

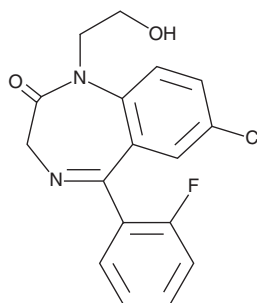
CERTIFICATE of ANALYSIS



ACCREDITED
ISO 17034 #AR-1774

2-Hydroxyethylflurazepam (CRM)

Certified Reference Material



Item No.:	40620
Batch No.:	0706249
CAS Registry No.:	20971-53-3
Molecular Formula:	C ₁₇ H ₁₄ ClFN ₂ O ₂
Formula Weight:	332.80 amu
Expiry Date:	02APR2027 (valid from date of certification)
Supplied as:	A 1 mg/ml (nominal) solution in methanol
Volume per Ampule:	Not less than 1 ml. Ampules are overfilled.
Storage:	Unopened at -20°C ± 10°C
Safety:	Refer to Safety Data Sheet
Intended Use:	For analytical testing purposes only, not intended for human or animal use.
Instructions for Use:	This product is designated for one-time use and should be used immediately after opening. It is advised that laboratories warm the vial to room temperature prior to opening and use measured volumes.

Certified Concentration · 1.000 mg/ml ± 0.005 mg/ml

Concentration is calculated based on product mass, solution mass, corrected purity, and density at 20°C. It is traceable to SI units through an unbroken chain of measurements. Uncertainty of concentration is expressed as an expanded uncertainty in accordance with ISO standards for Testing Laboratories and Reference Material Producers at the approximate 95% confidence interval using a coverage factor of k=2 and incorporates uncertainties from the corrected purity, solution preparation, homogeneity, and long- and short-term stability. Concentration was verified by comparison to an independently prepared calibration standard.

Corrected Purity · 98.86%

Corrected purity is determined as follows: Corrected Purity = [(100 - % LOD - % ROI)*Chromatographic Purity/100] or [(100 - % KF - % RS - % ROI)*Chromatographic Purity/100]. Where applicable, optical rotation, chiral purity, and/or isotopic purity testing are performed to support the identification of the reference material, therefore the uncertainty is considered null.

Approval:

Roxanne Franckowski

Title: Senior Manager of ISO Quality

Certification Date: 02APR2024

Cayman Chemical 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 when stored unopened as recommended.



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CRM Assay

Method Parameters									
Cayman Method	TST F0544								
Column	4.6 x 150 mm, 5 µm Gemini C18								
Mobile Phase	A: 90:10:0.1% Water:Acetonitrile:Ammonium Formate B: 10:90:0.1% Water:Acetonitrile:Ammonium Formate								
Gradient	<table><tr><th>Time (min)</th><th>%B</th></tr><tr><td>0-10</td><td>31.2-100%</td></tr><tr><td>10-13</td><td>100%</td></tr><tr><td>13.1-20</td><td>31.2%</td></tr></table>	Time (min)	%B	0-10	31.2-100%	10-13	100%	13.1-20	31.2%
Time (min)	%B								
0-10	31.2-100%								
10-13	100%								
13.1-20	31.2%								
Flow Rate	1 ml/min								
Column Temp	30°C								
Wavelength	UV monitored at 250 nm								

Homogeneity

A minimum sample size of 1.0 µg was used to determine homogeneity. Homogeneity was determined by HPLC using ampules selected from a random sampling plan from early, middle, and late fill positions.

%RSD	Acceptance Criteria
0.39%	≤3%

The recommended minimum quantity for use is 1.0 µg. Quantities below this have not been evaluated.

