Optical Reference Materials for UV/Vis Spectroscopy

PRODUCT CATALOG WITH USAGE GUIDELINES

GLASS FILTERS
LIQUID FILTERS
REFERENCE PLATES

DAkkS
Deutsche Akkreditierungsstelle
D-K-18752-01-00

NIST TRACEABLE

MADE IN GERMANY

Hellma®

YOUR PROCESS. OUR SOLUTIONS.

As a leading supplier of high-precision, optical solutions that are 'Made in Germany' from glass, quartz glass and synthetic crystals, Hellma has been a by-word for outstanding quality for over 95 years. A key supplier, the company is an integral part of its clients' value chains. Reliability, trust and continuity are inherent in Hellma's work, and the company believes it has both a duty and responsibility to ensure these principles are upheld. Clients from more than 40 countries world-wide put their confidence in Hellma's exceptional level of performance and problem-solving skills to meet and exceed regulatory requirements and to make their products safer.

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Optical components and assemblies used in devices and systems in analytical technology.

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Hellma Optics

Premium-quality precision optics for use in laser technology as well as all areas of photonics and the optical industry.

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NIST, PTB
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DAkkS
accredited calibration laboratory

DAkkS Deutsche Akkreditierungsstelle D-K-18752-01-00

ACCREDITED ACCORDING TO
DIN EN ISO / IEC 17025

30 YEAR GUARANTEE
with regular recalibration

COMPLIANT
with the most important Pharmacopeia

Benefit from more than 95 years of experience.

EXPERTS TIP:

Where you see this icon you will find useful information to make your processes safer and more effective.

CONTENTS

1. INTRODUCTION

Dear Readers,

Although checking measuring equipment to ensure that results are accurate has long been common practice for analytical balances, it still tends to take something of a backseat where spectrophotometers are concerned. Spectrophotometers are important instruments that play a major role in health care, the life sciences, environmental analysis and processes such as production control and ensuring product quality. Over the last two years, many laboratories have become considerably more aware of the need to check their spectrophotometers, **making it all the more important to know that these precision tools are also subject to mandatory checks under DIN EN ISO 9001**. The standard clearly stipulates that measuring equipment be calibrated or verified, either at regular intervals or before use, using measurement standards that can be traced back to international or national standards. For an overview of the measurement standards for UV-Vis spectrophotometers, please refer to our reference materials in this brochure. An increasing number of laboratories are turning to this easy method for ensuring high standards of work – not only to satisfy requirements in time for their next audit, but also to be safe in the knowledge that they are taking accurate measurements and thus basing their actions and responses on correct results. We are delighted that our products are helping to achieve this. Details of our product range, as well as usage guidelines, helpful tips, and recommendations, are all included in this Handbook. **WE VERY MUCH HOPE THAT YOU ENJOY READING IT.**

FOR INFORMATION
Proven reliability, completely documented. The Hellma Analytics calibration laboratory is the only calibration laboratory in Germany accredited for the certification of UV/Vis reference materials.

1.1 Hellma Analytics calibration laboratory: accredited to DIN EN ISO 17025

Our lab is a DAkkS calibration laboratory and is accredited to DIN EN ISO 17025, a comprehensive quality management system that acts as a seamless continuation of other systems such as ISO 9001. By achieving this accreditation, we have demonstrated proof of expertise in the calibration activities that we perform and are authorized to issue internationally recognized DAkkS calibration certificates. The accreditation is the key to high quality measurements, international comparability, and trust in both the work of the calibration laboratory and the transparency of results.

DAkkS Deutsche Akkreditierungsstelle D-K-18752-01-00

NIST TRACEABLE

DIN EN ISO 17025

With the certified UV/Vis reference materials from Hellma Analytics, we create the basis for reliable measurements for our customers.

Birgit Kehl,
Compliance Representative
Calibration Laboratory

1. INTRODUCTION

1.2 Certified test equipment

Quality assurance and quality control regulations, such as ISO 9001, GLP, GMP, and Pharmacopeias, require companies to verify the consistently excellent performance of any spectrophotometer in use. The two most important factors for obtaining precise spectrophotometer data are the photometric accuracy (absorbance accuracy) and wavelength accuracy of the spectrophotometer, which should be tested on a regular basis.

In the Hellma Analytics calibration laboratory, which is accredited to DIN EN ISO 17025, we manufacture certified reference materials based on the regulatory codes issued by NIST (National Institute of Standards and Technology), ASTM (American Society for Testing and Materials) and Pharmacopeias (Ph. Eur., USP). All certified measurement results can be traced back to NIST (photometric accuracy) or to PTB (Physikalisch-Technische Bundesanstalt) (wavelength accuracy) standard reference materials. (Photometric accuracy: NIST SRM® 930e, NIST SRM® 1930. Hellma 666S300; wavelength accuracy: Hellma 667005)

FOR INFORMATION

Hellma Analytics' DAkkS-certified reference materials comply with the requirements stipulated by quality management systems and Pharmacopeias, meeting the highest quality requirements and ensuring the international comparability of measurement results.

Choose between glass filters and liquid filters for your reference materials:

1.3 Glass filter applications

666 at the beginning of the article number identifies our glass filters. Glass filters are certified reference materials made of glass manufactured specifically for calibration. They are, above all, extremely robust. All glass filters certified by Hellma Analytics are traceable to NIST primary standards. Certified glass filters are suitable for checking the following parameters of your spectrophotometer:

- Wavelength accuracy
- Photometric accuracy (absorbance)

1.4 Liquid filter applications

667 at the beginning of the article number identifies our liquid filters. Liquid filters are certified liquid reference materials that are manufactured in compliance with Pharmacopeias and/or NIST standards and filled into quartz glass cuvettes under controlled conditions. The cuvettes are then permanently sealed to become airtight. Liquid filters have the distinct advantage of equating to real measuring conditions. Certified Hellma Analytics liquid filters are suitable for checking the following parameters of your spectrophotometer:

- Wavelength accuracy
- Photometric accuracy (absorbance)
- Stray light levels
- Spectral resolution

You should regularly check your UV/Vis spectrophotometer ter for all of these parameters, especially photometric and wavelength accuracy, while observing the relevant requirements in your device handbook. Thanks to their ease of use and long service life, certified Hellma Analytics reference materials provide an excellent aid for all routine checks.

HELLMA ANALYTICS REFERENCE MATERIALS IN ACCORDANCE WITH THE MOST IMPORTANT REGULATIONS

MATERIAL	CHECKING OF	RANGE	PH.EUR 8.0	USP <857>	ASTM
GLASS FILTERS					
Holmium glass	Wavelength accuracy	UV/Vis		×	×
Didymium glass	Wavelength accuracy	UV/Vis		×	×
Neutral density glass	Photometric accuracy	Vis		×	×
LIQUID FILTERS					
Holmium (solution)	Wavelength accuracy	UV/Vis	×	×	×
Didymium (solution)	Wavelength accuracy	UV/Vis		×	
Potassium dichromate (solution)	Photometric accuracy	UV/Vis	×	×	×
Toluene in hexane (solution)	Spectral resolution	UV	×	×	
Potassium chloride (solution)	Stray light	UV	×	×	×
Sodium iodide (solution)	Stray light	UV		×	×
Sodium nitrite (solution)	Stray light	UV		×	×
Acetone	Stray light	UV		×	×

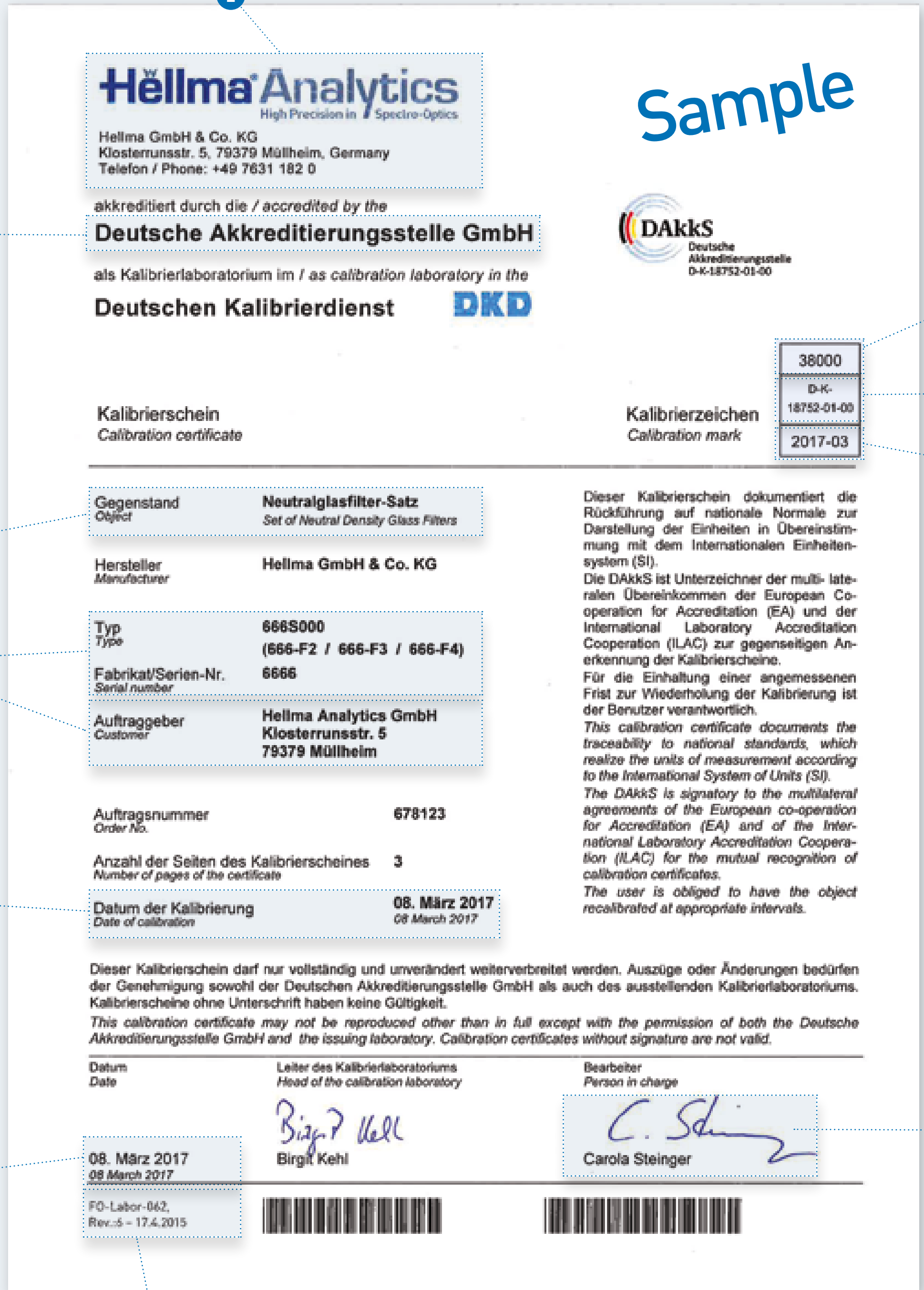
1. INTRODUCTION

1.5 DAkkS calibration certificate

After careful production, reference materials are certified in the Hellma Analytics calibration laboratory (accredited to DIN EN ISO 17025) using a high-performance UV-Vis/NIR spectrophotometer. Reference materials are only considered to be certified if they have been issued with a DAkkS calibration certificate and bear a calibration mark. Using the measurement values documented and certified in the calibration certificate, users can check and calibrate their spectrophotometers accordingly.

IMPORTANT INFORMATION

Only if the DAkkS calibration certificate has been issued and the calibration mark has been affixed, do the reference materials actually become certified reference materials.



Hellma Analytics
High Precision in Spectro-Optics
Hellma GmbH & Co. KG
Klosterrunsstr. 5, 79379 Müllheim, Germany
Telefon / Phone: +49 7631 182 0

Sample

akkreditiert durch die / *accredited by the*
Deutsche Akkreditierungsstelle GmbH
als Kalibrierlaboratorium im / *as calibration laboratory in the*
Deutschen Kalibrierdienst DKD

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Kalibrierschein
Calibration certificate

Kalibrierzeichen
Calibration mark

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Gegenstand *Object*	**Neutralglasfilter-Satz** *Set of Neutral Density Glass Filters*
Hersteller *Manufacturer*	**Hellma GmbH & Co. KG**
Typ *Type*	**666S000 (666-F2 / 666-F3 / 666-F4)**
Fabrikat/Serien-Nr. *Serial number*	**6666**
Auftraggeber *Customer*	**Hellma Analytics GmbH Klosterrunsstr. 5 79379 Müllheim**
Auftragsnummer *Order No.*	**678123**
Anzahl der Seiten des Kalibrierscheines *Number of pages of the certificate*	**3**
Datum der Kalibrierung *Date of calibration*	**08. März 2017** *08 March 2017*

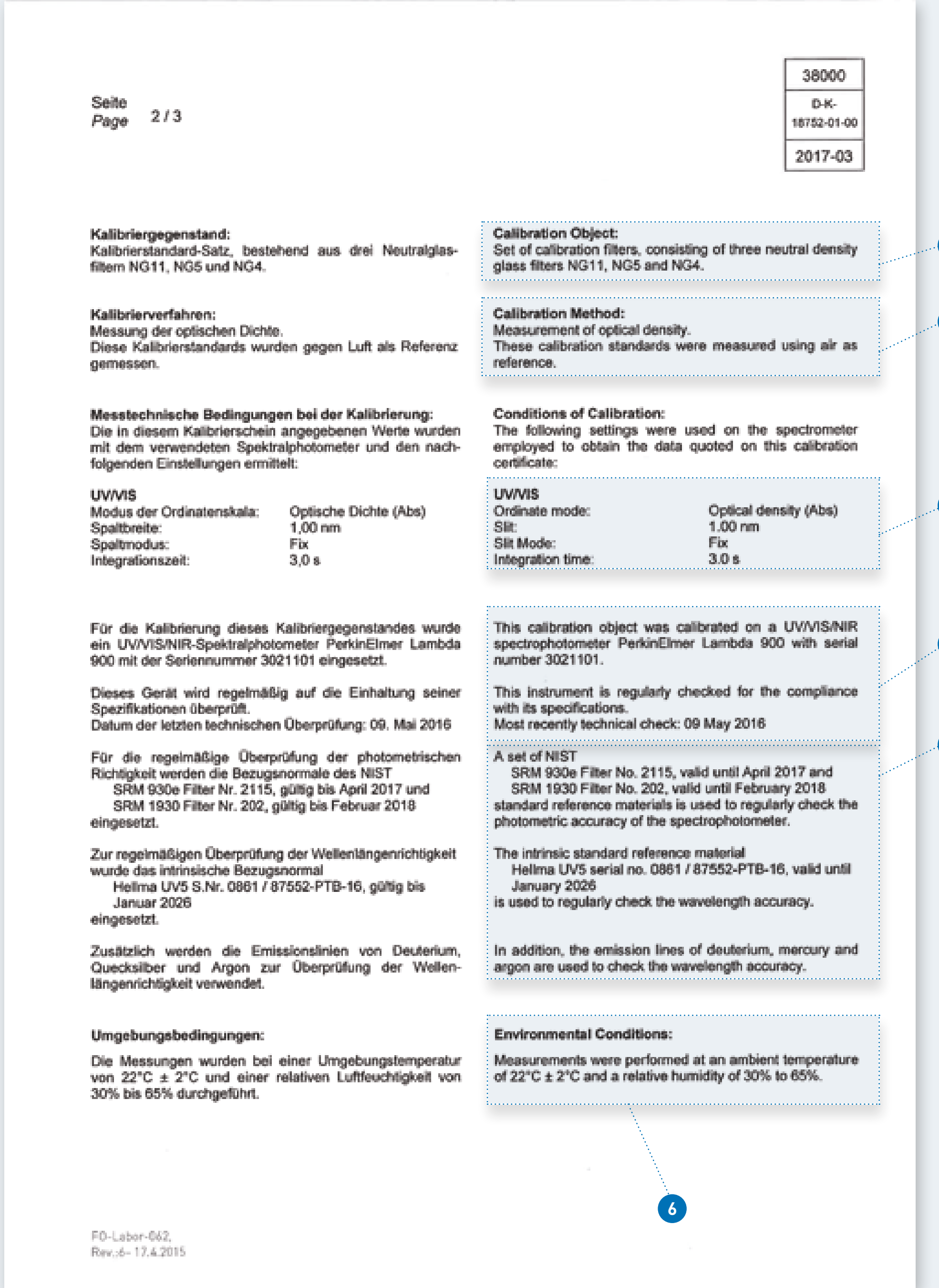
Dieser Kalibrierschein dokumentiert die Rückführung auf nationale Normale zur Darstellung der Einheiten in Übereinstimmung mit dem Internationalen Einheitensystem (SI).
Die DAkkS ist Unterzeichner der multilateralen Übereinkommen der European Co-operation for Accreditation (EA) und der International Laboratory Accreditation Cooperation (ILAC) zur gegenseitigen Anerkennung der Kalibrierscheine.
Für die Einhaltung einer angemessenen Frist zur Wiederholung der Kalibrierung ist der Benutzer verantwortlich.
This calibration certificate documents the traceability to national standards, which realize the units of measurement according to the International System of Units (SI).
The DAkkS is signatory to the multilateral agreements of the European co-operation for Accreditation (EA) and of the International Laboratory Accreditation Cooperation (ILAC) for the mutual recognition of calibration certificates.
The user is obliged to have the object recalibrated at appropriate intervals.

Dieser Kalibrierschein darf nur vollständig und unverändert weiterverbreitet werden. Auszüge oder Änderungen bedürfen der Genehmigung sowohl der Deutschen Akkreditierungsstelle GmbH als auch des ausstellenden Kalibrierlaboratoriums. Kalibrierscheine ohne Unterschrift haben keine Gültigkeit.
This calibration certificate may not be reproduced other than in full except with the permission of both the Deutsche Akkreditierungsstelle GmbH and the issuing laboratory. Calibration certificates without signature are not valid.

Datum *Date*	Leiter des Kalibrierlaboratoriums *Head of the calibration laboratory*	Bearbeiter *Person in charge*
08. März 2017 *08 March 2017*	Birgit Kehl	Carola Steinger

FO-Labor-062,
Rev.:6 – 17.4.2015

1. Name of accreditation body
2. Name and address of issuing laboratory
3. Description of calibrated item
4. Clear details of measured item
5. Customer
6. Date of measurement
7. Internal document reference, including date of issue and last modified date
8. Date of issue of calibration certificate
9. Calibration certificate serial numbers
10. DAkkS registration number
11. Year and month of calibration
12. Authorized persons



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Kalibriergegenstand:
Kalibrierstandard-Satz, bestehend aus drei Neutralglasfiltern NG11, NG5 und NG4.

Calibration Object:
Set of calibration filters, consisting of three neutral density glass filters NG11, NG5 and NG4.

Kalibrierverfahren:
Messung der optischen Dichte.
Diese Kalibrierstandards wurden gegen Luft als Referenz gemessen.

Calibration Method:
Measurement of optical density.
These calibration standards were measured using air as reference.

Messtechnische Bedingungen bei der Kalibrierung:
Die in diesem Kalibrierschein angegebenen Werte wurden mit dem verwendeten Spektralphotometer und den nachfolgenden Einstellungen ermittelt:

Conditions of Calibration:
The following settings were used on the spectrometer employed to obtain the data quoted on this calibration certificate:

UV/VIS	
Modus der Ordinatenskala:	Optische Dichte (Abs)
Spaltbreite:	1,00 nm
Spaltmodus:	Fix
Integrationszeit:	3,0 s

UV/VIS	
Ordinate mode:	Optical density (Abs)
Slit:	1.00 nm
Slit Mode:	Fix
Integration time:	3.0 s

Für die Kalibrierung dieses Kalibriergegenstandes wurde ein UV/VIS/NIR-Spektralphotometer PerkinElmer Lambda 900 mit der Seriennummer 3021101 eingesetzt.

Dieses Gerät wird regelmäßig auf die Einhaltung seiner Spezifikationen überprüft.
Datum der letzten technischen Überprüfung: 09. Mai 2016

Für die regelmäßige Überprüfung der photometrischen Richtigkeit werden die Bezugsnormale des NIST
SRM 930e Filter Nr. 2115, gültig bis April 2017 und
SRM 1930 Filter Nr. 202, gültig bis Februar 2018
eingesetzt.

Zur regelmäßigen Überprüfung der Wellenlängenrichtigkeit wurde das intrinsische Bezugsnormal
Hellma UV5 S.Nr. 0861 / 87552-PTB-16, gültig bis Januar 2026
eingesetzt.

Zusätzlich werden die Emissionslinien von Deuterium, Quecksilber und Argon zur Überprüfung der Wellenlängenrichtigkeit verwendet.

