Determination of Acrylamide Monomer in Waters and Polymers 1987

Methods for the Examination of Waters and Associated Materials

Her Majesty's Stationery Office

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Chromatographic methods are very sensitive to minor physical and chemical variations in the quality of the materials and apparatus used. Hence this method mentions the actual materials used for the evaluation tests. This in no way endorses these materials as superior to other similar materials. Equivalent materials are acceptable, though it must be understood that the performance characteristics may be different, and can vary with batch. It is left to the senior supervising analyst to evaluate and choose from the appropriate brands available.

Contents

About this Series

This booklet is part of a series intended to provide both recommended methods for the determination of water quality, and in addition, short reviews of the more important analytical techniques of interest to the water and sewage industries.

In the past, the Department of the Environment and its predecessors, in collaboration with various learned societies, have issued volumes of methods for the analysis of water and sewage culminating in 'Analysis of Raw, Potable and Waste Waters'. These volumes inevitably took some years to prepare, so that they were often partially out of date before they appeared in print. The present series will be published as series of booklets on single or related topics; thus allowing for the replacement or addition of methods as quickly as possible without need of waiting for the next edition. The rate of publication will also be related to the urgency of requirement for that particular method, tentative methods and notes being issued when necessary.

The aim is to provide as complete and up to date a collection of methods and reviews as is practicable, which will, as far as possible, take into account the analytical facilities available in different parts of the Kingdom, and the quality criteria of interest to those responsible for the various aspects of the water cycle. Because both needs and equipment vary widely, where necessary, a selection of methods may be recommended for a single determinand. It will be the responsibility of the users—the senior technical staff to decide which of these methods to use for the determination in hand. Whilst the attention of the user is drawn to any special known hazards which may occur with the use of any particular method, responsibility for proper supervision and the provision of safe working conditions must remain with the user.

The preparation of this series and its continuous revision is the responsibility of the Standing Committee of Analysts (to review Standard Methods for Quality Control of the Water Cycle). The Standing Committee of Analysts is a committee of the Department of the Environment set up in 1972. Currently it has 9 Working Groups, each responsible for one section or aspect of water cycle quality analysis. They are as follows:

1.0 General principles of sampling and accuracy of results
2.0 Microbiological methods
3.0 Empirical and physical methods
4.0 Metals and metalloids
5.0 General nonmetallic substances
6.0 Organic impurities
7.0 Biological monitoring
8.0 Sewage Works Control Methods
9.0 Radiochemical methods

The actual methods and reviews are produced by smaller panels of experts in the appropriate field, under the overall supervision of the appropriate working group and the main committee.

The names of those associated with this method are listed inside the back cover. Publication of new or revised methods will be notified to the technical press, whilst a list of Methods in Print is given in the current HMSO Sectional Publication List No 5.

Whilst an effort is made to prevent errors from occurring in the published text, a few errors have been found in booklets in this series. Correction notes and minor additions to published booklets not warranting a new booklet in this series will be issued periodically as the need arises. Should an error be found affecting the operation of a method, the true sense not being obvious, or an error in the printed text be discovered prior to sale, a separate correction note will be issued for inclusion in that booklet.

L R PITTWELL
Secretary

1 July 1987

Warning to Users

The analytical procedures given in this booklet should only be carried out by competent trained persons, with adequate supervision when necessary.

Local Safety Regulations must be observed.

Laboratory procedures should be carried out only in properly equipped laboratories.

Field Operations should be conducted with due regard to possible local hazards, and portable safety equipment should be carried.

Care should be taken against creating hazards for one's self, one's colleagues, those outside the laboratory or work place, or subsequently for maintenance or waste disposal workers. Where the Committee have considered that a special unusual hazard exists, attention has been drawn to this in the text so that additional care might be taken beyond that which should be exercised at all times when carrying out analytical procedures. Reagents of adequate purity must be used, along with properly maintained apparatus and equipment of correct specifications. Specifications for reagents, apparatus and equipment are given in manufacturers catalogues and various published standards. If contamination is suspected, reagent purity should be checked before use.

