Determination of airborne fibre number concentrations

A recommended method, by phase-contrast optical microscopy (membrane filter method)

World Health Organization
Geneva
1997
Determination of airborne fibre number concentrations: a recommended method, by phase-contrast optical microscopy (membrane filter method).

1. Air pollutants, Occupational — analysis
2. Environmental monitoring — methods
3. Microscopy, Phase-contrast

ISBN 92 4 154496 1 (NLM Classification: WA 450)
Contents

Preface v
Outline of the method specification vii

1. Introduction 1

2. Scope of application 3

3. Specifications of parameters 5
   3.1 Sampling 5
      3.1.1 Filter 5
      3.1.2 Filter holder 7
      3.1.3 Storage and transport 9
      3.1.4 Sampling pump 9
      3.1.5 Optimal filter fibre loadings 10
      3.1.6 Flow rate 11
      3.1.7 Single sample duration 14
      3.1.8 Blanks 15
   3.2 Evaluation 16
      3.2.1 Sample preparation 16
      3.2.2 Optical requirements 22
         Microscope 22
         Eyepiece graticule 23
         Stage micrometer 25
         Test slide 26
      3.2.3 Counting and sizing fibres 27
         Low-power scanning 27
         Graticule field selection 28
         Laboratory working conditions 28
         Counting rules 29
      3.2.4 Calculation of fibre concentration 33

4. Accuracy, precision and lower limit of measurement 36
   4.1 Accuracy 36
   4.2 Precision 37
4.3 Lower limit of measurement

5. Quality assurance

References

Related reading

Annex 1. Static monitoring

Annex 2. Characterization of fibres

Annex 3. List of participants at the final meeting
Preface

The inhalation of airborne fibres in the workplace can cause a variety of occupational respiratory diseases, which contribute appreciably to morbidity and mortality among workers in both developing and developed countries. Monitoring airborne fibre concentrations is an important tool for occupational health professionals for assessing exposure and establishing the need to control it, evaluating the efficiency of control systems, and characterizing exposure in epidemiological studies. However, a proliferation of methods has hitherto hindered the comparability of results, as well as the possibility of having worldwide proficiency testing to ensure the reliability of obtained data.

A methodological framework that allows for meaningful comparisons between results obtained by different researchers and laboratories is of immense benefit for all aspects of occupational hygiene, but particularly for exposure assessment and environmental monitoring. In addition to proficiency testing, quality assurance schemes, comparisons and exchanges of data and international collaborative studies depend on the use of compatible methodologies. Furthermore, to ensure that preventive control systems in the workplace are adequate and effective, the reliability and comparability of monitoring and exposure data are essential. Occupational health surveillance and the ability to establish correlations between epidemiological and environmental indicators also depend on the ability to compare data from diverse sources.

A project aiming to establish a unified methodology for the evaluation of airborne fibres in the work environment was therefore carried out by the World Health Organization (WHO). A draft text of the present method specification was initially prepared by Dr N. P. Crawford, Institute for Occupational Medicine, Edinburgh, Scotland. During the course of four meetings of an international working group of experts (see Annex 3 for a list of participants at the final meeting),
current evaluation methods were compared and their differences identified and analysed, with a view to understanding the effect of these differences on the results of counting airborne fibres. Consensus was reached by the working group on a recommended method for the determination of airborne fibre number concentrations by phase-contrast optical microscopy. Dr Crawford's role as rapporteur at the final meeting and his work in revising the draft manuscript are gratefully acknowledged.

Collaboration from the International Labour Office (ILO), the European Commission (EC), the International Organization for Standardization (ISO), the European Committee for Standardization (CEN), the International Fibre Safety Group (IFSG), as well as national agencies, particularly the Health and Safety Executive (HSE), United Kingdom, and the National Institute for Occupational Safety and Health (NIOSH), USA, has been fundamental to this project and is also gratefully acknowledged. Special thanks are offered for the financial support provided by the EC and the IFSG.

Particular acknowledgement should also be made of the valuable personal contribution of several members of the working group, particularly Dr N. G. West (HSE), Dr P. Baron (NIOSH), and Mr S. Houston (IFSG), as well as Mrs B. Goelzer, Occupational Health, WHO.

In its first phase, this project has focused on methodology, so that authoritative scientific knowledge can be utilized to ensure accurate and precise measurements of airborne fibre number concentrations. In its second phase, the project will consider worldwide efforts for proficiency testing, quality assurance, and technical cooperation, including training and education.

Dr M. I. Mikheev
Chief, Occupational Health
World Health Organization
Outline of the method specification

Principle of the method
A sample is collected by drawing a known volume of air through a membrane filter by means of a sampling pump. The filter is rendered transparent ("cleared") and mounted on a microscope slide. Fibres on a measured area of the filter are counted visually using phase-contrast optical microscopy (PCOM), and the number concentration of fibres in the volume of air is calculated.

Sampling
Filter: Membrane of mixed esters of cellulose or cellulose nitrate, 0.8–1.2μm pore size, 25mm diameter.
Filter holder: Fitted with an electrically conducting cowl.
Transport: In closed holders.
Flow rate: 0.5–16 litres·min⁻¹. Adjust to give 100–650 fibres·mm⁻².
Blanks: Sampling media, 4% of filters. Field, ≥2% of samples. Laboratory, optional.

Sample preparation
Acetone-triacetin for fibres with a refractive index >1.51; stable for ≥1 year.
Acetone/etch/water for fibres with a refractive index ≤1.51; unstable.

Sample evaluation
Technique: Phase-contrast optical microscopy.
Microscope: Positive phase contrast, ×40 objective, ×400–600 magnification.
Walton–Beckett graticule, type G-22 (100 ± 2μm diameter).

