Standards for qNMR

qNMR Standards for External and Internal Referencing

Quantitative ^{1}H-NMR (qNMR) continues to be utilized with much success in the pharmaceutical, chemical and food industries and in many facets of academic research. Regardless of the application, all qNMR methods require a calibration signal whose integrated signal intensity originates or is traceable to a known number of protons. Calibration for qNMR is made using either internal or external referencing methods. External methods rely on the use of a standard solution packaged in a defined NMR tube or capillary to obtain an integral that can be used for sample quantification, whereas internal methods rely on the use of a known amount of standard that is co-dissolved in the sample itself.

External Calibration Standards

CIL is pleased to offer external calibration standards for qNMR. The standards are formulated using CIL's high-quality DMSO-d_6 and benzoic acid from NIST (SRM 350(b)), a standard reference material for acidometry. Both 5 mM and 15 mM benzoic acid concentrations are available. The concentration and associated expanded uncertainty of the benzoic acid has been accurately determined using metrological techniques and verified using qNMR. The ^{1}H-NMR spectrum of benzoic acid in DMSO-d_6 is presented in Figure 1.

CIL is currently offering these standards in presealed NMR tubes. Please see the information below for details regarding NMR tubes and fill volumes. Other NMR tubes and concentrations may be available upon request.

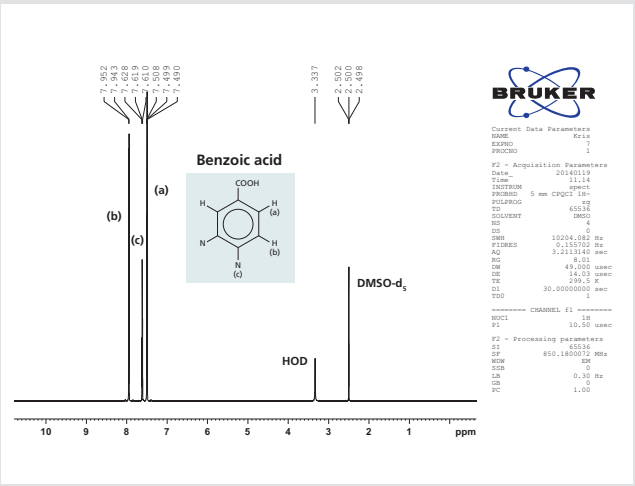


Figure 1. 850 MHz ^{1}H-NMR spectrum of benzoic acid in DMSO-d_6. Resonances from the aromatic protons of benzoic acid, HOD and DMSO-d_5 are assigned. The acid proton resonance from benzoic acid (~12-13 ppm) is not shown. (Courtesy Joe Ray, Baxter Healthcare Corporation, Round Lake, IL)

qNMR Standard for External Referencing

Catalog No.	Description*	NMR Tube**	Part No.	Fill Volume
DLM-9491A	5 mM Benzoic acid in DMSO-d_6	1.7 mm O.D.	Bruker Part No. Z106462	50 µL
DLM-9491B	5 mM Benzoic acid in DMSO-d_6	3 mm O.D.	Wilmad Part No. 335-PP-9	160 µL
DLM-9491C	5 mM Benzoic acid in DMSO-d_6	5 mm O.D.	Wilmad Part No. 528-PP-8	750 µL
DLM-7061A	15 mM Benzoic acid in DMSO-d_6	1.7 mm O.D.	Bruker Part No. Z106462	50 µL
DLM-7061B	15 mM Benzoic acid in DMSO-d_6	3 mm O.D.	Wilmad Part No. 335-PP-9	160 µL
DLM-7061C	15 mM Benzoic acid in DMSO-d_6	5 mm O.D.	Wilmad Part No. 528-PP-8	750 µL

Other NMR fill volumes and tubes are available. Please inquire.

* The benzoic acid concentration and associated uncertainty are reported.

** All tubes are flame-sealed to ensure longevity.

Internal Calibration Standards

The internal reference method commonly gives errors of <1% and is considered to be the most accurate and reproducible method available to obtain quantitative ^{1}H-NMR spectra. Unfortunately, the reference standard is typically weighed into each sample solution, an action that requires time and effort, and has been reported to the largest source of error with this method.