Lone working, whether in the laboratory or field, should be discouraged.

The best safeguard is a thorough consideration of hazards and the consequent safety precautions and remedies well in advance. Without intending to give a complete checklist, points that experience has shown are often forgotten include; laboratory tidiness, stray radiation leaks (including ultra violet) use of correct protective clothing and goggles, removal of toxic fumes and waste, containment in the event of breakage, access to taps, escape routes and the accessibility of the correct and properly maintained first-aid, fire-fighting, and rescue equipment. Hazardous reagents and solutions should always be stored in plain sight and below face level. Attention should also be given to potential vapour and fire risks. If in doubt, it is safer to assume that the hazard may exist and take reasonable precautions, rather than to assume that no hazard exists until proved otherwise.

There are numerous handbooks on first aid and laboratory safety. Among such publications are: 'Guide to Safe Practices in Chemical Laboratories' and 'Hazards in the Chemical Laboratory', issued by the Royal Society of Chemistry, London: 'Safety in Biological Laboratories' (Editors Hartree and Booth), Biochemical Society Special Publication No. 5, The Biochemical Society, London, which includes biological hazards; and 'The Prevention of Laboratory Acquired Infection' Public Health Laboratory Service Monograph 6, HMSO, London.

It cannot be too strongly emphasised that prompt first aid decontamination, or administration of the correct antidote can save life; but that incorrect treatment can make matters worse. It is suggested that both supervisors and operators be familiar with emergency procedures before starting even a slightly hazardous operation, and that doctors consulted after any accident involving chemical contamination, ingestion, or inhalation, be made familiar with the chemical nature of the injury, as some chemical injuries require specialist treatment not normally encountered by most doctors. Similar warning should be given if a biological or radiochemical injury is suspected. Some very unusual parasites, viruses and other micro-organisms are occasionally encountered in samples and when sampling in the field. In the latter case, all equipment including footwear should be disinfected by appropriate methods if contamination is suspected. If an ambulance is called or a hospital notified of an incoming patient give information on the type of injury, especially if poisoning is suspected, as the patient may be taken directly to a specialised hospital.

Determination of Acrylamide Monomer in Waters and Polymer

Introduction

Acrylamide polymers are used extensively as retention aids in paper making, boiler descalers and emulsifiers, to aid water clarification at water treatment works and for conditioning sludges, etc. The manufacture of these polyelectrolytes results in contamination of water and effluents by acrylamide monomer. The high chronic toxicity of acrylamide makes it undesirable in potable water supplies, the maximum recommended concentration being 0.25 μg/l.

The use of polyelectrolytes containing acrylamide monomer may lead to unacceptable levels in treated water since the monomer remains in aqueous solution after the polymer has coagulated suspended matter owing to its high solubility in water (2 g/l). Commercial polyelectrolytes used as coagulants in potable water treatment are not permitted to contain in excess of 0.05% of acrylamide monomer. Unregulated polyelectrolytes however, may be used for effluent or sludge treatment and these may contain up to 5.0% of monomer, although advances in the polymerization procedures have reduced monomer levels in unregulated polymers to less than 0.3% in many products. Note that some polyelectrolyte polymer treatment aids contain other acrylic derivatives such as esters, amines and acids in order to vary their properties. Many are copolymers.

Acrylamide, (CH_2: $CH.CO.NH_2$) official IUPAC name—Propenamide, is a white crystalline solid, melting at 84°C. For more information see Beilstein, 'Handbuch der organische Chemie', volume 2, second supplement p388.

This booklet contains two methods. One, Part A with a detection limit of 5 μg/l acrylamide, is intended for monitoring effluents prior to dilution by river or sewer; the other, Part B is a method for determining monomer in polymer. Use of more sensative electrochemical detectors may enable method A to achieve the maximum limits required by potable water suppliers which are of the order of 0.25 μg/l, but sufficient testing has not yet been possible.