This calibration object was calibrated on a UV/VIS/NIR spectrophotometer PerkinElmer Lambda 900 with serial number 3021101.

This instrument is regularly checked for the compliance with its specifications.
Most recently technical check: 09 May 2016

A set of NIST
SRM 930e Filter No. 2115, valid until April 2017 and
SRM 1930 Filter No. 202, valid until February 2018
standard reference materials is used to regularly check the photometric accuracy of the spectrophotometer.

The intrinsic standard reference material
Hellma UV5 serial no. 0861 / 87552-PTB-16, valid until January 2026
is used to regularly check the wavelength accuracy.

In addition, the emission lines of deuterium, mercury and argon are used to check the wavelength accuracy.

Umgebungsbedingungen:
Die Messungen wurden bei einer Umgebungstemperatur von 22°C ± 2°C und einer relativen Luftfeuchtigkeit von 30% bis 65% durchgeführt.

Environmental Conditions:
Measurements were performed at an ambient temperature of 22°C ± 2°C and a relative humidity of 30% to 65%.

FO-Labor-062,
Rev.:6 – 17.4.2015

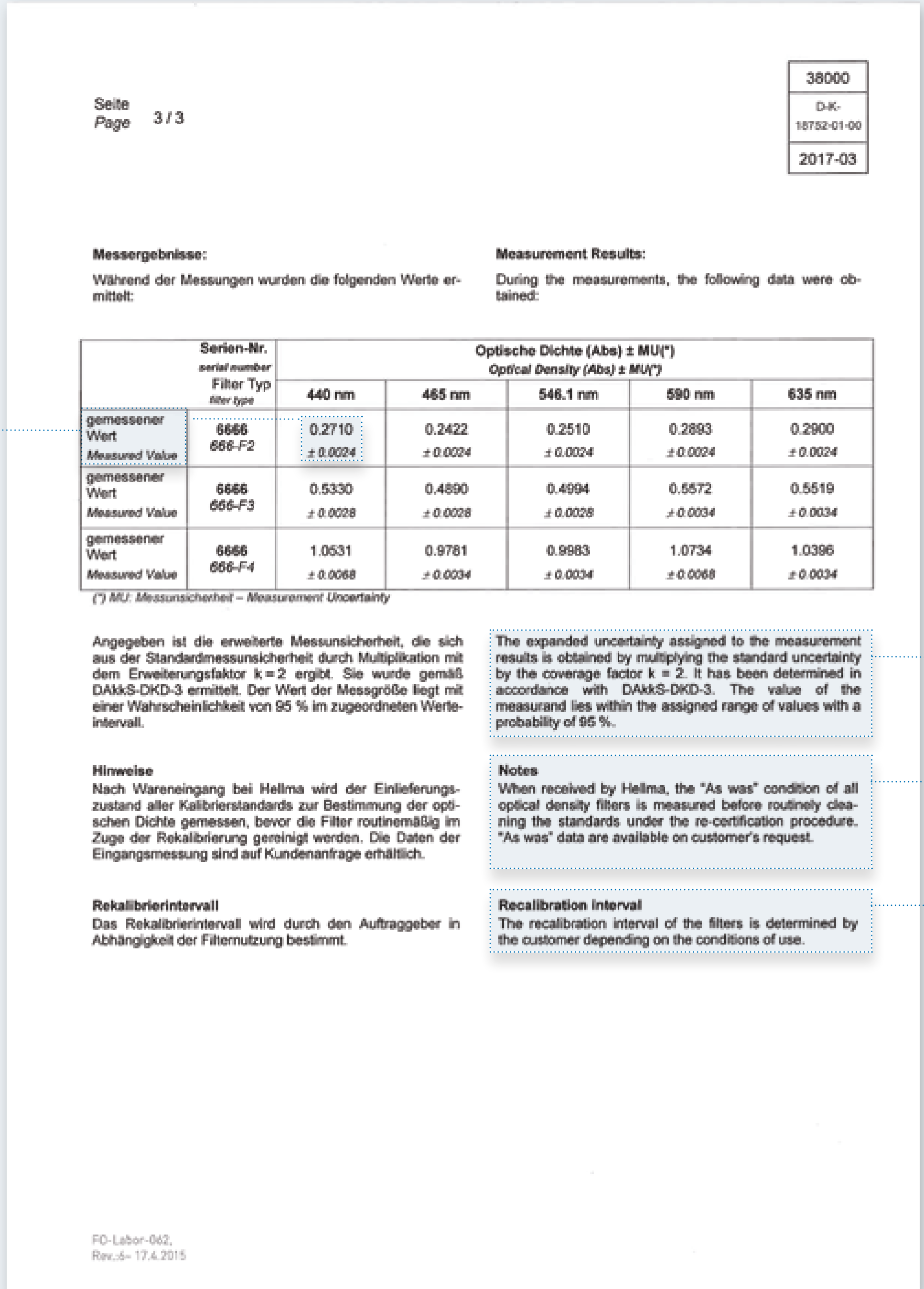
DAkkS Deutsche Akkreditierungsstelle D-K-18752-01-00

1. Description of calibrated item
2. Naming the reference
3. Measurement conditions (device settings)
4. Type of device used to carry out measurements
5. Type, serial number and validity of calibrated NIST/PTB reference standards used to regularly check reference photometers; details of additional checking methods
6. Ambient conditions during measurement

1. INTRODUCTION

1.5 DAkkS calibration certificate

1. Measurement value and smallest attributed measurement uncertainty that can be specified. This value only refers to Hellma Analytics measurements and applies solely to the company's specific measurement conditions. In justified cases, calibration certificates may also show measurement results that do not fall within the calibration laboratory's scope of accreditation. These must be clearly labeled as such on the calibration certificate.
2. Notes on determining expanded measurement uncertainty.
3. Notes on initial measurements of filters used to determine optical density. Initial measurements are not taken for filters used to determine wavelength accuracy.
4. Calibration certificates shall not contain recommended recertification intervals (in accordance with DAkkS-DKD-5). Exceptions may be made if requested by the customer or required by legislation.



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Messergebnisse:
Während der Messungen wurden die folgenden Werte ermittelt:

Measurement Results:
During the measurements, the following data were obtained:

	Serien-Nr. *serial number* Filter Typ *filter type*	Optische Dichte (Abs) ± MU(*) *Optical Density (Abs) ± MU(*)*				
		440 nm	465 nm	546.1 nm	590 nm	635 nm
gemessener Wert *Measured Value*	6666 *666-F2*	0.2710 *± 0.0024*	0.2422 *± 0.0024*	0.2510 *± 0.0024*	0.2893 *± 0.0024*	0.2900 *± 0.0024*
gemessener Wert *Measured Value*	6666 *666-F3*	0.5330 *± 0.0028*	0.4890 *± 0.0028*	0.4994 *± 0.0028*	0.5572 *± 0.0034*	0.5519 *± 0.0034*
gemessener Wert *Measured Value*	6666 *666-F4*	1.0531 *± 0.0068*	0.9781 *± 0.0034*	0.9983 *± 0.0034*	1.0734 *± 0.0068*	1.0396 *± 0.0034*

(*) MU: Messunsicherheit – Measurement Uncertainty

Angegeben ist die erweiterte Messunsicherheit, die sich aus der Standardmessunsicherheit durch Multiplikation mit dem Erweiterungsfaktor k = 2 ergibt. Sie wurde gemäß DAkkS-DKD-3 ermittelt. Der Wert der Messgröße liegt mit einer Wahrscheinlichkeit von 95 % im zugeordneten Werteintervall.

The expanded uncertainty assigned to the measurement results is obtained by multiplying the standard uncertainty by the coverage factor k = 2. It has been determined in accordance with DAkkS-DKD-3. The value of the measurand lies within the assigned range of values with a probability of 95 %.

Hinweise
Nach Wareneingang bei Hellma wird der Einlieferungszustand aller Kalibrierstandards zur Bestimmung der optischen Dichte gemessen, bevor die Filter routinemäßig im Zuge der Rekalibrierung gereinigt werden. Die Daten der Eingangsmessung sind auf Kundenanfrage erhältlich.

Notes
When received by Hellma, the "As was" condition of all optical density filters is measured before routinely cleaning the standards under the re-certification procedure. "As was" data are available on customer's request.

Rekalibrierintervall
Das Rekalibrierintervall wird durch den Auftraggeber in Abhängigkeit der Filternutzung bestimmt.

Recalibration Interval
The recalibration interval of the filters is determined by the customer depending on the conditions of use.

FO-Labor-062,
Rev.:6– 17.4.2015

1.6 Warranty
30-year manufacturer's warranty on all Hellma Analytics reference materials

We're confident of our quality and you can be confident of reliable measurement results!

All **Hellma Analytics reference materials** come with a **30-year warranty**, provided that they are regularly recertified – every two years – at the Hellma Analytics calibration laboratory. Certified reference materials sent for recertification are carefully cleaned and recertified before being sent back with a new DAkkS calibration certificate and calibration mark. Damaged filters and filters that deviate significantly from nominal values are usually replaced in consultation with the customer.

Details on recertification can be found on page 18 and 19.

All of our reference materials come with a 30-year warranty, provided that they are regularly recertified (at least every two years) at the Hellma Analytics calibration laboratory.

2. GLASS FILTERS

WAVELENGTH ACCURACY

2.1 Checking wavelength accuracy

APPLICATION

To measure wavelength accuracy, the filter absorbs the light beam of the spectrophotometer to a greater extent at certain wavelengths creating absorbance peaks. Ideally, any reference materials used to determine wavelength accuracy should have narrow, well-defined peaks at a variety of wavelengths in the UV and visible range.

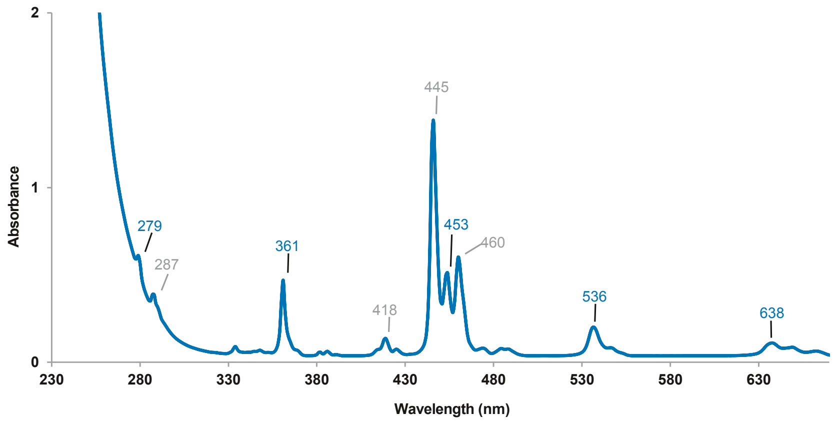
NOTE

The positions of holmium peaks may vary slightly depending on the glass batch used. This is why Hellma Analytics certifies each holmium glass filter individually.

2.1.1 Holmium glass filter

PRODUCT DESCRIPTION

The holmium glass filter 666-F1 has a range of narrow, well-defined peaks in the UV and visible range, making holmium an excellent choice for checking the wavelength scale of spectrophotometers. In comparison to filters that use holmium solution, the holmium glass filter has a somewhat weaker spectrum with fewer peaks. In the low UV range in particular, the absorbance behavior of the glass matrix is superimposed on the holmium peaks. The main advantage of using a glass filter over a liquid filter is that it is more robust.



Typical spectrum of a holmium glass filter

ARTICLE NO.	666F1-339
APPLICATION	Testing the wavelength accuracy in the UV and Vis range (279 nm to 638 nm) at a spectral bandwidth up to 2 nm.
CONTENT	Holmium glass filter with metal frame
STANDARD CERTIFICATION	**Wavelengths:** 279; 361; 453; 536; 638 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Wavelengths:** 279; 287; 361; 418; 445; 453; 460; 536; 638 nm **Slit width:** all possible up to 2 nm

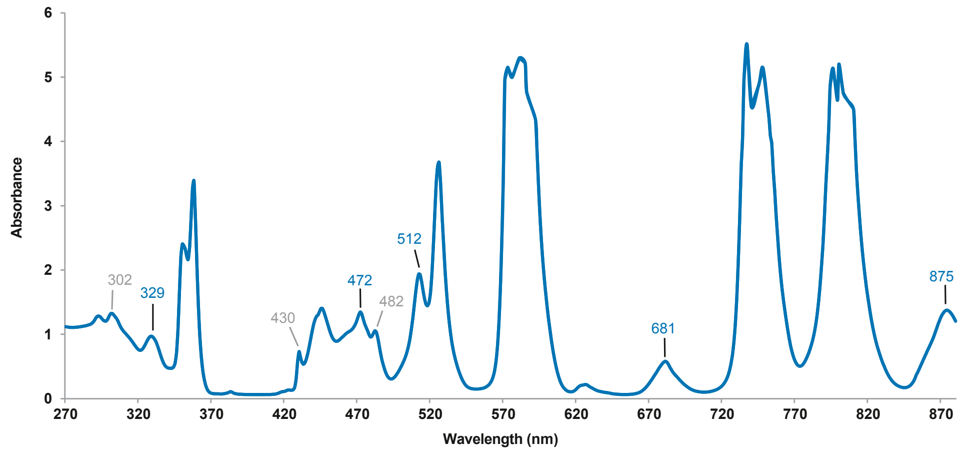
2.1.2 Didymium glass filter

PRODUCT DESCRIPTION

The didymium glass filter 666-F7W is made from material specially manufactured by Schott AG. Like holmium glass, didymium glass has a variety of characteristic peaks in the ultraviolet and visible range and is therefore typically used for checking wavelength accuracy. Its peaks are not as narrow as those of holmium glass filters, however. The filter's absorbance behavior in the ultraviolet range also makes it suitable as an absorbance filter for checking photometric accuracy (see page 16, 666-F7A).

NOTE

The positions of didymium glass peaks may vary slightly depending on the glass batch used. This is why Hellma Analytics certifies each didymium glass filter individually.



Typical spectrum of a didymium glass filter

ARTICLE NO.	666F7W-323 or 666F7-323
APPLICATION	Testing the wavelength accuracy in the UV and Vis range (329 nm to 875 nm) at a spectral bandwidth up to 2 nm.
CONTENT	Didymium glass filter with metal frame
STANDARD CERTIFICATION	**Wavelengths:** 329; 472; 512; 681; 875 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Wavelengths:** 302; 329; 430; 472; 482; 512; 681; 875 nm **Slit width:** all possible up to 2 nm

2. GLASS FILTERS

PHOTOMETRIC ACCURACY

2.2 Checking the photometric accuracy

APPLICATION

To measure photometric accuracy (absorbance), the filter reduces the light beam from the spectrophotometer. An absorbance value (Abs) can be deduced from the light extinction caused by the filter.

2.2.1 Didymium glass filter F7A

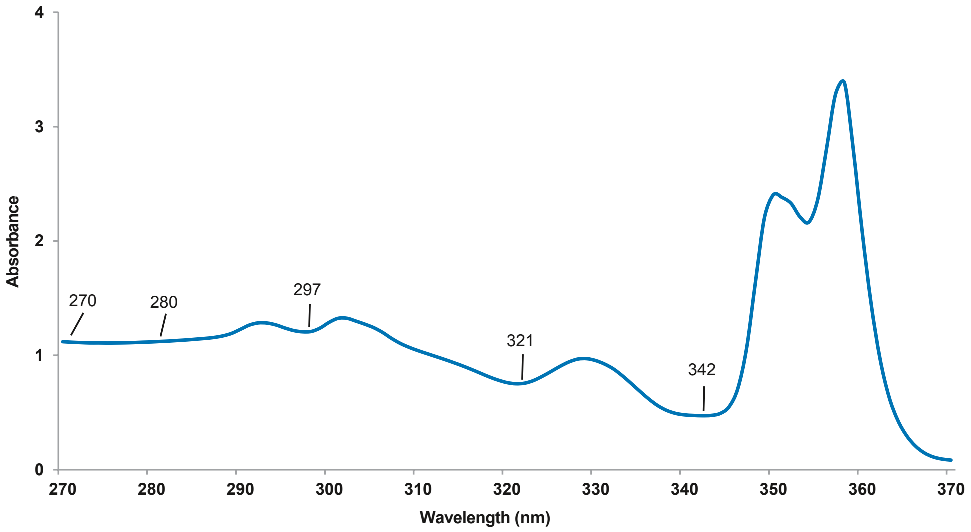
PRODUCT DESCRIPTION

Didymium glass filter 666-F7A is made from material specially manufactured by Schott AG. The didymium glass filter's absorbance behavior in the ultraviolet range also makes it suitable for use as an absorbance filter. Didymium glass filters are therefore suitable for checking wavelength accuracy in the UV-Vis range as well as checking photometric accuracy in the UV range.

Absorbance behavior in the UV range can be checked at 270 nm, 280 nm, 297 nm, 321 nm, and 342 nm. Filters are routinely set at a thickness that produces a nominal optical density of 0.5 Abs at 342 nm. This results in increasingly larger absorbances the shorter the wavelengths become.

NOTE

These absorbance values vary greatly depending on the glass batch used and can only be compared for filters derived from the same glass melting process. This is why all didymium glass filters are certified individually.



Selected spectrum for a didymium glass filter between 270 nm and 370 nm

ARTICLE NO.	666F7A-323 or 666F7-323
APPLICATION	Testing the wavelength accuracy in the UV and Vis range(270 nm to 340 nm)
CONTENT	Didymium glass filter with metal frame
STANDARD CERTIFICATION	**Photometric accuracy:** approx. 0.5 to 1 Abs. **Wavelengths:** 270; 280; 297; 321; 342 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Additional possible wavelengths:** from 270 to 280 nm **Slit width:** all possible up to 3 nm

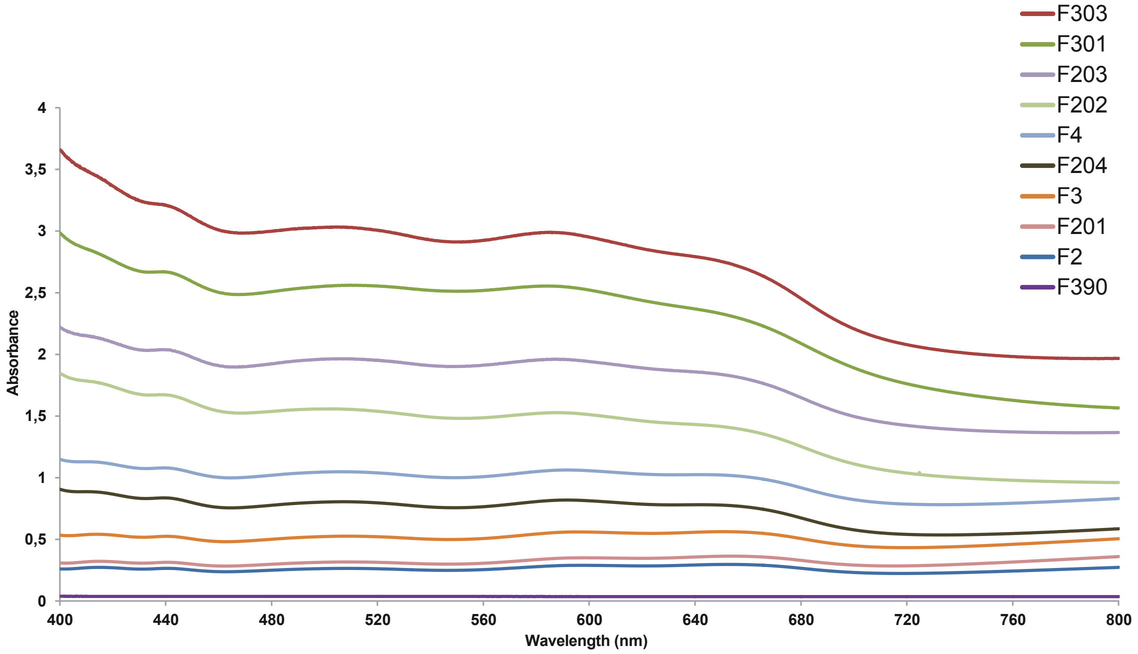
2.2.2 Neutral density glass filter

PRODUCT DESCRIPTION

Hellma Analytics neutral density glass filters are made from filter materials produced by Schott AG, which were selected on account of their homogeneity and stability. Thanks to a relatively constant transmittance within the wavelength range of 405 nm to 800 nm, they have been used to check photometric accuracy and linearity in the visible wavelength range (> 405 nm) for decades.

NOTE

If you have several Neutral density glass filters with different nominal absorbances, you can check the linearity of your absorbance scale by plotting the absorbance values measured for each wavelength against the measurement values on the DAkkS calibration certificate in a diagram.



