

A fast, cost-effective HS-GC-FID method for the analysis of ethylene oxide in surgical-style face masks

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Goal

To develop a fast, cost-effective analytical method suitable for the analysis of residual ethylene oxide and 2-chloroethanol in face masks using static headspace (HS) gas chromatography (GC) with flame ionization detection (FID).

Introduction

Face masks are ubiquitously worn by medical staff while performing duties such as surgery in order to reduce the risk of infection to the patient and to protect themselves from splashes of bodily fluids.¹ With the recent spread of Covid-19, many governments around the world have made the wearing of some form of face covering compulsory. Surgical face masks, along with other medical devices, are required to be sterilized to destroy bacteria and viruses. Steam could be used for sterilization, however many medical devices are sensitive to heat or moisture, meaning that steam is not always suitable.² In some



parts of the world, sterilization using ethylene oxide is commonly utilized due to its ability to alter pathogen DNA.³ However, in other countries and in parts of the US, the use of ethylene oxide as a sterilizing agent has not been approved by the US Department of Labor's Occupational Safety and Health Administration (OSHA) to sterilize filtering face piece respirators.⁴ Moreover, the Centers for Disease Control & Prevention (CDC) states that "Ethylene oxide is not recommended for cleaning filtering face piece respirators as it may be harmful to the wearer." Ethylene oxide is known to be toxic, flammable, and carcinogenic and is easily absorbed by many materials. Exposure to ethylene oxide can lead to irritation, central nervous system depression, spontaneous abortion, and various cancers.³

Due to these significant health effects, it is important that no ethylene oxide traces are found in the face masks or other respiratory devices that were potentially subjected to sterilization with this agent. To ensure that no ethylene oxide is present in the final product, mechanical aeration is usually performed over a period of 8 to 12 hours at a temperature of 50–60 °C. During the ethylene oxide sterilization process, it is also possible to form 2-chloroethanol.⁵ 2-chloroethanol is flammable and fatal if swallowed, comes in contact with skin, or is inhaled. To ensure that the ethylene oxide and 2-chloroethanol have been sufficiently removed, testing of the devices needs to be performed. GBT 16886.7-2015⁶ and ISO 10993-7⁷ both set limits for ethylene oxide and 2-chloroethanol in a variety of different materials, and GB 19083-2010⁸ describes a method for the analysis of ethylene oxide using GC-FID with a packed column setting a specific limit of 10 µg/g in face masks.

In the experiments described in this application note, a cost-effective and sensitive, capillary column, GC-FID analytical method for the analysis of ethylene oxide and 2-chloroethanol in face masks is demonstrated.

Experimental

Instrument and method setup

The Thermo Scientific™ TriPlus™ 500 headspace autosampler was coupled to a Thermo Scientific™ TRACE™ 1310 gas chromatograph fitted with a Thermo Scientific™ Instant Connect FID. The TriPlus 500 HS autosampler is directly connected to the analytical column, bypassing the GC inlet, significantly reducing the sample path and optimizing the sample transfer. Chromatographic separation was achieved using a Thermo Scientific™ TraceGOLD™ TG-1MS 30 m x 0.32 mm x 3 µm column (P/N 26099-4840). Nitrogen was used as the carrier gas and to pressurize the headspace vials. This significantly reduces analysis costs as compared to traditional, more expensive carrier gases such as helium, which can cost 50 times more than nitrogen. Full instrument conditions can be found in Table 1.

Standards and samples preparation

Ethylene oxide and 2-chloroethanol solutions in methanol were purchased from Sigma-Aldrich and diluted in deionized water to obtain mixed spiking solutions in the range 0.2 µg/mL to 20 µg/mL ethylene oxide and 1 µg/mL to 20 µg/mL 2-chloroethanol. 1 mL of spiking solution and