A Determination of Acrylamide Monomer in Waters

A1 Performance Characteristics of the Method

A1.1	Substance determined.	Acrylamide monomer.
A1.2	Type of sample.	Natural and polluted waters.
A1.3	Basis of method.	Direct analysis by high performance liquid chromatography using ultraviolet detection. (See also A7.2).
A1.4	Range of application.	Direct analysis; 5 μg/l to 100 mg/l.
A1.5	Calibration curve.	Linear to 100 mg/l.
A1.6	Standard deviation.	0.5 mg/l synthetic 0.023 mg/l(9)
A1.7	Limit of detection.	5 μg/l for freshwater samples. (See also A7.2).
A1.8	Sensitivity	Depends upon the instrument used.
A1.9	Bias	Not determined.
A1.10	Interferences.	See Section A4.
A1.11	Time required for analysis.	20 minutes

A2 Principle

The methods described are based upon experimental work carried out at Plymouth Polytechnic. Acrylamide may be analysed directly by high performance liquid chromatography with ultraviolet detection following filtration and removal of interferences (mixed resin method) at concentration in excess of 5 μg/l. This method is not amenable to seawater analysis.

A3 Field of Application

The methods are applicable to (i) sewage effluents which may contain acrylamide as a residue from the manufacture of polyelectrolytes or the treatment of sludge with polyacrylamide flocculants, (ii) treated waters for which polyacrylamides were used as an aid to coagulation, and (iii) river water which may have received effluents flocculated with polyacrylamide or otherwise containing acrylamide.

A4 Interferences

Natural organic material which interferes by absorption at 196 nm or 202 nm must be removed with a mixed resin bed, (Section A9). Use of a second column type may also be tried. Interference by contaminants in the resin must be removed by washing with ethanol and regeneration with sodium chloride. Soxhlet extraction with both water and ethanol in succession may be necessary. Compounds absorbed onto glassware may interfere and must be excluded by soaking all glassware in chromic acid and rinsing with distilled water prior to use.

Distilled water prepared from tap water may contain acrylamide, thus it may be necessary to use spring or ground water for the preparation of distilled water. If this is not available, aeration followed by ultra-violet irradiation can be used prior to redistillation.

Acrylamide stock solutions should be prepared freshly for each batch of determinands as loss of acrylamide may occur from 1.0 mg/l standards.

A5 Hazards

Chromic acid is hazardous and requires care in its use. The procedure described uses acrylamide which is toxic. Avoid inhalation, skin contact and ingestion. Acrylamide affects the peripheral nerve endings and may cause dizziness and hallucinations.

A6 Reagents

All standards and solvents should be of Analytical Reagent or HPLC grade quality, unless otherwise stated.

A6.1 Amberlite resins XAD-2, IRA401 (Cl), IR120 (Na)7.1

The resins are washed with ethanol, sodium chloride (1M) and finally distilled water.

A6.2 Ethanol (absolute).

A6.3 Distilled water.

Prepared from water known to contain no acrylamide.

A6.4 Sodium chloride 1M.

Dissolve 7 ± 1 g sodium chloride analytical reagent grade in distilled water. Make up to 1 litre in a measuring cylinder.

A6.5 Acrylamide monomer.

Electrophoresis grade, greater than 99% pure.

A7 Apparatus

A7.1 A high performance liquid chromatograph fitted with a solvent delivery system capable of operating under constant flow, and with a remote sample injector loop of at least 110 μl capacity. The Chromatograph must be fitted with a reverse phase octadecyl silane (ODS) column (see Section A9, note a).

A7.2 Detector: A variable wavelength UV spectrophotmeter capable of analysis at 196 nm and 202 nm with a proven UV sensitivity to acrylamide as verified by trial prior to its use for quantitative work. The detector must be fitted with a chart recorder. An electrochemical detector may prove to be more sensitive, but the analyst should evaluate this. In which case lower limits of detection may be achieved.

A7.3 Microsyringe 100 ± 1μl of suitable design for injection onto a liquid chromatograph.