Typical spectrum for Neutral density glass filters, measured with a slit width of 1 nm

ARTICLE NO.	666F390-25, 666F2-39, 666F201-39, 666F3-38, 666F204-37, 666F4-37, 666F202-36, 666F203-36, 666F301-361, 666F303-361
APPLICATION	Checking the photometric accuracy in the Vis range (405 nm to 890 nm)
CONTENT	**Neutral density glass filter:** F390 (0.04 Abs); F2 (0.25 Abs); F201 (0.3 Abs); F3 (0.5 Abs); F204 (0.7 Abs); F4 (1.0 Abs); F202 (1.5 Abs); F203 (2.0 Abs); F301 (2.5 Abs); F303 (3.0 Abs)
STANDARD CERTIFICATION	**Wavelengths:** 440; 465; 546.1; 590; 635 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	All wavelengths possible from 405 to 890 nm. Also possible above 890 nm, with Hellma Analytics calibration certificate **Slit width:** all possible up to 5 nm

2. GLASS FILTERS

SETS

2.3 Glass filter sets

Specifically created to meet customer requirements, Hellma Analytics glass filter sets consist of existing individual filters suitable for standard or custom validation procedures.

To ensure that filters can be easily identified, the set number is engraved on each filter frame. The absorbance/peak position values measured for each filter can be found on the DAkkS calibration certificate provided.

ARTICLE NO.	666S000	666S001
APPLICATION	Complete Glass Filter Set for testing the photometric accuracy and the wavelength accuracy of the spectrophotometer	Glass Filter Set for checking the wavelength accuracy and the photometric accuracy of the spectrophotometer
CONTENT	F1, Holmium glass filter; F2, Neutral density glass filter (0.25 Abs); F3, Neutral density glass filter (0.5 Abs); F4, Neutral density glass filter (1.0 Abs); F0, Filter frame without glass (reference filter)	F3, Neutral density glass filter (0.5 Abs); F4, Neutral density glass filter (1.0 Abs); F7, Didymium glass filter (0.5 – 1.0 Abs)
STANDARD CERTIFICATION	**F1, Holmium glass filter:** **Wavelength accuracy:** **Wavelengths accuracy at:** 279; 361; 453; 536; 638 nm **Slit width:** 1 nm **F2, F3, F4, Neutral density glass filter:** **Photometric accuracy:** **Wavelengths:** 440; 465; 546.1; 590; 635 nm **Slit width:** 1 nm	**F3, F4 Neutral density glass filter:** **Photometric accuracy:** **Wavelengths:** 440; 465; 546.1; 590; 635 nm **Slit width:** 1 nm **F7 Didymium glass filter:** **Wavelength accuracy:** **Wavelengths:** 329; 472; 512; 681; 875 nm **Photometric accuracy:** approx. 0.5 to 1.0 Abs **Wavelengths:** 270; 280; 297; 321; 342 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**F1, Holmium glass filter:** **Wavelength accuracy:** **Wavelengths:** 279; 287; 361; 418; 445; 453; 460; 536; 638 nm **Slit width:** all up to 2 nm **F2, F3, F4 Neutral densitiy glass filter:** **Photometric accuracy:** **Wavelengths:** all possible between 405 to 890 nm. Above 890 nm also possible with Hellma Analytics calibration certificate **Slit width:** all possible up to 5 nm	**F3, F4 Neutral density glass filter:** **Photometric accuracy:** **Wavelengths:** all possible from 405 to 890 nm. Also possible above 890 nm, with Hellma Analytics calibration certificate **Slit width:** all possible up to 5 nm **Didymium glass filter:** **Wavelength accuracy:** **Wavelengths:** 291; 302; 329; 430; 446; 472; 482; 512; 626; 681; 875 nm **Slit width:** all up to 2 nm **Photometric accuracy:** **Wavelengths:** from 270 to 280 nm **Slit width:** all possible up to 3 nm

More sets, as well as the complete product overview see page 60 onwards.

2.4 General usage guidelines for glass filters

Glass filters are made of glass doped with metal ions/rare earth metals, which are assembled stress-free in black anodized precision frames made of aluminum. They are designed to fit into all spectrophotometers equipped with a holder for standard cuvettes with a 10-mm optical path length. To ensure that filters can be easily identified, each filter frame is engraved with the filter type and serial number. Details of the absorbance and peak position values measured for each filter can be found on the respective calibration certificate. Please ensure that you do not touch the glass surfaces of the filter. Dirt, dust, and damage can significantly impair the accuracy of measurement results. Anodized aluminum holders should not come into contact with acids or alkalis.

STORAGE

After use, we strongly recommend storing the filters at room temperature, in their packaging and at a dry and dust-free place.

OTHER FACTORS THAT MAY INFLUENCE MEASUREMENTS

Dirt (e.g. fingerprints) and dust on, or damage (scratches, corrosion) to, polished optical surfaces can significantly impair the accuracy of measurement results. Always store the filters in their original packaging and protect the optical windows from contamination. Only handle the filters by their frames.

CLEANING

Dirt often accumulates on optical surfaces as a result of regular use. This is best removed using a lint-free cloth and alcohol.

INFLUENCE OF TEMPERATURE ON MEASUREMENTS

Temperature has a very small influence on certified measurement values, and temperatures between 20°C und 24°C fall within the measurement uncertainty stated on the calibration certificate. Measurements should therefore be taken in this range to keep any potential temperature influence on the results to a minimum.

VIDEO-TUTORIAL
Preparation and execution of measurements with glass filters

2. GLASS FILTERS

2.5 Calibration with glass filters

2.5.1 Preparations

1. Warm up the spectrophotometer until the correct operating temperature has been reached and remains constant (e.g. for one hour), taking care to observe the device manufacturer's guidelines.
2. Make sure that you use a stable cuvette holder for 10 mm standard cuvettes to measure the filters, as this is the only way to guarantee the best positioning of the filters in the light path. Check that the holder is secure and stable in the sample compartment.
3. To begin with, carry out a baseline correction with an empty sample compartment.
4. Check that the filter is correctly positioned in the light path by first placing empty filter holder F0 in the cuvette holder. The F0 marking must be visible from above. Ensure that all filter frames are always positioned in the same way, i.e. with serial numbers facing the light source.
5. Check that the device's display has not changed. In spectrophotometers with very large beams, the measurement beam may touch the filter frame (beam clipping). If this is the case, you will notice a change in the device's display.
   - » If necessary, adjust the height of the cuvette holder until the light beam shines through the aperture unimpeded. To help, you can switch the device's measurement beam to visible i.e. by adjusting the monochromator to 500 nm. There may be other ways of doing this depending on the device.
   - » If the light beam touches the sides of the aperture, adjust the horizontal position of the cuvette holder until the light beam shines through the center of the aperture. The filter frame is correctly positioned if the display values, from the zero adjustment performed in step 3 (baseline correction), do not change. In rare cases where the zero reading cannot be retained after inserting the empty filter frame and carrying out the above procedures it is permissible to re-zero the instrument with the empty filter frame in place and then continue with the filter measurements.
6. Carry out the filter measurement in a closed sample compartment as carefully as you would carry out a sample measurement (open sample compartments produce incorrect results).
7. Please note that, if you are using a diode array spectrophotometer with a stand-alone cuvette holder connected via a fiber-optic cable, extraneous light and vibrations (e.g. movement of fiber-optic cables) may also impair the accuracy of measurement results.

Watch a video of the individual steps here.

2.5.2 Steps for checking wavelength accuracy with holmium glass or didymium glass filter

1. First, carry out the "Preparations" according to chap. 2.5.1
2. Run the scan program on your spectrophotometer, observing the guidelines in the user manual. Select a scanning range that covers all of the peaks listed on the filter's calibration certificate.
3. Set your spectrophotometer to the measurement parameters that appear on the calibration certificate provided. Select – if possible – a slow scanning speed and a small data interval.
4. If possible, carry out a baseline correction.
5. Measurements are taken using an air blank which means, which means that the reference cuvette holder remains empty in double beam photometers, while a reference measurement is taken using the empty cuvette holder in single beam photometers.
6. Insert the holmium glass or didymium glass filter into the cuvette holder. Ensure that the filter is inserted into the holder as far as it will go, and that the filter ID can be seen from above. The filters must always be positioned in the cuvette holder in the same way, i.e. with the serial number facing the light source.
7. Start the measurement.
8. Calculate the positions of the peaks at the wavelengths stated on the calibration certificate. (Take several measurements and then use the mean of the measured values to avoid errors).
9. Compare your measurement values with the certified ones.

MEASUREMENT PARAMETERS FOR CHECKING WAVELENGTH ACCURACY

Ensure that you have selected the correct measurement parameters before scanning the absorbance curve to calculate peak positions. Incorrect parameters may distort the absorbance curve and thus shift the actual positions of peaks. Please use the settings stated on the accompanying calibration certificate. It should be noted that changing the slit width of the spectrophotometer can cause the absorption maxima to shift slightly. Ignore any influence that the spectral bandwidth from 1 nm to 2 nm has on peak positions. Peak heights, however, may vary greatly following changes to the slit width due to their narrow nature. As a result, filters for checking wavelength accuracy are usually unsuitable for checking absorbance accuracy.

EXPERTS TIP:

Measuring with deviating slit width:

"Generally speaking, filters can also be measured using a slit width that differs from the information provided on the calibration certificate. However, please note that large slit widths will prevent peaks lying close together from being resolved."

Thomas Brenn,
Product Manager

2. GLASS FILTERS

2.5.3 Steps for checking photometric accuracy with a neutral density glass filter or a didymium glass filter (F7A)

1. First, carry out the "Preparations" according to chap. 2.5.1
2. Run the wavelength selection program on your spectrophotometer, observing the guidelines in the user manual. Select the wavelengths provided on the calibration certificate.
3. Set your spectrophotometer to the measurement parameters that appear on the calibration certificate provided.
4. Adjust to zero.
5. Measurements are taken using an air blank which means that the reference cuvette holder remains empty in double beam spectrophotometers, while a reference measurement is taken using the empty cuvette holder in single beam spectrophotometers.
6. Insert the neutral density glass or didymium glass filter into the cuvette holder. Ensure that the filter is inserted into the holder as far as it will go, and that the filter ID can be seen from above. The filters must always be positioned in the cuvette holder in the same way, i.e. with the serial number facing the light source.
7. Start the program for measuring the absorbance values at the wavelengths stated on the calibration certificate. (Take several measurements and then use the mean of the measured values to avoid errors).
8. Compare your measurement values with the certified ones.

Watch a video of the individual steps here.

Factors in measurement uncertainty

"Measurement uncertainty arises in particular from the device-specific measurement deviation of the spectrophotometer used and from the measurement uncertainties listed on the calibration certificate."

Carola Steinger,
Chemistry lab technician

2.5.4 Calibration with glass filters – interpreting measurement results

The measurement uncertainties that appear on the calibration certificate only refer to measurements conducted by Hellma Analytics and apply solely to the measurement conditions at the company (spectrophotometer used, environmental influences such as temperature, air humidity, user influence, and reference materials used).

The smallest possible measurement uncertainty can then be derived by statistically combining the measurement uncertainty stated on the calibration certificate **and all** of the user's uncertainty contributions. These include the wavelength scale tolerance of the spectrophotometer used and other influences on measurement accuracy (environmental factors such as temperature, air humidity, user influence, etc.). For further literature on correctly calculating measurement uncertainty, please refer to chapter 8 of this user manual.

3. LIQUID FILTERS

WAVELENGTH ACCURACY

3.1 Checking wavelength accuracy

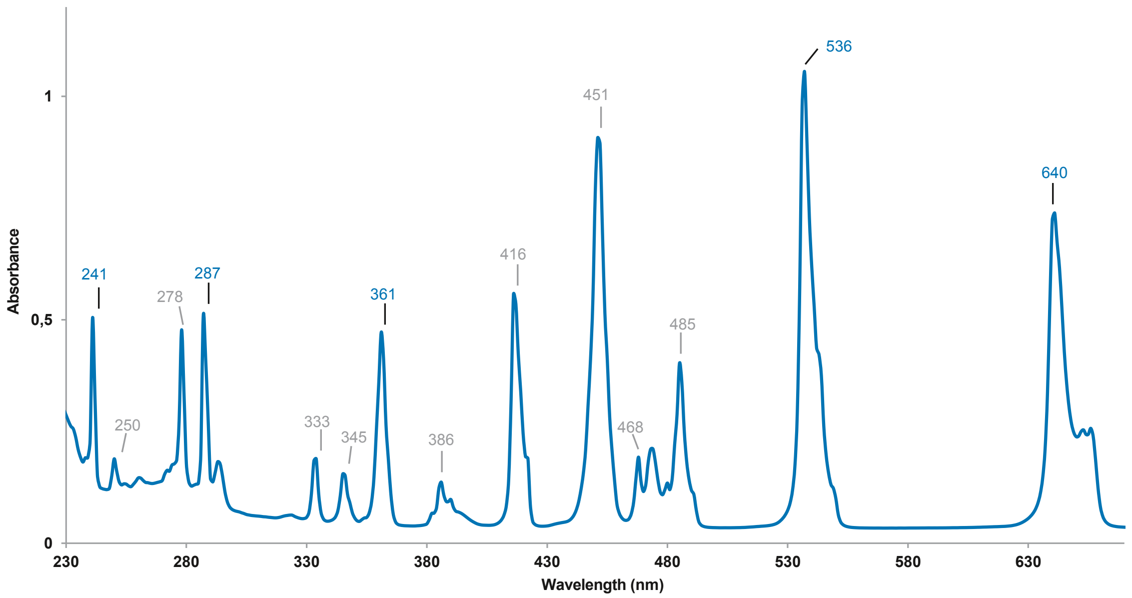
APPLICATION

To measure wavelength accuracy, the filter reduces the light beam of the spectrophotometer to a greater extent at certain wavelengths (peaks). Ideally, any standards used to determine wavelength accuracy should have narrow, well-defined peaks at a variety of wavelengths in the UV and visible range.

3.1.1 Holmium liquid filter

PRODUCT DESCRIPTION

The holmium liquid filter consists of a solution of holmium dissolved in perchloric acid. This filter is ideally suited to checking the wavelength accuracy of spectrophotometers in the UV and visible range. It has a spectrum with a variety of characteristic, very well-defined peaks in the range between 240 nm and 650 nm.



Typical spectrum of holmium dissolved in perchloric acid, measured at a slit width of 1 nm.

ARTICLE NO.	667005
APPLICATION	Checking the wavelength accuracy according to **Ph. Eur.** in the UV and Vis range
CONTENT	Holmium in perchloric acid
STANDARD CERTIFICATION	**Wavelengths:** 241, 287, 361, 536, 640 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Wavelengths:** 241, 250, 278, 287, 333, 345, 361, 386, 416, 451, 468, 485, 536, 640 nm **Slit width:** all up to 2 nm, above peaks become indistinct

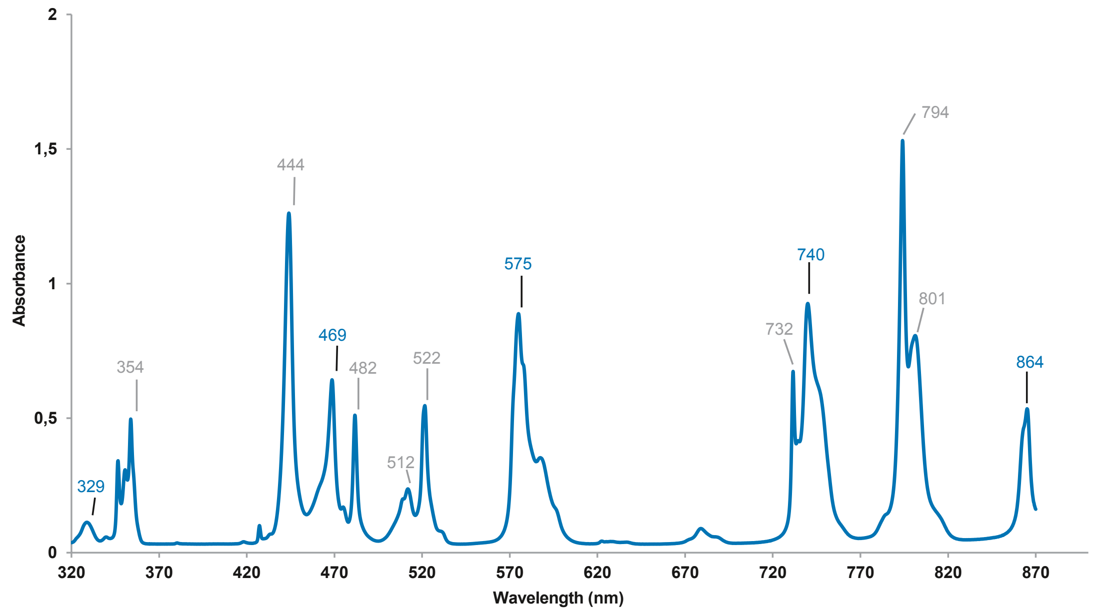
ARTICLE NO.	667005USP
APPLICATION	Checking the wavelength accuracy according to **USP <857>** in the UV and Vis range
CONTENT	Holmium in perchloric acid
STANDARD CERTIFICATION	**Wavelengths:** 241, 250, 278, 287, 333, 345, 361, 386, 416, 451, 468, 485, 536, 640 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Slit width:** all up to 2 nm

3.1.2 Didymium liquid filter

PRODUCT DESCRIPTION

The didymium liquid filter consists of praseodymium and neodymium, dissolved in perchloric acid. This filter is ideally suited for checking the wavelength accuracy of spectrophotometers in the UV and visible range. It has a spectrum with a variety of characteristic, very well-defined peaks in the range between 320 nm and 870 nm.

In USP 857, this filter is recommended for checking the wavelength accuracy above 640 nm.



Typical spectrum of Didymium (praseodymium and neodymium) dissolved in perchlroic acid measured at a slit width of 1 nm.

ARTICLE NO.	667025
APPLICATION	Checking the wavelength accuracy in the UV and Vis range
CONTENT	Didymium in perchloric acid
STANDARD CERTIFICATION	**Wavelengths:** 329, 469, 575, 740, 864 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Wavelengths:** 329, 354, 444, 469, 482, 512, 522, 575, 732, 740, 794, 801, 864 nm **Slit width:** all up to 2 nm, above peaks become indistinct

ARTICLE NO.	667025USP
APPLICATION	Checking the wavelength accuracy according to **USP <857>**
CONTENT	Didymium in perchloric acid
STANDARD CERTIFICATION	**Wavelengths:** 732, 740, 794, 801, 864 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Slit width:** all up to 2 nm

3. LIQUID FILTERS

WAVELENGTH ACCURACY

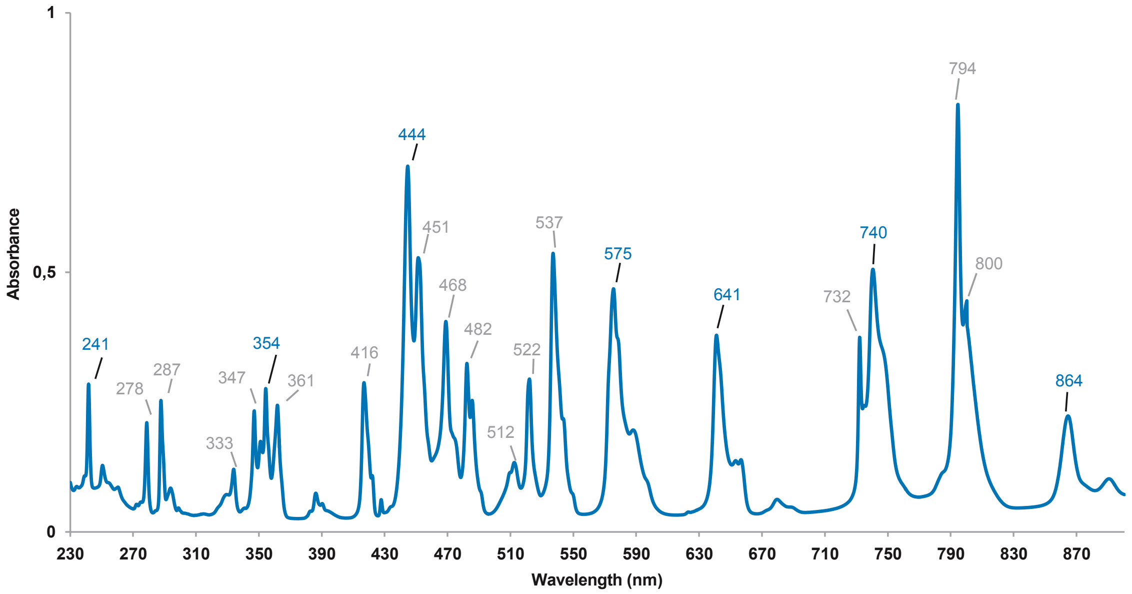
3.1.3 HoDi liquid filter

PRODUCT DESCRIPTION

The HoDi liquid filter consists of a solution of holmium and didymium (praseodymium and neodymium) in perchloric acid. This filter features an especially broad wavelength spectrum and is therefore ideally suited to checking the wavelength accuracy of spectrophotometers in the UV and visible range. It has a broad spectrum with a variety of characteristic, very well-defined peaks in the range between 241 nm and 864 nm. Depending on the performance of the spectrophotometers used, up to 22 peaks can be detected at a slit width of 1 nm.

