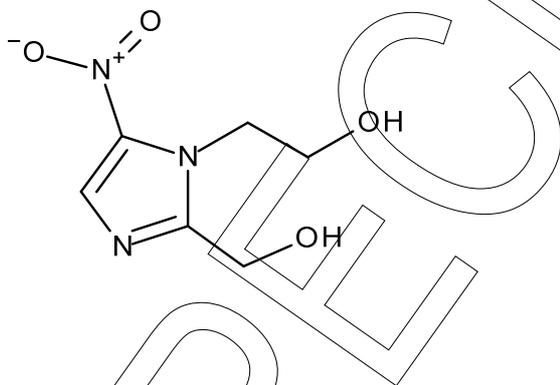




Certificate of Analysis

Reference Standard

Hydroxymetronidazole (1-(2-Hydroxyethyl)-2-hydroxymethyl-5-nitroimidazole)



Molecular Formula: $C_6H_9N_3O_4$
Molecular Weight: 187.15
CAS Number: 4812-40-2

Catalogue Number: LGCFOR0012.04
Lot Number: 173207
Long-term Storage: 2 to 8 °C, dark
Appearance: white solid
Melting Point: 119 °C
Assay 'as is': 98.2 %

Date of shipment: **2020-November-30**

This certificate is valid for two years from the date of shipment provided the substance is stored under the recommended conditions unopened in the original container.

LGC Quality | ISO 9001:2008
DQS 102448 QM08

LoGiCal[®]
produced by LGC

LGC GmbH, Louis-Pasteur-Str. 30, D-14943 Luckenwalde, Germany

© 2012 LGC limited. All rights reserved. LGC Standards is part of the LGC Group.
LoGiCal is a registered trademark of LGC Standards GmbH

6 Pages

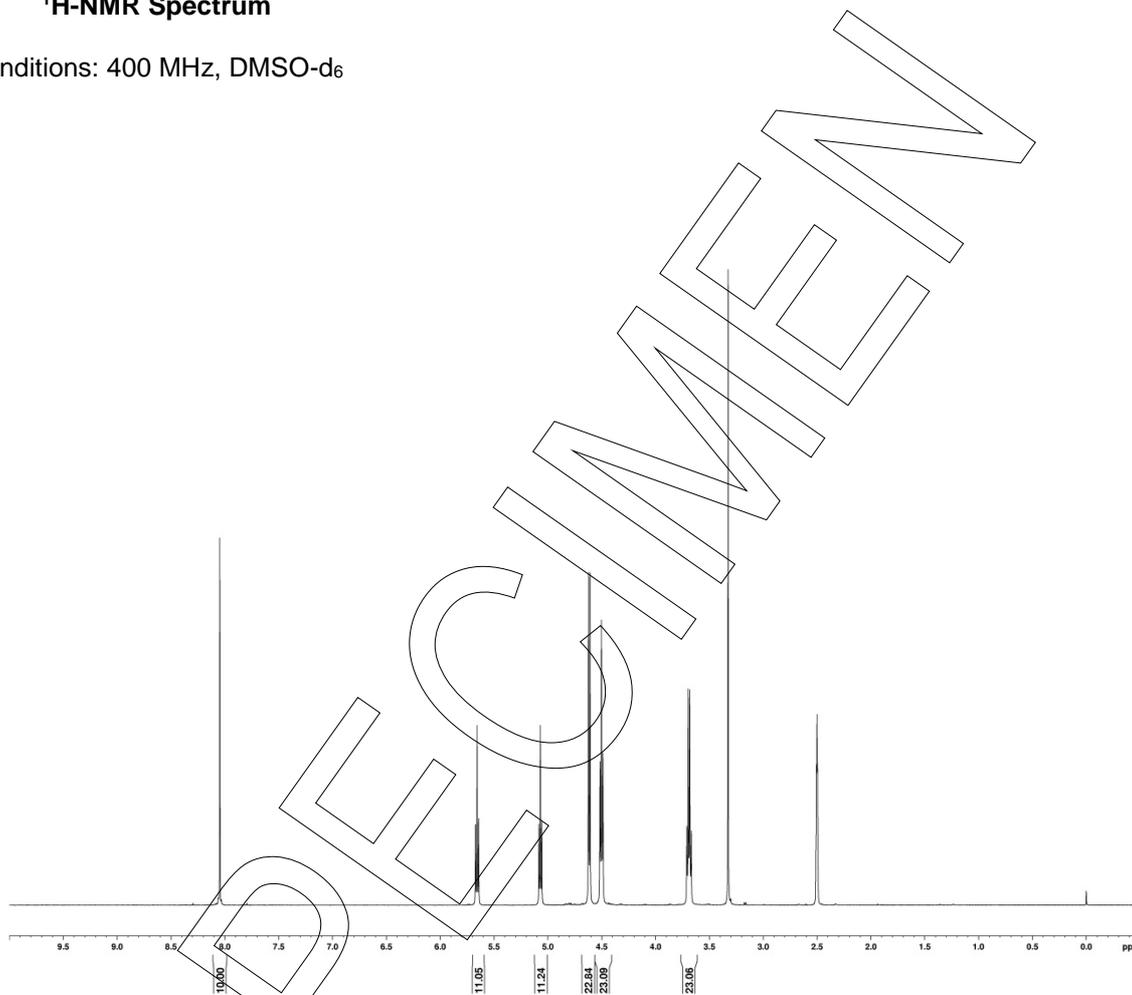


I. Identity

The identity of the reference substance was established by following analyses.

