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# Cigarettes — Determination of selected volatile organic compounds in the mainstream smoke of cigarettes with an intense smoking regime — Method using GC/MS

*Cigarettes — Dosage de composés organiques volatils sélectionnés dans le courant principal de la fumée de cigarette avec un régime de fumage intense — Méthode par CG-SM*



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# Foreword

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The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights. Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see www.iso.org/patents).

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For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT), see www.iso.org/iso/foreword.html.

This document was prepared by Technical Committee ISO/TC 126, *Tobacco and tobacco products*.

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.



# Introduction

The CORESTA (www.coresta.org) Special Analytes Sub-Group (since 2017 the Sub-Group changed its name to Smoke Analytes Sub-Group) carried out a collaborative study in 2005 to compare smoke analyte yields of selected volatile organic compounds (volatiles) obtained from different laboratories using their own preferred methodologies. This study reported significant and unacceptable differences in volatiles yields, especially for 1,3-butadiene and acrylonitrile and suggested that further work was required to understand factors influencing the variability. Key parameters of existing methodologies were reviewed and further studies were conducted on selected volatiles between 2008[1] and 2009[2]. These studies investigated critical method steps that required optimization before incorporation into a CORESTA Recommended Method (CRM). The CRM was based on collecting the volatiles from mainstream cigarette smoke in cryogenically cooled impinger traps containing methanol. The impinger solutions were fortified with benzene-$D_6$ and analysed by gas chromatography/mass spectrometry (GC-MS).

This document was produced through a CORESTA collaborative study conducted in 2011, involving 17 laboratories from 11 countries and included 10 samples with different tar yields[3]-[5]. Cigarettes were smoked with the intense smoking regime specified in Health Canada Official Method T-115 (equivalent to ISO 20778). Statistical evaluations carried out according to ISO 5725-1 and ISO 5725-2 are included.

No machine smoking regime can represent all human smoking behaviour.

— It is recommended that cigarettes also be tested under conditions of a different intensity of machine smoking than those specified in this document.

— Machine smoking testing is useful to characterize cigarette emissions for design and regulatory purposes, but communication of machine measurements to smokers can result in misunderstandings about differences in exposure and risk across brands.

— Smoke emission data from machine measurements may be used as inputs for product hazard assessment, but they are not intended to be nor are they valid as measures of human exposure or risks. Communicating differences between products in machine measurements as differences in exposure or risk is a misuse of testing using International Standards

# Cigarettes — Determination of selected volatile organic compounds in the mainstream smoke of cigarettes with an intense smoking regime — Method using GC/MS

**WARNING — The use of this document can involve hazardous materials, operations and equipment. This document does not purport to address all the safety problems associated with its use. It is the responsibility of the user of this document to establish appropriate safety and health practices and determine the applicability of any other restrictions prior to use.**



## 1 Scope

This document specifies a method for the quantification of selected volatile organic compounds (VOCs: 1,3-butadiene, isoprene, acrylonitrile, benzene and toluene) by gas chromatography/mass spectrometry (GC-MS) in mainstream cigarette smoke using ISO 20778 smoking parameters.

## 2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 3402, *Tobacco and tobacco products — Atmosphere for conditioning and testing*

ISO 8243, *Cigarettes — Sampling*

ISO 20778, *Cigarettes — Routine analytical cigarette smoking machine — Definitions and standard conditions with an intense smoking regime*

ISO 20779, *Cigarettes — Generation and collection of total particulate matter using a routine analytical smoking machine with an intense smoking regime*

## 3 Terms and definitions

No terms and definitions are listed in this document.

ISO and IEC maintain terminological databases for use in standardization at the following addresses:

— ISO Online browsing platform: available at https://www.iso.org/obp

— IEC Electropedia: available at http://www.electropedia.org/

## 4 Principle

Selected volatiles are collected by passing the mainstream smoke of cigarettes through a glass fibre filter pad as specified in ISO 20778 into cryogenic traps containing methanol.

