



# A Rapid iMethod<sup>™</sup> Test for the Quantification of Acrylamide in Food

## iMethod™ Test for Acrylamide Version 1.0 for Cliquid<sup>®</sup> Software

Acrylamide has been classified as "a probable carcinogenic to humans" by the International Agency for Research on Cancer (IARC). The carcinogenic effect of acrylamide has been proven in different animal studies. While it is not entirely known how acrylamide is produced, Tareke et al. (2002) have found increased amounts of acrylamide in many starch enriched food such as potatoes, beets and potato chips.

The following description outlines the instrument requirements and expected results obtainable from the AB SCIEX iMethod<sup>™</sup> Test for the Analysis of Acrylamide in Food, when using an AB SCIEX 3200 Series instrument. This method has also been developed and verified for use with AB SCIEX 4000 Series instrumentation. More in-depth sample preparation and instrument parameter information is included as part of the standard operating procedure provided with the acrylamide iMethod test upon purchase.

Example sample preparation procedures are provided for milk and starch-enriched foods. While the actual protocol outlined is dependent upon the matrix to be used, most protocols are based upon a simple sample homogenization, centrifugation, extraction and dilution. A deuterated internal standard of acrylamide at a known concentration is added during sample preparation to monitor sample recovery.

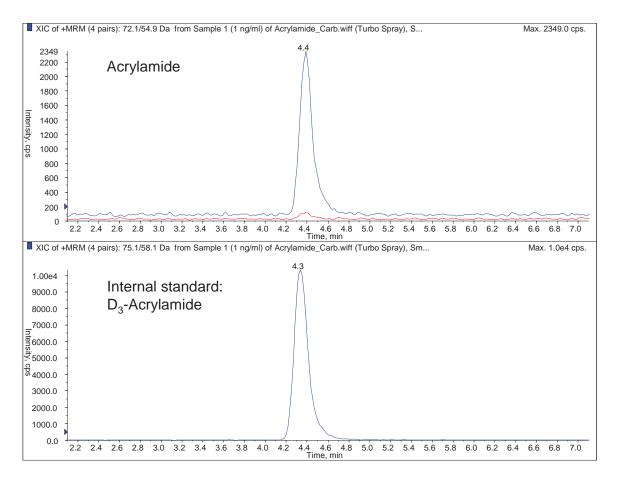


Figure 1: Chromatogram of a 1 ng/mL acrylamide spiked analyzed by LC/MS/MS.



## Results

An example chromatogram of acrylamide is shown in Figure 1; retention times and the MRM transitions used are listed in Table

1. The superior sensitivity of the method is highlighted by the limits of detection shown (< 1 ng / mL).

Table 1: Multiple Reaction Monitoring (MRM) Transitions, Retention Times, and Limit of Detection (LOD) based on the Qualifier MRM Transition and Coefficient of Variation (n=3) of all Detected Substances

Compound	MRM 1	MRM 2	RT (min)	LOD
Acrylamide	72 / 55	72 / 44	4.1	<1 ng/mL
D <sub>3</sub> -Acrylamide	75 / 58			

# Calibration

The following calibration curves using the calibrator, low and high level controls are provided as examples, showing the range and linearity expected for the assay.

Please note that the results were obtained using a single instrument and single set of standards and samples, and the results here may not be typical for all instruments. Prior to production use, the method should be fully validated with real samples. Variations in LC column properties, chemicals, environment, instrument performance and sample preparation procedures will impact performance, thus these results should be considered as informative rather than representative.

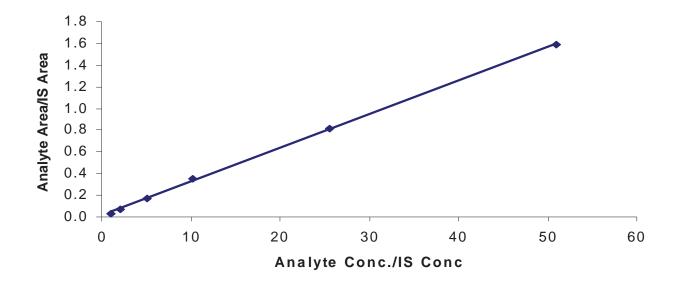


Figure 2: Representative calibration curve for acrylamide.

## **System Requirements**

In order to run this method as outlined above, the following equipment and reagents are required:

- An AB SCIEX 3200 Series (3200 QTRAP<sup>®</sup> or API 3200<sup>™</sup>) or 4000 Series (4000 QTRAP<sup>®</sup> or API 4000<sup>™</sup>) LC/MS/MS System
- A Shimadzu Prominence 20A LC System with reservoir tray and bottles, system controller CBM-20A, 100 µL mixer, 2 isocratic pumps LC-20AD, 3-channel degasser autosampler SIL-20AC, column oven CTO-20AC or Agilent 1100/1200 LC system with binary pump G1312A (without static mixer), well plate autosampler, and thermostated column oven
- D<sub>3</sub>-Acrylamide internal standard (www.sigmaaldrich.com)
- · LC/MS-grade water, methanol, and formic acid
- A Phenomenex Luna 3µ C18(2) 150 x 3 mm column
- Potassium hexacyanoferrate(II) trihydrate and zinc sulfate heptahydrate sample preparation solutions (www.sigmaaldrich.com)
- 1.5 mL Eppendorf tubes
- A centrifuge able to accommodate Eppendorf tubes and run at 14000 rpm
- · Pipettes and standard laboratory glassware

• Acrylamide standard (www.sigmaaldrich.com)



# **Ordering Information**

Product Name	Part Number
iMethod <sup>™</sup> Test for Acrylamide Version 1.0 for Cliquid <sup>®</sup> Software	1034371

#### **Important Note**

The iMethod<sup>™</sup> test described above has been designed by AB SCIEX to provide the sample prep and instrument parameters required to accelerate the adoption of this method for routine testing. This method is provided for information purposes only. The performance of this method is not guaranteed due to many different potential variations, including instrument performance, tuning, and maintenance, chemical variability and procedures used, technical experience, sample matrices, and environmental conditions. It us up to the end user to make adjustments to this method to account for slight differences in equipment and/or materials from lab-to-lab as well as to determine and validate the performance of this method for a given instrument and sample type. Please note that a working knowledge of Analyst<sup>®</sup> Software may be required to do so.

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