HSE/NPL Mark II test slide.

Stage micrometers (1 mm long, 2-μm divisions).

Calibration: To meet visibility requirements of the test slide.

Analyte: Fibres (visual count).

Counting rules: Select counting fields at random, subject to defined criteria.

A countable fibre is >5μm long, <3μm wide and with a length:width ratio >3:1, subject to defined rules when it overlaps the graticule perimeter and when it touches other fibres or particles.

Lower limit of measurement: 10 fibres per 100 graticule fields.

Bias and reproducibility: See sections 4 and 5.
1. Introduction

Many countries have established personal exposure limits for airborne fibres in workplace atmospheres in terms of fibre number concentrations, i.e. the number of fibres in one millilitre of air. The method typically used to determine these concentrations for comparison with exposure limits is the so-called membrane filter method. However, experience has shown that this method does not always produce comparable results when used by different laboratories and different analysts. In fact, its precision is among the poorest of any occupational hygiene assessment method. Differences in results can arise through variations in sampling method, sample preparation, optical microscopy, the calculation of results and other factors, but particularly as a result of subjective effects associated with the visual counting of fibres. Such differences have both systematic and random components. The application of standard procedures and the establishment of a reproducible routine is the only way of controlling most of the sources of error inherent in the membrane filter method, which despite its limitations is the only method suitable for widespread international use in developed and developing countries.

Various specifications for the membrane filter method have been published by international, regional and national organizations. Differences in detail between the specifications are found when they are applied both to different fibre types and to the same fibre type (e.g. EEC, 1983; WHO, 1985; ISO, 1993; NIOSH, 1994). There are now fewer differences in the specifications than in the past, but such differences as still exist can have important systematic effects on results. The magnitude of this systematic variation depends on the sampling method, filter type and process employed. Further harmonization is needed to eliminate method specification as a source of variation and, with proper training and quality control, to ensure comparability between results produced by different microscopists and laboratories.

The method recommended by WHO, as set out in the main text of
this publication, relates to measurement of the number concentration of airborne fibres of all types for the purpose of assessing personal exposure in the occupational environment. Modifications are needed for application to static monitoring; these are described in Annex 1. These specifications have benefited from a review of the relevant literature previously prepared (Crawford, 1992).

Sampling strategies are not covered by this publication, but a training manual on this method, as well as sampling strategies specific to fibres, is envisaged. Sampling strategies have also been well covered in the specialist literature (e.g. NIOSH, 1977; ACGIH, 1991; AIHA, 1991; BOHS, 1993; NIOSH, 1994; ACGIH, 1995; CEN, 1995).

Reliable results depend on participation in a suitable quality assurance programme. The general requirements for the technical competence of testing laboratories published by the International Organization for Standardization (ISO, 1990) should therefore be followed. Microscopists should participate in intralaboratory counting checks, and laboratories should participate in a proficiency testing scheme.

It is hoped that this publication will motivate a review of the various specifications currently in use and that the method presented here will eventually be used by all countries, irrespective of the fibre type being assessed.
2. Scope of application

The method set out in the following pages is applicable to the assessment of concentrations of airborne fibres in workplace atmospheres—most commonly personal exposures—for all natural and synthetic fibres, including the asbestos varieties, other naturally occurring mineral fibres and man-made mineral fibres. The method can be used in sampling or monitoring carried out for the purposes of:

- comparison with occupational exposure limits
- epidemiology
- assessing the effectiveness of control measures and monitoring the effects of process modifications.

The method is appropriate for the above applications when information is required about the number concentration of airborne fibres. Occupational exposure limits for some fibres may also be expressed in gravimetric units (mg·m⁻³); in these cases, mass concentrations are measured by other methods referred to in *Safety in the use of mineral and synthetic fibres* (ILO, 1990). Unlike the membrane filter method, these gravimetric methods are not specific for fibres, since particles and non-countable fibres are included in the mass.

The method presented here measures the number concentration of airborne fibres, defined as objects with a length >5µm, a width <3µm and a length:width ratio (aspect ratio) >3:1, using a phase-contrast optical microscope.

Setting an upper width limit means that, for some fibre types, some wide fibres will not be counted. All fibres satisfying the dimensional criteria and counting rules defined in section 3.2.3 of this method should be counted.

Many fibres are too small to be visible by optical microscopy. The minimum visible width depends on the resolving power of the optical system, the difference in refractive index between the fibre and the surrounding medium, and the visual acuity of the microscopist. With
a good, correctly adjusted microscope which conforms to the specification of this method, the limit of visibility is in principle about 0.13–0.15μm. However, in practice, the smallest visible fibres will be about 0.2–0.25μm wide. Since some fibres fall below the limit of visibility, the PCOM fibre count represents only a certain proportion of the total number of fibres present (the exact proportion varies and depends on factors such as the sample type and the analyst). Thus the count represents only an index of the numerical concentration of fibres and is not an absolute measure of the number of fibres present.

Use of this method has other limitations when applied to samples containing “platy” (flat) or acicular (needle-shaped) particles and consequently should not be implemented without a full understanding of the workplace atmosphere (ISO, 1993). The method does not permit the determination of the chemical composition or crystallographic structure of fibres and therefore cannot be used on its own to distinguish between different fibre types. However, supplementary information on fibre type or size may be obtained by using other methods when necessary, such as polarized light microscopy, scanning electron microscopy and transmission electron microscopy. Annex 2 provides guidance on the scope and application of these methods. Such methods may be particularly appropriate when different fibre types are present in the same workplace and when airborne dust is a mixture of fibres and other types of particle. Supplementary methods are also useful for epidemiological studies, where a more detailed characterization of the properties of airborne contaminants is usually required.