The impinger solutions are fortified with internal standard and analysed by GC-MS.

## 5 Apparatus

In addition to the list provided below, usual laboratory apparatus and equipment are needed for preparation of samples and standards.

**5.1 Impinger trapping system**, capable of being connected in series, a cryogenically cooled liquid impinger to efficiently trap volatiles present in the vapour phase of mainstream smoke.

**5.2 GC-MS system**, to obtain chromatographic data.

The GC shall be configured to perform split injections on a capillary column and operated in Selected Ion Monitoring mode (SIM or equivalent).

**5.3 Gas tight syringes**, of appropriate volumes.

**5.4 Fused silica capillary column**, for example DB-624, length 60 m with internal diameter of 0,25 mm and 1,4 µm film thickness, or equivalent.

**5.5 Spectrophotometer,** to estimate 1,3-butadiene concentration in secondary stock solution.

## 6 Reagents

**6.1 Dry ice**.

**6.2 Isopropanol**, for Dewar flasks.

**6.3 Methanol**, HPLC grade or better.

The methanol should be checked to ensure the background levels of the analytes will not negatively affect the analysis.

**6.4 Ethanol**, reagent grade or equivalent.

**6.5 Benzene-$D_6$**, min. 99 atom % D; checked for the absence of not-labelled analogue.

**6.6 Toluene-$D_8$**, min. 99 atom % D; checked for the absence of not-labelled analogue.

**6.7 1,3-Butadiene**, min. 99 %.

**6.8 Isoprene**, min. 99 %.

**6.9 Acrylonitrile**, min. 99 %.

**6.10 Benzene**, min. 99 %.

**6.11 Toluene**, min. 99 %.

## 7 Preparation

### 7.1 Preparation of glassware

Glassware shall be cleaned and dried in such a manner which ensures that contamination from glassware does not occur.

It is important that all possible sources of contamination are removed from the work area.

## 7.2 Preparation of standards

### 7.2.1 General

Where available, certified reference solutions of the selected volatile organic compounds and internal standards can be used.

### 7.2.2 Preparation of internal standard spiking solution

#### 7.2.2.1 Internal standard stock solution

Transfer the contents of a 1 g ampoule of benzene-$D_6$ into a 10 ml amber volumetric flask. Dilute to volume with methanol.

The inclusion of other internal standards, such as toluene-$D_8$, may also be suitable. Laboratories shall demonstrate the suitability of the inclusion of additional internal standards.

#### 7.2.2.2 Internal standard spiking solution

Using a volumetric pipette, transfer 4 ml of the stock solution (7.2.2.1) into a 100 ml volumetric flask and dilute to volume with methanol. This solution has a concentration of 4 000 µg/ml.

#### 7.2.2.3 Storage

Store the diluted solutions in 25 ml vials with PTFE-lined caps in freezer at approximately –20 °C

### 7.2.3 Preparation of working standards for isoprene, acrylonitrile, benzene, and toluene

#### 7.2.3.1 Primary isoprene, acrylonitrile, benzene and toluene stock solutions

Using gas tight syringes, weigh accurately 100 mg of isoprene, acrylonitrile, benzene, and toluene into separate 10 ml amber volumetric flasks that are half filled with methanol. Dilute each compound to volume with methanol. Each solution has a nominal concentration of 10 mg/ml.

NOTE Approximate volumes corresponding to 100 mg are: isoprene = 150 µl, acrylonitrile = 140 µl, benzene = 130 µl, toluene = 120 µl.

#### 7.2.3.2 Secondary stock solution (mixture of isoprene, acrylonitrile, benzene and toluene primary stock solutions)

A combined secondary stock solution is prepared by transferring appropriate amounts (Table 1) of isoprene, acrylonitrile, benzene, and toluene primary stock solutions (7.2.3.1) into a 50 ml volumetric flask that is a third full with methanol. Dilute to volume with methanol.

