Health and Safety
Executive

Aromatic amines in air and on surfaces

Laboratory method using pumped acid coated filters, moistened swabs and HPLC

MDHS75/2

Methods for the
Determination of
Hazardous Substances

Health and Safety
Laboratory

Scope

1 This procedure describes the determination of time-weighted average concentrations of aromatic amines in air and the identification of amine contamination on surfaces.

2 The procedure is suitable for a range of aromatic amines including: 4,4’methylene dianiline (MDA, CAS 101-77-9), 2,2’-dichloro-4,4’-methylene dianiline (MbOCA, CAS 101-14-4), aniline (Phenylamine, CAS 62-53-3), o-chloroaniline (2-chloroaniline, OCA, CAS 95-51-2) and o-toluidine (2-methylaniline, CAS 95-53-4).

Summary

3 A measured volume of air is drawn through an acid-coated glass fibre filter and, if required, a sorbent back-up tube in series to trap aromatic amines with significant vapour pressure (not required for MDA, MbOCA and aniline). After sampling, the filters are desorbed in sodium hydroxide solution and the sorbent tube in methanol. The resultant solutions are analysed by HPLC with UV detection.

4 Surface contamination can be estimated using wipes moistened with methanol. The wipes are subsequently desorbed in methanol and analysed by HPLC.

5 The use of alternative methods not included in the MDHS series is acceptable provided they can demonstrate the accuracy and reliability appropriate to the application.

Recommended sampling

6 For airborne sampling: Sampling Time: 10 minutes to 8 hours; Sampling Rate: 2 l.min^{-1}; Sampled volume: up to 200 litres.

7 For more volatile aromatic amines, a back-up sorbent tube can be connected in series after the filter, but this is not required for MDA, MbOCA or aniline. The sampling efficiency for inhalable particulate matter will be compromised if a flow rate less than 2 l.min^{-1} is used for the filter/tube combination.

8 Surface contamination can be estimated using methanol-moistened cotton wipes, which collect a proportion of the amine present in a given measured surface area.

Prerequisites

9 Users of this procedure will need to be familiar with the content of MDHS14.[1]

Safety

10 Users should be familiar with standard laboratory practice and carry out a suitable risk assessment. Some aromatic amines are classified as carcinogenic and have been assigned the risk phrase ‘R45’.[2] It is the user’s responsibility to establish appropriate health and safety practices and to ensure compliance with regulatory requirements.

Equipment

11 An IOM inhalable dust sampler, or equivalent, as described in MDHS14.

12 Binder-free acid-coated glass fibre filters (25 mm GF/A have been found suitable for the IOM sampler).

13 Two section sorbent tube containing Tenax®, 30 mg (front section) and 15 mg (back-up section). This can be connected in series to the outlet of the IOM sampler using a short piece of flexible tubing.

14 Personal sampling pumps that meet the requirements of BS EN 1232.[3]

15 A portable flow meter, calibrated against a primary standard, with a measurement uncertainty typically less than ±2%.

16 Flexible plastic tubing for making a leak-proof connection from the sampling train to the pump; belts or harnesses to facilitate attachment of sampling apparatus to sample subjects; flat-tipped tweezers for loading and unloading the filters into samplers; and filter transport cassettes and bottles to transport samples to the laboratory.

17 Filmated cotton wipes (10 x 10 cm) for surface sampling: these may be halved to facilitate sample desorption.

Laboratory apparatus and reagents

18 During the analysis use only reagents of a recognised analytical grade.

19 Ehrlich’s reagent: Dissolve 2.5 g of p-dimethylaminobenzaldehyde (CAS No. 100-10-7) in 1 litre of general purpose grade methanol to which 17 ml of concentrated hydrochloric acid has been added. Store in the dark until required and use within one week. Decontaminate all glassware, surfaces and waste with Ehrlich’s reagent before disposal.

20 Methanol: general purpose grade.

21 Methanol: HPLC grade.

22 Water: HPLC grade.

23 0.135M Sulphuric acid.

