8.1.

[14] Eide, M.; Simmons, M.; Hendricks, W. Validation Guidelines for Air Sampling Methods Utilizing Chromatographic Analysis, 2010. United States Department of Labor, Occupational Safety & Health Administration Web site. http://www.osha.gov/dts/sltc/methods/chromguide/chromguide.pdf (accessed December 2013).

Table 4.8.1
Extraction Efficiency of EtO

level × target concn	µg per sample	sample number 1	2	3	4	mean
0.1	2.1	99.6	95.1	95.6	95.2	96.4
0.25	5.1	97.6	96.1	96.6	97.6	97.0
0.5	10.5	98.6	97.5	96.0	97.0	97.3
1.0	21.0	95.5	95.9	96.2	96.3	96.0
1.5	31.6	97.2	96.7	96.5	96.6	96.8
2.0	42.1	97.8	95.3	96.6	96.1	96.5
RQL	0.034	105.5	98.4	96.9	100.8	100.4
1.0 (wet)	21.0	94.1	94.2	95.7	95.1	94.8
1.0 (B section)	21.0	99.2	101.1	98.9	98.7	99.5

Stability of extracted samples

The stability of extracted samples was examined by reanalyzing the target concentration samples 24, 48, and 72 hours after the initial analysis. After the original analysis was performed two vials were recapped with new septa which were replaced after each reanalysis. The remaining two vials retained their punctured septa throughout the test. All samples were allowed to stand in the autosampler tray at ambient temperature. The samples were reanalyzed with freshly prepared standards. Diff is the difference between the initial analysis and the subsequent analysis. Each septum was punctured four times for each injection. The data obtained are shown in Table 4.8.2. Sample results are not corrected for extraction efficiency.

Table 4.8.2
Stability of Extracted Samples for EtO

punctured septa replaced							punctured septa retained						
initial (%)	24 h (%)	diff (%)	48 h (%)	diff (%)	72 h (%)	diff (%)	initial (%)	24 h (%)	diff (%)	48 h (%)	diff (%)	72 h (%)	diff (%)
96.2	96.3	0.1	99.1	2.9	99.4	3.2	95.5	95.4	-0.1	97.7	2.2	97.6	2.1
96.3	99.8	3.5	97.7	1.4	98.0	1.7	95.9	96.1	0.2	98.8	2.9	98.8	2.9
			mean							mean			
96.2	98.0	1.8	98.4	2.2	98.7	2.5	95.7	95.8	0.1	98.2	2.5	98.2	2.5

4.9 Sampling interferences

The tested sampling interferences had no significant effect on the ability of HBr-CB beads to collect or retain EtO.

Retention

Retention was tested by sampling a dynamically generated controlled test atmosphere containing two times the target concentration (1.96 ppm or 3.53 mg/m^3) of EtO at 81% relative humidity and 22 °C. The test atmosphere was sampled with six samplers at 50 mL/min for 60 min. Sampling was discontinued and the samplers were separated into two sets of 3 samplers each. The generation system was flushed with contaminant-free air. Contaminant-free air is laboratory conditioned air at known relative humidity and temperature but without any added chemicals except water. One set of samplers was set aside (first set). Sampling was resumed with the

Table 4.9
Retention of EtO

set	recovery (%) 1	2	3	mean
first	97.0	99.0	97.1	97.7
second	95.5	95.3	95.5	95.4
second/first				102.4

second set of three samples and contaminant-free air at 82% relative humidity and 22 °C at 50 mL/min for 180 min. All six samplers were analyzed and the data obtained are shown in Table 4.9.

Low humidity

The effect of low humidity was tested by sampling a dynamically generated controlled test atmosphere containing two times the target concentration (2.0 ppm or 3.60 mg/m^3) of EtO at 20% relative humidity and 22 °C. The test atmosphere was sampled with three samplers at 50 mL/min for 240 min. All of the samples were immediately analyzed. Sample results were 102.8%, 103.8%, and 104.6% of theoretical.

Low concentration

The effect of low concentration is normally tested by sampling a dynamically generated controlled test atmosphere, containing about 0.1 times the target concentration, using the recommended sampling parameters. For EtO this would be a 0.1 ppm (0.18 mg/m^3) test atmosphere, sampling for 240 min at 50 mL/min, resulting in a sample loading of 2.16 ug. The test atmosphere, described in Section 4.11, was only capable of generating down to 0.36 ppm (0.65 mg/m^3) of EtO. To get the equivalent loading, a dynamically generated controlled test atmosphere containing 0.36 ppm (0.65 mg/m^3) of EtO, at 80% relative humidity and 22 °C, was sampled with three samplers for 60 min at 50 mL/min. The generation system was then flushed with contaminant-free air. Contaminant-free air is laboratory conditioned air at known relative humidity and temperature but without any added chemicals except water. Sampling was resumed for an additional 180 min at 50 mL/min, at 80% relative humidity and 22 °C. This resulted in a sample loading of 1.94 µg of EtO. All of the samples were immediately analyzed. Sample results were 95.5%, 96.6%, and 93.3% of theoretical.