Table 1a. GC instrument conditions

| TRACE 1310 GC parameters | | | |
|--------------------------|---|-------------------|-----------------|
| Carrier gas | N ₂ | | |
| Column flow | 4 mL/min constant flow | | |
| Inlet mode | Split | | |
| Split flow | 40 mL/min | | |
| Column | TraceGOLD TG-1MS, 30 m × 0.32 mm × 3 µm | | |
| Oven temp. program | Rate (°C/min) | Target temp. (°C) | Hold time (min) |
| Temperature 1 | - | 50 | 1.5 |
| Temperature 2 | 40 | 280 | 1 |
| Run time | 8.25 min | | |
| FID temperature | 290 °C | | |
| Hydrogen flow | 35 mL/min | | |
| Air flow | 350 mL/min | | |
| Makeup gas flow | 40 mL/min | | |
| Data collection rate | 10 Hz | | |

Table 1b. HS autosampler parameters

| TriPlus 500 HS autosampler parameters | |
|---------------------------------------|----------|
| Vial incubation temperature | 85 °C |
| Vial incubation time | 30 min |
| Vial shaking | Medium |
| Vial pressurization mode | Pressure |
| Vial pressure | 150 kPa |
| Vial pressure equilibration time | 0.20 min |
| Loop/sample path temperature | 110 °C |
| Loop pressure | 100 kPa |
| Loop equilibration time | 0.20 min |
| Injection mode | Standard |
| Injection time | 0.10 min |

4 mL of deionized water were added to 20 mL headspace vials (P/N 20-CV) to obtain calibration standards in the range 0.2–20 µg/g ethylene oxide and 1–15 µg/g 2-chloroethanol. The vials were sealed with magnetic caps (P/N 20-MCBC-ST3) prior to analysis.

Surgical-style face masks were sourced from two different manufacturers and were cut into small pieces, approximately 1 cm². Unspiked samples were prepared in duplicate by adding 1 g of face mask and 5 mL of deionized water to a 20 mL headspace vial before sealing with magnetic caps. Spiked samples were prepared in duplicate at two different levels by adding 1 g of face mask, 1 mL of spiking solution, and then 4 mL of deionized water.

Data acquisition, processing, and reporting

The data were acquired, processed, and reported using the Thermo Scientific™ Chromeleon™ Chromatography Data System (CDS) software, version 7.3. Chromeleon CDS provides comprehensive system control, using the one software for data acquisition, real time data processing, and customized reporting, with all calculations performed within the software. Simplified Thermo Scientific™ eWorkflows™ allow for seamless implementation of methods into any laboratory, delivering effective data management, sample integrity, and full traceability. Chromeleon CDS also offers the option to scale up the entire analytical process in the laboratory from a single workstation to an enterprise environment.

Results and discussion

Chromatography

Consistent retention times, Gaussian peak shape, and repeatable peak areas are essential to provide reliable identification and quantitation. Examples of the achieved chromatographic separation are reported in Figures 1 and 2. Analyte peaks have chromatographic resolution >2.0 and peak asymmetry factors (AF) between 0.9 and 1.1, both calculated using the EP method, with all analytes eluting in <2.2 minutes. A total run time of 8.25 minutes was used to ensure that all compounds extracted from the sample were eluted.