**Table 1 — Preparation of secondary stock solution**

| Analyte | Volume of primary stock (ml) | Approximate concentration (µg/ml) |
|---|---|---|
| Isoprene | 3,0 | 600 |
| Acrylonitrile | 1,0 | 200 |
| Benzene | 1,0 | 200 |
| Toluene | 1,0 | 200 |

#### 7.2.3.3 Calibration standard solutions (for isoprene, acrylonitrile, benzene and toluene)

Prepare seven working standard solutions by mixing appropriate volumes of secondary stock solution (7.2.3.2) and internal standard spiking solution (7.2.2.2) with sufficient methanol to cover the concentration range of interest, i.e. (12 to 600) µg/ml (isoprene); (4 to 200) µg/ml (acrylonitrile); (4 to 200) µg/ml (benzene); (4 to 200) µg/ml (toluene) and 40 µg/ml of internal standard (e.g. benzene-$D_6$).

Transfer aliquots of each calibration standard solution into amber GC vials and fill each vial up to the shoulder of the vial to minimize headspace.

Adjust standard concentrations accordingly to reflect levels of volatiles found in smoke samples.

PTFE lined GC vial caps are recommended, although other materials may also be suitable.

#### 7.2.3.4 Storage

Store all solutions in glass vials with PTFE-lined caps in freezer at approximately –20 °C.

### 7.2.4 Preparation of working standards for 1,3-butadiene

#### 7.2.4.1 Primary 1,3-butadiene stock solution

Attach a piece of chemically resistant polymer tubing to the valve of a cylinder containing 1,3-butadiene. Place a Pasteur pipette on the other end of the tubing and immerse the tip of the pipette into a 100 ml amber glass volumetric flask containing methanol up to the base of the neck of the flask. Open the valve and gently bubble the 1,3-butadiene into the methanol for approximately 5 min. Dilute to volume using methanol and mix well.

#### 7.2.4.2 Secondary 1,3-butadiene stock solution

Pipette 1 ml of the primary 1,3-butadiene stock solution (7.2.4.1) into a 100 ml volumetric flask and dilute to volume with methanol. Mix well.

#### 7.2.4.3 Determination of secondary 1,3-butadiene stock concentration

Pipette 1 ml of the secondary 1,3-butadiene stock solution (7.2.4.2) into a 100 ml volumetric flask and dilute to volume using ethanol. This solution is used only to check the concentration of the secondary stock solution and shall not be used to prepare the working standards.

Measure the absorbance of the solution against an ethanol blank on a spectrophotometer (use 1 cm long cuvettes). Conduct a wave scan from 200 nm to 250 nm to determine the wavelength of maximum absorbance. 1,3-butadiene in hexane absorbs at 217 nm whereas 1,3-butadiene in ethanol can have a peak shift. Measure the absorbance at the peak maximum.

Repeat the above measurement three more times and calculate the mean absorbance, $A$ (at least three significant figures). The absorbance should be between 0,2 and 0,6 extinction units. If it is higher, make a new secondary stock solution using a smaller volume of the primary stock solution and repeat the spectrophotometer measurements to determine the concentration of the secondary stock. If the absorbance is lower, make a new secondary stock solution using a larger volume of the primary stock solution and repeat the spectrometer measurements to determine the concentration of the secondary stock.

The concentration of the secondary stock solution, in micrograms per millilitre, $c_{2S}$, is calculated by Formula (1):

$$c_{2S} = \frac{A}{20\ 893} \times 54 \times 100 \times 1\ 000 \quad (1)$$

where

| | |
|---|---|
| $A$ | is the mean absorbance; |
| 20 893 l $mol^{-1}$ $cm^{-1}$ | is the molar absorption coefficient of 1,3-butadiene; |
| 54 g/mol | is the molar mass of 1,3-butadiene; |
| 100 ml | is the volume of the solution; |
| 1 000 | is the units conversion factor. |

#### 7.2.4.4 1,3-butadiene calibration standard solutions

Prepare five working standard solutions by mixing 1,3-butadiene secondary stock solution (7.2.4.2) and internal standard spiking solution (7.2.2.2) with sufficient methanol to cover the concentration range of interest, i.e. (5 to 50) µg/ml for 1,3-butadiene and 40 µg/ml for internal standard.