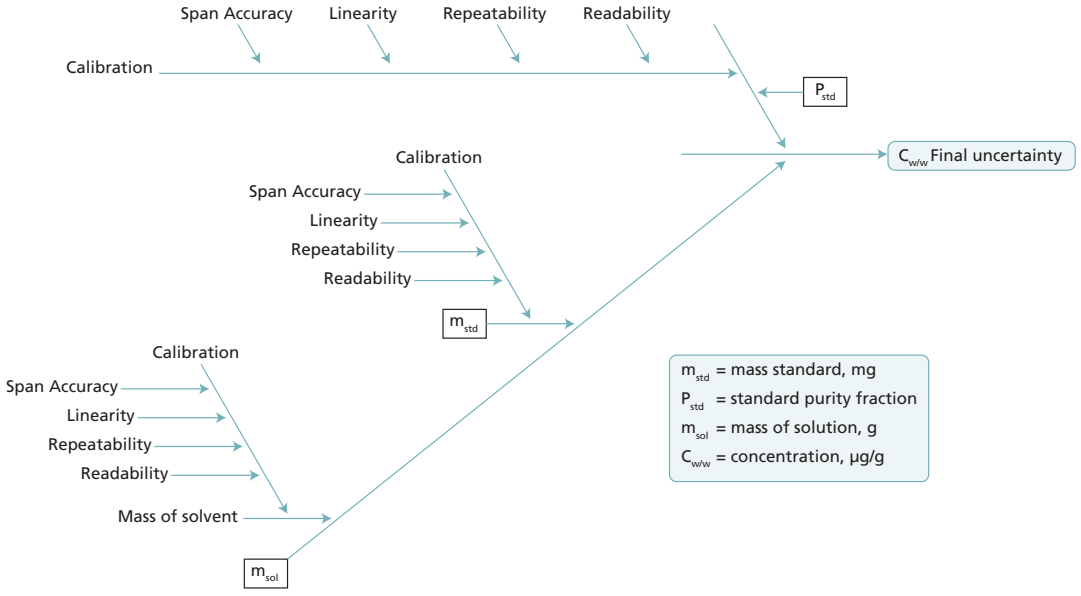
CIL is pleased to offer a ready-to-use DMSO-d_6 solution containing a known amount of benzoic acid for internal referencing. Because this solution is preformulated, the user does not need to weigh a standard material. The elimination of this step will reduce effort and time in sample preparation and also may bring about more accurate results than if the user performs this formulation. To use this product, the sample must be soluble in DMSO-d_6, physically and chemically inert toward benzoic acid and stable in acidic pH. Ideally, there will be no resonances from the sample in the region of benzoic acid aromatic protons (7.4-8.1 ppm), HOD (~3 ppm but is variable) and DMSO-d_5 (2.5 ppm). The benzoic acid concentration with associated uncertainty is presented on the certificate of analysis.

qNMR Standard for Internal Referencing

Catalog No.	Description	Ampoule	Comments
DLM-9491D	5 mM Benzoic acid in DMSO-d_6	1 g	The benzoic acid concentration and associated uncertainty is reported.
DLM-7061D	15 mM Benzoic acid in DMSO-d_6	1 g	The benzoic acid concentration and associated uncertainty is reported.

CIL Formulation Procedure

The procedure that CIL uses to formulate qNMR external calibration reference standard bulk solutions allows for the expanded uncertainty of the concentration of the calibration standard (e.g., benzoic acid) to be determined. Traceability to SI is maintained through the use of weight sets with calibration traceable to NIST and laboratory balances with NIST-traceable calibration certificates, maintaining an unbroken chain of calibration to the kilogram. The factors contributing to the uncertainty of the benzoic acid concentration[1] is shown in below.



Cause-and-effect diagram of factors contributing to the uncertainty of the benzoic acid concentration in the qNMR standard formulation.

Reference

1. EURACHEM CITAC Guide CG 4, "Quantifying Uncertainty in Analytical Measurement," Third Edition, QUAM:2012