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 Reference Substances
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 ref-substances@phytolab.de
 https://phyproof.phytolab.com

Certificate of analysis

Article:	82674 4',5,7-Trihydroxy 3,3',6,8-tetramethoxyflavone
Certificate # / Lot Number:	70191
Material batch:	5286
Sample-ID:	70780
End of analysis:	08/2023
Expiry date:	08/2028

Test	Unit	Specified value	Testresult
Appearance, SOP 100005		powder	conform
Color, SOP 100006		white - yellow	conform
Identification (1H-NMR-spectroscopy), (outsourced), SOP 206010		conform	conform
Identification (13C-NMR-spectroscopy), (outsourced), SOP 206020		conform	conform
Identification (HPLC-HR/MS), SOP 204125		conform	conform
Identification (IR-spectroscopy, Ph.Eur. 10.3, 2.2.24 / USP43 NF37 <197>), SOP 206000		conform	conform
Water content, (micro determination, coulometric titration), Ph.Eur. 10.0., 2.5.32, SOP 304291 Vers. 2018-01: Mean value	%		4.5
4',5,7-Trihydroxy-3,3',6,8-tetramethoxyflavone (HPLC), method 1 (% AU), SOP 440546	%	≥ 90.00	96.75
Peakpurity, (HPLC), SOP 401367		conform	conform

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Test	Unit	Specified value	Testresult
Inorganic impurities, (ICP-MS), for reference substances, SOP 811701:	%		<0.1
Calcium			
Potassium	%		<0.1
Magnesium	%		<0.1
Sulfur	%		<0.1
Sodium	%		<0.1
Phosphorus	%		<0.1
Aluminium	%		<0.1
Residual solvents, (headspace-GC), SOP 805765:	%		
Residual solvents (LOQ: 0.050)			<0.050
Content, SOP 890000, calculated in (%): (100 - water - residual solvents - inorganic impurities) x chromatographic purity / 100	%		92

This PhytoLab phyproof© reference standard is by definition a primary reference standard and does not need to be qualified against any other reference standard. The identity of the reference standard has been substantiated by at least two independent analytical methods such as IR, NMR, UV or MS 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 (see description of SOP 8900XX).

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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Vestenbergsgreuth, 02/Aug/2023

Sibylle Friess

QC Reference Substances

This is a computer print and valid without signature. A signed certificate of analysis can be taken on request.

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Material batch: 5286

Further information:

Shelf life/stability: The stated [expiry](#) date applies when the reference substance is stored in the original unopened container within the specified temperature range. PhytoLab does not guarantee the stability of the reference substance once the vial has been opened.

Long-term storage and handling: The reference standard should be stored in the original unopened vial, protected against light and humidity in an airtight container, within the temperature range given on the label and accompanying data sheet. If stored below room temperature, the vial should be warmed up to room temperature in a desiccator before it is opened in order to avoid condensation of humidity. The user assumes responsibility for deciding how previously opened reference standard vials should be used and the user must ensure that the contents of opened vials are still suitable for their intended use.

Exact weight: the exact weight of each vial is given on the label of the inner vial to two decimal places. This information may be used to produce stock solutions of a known concentration without having to weigh in the reference substance again. If used for this purpose, the content of the vial must be quantitatively transferred to a volumetric flask and filled up to the required level. Please note that PhytoLab is unable to guarantee the stability of the reference standard in solution.

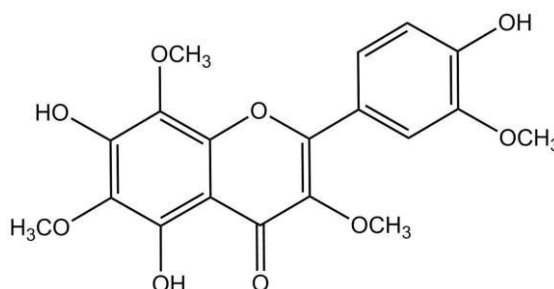
Intended use: this reference standard is solely intended for laboratory analytical purposes, research & development, and scientific teaching and training purposes. It may not be used for any other purpose and particularly not for use in, or the production of, food, animal feed, human or veterinary drugs, cosmetics, medicinal products or diagnostic agents, including in-vitro diagnostic agents. PhytoLab is unable to guarantee the suitability of this reference standard for any particular application other than its qualitative and quantitative use in chromatography and identification testing.

