Cerilliant Analytical Reference Standards
a SIGMA-ALDRICH company

Certificate of Analysis

Certified Reference Standard - NIST Traceable

Ethanol-400

Ethyl Alcohol

Cerilliant Quality
ISO GUIDE 34
ISO/IEC 17025
ISO 13485
ISO 15194
ISO 9001
GMP/GLP

Catalog Number: E-036
Solution Lot: FN11191402
Expiration Date: February 2020
Diluent: Water
Volume per Ampoule: 1.2 mL
Storage: Refrigerate. Do not freeze.
Intended Use: For R&D/ analytical purposes only. Not suitable for human or animal consumption.

- Expiration Date has been established through real time stability studies and applies to the ampoule stored unopened at the recommended storage condition.
- Ampoules are overfilled to ensure a minimum 1.2 mL volume fill. We advise laboratories to use measured volumes of this standard solution before diluting to the desired concentration. The standard should be used immediately after opening to avoid concentration changes due to evaporation.
- For quantitative applications, the minimum sample size for intended use is 100 µL.

Component	Solution Chromatographic Purity	Certified Concentration
Ethanol	> 99.9%	400.0 ± 1.4 mg/dL

- Uncertainty of the concentration, expressed in terms of volume, is an expanded uncertainty in accordance with ISO 17025 and ISO Guide 34 at the 95% confidence interval using a coverage factor of k=2 and has been calculated by statistical analysis of our production methods applicable to ethanol reference standards and incorporates uncertainty of the purity factor, material density and mass measurement. The dispensing process is sufficiently controlled as to not be a significant contributor to uncertainty calculations and is, therefore, excluded. Solution stability is established through real time stability studies and is, therefore, excluded.
- When expressed in percentage terms, the relative standard uncertainty of the concentration is 0.175% and the relative expanded uncertainty is 0.35% at the 95% confidence interval (k=2).
- The purity factor (PF) mass balance measurement equation is used to calculate the amount of ethanol required to achieve an accurate concentration of the solution standard, accounting for both purity and residual water content.
- Purity factor has been established through independent certification of the neat analyte to ISO 17025 standards – See page 2.
- Solution purity is verified post ampouling and demonstrates no contamination or degradation has occurred.

Traceability to SI through NIST:

- This standard has been prepared and certified under the ISO Guide 34 and ISO/IEC 17025 standards and meets the requirements of a Certified Reference Material as defined by ISO.
- Gravimetrically prepared using qualified balances calibrated semi-annually by Mettler Toledo to ISO 17025 requirements and using NIST traceable weights. Qualification of each balance includes the assignment of a minimum weighing by Mettler Toledo taking into consideration the balance and installed environmental conditions to ensure each weighing complies with USP tolerances of NMT 0.1% relative uncertainty.
- Balance calibration adjustments are performed weekly utilizing the balance's internal adjustment mechanism and with NIST traceable weights.
- Balance calibration is verified prior to each use and is performed utilizing NIST traceable weights. Weigh tapes from the balance calibration are included in the production batch record for this standard. Production data package available upon request.
- Fill volume is gravimetrically verified throughout the dispensing process using qualified balances calibrated with NIST traceable weights.
- Weight sets used for all balance calibrations are calibrated externally by an ISO 17025 accredited calibration laboratory to NIST standards.
- Concentration of this standard has been analytically verified against a NIST SRM and a Control using a validated method. See page 2.

Cerilliant certifies that this standard meets the specifications stated in this certificate and warrants this product to meet the stated acceptance criteria through the expiration date. Warranty applies to ampoules stored unopened and stored under the recommended storage conditions. Warranty and expiry do not extend to solutions into which this product has been incorporated. Establishment of shelf life of all such products is the responsibility of the user.