A7.4 Refrigerator operating at 4 ± 1°C (flame proof).

A7.5 Glass fibre filters, porosity 0.7μm, 7 cm diameter.

A7.6 Buchner filtration apparatus, 7 cm diameter.

A7.7 Glass ion exchange column 24 × 1 cm fitted with a glass sinter to retain the resin, and a solvent reservoir of 60 ml ± 5 ml capacity.

A8 Sample Collection and Preservation

Samples must be collected in 250 ml or larger glass stoppered bottles which have been previously cleaned by soaking in chromic acid, washed with distilled water and oven dried at 105°C. Samples should be stored in a refrigerator at 4 ± 1°C. If samples are to be stored for longer than 5–16 hours depending on likely microbial activity they should be boiled without loss of volatiles prior to storage.

Step	Procedure	Notes
	Determination of Retention Time	
A9.1	Dissolve 1.00 ± 0.01 mg acrylamide in 1 litre of distilled water. Inject 10 µl of this solution into the Hypersil ODS 100 mm × 0.5 mm column (note a and f) used with distilled water eluent and note the relative retention time. The instrument should be set to achieve a flow rate of 2 ml/min, detection wavelength 202 nm (note b) chart speed 0.5 cm/min.	(a) Only the Hypersil commercially available ODS column was found suitable for this analysis at the concentrations discussed, as acrylamide co-eluted with an unknown contaminant with the other columns tried. (but see method B). (b) A wavelength of 202 nm was used in preference to 196 nm (the wavelength of maximum absorbance maximum for acrylamide) because:— (i) For commercially available apparatus, with the high sensitivity setting of 0.004 Absorbance Units (fs) needed, at 196 nm, the UV source may provide insufficient quanta, which coupled with the reduced sensitivity of the photomultiplier and increased oxygen absorption from air may give poor baseline stability. Even though the acrylamide absorption is reduced at 202 nm, acceptable stability and sensitivity can be obtained. (ii) The elution peak of acrylamide lies close to those of certain inorganic anions such as nitrate. The use of 202 nm as a detection wavelength considerably reduced the interference from chloride. Interference from chloride at greater than 4,000 mg/l reduces the usefulness of the method for saline water. The interference from an unknown contaminant just after the acrylamide peak was also reduced at 202 nm. (c) A detector must be capable of giving a peak greater than 3 times base line noise for 100 µl injection of 5 µg/l acrylamine (sensitivity 0.004 aufs).
	Preparation of Calibration Standards	
A9.2	Using the acrylamide solution prepared in Step A9.1 (1 ml = 1 kg), by appropriate quantitative dilution with water, prepare a series of standard solutions containing 0.0, 5.0, 7.0, 10.0, 12.0, 15.0, 20.0, 50.0, 100 µg/l acrylamide.	
	General Procedure (see A9.8).	
	Preparation of the Exchange column	
A9.3	Prepare a mixed resin bed by filling the exchange column (A7.16) with distilled water, and adding equal volumes of anionic (note d), cationic and	(d) If nitrate causes problems use extra chloride form anion exchange resin. This resin must not be used in the nitrate form.