HODI LIQUID FILTER

- Checking the UV-Vis wavelength accuracy
- Broad wavelength spectrum from 241 – 864 nm
- Two filters in one: Holmium + Didymium = HoDi



Typical spectrum of a HoDi filter, measured at a slit width of 1 nm.

ARTICLE NO.	667045
APPLICATION	Checking the wavelength accuracy in the UV and Vis range
CONTENT	Holmium and Didymium in perchloric acid
STANDARD CERTIFICATION	**Wavelengths:** 241, 354, 444, 575, 641, 740, 864 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Wavelengths:** 241, 278, 287, 333, 347, 354, 361, 416, 444, 451, 468, 482, 512, 522, 537, 575, 641, 732, 740, 794, 800, 864 nm **Slit width:** all up to 2 nm

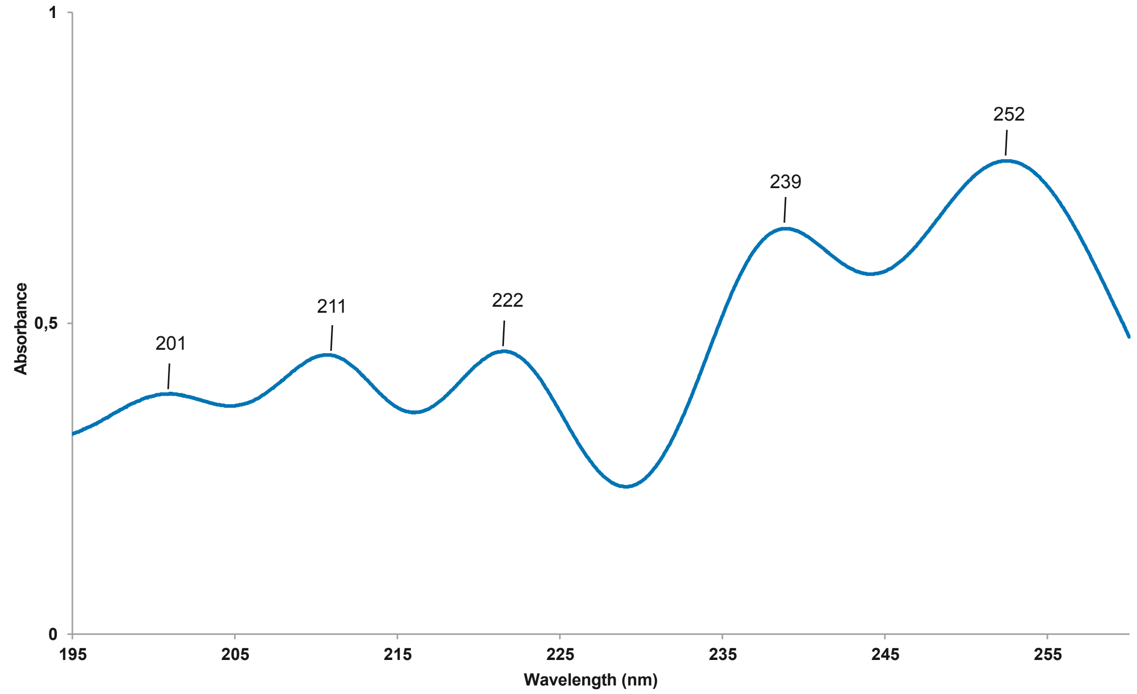
3.1.4 Rare Earth liquid filter

PRODUCT DESCRIPTION

The rare earth liquid filter consists of a solution of rare earth metals dissolved in perchloric acid. This filter is ideally suited to checking the wavelength accuracy of spectrophotometers in the low UV range. It has a spectrum with five characteristic peaks in the range from 201 nm to 252 nm.

RARE EARTH LIQUID FILTER

- Specially developed for checking wavelength accuracy in the low UV range
- Wavelength spectrum from 201 – 252 nm



Typical spectrum of a rare earth filter, measured at a slit width of 1 nm.

ARTICLE NO.	667035
APPLICATION	Checking the wavelength accuracy in the low UV range
CONTENT	Rare Earth in perchloric acid
STANDARD CERTIFICATION	**Wavelengths:** 201, 211, 222, 239, 252 nm **Slit width:** 1 nm

3. LIQUID FILTERS

PHOTOMETRIC ACCURACY

3.2 Checking the photometric accuracy

APPLICATION

Photometric accuracy (absorbance) is measured by shining a light beam from the spectrophotometer through the inserted filter. An absorbance value (Abs) can be deduced from the light attenuation caused by the filter.

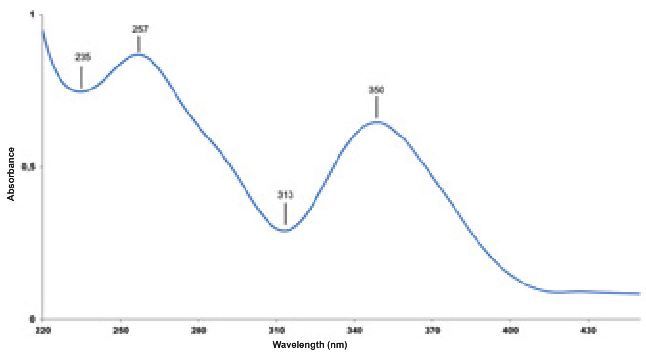
3.2.1 Potassium dichromate liquid filter for checking the photometric accuracy in accordance to Ph.Eur. and USP <857>

PRODUCT DESCRIPTION

Potassium dichromate in perchloric acid is very suitable for checking the photometric accuracy of spectrophotometers. In the UV range, the Potassium dichromate spectrum has characteristic maxima at 257 nm and 350 nm and characteristic minima at 235 nm and 313 nm. The spectrum reaches a plateau at 430 nm, which is used to determine photometric accuracy in the visible range. Hellma Analytics purchases the reference material for this filter directly from NIST (SRMR 935a "Potassium Dichromate"). The filter solutions are manufactured in strict compliance with NIST requirements and filled under controlled conditions. The cuvettes are then immediately fused to become airtight.

NOTE

As the filters are certified individually, measurement results are free from systematic errors made when preparing solutions and with regards to the optical path length of the cuvette. The measurement values of reference filter UV14 (perchloric acid measured against an air blank) appear separately on the DAkkS calibration certificate. To check absorbance linearity, take measurements using Potassium dichromate filters with different concentrations. Plot the absorbance values measured for each filter and wavelength against the measurement values that appear on the DAkkS calibration certificate in a graph.



Typical spectrum of a Potassium dichromate solution with 60 mg/l

ARTICLE NO.	667020, 667040, 667060, 667080, 6670100, 6670120, 6670140, 6670160, 6670180, 6670200, 667600, 667014 (reference filter)
APPLICATION	Checking the photometric accuracy in the UV range (235 nm to 350 nm) and Vis range (wavelength 430 nm) with a spectral bandwidth of 2 nm or less
CONTENT	UV20, 20 mg/l Potassium dichromate in $HClO_4$, (0.1 – 0.3 Abs), acc. to USP <857> UV40, 40 mg/l Potassium dichromate in $HClO_4$, (0.2 – 0.6 Abs), acc. to USP <857> UV60, 60 mg/l Potassium dichromate in $HClO_4$, (0.3 – 0.9 Abs), acc. to USP <857> and Ph. Eur. UV80, 80 mg/l Potassium dichromate in $HClO_4$, (0.4 – 1.2 Abs), acc. to USP <857> UV0100, 100 mg/l Potassium dichromate in $HClO_4$ (0.5 – 1.5 Abs), acc. to USP <857> UV0120, 120 mg/l Potassium dichromate in $HClO_4$ (0.6 – 1.8 Abs), acc. to USP <857> UV0140, 140 mg/l Potassium dichromate in $HClO_4$ (0.7 – 2.0 Abs), acc. to USP <857> UV0160, 160 mg/l Potassium dichromate in $HClO_4$ (0.8 – 2.3 Abs), acc. to USP <857> UV0180, 180 mg/l Potassium dichromate in $HClO_4$ (0.9 – 2.6 Abs), acc. to USP <857> UV0200, 200 mg/l Potassium dichromate in $HClO_4$ (1.0 – 3.0 Abs), acc. to USP <857> UV600, 600 mg/l Potassium dichromate in $HClO_4$ (1.0 Abs), acc. to Ph. Eur. UV14, perchloric acid ($HClO_4$), (reference filter)
STANDARD CERTIFICATION	**UV 20 to UV0200 at wavelengths:** 235; 257; 313; 350 nm (UV range) **UV600 at wavelength:** 430 nm (Vis range) **Slit width:** 2 nm
POSSIBLE CERTIFICATION	**Wavelengths:** fix **Slit width:** all up to 2 nm

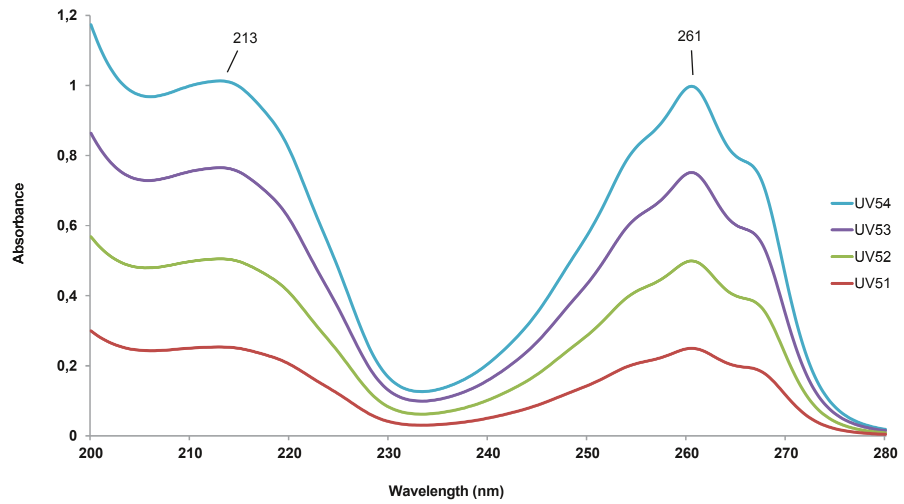
3.2.2 Niacin liquid filter

PRODUCT DESCRIPTION

Niacin (nicotinic acid) in hydrochloric acid is highly suitable for checking the photometric accuracy of spectrometers. The niacin spectrum shows in the UV range two characteristic peaks at 213 nm and 261 nm. The niacin filter solutions are filled and immediately fused under controlled conditions to become permanently airtight.

NOTE

The individually measured absorbance values are free from any systematic errors. The measured data of the reference filter UV59 (hydrochloric acid measured against air) appears separately on the DAkkS calibration certificate. To check absorbance linearity, perform the measurement with niacin filters of different concentrations. List the measured absorbance values for each filter and each wavelength in a diagram against the values on the DAkkS calibration certificate.



Typical scanlines of Niacin liquid filters, measured at a slit width of 1 nm

ARTICLE NO.	667051, 667052, 667053, 667054, 667059 (reference filter)
APPLICATION	Checking the photometric accuracy in UV range (213 nm and 261 nm) at a spectral bandwidth of 2 nm or less
CONTENT	UV51, 6 mg/l Niacin in HCl (approx. 0.25 Abs), UV52, 12 mg/l Niacin in HCl (approx. 0.50 Abs), UV53, 18 mg/l Niacin in HCl (approx. 0.75 Abs), UV54, 24 mg/l Niacin in HCl (approx. 1.0 Abs), UV59, hydrochloric acid (HCl), (reference filter)
STANDARD CERTIFICATION	**UV51 to UV54 at wavelengths:** 213 nm and 261 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Wavelengths:** fixed **Slit width:** all up to 2 nm

More sets, as well as the complete product overview see page 60 onwards.

3. LIQUID FILTERS

STRAY LIGHT

3.3 Checking for stray light

APPLICATION

In a spectrophotometer, stray light is light that passes by the sample and falls directly on the detector. This can lead to incorrect measurement results. Stray light may be caused by scattering or diffraction, by poor optical alignment, the use of incorrect or damaged cuvettes, incorrectly fitted sampling accessories or damaged seals around a light-tight sample chamber. Stray light is problematic, as it reduces the range of measurable absorbance and impairs the linearity between concentration and absorbance. Cut-off filters (filters with a strictly defined spectrum) are required to check the device for stray light.

Due to their strictly defined spectrum, Potassium Chloride filters, Sodium Iodide filters, Niacin and Sodium Nitrite filters are ideally suited to qualifying the stray light level of spectrophotometers in **compliance with pharmacopeias**. The steps are the same for all stray light filters.

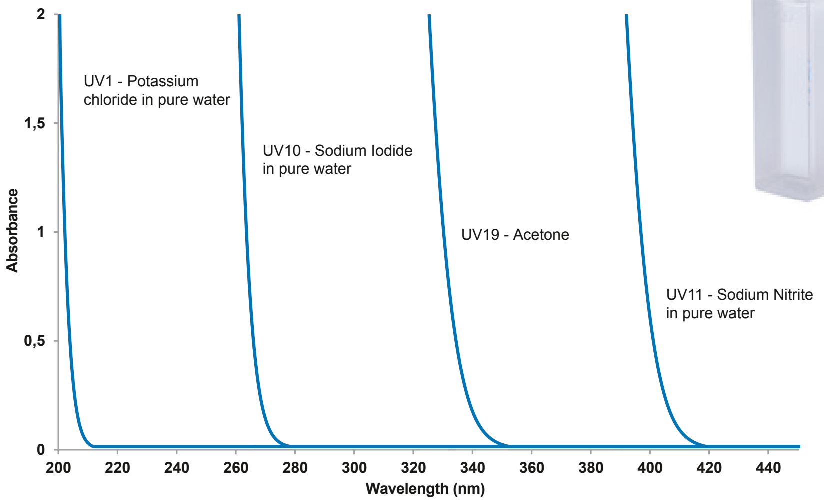
Measurement in accordance with Ph. Eur

3.3.1 Checking for stray light – measurement in accordance with Ph. Eur.

PRODUCT DESCRIPTION

Hellma Analytics stray light filters do not allow light to pass through them below a certain wavelength (cut-off wavelength). Any transmittance values displayed in the cut-off wavelength range therefore represent stray light.

According to the Ph. Eur method, the measurement is made against the reference filter which is filled with water.



Typical spectrum of the stray light filters in accordance to Ph. Eur.

ARTICLE NO.	667001, 667010, 667011, 667019, 667012 (reference for 667001, 667010, 667011)
APPLICATION	Checking for stray light in the UV range , measurement in accordance to Ph. Eur. (at wavelength 198 n. to 385 nm, depending on the selected filter)
CONTENT	UV1, Potassium chloride in pure water, nach Ph. Eur. UV10, Sodium Iodide in pure water, UV11, Sodium Nitrite in pure water, UV19, Acetone (measurement against air) UV12, pure water (reference filter for UV1, UV10, UV11)
STANDARD CERTIFICATION	**UV 1:** Cut-Off 200 nm **UV 10:** Cut-Off 259 nm **UV 11:** Cut-Off 385 nm **UV19:** Cut-Off 325 nm **UV12:** reference filter for UV1, UV10, UV11, measured against air at: 198, 200, 300, 400 nm **Slit width:** 2 nm
POSSIBLE CERTIFICATION	**Wavelength:** fix **Slit width:** all up to 5 nm

Measurement in accordance with USP <857>

3.3.2 Checking for stray light – measurement in accordance to USP <857>

PRODUCT DESCRIPTION

The new chapter <857> of the USP describes a new measuring method for checking for stray light: "When using a 5 mm path length cell (filled with the same filter) as the reference cell and then measuring the 10 mm cell over the required spectral range, analysts can calculate the stray light value from the observed maximum absorbance using the formula" (Quoted from USP <857>).

This means that in a single-beam photometer, first of all, the reference filter with 5 mm path length (filled with the same solution) is measured. The 10 mm stray light filter is then measured for the same required spectral range.

In a double-beam spectrophotometer, the stray light filter with 10 mm path length is measured against the reference filter (filled with the same solution) with 5 mm path length.

The stray light value can now be calculated from the absorbance maximum obtained, using the following formula:

$S_\lambda = 0.25 \times 10^{-2A_\lambda}$

The following acceptance criteria apply:

$A_\lambda \geq 0.7$ Abs and $S_\lambda \leq 0.01$

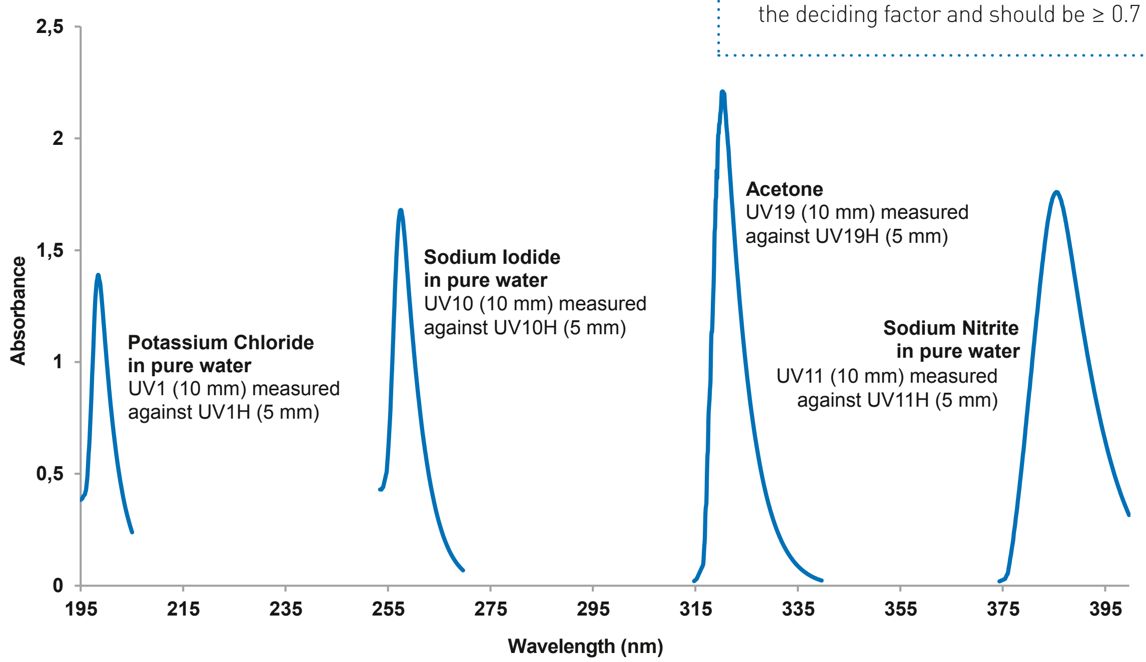
A_λ = absorbance measured at peak maximum at wavelength **λ**

S_λ = stray light value calculated at wavelength **λ**

The measurement indicated on the calibration certificate for the wavelength at the peak maximum refers only to the measurement taken with the UV-Vis-NIR spectrophotometer shown on the calibration certificate. This wavelength is instrument-dependent due to the different optical components installed and the resulting differences in performance, and so it is not applicable to other UV-Vis-NIR spectrophotometers. The indicated measurement of the wavelength at the peak maximum is not suitable for checking the wavelength scale.

NOTE

Experience from everyday use shows that the values obtained using this stray light measurement method are extremely instrument-dependent, i.e. the wavelength of the peak position varies depending on the type of instrument and its performance. The important thing for the user to know is that, when using this testing method, the maximum absorbance measured in the testing range is the deciding factor and should be ≥ 0.7 Abs.



Typical spectrum of stray light filters according to USP <857>
The reference filters with 5 mm light path are marked with the letter "H" for "half"

3. LIQUID FILTERS

STRAY LIGHT

3.3.2 Checking for stray light measurement in accordance with USP <857>

The Hellma Analytics USP stray light filter sets each consist of a 10 mm stray light filter and a 5 mm reference filter, both filled with the same solution.
These filter sets meet the criteria of USP <857> and are therefore ideal for qualifying the stray light level of spectrophotometers in accordance with the requirements of the USP.
The procedure for determining the stray light value is the same for all stray light filter sets; the difference lies in the cut-off range for each set.

