UV/VIS Spectrophotometers

Calibration Guide

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Calibration set EMC-SET-100

Glass filter set EMC-SET-100 contains:

Wavelength Accuracy:
H1 Holmiumoxide filter

Photometric Accuracy:
N2 Neutral density glass filter 0.25 Abs
N3 Neutral density glass filter 0.5 Abs
N4 Neutral density glass filter 1.0 Abs
N0 Blank filter

Calibration Certificate
Microfiber cloth
Calibration label (2 pcs.)


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Secondary Spectrometric Calibration Standards
Certificate of Calibration - No.: H11N21N31N41

Calibration of Wavelength Accuracy

Calibration of Photometric Accuracy

Conditions of Calibration

Date
30 March 2016

General Information

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 plastic frames. 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. The plastic frames should not come into contact with acids or alkalis. When inserting the filter into the spectrophotometer, no pull, pressure or lever should act on the holder.

Storage

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

Cleaning

Dirt often accumulates on optical surfaces as a result of regular use. This is best removed using a lint-free cloth and alcohol (Attention: Never clean the plastic frame with alkaline concentrate solution!)

Influence of temperature and humidity on measurements

Temperature has only an exceedingly small influence on the certified measurement values, and temperatures between 20°C and 24°C fall within the measurement uncertainty stated on the calibration certificate. Measurements should therefore be performed within this range to avoid any potential temperature influence on the results to a minimum. Take care that the relative humidity is between 30% to 65%, especially when measuring within UV range.

Other factors that may influence the 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.

Steps to take before performing calibration

- Switch on and preheat the instrument for at least 20 minutes
- Only use a stable cell holder for 10 mm standard cells. Make sure that the holder is secure and stable in the sample compartment.
- Carry out a wavelength reset and dark current.
- Carry out a baseline correction with an empty sample compartment.
- Make sure all filters are always clean before using. Otherwise, please use the provided microfiber cloth for purifying.
- Check that the filter is correctly positioned in the light path by first placing the blank filter N0 in the cell holder. The N0 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 detector.
- Choose the slit width the filters are calibrated at (1 nm for EMC-SET-100) or the slit width that is closest. If you cannot adjust the slit width of the instrument, note that slight deviations to the values recorded in the calibration may appear. (recommended rough guide: double uncertainty if slit width differs from certified slit width)
- Carry out measurement of the filters just as carefully as you would carry out a sample measurement in a closed sample compartment (opening the sample compartment will cause incorrect results).

Neutral Density Glass Filter for checking Photometric Accuracy

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. The present Neutral Density Glass Filters are made from special selected filter materials. They show a relatively constant transmittance within the wavelength range of 405 nm to 800 nm, therefore they are an excellent choice to check photometric accuracy in the visible wavelength range (> 405 nm).

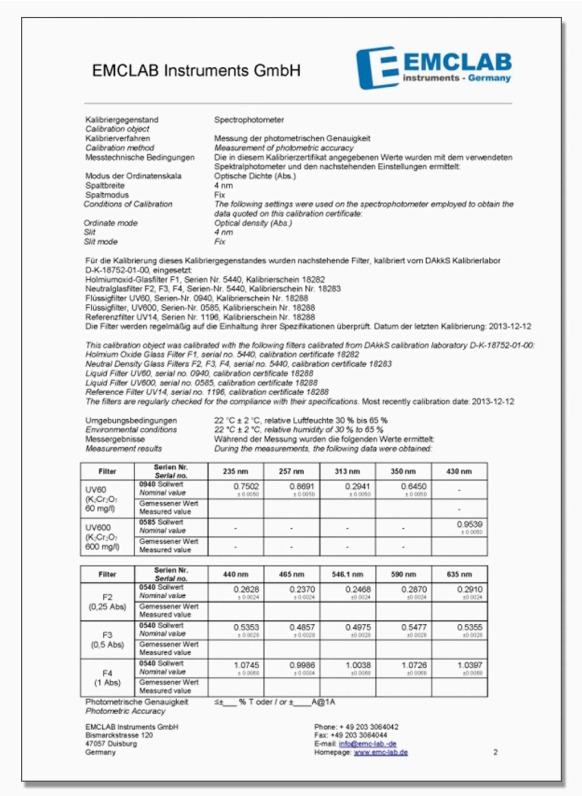
Type	N2, N3, N4, Neutral Density Glass Filter
Application	Testing the photometric accuracy in the VIS range
Content	N2: Neutral Density Glass Filter (0,25 Abs), N3: Neutral Density Glass Filter (0,5 Abs), N4: Neutral Density Glass Filter (1,0 Abs),
Standard Certification	Wavelengths: 440, 465, 546.1, 590, 635 nm Slit width: 1 nm

