Australian Government
Department of Industry, Innovation and Science

National Measurement Institute

NATA
ACCREDITED FOR TECHNICAL COMPETENCE

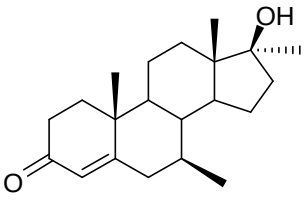
REFERENCE MATERIAL PRODUCT INFORMATION SHEET

NMIA D618: Calusterone

Report ID: D618.2017.02 (Ampouled 080214)

Chemical Formula: $C_{21}H_{32}O_2$

Molecular Weight: 316.2 g/mol



Property value

Batch No.	CAS No.	Mass per ampoule
99-S-26	17021-26-0	974 μg

IUPAC name: (7β,17β)-17-Hydroxy-7,17-dimethylandrost-4-en-3-one

Expiration of certification: The property values are valid till 3 March 2022, 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 under an atmosphere of argon. The reference material is intended for a single use to prepare a standard solution containing D618. 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.

Instructions for use: Open the ampoule and carefully rinse the interior at least three times with a suitable organic solvent (e.g. chloroform). This will transfer approximately 974 μg of anhydrous calusterone. The mass of analyte in each ampoule is calculated from the assigned purity of the bulk and the concentration of bulk material in a stock solution used to prepare the ampoules.

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: This material has demonstrated stability over a minimum period of three years. 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 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.
21 February 2020

This report supersedes any issued prior to 21 February 2020

NATA logo notice: Accredited for compliance with ISO 17034. Accreditation No. 198 / Corporate Site No. 20844. 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:

Warning: **This material is sensitive to the quality of the silanised glass liner when injected at elevated temperature (~ 250 ºC) into a GC instrument.**

GC-FID:	Instrument:	Agilent 6890
	Column:	HP-1 Capillary, 30 m × 0.32 mm I.D. × 0.25 μm
	Program:	180 °C (1 min), 10 °C/min to 220 °C (5 min), 20 °C/min to 300 °C (10 min)
	Injector:	250 °C
	Detector Temp:	320 °C
	Carrier:	Helium
	Split ratio:	20/1

Relative peak area of the main component as the free base:

Initial analysis:	Mean = 98.4%, s = 0.07% (5 ampoules in duplicate, February 2008)
Re-analysis:	Mean = 98.4%, s = 0.02% (5 ampoules in duplicate, April 2009)
Re-analysis:	Mean = 98.4%, s = 0.07% (5 ampoules in duplicate, March 2012)
Re-analysis:	Mean = 98.4%, s = 0.03% (5 ampoules in duplicate, March 2017)

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

Characterisation Report:

The identity was confirmed by a range of spectroscopic techniques, NMR, IR and MS. The 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.

Karl Fischer analysis:	Moisture content 0.15% mass fraction (March 2008)
Thermogravimetric analysis:	Volatiles content 0.18% and non-volatile residue < 0.2% mass fraction (March 2008)

Spectroscopic and other characterisation data

GC-MS:	Instrument:	HP6890/5973
	Column:	HP Ultra 2, 17 m x 0.22 mm I.D. x 0.11 μm
	Program:	140 °C (1 min), 8 °C/min to 250 °C, 30 °C/min to 300 °C (3 min)
	Injector:	280 °C
	Split ratio:	Splitless
	Transfer line temp:	300 °C
	Carrier:	Helium
	Scan range:	50-550 *m/z*
	The retention time of the parent compound 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.	
	Parent (13.8 min):	316 (M^+, 100), 301 (48), 298 (16), 283 (14), 259 (64), 243 (43), 91 (49) *m/z*
TLC:	Conditions:	Kieselgel 60F_{254}. Hexane/ethyl acetate (1:1) Single spot observed, R_f = 0.3
IR:	Instrument:	FT-IR, Biorad WIN FTS40
	Range:	4000-400 cm^{-1}, KBr pellet
	Peaks:	3426, 1655, 1613, 1375, 1230, 1159, 1128, 941, 860 cm^{-1}
^{1}H NMR:	Instrument:	Bruker Advance-300
	Field strength:	300 MHz
	Solvent:	$CDCl_3$ (7.26 ppm)
	Spectral data:	δ 0.91 (3H, s), 1.05 (3H, d, *J* = 5.7 Hz), 1.19 (3H, s), 1.20 (3H, s), 5.70 (1H, s) ppm
^{13}C NMR:	Instrument:	Bruker Advance-300
	Field strength:	75 MHz
	Solvent:	$CDCl_3$ (77.2 ppm)
	Spectral data:	δ 14.1, 17.4, 20.7, 22.9, 25.6, 26.9, 31.1, 33.9, 35.8, 38.3, 38.7, 39.0, 43.0, 43.7, 46.4, 50.1, 53.7, 80.3, 122.7, 170.8, 199.4 ppm
Melting point:		133-135 °C
Microanalysis:	Found:	C = 79.5%; H = 10.3%
	Calculated:	C = 79.7%; H = 10.2% (Calculated for $C_{21}H_{32}O_2$)