24 Filter desorbing solution: dissolve 0.54 g of sodium hydroxide in 100 ml of deionised water. Add 40 ml of this solution to 60 ml methanol and mix.

25 A selection of laboratory glassware, including Pasteur pipettes, beakers, 4 ml vials (with screw-top caps and teflon-silicone septa) and volumetric flasks, Class A, complying with the requirements of BS EN ISO 1042:2000.[4]

26 Solvent-resistant plastic syringe filters (2 µm pore size) for sample filtration prior to analysis.

27 Positive displacement micropipettes complying with the requirements of BS EN 8655-6:2002.[5]

28 A balance, calibrated against a primary standard, for the preparation of the calibration standards. The balance should be capable of weighing to ±0.1 mg over the range -100 g.

29 HPLC mobile phase: Dissolve 6 g of sodium dihydrogen orthophosphate and 1.43 g of disodium hydrogen orthophosphate in 1 litre of deionised water. Add -600 ml of methanol and mix. Adjust the pH to 7.5 with sodium hydroxide solution or orthophosphoric acid.

30 HPLC system with UV-detector: Suitable operating conditions for MbOCA are listed below, but the use of other columns and conditions are acceptable provided they have the accuracy and reliability appropriate to the application.

Column dimensions	100 mm x 4.5 mm ID
Column packing	S3 ODS2
Column temperature °C	20 °C
Mobile phase	60:40 methanol:buffer
Flow rate	0.8 $ml.min^{-1}$
UV detector	242 nm

31 Under these conditions, the retention time of MbOCA was 10.4 minutes.

Preparation and sampling

32 Lay out clean glass fibre filters, in a fume cupboard, so that they are supported with a minimum amount of their surface area in contact with any solid surface. With 25 mm GF/A filters this may be achieved by resting the filter on the top rim of a 10 ml beaker. Add 0.20 ml of 0.135M sulphuric acid to each filter and allow to dry at room temperature or in an oven at 100 °C overnight. The filters may be stored in a desiccator or airtight container for several weeks before use.

Air sampling

33 Sampling should be carried out in accordance with the procedures described in MDHS14 for inhalable dusts using an IOM sampler. Remove the ends of the sorbent tube before sampling (if using) and place in series behind the filter head using a short piece of flexible tubing. Maximising the air volume will improve the detection limit for the procedure. Select a sampling period that avoids overloading or use two or more consecutive samples.

34 After sampling remove the filter from the sampling head using clean flat-tipped tweezers and place in a labelled transport cassette for transfer to the laboratory. Sorbent tubes should be capped (if used) and transported with the blanks and filter cassettes.

Wipe sampling

35 The area of each surface to be sampled should be measured then wiped in a circular motion using a wipe moistened with 1 ml methanol.

36 Seal the wipe samples in individual glass bottles and refrigerate in the laboratory until analysed. Analyse within one week of collection.

Blanks

37 Blanks of each type of sampling medium; filters, sorbent tubes and wipes from the same batch as the samples should be subjected to the same sampling procedure except for the actual period of sampling. A minimum of two blanks of each type should be included with each batch of ten samples.

Calibration

38 Prepare at least five standard solutions of the aromatic amine in methanol, over a suitable calibration range, by accurately weighing an aliquot of the amine into a volumetric flask and diluting as appropriate.

39 Analyse each standard solution in an identical manner to the samples and measure the peak area of the target compound. Plot the peak areas against the corresponding amine concentration of the standard, in $\mu g.ml^{-1}$, and construct the line of best fit. The slope of this line is the detector response (R_F) for the amine.

40 Modern HPLC equipment is usually sufficiently stable that a new calibration is not required with each set of samples. However, to verify the equipment, a quality assurance (QA) solution of known concentration must be analysed with each set of samples. The QA solution may be prepared using the procedure in paragraph 38.

Sample analysis

41 Analyse samples and blanks in an identical manner.

42 Desorb each filter in a sealed glass vial containing 2 ml of the desorbing solution. Rolling gently for 3 hours. Remove an aliquot and filter before analysis.

