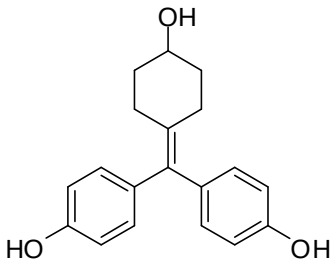
Australian Government
Department of Industry, Science and Resources

National Measurement Institute

NATA
WORLD RECOGNISED ACCREDITATION

REFERENCE MATERIAL PRODUCT INFORMATION SHEET

NMIA D930: 4-Hydroxycyclofenil



Report ID: D930.2024.01 (Ampouled 180712)

Chemical Formula: $C_{19}H_{20}O_3$

Molecular Weight: 296.4 g/mol

Property value

Batch No.	CAS No.	Mass per ampoule
08-S-10	Not available	960 ± 43 μg

SYNONYMS: 4-[Bis(4-hydroxyphenyl)methylene]cyclohexanol
Cyclofenil metabolite M1

Expiration of certification: The property values are valid till 19 February 2029, i.e. five years from the date of re-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 ampoules that have been opened. In such cases it is recommended that the end-user conduct their own in-house stability trials.

Description: The compound is supplied as a dried aliquot in a sealed ampoule and is intended for a single use to prepare a standard solution containing D930. Material was sourced from an external supplier, and certified for identity and purity by NMIA.

Intended use: This reference material is recommended for qualitative analysis only and is not intended for use as a calibrator. The material does not have certified reference material status as metrological traceability of the stated purity value to the SI unit for mass (kg) has not been established.

Instructions for use: Open the ampoule and carefully rinse the interior at least three times with a suitable organic solvent (e.g. acetonitrile). This will transfer 960 μg of anhydrous 4-hydroxycyclofenil.

Recommended storage: When not in use, this material should be stored at or below 4 ºC in a closed container in a dry, dark area.

Stability: The ampouled material has been shown to degrade slowly over time. 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 HPLC with UV detection on seven randomly selected ampoules 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 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.
22 February 2024

This report supersedes any issued prior to 22 February 2024.

NATA Accreditation No. 198 / Corporate Site No. 14214.

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

Characterisation Report:

HPLC:	Instrument:	Waters Model 1525 Binary pump, 717 plus autosampler or Shimadzu Binary pump LC-20AB, SIL-20 A HT auto sampler
	Column:	Alltima C-18, 5 μm (4.6 mm × 150 mm)
	Mobile Phase:	A = Milli Q water, B = Acetonitrile 0-17 min 29% B, 17-19 min 29-80% B, 19-23 min 80% B, 23-25 min 80-29% B, 25-30 min 29% B
	Flow rate:	1.0 mL/min, Gradient
	Detector:	Waters PDA 2998 operating at Max plot or Shimadzu SPD-M20A PDA operating at Max plot
	Relative peak area of the main component:	
	Initial analysis:	Mean = 97.9%, s = 0.04% (7 ampoules in duplicate, July 2018)
	Re-analysis:	Mean = 97.7%, s = 0.10% (5 ampoules in duplicate, July 2019)
	Re-analysis:	Mean = 95.2%, s = 0.43% (5 ampoules in duplicate, February 2024)

The following analytical data was obtained on the bulk material subsequently used in the preparation of the ampoules.

Characterisation Report:

Purity estimate obtained from quantitative nuclear magnetic resonance (QNMR). The purity estimate by QNMR was obtained using a certified internal standard of dimethylsulfone. Supporting evidence is provided by HPLC with UV detection, thermogravimetric analysis, Karl Fischer, ^{1}H NMR and elemental microanalysis.

HPLC:	Instrument:	Waters Model 1525 Binary pump, 717 plus autosampler
	Column:	Alltima C-18, 5 μm (4.6 mm × 150 mm)
	Mobile Phase:	Acetonitrile/MilliQ water (29:71)
	Flow rate:	1.0 mL/min
	Detector:	Waters PDA 996 operating at Max plot
	Relative peak area of the main component:	
	Initial analysis:	Mean = 98.2%, s= 0.08% (10 sub samples in duplicate, September 2008)
	Re-analysis:	Mean = 98.5%, s= 0.00% (1 sub sample in duplicate, December 2009)
HPLC:	Instrument:	Waters Model 1525 Binary pump, 717 plus autosampler
	Column:	Alltima C-18, 5 μm (4.6 mm × 150 mm)
	Mobile Phase:	A = MilliQ water, B = Acetonitrile 0-17 min 29% B, 19-23 min 80% B, 25-30 min 29% B
	Flow rate:	1.0 mL/min, Gradient
	Detector:	Waters PDA 2998 operating at Max plot
	Relative peak area of the main component:	
	Initial analysis:	Mean = 97.9%, s = 0.00% (1 sub sample in duplicate, July 2018)
Thermogravimetric analysis:		Volatile content unable to be determined and non volatile residue 0.5% mass fraction (August 2008)
Karl Fischer analysis:		Moisture content 0.1% mass fraction (August 2008) Moisture content 0.2% mass fraction (June 2018)
QNMR:	Instrument:	Bruker DMX-600
	Field strength:	600 MHz
	Solvent:	MeOH-d_4 (3.31 ppm)
	Internal standard:	Dimethylsulfone (100% m/m)
	Initial analysis:	Mean = 96.8%, s = 1.27% (4 sub samples, August 2008)

Spectroscopic and other characterisation data

GC-MS:	*Tris*-TMS:		
	Instrument:	Agilent 6890/5973	
	Column:	HP Ultra 1, 17 m × 0.22 mm I.D. × 0.11 μm	
	Program:	180 °C (0.2 min), 3 °C /min to 233 °C, 10 °C /min to 265 °C, 30 °C /min to 310 °C (2 min)	
	Injector:	250 °C	Transfer line temp: 300 °C
	Carrier:	Helium, 1.0 mL/min	Split ratio: 14/1
	The retention time of the *tris*-TMS derivative 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.		
	Tris-TMS (16.2 min):	512 (M+, 18), 423 (39), 422 (100), 381 (8), 343 (22), 179 (5), 73 (24) *m/z*	
TLC:	Conditions:	Kieselgel 60F_{254}. Methanol/ethyl acetate (1:4) Single spot observed, R_f = 0.7. Visualisation with UV at 254 nm	
IR:	Instrument:	Biorad FTS3000MX FT-IR	
	Range:	4000-400 cm^{-1}, KBr powder	
	Peaks:	3516, 3464, 3329, 2934, 1608, 1511, 1451, 1264, 1040, 836, 569 cm^{-1}	
^{1}H NMR:	Instrument:	Bruker DMX600	
	Field strength:	600 MHz	
	Solvent:	MeOH-d_4 (3.31 ppm)	
	Spectral data:	δ 1.44 (2H, m), 1.92 (2H, m), 2.05 (2H, m), 2.582 (2H, m), 3.81 (1H, m), 6.71 (4H, d, *J* = 8.5 Hz), 6.91 (4H, d, *J* = 8.5 Hz) ppm	
^{13}C NMR:	Instrument:	Bruker DMX600	
	Field strength:	151 MHz	
	Solvent:	MeOH-d_4 (49.9 ppm)	
	Spectral data:	δ 30.8, 38.1, 71.4, 116.4, 132.6, 136.8, 137.2, 137.6, 157.7 ppm	
Melting point:		203-207 °C	
Microanalysis:	Found:	C = 77.1 %; H = 6.8 % (August 2008)	
	Calculated:	C = 77.0 %; H = 6.8 % (Calculated for $C_{19}H_{20}O_3$)	