Further information about this reference standard can be found on the accompanying data sheet or in our webshop. Spectral and chromatographic data, and a description of the applied chromatographic method, are provided in the attachments to this COA. A detailed explanation of all data given on the COA can be found in the guide that is available from the download area in our webshop, where you can also download all of the safety data sheets.

Product Data Sheet

4',5,7-Trihydroxy 3,3',6,8-tetramethoxyflavone

Product #: 82674



Physicochemical Data

CAS #:	58130-91-9
Molecular formula:	C ₁₉ H ₁₈ O ₉
Molecular weight [g/mol]:	390.35
Substance class:	Flavonoids
Subgroup 1:	Flavones
Subgroup 2:	3-Hydroxyflavones
Solubility:	soluble in methanol Please note that this solubility information is based on in-house experience or taken from published data. It is not meant to guarantee solubility up to a specific concentration, nor does it guarantee stability of the reference substance in solution.

Additional Information

Source:	botanical origin	
Long-term storage conditions:	15-25 °C	
Manufacturer:	Phytolab GmbH & Co.KG Dutendorfer Straße 5-7 91487 Vestenbergsgreuth Germany	Tel.: +49 9163 88-395 Fax: +49 9163 88-456 Mail: ref-substances@phytolab.de Shop: https://phyproof.phytolab.com



Supplements

4',5,7-Trihydroxy 3,3',6,8-tetramethoxyflavone
Product # 82674

Batch # 5286

Identity tests:

IR spectrum

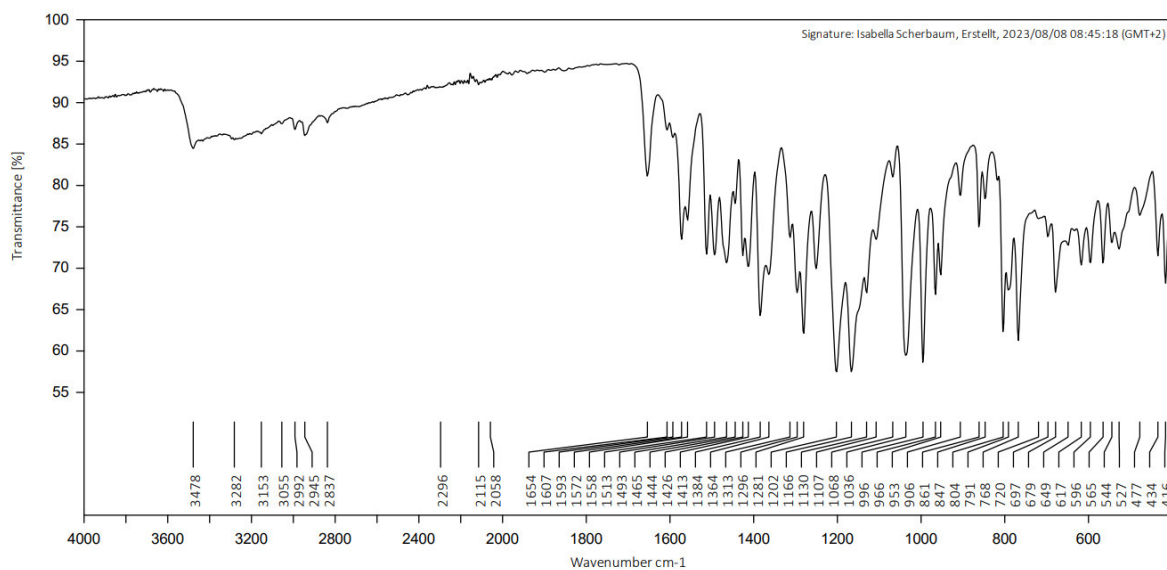


PhytoLab

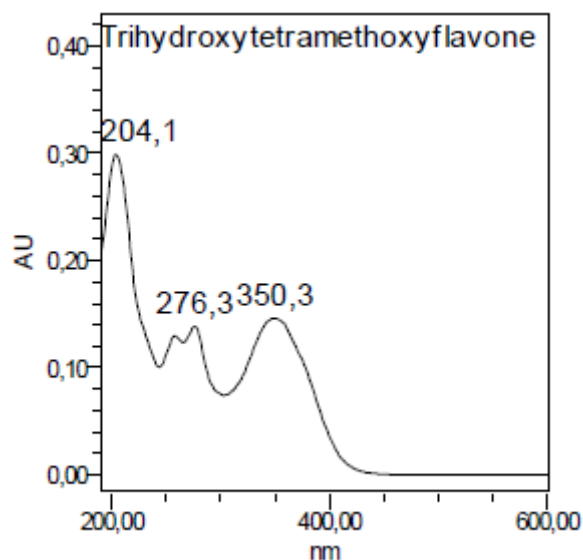
SAFEGUARDING BOTANICAL QUALITY

82674_4,5,7-Trihydroxy-3,3,6,8-tetramethoxyflavon_5286

Signature: Isabella Scherbaum, Erstellt, 2023/08/08 08:45:18 (GMT+2)



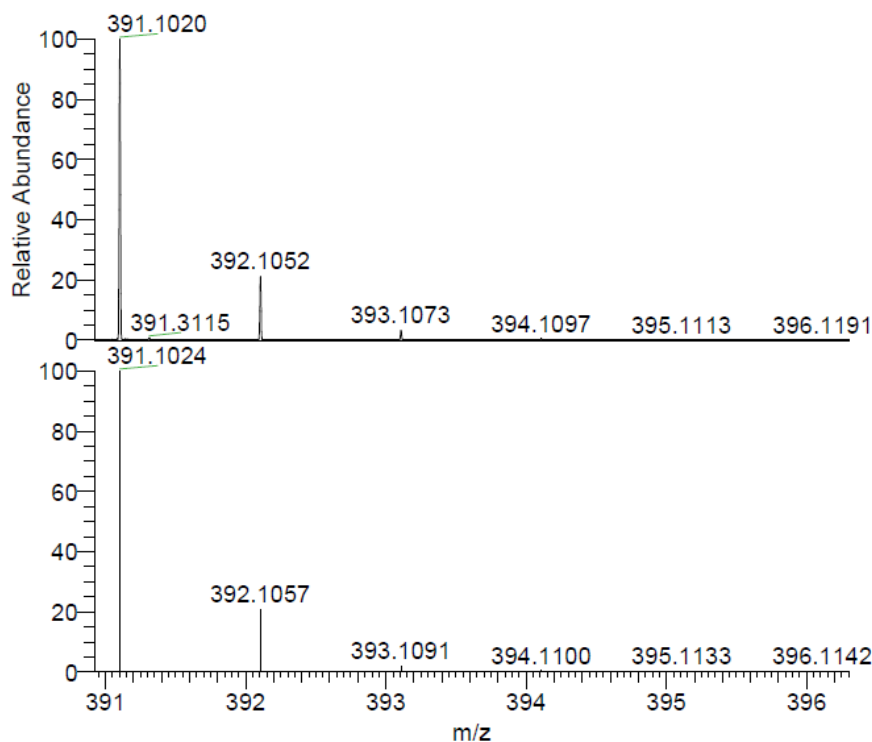
UV spectrum (derived from HPLC/PDA)





MS spectrum (ESI)

