Australian Government
Department of Industry, Science and Resources

National Measurement Institute

NATA
WORLD RECOGNISED ACCREDITATION

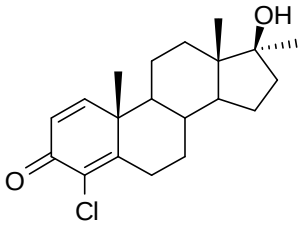
# CERTIFIED REFERENCE MATERIAL CERTIFICATE OF ANALYSIS

## NMIA D613: Turinabol

Report ID: D613.2023.01 (Ampouled 200423)

Chemical Formula: $C_{20}H_{27}ClO_2$

Molecular Weight: 334.9 g/mol




## Certified value

| Batch No. | CAS No. | Mass per ampoule |
|---|---|---|
| 99-S-28 | 2446-23-3 | 969 ± 15 µg |

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

**IUPAC name:** (17β)-4-Chloro-17-hydroxy-17-methylandrosta-1,4-dien-3-one.

**Expiration of certification:** The property values are valid till 07 June 2026, three 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 CRM is intended for a single use to prepare a standard solution containing D613. This material was prepared by sourced from an external supplier and certified for identity and purity by NMIA.

**Intended use:** This certified reference material is suitable for use as a primary calibrator.

**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 969 ± 15 µg of anhydrous turinabol. 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.

**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 all impurities are quantified as a mass fraction and subtracted from 100%.

**Stability:** This material has demonstrated stability over a minimum period of three 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 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.
9 June 2023

This report supersedes any issued prior to 09 June 2023

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:

| | | |
|---|---|---|
| GC-FID: | Instrument: | Agilent 7890 |
| | Column: | HP-5, 30 m × 0.32 mm I.D. × 0.25 μm |
| | Program: | 200 °C (1 min), 20 °C/min to 250 °C, 3 °C/min to 280 °C, 30 °C/min to 300 °C (3 min) |
| | Injector: | 250 °C |
| | Detector Temp: | 320 °C |
| | Carrier: | Helium |
| | Split ratio: | 20/1 |
| | Relative mass fraction of the main component: | |
| | Initial analysis: | Mean = 96.9%, s = 0.02% (7 ampoules in duplicate, June 2020) |
| | Re-analysis: | Mean = 96.7%, s = 0.02% (5 ampoules in duplicate, May 2021) |
| | Re-analysis: | Mean = 96.8%, s = 0.02% (5 ampoules in duplicate, June 2023) |

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

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 GC-FID, thermogravimetric analysis, Karl Fischer analysis and $^{1}$H NMR spectroscopy. The purity value is calculated as per Equation 1.

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

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

Supporting evidence is provided by qualitative elemental microanalysis.

| | | |
|---|---|---|
| GC-FID: | Instrument: | Varian CP-3800 or Agilent 7890 |
| | Column: | HP-5, 30 m × 0.32 mm I.D. × 0.25 μm |
| | Program: | 200 °C (1 min), 20 °C/min to 250 °C, 3 °C/min to 280 °C, 30 °C/min to 300 °C (3 min) |
| | Injector: | 250 °C |
| | Detector Temp: | 320 °C |
| | Carrier: | Helium |
| | Split ratio: | 20/1 |
| | Relative mass fraction of the main component: | |
| | Initial analysis: | Mean = 97.1%, s = 0.06% (7 sub samples in duplicate, January 2000) |
| | Re-analysis: | Mean = 96.7%, s = 0.02% (5 sub samples in duplicate, January 2009) |
| | Re-analysis: | Mean = 97.0%, s = 0.02% (7 sub samples in duplicate, June 2020) |
| Karl Fischer analysis: | | Moisture content 0.4% mass fraction (January 2009)<br>Moisture content 0.6% mass fraction (June 2020) |
| Thermogravimetric analysis: | | Volatile content not determined due to the nature of the material<br>Non-volatile residue < 0.2% mass fraction (January 2009) |

## Spectroscopic and other characterisation data

| | | |
|---|---|---|
| GC-MS: | Instrument: | Agilent 6890/5973 |
| | Column: | HP Ultra 1, 17 m x 0.2 mm I.D. x 0.11 μm |
| | Program: | 185 °C (0.2 min), 3 °C/min to 236 °C, 10 °C/min to 265 °C, 30 °C/min to 310 °C (2 min) |
| | Injector: | 280 °C |
| | Split ratio: | 15/1 |
| | 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 (14.9 min): | 336 ($M^+$, 24), 334 ($M^+$, 62), 281 (23), 265 (20), 240 (54), 179 (35), 161 (76), 157 (39), 155 (100), 121 (77), 91 (95) *m/z* |
| TLC: | Conditions: | Kieselgel 60$F_{254}$. Hexane/ethyl acetate (30:20)<br>Single spot observed, $R_f$ = 0.25 (5 sub samples) |
| IR: | Instrument: | FT-IR, Biorad WIN FTS40 |
| | Range: | 4000-400 $cm^{-1}$, Nujol mull |
| | Peaks: | 3502, 1649, 1632, 1584, 1457, 1373, 1066 $cm^{-1}$ |
| $^1$H NMR: | Instrument: | Bruker Advance-300 |
| | Field strength: | 300 MHz |
| | Solvent: | $CDCl_3$ (7.26 ppm) |
| | Spectral data: | δ 0.94 (3H, s), 1.19 (3H, s), 1.31 (3H, s), 6.35 (1H, d, *J* = 12 Hz), 7.16 (1H, d, *J* = 12 Hz) ppm |
| $^{13}$C NMR: | Instrument: | Bruker Advance-300 |
| | Field strength: | 75 MHz |
| | Solvent: | $CDCl_3$ (77.2 ppm) |
| | Spectral data: | δ 14.4, 19.5, 23.3, 23.7, 26.2, 29.3, 31.7, 32.6, 36.7, 39.2, 46.0, 46.7, 50.1, 53.6, 81.7, 126.6, 128.6, 155.8, 163.0, 178.6 ppm |
| Melting point: | | 145-146 °C |
| Microanalysis: | Found: | C = 71.8%; H = 8.3%; Cl = 10.9% (October 1999) |
| | Calculated: | C = 71.7%; H = 8.1%; Cl = 10.6% (Calculated for $C_{20}H_{27}ClO_2$) |