Preparation for calibration

Holmium Glass Filter for checking Wavelength Accuracy

To measure wavelength accuracy, the filter absorbs the light beam of the spectrophotometer to a greater extent at certain wavelengths creating absorbance peaks. The Holmium Glass Filter is an excellent choice for checking the wavelength scale of a spectrophotometer since it has a range of narrow, well defined peaks in the UV and visible range.

Typ	H1, Holmiumoxide Glass Filter
Application	Testing the wavelength accuracy in the UV and VIS range
Standard Certification	Wavelength accuracy at 279 nm to 638 nm Slit width: 1 nm



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Kalibriergegenstand *Calibration object*	Spectrophotometer
Kalibrierverfahren *Calibration method*	Messung der photometrischen Genauigkeit *Measurement of photometric accuracy*
Messtechnische Bedingungen	Die in diesem Kalibrierzertifikat angegebenen Werte wurden mit dem verwendeten Spektralphotometer und den nachstehenden Einstellungen ermittelt:
Modus der Ordinatenskala	Optische Dichte (Abs.)
Spaltbreite	4 nm
Spaltmodus	Fix
Conditions of Calibration	*The following settings were used on the spectrophotometer employed to obtain the data quoted on this calibration certificate:*
Ordinate mode	*Optical density (Abs.)*
Slit	*4 nm*
Slit mode	*Fix*

Für die Kalibrierung dieses Kalibriergegenstandes wurden nachstehende Filter, kalibriert vom DAkkS Kalibrierlabor D-K-18752-01-00, eingesetzt:
Holmiumoxid-Glasfilter F1, Serien Nr. 5440, Kalibrierschein 18282
Neutralglasfilter F2, F3, F4, Serien-Nr. 5440, Kalibrierschein Nr. 18283
Flüssigfilter UV60, Serien-Nr. 0940, Kalibrierschein Nr. 18288
Flüssigfilter, UV600, Serien-Nr. 0585, Kalibrierschein Nr. 18288
Referenzfilter UV14, Serien Nr. 1196, Kalibrierschein Nr. 18288
Die Filter werden regelmäßig auf die Einhaltung ihrer Spezifikationen überprüft. Datum der letzten Kalibrierung: 2013-12-12

This calibration object was calibrated with the following filters calibrated from DAkkS calibration laboratory D-K-18752-01-00:
Holmium Oxide Glass Filter F1, serial no. 5440, calibration certificate 18282
Neutral Density Glass Filters F2, F3, F4, serial no. 5440, calibration certificate 18283
Liquid Filter UV60, serial no. 0940, calibration certificate 18288
Liquid Filter UV600, serial no. 0585, calibration certificate 18288
Reference Filter UV14, serial no. 1196, calibration certificate 18288
The filters are regularly checked for the compliance with their specifications. Most recently calibration date: 2013-12-12

Umgebungsbedingungen *Environmental conditions*	22 °C ± 2 °C, relative Luftfeuchte 30 % bis 65 % *22 °C ± 2 °C, relative humidity of 30 % to 65 %*
Messergebnisse *Measurement results*	Während der Messung wurden die folgenden Werte ermittelt: *During the measurements, the following data were obtained:*

Filter	Serien Nr. *Serial no.*	235 nm	257 nm	313 nm	350 nm	430 nm
UV60 ($K_2Cr_2O_7$ 60 mg/l)	0940 Sollwert *Nominal value*	0.7502 ± 0.0050	0.8691 ± 0.0050	0.2941 ± 0.0050	0.6450 ± 0.0050	-
	Gemessener Wert Measured value					-
UV600 ($K_2Cr_2O_7$ 600 mg/l)	0585 Sollwert *Nominal value*	-	-	-	-	0.9539 ± 0.0050
	Gemessener Wert Measured value	-	-	-	-	