Neat Material Quality Information (Item No.: 40185, Batch No.: 0694750)

Qualifier	Method	Result
Chromatographic Purity, HPLC	Cayman Method F0544	>99.90%
Identity, LC-MS	Cayman Method TST SD13, +ESI	333.0 amu
Identity, GC-MS	Cayman Method TST SD12	Conforms
Identity, FTIR	Cayman Method TST SD03	Conforms
% LOD	Cayman Method TST SD24	0.94%
% ROI	Cayman Method TST SD06	<0.10%
Identity, NMR	¹ H NMR	Conforms

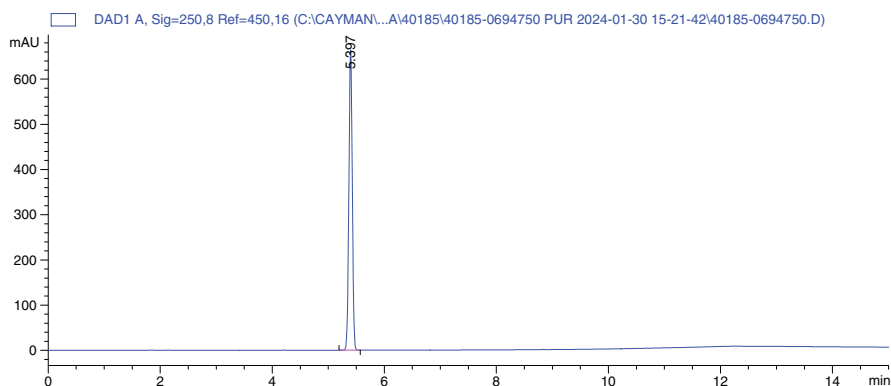
NMR and optical rotation (if applicable) are provided as supplemental information but are not within scope of ISO accreditation. Property values are traceable to SI units through an unbroken chain of measurements.

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Supplemental Data (Neat Material)

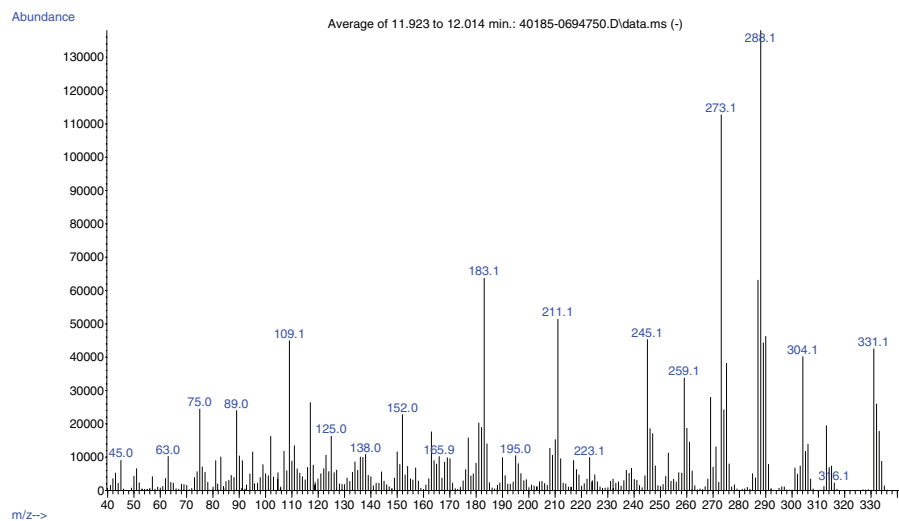
HPLC-UV



Conditions

Instrument	Agilent 1100/1200 Series								
Column	4.6 x 150 mm, 5 µm Gemini C18								
Mobile Phase	A: 90:10:0.1% Water:Acetonitrile:Ammonium Formate B: 10:90:0.1% Water:Acetonitrile:Ammonium Formate								
Gradient	<table border="1"> <thead> <tr> <th>Time (min)</th> <th>%B</th> </tr> </thead> <tbody> <tr> <td>0-10</td> <td>31.2-100%</td> </tr> <tr> <td>10-13</td> <td>100%</td> </tr> <tr> <td>13.1-20</td> <td>31.2%</td> </tr> </tbody> </table>	Time (min)	%B	0-10	31.2-100%	10-13	100%	13.1-20	31.2%
Time (min)	%B								
0-10	31.2-100%								
10-13	100%								
13.1-20	31.2%								
Flow Rate	1 ml/min								
Column Temp	30°C								
Wavelength	UV monitored at 250 nm								