Measurement in accordance with USP <857>

For improved handling of the reference filters with 5 mm light path, our USP stray light reference filters have the same external dimensions as a cuvette with 10 mm path length. The reference filters with 5 mm path length are marked with the letter H. There is no need for an additional spacer.

ARTICLE NO.	667001/667001H, 667010/667010H, 667011/667011H, 667019/667019H **Letter H indicates the reference filter with 5 mm path length**
APPLICATION	Checking for stray light in the UV range , measurement acc. to USP <857> (at wavelength 198 n. to 385 nm, depending on the selected filter)
CONTENT	UV1/UV1H, Potassium chloride in pure water, acc. to USP <857> UV10/UV10H, Sodium iodide in pure water, acc. to USP <857> UV11/UV11H, Sodium nitrite in pure water, acc. to USP <857> UV19/UV19H, pure acetone, acc. to USP <857>
STANDARD CERTIFICATION	UV1/UV1H: Cut-Off approx. 198 nm*, spectral range 190 – 205 nm UV10/UV10H: Cut-Off approx. 259 nm*, spectral range 210 – 259 nm UV11/UV11H: Cut-Off approx. 385 nm*, spectral range 300 – 386 nm UV19/UV19H: Cut-Off approx. 322 nm*, spectral range 250 – 324 nm **Slit width: 2 nm**
POSSIBLE CERTIFICATION	**Wavelength:** fixed **Possible slit width:** 1 to 5 nm

* depending on the device

3. LIQUID FILTERS

SPECTRAL RESOLUTION

3.4 Checking the spectral resolution

APPLICATION

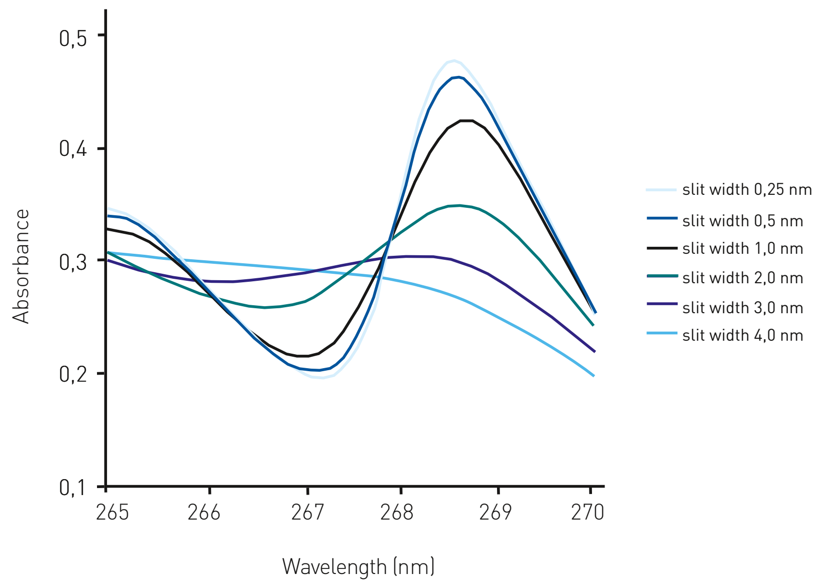
Regularly checking the spectral resolution of spectrophotometers ensures, for example, that neighbouring peaks are resolved and not superimposed on the peaks of bordering wavelengths. This also prevents absorbance errors.

PRODUCT DESCRIPTION

The Toluene in hexane liquid filter has a prominent point in its spectrum, which is excellent for determining the spectral resolution and/or actual slit width of spectrophotometers in compliance with the European Pharmacopeia, and also the USP <857>.

NOTES

A spectrophotometer's spectral resolution is very closely connected to the correct slit width setting and characterized by its ability to resolve (recognize) two very closely related peaks. The smaller the slit and corresponding spectral bandwidth, the higher the resolution.
As a rule of thumb, the slit width should be no more than 10 % of the peak width at half maximum in order to be able to determine its absorbance with an accuracy of 99.5 %. Two peaks are deemed to be resolved separately if the minimum absorbance between them amounts to less than 80 % of the peak maximum. If the spectrophotometer's spectral resolution is impaired, two different peaks will be shown as a combined peak, leading to inaccurate measurement results.



Typical spectum of the toluene liquid filter, measured with difenrent slit width.

ARTICLE NO.	667006, 667009; set: 667200
APPLICATION	Testing the resolution in accordance to Ph. Eur. and USP <857>
CONTENT	UV6, Toluene in hexane UV9, hexane (reference filter)
STANDARD CERTIFICATION	**Wavelengths:** scan from 265 to 270 nm **Slit width:** 0.5; 1.0; 1.5; 2.0; 3.0 nm with Hellma Analytics Calibration Certificate (no DAkkS Calibration Certificate)
POSSIBLE CERTIFICATION	**Wavelength:** fixed **Possible slit width:** all between 0.5 to 3 nm

Checking spectral resolution

"If the spectrophotometer's spectral resolution is impaired, two peaks may be shown as a combined peak, for example, leading to inaccurate measurement results."

Benjamin Brix,
Biological-technical assistant

3. LIQUID FILTERS

SETS

3.5 Liquid filter sets

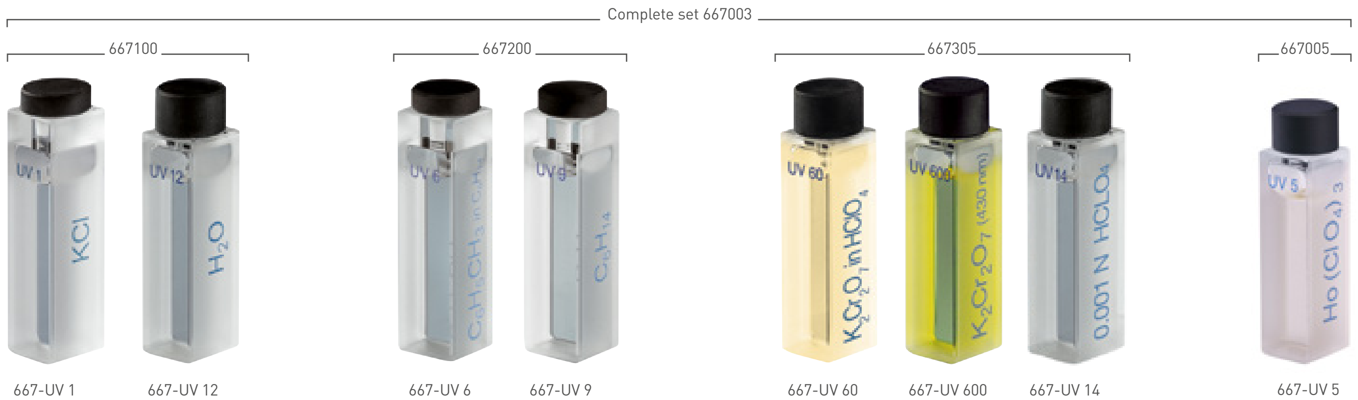
3.5.1 Set in accordance to Ph. Eur.

The complete liquid filter set – art. no. 667003 – was compiled on the basis of European Pharmacopeia requirements and contains all filters required to carry out a complete spectrophotometer check.

- ✔ **Checking wavelength accuracy (UV5)**
- ✔ **Checking photometric accuracy (UV60, UV600, UV14)**
- ✔ **Checking stray light (UV1 and UV12)**
- ✔ **Checking spectral resolution (UV6 and UV9)**

Measurement in accordance with Ph. Eur.

All liquid filters consist of reference materials that are filled into precision Hellma cuvettes made of high performance quartz glass. These cuvettes are permanently sealed, and the complete set is delivered in a high-quality storage box. To ensure easy identification, each filter is engraved with its serial number. The calibration values measured for each filter can be found on the DAkkS and Hellma Analytics calibration certificates provided.

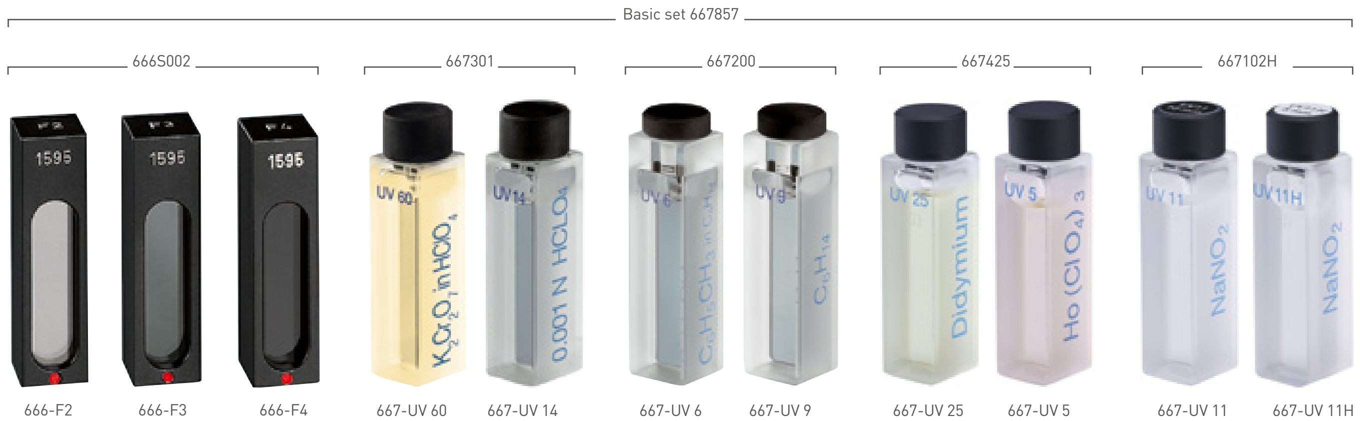


ARTICLE NO.	667003
APPLICATION	Complete set for checking spectrophotometers acc. Ph. Eur. Wavelength accuracy, photometric accuracy, stray light and spectral solution
CONTENT	UV1, Potassium Chloride in pure water; UV12, pure water (reference filter); UV5, Holmium in perchloric acid; UV6, Toluene in hexane; UV9, Hexane (reference filter); UV60, 60 mg/l Potassium Dichromate in perchloric acid with 0.3 – 0.9 Abs; UV600, 600 mg/l Potassium dichromate in perchloric acid; UV14, Perchloric acid (reference filter)
STANDARD CERTIFICATION	**UV1:** carried out against reference water (UV12), **wavelengths:** Cut-Off 200 nm; **slit width:** 2 nm **UV12: wavelengths:** 198, 200, 300, 400 nm, **slit width:** 2 nm (measurements against air) **UV5: wavelengths:** 241; 287; 361; 536; 640 nm; **slit width:** 1 nm **UV6/UV9: wavelengths:** Scan from 265 to 270 nm; **slit width:** 0.5; 1.0; 1.5; 2.0; 3.0 nm **UV60:** approx. 0.3 – 0.9 Abs **wavelengths:** 235; 257; 313; 350 nm; **slit width:** 2 nm **UV600:** approx. 1.0 Abs **wavelengths:** 430 nm; **slit width:** 2 nm
POSSIBLE CERTIFICATION	**UV1/UV12: wavelengths:** fixed; **slit width:** all until 5 nm **UV5: wavelengths:** 241; 250; 278; 287; 333; 345; 361; 386; 416; 451; 468; 485; 536; 640 nm; **slit width:** all until 2 nm, above peaks become indistinct **UV6/UV9: wavelengths:** fixed; **slit width:** 0.5 nm to 3 nm **UV60: wavelengths:** fixed; **slit width:** up to 2 nm **UV600: wavelengths:** fixed; **slit width:** up to 2 nm

Measurement in accordance with USP <857>

3.5.2 Set in accordance to USP <857>

The USP basic filter set – art.-no. 667857 – was compiled based on the specifications of the USP. The set contains a basic selection of filters for all parameters to be checked in the USP <857>. Depending on the intended operational range, this basic set can be supplemented accordingly by the user.



ARTICLE NO.	667857
APPLICATION	Basic set for checking spectrophotometers in accordance to USP <857>
CONTENT	F2, Neutral density glass filter with 0.25 Abs F3, Neutral density glass filter with 0.5 Abs F4, Neutral density glass filter with 1.0 Abs UV60, 60 mg/l Potassium dichromate in perchloric acid with 0.3 to 0.9 Abs UV14, Perchloric acid, (reference filter) UV5, Holmium in perchloric acid UV25, Didymium in perchloric acid UV11, Sodium nitrite in pure water with 10 mm path length UV11H, sodium nitrite in pure water with 5 mm path length (reference filter) UV6, Toluene in hexane UV9, Hexane (reference filter)
STANDARD CERTIFICATION	**F2, F3, F4 at wavelengths:** 440, 465, 546.1, 590, 635 nm; **slit width:** 1 nm **UV60 at wavelengths:** 235, 257, 313, 350 nm; **slit width:** 2 nm **UV5 at wavelengths:** 241, 250, 278, 287, 333, 345, 361, 386, 416, 451, 468, 485, 536, 640 nm; **slit width:** 1 nm **UV25 at wavelengths:** 732, 740, 794, 801, 864nm; **slit width:** 1 nm **UV11/UV11H, cut-off wavelengths:** approx. 385 nm; **slit width:** 2 nm **UV6/UV9:** scan at 265 – 270 nm; **slit width:** 0.5; 1.0; 1.5; 2.0; 3.0 nm
POSSIBLE CERTIFICATION	**F2, F3, F4; wavelengths: free selectable between** 405 – 890 nm**, slit width:** all up to 5 nm; **UV60: wavelengths:** fix; **slit width:** all up to 2 nm; **UV11/UV11H: wavelengths:** fix; **slit width:** 1 to 5 nm; **UV5:** no further wavelengths possible; **slit width:** all up to 2 nm; **UV25: wavelengths:** 329; 354; 444; 469; 482; 512; 522; 575; 732; 740; 794; 801; 864 nm **slit width:** all up to 2 nm; **UV6/UV9:** scan area is fixed; **slit width:** all between 0.5 to 3 nm

3. LIQUID FILTERS

3.6 General usage guidelines

Liquid filters bear a marking on one side showing the chemical formula of the substance contained in the cuvette. If a filter breaks, please observe the codes of conduct and safety instructions that apply to this substance. This information can be found in the safety instructions. Up-to-date safety instructions for all substances used to manufacture liquid filters are available at **www.hellma-analytics.com/download**

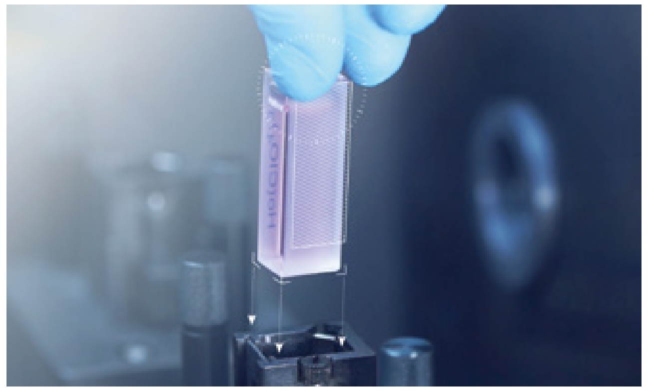
STORAGE
After use, we strongly recommend storing the filters at room temperature, in their storage box, and in a dry, dust-free area. **Liquid filters must not be exposed to temperatures below 4°C or above 40°C**. This also applies when **transporting** and **delivering** liquid filters for recertification. Especially during the winter months, attention must be paid to adequate packaging.

OTHER FACTORS THAT MAY INFLUENCE MEASUREMENTS
Dirt (e.g. fingerprints) and dust on, or damage (scratches, corrosion) to polished surfaces can significantly impair the accuracy of measurement results. Always store the filters in their original packaging and protect the optical windows from contamination. Only handle the filters by their caps or matt surfaces.

CLEANING
Dirt often accumulates on optical surfaces as a result of regular use. This is best removed using a lint-free cloth and alcohol.

INFLUENCE OF TEMPERATURE ON MEASUREMENTS
Temperature has a very small influence on certified measurement values, and temperatures of between 20°C und 24°C fall within the measurement uncertainty stated on the calibration certificate. Measurements should therefore be taken in this range to keep any potential temperature influence on the results to a minimum.

FOR INFORMATION
Always take great care when placing liquid filters in the sample holder of your spectrophotometer. Wherever possible, only touch filters by their caps or matt sides. Take care not to touch the polished surfaces. Grease on the fingers may cause a greasy film on the polished surfaces, which may affect the measurement results. The filters are fragile and should be handled with the utmost care.



3.7 Calibration with liquid filters (wavelength accuracy and photometric accuracy

3.7.1 Preparations

1. Warm up the spectrophotometer until the correct operating temperature has been reached and remains constant (e.g. for one hour). Please also note the instructions of your instrument manufacturer.
2. Make sure that you use a stable cuvette holder for 10 mm standard cuvettes to measure the liquid filters, as this is the only way to guarantee the best positioning of the filters in the light path. Check that the holder is secure and stable in the sample compartment.
3. The filters should always be positioned in the cuvette holders in the same way, i.e. with the Hellma lettering facing the light source. The light beam must pass through the part of the filter filled with liquid (solution).
4. Carry out the filter measurement in a closed sample compartment as carefully as you would carry out a sample measurement (open sample compartments produce incorrect results).
5. Please note that, if you are using a diode array spectrophotometer with a stand-alone cuvette holder connected via a fiber-optic cable, extraneous light and vibrations (e.g. movement of fiber-optic cables) may also impair the accuracy of measurement results.

VIDEO-TUTORIAL
Calibration of wavelength accuracy with liquid filter

3. LIQUID FILTERS

3.7.2 Steps for checking **wavelength accuracy** with Holmium, Didymium, Rare Earth or HoDi liquid filter

1. First of all, follow the 'steps to take before performing calibration with liquid filters' (see chapter 3.7).
2. Run the scan program on your spectrophotometer, observing the guidelines in the user manual. Select a scanning range that covers all of the peaks listed on the filter's calibration certificate.
3. Set your spectrophotometer to the measurement parameters that appear on the calibration certificate provided. Select the slowest scanning speed and a small data interval.
4. If possible, carry out a baseline correction.
5. Measurements are taken against the Perchloric Acid blank UV14, which means that the reference cuvette holder remains empty in double beam photometers, while a reference measurement is taken using the empty cuvette holder in single beam photometers.
6. Insert the holmium liquid filter into the cuvette holder, observing the general usage guidelines for liquid filters. The filters should always be positioned in the cuvette holders in the same way, i.e. with the Hellma lettering facing the light source.
7. Start the measurement.
8. Calculate the positions of the peaks at the wavelengths stated on the calibration certificate. (Take several measurements and then use the mean of the measured values to avoid errors).
9. Compare your measurement values with the certified ones, suitable for this purpose is, for example, a control chart (see page 41).

MEASUREMENT PARAMETERS FOR CHECKING WAVELENGTH ACCURACY

Ensure that you have selected the correct measurement parameters before plotting the absorbance curve to calculate peak positions. Incorrect parameters may distort the absorbance curve and thus shift the actual positions of peaks. Please use the settings stated on the accompanying calibration certificate. It should be noted that changing the slit width of the spectrophotometer can cause the absorption maxima to shift slightly. Ignore any influence that the spectral bandwidth from 1 nm to 2 nm has on peak positions. Peak heights, however, may vary greatly following changes to the slit width due to their narrow nature. A s a result, filters for checking wavelength accuracy are usually unsuitable for checking absorbance accuracy.

3.7.3 Steps for checking **photometric accuracy** with Potassium Dichromate or Niacin liquid filter

1. First of all, follow the 'steps to take before performing calibration with liquid filters' (see chapter 3.7).
2. Run the scan program on your spectrophotometer, observing the guidelines in the user manual. Select a scanning range that covers all of the peaks listed on the filter's calibration certificate.
3. Set your spectrophotometer to the measurement parameters that appear on the calibration certificate provided. If this is not possible, please ensure that the integration time is not too short.
4. If possible, carry out a baseline correction.
5. The measurements are usually carried out against a filled with perchloric acid or hydrochloric acid reference filter. Observe the general handling instructions for liquid filters. The filters should always be placed in the same orientation in the cuvette holder, eg. always with the Hellma logo to the light source.
6. **Measurement in a single-beam spectrophotometer:** Carefully insert the supplied perchloric acid or hydrochloric acid reference filter into the cuvette holder. Start the measurement. Then measure the certified reference material, which contains potassium dichromate dissolved in perchloric acid or niacin in hydrochloric acid. Then subtract the values of the reference measurement from the values of the measurement of the certified reference material.
7. **Measurement in a two-beam spectrophotometer:** Put the certified reference material with dissolved potassium dichromate in perchloric acid or niacin in hydrochloric acid, carefully into the sample holder and the perchloric acid or hydrochloric acid reference filter in the reference sample holder.
8. Start the program for measuring absorbance values at the wavelengths indicated on the calibration certificate. Take several measurements and average your measured values to avoid errors.
9. Compare your measurement values with the certified ones, suitable for this purpose is, for example, a control chart (see page 41).

