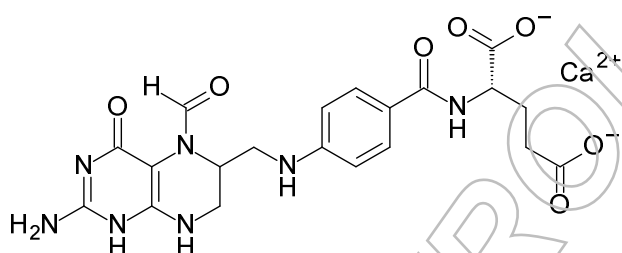




Certificate of Analysis

Reference Standard

Calcium Folate



Molecular Formula: $C_{20}H_{24}N_7O_7$ Ca
Molecular Weight: 511.50
CAS Number: 1492-18-8

Catalogue Number: MM0324.00
Lot Number: 71760
Long-term Storage: 2 to 8 °C, dark
Appearance: light yellow solid
hygroscopic
Melting Point: > 340°C (dec.)
Assay 'as is': 88.3 %

Date of shipment: **2017-September-05**

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

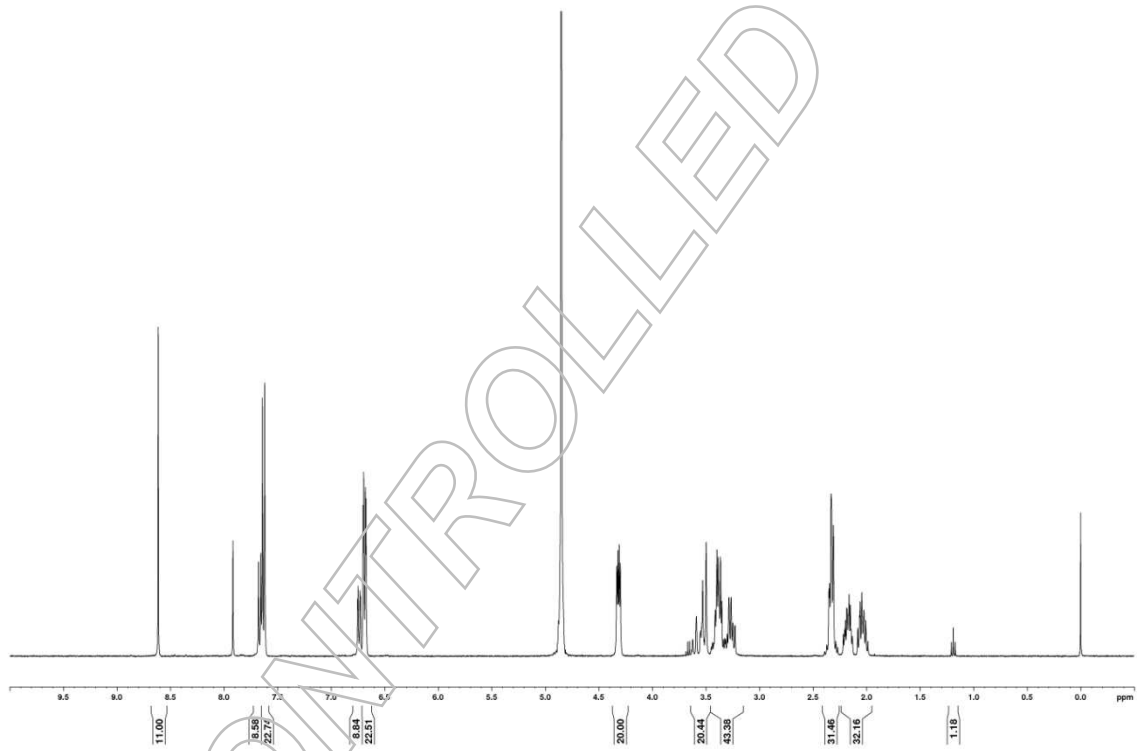


I. Identity