Ia. ¹H-NMR Spectrum

Conditions: 400 MHz, DMSO-d₆

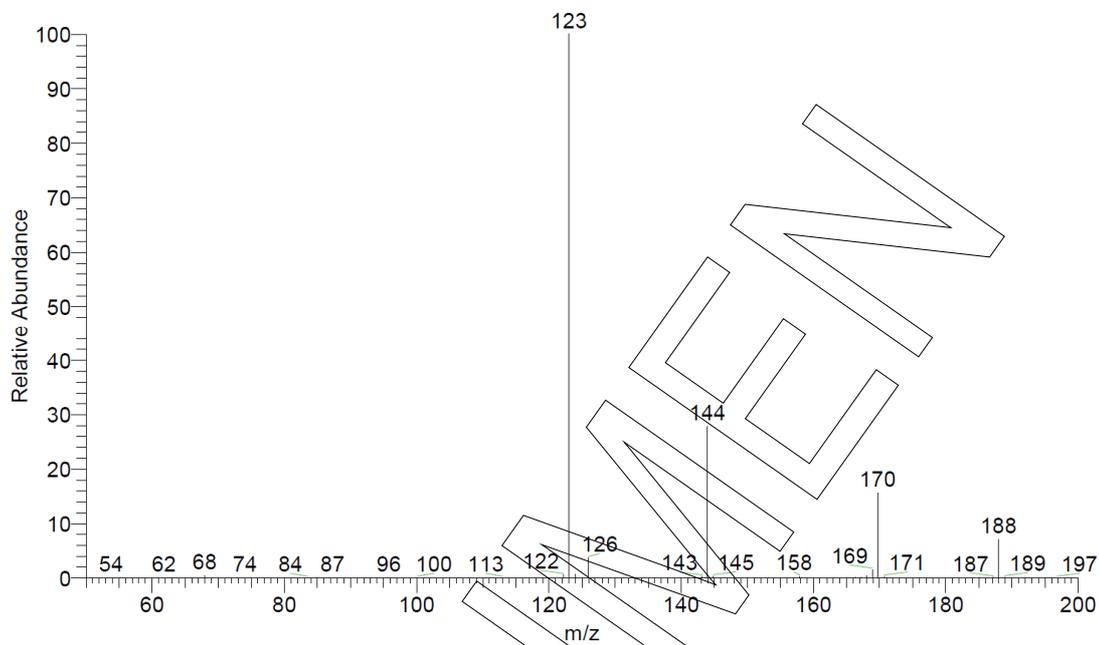


The structure is confirmed by the signals of the spectrum and their interpretation.



Ib. Mass Spectrum

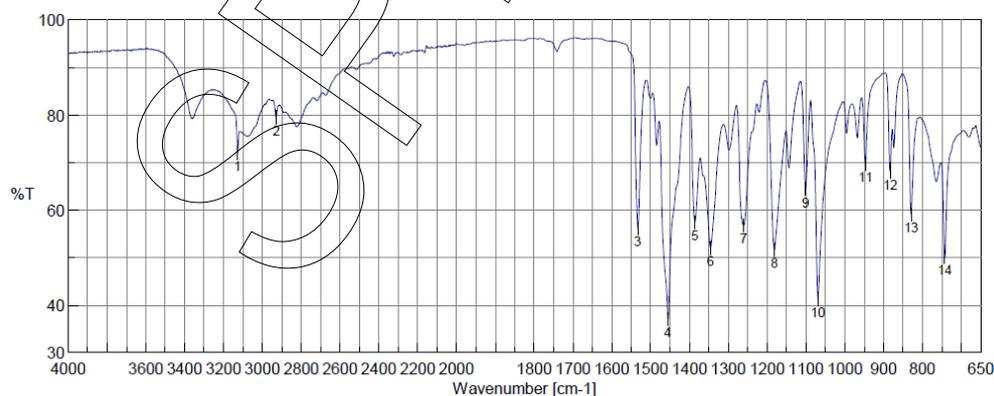
Method: 4.5 kV ESI+; vaporization temperature: 200 °C



The signals of the mass spectrum and their interpretation are consistent with the structural formula.