EXPERTS TIP:

Measuring with differing slit width

"Generally speaking, filters can also be measured using a slit width that differs from the information provided on the calibration certificate. However, please note that large slit widths will prevent peaks lying close together from being resolved."

Carola Steinger,
Chemistry lab technician

3. LIQUID FILTERS

3.7.4 Calibration with liquid filters – interpreting the measurement results (wavelength accuracy and photometric accuracy)

MEASUREMENT PARAMETERS FOR CHECKING PHOTOMETRIC ACCURACY

As the difference between the maxima and minima in the absorbance spectrum is relatively large, the Potassium dichromate liquid filters or the Niacin filters may also be measured with a slit width that differs from the one on the calibration certificate. However, please note that using large slit widths (> 2 nm) may result in slight deviations from the values stated on the calibration certificate. In cases of doubt, it is therefore advisable to choose the slit width quoted on the calibration certificate. We recommend taking several measurements and then using the mean value to avoid errors during evaluation.

INTERPRETING THE MEASUREMENT RESULTS OF LIQUID FILTERS FOR CHECKING PHOTOMETRIC AND WAVELENGTH ACCURACY

The measurement uncertainties that appear on the calibration certificate only refer to measurements conducted by Hellma Analytics and apply solely to the measurement conditions at the company (spectrophotometer used, environmental influences such as temperature, air humidity, user influence, and reference materials used).

The smallest possible measurement uncertainty that can be achieved by the user can then be derived by statistically combining the measurement uncertainty stated on the calibration certificate **plus all the user's uncertainty contributions**, such as the wavelength scale tolerance of the spectrophotometer used and other influences on measurement accuracy (environmental factors such as temperature, air humidity, user influence, etc.). For further literature on correctly calculating measurement uncertainty, please refer to chapter 8 of this user manual.

Documentation with target value chart

"Documenting the measurement results on the target value chart gives a valuable overview and helps to quickly identify trends and deviations."

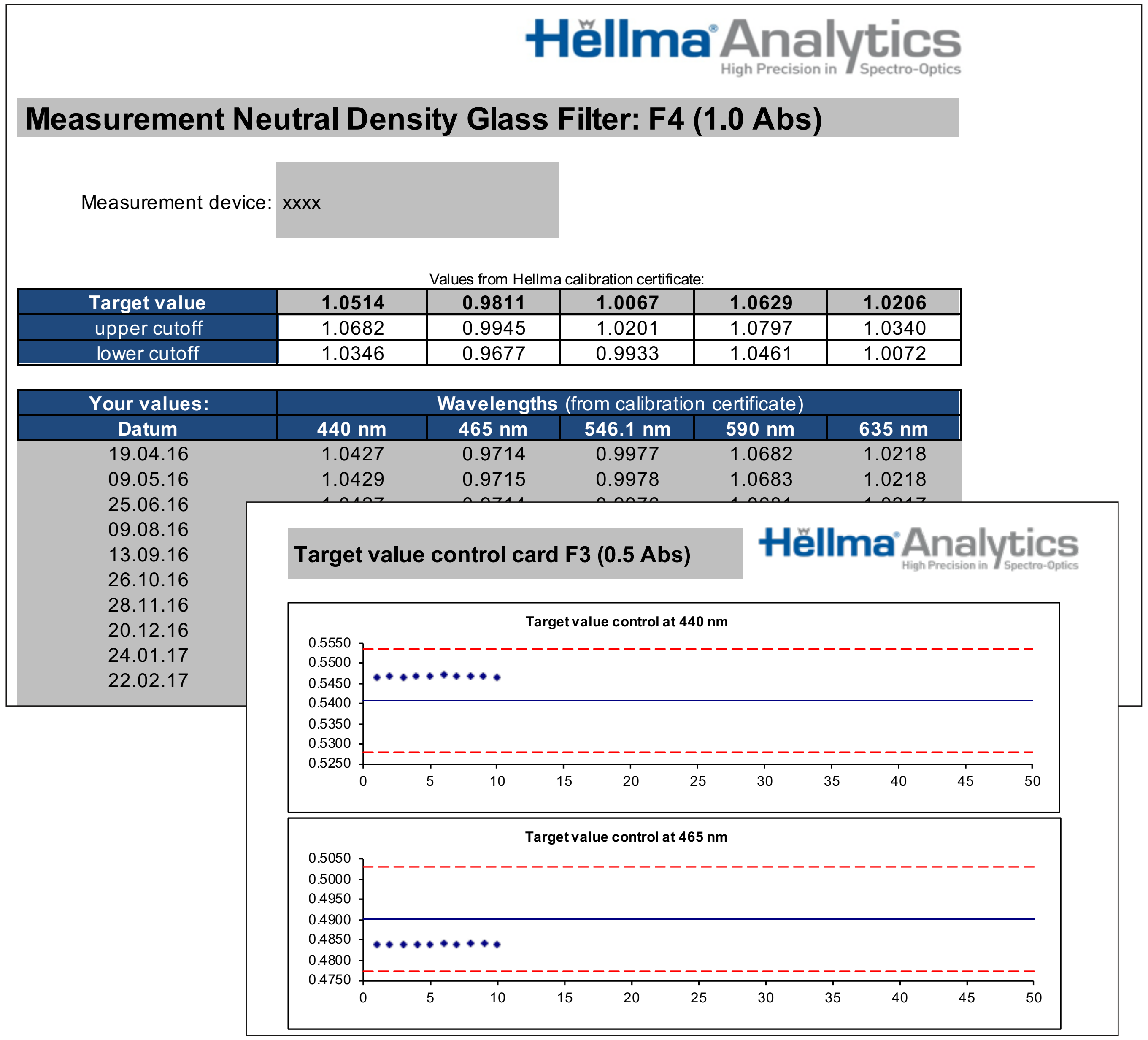
Birgit Kehl,
Compliance Representative Calibration Laboratory

Control chart for certified reference materials

In order to achieve exact measurement results, it is important to test the spectrophotometer at regular intervals and to document the measurement results achieved. The results can be documented using control charts, for example, which also display the measurement values graphically. One example of a type of control chart is the target value chart used below.

Here, the value given on the Hellma Analytics calibration certificate is set as the target value. As the exclusion limit, you should use the measurement uncertainty you have determined (measurement uncertainty from the calibration certificate plus own measurement uncertainty), i.e. all measured values must lie within the margin of measurement uncertainty in order to avoid an out-of-control situation.

To help you in the analysis, you can download an example control chart template from our website
http://www.hellma-analytics.com/control-charts



Target value	1.0514	0.9811	1.0067	1.0629	1.0206
upper cutoff	1.0682	0.9945	1.0201	1.0797	1.0340
lower cutoff	1.0346	0.9677	0.9933	1.0461	1.0072

Your values:	Wavelengths (from calibration certificate)				
Datum	440 nm	465 nm	546.1 nm	590 nm	635 nm
19.04.16	1.0427	0.9714	0.9977	1.0682	1.0218
09.05.16	1.0429	0.9715	0.9978	1.0683	1.0218
25.06.16					
09.08.16					
13.09.16					
26.10.16					
28.11.16					
20.12.16					
24.01.17					
22.02.17					

3. LIQUID FILTERS

Measurement in accordance with Ph. Eur.

3.8 Calibration with liquid filters (stray light and spectral resolution)

3.8.1 Steps for checking the stray light level in accordance to Ph. Eur. + interpretation

1. First of all, follow the 'steps to take before performing calibration with liquid filters'.
2. Run the scan program on your spectrophotometer, observing the guidelines in the user manual. Select a scanning range that covers all of the values listed on the filter's calibration certificate.
3. Set your spectrophotometer to the measurement parameters quoted on the calibration certificate provided.
4. Set the spectrophotometer to a wavelength of approx. 20 nm above the cut-off wavelength for the stray light filter used (for Potassium chloride (UV1), for example, start at 220 nm) and scan down to the wavelength for which you wish to determine the stray light level.
5. If possible, carry out a baseline correction.
6. The measurement is usually carried out against a reference filter (UV12) filled with water (in the case of acetone measurement against air). Please note the general guidelines for liquid filters. The filters should always be positioned in the cuvette holders in the same way, i.e. with the Hellma Analytics lettering facing the light source.
7. **Measurements in a single beam spectrophotometer:** Carefully insert the reference filter provided into the cuvette holder. Start the measurement. Next, measure the certified reference material. Then subtract the reference measurement values from the measurement values of the certified reference material.
8. **Measurements in a double beam spectrophotometer:** Carefully insert the certified reference material into the sample holder and the reference filter into the reference sample holder. Start the measurement.
9. Scan down to the wavelength for which you wish to determine the stray light level.
10. The light level (remaining transmittance value) measured below the cut-off wavelength represents stray light.

MEASUREMENT PARAMETERS FOR CHECKING STRAY LIGHT LEVEL

For a realistic calculation of the stray light level, choose a filter with a cut-off wavelength as close above the required wavelength as possible. This enables the stray light test to be carried out at the wavelength at which the stray light filter can fully absorb light. The remaining transmittance displayed by the device at the measurement wavelength represents the stray light level. Since this value differs depending on the properties of the measuring system, filters can only be certified with regard to their suitability for use as a stray light filter. Certification therefore demonstrates that filters have virtually full absorbance in the measuring range and steep peaks at high transmittance values.

FOR INFORMATION

Please note that the reference for the measuring system to be tested is not provided by the entire transmittance characteristic of the stray light filter, but solely by the transmittance value measured in the range of virtually full absorbance.

FOR INFORMATION

You can check the lower absorbance range of your spectrophotometer using reference filter 667-UV12, which is filled with ultrapure water. The filter's absorbance characteristics from 200 nm to NIR are practically only determined by the reflection losses on the two air/glass surfaces. You can check your device's display at very low absorbance values against the certified values at 198 nm, 200 nm, 300 nm and 400 nm. If your results differ significantly from the certified values, particularly if the measured values are smaller than 0.02 A , you should contact the service engineer.

NOTE:

Interpreting measurement results when checking for stray light. To estimate the sample measurement error due to stray light, compare the calculated stray light level to the signal strength from the sample measurement. For example, a stray light value of 0.1% transmittance and a sample with an absorbance of around 1 Abs would equate to a measurement error due to stray light of around 0.4%. If you have calculated a stray light level that is considerably higher than the level stated in the device specifications, check whether extraneous light could have interfered with this result. If you can rule out extraneous light, please contact a service technician.

3. LIQUID FILTERS

Measurement in accordance with USP <857>

3.8.2 Procedure for checking the stray light level according to USP <857> + Interpretation

1. First, carry out the "Steps to take before performing calibration with liquid filters" according to Chap. 3.7.
2. Run the scan program on your spectrophotometer, observing the guidelines in the user manual. Select the limits of the scanning range so that all values listed on the filter's calibration certificate are recorded.
3. If possible, set your spectrophotometer to the measurement parameters quoted on the calibration certificate provided.
4. Set the spectrophotometer to a wavelength of approx. 20 nm above the cut-off wavelength for the stray light filter used in each case with 10 mm path length (for Potassium chloride (UV1), for example, start at 220 nm) and scan to approx. 20 nm below the cut-off wavelength.
5. If possible, carry out a baseline correction.
6. The measurement is carried out against a reference filter with 5 mm path length, filled with the same solution. Follow the general usage guidelines for liquid filters. The filters should always be positioned in the cuvette holders in the same way, e.g. always with the Hellma lettering facing the light source.
7. **Measurements in a single-beam spectrophotometer:** Carefully insert the reference filter provided, with 5 mm path length, into the cuvette holder. Start the measurement. Next, measure the filter with 10 mm path length, filled with the same solution.
8. **Measurements in a double-beam spectrophotometer:** Carefully insert the filter with 10 mm path length into the sample holder, and the reference filter with 5 mm path length, filled with the same solution, into the reference sample holder. Start the measurement.
9. Scan a range of 20 nm around the cut-off.
10. Record the maximum absorbance value measured at wavelength λ (= A_λ)
11. Check whether the recorded absorbance value is ≥ 0.7 Abs.
12. Next, use the following formula to calculate the stray light level:
    $S_\lambda = 0.25 \times 10^{-2A_\lambda}$
    S_λ is the stray light value calculated for wavelength λ.
    A_λ = maximum absorbance value measured at wavelength λ.
13. Check whether $S_\lambda \leq 0.01$.

MEASUREMENT PARAMETERS FOR CHECKING STRAY LIGHT LEVEL

To enable a realistic estimate of the stray light level, choose a filter set with a cut-off wavelength as close above the required wavelength as possible.

The stray light test is then carried out with the appropriate filter set, consisting of a stray light filter with 10 mm path length and the associated reference filter with 5 mm path length, both filters being filled with the same solution.

The stray light level S_λ calculated using the formula corresponds to the stray light level of the device at the measurement wavelength. Since this value as well as the location of the peak maximum differ depending on the properties of the measuring system, the filter set can only be certified with regard to its suitability for use as a stray light filter in accordance with USP <857>.

Measurement in accordance with USP <857>

667-UV105H liquid filter set for checking stray light acc. to USP <857>

INTERPRETING MEASUREMENT RESULTS WHEN CHECKING FOR STRAY LIGHT

If the stray light level you have observed does not correspond to the default value criteria of USP <857>, i.e. the absorbance at the peak maximum is > 0.7 Abs and the stray light value calculated for the wavelength of the peak maximum S_λ is > 0.01, first check whether your result was caused by extraneous light. If you can rule out extraneous light, please contact one of your device manufacturer's service technicians.

3. LIQUID FILTERS

3.8.3 Steps for checking spectral resolution + interpretation

1. First of all, follow the 'steps to take before performing calibration with liquid filters' – see chapter 3.7.
2. Run the scan program on your spectrophotometer, observing the guidelines in the user manual. Select a scanning range that covers both of the required peaks.
3. Set your spectrophotometer to the measurement parameters stated on the calibration certificate provided.
4. If possible, carry out a baseline correction.
5. Take the measurement using a reference filter filled with hexane – if the spectrum is corrected to zero at 300 nm, measurements can also be taken using air – observing the general usage guidelines for liquid filters. The filters should always be positioned in the cuvette holders in the same way, i.e. with the Hellma Analytics lettering facing the light source.
6. **Measurements in a single beam spectrophotometer:** Carefully insert the hexane reference filter provided into the cuvette holder. Start the measurement. Next, measure the certified reference material, which contains Toluene in hexane. Then subtract the reference measurement values from the measurement values of the certified reference material.
7. **Measurements in a double beam spectrophotometer:** Carefully insert the Toluene in hexane liquid filter into the sample holder and the hexane reference filter into the reference sample holder. Start the measurement.
8. Measure the actual minimum of the absorbance values at 266 nm and the actual maximum at 269 nm. (Take several measurements and then use the mean of the measured values to avoid errors).
9. If possible, carry out a baseline correction each time you change the slit width.
10. Determine the ratio using both of the measured values, as stated on the calibration certificate provided.

MEASUREMENT PARAMETERS WHEN CHECKING SPECTRAL RESOLUTION

When measuring spectral resolution, the liquid filter absorbs the light beam from the spectrophotometer to significantly different extents in a narrow wavelength range (5 nm). The filter will show a clear maximum and minimum within the narrow range. After placing the liquid filter in the spectrophotometer, run the scan program in the defined wavelength range and divide the maximum peak measured at λmax = 269 nm by the minimum peak measured at λmin = 266 nm. The resulting ratio represents the absorbance ratio, which is directly linked to the slit width. If the ratio is considerably lower (e.g. 15%), please contact the device manufacturer. Please note, however, that the result also depends on the measurement conditions. Therefore, please make sure that you select a sufficiently long integration time, particularly if using a small slit width.

INTERPRETING MEASUREMENT RESULTS WHEN CHECKING SPECTRAL RESOLUTION

Regulatory codes or internal applications and measuring procedures may place requirements on the ratios that must be achieved. In addition, comparing calculated ratios with certified values may provide an indication of the actual slit width of the device used.

OVERVIEW: ABSORBANCE RATIO OF MAXIMUM/MINIMUM PEAK IN RELATION TO SLIT WIDTH

SLIT WIDTH	ABSORBANCE RATIO
0.5	2.2
1.0	2.0
2.0	1.4
3.0	1.1

(see: Standards and Best Practice in Absorption Spectrometry, edited by C. Burgess & T. Frost)

FOR INFORMATION

Please note that the filter set for determining spectral resolution does not fall within our scope of accreditation, and therefore cannot be issued with a DAkkS calibration certificate or calibration mark.

4. REFERENCE PLATES

PHOTOMETRIC ACCURACY

4.1 Checking the photometric accuracy

APPLICATION

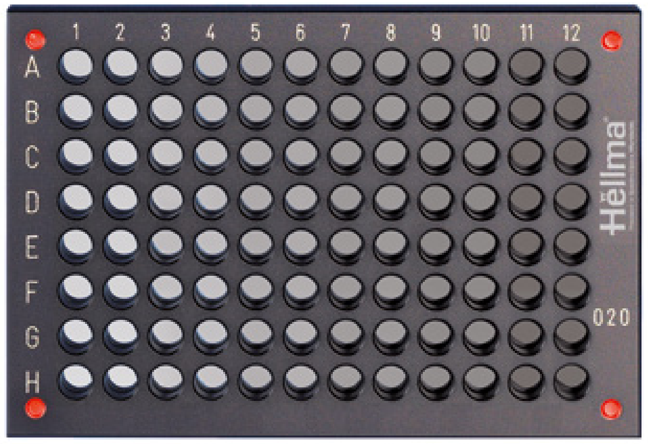
The Hellma Analytics reference plate 666R013 can be used to check the photometric accuracy of microplate readers.

PRODUCT DESCRIPTION

The reference plate dimensions are equivalent to a 96-well microplate with a diameter of 6.6 mm per window (H 14.5 mm x D 125 mm x L 85.5 mm). Each of the five Neutral density glass filters (columns 3 – 12) in the reference plate can measure the absorbance value for 16 windows. The other 16 windows do not contain glass (column 1 + 2) and serve as references.

NOTE

The reference plate has five Neutral density glass filters with different nominal absorbance values, allowing you to check the linearity of your absorbance scale by plotting the absorbance values measured for each wavelength against the measurement values on the DAkkS calibration certificate in a diagram.



ARTICLE NO.	666R013
APPLICATION	Reference plate for microplate readers for testing the photometric accuracy
CONTENT	Neutral density glass filter (0.25 Abs); column 3 + 4 Neutral density glass filter (0.5 Abs); column 5 + 6 Neutral density glass filter (1.0 Abs); column 7 + 8 Neutral density glass filter (1.5 Abs); column 9 + 10 Neutral density glass filter (2.5 Abs); column 11 + 12 column 1 + 2 without glass (reference filter)
STANDARD CERTIFICATION	**Photometric accuracy certified at wavelengths:** 405; 450; 490; 650 nm; at 8 points in a column **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Wavelengths:** all possible from 405 to 890 nm Also possible above 890 nm, but only with a Hellma Analytics calibration certificate **Slit widths:** all possible up to 5 nm

PHOTOMETRIC ACCURACY AND WAVELENGTH ACCURACY

4.2 Checking photometric and wavelength accuracy

APPLICATION

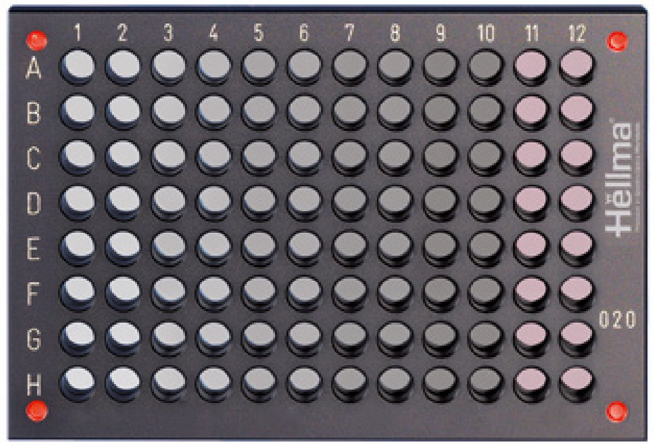
The Hellma Analytics reference plate 666R113 can be used to check the photometric and wavelength accuracy of microplate readers.