Step	Procedure	Notes
	finally hydrophobic resin to achieve approximately 8 cm column lengths of each resin. The resins should be added in the above order as this relates to their decreasing densities. Insert a glass wool plug at the top of the column.	
A9.4	Filter each solution through a glass fibre filter (note e).	(e) Glass fibre filters must be used. Cellulose filters tend to leach detectable traces of acrylamide owing to the use of cationic polyacrylamides as retention aids in paper making. A pore size of 0.7 μm is specified since larger pore size filters allow unacceptable quantities of fines through, which will adversely effect the performance of the injection valve and column.
A9.5	Pass the solution through the mixed resin bed (note f), discard 1st bed volume, collect 2nd bed volume, flush column with at least 3 bed volumes of distilled water prior to the next analysis.	(f) The column should be checked at appropriate intervals (dependent of type of sample being analysed) for retention of acrylamide by passage of a 5 μg/l acrylamide standard followed by the appropriate analysis. Should any retention of acrylamide be noted or if the interference removal efficiency falls, the column should be regenerated using ethanol, and sodium chloride 1M. Should this clean up prove unsatisfactory, replacement by another column is advised. Flow rate through the column is limited by the packing, flows of between 2 and 10 ml/min have proved suitable for the analysis.
A9.6	Inject a 10.0 ± 0.1 to 100.0 ± 0.1 μl aliquot of the percolate into the liquid chromatograph under the instrument settings given for step A9.1. (note g).	(g) For maximum sensitivity inject 100 μl for an absorbance range of 0.004 Au full scale. At this high sensitivity the detector response may need to be adjusted to bring it on scale prior to the acrylamide peak.
A9.7	Measure the height of the peaks (note h).	(h) Peak area may be used as an alternative.
A9.8	Carry out steps A9.4 to A9.7 with at least duplicate portions of sample and calibration standards, noting the amount injected at step A9.6 in each case.	
A9.9	Convert the peak height or area measurement to a theoretical injection of 100 μl at an appropriate sensitivity by multiplying by $\frac{100}{\text{volume injected}}$	
A9.10	Using data from the calibration standards, prepare a calibration graph of acrylamide concentration versus response.	
A9.11	Read the concentration of acrylamide in the samples analysed from the calibration curve.	

B Determination of residual Acrylamide Monomer in Polyelectrolytes

B1 Performance Characteristics of the Method

B1.1	Substance determined	Acrylamide monomer
B1.2	Type of sample	Commercial polymers based on polyacrylamide
B1.3	Basis of method	Solvent extractions, separations using High Performance Liquid Chromatography with detection by Ultraviolet absorption at 215 nm.
B1.4	Range of application	Tested up to 100 mg/l in extract, up to 0.1% in the original sample.
B1.5	Calibration curve	Linear to at least 100 mg/l in extract.

B1.6 Stndard deviation

For HPLC Stage only

Acrylamide Concentration mg/l	Standard deviation mg/l	Degrees of freedom
0.5	0.021	9
20	0.242	7
40	0.628	7
Typical sample (approx. 20)	0.283	7
Spiked sample (approx. 40)	1.955	7

Information obtained using liquid standards and solid samples spiked with standards before extraction.

B1.7	Limit of detection	0.1 mg/l in extract
B1.8	Bias	Not known
B1.9	Interferences	Compounds eluting at the same time as the acrylamide monomer. These will be dependant on the nature of the sample and the chromatographic conditions used.
B1.10	Time required for analysis	The total analytical time for 1 sample is approximately 25 hours. Operator time is about 1 hour. For 8 samples the operator time is about 4 hours.

B2 Principle

B2.1 The method described is that used by the Water and Waste Water Sub-Division of the Laboratory of the Government Chemist.

B2.2 Acrylamide monomer is extracted from the polyelectrolyte by the procedure described in the WRA Paper TIR 171. The acrylamide is then determined by High Performance Liquid Chromatography using a reverse phase system and ultraviolet detection at 215 nm. Identification is made by comparison with an external standard and concentration determined by comparison with a calibration curve. By varying the chromatographic conditions interferance by co-eluting compounds may be minimized.

B3 Hazards

Acrylamide is toxic and should be handled with care, avoid inhalation, skin contact and ingestion.

B4 Reagents

Only analytical reagent grade or HPLC grade chemicals should be used unless otherwise stated.

B4.1 Water

The water used for blank determinations and for preparing standards and solvents should contain a negligible amount of acrylamide, or substances of similar retention time. Deionized water and double distilled water have been found satisfactory. (But see first method Section A4).

B4.2 Methanol HPLC Grade

B4.3 Methanol/Water Solvent (80/20 v/v)

Thoroughly mix, without violent agitation, 800 ml of methanol with 200 ml of water, degas by use of an ultrasonic bath or by passing nitrogen through the mixture and store in an amber glass reagent bottle.