43 Add 10 ml of methanol to each wipe sample, recap and roll or shake gently for 3 hours. Remove an aliquot and filter before analysis.

44 Place each sorbent tube vertically in a suitable holder and add methanol to the top of the tube with a pipette. Collect the first 2 ml of methanol washings and filter an aliquot before analysis.

45 The calibration standards may be used to estimate analyte recovery from sampling media by spiking acid-coated filters, sorbent tubes and wipes (6 replicates of each recommended) at appropriate levels.

46 Analyse the sample solutions by HPLC. Measure the chromatographic peak area of the target compound and convert this peak area to an aromatic amine concentration, in µg.ml^{-1}, by dividing by the R_F value obtained from the calibration standards.

Calculation of results

Mass concentration of analyte in air samples

47 Calculate the volume, V_S, in litres, of each air sample using the procedure described in MDHS14.

48 Calculate the airborne concentration of aromatic amine, C, in mg.m^{-3}, using the equation:

$$C = D \times (M_S - M_B) / V_S$$

Estimation of mass of analyte on wipe samples

49 Calculate the mass of aromatic amine on the wipe sample, M_W, in µg, using the equation:

$$M_W = D \times (M_S - M_B)$$

Where:

M_S = Concentration of TGIC in sample, in µg.ml^{-1}

M_B = Mean concentration of TGIC in blanks, in µg.ml^{-1}

D = Volume of desorbing solution (2 ml for filters and sorbent tubes; 10 ml for wipe samples)

Appendix 1: Additional information

Detection limit of the method

1 The procedure is suitable for measurement of a range of airborne aromatic amines in the concentration range 50–5500 µg.m^{-3} for 10 litre air samples. For 10 litre air samples, the following nominal limits of detection have been determined:

Aromatic amine	Limit of detection (µg.m^{-3})
MDA	4.0
MbOCA	0.2
OCA	0.1
Aniline	3.8
o-toluidine	7.2

Overall uncertainty

2 For air samples the overall uncertainty for this measurement procedure as defined in BS EN 482[6] is less than 25%.

Interferences

3 Methylene diphenyl diisocyanates (MDI, CAS No. 101-68-8) will hydrolyse to MDA on the acid-coated filters if present, leading to a high or false positive result. Parallel isocyanate measurements[7] will be required to determine the extent of interference.

Stability

4 Filters may be stored in sealed containers for up to one week without sample loss. Wipe samples do not contain acid for amine stabilisation and should be desorbed as soon as possible after sampling. These extracts are stable on refrigeration for several weeks.

References

1 *General methods for sampling and gravimetric analysis of respirable, thoracic and inhalable aerosols* MDHS14/4 HSE Books 2014 www.hse.gov.uk/pubns/mdhs/index.htm

2 *Approved Classification and Labelling Guide (Sixth edition). Chemicals (Hazard Information and Packaging for Supply) Regulations 2009 (CHIP 4). Approved Guide* L131 (Sixth edition) HSE Books 2009 ISBN 978 0 7176 6370 5 www.hse.gov.uk/pubns/books/l131.htm

3 BS EN 1232:1997 *Workplace atmospheres: Pumps for personal sampling of chemical agents. Requirements and test methods* British Standards Institution

4 BS EN ISO 1042:2000 *Laboratory glassware: One-mark volumetric flasks* British Standards Institution

5 BS EN ISO 8655-6:2002 *Piston-operated volumetric apparatus. Gravimetric methods for the determination of measurement error* British Standards Institution

6 BS EN 482:2012 *Workplace atmospheres: General requirements for the performance of procedures for the measurement of chemical agents* British Standards Institution

7 *Organic isocyanates in air: Laboratory method with derivatisation in situ either on treated glass fibre filters or in solution using impingers with a treated back up filter in series, followed by high-performance liquid chromatography analysis* MDHS25/4 HSE Books 2014 www.hse.gov.uk/pubns/mdhs/index.htm

You should use the most current edition of any standards listed.

Further information

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