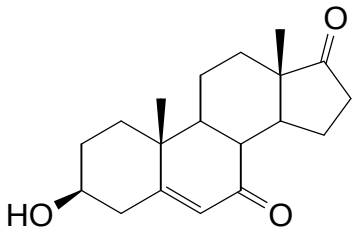
REFERENCE MATERIAL PRODUCT INFORMATION SHEET

Report ID: D833.2018.01

Compound Name: **7-Ketodehydroepiandrosterone**
Collection Number: D833
Chemical Formula: $C_{19}H_{26}O_3$
CAS Registry Number: 566-19-8
Structure:



Description: White powder
Batch Number: 03-S-10
Formula Weight: 302.4
Release date: 21st April 2004

Synonym: 7-Keto DHEA

Purity (mass fraction %): 98.9 ± 1.5% (95% coverage interval)

The purity value was obtained by subtraction from 100% of total impurities using a combination of traditional analytical techniques including HPLC with UV detection, thermogravimetric analysis, Karl Fischer analysis and ^{1}H NMR. Supporting evidence is provided by and elemental microanalysis.

HPLC:	Column:	Alltech C-18 5 µm (4.6 mm x 150 mm)
	Mobile Phase:	0-5 min 40% B; 5-8 min 40-60% B; 8-15 min 60% B; 15-16 min 60-40% B; 16-20 min 40% B
	Flow Rate:	1.0 mL/min
	Detector:	UV at 241 nm
	Relative peak area of main component:	
	Initial analysis:	Mean = 99.7%, s = 0.01% (7 sub samples in duplicate, August 2003)
	Re-analysis:	Mean = 99.4%, s = 0.05% (5 sub samples in duplicate, June 2008)
	Re-analysis:	Mean = 99.1%, s = 0.03% (5 sub samples in duplicate, May 2013)
	Re-analysis:	Mean = 99.3%, s = 0.03% (5 sub samples in duplicate, May 2018)
Thermogravimetric analysis:		Volatiles content < 0.1% and non-volatile residue < 0.2% mass fraction. (October 2003 and August 2006)
Karl Fischer moisture analysis:		Moisture content is ≤ 0.2% mass fraction. (May 2008 & 2013) Moisture content is ≤ 0.2% mass fraction. (May 2018)

Spectroscopic and other characterisation data

GC-MS:

Instrument:	HP6890/5973	
Column:	Zebron ZB-5, 30 m x 0.25 mm I.D. x 0.30µm	
Program:	180 °C (1 min), 10 °C/min to 210 °C, 20 °C/min to 310 °C (6 min)	
Injector:	250 °C	Transfer line temp: 310 °C
Carrier:	Helium, 1.0 mL/min	Split ratio: 15/1

The retention time of the parent compound is reported with the major peaks observed in the mass spectrum. The latter are reported as mass/charge and (in brackets) as a percentage relative to the base peak.

10.5 min: 302 (M^+, 100), 269 (8), 205 (11), 187 (14), 161 (36), 91 (21) m/z

Instrument:	HP5890/5971A	
Column:	BPX-5, 30 m × 0.25 mm I.D. × 0.25 µm	
Program:	180 °C (1 min), 10 °C/min to 210 °C, 20 °C/min to 310 °C (6 min)	
Injector:	250 °C	Transfer line temp: 310 °C
Carrier:	Helium, 1.0 mL/min	Split ratio: 15/1

The retention time of the mono-TMS derivative is reported with the major peaks observed in the mass spectrum. The latter are reported in a.m.u. and (in brackets) as a percentage relative to the intensity of the base peak.

12.5 min: 374 (M^+, 35), 207 (10), 163 (27), 129 (100), 73 (84) m/z

TLC: Conditions: Kieselgel 60F_{254}. Hexane/ethyl acetate (10:1) developed in ammonium molybdate/acidified ceric sulfate. Single spot observed, R_f = 0.23

IR:

Instrument:	Biorad WIN FTS3000MX FTIR
Range:	4000-400 cm^{-1}, KBr powder
Peaks:	3482, 2941, 2868, 1724, 1653, 1457, 1298, 1218, 1185, 1064 cm^{-1}

^{1}H NMR:

Instrument:	Bruker DMX-600	
Field strength:	600 MHz	Solvent: $CDCl_3$
Key spectral data:	δ 0.89 (3H, s), 1.22 (3H, s), 2.55 (1H, ddd), 2.81 (1H, m), 3.69 (1H, m), 5.74 (1H, d) ppm	

^{13}C NMR:

Instrument:	Bruker DMX-300	
Field strength:	75 MHz	Solvent: DMSO-d_6
Spectral data:	δ 13.8, 17.4, 20.5, 24.2, 30.8, 31.4, 35.4, 36.3, 38.4, 42.1, 44.1, 45.4, 47.5, 49.8, 69.4, 125.1, 157.7, 200.8, 219.4 ppm	

Melting point: 243-245 °C

Microanalysis: Found: C = 75.7%; H = 8.9%
Calc.: C = 75.5%; H = 8.7% (Calculated for $C_{19}H_{26}O_3$)

Expiration of certification
The property values are valid till 15th May 2023, 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 sample bottles that have been opened. In such cases, it is recommended that the end-user conduct their own in-house stability trials.

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

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

Homogeneity assessment
The homogeneity of the material was assessed using purity assay by HPLC with UV detection on five 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.

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.

Intended use
For ***in vitro*** laboratory analysis only.

Caution
Treat as hazardous substance. Use appropriate work practices when handling to avoid skin or eye contact, ingestion or inhalation of dust.

Legal notice
Neither NMI nor any person acting on NMI's behalf assumes any liability with respect to the use of, or for damages resulting from the use of, this reference material or the information contained in this certificate.

Authorised by:

S. R. Davies

Dr Stephen R. Davies,
Team Leader,
Chemical Reference Materials, NMI.
Dated: 25 May, 2018.

Characterisation data and property values specified in this report supersede those in all reports issued prior to 25th May 2018.