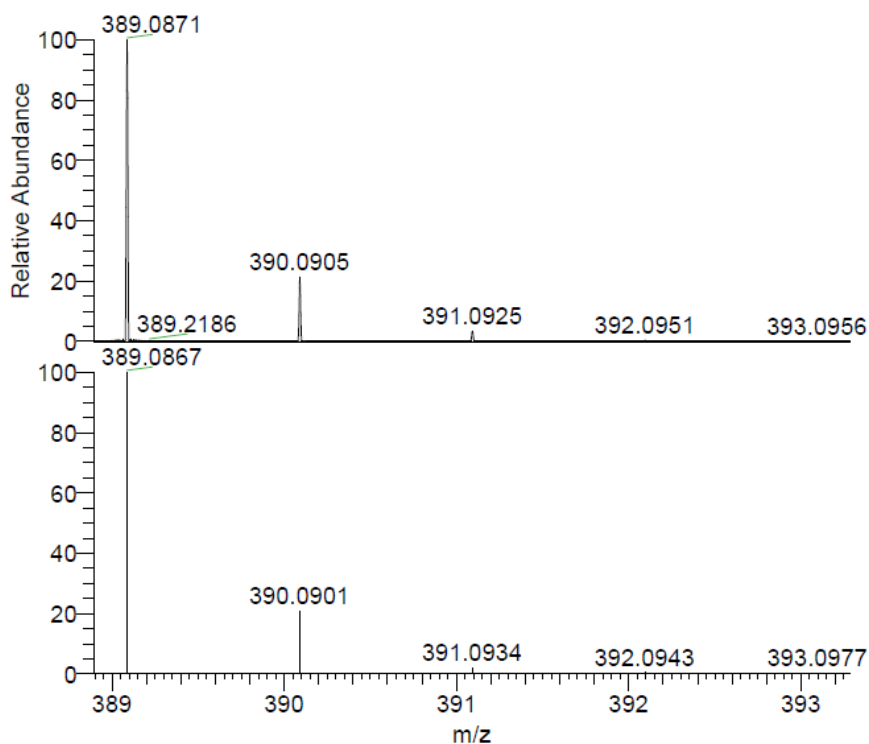
Detection: positive mode (compared with predicted spectrum)



NL:
1.13E7
201008_004#1074 RT: 11.61
AV: 1 SB: 159 7.72-8.47 ,
10.12-11.16 T: FTMS {1,1} +
p ESI Full ms
[100.00-1500.00]

NL:
7.96E5
C₁₉H₁₉O₉:
C₁₉H₁₉O₉
pa Chrg 1

Detection: negative mode (compared with predicted spectrum)



NL:
2.17E6
201008_005#1016 RT: 11.58
AV: 1 SB: 158 7.73-8.47 ,
10.12-11.16 T: FTMS {1,1} - p
ESI Full ms [100.00-1500.00]

