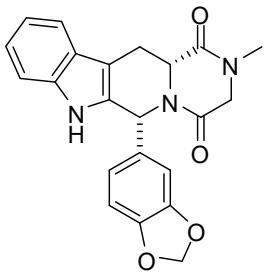
Australian Government
Department of Industry, Science and Resources

National Measurement Institute

NATA
WORLD RECOGNISED ACCREDITATION

CERTIFIED REFERENCE MATERIAL CERTIFICATE OF ANALYSIS

NMIA D941: Tadalafil



Report ID: D941.2025.01 (Bottled 110711)

Chemical Formula: $C_{22}H_{19}N_3O_4$

Molecular Weight: 389.4 g/mol

Certified value

Batch No.	CAS No.	Purity (mass fraction)
09-D-08	171596-29-5	99.7 ± 0.4%

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

IUPAC name: (6R, 12aR)-6-(benzodioxol-5-yl)-2,3,6,7,12,12a-hexahydro-2-methylpyrazinol[1',2':1,-6]pyrido[3,4-b]indole-1,4-dione

Expiration of certification: The property values are valid till 20 October 2035, ten 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 sample bottles that have been opened. In such cases it is recommended that the end-user conduct their own in-house stability trials.

Description: Off-white powder sourced from an external supplier, certified for identity and purity by NMI Australia. Packaged in amber glass bottles with a septum and crimped aluminium cap or screw top 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%. Quantitative NMR provides an independent direct measure of the mass fraction of the analyte of interest, calibrated with an internal standard certified for purity (mass fraction).

Stability: This material has demonstrated stability over a minimum period of ten years. The measurement uncertainty at the 95% confidence interval includes a stability component which has been estimated from annual stability trials. 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 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.

Caution: 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.
21 October 2025

This report supersedes any issued prior to 21 October 2025.

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.

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 HPLC with UV detection, 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 quantitative NMR (qNMR) and elemental microanalysis. The purity value obtained by qNMR was determined using a combination of the one-proton doublet at 5.86 ppm and the one-proton singlet at 6.14 ppm measured against a certified internal standard of dimethyl terephthalate.

HPLC:	Column:	X-Terra C-18, 5 μm (3.9 mm × 150 mm), Alltima C-18, 5 μm (4.6 mm × 150 mm), or ACE Excel 5 super C18 (4.6 mm x 250 mm)
	Mobile Phase:	Solvent A: Milli Q water Solvent B: acetonitrile Gradient 0-5 min 20% B, 5-28 min 20-80% B, 28-29 min 80% B, 29-30 min 80-20% B, 30-40 min 20% B
	Flow Rate:	1.2 mL/min
	Detector:	UV at 283 nm
	Relative mass fraction of the main component:	
	Initial analysis:	Mean = 99.7%, s = 0.02% (10 sub samples in duplicate, June 2009)
	Re-analysis:	Mean = 99.3%, s = 0.02% (5 sub samples in duplicate, June 2011)
	Re-analysis:	Mean = 99.7%, s = 0.02% (5 sub samples in duplicate, May 2012)
	Re-analysis:	Mean = 99.1%, s = 0.05% (5 sub samples in duplicate, June 2015)
	Initial analysis:	Mean = 99.3%, s = 0.03% (5 sub samples in duplicate, June 2018)
	Re-analysis:	Mean = 99.5%, s = 0.04% (5 sub samples in duplicate, June 2021)
	Re-analysis:	Mean = 99.7%, s = 0.01% (5 sub samples in duplicate, October 2025)
Karl Fischer analysis:		Moisture content 0.3% mass fraction (February 2009), Moisture content < 0.1% mass fraction (March 2010, July 2011, May 2012, June 2015 and October 2025), Moisture content 0.14% mass fraction (June 2018) Moisture content 0.18% mass fraction (June 2021)
Thermogravimetric analysis:		Initial volatile content < 0.1% and non volatile residue < 0.2 % mass fraction (April 2009)
QNMR:	Instrument:	Bruker Avance 400
	Field strength:	400 MHz
	Solvent:	d_6-DMSO (2.50 ppm)
	Internal standard:	Dimethyl terephthalate (100% mass fraction)
	Purity estimate:	Mean = 99.7%, s = 0.9 (5 sub samples, May 2009)

Spectroscopic and other characterisation data

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:	650 V
	Cone voltage:	15 V
	Peak:	390 ($M + H^+$) *m/z*
TLC:	Conditions:	Kieselgel 60F_{254}. Hexane/ethyl acetate/methanol (4/3/1) Single spot observed, R_f = 0.5. Visualisation with UV at 254 nm
IR:	Instrument:	Biorad FTS300MX FT-IR
	Range:	4000-400cm^{-1}, KBr powder
	Peaks:	3328, 2904, 1677, 1650, 1490, 1437, 1324, 1269, 1242, 1152, 1042, 940, 746 cm^{-1}
^{1}H NMR:	Instrument:	Avance 400
	Field strength:	400 MHz
	Solvent:	$CDCl_3$ (7.26 ppm)
	Spectral data:	δ 3.04 (3H, s), 3.21 (1H, ddd, *J* = 1.4, 11.6, 16.0 Hz), 3.78 (1H, dd, *J* = 4.4, 16 Hz), 3.93 (1H, d, *J* = 17.4 Hz), 4.11 (1H, dd, *J* = 1.5, 17.5 Hz), 4.30 (1H, dd, *J* = 4.2, 11.4 Hz), 5.85 (1H, d, *J* = 8.9 Hz), 5.86 (1H, d, *J* = 8.9 Hz), 6.14 (1H, s), 6.68 (1H, d, *J* = 8.1 Hz), 6.73 (1H, d, *J* = 1.7 Hz), 6.84 (1H, dd, *J* = 1.7, 8.1 Hz), 7.17 (2H, m), 7.27 (1H, m), 7.61 (1H, m), 7.95 (1H, s) ppm
^{13}C NMR:	Instrument:	Bruker Gyro-300
	Field strength:	75 MHz
	Solvent:	$CDCl_3$ (77.0 ppm)
	Spectral data:	δ 23.8, 33.6, 52.1, 56.2, 56.7, 101.1, 106.5, 107.4, 108.2, 111.2, 118.6, 120.1, 120.7, 122.5, 126.2, 132.7, 135.3, 136.5, 147.1, 147.8, 166.4, 166.8 ppm
Microanalysis:	Found:	C = 67.7%; H = 4.7%; N = 10.8% (February 2009)
	Calculated:	C = 67.9%; H = 4.9%; N = 10.8% (Calculated for $C_{22}H_{19}N_3O_4$)
Specific rotation:		$[\alpha]_D$ = + 69.4° (July 2009)