Transfer aliquots of each calibration standard solution into amber GC vials and fill each vial up to the shoulder of the vial to minimize headspace.

NOTE Certified concentrations of 1,3-butadiene in methanol can be purchased and used to prepare the standards.

#### 7.2.4.5 Storage

Store all solutions in glass vials with PTFE-lined caps in freezer at approximately –20 °C.

Stability of all standard solutions for all components should be assessed by laboratories under their own in-house conditions before use as stability is dependent on the actual storage conditions of each laboratory.

## 8 Sampling

Carry out sampling in accordance with ISO 8243.

## 9 Tobacco product preparation

Condition the cigarettes in accordance with ISO 3402.

## 10 Sample generation — Smoking of cigarettes

### 10.1 General

The smoking parameters for which the method has been studied are defined in ISO 20778.

### 10.2 Smoking machine setup

An analytical cigarette-smoking machine complying with the requirements of ISO 20778 is required.

A methanol-filled impinger system that efficiently traps the VOCs of interest is required. An example using two impingers is provided in Figure 1; however, other trapping systems using a different number of impingers, different impinger tip styles (capillary, fritted, etc.) and a different volume of trapping solution can also provide suitable trapping efficiency.

Fill all coolant reservoirs with one-third full of isopropanol. Add dry ice until each reservoir is filled halfway. The number of reservoirs required is dependent on the impinger design and shall be optimized to ensure that all volatiles are trapped efficiently.

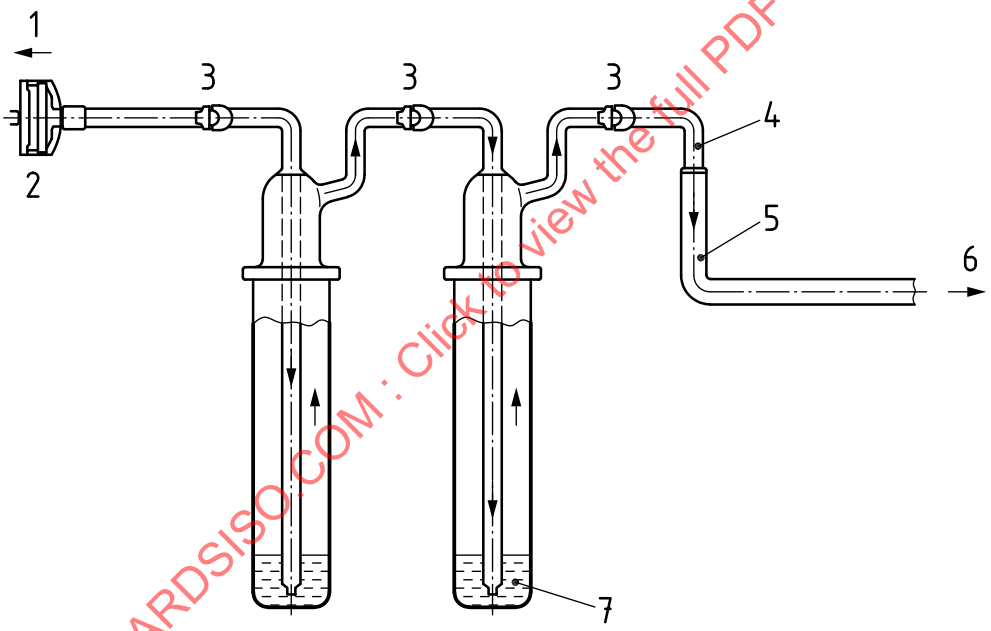
Add 20 ml of methanol to each impinger and place the impingers into the coolant reservoir containing the dry ice/isopropanol solution. Check each coolant reservoir to ensure that the temperature is at or below –70 °C.