Filter	Serien Nr. *Serial no.*	440 nm	465 nm	546.1 nm	590 nm	635 nm
F2 (0,25 Abs)	0540 Sollwert *Nominal value*	0.2828 ± 0.0024	0.2370 ± 0.0024	0.2468 ±0.0024	0.2870 ±0.0024	0.2910 ±0.0024
	Gemessener Wert Measured value					
F3 (0,5 Abs)	0540 Sollwert *Nominal value*	0.5353 ± 0.0028	0.4857 ± 0.0028	0.4975 ±0.0028	0.5477 ±0.0028	0.5355 ±0.0028
	Gemessener Wert Measured value					
F4 (1 Abs)	0540 Sollwert *Nominal value*	1.0745 ± 0.0050	0.9986 ± 0.0004	1.0038 ±0.0050	1.0726 ±0.0050	1.0397 ±0.0050
	Gemessener Wert Measured value					

Photometrische Genauigkeit ≤±___ % T oder / or ±___A@1A
Photometric Accuracy

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Fig. 1 Calibration Certificate (sample) - Photometric Accuracy

1. Photometric Accuracy Checking

Filters:	**0.25 Abs, 0.5 Abs, 1.0 Abs neutral density glass filters N2, N3, N4**
Test mode:	**Photometric**
Reference setting:	**Testing wavelengths 440, 465, 546.1, 590, 635 nm**
Display mode:	**A**

1. First of all, follow the steps to take before performing calibration (page 4)
2. Set the display mode to absorbance (Abs).
3. Run the wavelength selection program on your spectrophotometer, observing the guidelines in the user manual. Select the wavelengths provided on the Calibration Certificate (see above reference setting).
4. Insert blank filter N0 as reference and carry out blank (100%T/0Abs).
5. Insert the neutral density filters N2, N3, N4. Ensure that the filters are inserted into the holder as far as possible. The N2, N3, N4 marking must be visible from above (serial number facing the detector).
6. Start the program for measuring the absorbance values at the aforementioned wavelengths stated on the calibration certificate. Record the values. Take several measurements (we recommend at least 3) and average the measured values to avoid errors.
7. Compare the measured values with the standard values in the calibration certificate.
8. Repeat the steps 1 to 7 for every given wavelength in the calibration certificate.

If you are using the PC software PROFESSIONAL, Easy UV or ANALYST, you can carry out the calibration and save results automatically by executing the “Multi Wavelength Scan” application.

Testing and calibration

2. Wavelength Accuracy Checking

Fig. 2

Filters:	**Holmiumoxide filter H1 (Fig. 2)**
Test mode:	**Spectrum Scan**
Reference setting:	**Peak wavelengths* see calibration certificate**
Scanning interval:	**0.5 nm**
Display mode:	**A**

1. First of all, follow the **steps to take before performing calibration.**
2. Set the display mode to absorbance (Abs).
3. Run the scan program on your spectrophotometer, observing the guidelines in the user manual. Select a scanning range that covers all the peaks listed on the Calibration Certificate (see above reference setting). If you cannot set a range, please follow the steps below for manual measurement.
4. Adjust your spectrophotometer to the measurement parameters that appear on the calibration certificate. Select a slow scanning speed and a small data interval.
5. Measurements are taken using an air blank which means reference measurement is taken using the blank filter N0.
6. Insert the Holmiumoxide filter H1. Ensure that the filter is inserted into the holder as far as it will go. The H1 marking must be visible from above (serial number facing the detector).
7. Start the measurement.
8. Take several measurements (we recommend at least 3) and average the measured values to avoid errors.
9. Compare the positions of the peaks to the wavelengths stated on the calibration certificate.

If the scan program is not applicable you can do it manually:
e.g. peak 360,90:

1. Set the wavelength to 355 nm, insert blank filter N0 as reference and carry out blank (100%T/0Abs).
2. Insert Holmiumoxide filter H1. Ensure that the filter is inserted into the holder as far as it will go. The H1 marking must be visible from above (serial number facing the detector). Record the value.
3. Set the wavelength to the next higher value (+0.5 nm), repeat step 1 and 2 get the value during 355 nm to 366 nm.
4. Compare the measurement values with the certified ones.
5. Repeat steps 1 to 4 for every given peak position from the calibration certificate.