ACLASS ACCREDITED REFERENCE MATERIAL PRODUCER

Darron Ellsworth, Quality Assurance Manager

June 25, 2015
Date

811 Paloma Drive. Suite A. Round Rock. TX 78665 800-848-7837 / 512-238-9974

Analytical Verification of Solution Standard Concentration and Batch Homogeneity:

Solution Standard	Lot Number	Results compared to NIST SRM Lot 2896 (mg/dL)	Homogeneity (ampoule to ampoule consistency) %RSD
New Lot	FN11191402	399.2	0.54%
Prior Lot	FN012712-01	400.4	1.56%
Acceptance Criteria		**± 2%**	**≤ 2%**

- Concentration is calculated as the average of multiple analyses conducted using a validated Headspace GC/FID method. The validated GC/HS method has been demonstrated to adequately detect and quantitate ethanol concentrations ranging from 5 to 600 mg/dL. Relative standard uncertainty of the analysis is 1.675% and includes both uncertainty of the analytical method and uncertainty of the NIST SRM concentration.
- The Control is independently prepared from a different lot of neat ethanol to ensure no bias in the analysis and independently qualified against a NIST SRM.
- Homogeneity is ensured through rigorous production process controls statistically analyzed to evaluate risk and verified by analysis. The %RSD of samples pulled from across the lot using a stratified random sampling plan demonstrates ampoule to ampoule consistency or homogeneity of the New Lot.
- The %RSD of the Prior Lot represents system suitability on the date of analysis. Triplicate injections of the Prior Lot are bracketed at the beginning and end of the sequence. %RSD criteria ensures proper system performance throughout the sequence.
- All instruments used for certification of the neat materials and verification of the solution concentration and homogeneity are fully qualified through an Installation Qualification and an Operational Qualification which is repeated annually. System suitability is performed daily with rigorous acceptance criteria to ensure the system continues to perform within the validated parameters.

Solution Standard Assay Parameters

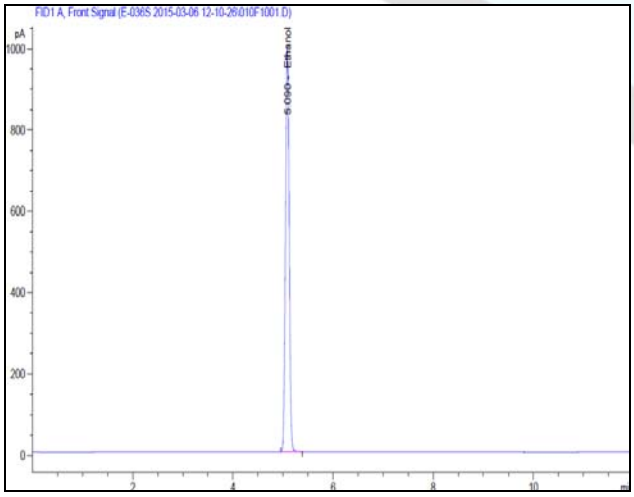
Analysis Method: GC/FID Headspace

Column: DB-ALC1 30 m x 0.53 mm ID, 3.0 µm film thickness

Temp Program: 40°C hold for 12 min

Injector Temp: 200°C

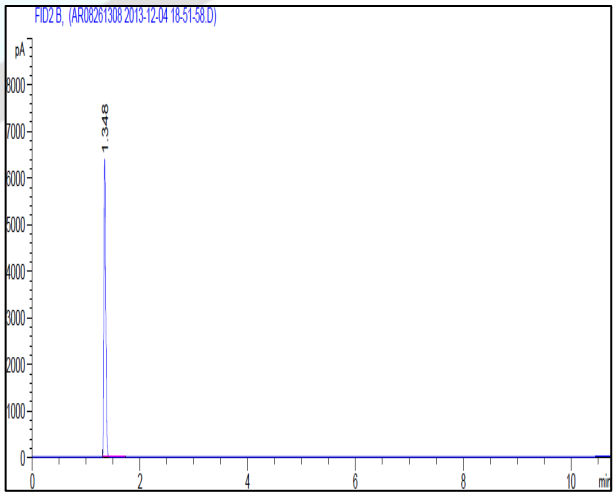
Detector Temp: 250°C



Neat Material Analysis

Purity by GC/FID Analysis: 100.0%

Water Content by Karl Fischer: 0.0%

Purity Factor: 100.0%

The purity factor (PF) mass balance measurement equation is used to calculate the amount of ethanol required to achieve an accurate concentration of the solution standard, accounting for both purity and residual water content.