A volume other than 20 ml of methanol may need to be added to each impinger depending on the particular style of impinger used.

The methanol-filled impingers shall be given sufficient time to cool to –70 °C or below before starting smoke collection.

Connect the impingers to the smoking machine (see Figure 1).

Check and adjust the puff volume drawn by the smoking machine at all channels.




**Key**

1 to cigarette
2 glass fibre filter pad
3 glass ball joint or tubing connections
4 glass elbow or tubing
5 tubing
6 to smoking machine
7 100 ml impingers each containing 20 ml methanol

**Figure 1 — Example of an impinger setup for smoking machines**

To determine whether a leak has occurred in the smoking machine impinger setup, use a leak tester. If the fluid column does not maintain its position but drops, then there is a leak in the system.

It is recommended that tubing other than silicone tubing is used for connections between the smoking machine and the impingers (i.e. polyethylene, polyvinyl chloride, polypropylene). Methyl silicone tubing is not recommended since adsorption of the analytes can occur. Tubing should be as short as possible to minimize the potential for any adsorption. Connectors made of either glass or stainless steel are preferred.

It is recommended that the trapping efficiency is checked when validating this method. To check the trapping efficiency of the method, add an additional impinger and follow the method accordingly. Analyse each impinger individually for the VOCs of interest. If no VOCs are detected in the additional impinger, then only the prescribed number of impingers is required to trap all the VOCs effectively. Poor trapping efficiency can be due to the impinger or impinger tip design.

If a carryover occurs, it is a responsibility of each laboratory to assess the carryover with respect to the specific trapping system design and decide how to manage it. Carryover should be repeatable, less than 5 % (ideally less than 1 %).

### 10.3 Smoking

#### 10.3.1 General

The cigarettes are smoked according to ISO 20778 and ISO 20779 with the following modifications.

Glass fibre filter pads of 44 mm diameter are capable of retaining up to 150 mg of TPM and pads of 92 mm diameter are capable of retaining 600 mg of TPM. If, during smoking, this mass is exceeded, the number of cigarettes should be reduced and a calculation made to allow for the reduced number of cigarettes smoked.

#### 10.3.2 Linear smoking

Typically, three cigarettes are smoked per trap onto 44 mm diameter glass fibre filter pad.

#### 10.3.3 Rotary smoking

Typically, five or 10 cigarettes are smoked per trap onto 92 mm diameter glass fibre filter pad.

## 11 Sample analysis

### 11.1 Preparation of sample

After all samples have been smoked, the TPM shall be determined on the glass fibre filter pad as a quality control measure and the glass fibre filter pad discarded. Laboratories should evaluate the trapping system for losses in the tubing that connects the pad holder to the impinger(s) and the connections between impingers (if more than one impinger is used). If there are losses, the tubing may be rinsed, or extra clearing puffs may be taken. The impingers shall be kept in the cooling reservoir until sampling is complete.

After all samples have been smoked following ISO 20778, each impinger is spiked with 200 µl of internal standard spiking solution. The impingers are stoppered and mixed well to ensure that the extract is well mixed. If the impinger setup requires more than one impinger, then the trapping solutions are combined in such a way as to ensure complete mixing of both impingers. Transfer an aliquot of the impinger solution into an amber GC vial for GC-MS analysis. Fill each vial up to the shoulder of the vial to minimize headspace and cap tightly. Prepare all samples in duplicate and keep a set in the freezer at or below –20 °C in case repeated analysis is required.

Samples have been shown to be stable when stored in the freezer (temperature below –20 °C) for a maximum of 48 h. It is recommended that sample stability is determined under storage conditions when validating this method.

## 11.2 Determination

### 11.2.1 GC-MS operating conditions

Set up and operate the GC-MS system in accordance with the manufacturer's instruction.

The following parameters have been found to be suitable for separation.