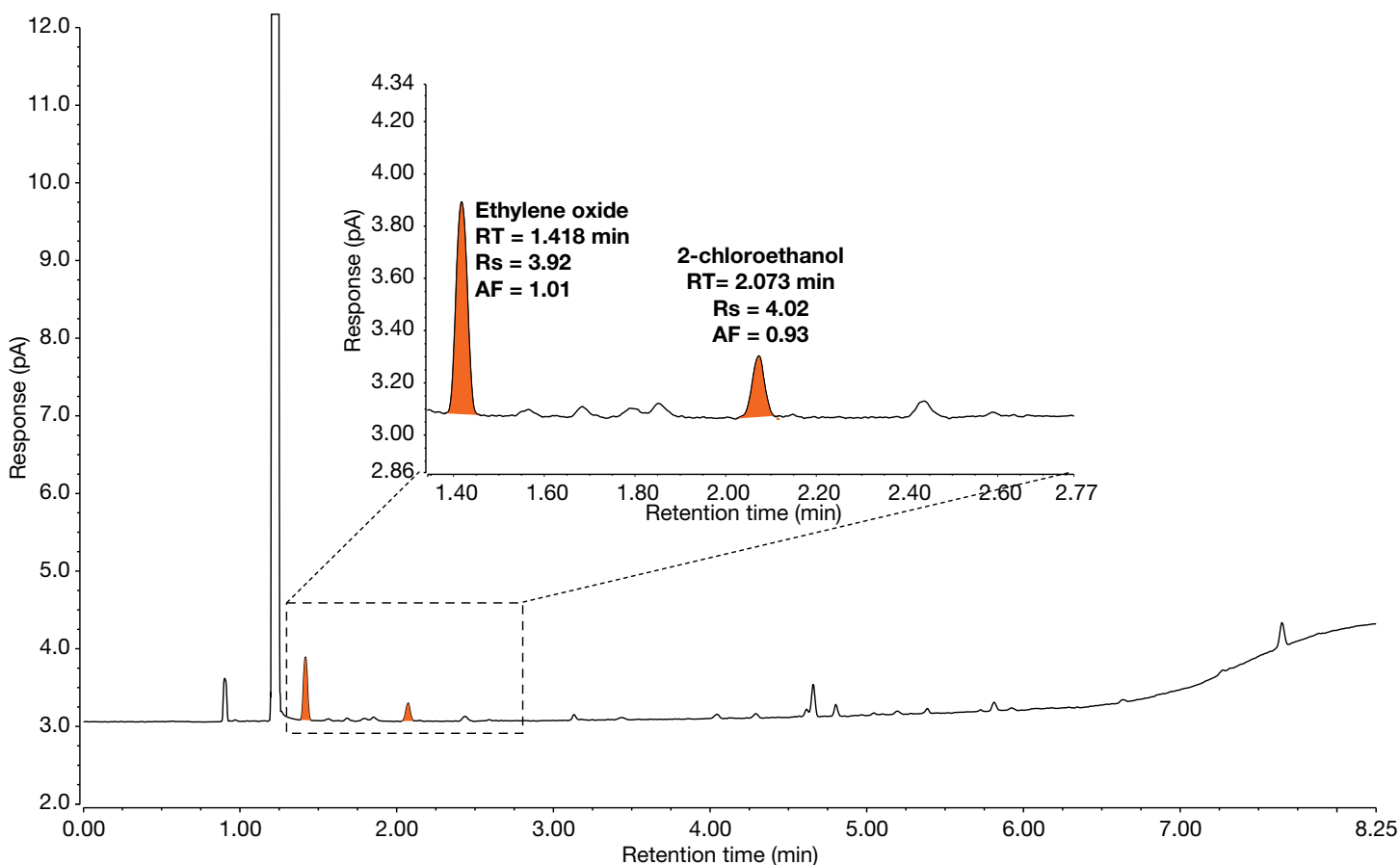


Figure 1. GC-FID chromatogram of a mixed 1 $\mu\text{g/g}$ ethylene oxide and 5 $\mu\text{g/g}$ 2-chloroethanol standard annotated with compound name, retention time (RT), chromatographic resolution (Rs), and peak asymmetry factor (AF)