Chemical interference (From Version 1.0)

Three HBr-CB sampling tubes were used to sample an atmosphere containing 1 ppm (1.8 mg/m^3) EtO and 80% relative humidity at 23 °C at 50 mL/min for 240 min. The sampling media were transferred to glass vials and each vial was spiked with 2 mL of 25 ppm ethylene gas. All of the samples were analyzed after 2 days. The recoveries were 99.9%, 104.6% and 97.8% of theoretical. This test showed that ethylene gas did not form a reaction product with HBr that interfered with the analysis of EtO.

4.10 Qualitative analysis

When necessary, the identity or purity of an analyte peak can be confirmed by GC/MS or by another analytical procedure. For the levels analyzed in this method the use of GC/MS with selected ion monitoring (SIM) is recommended. A mass spectrum, obtained using the analytical conditions described below, is shown in Figure 4.10.

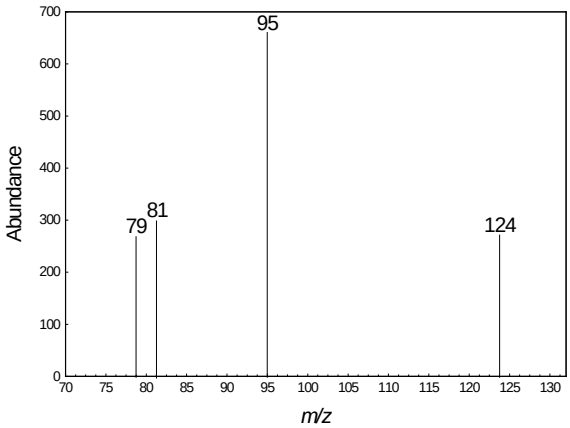

Figure 4.10. Mass spectrum of 2-bromoethanol using SIM.

Analytical conditions

GC column conditions

column:	Restek Rtx-Volatiles, 30-m × 0.25-mm i.d., (1-µm df), (Restek catalog no. 10900, or equivalent)
flow:	0.96 mL/min (helium)
column mode:	constant flow
initial average velocity:	36.1 cm/sec

GC oven conditions

oven temperature:	70 °C (hold 6.66 min), ramp to 105 °C at 4.88 °C/min (hold 0 min), ramp to 250 °C at 35 °C/min (hold 1 min)
run time:	18.97 min

GC autosampler conditions

injection volume:	1 µL
sample depth:	**10 mm**

GC inlet conditions

liner:	Restek Sky 4.0 mm ID Low Pressure Drop Precision Inlet Liner w/wool (Restek catalog no. 23309.1, or equivalent)
temperature:	250 °C
split ratio (flow):	100:1 (96 mL/min)
septum purge:	3 mL/min (helium)
total flow:	99.96 mL/min

MS acquisition parameters

mode:	EI
solvent delay:	5.00 min
EMV mode:	gain factor
gain factor:	1.00
MS source:	250 °C
MS quad:	200 °C
MSD transfer line:	250 °C

SIM parameters:

mass (*m/z*)	dwell (ms)
79	75
81	75
95	75
124	75

retention times

2-bromoethanol:	6.4 min
ISTD:	13.9 min

4.11 Generation of test atmospheres

A test atmosphere generator, as diagramed in Figure 4.11, was set up in a walk-in hood. House air was regulated using a Miller Nelson Model 401 Flow-Temperature-Humidity Control System. A measured flow of a certified standard of EtO gas was introduced into a measured flow of dilution air coming from the Miller Nelson control system. The EtO gas and dilution air flowed into a mixing chamber (76-cm × 15-cm) and then into a sampling chamber (56-cm × 9.5-cm). Samples were collected through sampling ports on the sampling chamber. Temperature and humidity were measured near the exit of the sampling chamber using a Vaisala HUMICAP® Hand-Held Humidity and Temperature Meter HM70. The certified EtO standard (1000 ppm in nitrogen) used in this validation was purchased from Praxair (Geismar, LA).

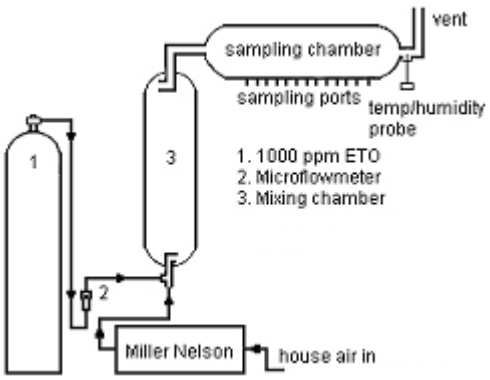

Figure 4.11. Diagram of apparatus used to generate EtO test atmospheres.