**GC parameters:**

| | |
|---|---|
| Injector temperature: | 150 °C |
| Column temperature: | 40 °C (6 min) 20 °C/min to 225 °C (6 min) |
| Carrier gas: | Helium |
| Carrier gas flow: | 1,5 ml/min (160 kPa) |
| Injection mode: | Split |
| Injection split ratio: | 30:1 |
| Injection split flow: | 30 ml/min |
| Injection volume: | 3 µl |

**MS parameters:**

| | |
|---|---|
| Transfer line temperature: | 240 °C |
| Source temperature: | 240 °C |
| Acquisition mode: | SIM (or SCAN) |
| Solvent delay: | Column dependent |
| Low mass: | 40,0 |
| High mass: | 200,0 |

| **Ion traces (*m/z*):** | **Quantification** | **Confirmation** |
|---|---|---|
| 1,3-butadiene | 54 | 53 |
| Isoprene | 67 | 68 |
| Acrylonitrile | 52 | 53 |
| Benzene | 78 | 77 |
| Benzene-$D_6$ | 84 | 83 |
| Toluene | 91 | 92 |

Chromatographic separation should be similar to example chromatograms shown in Annex A, Figures A.1 to A.4.

NOTE The choice of chromatographic conditions and data acquisition parameters can differ for different instrument configurations.

### 11.2.2 Calibration

Analyse, successively, each working standard solution (7.2.3.3 and 7.2.4.4) by GC-MS. Record the area of each of the analysed compounds and the internal standard peaks. Generate a calibration curve for each of the compounds by calculating a linear regression equation of the peak area ratios of the analysed compounds to the internal standard against their concentration. The intercept of these regression lines should be close to zero.

### 11.2.3 Calculation

The yield of individual selected volatile compounds in the mainstream smoke of cigarettes, $m_i$, expressed in micrograms per cigarette, is given by Formula (2):

$$m_i = \frac{C_i V}{N_{cig}} \tag{2}$$

where

$C_i$ is the concentration of the analyte, in micrograms per millilitre, in the sample;

$V$ is the volume of methanol in the impinger, in millilitres;

$N_{cig}$ is the number of cigarettes smoked.

The expression of the laboratory data depends on the purpose for which the data are required, and the level of laboratory precision. Any further statistical analyses should be calculated and expressed on the basis of the laboratory data before any rounding has taken place.

The yield of individual selected volatile compounds in the mainstream smoke of cigarettes is reported in micrograms per cigarette (µg/cig) to the nearest 0,1 µg.

## 12 Repeatability and reproducibility

### 12.1 General

An international collaborative study was conducted in 2012 involving 17 laboratories and 10 cigarette samples including the reference cigarettes KR 1R5F, KR 3R4F and the CORESTA Monitor 6[5]. The cigarettes were smoked with the intense smoking regime specified in Health Canada Official Method T-115 (equivalent to ISO 20778). Five replicate analyses were conducted for each sample type. Description of the samples are provided in Table 2. The values for repeatability, *r*, and reproducibility, *R*, obtained with this method are provided in Tables 3 to 7. The statistical evaluation was performed according to ISO 5725-2.

The difference between two single results found on matched cigarette samples by one operator using the same apparatus within the shortest feasible time interval will exceed the repeatability limit, *r*, on average not more than once in 20 cases in the normal and correct operation of this method.

Single results on matched cigarette samples reported by two laboratories will differ by more than the reproducibility limit, *R*, on average not more than once in 20 cases in the normal and correct operation of the method.

**Table 2 — Cigarette test samples of 2012 the collaborative study**

| Sample | Intense TPM yield (mg/cig) | Product characterization |
|---|---|---|
| Sample 1 | 39,5 | Dark air-cured |
| Sample 2 | 35,9 | American blended |
| Sample 3 | 31,9 | American blended |
| Sample 4 | 27,7 | Virginia blended |
| Sample 5 | 19,4 | Virginia blended |
| Sample 6 | 35,8 | Virginia blended |
| Sample 7 | 22,4 | Charcoal filtered |
| 3R4F | 41,7 | Kentucky Reference 3R4F |
| 1R5F | 27,2 | Kentucky Reference 1R5F |
| CM6 | 43,7 | CORESTA Monitor 6 Test piece |

## 12.2 Results of the 2012 collaborative study

Repeatability and reproducibility values for the individual selected volatiles are given in Tables 3 to 7.