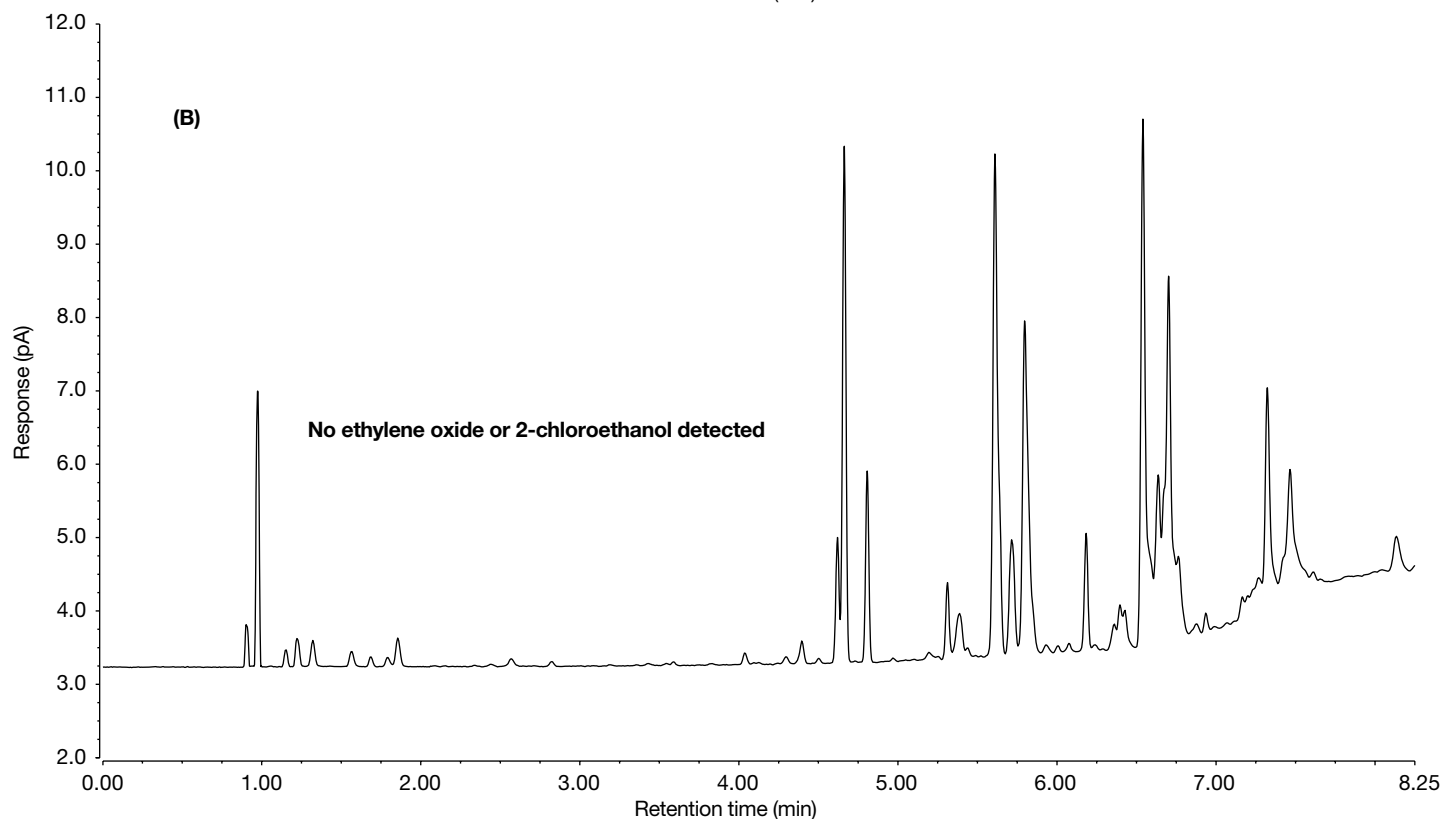