GC-MS



Conditions

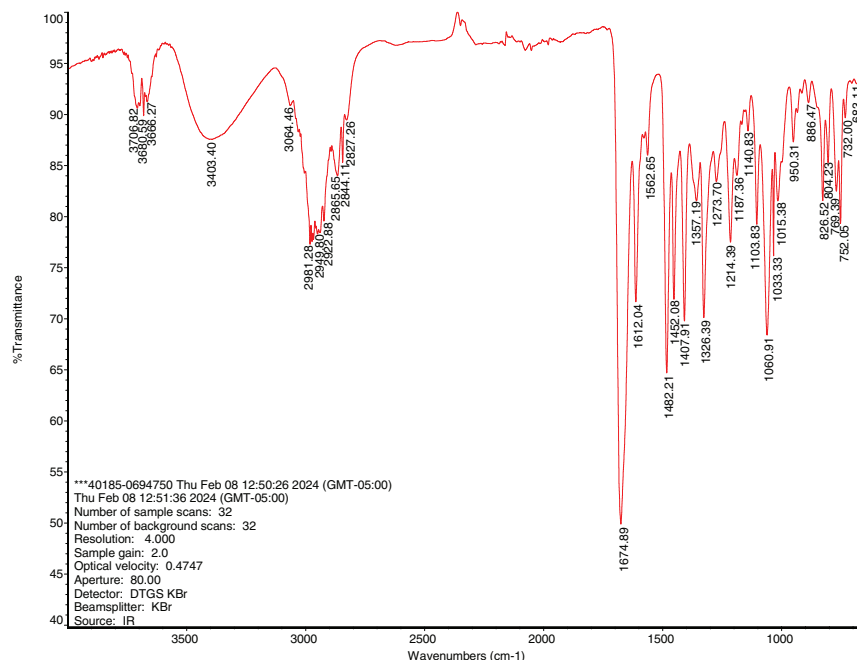
Instrument	Agilent GC MSD
Column	30 m x 0.32 mm, 0.5 µm Rtx-5MS
Carrier Gas	He
Flow Rate	2 ml/min
Inlet Temp	300°C
Split Ratio	15:1
Oven Program	50°C hold for 1 min, ramp to 300°C at 30°C per min, hold at 300°C to 25 min
Transfer Line Temp	300°C
Voltage	70eV EI MS
Scan Range	35-500 m/z
Tune File	stune

Apex spectrum – background (1 min window in front of peak)

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FTIR

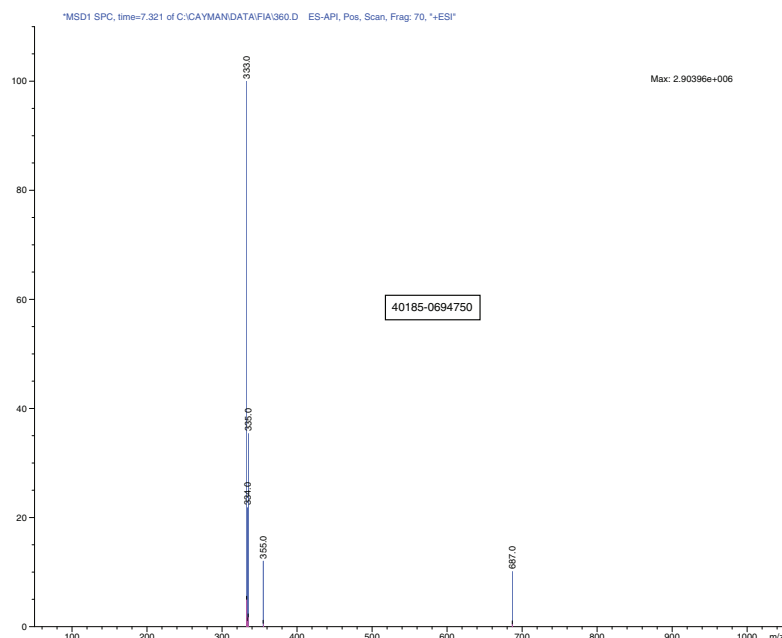


Conditions

Instrument	Thermo Nicolet iS10 FTIR / Diamond SmartATR (single bounce)
Scans	32 scans / 32 background scans
Range	650-4,000 cm-1
Resolution	4.000