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

1a. ¹H-NMR Spectrum

Conditions: 400 MHz, D₂O

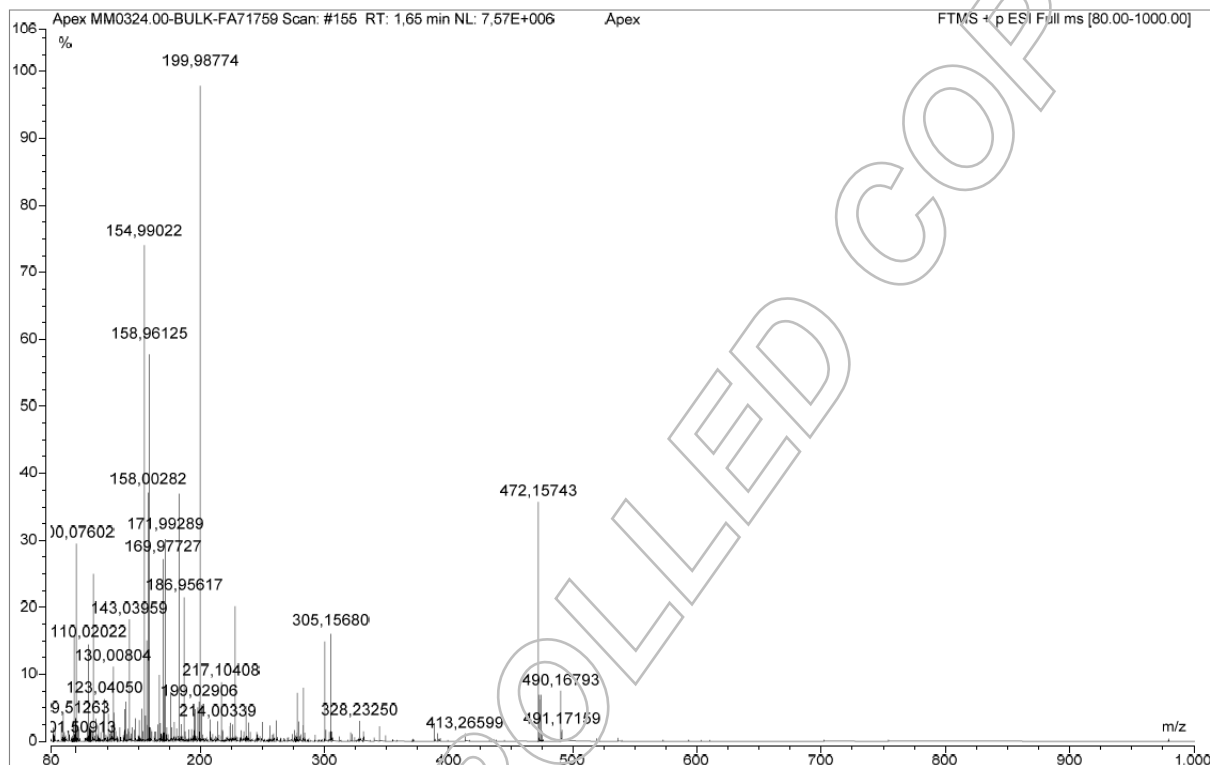


The structure is confirmed by the signals of the spectrum and their interpretation.



Ib. Mass Spectrum

Method: 3.5 kV ESI+; capillary temperature: 269 °C



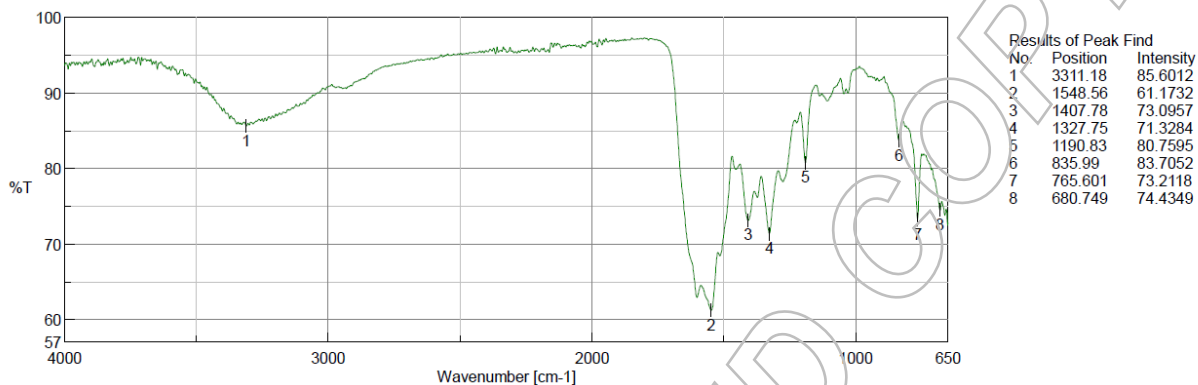
Theoretical value: 472.15752

The signal of the MS spectrum is consistent with the theoretical value and its interpretation is consistent with the structural formula.