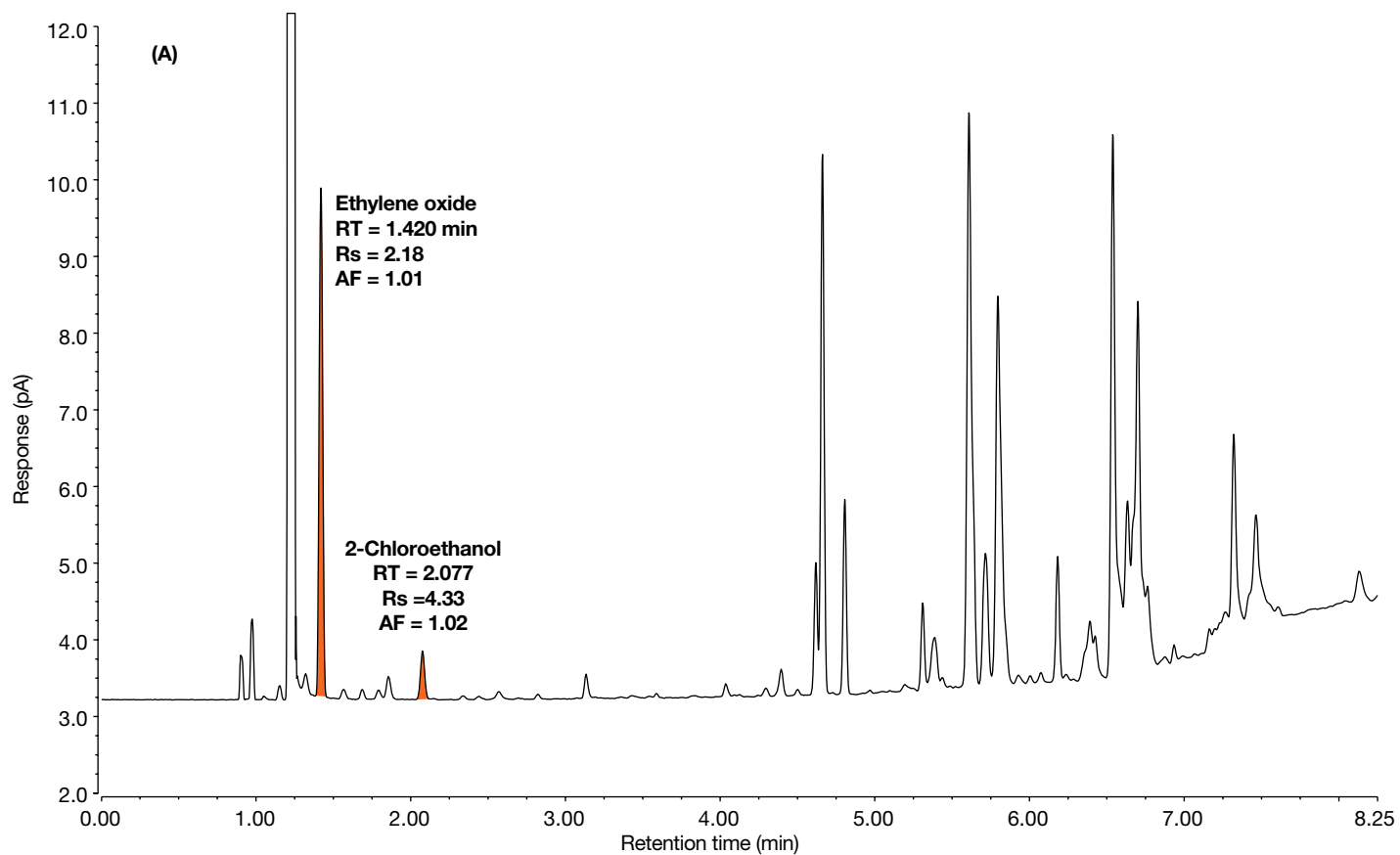