B4.4 Acrylamide Standard Solutions

B4.4.1 Stock Solution 1 g/l

Dissolve 0.100 ± 0.001 g, of acrylamide (GPR) in 50 ml of methanol/water solvent (B4.3), then transfer the solution to a 100 ml volumetric flask and make up to the mark with the solvent. Store in a tightly-stoppered glass reagent bottle in a refrigerator. The solution is stable for at least four weeks.

Calibration Standard Solutions

B4.4.2 Solution A 50 mg/l

Pipette 5 ml of stock standard solution (4.4.1) into a 100 ml volumetric flask and make up to the mark with methanol/water solvent (B4.3). This solution should be prepared freshly each day.

B4.4.3 Solution B 100 mg/l

Pipette 10 ml of stock standard solution (B4.4.1) into a 100 ml volumetric flask and make up to the mark with solvent (B4.4.3).

B4.4.4 Other concentrations (for example 0, 10 and 20 mg/l) may be required for checking the linearity of the calibration. These are prepared by similar quantitative dilution (by pipetting 0, 1 and 2 ml respectively and diluting to 100 ml as above).

B5 Apparatus

B5.1 Glassware—all glassware should be thoroughly cleaned using a suitable detergent solution, then rinsed with water and finally acetone and dried in an oven at about 200°C.

B5.2 Pipettes should be grade A with a suitable filling device.

B5.3 Microlitre syringe, 10 μl capacity.

B5.4 Glass vials (approx 40 ml capacity) with PTFE lined screw caps.

B5.5 Laboratory Shaker

B5.6 A high performance liquid chromatograph fitted with a constant flow solvent delivery system and a remote sample injection valve of 20 μl capacity.

B5.7 Detector. A variable wavelength UV spectrophotometric detector capable of analysis at 215 nm or an adjacent suitable wavelength and operated according to manufacturer's instructions.

B5.8 Chart recorder, connected to the detector.

B5.9 Stainless steel HPLC column, 250×4.6 mm packed with a suitable 5 μm Octadecyl silane (ODS) support material (Spherisorb S 5 ODS has been found to be suitable).

B6 Sample Pretreatment

Step	Procedure	Notes
	Sample Extraction	
B6.1	Weigh 1.00 of air dried sample into a 40 ml glass vial (B5.4) and pipette in 10 ml of methanol/water solvent (B4.3). Shake vigorously on a mechanical shaker (B5.5) for 24 hours (Note a) and allow to settle. Filter off the liquid, through a cotton wool plug in a funnel, and store the liquid in another vial in a refrigerator until required for analysis (Note b).	(a) This procedure has been found to be satisfactory for a variety of polyelectroytes but may not necessarily be the most suitable for all the many different types of polyelectroloyte products now available. (b) The sample is stable for at least a week under these conditions.

B7 Analytical Procedure

Step	Procedure	Notes
B7.1	Inject 10 μl of sample extract solution into the HPLC column via the injection valve (Note c). The eluent used is water at a flow rate of 1 ml/min and detection wavelength 215 nm (Note d).	(c) Smple residues tend to build up on the column and the efficiency of the column deteriorates. Therefore providing satisfactory precision can be obtained and the concentration of acrylamide is sufficient, smaller injection volumes may be used for instance 5 μl.
B7.1.1	Measure the height of the peak which has a retention time corresponding to that of the acrylamine standard Calculate the concentration of acrylamide from the calibration graph. (Section B8)	(d) The maximum absorbance wavelength for acrylamide is 196 nm but for filter spectrophotometers the nearest filter is 215 nm. Use the wavelength giving the best sensitivity. See also A9.11(note b).
	Blank Determination	
B7.2	Repeat steps (B7.1) and (B7.1.1) omitting the sample and substituting (Note e), the methanol/water solvent (B4.3).	(e) The detection limit and accuracy of the result depends on the condition and nature of the column at the time, the injection volume (see Note c), the level of interfering substances in blanks) and also upon the variability of the blanks. Therefore at least two blanks should be analysed with each batch of samples. The blank analysis must be carried out in an identical manner to that used for the samples.
	Calibration Standard	
B7.3	A calibration standard should be run after every four or five samples to check the HPLC column's performance. Prepare a 50 mg/l standard solution as described in section B8 for this purpose.	