ATR and background corrected

ESI-MS



Conditions

Instrument	Agilent HPLC MSD
Mobile Phase	50:50:0.1 Methanol/Water/Acetic Acid
Flow Rate	0.5 ml/min
Ionization Mode	+ESI
Mass Range	100-1,000 m/z
Nebulizer	60 psi
Desolvation Gas	13 L/min
Desolvation Temp	350°C
Electrospray Voltage	4kV

MS collected across peak width at half height

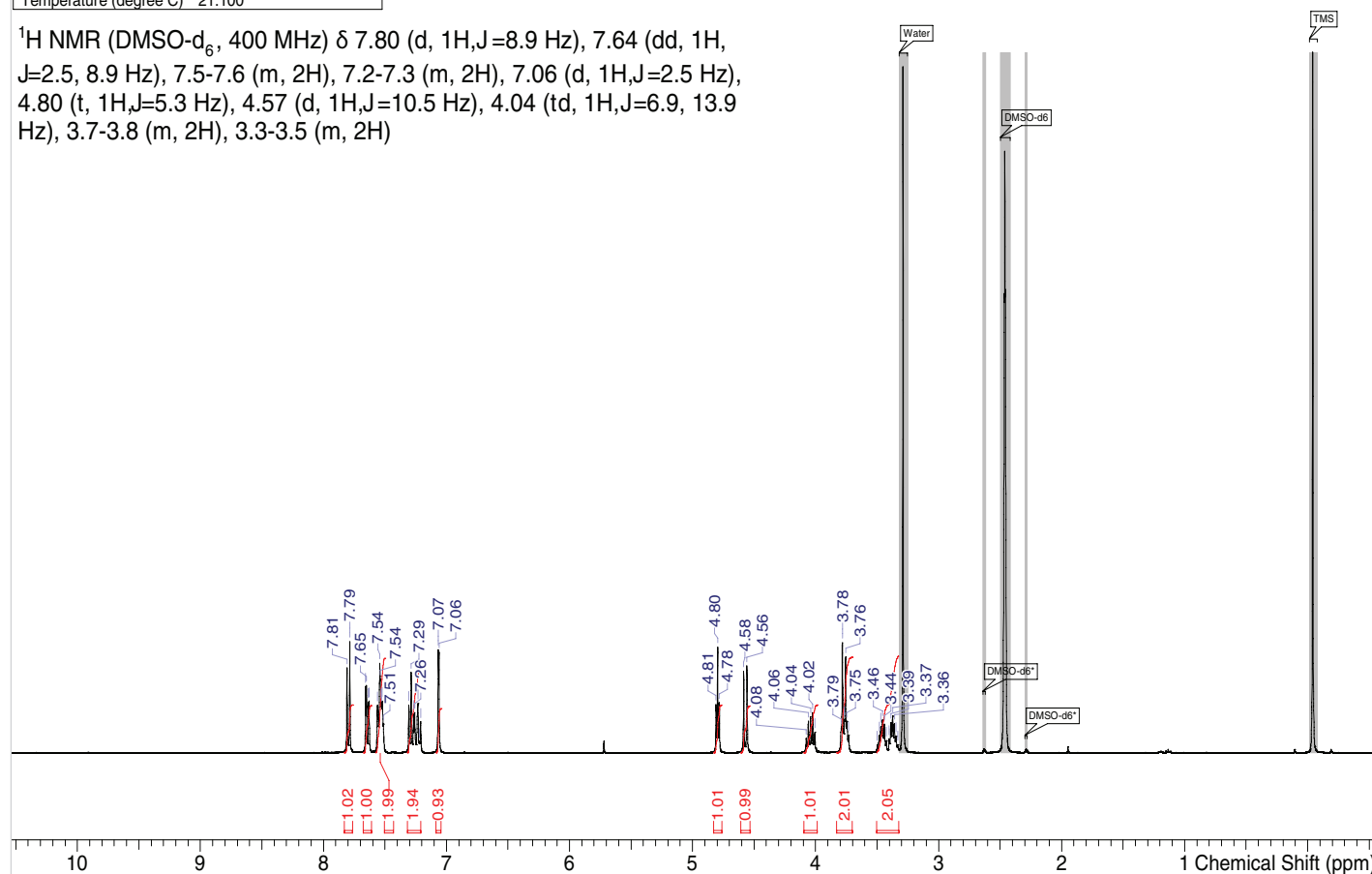
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NMR (not within scope of ISO accreditation)