Figure 2. (A) GC-FID chromatogram of face mask sample A spiked with 10 $\mu\text{g/g}$ ethylene oxide and 15 $\mu\text{g/g}$ 2-chloroethanol annotated with compound name, retention time (RT), chromatographic resolution (Rs), and peak asymmetry factor (AF) with matrix peaks present after 4 minutes and (B) GC-FID chromatogram of face mask sample A unspiked showing no ethylene oxide or 2-chloroethanol

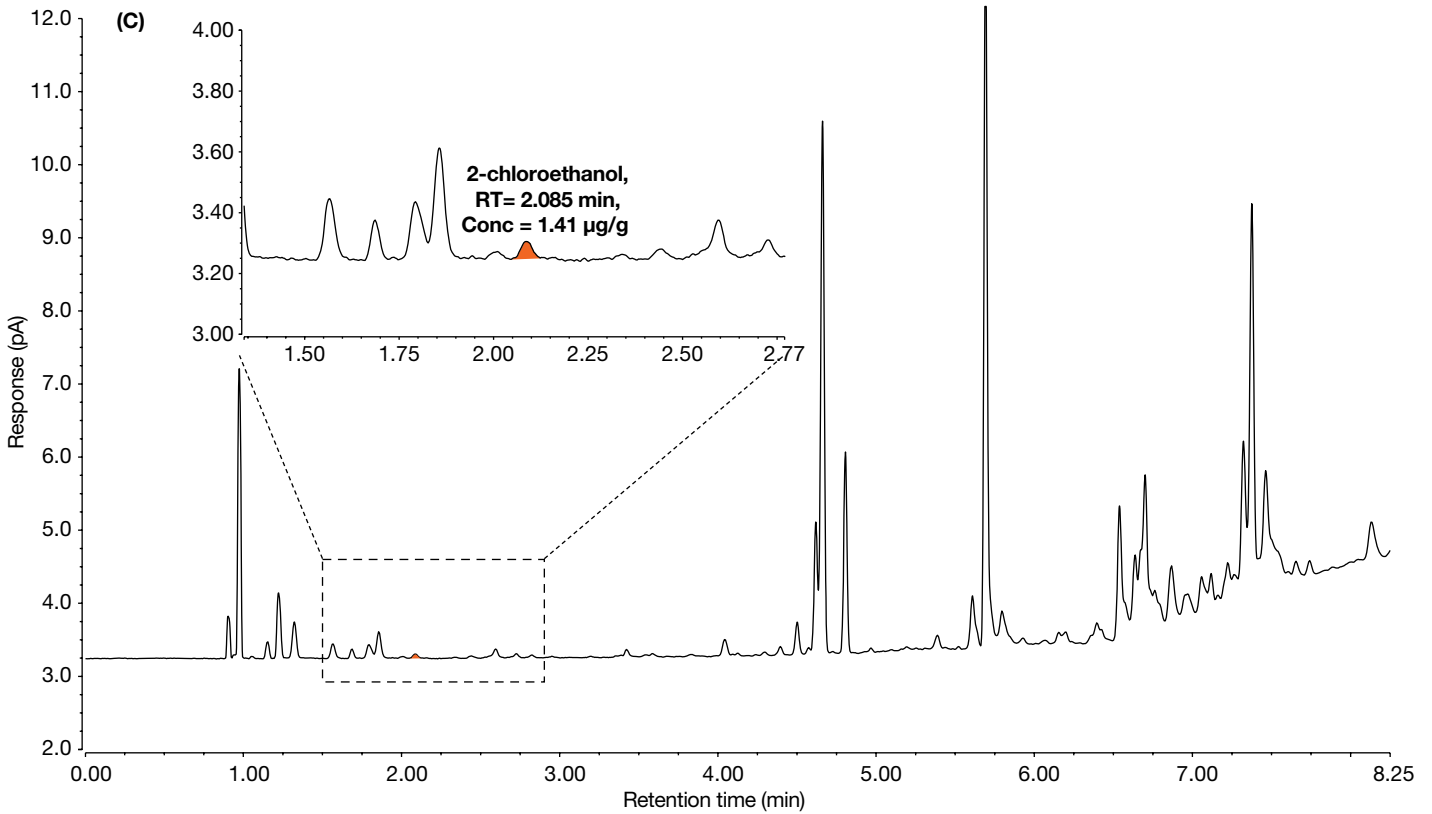


Figure 2. (C) GC-FID chromatogram of face mask sample B unspiked showing no ethylene oxide and 1.41 µg/g 2-chloroethanol

Linearity

To obtain accurate quantification of results, a calibration curve is essential. Linearity was assessed by performing triplicate standard injections across the range 0.2–20 µg/g for ethylene oxide and 1–15 µg/g for

2-chloroethanol. Examples of the calibration curves produced are shown in Figure 3. Excellent linearity was achieved across the range with R^2 values >0.998 and average calibration factor (AvCF) %RSDs <4.