B8 Preparation of the Calibration Curve

Initially the linear range of the method should be established by the analyst. A series of calibration standard solutions is prepared from the stock standard (B4.4.1) (See B4.4.2–B4.4.4). The solutions thus prepared usually contain 0, 10, 20, 50, 100 mg/l of acrylamide.

Inject each of these solutions into the HPLC and measure the peak height as for the sample extracts. Construct a calibration curve by plotting peak height against concentrations of acrylamide.

In subsequent batch analysis it will only be necessary to check the slope of the calibration curve by the use of a single standard in the upper range of sample concentrations, provided it is within the linear range. Standards of 50 or 100 mg/l are normally used. (See section B7.3)

B9 Calculation of Results

The peak height of the HPLC peak corresponding to acrylamide is measured and using the calibration graph described above (Section B8) the concentration of acrylamide in the sample extract may be found. Any blank values must be deducted from these results. The concentrations of acrylamide monomer in the original sample may be calculated as follows:

$[(\text{Conc. in extract}) - (\text{Blank conc.})] \times 10$ mg/kg.

Sources of Error (applicable to both methods)

The attention which it is necessary to pay to sources of error depends on the accuracy required of the analytical results. The following sub-sections summarize the main sources of error.

1 Contamination

It is desirable to carry out the analysis in a laboratory in which no appreciable amounts of acrylamide or its polymers are handled. The technique and working conditions should be critically examined and any sources of contamination eliminated or minimized. In particular, it is desirable to reserve the glass apparatus used for these determinations solely for this purpose and to carry out a preliminary series of blank determinations to ensure low blank values before analysing any samples.

2 Acrylamide Monmomer Content of the Water used for Blank Determinations

If the water used for the blank determinations contains acrylamide the results will be falsely low. Ideally the concentration in the water used for each blank determination should be measured and an appropriate correction made.

Whether the acrylamide content of the water used is negligeable may be ascertained by obtaining a sample of water known from its origin or pretreatment to be free from acrylamide, and comparing analyses.

3 Interfering substances

The effect of possible interfering substances may be determined by analysing samples spiked with acrylamide and various concentrations of the potential interfering substances.

4 Calibration standards

The procedure assumes a linear calibration curve and the linearity must be checked. A calibration standard should be run for each batch of analyses.

Estimation of the Accuracy of Analytical Results

1 Introduction

Quantitative investigation of the accuracy achievable when these methods are used appears to be limited to work in only a few laboratories. Before firmly recommending the method for general use, it is desirable to know the accuracy achievable in other laboratories. It would, therefore, be of great value if any laboratory using or considering the use of this method could estimate the accuracy of its own analytical results and report the findings to the Secretary of the Department of the Environment's Standing Committee of Analysts.

The precision achieved and the effects of any interfering substances that may be present in samples are of particular interest. Any information on these aspects would be useful, but the value of such information would be greatly enhanced if it were obtained to a common plan so that the information can be compared and valid conclusions drawn. Accordingly, suggestions for a suitable experimental design and analysis of results are given in the following sections and it is strongly urged that laboratories follow this design whenever possible. The design has been chosen to be as simple as possible; more complex designs are possible and would give more information.

2 Basis of suggested Tests

The limit of detection is governed by the within-batch variability of blank determinations. The precision of analytical results may depend on the concentration of acrylamide in the sample analysed and on the type of sample, eg worse precision may be obtained with samples than with standard solutions. For these reasons the basic design recommended is the analysis of one portion of each of the following solutions on each of n days, where n is at least 5 and preferably up to 10.