PRODUCT DESCRIPTION

The reference plate dimensions are equivalent to a 96-well microplate with a diameter of 6.6 mm per window (H 14.5 mm x D 125 mm x L 85.5 mm). Each of the four Neutral density glass filters used (columns 3 – 10) can measure the absorbance value for 16 windows. Holmium glass filter (column 11+12) are used to test the wavelength accuracy in 16 windows while a further 16 windows (columns 1+2) do not contain glass and serve as references.

NOTE

Filters are routinely set at a thickness that produces the indicated nominal optical density (ranging from 0.04 – 3.0 Abs) at 546.1 nm. This results in increasingly larger absorbances the shorter the wavelengths become.



ARTICLE NO.	666R113
APPLICATION	Reference plate for microplate readers for testing the photometric and wavelength accuracy.
CONTENT	Neutral density glass filter (0.5 Abs), column 3+4 Neutral density glass filter (1.0 Abs), column 5+6 Neutral density glass filter (1.5 Abs), column 7+8 Neutral density glass filter (2.0 Abs), column 9+10 Holmium glass filter, column 11+12 column 1 + 2 without glass (reference filter)
STANDARD CERTIFICATION	**Photometric accuracy certified at 8 points in column at wavelengths:** 405; 450; 490; 650 nm; **Wavelength accuracy certified at:** 279; 361; 453; 536; 638 nm **Slit width:** 1 nm
POSSIBLE CERTIFICATION	**Photometric accuracy:** **Wavelengths:** additional wavelengths: all possible from 405 to 890 nm also possible above 890 nm, but only with a Hellma Analytics Calibration Certificate **Slit widths:** all possible up to 5 nm **Wavelength accuracy:** **Wavelengths:** 279; 287; 361; 418; 445; 453; 460; 536; 638 nm **Slit widths:** all up to 2 nm recommended

4. REFERENCE PLATES

4.3 General usage guidelines for reference plates

Reference plates are made of glass doped with metal ions or rare earth metals, which is annealed and assembled in black anodized precision frames made of aluminum. They are designed to fit into all **microplate readers**. To ensure easy identification, each reference plate is engraved with the reference plate type and serial number. Details of the absorbance and peak position values measured for each filter can be found on the respective calibration certificate. Please ensure that you do not touch the glass surfaces of the filter. Dirt, dust, and damage can significantly impair the accuracy of measurement results. Anodized aluminum frames should not come into contact with acids or alkalis.

STORAGE

After use, we recommend storing reference plates at room temperature, in their packaging, and in a dry, dust-free area.

OTHER FACTORS THAT MAY INFLUENCE MEASUREMENTS

Dirt (e.g. fingerprints) and dust on, or damage (scratches, corrosion) to, polished surfaces can significantly impair the accuracy of measurement results. Always store reference plates in their original packaging and protect the optical windows from contamination. Only handle reference plates by their frames.

CLEANING

Dirt often accumulates on optical surfaces as a result of regular use. This is best removed using a lint-free cloth and alcohol.

INFLUENCE OF TEMPERATURE ON MEASUREMENTS

Temperature has a very small influence on certified measurement values. Measurements taken at temperatures between 20°C und 24°C fall within the measurement uncertainty stated on the calibration certificate. Measurements should therefore be taken in this range to keep any potential temperature influence on the results to a minimum.

4.4 Calibration with reference plates

4.4.1 Preparations

1. Warm up the microplate reader until the correct operating temperature has been reached and remains constant (e.g. for one hour), taking care to observe the device manufacturer's guidelines.
2. To begin with, carry out a baseline correction with an empty sample compartment.
3. Check that the reference plate is correctly positioned in the light path by first measuring the windows without glass (usually columns 1 and 2). The label showing the reference plate type must be visible from above.
4. Check that the device's display has not changed. In microplates with very large beams, the measurement beam may touch the window frame. If this is the case, you will notice a change in the device's display.
   » If necessary, adjust the position of the reference plate holder until the light beam shines through the empty window unimpeded.
   » The reference plate is correctly positioned if the display values from the zero adjustment performed in step 2 (baseline correction) do not change.
5. Carry out the filter measurement in a closed sample compartment as carefully as you would carry out a sample measurement (open sample compartments produce incorrect results).

A careful handling of the reference plates is very important. An impurity on the glass surfaces, leads to wrong measurement results.

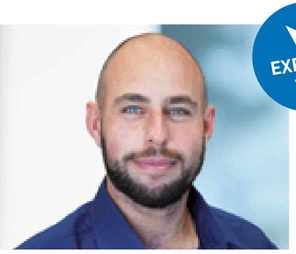
4. REFERENCE PLATES

4.4.2 Steps for checking photometric accuracy with reference plates

1. First of all, follow the 'steps to take before performing calibration with reference plates'.
2. Run the wavelength selection program on your microplate reader, observing the guidelines in the user manual. Select the wavelengths stated on the calibration certificate.
3. Set your microplate reader to the measurement parameters quoted on the calibration certificate provided.
4. Adjust to zero.
5. Place the reference plate in the plate holder. Ensure that the reference plate ID is visible from above. Reference plates must always be positioned in the plate holders in the same way.
6. Start the program for measuring absorbance values at the wavelengths stated on the calibration certificate – the positions measured are those where Neutral density glass filters are inserted.
7. Take several measurements and then use the mean of the measured values to avoid errors.
8. Compare your measurement values with the certified ones.

MEASUREMENT PARAMETERS FOR CHECKING PHOTOMETRIC ACCURACY

Generally speaking, reference plates can also be measured using a slit width that differs from the information provided on the calibration certificate. However, please note that using large slit widths may result in slight deviations from the values stated on the calibration certificate. In cases of doubt, it is therefore advisable to choose as small a slit width as possible. We recommend taking several measurements and then using the mean value to avoid errors during evaluation.



EXPERTS TIP:

Composition of the measurement uncertainty

The measurement uncertainty consists in particular of the device-specific measurement deviation of the used spectrophotometer and the in the calibration certificate listed measurement uncertainties together.

Benjamin Brix,
Biological-technical assistant

4.4.3 Steps for checking wavelength accuracy with reference plates

1. First of all, follow the 'steps to take before performing calibration with reference plates' see chap. 4.4.1
2. Run the scan program on your microplate reader, observing the guidelines in the user manual. Select a scanning range that covers all of the peaks listed on the filter's calibration certificate.
3. Set your microplate reader to the measurement parameters quoted on the calibration certificate provided. Select a slow scanning speed and a small data interval.
4. If possible, carry out a baseline correction.
5. Place the reference plate in the plate holder. Ensure that the filter ID is visible from above. Reference plates must always be positioned in the plate holders in the same way.
6. Start the measurement for the positions where holmium glass filters are inserted (usually column 11 + 12).
7. Detect the positions of the peaks at the wavelengths stated on the calibration certificate.
8. Take several measurements and then use the mean of the measured values to avoid errors.
9. Compare your measurement values with the certified ones.

MEASUREMENT PARAMETERS FOR CHECKING WAVELENGTH ACCURACY

Ensure that you have selected the correct measurement parameters before scanning the absorbance curve to detect peak positions. Incorrect parameters may distort the absorbance curve and thus shift the actual positions of peaks. Please use the settings stated on the accompanying calibration certificate. It should be noted that changing the slit width of the microplate reader can cause the absorbance maxima to shift slightly. Ignore any influence that the spectral bandwidth from 1 nm to 2 nm has on peak positions. Peak heights, however, may vary greatly following changes to the slit width due to their narrow nature. As a result, filters for checking wavelength accuracy are usually unsuitable for checking absorbance accuracy.

INTERPRETING MEASUREMENT RESULTS WITH REFERENCE PLATES FOR CHECKING PHOTOMETRIC AND WAVELENGTH ACCURACY

The measurement uncertainties stated on the calibration certificate only refer to measurements conducted by Hellma Analytics and apply solely to the measurement conditions at the company (spectrophotometer used, environmental influences such as temperature, air humidity, user influence, and reference materials used).

The smallest possible measurement uncertainty that can be achieved by the user can then be derived by statistically combining the measurement uncertainty stated on the calibration certificate with all the user's uncertainty contributions, such as the wavelength scale tolerance of the microplate reader used and other influences on measurement accuracy (environmental factors such as temperature, air humidity, user influence, etc.). For further literature on correctly calculating measurement uncertainty, please refer to chapter 8 of this user manual.

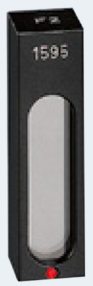
5. RECERTIFICATION

Continuously assured quality: Recertification intervals for reference materials

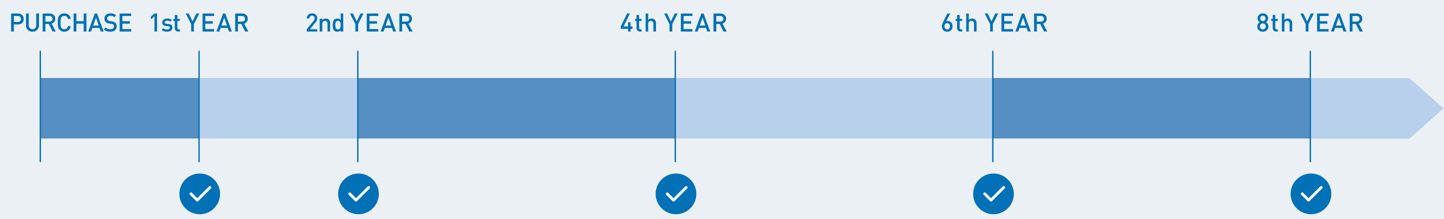
As is the case for all measuring devices, the **reference materials used to verify spectrophotometers must also be chekked and recertified at regular intervals** (see for example ISO 9001:2008 "Control of Monitoring and Measuring Equipment"). This allows you to ensure that you consistently fulfill your in-house **quality requirements and guarantees high levels of accuracy and reliability in your measurements**.

Important parameters for recertification

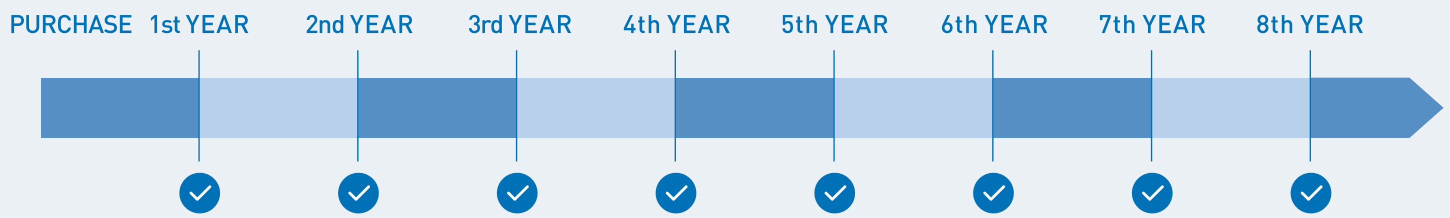
The **length of intervals** between the recertification of reference materials depends on how frequently materials are used, the wear associated with this, accuracy requirements, and the requirements of a company's internal auditing. In general, a recertification interval of **12 months** is recommended for checking and recertifying **glass filters** during the first two years of use, with an interval of **24 months** thereafter. We recommend verifying and recertifying **liquid filters** within a maximum of **12 months**. Intervals should be specified individually in accordance with your QM system.



GLASS FILTERS RECERTIFICATION EVERY 24 MONTHS



LIQUID FILTERS RECERTIFICATION INTERVAL OF 12 MONTHS IS RECOMMENDED



Fast and reliable – recertification service

In our DAkkS-accredited calibration laboratory, your reference materials are cleaned and are recertified in accordance with your requirements using a high-performance spectrophotometer. If necessary, filters are repaired, or are exchanged following a consultation.
You will receive your filters with a new DAkkS calibration certificate or Hellma Analytics calibration certificate.
Filters are usually recertified within five working days of their arrival at the calibration laboratory.

Recertification of reference materials from other manufacturers.
We also recertify reference materials for UV/Vis spectroscopy from other suppliers. If you require a quotation first, **please send your inquiry via email to your local Hellma partner**.

PLEASE NOTE:
Liquid filters can only be sent when the external temperature is above 4°C, as the liquid can freeze, which will destroy the reference materials.

Returning your reference materials for recertification

Efficient processing of the reference materials you send us ensures that you will be able to use your filters again within just a few days.
We need your support to make this possible. Please include all information needed to process the reference materials:

- **Article no.***
- **Serial no.***
- **Wavelength(s) to be measured***
- **Slit width(s) to be measured***
- **Documentation of measurement data prior to cleaning** Yes / No**
- **Quotation no.** (if you have already received a quotation from us)
- **Billing address**
- **Delivery address** (if different from billing address)
- **Special requests, e.g. additional wavelengths etc**

Please enclose a copy of your order or send this via email to your local Hellma partner.
If you send your reference materials with only a delivery note, **it is essential that you indicate your order number**.
Please include this on the delivery note, otherwise we will be unable to process your order.

*This information is not necessary if you enclose a copy of the current calibration certificate.
** Documentation of measurement data prior to cleaning
If you require documentation of measurement data prior to cleaning, please note this on your order. Depending on your quality management requirements, you have two options:
1. Documentation of measurement data prior to cleaning with DAkkS certificate.
2. Documentation of measurement data prior to cleaning with simple measurement report.

6. FAQ

6.1 How does the recertification of my filters work?

Hints for the return and recertification of your reference materials can be found on page 55.

6.2 Why do holmium glass filters become cloudy? Will this interfere with the measurement?

The glass material used for this filter is somewhat hygroscopic, which means that the filters become coated with a kind of water film. The film does not interfere with measurements or change the characteristic peak positions of holmium. The filter can be easily wiped down using alcohol and a soft cloth. The filter should generally be stored in a dry place.

6.3 How long can a calibration standard be used for in total?

Depending on the conditions in which they are used and stored, as well as how they are maintained, filters usually last for many years. We recommend having filters regularly recertified so that any signs of deterioration can be recognized at an early stage.

6.4 How often should filters be recertified?

Certified reference materials should be recertified at regular intervals to check that the values stated on the calibration certificate are still valid. It is up to the user to decide on the regularity of these intervals, which should take into account the use, storage and usage conditions of the filter in the laboratory. To establish a statistical database for determining recertification intervals, we recommend having all reference materials recertified at least every 12 months during their first two years of use, and then selecting a suitable recertification interval based on the values measured. (Please see chapter 5).

6.5 What do the tolerances on the calibration certificate tell us and how can they be correctly interpreted?

The measurement uncertainties that appear on calibration certificates only refer to measurements conducted by Hellma Analytics and apply solely to the measurement conditions at the company (spectrophotometer used, environmental influences such as temperature, air humidity, user influence, reference materials used, etc.). Consequently, the measurement uncertainties of the NIST reference materials used to ensure traceability have been mathematically combined with the measurement uncertainty statistics calculated by Hellma Analytics. The value provided is therefore an expanded measurement uncertainty (double standard deviation, coverage factor k=2). This means that the actual value is 95% certain to fall within this range. To correctly calculate the measurement uncertainties valid for their measuring system, reference material users should follow the same steps, mathematically/statistically combining the measurement uncertainties provided with the measurement uncertainty statistics they have calculated themselves for a particular spectrophotometer and relevant conditions (see ISO/IEC Guide 98-3:2008 'Guide to the Expression of Uncertainty in Measurement').

6.6 What is a baseline correction?

Baseline corrections are carried out with an empty cuvette holder to compensate for the lamps. Since lamps emit light at different strengths at various wavelengths, baseline corrections (also known as auto zero) are carried out to determine a zero value. Baseline corrections are usually performed automatically when the spectrophotometer is started up, but can also be carried out manually.

6.7 What is background correction?

Background correction is carried out to eliminate any influences that extend beyond the sample's properties. In double beam photometers, background correction is performed by simultaneously measuring the comparison cuvette in the reference beam path. This comparison cuvette usually contains pure solvent. In single beam photometers, background correction is carried out before the actual sample measurement is taken by measuring the comparison cuvette. The values obtained for the comparison cuvette are then deducted from the values of the sample measurement.

6.8 Why does the calibration certificate for the filter set used to determine spectral resolution look different to other calibration certificates?

Determining spectral resolution does not fall within our scope of accreditation. The filter set for determining spectral resolution therefore cannot be issued with a DAkkS calibration certificate or calibration mark. That is why this calibration certificate looks different from other calibration certificates for filter sets.

6.9 Why does Hellma Analytics no longer offer Potassium dichromate filters for checking photometric accuracy with sulfuric acid as a solvent, as described in the European Pharmacopoeia?

In the past, certified reference materials for checking photometric accuracy in the UV range contained a solution of Potassium dichromate in sulfuric acid and were manufactured in strict compliance with European Pharmacopoeia requirements. Over a number of years, Hellma Analytics noticed a continuous decrease in the absorbance values of the 'Potassium dichromate dissolved in sulfuric acid' filter during daily calibrations. We do not have a sufficient explanation for why this happens, but we assume that the comparatively high ionic strength of sulfuric acid causes mixed chromium (VI) complexes to form. To compensate for this behavior, over which we have no control, filters would need to be recertified much more regularly. Another possibility would be preparing new solutions every time the spectrophotometer is chekked. As a simple alternative, we offer a liquid filter that uses 'Potassium dichromate dissolved in perchloric acid'. This type of liquid filter for checking photometric accuracy has proven itself as a reliable and very stable standard for many years. No changes in absorbance properties comparable to those of the sulfuric acid model are known for this filter. Hellma Analytics cuvettes are permanently sealed, eliminating concerns about the toxicity of perchloric acid. Furthermore, the European Pharmacopoeia states that "suitable certified reference materials" may also be used, which undoubtedly applies to our perchloric acid solvent model. This model also contains a formulation described by NIST.

6.10 Why does the weight of Potassium dichromate filters seem to change after every recertification?

Due to measurement uncertainties, measurement values may fall within a specific range. This leads to an apparent change in weight from qualification to qualification, as the initial weight is calculated directly from the measured absorbance values. Earlier versions of regulatory codes stipulated that filters for checking photometric accuracy had to contain 60.06 mg/l Potassium dichromate, and allowed a tolerance of 0.01 Abs. More current versions of the European Pharmacopoeia have replaced this very strict provision, now accepting weights between 57.0 mg/l and 63.0 mg/l. The specific absorbance calculated (see European Pharmacopoeia, chapter 2.2.25) is now stated with a margin of tolerance.

6.11 Why are these peaks measured for certifying holmium glass and didymium glass filters?

Measurement errors are low in medium to high transmittance ranges. As a result, peaks in the range from 0 Abs to 1.0 Abs (corresponds to 100% -T to 10% T) are preferred for certification.

6.12 How do I calculate my measurement uncertainty?

The measurement uncertainties stated on the calibration certificate only refer to measurements conducted by Hellma Analytics and apply solely to the measurement conditions at the company (spectrophotometer used, environmental influences such as temperature, air humidity, user influence, reference materials used, etc.). The smallest possible measurement uncertainty that can be achieved by the user can then be derived by statistically combining the measurement uncertainty stated on the calibration certificate with all the user's uncertainty contributions, such as the wavelength scale tolerance of the spectrophotometer used and other influences on measurement accuracy (environmental factors such as temperature, air humidity, user influence, etc.).

EXAMPLE OF CALCULATING STANDARD MEASUREMENT UNCERTAINTY FOR A NEUTRAL DENSITY GLASS FILTER (HIGHLY SIMPLIFIED): THE CALIBRATION CERTIFICATE STATES THE FOLLOWING MEASUREMENT VALUES AND MEASUREMENT UNCERTAINTIES:

ARTICLE NO.	3524	OPTISCHE DICHTE (Abs) Optical Density (Abs)				
		440 nm	465 nm	546.1 nm	590 nm	635 nm
MEASURED VALUE	666-F2	0.2542 ± 0.0024	0.2254 ± 0.0024	0.2254 ± 0.0024	0.2415 ± 0.0024	0.2416 ± 0.0024

Here, a wavelength of 440 nm produces the following parameters:

Target measurement value (x_s) : 0.2542 Abs
Expanded measurement uncertainty: +/- 0.0024 Abs (coverage factor k=2)
Standard measurement uncertainty (x_a): +/- 0.0012 Abs

Next, you must calculate the measuring error specific to your spectrophotometer (xb) – refer to the operating instructions for more details – and define a value for the measuring error due to environmental influences at your company (xu) (such as temperature and air humidity).

Example of measuring error parameters:
Spectrophotometer (x_b): +/- 0.01 Abs
Environmental influences (x_u): +/- 0.001 Abs
Calculating standard measurement uncertainty (MU):
$MU = \sqrt{x_a^2 + x_b^2 + x_u^2} = 0.0101$
Expanded measurement uncertainty is calculated by multiplying this value by coverage factor k.