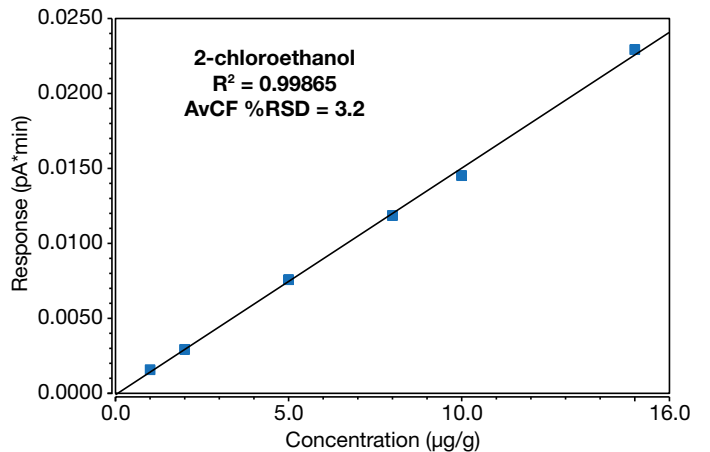
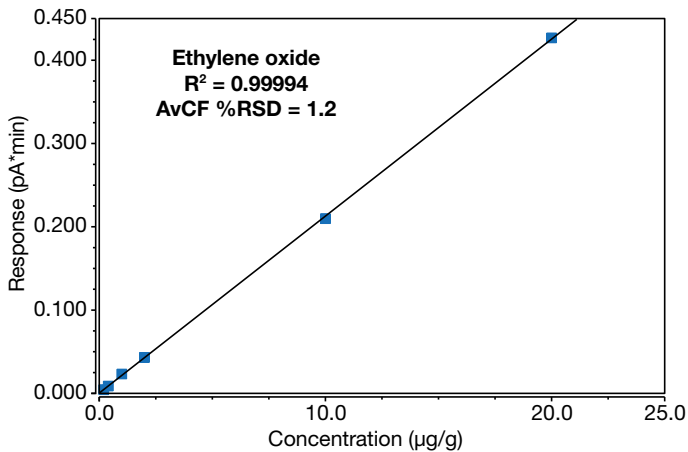


Figure 3. Calibration curves generated for ethylene oxide and 2-chloroethanol annotated with coefficient of determination (R^2) and AvCF %RSD across the range 0.2–20 µg/g for ethylene oxide and 1–15 µg/g for 2-chloroethanol for triplicate standard injections

Sensitivity

Sensitivity was assessed as method detection limits (MDLs) calculated by performing n=9 consecutive injections of the lowest calibration standard (0.2 µg/g ethylene oxide and 1 µg/g 2-chloroethanol). The MDLs were calculated considering the Student's-t critical values for the corresponding degrees of freedom (99% confidence) and the standard deviation of the calculated amount. Excellent sensitivity was obtained with an MDL of 0.027 µg/g achieved for ethylene oxide and an MDL of 0.104 µg/g achieved for 2-chloroethanol. This is significantly below the 10 µg/g limit set within the GB19083-2010 standard. Overlaid chromatograms of the lowest calibration standard are shown in Figure 4.

