HamyLab

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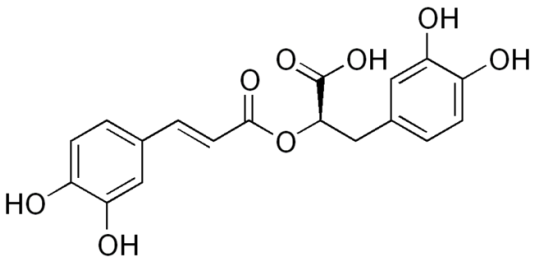
Certificate of Analysis

Product Name: Rosmarinic acid
Grade Working standard
Product Number: R0002
Lot Number: -
Brand: HamyLab
CAS Number: 20283-92-5
Formula: $C_{18}H_{16}O_8$
Formula Weight: 360.3 g/mol
Long term storage: 2-8 °C
Quality Release Date: 00-???-2022
PubChem Preview ID: -



Application Note: For peak identification. It is freely soluble in ethanol, DMSO or dimethyl formamide to approximately 25 mg/mL, and has a pKa = 3.57.

Test	Specification	Test Result
Appearance	Crystalline	Crystalline
Color	light beige	light beige
Solubility	ethanol, DMSO or dimethyl formamide to approximately 25 mg/mL	ethanol, DMSO or dimethyl formamide to approximately 25 mg/mL
Purity, (HPLC)	≥ 99.2 ± 0.4%	≥ 99.2 ± 0.4%
Identification (UV spectrum)	Confirm	Conform
Identification (IR-spectroscopy)	Confirm	Conform
Identification (1H-NMR-spectroscopy)	Confirm	Conform
Identification (13C-NMR-spectroscopy),		
Peak purity, (HPLC)	Confirm	Conform
Residual solvents, (headspace-GC)		
Inorganic impurities, (ICP-MS), for reference substances		
Water content, (micro determination, coulometric titration),		
Melting point	171-175 °C	171-175 °C

The identity of the Rosmarinic acid has been substantiated by at least two independent analytical methods: IR, NMR, UV and HPLC analysis. A mass balance approach, which takes chromatographic purity into account, as well as the contents of water, residual solvents, inorganic impurities, and the counter ion (if the reference standard is present as a salt) is applied in the calculation of the absolute purity as given in this COA

The absolute purity value (and not just the chromatographic purity result obtained by means of HPLC or GC) must be used in all quantitative calculations as the chromatographic techniques do not yet account for water, residual solvents and inorganic impurities.

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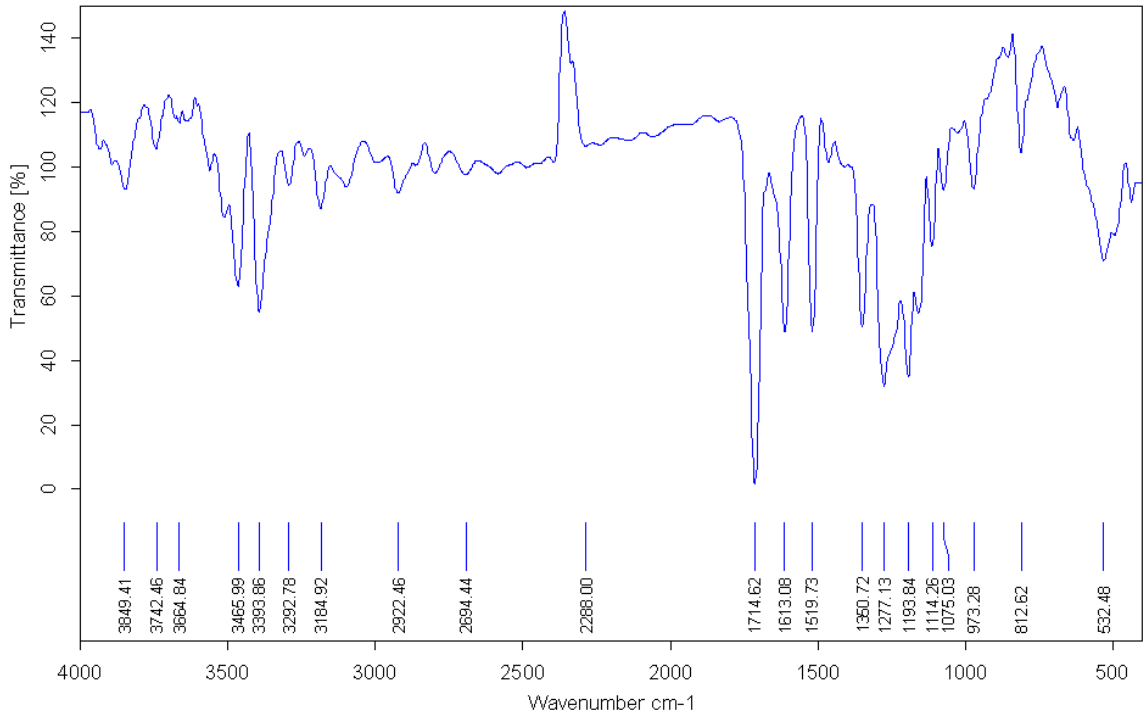
Identity:

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

1 IR Spectrum

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

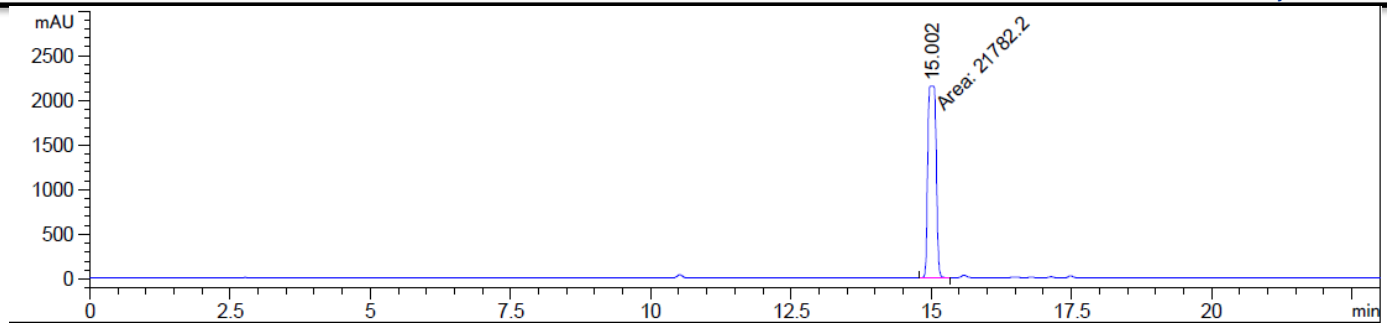


The signals of the IR spectrum and their interpretation are consistent with structural formula.

2 HPLC

Method: The purity of the reference substance was analysed by high performance liquid chromatography (HPLC). Analytical HPLC analysis was carried out using reverse-phase chromatography under the following conditions:

Column:	Ultisil XB-C18, 5 µm, 4.6 mm X 250 mm, 1/pk
Detector:	Photodiode-Array Detection (PDA)
Flow:	1 ml/min
Injector:	Manually
mob. Phase A:	Buffer phosphate (Ph=2.7)
mob. Phase B:	Methanol



Area Percent Report - Sorted by Signal						
No	**Retention Time**	**Area**	**Amt/Area**	**Amount [ppm]**	**Type**	**Real area %**
1	15.005	1.45669e4	6.73766e-2	981.46651	MM	100%
Total:						
For the calculation the system peaks were ignored. The content of the analyte was determined as the ratio of the peak area of the analyte and the cumulative areas of the purities, added up to 100 %.						
Results:						
Average:		15.049				
Number of results:		n=5				
Standard deviation:		< 0.01 %				

3 ^{1}H-NMR Spectrum

Conditions: 400 MHz, DMSO-d_6

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

4 Water Content

Method: Karl Fischer titration

Results:	
Average	0.55 %
Number of results	n=3
Standard deviation	0.08 %

5 Residual Solvents

Residual Solvent	Average	Method
Dioxane	0.08 %	^{1}H-NMR

Final Result

Chromatographic purity (HPLC) 99.42 %
Water content 0.55 %
Residual solvents 0.08 %
Assay (100 % method)[1] 98.79 %

The assay is assessed to be 98.8 % 'as is'

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

Maryam Yousefi /PhD/PhD,
Supervisor Quality Assurance

[1]The calculation of the 100% method follows the formula:
Assay (%)= (100%-volatile contents) * Purity (%)/100%
Volatile contents are considered as absolute contributions, purity is considered as relative contribution