File Name	\\sulfur\private\nmrdata\JEOL_2023\YL2340-112-1\YL2340-112-1_PROTON_14-Dec-2023-1-1.jdf		
Date	14 Dec 2023 15:54:33	Nucleus	1H
Solvent	DMSO-d6	Frequency (MHz)	399.7822
Temperature (degree C)	21.100	Number of Transients	32
		Origin	JEOL ECZ400S Ca v601

¹H NMR (DMSO-d₆, 400 MHz) δ 7.80 (d, 1H, J=8.9 Hz), 7.64 (dd, 1H, J=2.5, 8.9 Hz), 7.5-7.6 (m, 2H), 7.2-7.3 (m, 2H), 7.06 (d, 1H, J=2.5 Hz), 4.80 (t, 1H, J=5.3 Hz), 4.57 (d, 1H, J=10.5 Hz), 4.04 (td, 1H, J=6.9, 13.9 Hz), 3.7-3.8 (m, 2H), 3.3-3.5 (m, 2H)



Conditions

Instrument	JEOL ECZ 400S
Scans	32 scans

Stability

The effect of the components of stability on the combined standard uncertainty of the CRM property value are considered negligible unless indicated in stability studies.

Short-Term Stability

A decrease in property value was observed at 60°C during the two-week stability study. No decrease was observed at ambient temperature during the study. This data supports cold shipment of this product.

Long-Term Stability

Long-term stability data confirmed three years stability at the -20°C storage temperature.

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Quality Standard Documentation

The manufacturer of this Certified Reference Material is accredited under ISO 17034:2016 accreditation issued by ANAB. Refer to ANAB certificate and scope of accreditation AR-1774.

The manufacturer of this Certified Reference Material is accredited under ISO/IEC 17025:2017 accreditation issued by ANAB. Refer to the ANAB certificate and scope of accreditation AT-1773.

Revision History

Revision No.	Date	Reason for Revision
01	02APR2024	Initial version

Disclaimers

Material Safety Data

This material should be considered hazardous until information to the contrary becomes available. Do not ingest, swallow, or inhale. Do not get in eyes, on skin, or on clothing. Wash thoroughly after handling. This information contains some but not all of the information required for the safe and proper use of this material. Before use, review the complete Safety Data Sheet, which has been sent *via* email to your institution.

Warranty and Limitation of Remedy

Cayman Chemical Company makes no warranty or guarantee of any kind, whether written or oral, expressed or implied, including without limitation, any warranty of fitness for a particular purpose, suitability and merchantability, which extends beyond the description of the chemicals hereof. Cayman warrants only to the original customer that the material will meet our specifications at the time of delivery.

Cayman will carry out its delivery obligations with due care and skill. Thus, in no event will Cayman have any obligation or liability, whether in tort (including negligence) or in contract, for any direct, indirect, incidental or consequential damages, even if Cayman is informed about their possible existence.

This limitation of liability does not apply in the case of intentional acts or negligence of Cayman, its directors or its employees.

Buyer's exclusive remedy and Cayman's sole liability hereunder shall be limited to a refund of the purchase price, or at Cayman's option, the replacement, at no cost to Buyer, of all material that does not meet our specification.

Said refund or replacement is conditioned on Buyer giving written notice to Cayman within thirty (30) days after arrival of the material at its destination. Failure of Buyer to give said notice within thirty (30) days shall constitute a waiver of Buyer of all claims hereunder with respect to said material.

For further details, please refer to our Warranty and Limitations of Remedy located on our website and in our catalog.

This Certificate shall not be reproduced except in full, without written approval from the Cayman Chemical ISO Quality Manager.

ISO CRT SD02 v 5.1

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