**Table 3 — 1,3-Butadiene**

| Sample | $N^a$ | Mean | $r$ | $R$ |
|---|---|---|---|---|
| | | (µg/cigarette) | | |
| CM6 | 14 | 108,1 | 17,1 | 59 |
| 1R5F | 16 | 91,1 | 19,2 | 66 |
| 3R4F | 15 | 100,5 | 17,5 | 55 |
| 1 | 12 | 73,8 | 12,7 | 30 |
| 2 | 11 | 94,9 | 18,2 | 49 |
| 3 | 10 | 105,8 | 17,4 | 41 |
| 4 | 11 | 85,4 | 18,2 | 49 |
| 5 | 11 | 62,7 | 13,0 | 51 |
| 6 | 11 | 89,9 | 17,9 | 41 |
| 7 | 12 | 89,7 | 20,6 | 68 |

[a] $N$ = number of data sets taken for statistical analysis after removal of outliers.

**Table 4 — Isoprene**

| Sample | $N^a$ | Mean | $r$ | $R$ |
|---|---|---|---|---|
| | | (µg/cigarette) | | |
| CM6 | 15 | 995,1 | 147,0 | 356 |
| 1R5F | 16 | 885,8 | 131,0 | 590 |
| 3R4F | 15 | 912,5 | 113,2 | 392 |
| 1 | 11 | 415,8 | 64,7 | 114 |
| 2 | 12 | 647,3 | 112,8 | 197 |
| 3 | 11 | 814,0 | 97,1 | 261 |
| 4 | 12 | 688,7 | 166,5 | 329 |
| 5 | 12 | 394,9 | 95,2 | 225 |
| 6 | 12 | 499,7 | 73,2 | 170 |
| 7 | 12 | 632,8 | 134,6 | 410 |

[a] $N$ = number of data sets taken for statistical analysis after removal of outliers.

**Table 5 — Acrylonitrile**

| Sample | $N^a$ | Mean | $r$ | $R$ |
|---|---|---|---|---|
| | | (µg/cigarette) | | |
| CM6 | 14 | 25,3 | 4,1 | 10 |
| 1R5F | 16 | 28,0 | 4,5 | 23 |
| 3R4F | 14 | 27,0 | 3,5 | 11 |
| 1 | 12 | 27,2 | 5,0 | 10 |
| 2 | 12 | 24,3 | 5,2 | 11 |
| 3 | 11 | 24,2 | 5,3 | 12 |
| 4 | 12 | 15,8 | 3,5 | 11 |
| 5 | 11 | 13,4 | 2,4 | 12 |
| 6 | 11 | 18,1 | 2,8 | 8 |
| 7 | 11 | 20,1 | 4,7 | 9 |

[a] $N$ = number of data sets taken for statistical analysis after removal of outliers.

**Table 6 — Benzene**

| Sample | $N^a$ | Mean | $r$ | $R$ |
|---|---|---|---|---|
| | | (µg/cigarette) | | |
| CM6 | 15 | 108,5 | 15,2 | 37 |
| 1R5F | 16 | 79,3 | 10,6 | 35 |
| 3R4F | 16 | 97,4 | 9,9 | 35 |
| 1 | 12 | 77,7 | 11,8 | 31 |
| 2 | 12 | 85,3 | 15,2 | 30 |
| 3 | 11 | 91,1 | 10,4 | 35 |
| 4 | 11 | 65,0 | 10,7 | 34 |
| 5 | 12 | 51,2 | 7,3 | 31 |
| 6 | 11 | 80,4 | 10,1 | 33 |
| 7 | 11 | 63,8 | 11,6 | 32 |

[a] $N$ = number of data sets taken for statistical analysis after removal of outliers.