Solution No	Description
1	Blank
2	Another blank
3	Standard solution (low range)
4	Standard solution (high range)
5	Typical sample
6	Same sample spiked

It is essential that these solutions be treated exactly as if they were samples and the procedure specified in the method be rigidly followed. These solutions should be analysed in random order in each batch of analyses. Solutions 1 to 4 should be prepared each day exactly as described in the method and should always contain the same concentrations of acrylamide. The same batch of water should be used on each day to prepare all four solutions. For solutions 5 and 6 a total of 5 litres of typical sample are required. Prepare solution 6 each day when required by spiking solution 5.

When analysing solution 6 it may be necessary to take an appropriately smaller aliquot. The total period of the tests may be any convenient time so long as the concentration of solution 5 does not change appreciably. The results of the analyses of solutions 5 and 6 will provide a check on the effect of sample type on precision. Any deviation of the recovery of spike addition from 100% may give an indication of the presence of interfering substances.

3 Evaluation of Results

The raw experimental results should be sent direct to the Department of the Environment for evaluation together with the results obtained from the standards used to

establish the calibration curve in each batch of analyses. However, for those laboratories wishing to make the calculations themselves, the details are given below.

3.1 Convert all results to concentrations as described in the method. Deduct the first of the two blank values (solution 1) from each of the other solution values.

3.2 Calculate the mean concentration of the n results for each solution.

3.3 Calculate the standard deviation, s, of the n results for each solution from:

$$s = \sqrt{\frac{(x_i - \bar{x})^2}{n-1}}$$

where x_i = the result from the ith batch

$\bar{x}$ = the mean value of x_i.

3.4 Calculate the within-batch standard deviation, s_w, of the blank from:

$$s_w = \sqrt{\frac{(x_{1i} - x_{2i})^2}{2n}}$$

where x_{1i} = the 1st blank result (solution 1) from the ith batch

x_{2i} = the 2nd blank result (solution 2) from the ith batch.

3.5 Calculate the mean percentage recovery, R, of the spike in solution 6 from:

$$R = \frac{(\bar{x}_6 - \bar{x}_5)}{10} \times 100$$

where $\bar{x}_5$ = the mean value of the results for solution 5

$\bar{x}_6$ = the mean value of the results for solution 6.

3.6 Summarize the results as in the following table:

Solution	No of results n	Mean Concentration µg/l	Standard Deviation µg/l	Mean Recovery %
2 Blank				—
3 Standard,				—
4 Standard,				—
5 Sample.....				—
6 Solution 5+				

The appropriate sample description should be entered in the space for solution 5. The standard deviation from step 3.4 is entered for the blank solution 2 and the standard deviations from step 3.3 are entered for solutions 3 to 6.

4 Checking the Accuracy of Analytical Results

Once the methods have been put into normal routine operation many factors may subsequently adversely affect the accuracy of the analytical results. It is recommended that experimental tests to check certain sources of inaccuracy should be made regularly. Many types of tests are possible and they should be used as appropriate. As a minimum, however, it is suggested that a standard sample of suitable concentration should be analysed at the same time and in exactly the same way as normal samples.* The results obtained should then be plotted on a quality control chart which will facilitate detection of inadequate accuracy, and will also allow the standard deviation of routine analytical results to be estimated.

*As acrylamide monomer solutions are not very stable, it is suggested that the standard be prepared afresh when required and that an additional allowance be made for variation of the standard.

5 Polymer Method

For polymers the analysis can be checked in two sections: Extraction—Section B6, for which use of several different grades of polymer could be used; and the main analysis—Sections B7, 8 and 9, which can be tested as for water, using synthetic extracts of known concentration.

Address for Correspondence

However thoroughly a method may be tested, there is always the possibility of a user discovering a hitherto unknown problem. Users with information on this method are requested to write to:

The Secretary
The Standing Committee of Analysts
The Department of the Environment
Romney House
43 Marsham Street
LONDON SW1P 3PY
England

Department of the Environment

Standing Committee of Analysts

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