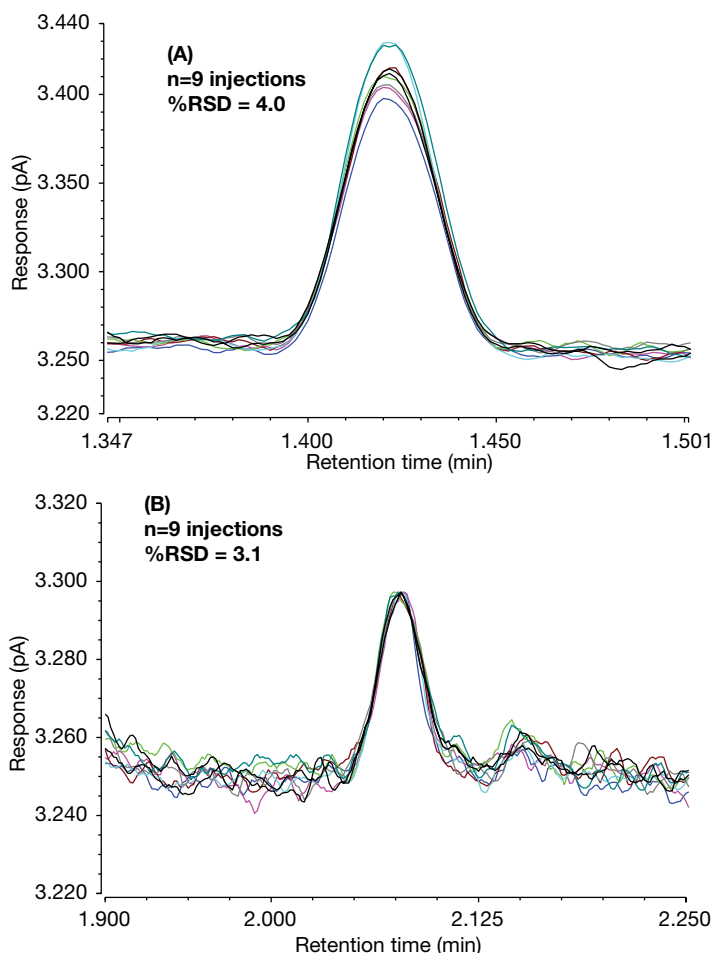


Figure 4. Overlaid chromatograms of n=9 repeat injections of a 0.2 µg/g ethylene oxide standard (a) and of a 1 µg/g 2-chloroethanol standard (b)

Precision

Precision was assessed using n=9 standard injections at low and high levels (0.2 µg/g and 10 µg/g ethylene oxide and 1 µg/g and 10 µg/g 2-chloroethanol). A %RSD (n=9) of the peak areas of <4 was obtained at both levels for both compounds. The results are summarized in Table 2.

Table 2. Obtained %RSD peak area values for n=9 standard injections

| Compound | %RSD Peak area (n=9) | | |
|-----------------|----------------------|--------|---------|
| | 0.2 µg/g | 1 µg/g | 10 µg/g |
| Ethylene oxide | 4.0 | N/A | 2.8 |
| 2-chloroethanol | N/A | 3.1 | 1.7 |

Quantification of target compounds in real samples

Surgical style face masks were obtained from 2 different manufacturers and analyzed in duplicate as spiked (0.4 µg/g and 10 µg/g ethylene oxide and 2 µg/g and 15 µg/g 2-chloroethanol) and unspiked samples. No ethylene oxide was detected in either unspiked sample, however, a small amount of 2-chloroethanol was detected in sample B (Figure 2C). The mean % spike recoveries were all within the range 80–120%. The full results are shown in Table 3.

Table 3. Mean spike recovery results for two different samples

| Compound | Sample A recovery (%) | | Sample B recovery (%) | |
|-----------------|-----------------------|------------|-----------------------|------------|
| | Low spike | High spike | Low spike | High spike |
| Ethylene oxide | 93 | 98 | 82 | 83 |
| 2-chloroethanol | 105 | 89 | 89 | 86 |

Conclusions

The Thermo Scientific TriPlus 500 HS with the TRACE 1310 GC and Instant Connect FID is an easy to use, reliable system with no sample preparation required. The method described here allows for a fast, simple, and cost-effective analysis of hazardous volatile impurities such as ethylene oxide and 2-chloroethanol in face masks. The analytical results obtained in the experiments described here demonstrate:

- Chromatographic resolution of the analytes >2.0 in both standards and samples, indicating sufficient separation between the target analytes.
- Excellent linearity over the tested ranges with the coefficient of determination, R^2 , values >0.998 and AvCF %RSD <4, allowing precise quantification of the target compound.
- Outstanding sensitivity was obtained with calculated MDLs of 0.03 µg/g and 0.10 µg/g for ethylene oxide and 2-chloroethanol, respectively, well below the 10 µg/g ethylene oxide requirement of the GB 19083-2010 standard.

- Precision values as %RSD peak area were <4 across the calibration range for both analytes.
- Accuracy was demonstrated with two different samples spiked at 0.4 µg/g and 10 µg/g ethylene oxide and 2 µg/g and 15 µg/g 2-chloroethanol with calculated results within 80–120% of the spiked amounts.

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