Ic. IR Spectrum

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



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

II. Purity

Ila. High Performance Liquid Chromatography (HPLC)

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

HPLC Conditions:

Column:

Kinetex Phenyl-Hexyl
1.7 µm, 100 x 2.1 mm

Conditions:

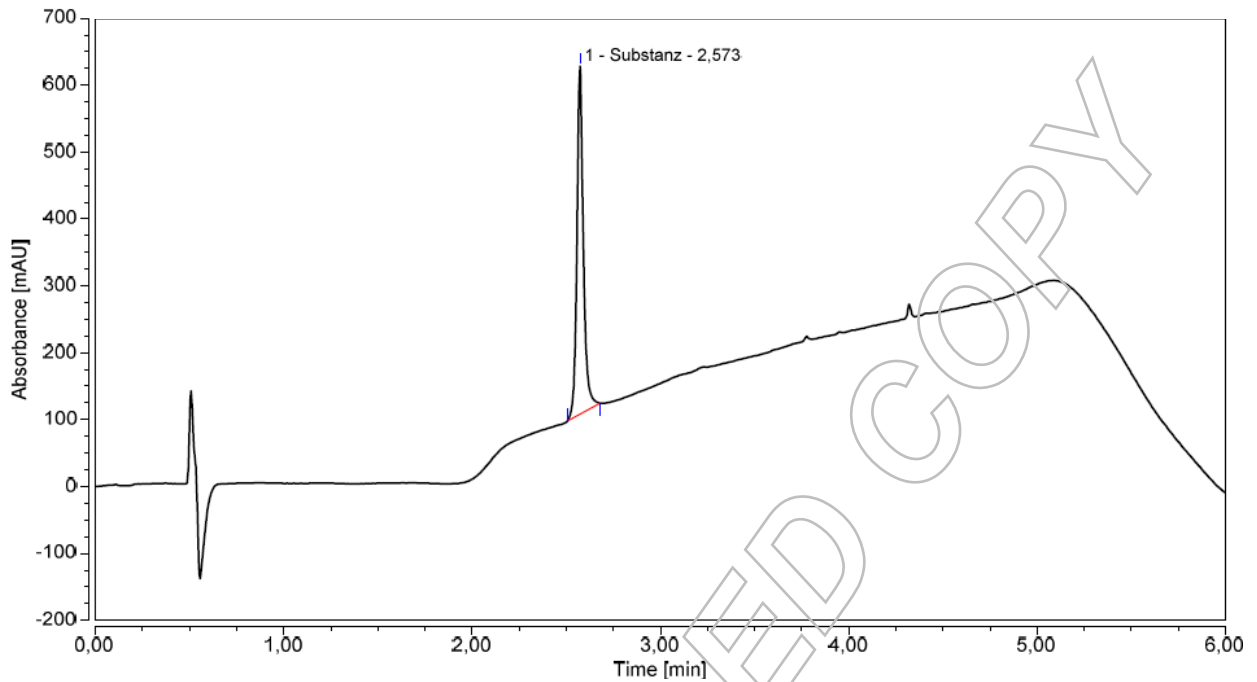
0.5 ml/min, 40 °C
0-1 min Water/Acetonitrile 98/2
1-4 min Water/Acetonitrile to 2/98
4-5 min Water/Acetonitrile to 98/2
5-6 min Water/Acetonitrile 98/2 (v/v);
0.1 % HCOOH

Detector:

DAD
220 nm

Injector:

Auto
2 µl; 0.044 mg/ml in
Water/Acetonitrile 50/50 (v/v)



Area Percent Report - Sorted by Signal

Pk #	Retention Time	Area	Area %
1	2.573	20.400	100.00
Totals		20.400	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 100 %
Number of results n=3
Standard deviation < 0.01 %

IIb. Water Content

Method: Karl Fischer titration

Results:

Average 11.40 %
Number of results n=3
Standard deviation 0.04 %



IIc. Residual Solvents

Method: ¹H-NMR

Result: 0.35 % Ethanol

III. Final Result

Chromatographic purity (HPLC)	100 %
Water content	11.40 %
Residual solvents	0.35 %
Assay (100 % method)¹	88.25 %

The assay is assessed to be 88.3 % 'as is'

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

Release Date:

Luckenwalde, 2015-07-30

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.