ASPEC® 274: Automated Extraction of Acrylamide from Underground Water Prior to HPLC-DAD Analysis

APPLICATION NOTE AN1026

APPLICATION BENEFITS

Acrylamide is a known neurotoxin and a potential carcinogen. The World Health Organization has established 0.5 µg/L as the maximum limit of acrylamide in drinking water. This application note describes an alternative to tedious and time-consuming trace analysis by gas chromatography.

SOLUTIONS

This application note describes a sample concentration method by solid phase extraction (SPE) using the Gilson ASPEC 274 system. Water contaminants were concentrated 150-fold using SPE cartridges and analyzed by HPLC. An assay with a limit of quantification of 0.3 µg/L was developed with this technology.

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INTRODUCTION

Acrylamide is a synthetic compound widely used in the plastics industry and in water treatment. It is considered by the World Health Organization (WHO) as a potential carcinogen and therefore the maximum level allowed in water for human consumption is limited to 0.5 µg/L[1]. The Brazilian water legislation, regulated by the Brazilian Health Regulatory Agency (ANVISA), in Resolution RDC 274, established the same maximum allowed level for acrylamide defined by WHO. In order to achieve such a low limit of quantification, a sample pre-concentration step is necessary before determination of the acrylamide content.

Most of the acrylamide produced is used as a chemical intermediate or as a monomer in the production of polyacrylamide. Both acrylamide and polyacrylamide are used mainly in the production of flocculants for the clarification of potable water and in the treatment of municipal and industrial effluents. They are also used as grouting agents in the construction of drinking-water reservoirs and wells. Acrylamide is highly mobile in aqueous environments and readily leachable in soil. As it has a higher mobility and lower rate of degradation in sandy soils than in clay soils, it may contaminate groundwater. The most important source of drinking-water contamination by acrylamide is the use of polyacrylamide flocculants containing residual levels of acrylamide monomer[1].

This application note describes a sample preparation method by solid phase extraction (SPE) using the Gilson ASPEC 274 system (Figure 1). Method validation was done by the evaluation of the following metrics: limit of quantification, repeatability, reproducibility and recovery. A limit of quantification of 0.3 µg/L was achieved.

MATERIALS & METHODS

Sample concentration by solid phase extraction

Samples were collected in 250 mL amber glass flasks and stored at 2°C–8°C. A control sample of 0.3 µg/L acrylamide was prepared in 150 mL of ultrapure water. CHROMABOND SPE cartridges (6 mL, 500 mg porous graphitic carbon phase, 730512) and collection tubes were arranged in a Gilson Code 386 rack. Each collection tube contained 0.5 mL of ultrapure water to avoid recovery loss. The SPE cartridge was conditioned with 10 mL of methanol and 10 mL of ultrapure water at 5 mL/min. A sample volume of 150 mL was loaded at 1 mL/min and the column was eluted with

9.3 mL of methanol at 0.6 mL/min. The eluates were concentrated in a TurboVap LV (Caliper Life Sciences) at 62˚C with 10 psi N_2. Samples were concentrated to less than 1 mL and the volume adjusted to 1.0 mL with ultrapure water and transferred to 1.5 mL vials.

Chromatographic analysis by HPLC-DAD

Vials containing concentrated sample were transferred to an Ultimate 3000 RS autosampler. A control sample was included for each sample sequence for evaluation of the extraction. HPLC was conducted on an Ultimate 3000 RS UHPLC system (Thermo Fisher) with an Acclaim C18 column (250 mm x 4.6 mm, 5 µm particle size) with water/methanol (60:40 v/v) at a flow rate of 0.8 mL/min at 35˚C. The run was monitored with a DAD detector (Thermo Fisher) at 202 nm.

