



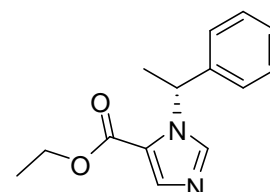
# CERTIFIED REFERENCE MATERIAL CERTIFICATE OF ANALYSIS

## NMIA D976: Etomidate

Report ID: D976.2020.03

Chemical Formula: C<sub>14</sub>H<sub>16</sub>N<sub>2</sub>O<sub>2</sub>

Molecular Weight: 244.3 g/mol



## Certified value

Batch No.	CAS No.	Purity (mass fraction)
11-D-27	33125-97-2	99.5 ± 0.6%

**IUPAC name:** Ethyl 1-[(1R)-1-phenylethyl]-1H-imidazole-5-carboxylate.

**Expiration of certification:** The property values are valid till 13 May 2028, i.e. eight 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.

**Description:** Off-white solid 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 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 20 °C 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 eight 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 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.  
20 September 2022

This report supersedes any issued prior to 20 September 2022.

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.

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## Characterisation Report:

The identity was confirmed by a range of spectroscopic techniques, NMR, IR and MS. Impurities of related structure were assessed by GC-FID. The purity value was obtained by mass balance from a combination of traditional analytical techniques, including GC-FID, thermogravimetric analysis, Karl Fischer analysis and <sup>1</sup>H NMR spectroscopy. The purity value is calculated as per Equation 1.

$$\text{Purity} = (100\% - I_{\text{ORG}}) \times (100\% - I_{\text{VOL}} - I_{\text{NVR}}) \quad \text{Equation 1}$$

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

Supporting evidence is provided by quantitative NMR, headspace GC-MS analysis of occluded solvents, and elemental microanalysis. The purity estimate by QNMR was obtained using a combination of the three proton triplet at 1.31 ppm, the two proton multiplet at 4.25 ppm and the one proton quartet at 6.35 ppm against a certified internal standard of dimethyl sulfone.

GC-FID: Instrument: Varian CP-3800  
 Column: VF-1MS, 29.58 m × 0.32 mm I.D. × 0.25 μm  
 Program: 150 °C (1 min), 10 °C/min to 300 °C (5 min)  
 Injector: 250 °C  
 Detector Temp: 320 °C  
 Carrier: Helium  
 Split ratio: 20/1  
 Relative peak area response of main component:  
 Initial analysis: Mean = 100.0%, s = 0.006% (10 sub samples in duplicate, November 2011)  
 Re - analysis: Mean = 100.0%, s = 0.001% (5 sub samples in duplicate, May 2020)

GC-FID: Instrument: Varian CP-3800  
 Column: HP-5, 30 m × 0.32 mm I.D. × 0.25 μm  
 Program: 150 °C (1 min), 10 °C/min to 300 °C (5 min)  
 Injector: 250 °C  
 Detector Temp: 320 °C  
 Carrier: Helium  
 Split ratio: 20/1  
 Relative peak area response of main component:  
 Initial analysis: Mean = 99.9%, s = 0.01% (10 sub samples in duplicate, November 2011)

Karl Fischer analysis: Moisture content < 0.1% mass fraction (November 2011, May 2020)

Thermogravimetric analysis: Non-volatile residue < 0.2% mass fraction (November 2011). The volatile content (e.g. organic solvents and/or water) could not be determined because of the inherent volatility of the material.

QNMR: Instrument: Bruker DMX-600  
 Field strength: 600 MHz  
 Solvent: CDCl<sub>3</sub> (7.26 ppm)  
 Internal standard: Dimethyl sulfone (100% mass fraction)  
 Initial analysis: Mean (1.31 ppm) = 99.7%, s = 0.3% (5 sub samples, November 2011)  
 Initial analysis: Mean (4.25 ppm) = 99.5%, s = 0.4% (5 sub samples, November 2011)  
 Initial analysis: Mean (6.35 ppm) = 99.9%, s = 0.4% (5 sub samples, November 2011)

## Spectroscopic and other characterisation data

GC-MS:	Instrument:	Agilent 6890/5973
	Column:	TG-1MS, 30 m x 0.25 mm I.D. x 0.25 $\mu$ m
	Program:	90 °C (1 min), 10 °C/min to 180 °C (7 min), 30 °C/min to 300 °C (3 min)
	Injector:	250 °C
	Split ratio:	20/1
	Transfer line temp:	300 °C
	Carrier:	Helium, 1.0 mL/min
	Scan range:	50-550 <i>m/z</i>
	The retention time of the parent compound is reported along with the major peaks in the mass spectrum. The latter are reported as mass/charge ratios and (in brackets) as a percentage relative to the base peak.	
	Parent (16.1 min):	244 ( $M^+$ , 12), 105 (100), 104 (20), 77 (11) <i>m/z</i>
HS-GC-MS:	Instrument:	Agilent 6890/5973/G1888
	Column:	DB-624, 30 m x 0.25 mm I.D. x 1.4 $\mu$ m
	Program:	50 °C (5 min), 7 °C/min to 120 °C, 15 °C/min to 220 °C (8.3 min)
	Injector:	150 °C
	Split ratio:	50/1
	Transfer line temp:	280 °C
	Carrier:	Helium, 1.2 mL/min
	Solvents detected:	None
TLC:	Conditions:	Kieselgel 60F <sub>254</sub> . Hexane/ethyl acetate (1:1) Single spot observed, $R_f$ = 0.16. Visualisation with UV at 254 nm.
IR:	Instrument:	Biorad FTS3000MX FT-IR
	Range:	4000-400 $cm^{-1}$ , KBr powder
	Peaks:	3399, 3130, 3098, 3063, 2999, 2975, 2931, 2881, 1709, 1523, 1456, 1391, 1349, 1278, 1211, 1109, 1049, 922, 864, 769, 712, 665 $cm^{-1}$
<sup>1</sup> H NMR:	Instrument:	Bruker Avance-400
	Field strength:	400 MHz
	Solvent:	CDCl <sub>3</sub> (7.26 ppm)
	Spectral data:	$\delta$ 1.31 (3H, t, $J$ = 7.1 Hz), 1.85 (3H, d, $J$ = 7.1 Hz), 4.25 (2H, m), 6.35 (1H, q, $J$ = 7.1 Hz), 7.18 (2H, m), 7.27 (1H, m), 7.32 (2H, m), 7.71 (1H, s), 7.77 (1H, d, $J$ = 0.8 Hz) ppm Di-isopropyl ether estimated at 0.3% mass fraction was observed in the <sup>1</sup> H NMR.
<sup>13</sup> C NMR:	Instrument:	Bruker Avance-400
	Field strength:	100 MHz
	Solvent:	CDCl <sub>3</sub> (77.0 ppm)
	Spectral data:	$\delta$ 14.2, 22.2, 55.2, 60.4, 122.6, 126.2, 127.9, 128.8, 138.1, 139.7, 141.1, 160.3 ppm
Melting point:		67-69 °C
Microanalysis:	Found:	C = 68.9%; H = 6.6%; N = 11.6% (November, 2011)
	Calculated:	C = 68.8%; H = 6.6%; N = 11.5% (Calculated for C <sub>14</sub> H <sub>16</sub> N <sub>2</sub> O <sub>2</sub> )