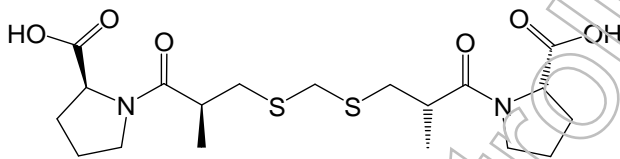




# Certificate of Analysis

## Reference Standard

1,1'-[Methylenebis[sulfanediyl-  
[(2S)-2-methyl-1-oxopropane-3,1-diyl]]]-  
bis[(2S)-pyrrolidine-2-carboxylic] Acid



Molecular Formula:  $C_{19}H_{30}N_2O_6S_2$   
Molecular Weight: 446.58  
CAS Number: [ unlisted ]

Catalogue Number: MM0067.13  
Lot Number: 1839  
Long-term Storage: 2 to 8 °C, dark  
Appearance: white solid  
Melting Point: 54 °C  
hygroscopic  
Assay 'as is': 94.7 %

Date of shipment: **2015-July-04**

This certificate is valid for one year from the date of shipment provided the substance is stored under the recommended conditions.



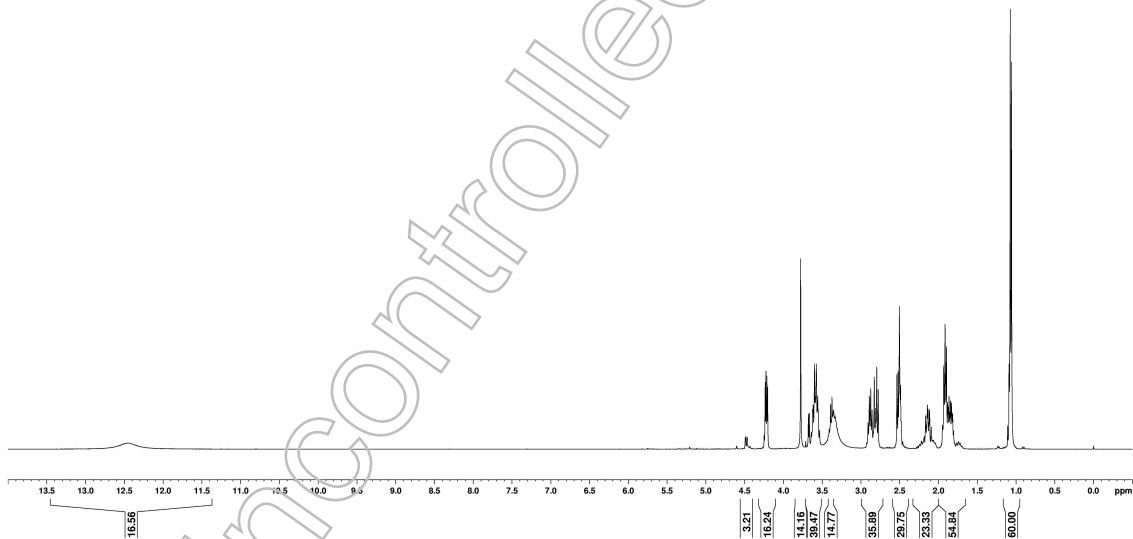
## I. Identity

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

### Ia. <sup>1</sup>H-NMR Spectrum

Conditions: 400 MHz, DMSO-d<sub>6</sub>

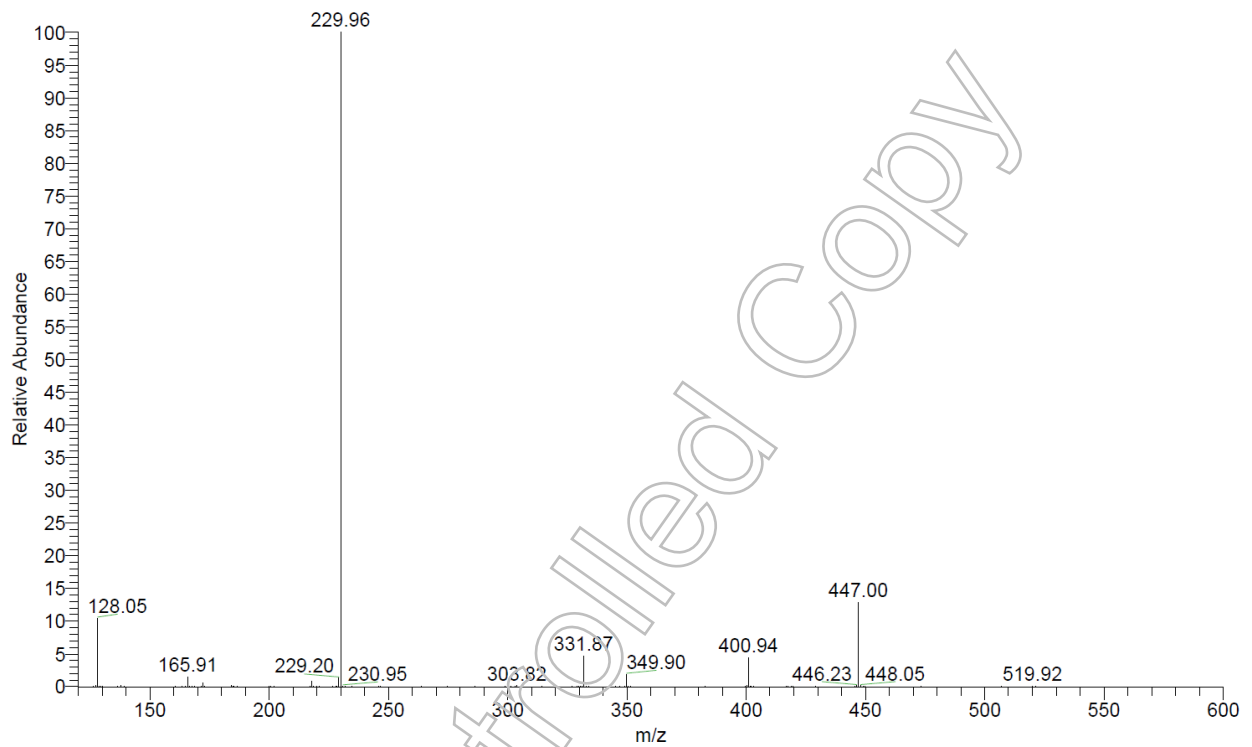
The structure is confirmed with the signals of the spectrum and their interpretation.





## Ib. Mass Spectrum

Method: 4.5 kV ESI; vaporization temperature: 200 °C, direct inlet



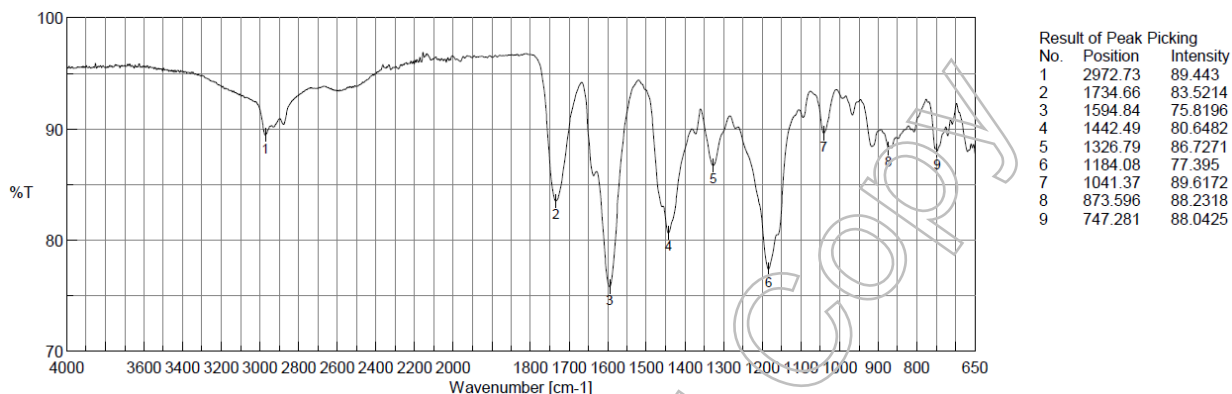
m/z	fragments
447	[ MH ]
401	[ M - COOH ]
230	[ C <sub>10</sub> H <sub>16</sub> NO <sub>3</sub> S ]