RESULTS AND DISCUSSION

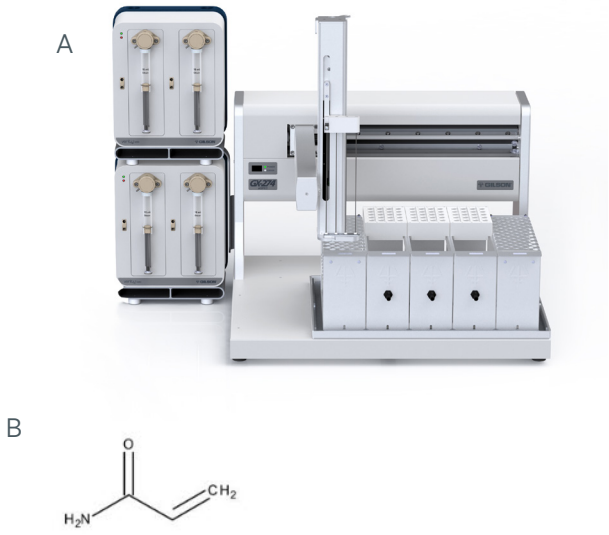

Figure 1
(a) Gilson ASPEC® 274 and
(b) chemical structure of acrylamide

The ASPEC 274 (Figure 1a) system was used to sufficiently concentrate acrylamide (Figure 1b) to levels detectable by HPLC. The positive control HPLC chromatogram is a 0.3µg/L sample of acrylamide in ultrapure water which was concentrated using an ASPEC 274. HPLC analysis of the concentrated sample detected acrylamide at 4.2 min (Figure 2). A negative control consisting of an environmental sample showed that acrylamide was not detected (Figure 3).

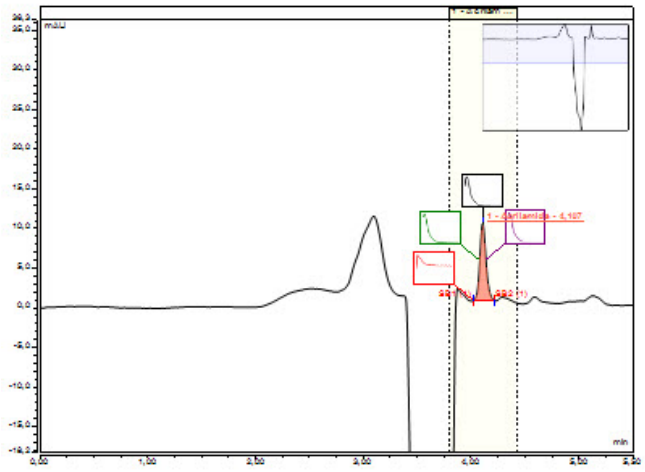

Figure 2
HPLC chromatogram at 202 nm of a control sample of acrylamide at 0.3 µg/L in demineralized water

The highlighted boxes represent the spectra of the peak at: baseline (red), 50% before apex (green), apex (black), and 50% height after apex (purple). Polar organic compound present in the matrix was detected at 3 minutes.

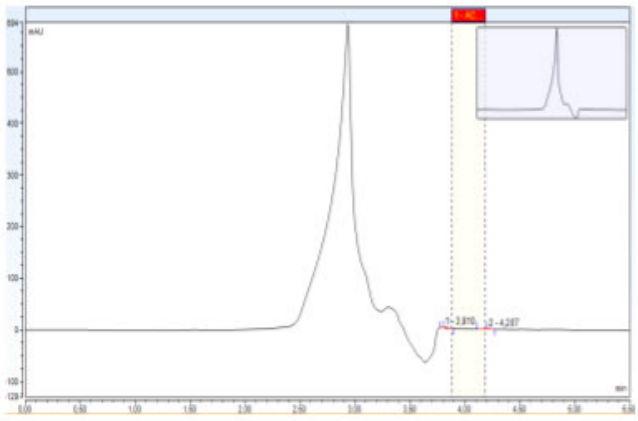

Figure 3
Chromatogram of an environmental sample without acrylamide at 202 nm

The limit of quantification, repeatability, reproducibility, and recovery of the developed method were determined in accordance with the guidelines on validation of analytical methods in the document DOQ-CGCRE-008 INMETRO[3]. Each of these parameters were determined experimentally with seven independent acrylamide solutions that were subjected to the described solid phase extraction, volume reduction, resuspension, and chromatographic analysis. The limit of quantification was measured using a solution of 0.3 µg/mL acrylamide. The recovery and the relative standard