Ic. IR Spectrum

Method: Attenuated Total Reflection Fourier Transform Infrared (ATR-FTIR) Spectroscopy



No.	Position	Intensity
1	3127.01	71.9918
2	2929.34	79.4965
3	1533.13	56.149
4	1455.99	37.0027
5	1385.6	57.3596
6	1346.07	52.0792
7	1260.25	56.7407
8	1181.19	51.6089
9	1101.15	64.3722
10	1069.33	41.3399
11	946.877	69.7318
12	883.238	67.926
13	829.241	59.031
14	742.46	50.0959

The signals of the IR spectrum and their interpretation are consistent with the structural formula.



II. Purity

IIa. High Performance Liquid Chromatography (HPLC)

The purity of the reference substance was analysed by high performance liquid chromatography (HPLC).

HPLC Conditions:

Column:

Discovery HS F5
3 μm , 150 x 4 mm

Conditions:

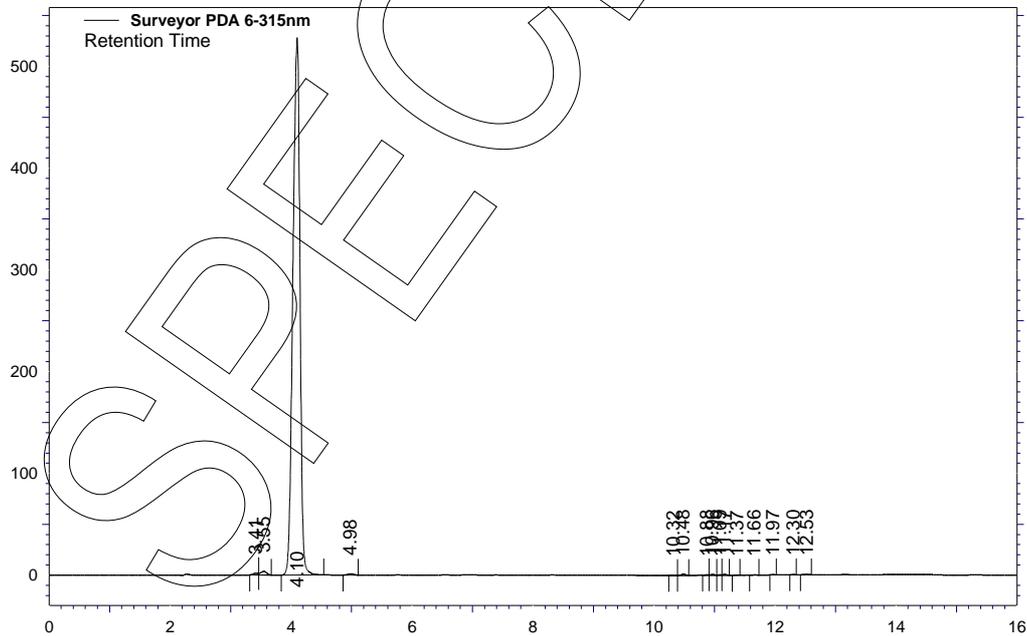
1.0 ml/min, 40 °C
0 – 6 min Water/Acetonitrile 97/3
6 – 9 min Water/Acetonitrile to 70/30
9 – 11 min Water/Acetonitrile to 97/3
11 – 16 min Water/Acetonitrile 97/3 (v/v);
0.1 % H_3PO_4

Detector:

DAD
315 nm

Injector:

Auto
3 μl ; 0.1344 mg/ml in
Water/Acetonitrile 50/50 (v/v)





Area Percent Report - Sorted by Signal

Pk #	Retention Time	Area	Area %
1	3.41	10517	0.24
2	3.55	25167	0.58
3	4.10	4283223	98.44
4	4.98	8611	0.20
5	10.32	594	0.01
6	10.48	6183	0.14
7	10.88	1513	0.03
8	10.96	3684	0.08
9	11.09	1892	0.04
10	11.17	3986	0.09
11	11.37	492	0.01
12	11.66	1144	0.03
13	11.97	1092	0.03
14	12.30	889	0.02
15	12.53	2332	0.05
Totals		4351319	100.00

For the calculation the system peaks were ignored. The content of the analyte was determined as ratio of the peak area of the analyte and the cumulative areas of the purities, added up to 100 %.

Results:

Average 98.43 %
Number of results n=3
Standard deviation 0.02 %

IIb. Water Content

Method: Karl Fischer titration

Results:

Average 0.12 %
Number of results n=3
Standard deviation 0.01 %



IIc. Residual Solvents

Method: ¹H-NMR

Result: 0.08 % Methanol

III. Final Result

Chromatographic purity (HPLC)	98.43 %
Water content	0.12 %
Residual solvents	0.08 %
Assay (100 % method)¹	98.23 %

The assay is assessed to be 98.2 % 'as is'

The assay 'as is' is equivalent to the assay based on the not anhydrous and not dried substance respectively.

Release Date:

Luckenwalde, 2018-02-23

Dr. Sabine Schröder
Product Release

¹ The calculation of the 100 % method follows the formula:

$$\text{Assay (\%)} = (100 \% - \text{volatile contents}) * \frac{\text{Purity (\%)}}{100 \%}$$

Volatile contents are considered as absolute contributions, purity is considered as relative contribution.