NL:
7.96E5
C₁₉H₁₇O₉:
C₁₉H₁₇O₉
pa Chrg 1

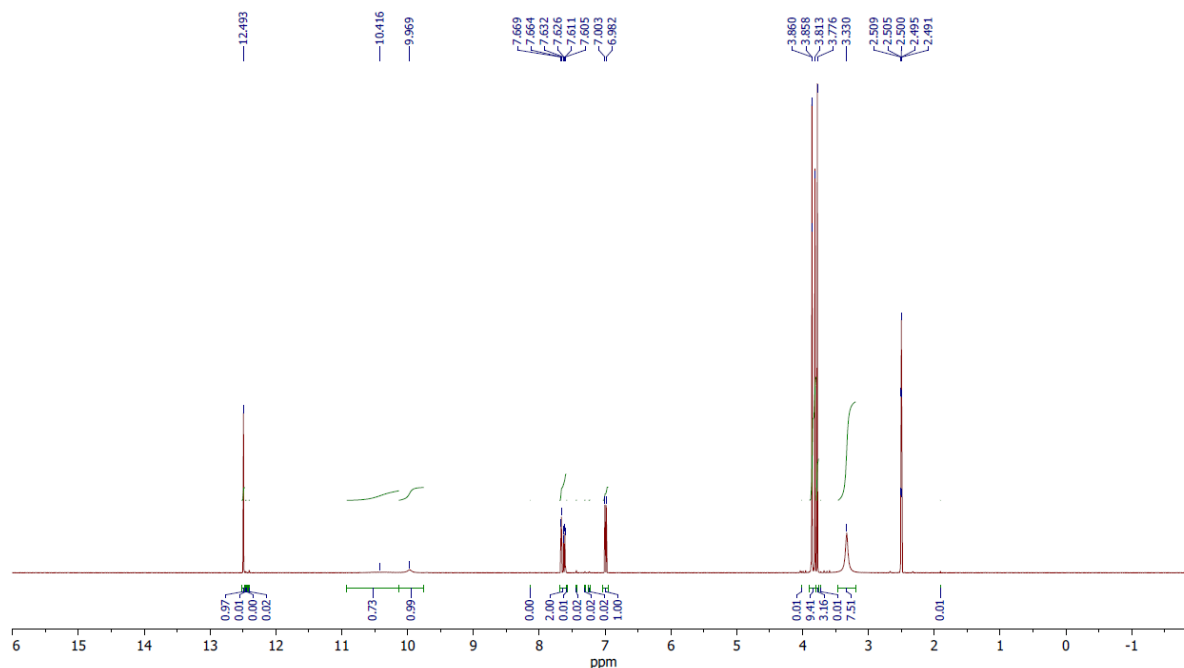


NMR spectra

¹H-NMR

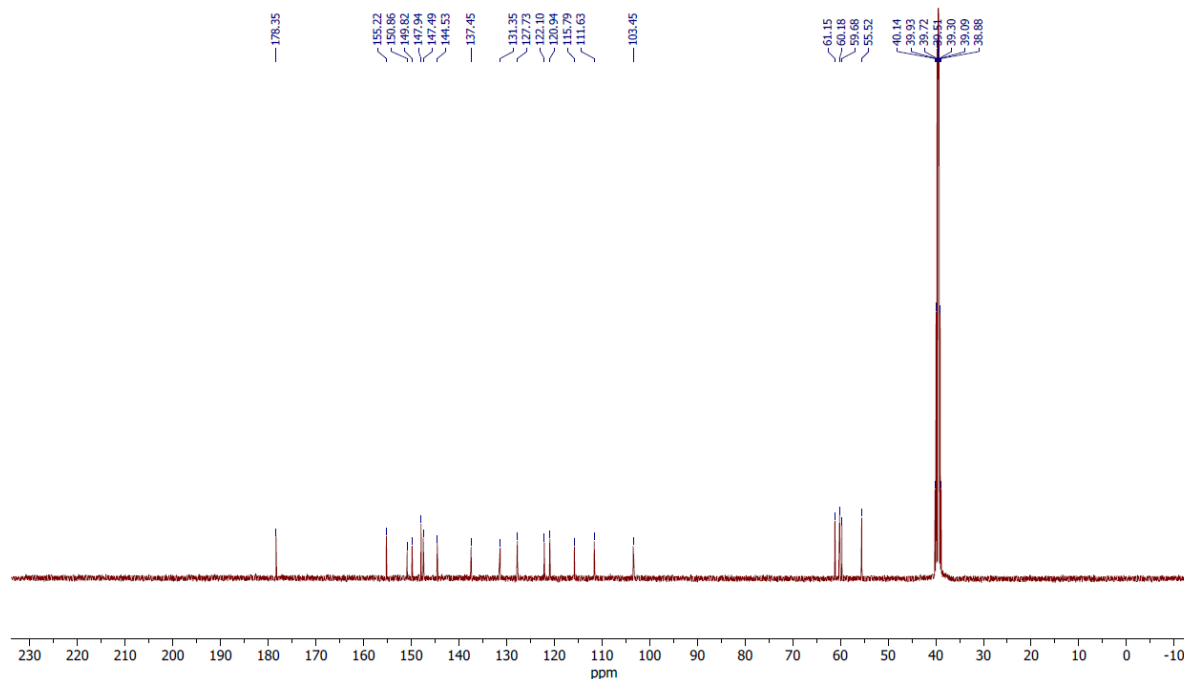
PhytoLab GmbH & Co. KG
4',5,7-Trihydroxy-3,3',6,8-tetramethoxyflavon, Charge: 5286
10.2 mg ad 0.75 ml DMSO-d₆