deviation (RSD) were equal to 93.3% and 5.7% respectively. As the recovery and RSD values met the criteria previously established by the laboratory, the limit of quantification of the method was established as being equal to 0.3 µg/L. The repeatability was evaluated with 0.5 µg/L acrylamide and analyzed by the same analyst. The recovery and the relative standard deviation were 89.5% and 7.0%, respectively. The determination of intra-laboratory reproducibility was performed by different analysts on different days with 0.5 µg/L acrylamide. The recovery and the relative standard deviation were 90% and 6.8%, respectively (Table 1).

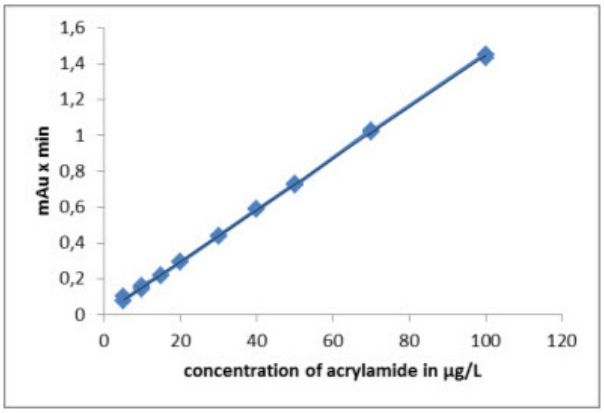

Figure 4
Calibration curve of acrylamide in water

Table 1
Summary of LOQ, repeatability, and intra-laboratory reproducibility of the SPE-based acrylamide assay

Test	% Recovery	RSD
Limit of quantification	93.3	5.7
Repeatability	89.5	7.0
Intra-laboratory reproducibility	90.0	6.8

CONCLUSIONS AND BENEFITS

- The ASPEC 274 is ideal for concentration of samples for improved sensitivity
- Accommodates large volumes – up to liters of sample can be processed
- Four probe Z-Arm: Process four SPE cartridges simultaneously

REFERENCES

1. Acrylamide in Drinking-water – Background document for development of WHO Guidelines for Drinking-water Quality. World Health Organization (2003).
2. KONTOMINAS, M.G.; PALEOLOGOS, E.K. - Determination of acrylamide and methacrylamide by normal phase high performance liquid chromatography and UV detection. Journal of Chromatography A. nº 1077, pgs 128-135. 2005.
3. Orientação sobre validação de métodos analíticos. DOQ-CGCRE-008, Revisão 4, 2011. Instituto Nacional de Metrologia INMETRO.

ACKNOWLEDGEMENTS

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ORDERING INFORMATION

Description	Part Number	Quantity
Gilson ASPEC 274 with two VERITY 4260 Dual Syringe Pumps	2614010	1
VALVEMATE II -GSIOC	331052AB	4
Valve, Prep, Multi Pos, 10 Port, PPS .06	49400006	4
Rack 338, Aluminum, 64 vials, 12 x 32 mm (2 mL)	260440106	1
Rack 345, Aluminum, 44 tubes, 16 x 150 mm	260440041	2
Extraction Rack 386, Aluminum,16 (6 mL) SPE Cartridges and 15 Tubes, Collection block holds 15 x 85 mm (10 mL) Tubes	260440109	1
TRILUTION LH Software, LIFETIME	21063023	1
Probe, 221 x 1.5 x 1.1 mm CON BEV .45 ID Tip	27067374	4