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



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

## II. Purity

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

### HPLC Conditions:

#### Column:

Hypersil Gold (C18)  
5 µm, 150 x 4.6 mm

#### Conditions:

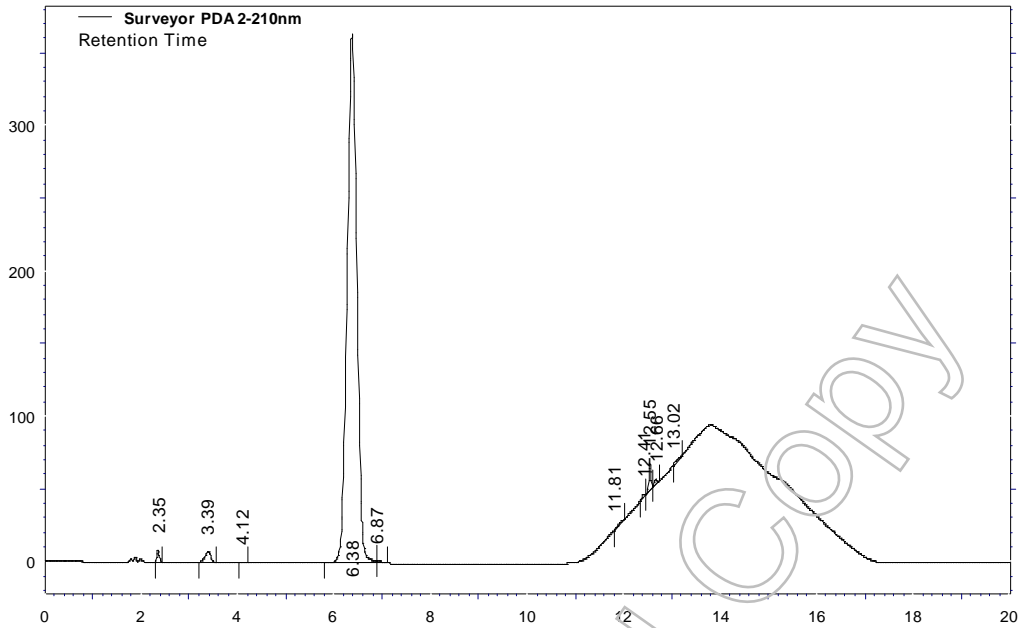
1.0 ml/min, 40 °C  
0 – 8 min Water/Acetonitrile 76/24  
8 – 11 min Water/Acetonitrile to 30/70  
11 – 14 min Water/Acetonitrile to 76/24  
14 – 20 min Water/Acetonitrile 76/24 (v/v);  
0.1 % H<sub>3</sub>PO<sub>4</sub>

#### Detector:

DAD  
210 nm

#### Injector:

Auto  
3.5 µl; 0.2552 mg/ml in  
Water/Acetonitrile 50/50 (v/v)



Area Percent Report - Sorted by Signal

Pk #	Retention Time	Area	Area %
1	2.35	33887	0.64
2	3.39	65127	1.23
3	4.12	918	0.02
4	6.38	5087673	96.01
5	6.87	12150	0.23
6	11.81	5548	0.10
7	12.41	8228	0.16
8	12.55	64425	1.22
9	12.66	13625	0.26
10	13.02	7379	0.14
Totals		5298960	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 96.00 %  
Number of results n=6  
Standard deviation 0.03 %



### III. Water Content

Method: Karl Fischer titration

#### Results:

Average	1.40 %
Number of results	n=3
Standard deviation	0.10 %

### IV. Residual Solvents

Method: <sup>1</sup>H-NMR

No significant amounts of residual solvents were detected (< 0.05 %).

### V. Final Result

Total impurities (HPLC)	4.00 %
Water content	1.40 %
Residual solvents	n. d. (not detected)
Assay (100 % method) <sup>1</sup>	94.66 %

The assay is assessed to be 94.7 % 'as is'

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

Release Date: 2013-05-07

LGC GmbH

Dr. Sabine Schröder  
Product Release

<sup>1</sup> The calculation of the 100 % method follows the formula:

$$\text{Assay (\%)} = (100 \% - \text{KF} - \text{RES}) \times \frac{\text{Purity HPLC (\%)}}{100 \%}$$

Water (KF) and Residual solvents (RES) are considered as absolute contributions, HPLC purity is considered as relative contribution.