**Table 7 — Toluene**

| Sample | $N^a$ | Mean | $r$ | $R$ |
|---|---|---|---|---|
| | | (µg/cigarette) | | |
| CM6 | 14 | 174,4 | 23,5 | 62 |
| 1R5F | 15 | 135,6 | 21,0 | 70 |
| 3R4F | 15 | 174,2 | 19,7 | 64 |
| 1 | 11 | 142,3 | 22,3 | 50 |
| 2 | 11 | 142,7 | 24,4 | 51 |
| 3 | 10 | 148,3 | 18,6 | 56 |
| 4 | 11 | 96,0 | 18,5 | 43 |
| 5 | 11 | 78,5 | 11,2 | 43 |
| 6 | 11 | 119,4 | 19,3 | 46 |
| 7 | 11 | 99,0 | 16,6 | 49 |

[a] $N$ = number of data sets taken for statistical analysis after removal of outliers.



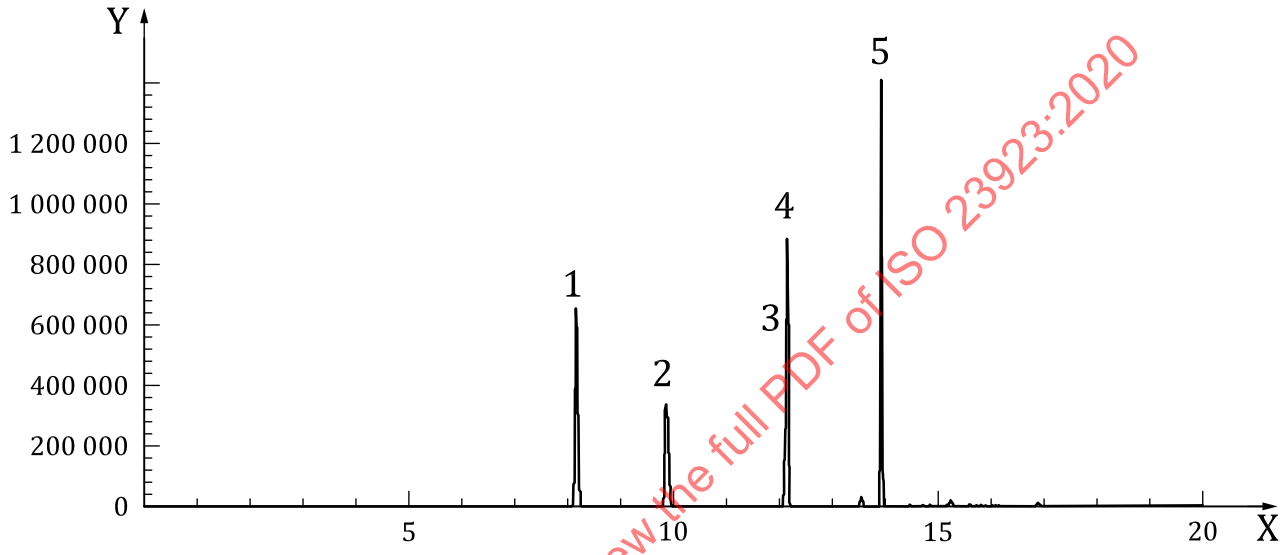
## 13 Test report

The test report shall state the yield of VOCs in µg/cigarette, the method used, and shall include all conditions not specified in this document or regarded as optional. It shall also give all details necessary for the identification of the cigarettes smoked.

# Annex A
(informative)

# Examples of chromatograms

Example chromatograms are given in Figures A.1 to A.4.




**Key**

| | | | |
|---|---|---|---|
| X | time, min | 3 | $D_6$-benzene |
| Y | abundance | 4 | benzene |
| 1 | isoprene | 5 | toluene |
| 2 | acrylonitrile | | |

**Figure A.1 — Example of a chromatogram of toluene, isoprene, benzene and acrylonitrile calibration standard (full scan mode)**