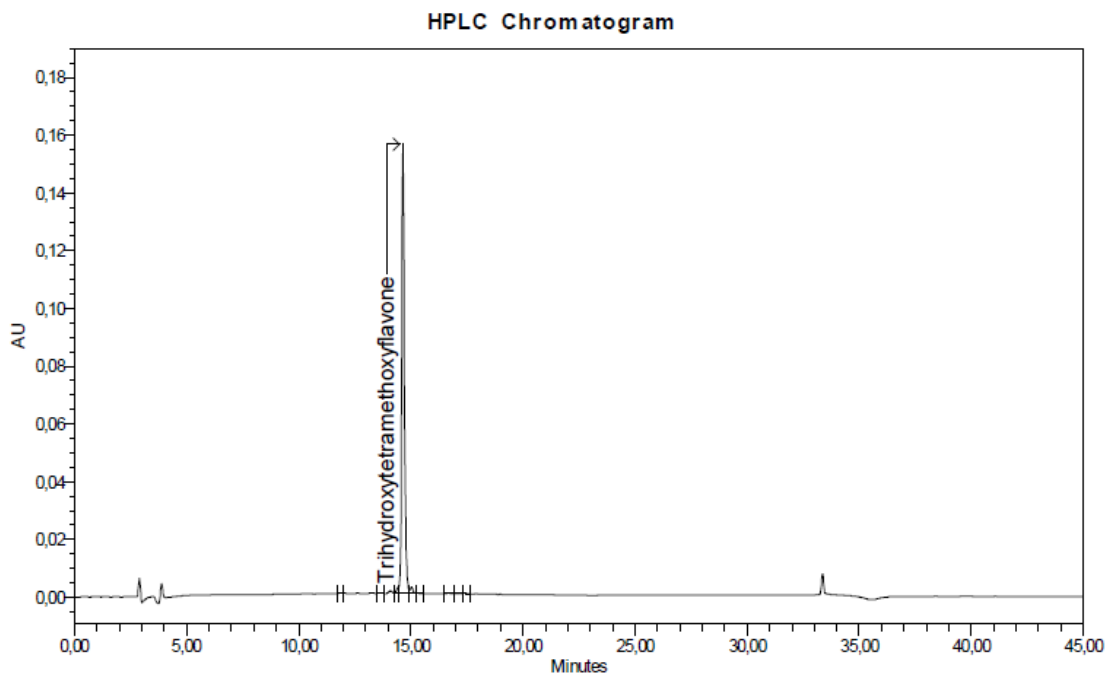
400 MHz ¹H-NMR, Agilent MR400

¹³C-NMR

PhytoLab GmbH & Co. KG
4',5,7-Trihydroxy-3,3',6,8-tetramethoxyflavon, Charge: 5286
10.2 mg ad 0.75 ml DMSO-d₆

100 MHz ¹³C-NMR, Agilent MR400



Chromatographic purity:**Peak Results**

	Name	RT	Area	% Area
1		11,875	1615	0,12
2		13,627	2359	0,17
3		14,450	12189	0,89
4	Trihydroxytetramethoxyflavone	14,649	1328817	96,75
5		15,030	19673	1,43

	Name	RT	Area	% Area
6		15,223	2243	0,16
7		16,692	2410	0,18
8		17,226	1827	0,13
9		17,402	2340	0,17
Sum				100,00

Analytical conditions

Column: Luna C18, 250 x 4.6 mm, 5 µm
Mobile Phase: eluent A: H₂O pH 3.0 (H₃PO₄)
 eluent B: CH₃CN
Mode: gradient

Time [min]	Eluent A [%]	Eluent B [%]
0	90	10
20	10	90
30	10	90
31	90	10
45	90	10

Flow: 1.0 ml/min
Injection Volume: 10 µl
Column Temperature: 23 °C
Sample concentration: approx. 5.1 mg/100 ml
Sample preparation: dissolved in CH₃OH
Detection: UV, 350 nm
Special note: -

Please note: Values on the certificate of analysis may vary as these are average values of at least six injections while above chromatogram and report is only one example. Non-integrated peaks originate from the blank injection.