As shown here, in practice it is often easier to simply add up uncertainty contributions than to combine them statistically. However, the method used to determine measurement uncertainty depends on the specifications of your quality system and your measurement accuracy requirements. For further literature on correctly calculating measurement uncertainty, please refer to the recommendations for further reading in chapter 8 of this user manual.

6.13 Control chart?

see page 41

7. GLOSSARY

Abbreviations:

A	absorbance
ASTM	American Society for Testing and Materials
BG	Specific term for Schott glass
DAB	Deutsches Arzneibuch (German Pharmacopoeia)
DAkkS	Deutsche Akkreditierungsstelle (National accreditation body for the Federal Republic of Germany)
DAR	Deutscher Akkreditierungsrat (German accreditation body)
DKD	Deutscher Kalibrierdienst (German calibration body)
Ph. Eur.	European Pharmacopoeia
FAQs	Frequently asked questions
GLP	Good laboratory practice
GMP	Good manufacturing practice
I	Intensity of light beam
I0	Original intensity of light beam
k	Coverage factor for measurement uncertainty
λmax	Maximum peak at defined wavelength
λmin	Minimum peak at defined wavelength
NG	Neutral density glass
NIR	Near-infrared
NIST	National Institute of Standards and Technology
PTB	Physikalisch-Technische Bundesanstalt (Germany's national metrology institute)
SRM®	Standard Reference Material (registered trademark of NIST)
USP	United States Pharmacopeia
UV	Ultraviolet (wavelength range 200 – 380 nm)
Vis	Visible (visible wavelength range 380 – 780 nm)

Absorbance (Abs):
When light falls on or passes through a sample, the quantity of absorbed light is equal to the difference between the original intensity I0 and the intensity I after interaction with the sample. This is because part of the irradiated light is transferred to the molecules, causing the beam to have a smaller output when it exits the sample. The extent to which light is absorbed is determined by the principles of the Beer-Lambert law. The amount of absorbed light can be expressed as transmittance (see definition) or absorbance. Absorbance is defined as: Abs = -logT. According to the relevant standard, this parameter is referred to as spectral optical density on transmittance ("optical density").

Optical density: see absorbance

Visible range: Part of the optical spectrum that stretches from 380 nm to 780 nm of the wavelength range of electromagnetic radiation. This range is generally referred to as light. This is the only range in which the human eye can 'see' electromagnetic radiation.

Spectral resolution: This refers to a measuring system's ability to separate individual wavelength ranges.

Spectral bandwidth: Wavelength range that appears with a continuum at the exit slit when the monochromator is exposed to irradiation. Spectral bandwidth is determined by the bandwidth of emitted radiation where the light has reached half the maximum intensity.

Spectral optical density on transmittance: see absorbance

Transmittance (T): When light falls on or passes through a sample, the quantity of absorbed light is equal to the difference between the original intensity I0 and the intensity I after interaction with the sample. This is because part of the irradiated light is transferred to the molecules, causing the beam to have a smaller output when it exits the sample. The extent to which light is absorbed is determined by the principles of the Beer-Lambert law. The amount of absorbed light can be expressed as transmittance (see definition) or absorbance. Transmittance is normally expressed as a fraction of 1 or as a percentage, and is defined as follows: T = I/I0 or %T = (I/I0) * 100.

Ultraviolet range (UV range): Also known as UV radiation, this is the short-wave part of the optical radiation spectrum. UV radiation has a wavelength range of 200 nm to 380 nm.

Wavelength: Wavelength is the distance between two identical, adjacent corresponding points of the same wave phase at a certain point in time.

8. LITERATURE REFERENCES

Standards and Best Practice in Absorption Spectrometry; Edited by C. Burgess and T. Frost UVSG, ISBN 0-632-05313-5 Blackwell Service

Qualitätssicherung in der Analytischen Chemie; Werner Funk, Vera Dammann, Gerhild Donnevert;
ISBN-10: 3-527-31112-2; Verlag: WILEY-VCH

ISO/IEC Guide 98-3:2008; **Evaluation of measurement data – Guide to the expression of uncertainty in measurement**

NIST Special Publication 260-54 Standard Reference Materials: **Certification and Use of Acidic Potassium Dichromate Solutions as an Ultraviolet Absorbance Standard – SRM 935**

NIST Special Publication 260-116 Standard Reference Materials: **Glass Filters as a Standard Reference Material for Spectrophotometry – Selection, Preparation, Certification, and Use of SRM 930 and SRM 1930**

NIST Special Publication 260-102: Standard Reference Materials: **Holmium Oxide Solution Wavelength Standard from 240 to 640 nm – SRM 2034**

European Pharmacopoeia (Ph.Eur.)

DKD3

United States Pharmacopeia (USP)

9. PRODUCT OVERVIEW

LIQUID FILTERS WITH DAKKS CERTIFICATE

TYPE	MATERIAL	WAVELENGTH nm	ARTICLE NO.
Glass Filters for testing the wavelength accuracy			
666-F1	Holmium Glass Filter F1	279; 361; 453; 536; 638	666F1-339
666-F7W	Didymium Glass Filter F7W	329; 472; 512; 681; 875	666F7W-323
Glass Filters for testing the photometric accuracy			
666-F390	Neutral Density Glass Filter F390; 0.04 Abs	440; 465; 546.1; 590; 635	666F390-25
666-F2	Neutral Density Glass Filter F2; 0.25 Abs	440; 465; 546.1; 590; 635	666F2-39
666-F201	Neutral Density Glass Filter F201; 0.3 Abs	440; 465; 546.1; 590; 635	666F201-39
666-F3	Neutral Density Glass Filter F3; 0.5 Abs	440; 465; 546.1; 590; 635	666F3-38
666-F204	Neutral Density Glass Filter F204; 0.7 Abs	440; 465; 546.1; 590; 635	666F204-37
666-F4	Neutral Density Glass Filter F4; 1.0 Abs	440; 465; 546.1; 590; 635	666F4-37
666-F202	Neutral Density Glass Filter F202; 1.5 Abs	440; 465; 546.1; 590; 635	666F202-36
666-F203	Neutral Density Glass Filter F203; 2.0 Abs	440; 465; 546.1; 590; 635	666F203-36
666-F301	Neutral Density Glass Filter F301; 2.5 Abs	440; 465; 546.1; 590; 635	666F301-361
666-F303	Neutral Density Glass Filter F303; 3.0 Abs	440; 465; 546.1; 590; 635	666F303-361
666-F7A	Didymium Glass Filter F7A; ca. 0.5 – 1.0 Abs	270; 280; 297; 321; 342	666F7A-323
Glass Filter for testing the photometric accuracy and wavelength accuracy			
666-F7	Didymium Glass Filter F7	A: 270; 280; 297; 321; 342 W: 329; 472; 512; 681; 875	666F7-323
Empty filter mount			
666-F0	Reference filter frame made of aluminum (without glass)		666F0-71
Sets for testing t he photometric accuracy and wavelength accuracy			
666-S000	Complete Glass Filter Set: F1, F2, F3, F4, F0 (Abs: 0.25; 0.5; 1.0)	A: 440; 465; 546.1; 590; 635 W: 279; 361; 453; 536; 638	666S000
666-S001	Glass Filter Set: F3, F4, F7 (Abs: 0.5; 1.0; F7: ca. 0.5 – 1.0)	A (F7): 270; 280; 297; 321; 342 A (F3, F4): 440; 465; 546.1; 590; 635 W (F7): 329; 472; 512; 681; 875	666S001
666-S002	Glass Filter Set: F2, F3, F4 (Abs: 0.25; 0.5; 1.0)	A: 440; 465; 546.1; 590; 635	666S002
666-S003	Glass Filter Set: F1, F2, F3, F4, F7; (Abs: 0.25; 0.5; 1.0; F7: ca. 0.5 – 1.0)	A (F7): 270; 280; 297; 321; 342; A (F2, F3, F4): 440; 465; 546.1; 590; 635 W (F1): 279; 361; 453; 536; 638 W (F7): 329; 472; 512; 681; 875	666S003
666-S004	Glass Filter Set: F201, F202, F203, F0 (Abs: 0.3; 1.5; 2.0)	A: 440; 465; 546.1; 590; 635	666S004
666-S005	Glass Filter Set: F0, F1, F3, F4; (Abs: 0.5; 1.0)	A: 440; 465; 546.1; 590; 635 W: 279; 361; 453; 536; 638	666S005
666-S006	Glass Filter Set: F0, F2, F3, F4; (Abs: 0.25; 0.5; 1.0)	A: 440; 465; 546.1; 590; 635	666S006
666-S300	Glass Filter Set: F390, F301, F303 (Abs: 0.04; 2.5; 3.0)	A: 440; 465; 546.1; 590; 635	666S300

A: Wavelength for absorbance W: Wavelength for wavelength accuracy

LIQUID FILTERS WITH DAKKS CERTIFICATE

TYPE	MATERIAL	WAVELENGTH nm	ARTICLE NO.
Liquid Filters for testing the photometric accuracy			
667-UV20	20 mg/l potassium dichromate in $HClO_4$ (0.1 – 0.3 Abs)	235; 257; 313; 350	667020
667-UV40	40 mg/l potassium dichromate in $HClO_4$ (0.2 – 0.6 Abs)	235; 257; 313; 350	667040
667-UV60	60 mg/l potassium dichromate in $HClO_4$ (0.3 – 0.9 Abs)	235; 257; 313; 350	667060
667-UV80	80 mg/l potassium dichromate in $HClO_4$ (0.4 – 1.2 Abs)	235; 257; 313; 350	667080
667-UV0100	100 mg/l potassium dichromate in $HClO_4$ (0.5 – 1.45 Abs)	235; 257; 313; 350	6670100
667-UV0120	120 mg/l potassium dichromate in $HClO_4$ (0.6 – 1.7 Abs)	235; 257; 313; 350	6670120
667-UV0140	140 mg/l potassium dichromate in $HClO_4$ (0.7 – 2.0 Abs)	235; 257; 313; 350	6670140
667-UV0160	160 mg/l potassium dichromate in $HClO_4$ (0.8 – 2.3 Abs)	235; 257; 313; 350	6670160
667-UV0180	180 mg/l potassium dichromate in $HClO_4$ (0.9 – 2.6 Abs)	235; 257; 313; 350	6670180
667-UV0200	200 mg/l potassium dichromate in $HClO_4$ (1.0 – 3.0 Abs)	235; 257; 313; 350	6670200
667-UV600	600 mg/l potassium dichromate in $HClO_4$ (1.0 Abs)	430	667600
667-UV14	Perchloric acid (reference filter)	235; 257; 313; 350	667014
667-UV301	Filter Set for UV-range: UV60, UV14	235; 257; 313; 350	667301
667-UV304	Filter Set for Vis-range: UV600, UV14	430	667304
667-UV305	Filter Set for UV/Vis-range: UV60, UV600, UV14	235; 257; 313; 350; 430	667305
Liquid Filter Set for testing the linearity of the absorption			
667-UV307	Filter Set: UV20, UV40, UV60, UV80, UV0100, UV14	235; 257; 313; 350	667307
Niacin Liquid Filters for testing the photometric accuracy			
667-UV51 NEW	6 mg/l Niacin in HCl (0.25 Abs)	213; 261	667051
667-UV52 NEW	12 mg/l Niacin in HCl (0.5 Abs)	213; 261	667052
667-UV53 NEW	18 mg/l Niacin in HCl (0.75 Abs)	213; 261	667053
667-UV54 NEW	24 mg/l Niacin in HCl (1.0 Abs)	213; 261	667054
667-UV59 NEW	Reference filter (HCl)	213; 261	667059
667-UV350 NEW	Filter Set: UV51, UV52, UV53, UV54, UV59	213; 261	667350
Liquid Filters for testing the wavelength accuracy			
667-UV5	Holmium in perchloric acid	241; 287; 361; 536; 640	667005
667-UV400	Filter Set: UV5, UV14	241; 287; 361; 536; 640	667400
667-UV25	Didymium in perchloric acid	329; 469; 575; 740; 864	667025
667-UV35	Rare Earth	201; 211; 222; 239; 252	667035
667-UV45	Holmium/Didymium in perchloric acid	241; 354; 444; 575; 641; 740; 864	667045

A: Wavelength for absorbance W: Wavelength for wavelength accuracy S: Wavelength for stray light R: Wavelength for spectral resolution
*with Hellma Analytics calibration certificate

9. PRODUCT OVERVIEW

LIQUID FILTERS WITH DAKKS CERTIFICATE

TYPE	MATERIAL	WAVELENGTH nm	ARTICLE NO.
Liquid Filters for testing the wavelength accuracy acc. to USP <857>			
667-UV5USP	Holmium in perchloric acid	241; 250; 278; 287; 333; 345; 361; 385; 416; 452; 468; 485; 536; 640	667005USP
667-UV25USP	Didymium in perchloric acid	732, 740, 794, 801, 864	667025USP
667-UV425	Filter Set: UV5, UV25	UV5: 241; 250; 278; 287; 333; 345; 361; 385; 416; 452; 468; 485; 536; 640 UV25: 732; 740; 794; 801; 864	667425
Liquid Filters for testing stray light			
667-UV1	Potassium chloride in pure water, LP 10 mm	200 (cut-off)	667001
667-UV1H*	Potassium chloride in pure water, reference filter, LP 5 mm	200 (cut-off)	667001H
667-UV10	Sodium iodide in pure water, LP 10 mm	259 (cut-off)	667010
667-UV10H*	Sodium iodide in pure water, reference filter, LP 5 mm	259 (cut-off)	667010H
667-UV11	Sodium nitrite in pure water, LP 10 mm	385 (cut-off)	667011
667-UV11H*	Sodium nitrite in pure water, reference filter, LP 5 mm	385 (cut-off)	667011H
667-UV12	Pure water, reference filter, LP 10 mm	198; 200; 300; 400	667012
667-UV19	Acetone, LP 10 mm	325 (cut-off)	667019
667-UV19H*	Acetone, reference filter, LP 5 mm	325 (cut-off)	667019H
Liquid Filter Sets for testing stray light according to Ph. Eur.			
667-UV100	Filter Set: UV1, UV12; LP 10 mm	200 (cut-off)	667100
667-UV101	Filter Set: UV10, UV12; LP 10 mm	259 (cut-off)	667101
667-UV102	Filter Set: UV11, UV12; LP 10 mm	385 (cut-off)	667102
667-UV103	Filter Set: UV1, UV10, UV11, UV12; LP 10 mm	200; 259; 385 (cut-off)	667103
667-UV104	Filter Set: UV10, UV11, UV12; LP 10 mm	259; 385 (cut-off)	667104
Liquid Filter Sets for testing stray light according to USP <857>			
667-UV100H	Filter Set: UV1, UV1H, LP 10 and 5 mm	200 (cut-off); SR: 190 – 205	667100H
667-UV101H	Filter Set: UV10, UV10H, LP 10 and 5 mm	259 (cut-off); SR: 210 – 259	667101H
667-UV102H	Filter Set: UV11, UV11H, LP 10 and 5 mm	385 (cut-off); SR: 300 – 386	667102H
667-UV119H	Filter Set: UV19, UV19H, LP 10 and 5 mm	325 (cut-off); SR: 250 – 324	667119H
667-UV105H	Filter Set: UV1/UV1H; UV10/UV10H; UV11/UV11H; UV19/UV19H, LP 10 and 5 mm	200, 259, 325, 385 (cut-off)	667105H
667-UV106H	Filter Set: UV1/UV1H; UV10/UV10H; UV19/UV19H; LP 10 mm and 5 mm	200; 259; 325 (cut-off)	667106H
Liquid Filters for testing the resolution			
667-UV6*	Toluene in hexane	Scan: 265 – 270	667006
667-UV9*	hexane (Reference Filter)	Scan: 265 – 270	667009
667-UV200*	Filter Set: UV6, UV9	Scan: 265 – 270 Slit widths: 0.5; 1.0; 1.5; 2.0; 3.0	667200

A: Wavelength for absorbance W: Wavelength for wavelength accuracy S: Wavelength for stray light R: Wavelength for spectral resolution
*with Hellma Analytics calibration certificate

FILTER SETS ACC. TO PH.EUR AND USP <857> WITH DAkkS CERTIFICATE

TYPE	CONTENT	WAVELENGTH nm	ARTICLE NO.
Complete Filter Set for testing the spectrophotometer according to Ph.Eur.			
667-UV003	Potassium dichromate filters: UV60/ UV600/UV 14 (Abs: 0.3 – 0.9; 1.0) Holmium liquid filter: UV5 Potassium chloride in H_2O: UV1/UV12 Toluene in hexane: UV6/UV9	A: 235; 257; 313; 350; 430 W: 241; 287; 361; 536; 640 S: 200 (cut-off) R: Scan 265 – 270	667003
Basic set for testing the spectrophotometer according to United States Pharmacopoeia (USP <857>)			
667-UV857 **NEU** Depending on the application, this basic set can be supplemented with further USP filters. More information: www.hellma-analytics.com/USP857	Neutral density glass filters: F2, F3, F4 (Abs: 0.25; 0.5; 1.0) Potassium dichromate 60mg/L: UV60/UV14 (Abs: 0.3 – 0.9) Holmium and Didymium liquid filter: UV5/UV25 Sodium nitrite in H_2O: UV11/UV11H PL: 10 mm and 5 mm Toluene in hexane: UV6/UV9	A (F2,F3,F4): 440; 465; 546.1; 590; 635 A (UV60/14): 235; 257; 313; 350 W (UV5): 241; 250; 278; 287; 333; 345; 361; 385; 416; 452; 468; 485; 536; 640 W (UV25): 732; 740; 794; 801; 864 S (UV11/11H): 385 (cut-off), R (UV6/9): Scan: 265 – 270	667857

A: Wavelength for absorbance W: Wavelength for wavelength accuracy S: Wavelength for stray light R: Wavelength for spectral resolution PL: Path length

REFERENCE PLATES FOR QUALIFYING MICROPLATE READERS WITH DAKKS CERTIFICATE

With reference plates from Hellma Analytics you can check the photometric and wavelength accuracy of microplate readers. They have the same dimensions as a microplate with 96 wells and a 6.6 mm diameter per window (height 14.5 x width 125 x length 85.5 mm).

TYPE	USAGE	MATERIAL Nominal value of absorption (Abs.)	WAVELENGTH nm	ARTICLE NO.
666-R013	To check photometric accuracy	Neutral density glass Filter NG 11 (0.25), NG 5 (0.5), NG 4 (1.0), NG 3 (1.5), (2.5)	A: 405; 450; 490; 650	666R013
666-R113	To check photometric accuracy and wavelength accuracy	Neutral density glass Filter NG 5 (0.5), NG 4 (1.0), NG 3 (1.5), (2.0) Holmium Glass Filter	A: 405; 450; 490; 650 W: 279; 361; 453; 536; 638	666R113

A: Wavelength for absorbance W: Wavelength for wavelength accuracy

Hellma Analytics
High Precision in Spectro-Optics

ASIA

Hellma Asia Pte Ltd
1 Commonwealth Lane
#09-33 One Commonwealth
Singapore 149544
phone +65 6397 4138
fax +65 6397 4139
info.asia@hellma.com

EUROPE

Hellma GmbH & Co. KG
Klosterrunsstraße 5
79379 Müllheim
Germany
phone + 49 7631 182 1010
fax + 49 7631 182 1011
info.de@hellma.com

Hellma Benelux BVBA
Hogen Akkerhoekstraat 14
9150 Kruibeke
Belgium
phone + 32 3 877 33 27
fax + 32 3 887 10 26
info.be@hellma.com

Hellma France S.A.R.L.
35 rue de Meaux
75019 Paris
France
phone + 33 1 42 08 01 28
fax + 33 1 42 08 13 65
info.fr@hellma.com

Hellma Italia S.r.l.
Via Gioacchino Murat, 84
20159 Milano
Italy
phone + 39 02 261 164 19
fax + 39 02 261 133 31
info.it@hellma.com

Hellma Schweiz AG
Schwäntenmos 15
8126 Zumikon
Switzerland
phone + 41 44 918 23 79
fax + 41 44 918 08 12
info.ch@hellma.com

Hellma UK LTD
Cumberland House
24-28 Baxter Avenue
Southend on Sea,
Essex SS2 6HZ
United Kingdom
phone + 44 1702 335 266
fax + 44 1702 430 652
info.uk@hellma.com

THE AMERICAS

Hellma USA INC.
80 Skyline Drive
Plainview, NY 11803
USA
phone + 1 516 939 0888
fax + 1 516 939 0555
info.us@hellma.com

Hellma Canada Ltd.
7321 Victoria Park Avenue,
Unit 108
Markham, Ontario L3R 2Z8
Canada
phone + 1 905 604 5013
fax + 1 905 604 5015
info.ca@hellma.com

For all other countries, please see our list of representatives and authorized dealers:
www.hellma-analytics.com/contacts

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