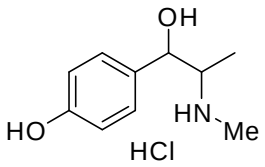
Australian Government
Department of Industry, Science, Energy and Resources

National Measurement Institute

NATA ACCREDITED FOR TECHNICAL COMPETENCE

CERTIFIED REFERENCE MATERIAL CERTIFICATE OF ANALYSIS

NMIA D892b: Oxilofrine hydrochloride



Report ID: D892b.2022.01 (Bottled 220301)

Chemical Formula: $C_{10}H_{15}NO_2$.HCl

Molecular Weight: 217.7 g/mol (HCl), 181.3 g/mol (base)

Certified value

Batch No.	CAS No.	Purity (mass fraction)
21-D-10	942-51-8 (HCl) 365-26-4 (base)	98.7 ± 1.6%

The uncertainty has been calculated according to ISO Guide 35 and is stated at the 95% confidence limit (k = 2).

IUPAC name: 4-[1-Hydroxy-2-(methylamino)propyl]phenol hydrochloride.

Expiration of certification: The property values are valid till 12 January 2025, i.e. three years from the date of certification provided the **unopened** material is handled and stored in accordance with the recommendations below. The material as issued in the unopened container and stored as recommended below should be suitable for use beyond this date, subject to confirmation of batch stability from the issuing body. The expiry date/shelf life does not apply to sample bottles that have been opened. In such cases it is recommended that the end-user conduct their own in-house stability trials. The material will be re-tested on an annual basis to ensure that the property values are still valid. In the event a product fails the stability trial, notification will be sent to all impacted customers.

Description: Off-white powder sourced from an external supplier, and certified for identity and purity by NMIA. Packaged in amber glass bottles with a septum and crimped aluminium cap.

Intended use: This certified reference material is suitable for use as a primary calibrator.

Instructions for use: Equilibrate the bottled material to room temperature before opening.

Recommended storage: When not in use this material should be stored at or below 25 ºC in a closed container in a dry, dark area.

Metrological traceability: The certified purity value is traceable to the SI unit for mass (kg) through Australian national standards via balance calibration. In the mass balance approach all impurities are quantified as a mass fraction and subtracted from 100%.

Stability: At the recommended storage conditions this material has demonstrated stability for a period of at least three years. The measurement uncertainty includes components for long term stability at the recommended storage conditions, and accelerated stability trials conducted at 40 ºC and 75% humidity for a 14 day period.

The long-term stability of the compound in solution has not been examined.

Homogeneity assessment: The homogeneity of the material was assessed using purity assay by GC-FID on ten randomly selected 1-2 mg sub samples of the material. The material was judged to be sufficiently homogeneous at this level of sampling as the variation in analysis results between samples was not significantly different at a 95% confidence level from that observed on repeat analysis of the same sample.

Safety: Treat as a hazardous substance. Use appropriate work practices when handling to avoid skin or eye contact, ingestion or inhalation of dust. Refer to the provided safety data sheet.

S. R. Davies

Dr Stephen R. Davies,
Team Leader,
Chemical Reference Materials, NMI.
16 March 2022

NATA logo notice: Accredited for compliance with ISO 17034. Accreditation No. 198 / Corporate Site No. 14214. The results of the tests, calibrations and/or measurements included in this document are traceable to Australian/national standards.

Legal notice: Terms and Conditions associated with the provision of this reference material can be found on the NMIA website.

Characterisation Report:

The identity was confirmed by a range of spectroscopic techniques, NMR, IR and MS. The certified purity value was obtained by mass balance from a combination of traditional analytical techniques, including GC-FID, thermogravimetric analysis, Karl Fischer analysis, and ^{1}H NMR spectroscopy. The purity value is calculated as per Equation 1.

Purity = (100 % - I_{ORG}) x (100 % - I_{VOL} – I_{NVR}) Equation 1

I_{ORG} = Organic impurities of related structure, I_{VOL} = volatile impurities, I_{NVR} = non-volatile residue.

Supporting evidence is provided by elemental microanalysis.

GC-FID:	Instrument:	Varian CP-3800
	Column:	DB-17, 30 m × 0.32 mm I.D. × 0.25 μm
	Program:	180 °C (10 min), 20 °C/min to 280 °C (10 min)
	Injector:	180 °C
	Detector Temp:	320 °C
	Carrier:	Helium
	Split ratio:	20/1
	Relative mass fraction of the main component as the free base:	
	Initial analysis:	Mean = 99.7%, s = 0.06% (10 sub samples in duplicate, January 2022)
Karl Fischer analysis:		Moisture content 0.5 % mass fraction (January 2022)
Thermogravimetric analysis:		Non-volatile residue 0.3% mass fraction (October 2021)

Spectroscopic and other characterisation data

GC-MS:	Instrument:	Agilent 6890/5973
	Column:	DB-5MS, 30 m x 0.25 mm I.D. x 0.25 μm
	Program:	180 °C (10 min), 20 °C/min to 300 °C (5 min)
	Injector:	250 °C
	Split ratio:	20/1
	Transfer line temp:	280 °C
	Carrier:	Helium, 1.0 mL/min
	Scan range:	50-550 *m/z*
	The retention time of oxilofrine free base is reported with the major peaks in the mass spectra. The latter are reported as mass/charge ratios and (in brackets) as a percentage relative to the base peak.	
	Free base (5.0 min):	134 (11), 133 (9), 121 (12), 107 (6), 77 (9), 65 (7), 58 (100), 56 (18), 42 (17) *m/z*
ESI-MS:	Instrument	Micromass Quatro Micro
	Operation:	Positive ion mode, direct infusion at 5 μL/min
	Ionisation:	ESI spray voltage at 3.2 kV negative ion
	EM voltage:	600 V
	Cone voltage:	250 V
	The ions observed are reported in mass/charge ratios and (in brackets) as a percentage relative to the base peak.	
	Peak:	204 ($M+Na^+$, 4), 182 ($M+H^+$, 67), 164 ($M+H^+-H_2O$, 100) *m/z*
TLC:	Conditions:	Kieselgel 60F_{254}. Dichloromethane/Methanol (1:1) Single spot observed, R_f = 0.3. Visualisation with UV at 254 nm
IR:	Instrument:	Bruker Alpha Platinum ATR
	Range:	4000-400 cm^{-1}, neat
	Peaks:	3410, 3094, 2971, 2841, 2758, 2469, 1615, 1596, 1511, 1462, 1442, 1213, 988, 851, 837, 670, 531 cm^{-1}
^{1}H NMR:	Instrument:	Bruker Avance III-500
	Field strength:	500 MHz
	Solvent:	MeOH-d_4 (3.31 ppm)
	Spectral data:	δ 1.09 (3H, d, *J* = 6.5 Hz), 2.75 (3H, s), 3.35 (1H, dq, *J* = 3.3, 6.6 Hz), 5.01 (1H, d, *J* = 3.3 Hz), 6.81 (2H, m), 7.23 (2H, m) ppm
		Methanol was quantified at 0.2% mass fraction.
^{13}C NMR:	Instrument:	Bruker Avance III-500
	Field strength:	126 MHz
	Solvent:	MeOH-d_4 (49.0 ppm)
	Spectral data:	δ 10.2, 31.6, 61.7, 71.8, 116.3, 128.3, 131.8, 158.4 ppm
Melting point:		207-209 °C
Microanalysis:	Found:	C = 55.2%; H = 7.5%; N = 6.4% (October, 2021)
	Calculated:	C = 55.2%; H = 7.4%; N = 6.4% (Calculated for $C_{10}H_{15}NO_2$.HCl)