Option 1: Wavelength scan with the On-board software (not applicable for all instruments)
Option 2: Wavelength scan with PC software PROFESSIONAL, Easy UV or ANALYST

*Wavelength 279 nm is within the UV range and not working in the VIS range

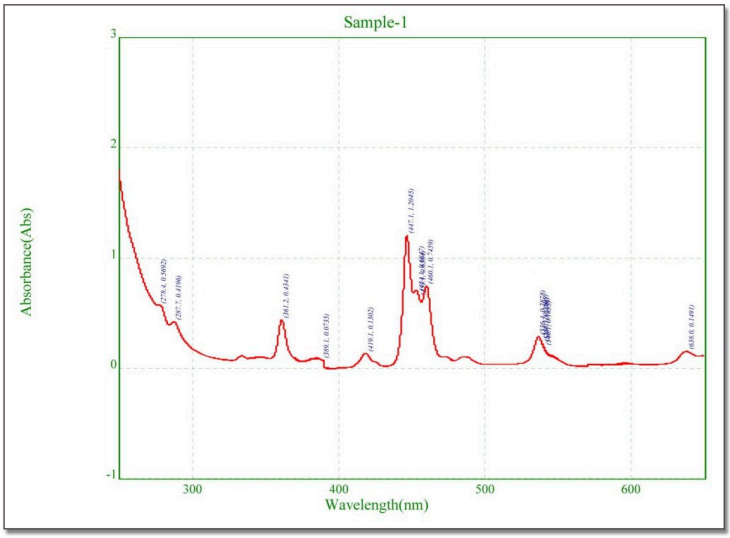


Fig. 3 Wavelength scan with Holmiumoxide filter H1 (ANALYST PC Software)

3. Stray Light Checking (filter optionally available**)

Only for spectrophotometers that can measure in the UV range!

Filters:	**UV1, UV12 (reference filter) (Fig. 4)**
Test mode:	**Spectrum Scan**
Setting:	**250 nm to 200 nm**
Display mode:	**A**

1. First of all, follow the **steps to take before performing calibration.**
2. Set the display mode to absorbance (Abs).
3. Run the scan program on your spectrophotometer, observing the guidelines in the user manual. Select the scanning range according to the setting above (250 nm to 200 nm).
4. Select a slow scanning speed and a small data interval.

Testing and calibration

5. Measurements are taken using a pure water blank which means reference measurement is taken using the filter UV12. Ensure that the filter is inserted into the holder as far as possible. Do a blank scan to set the instrument to zero.
6. Insert the potassium chloride filter UV1. Ensure that the filter is inserted into the holder as far as it will go.
7. Start the measurement.
8. Take several measurements (we recommend at least 3) to avoid errors.
9. Compare the curve shape to the characteristic curve (Fig. 5).

**Optionally available DAkkS and NIST certified reference filters:
UV1 (Potassium chloride in pure water) + UV12 (pure water (H2O) reference filter)

Fig. 4 UV1 (KCl) UV12 (H_2O)

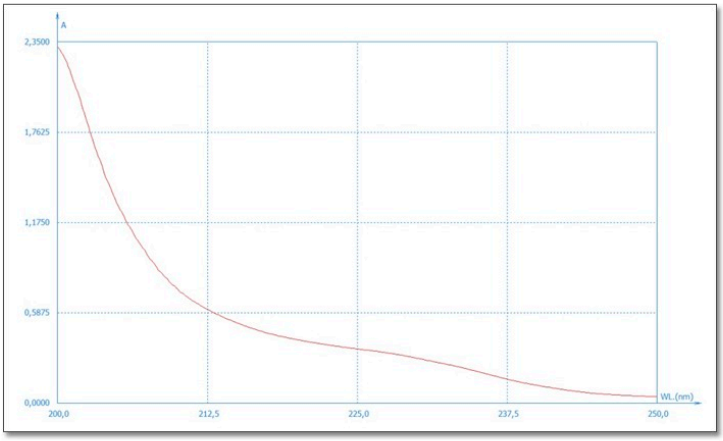


Fig. 5 Characteristic Curve Shape

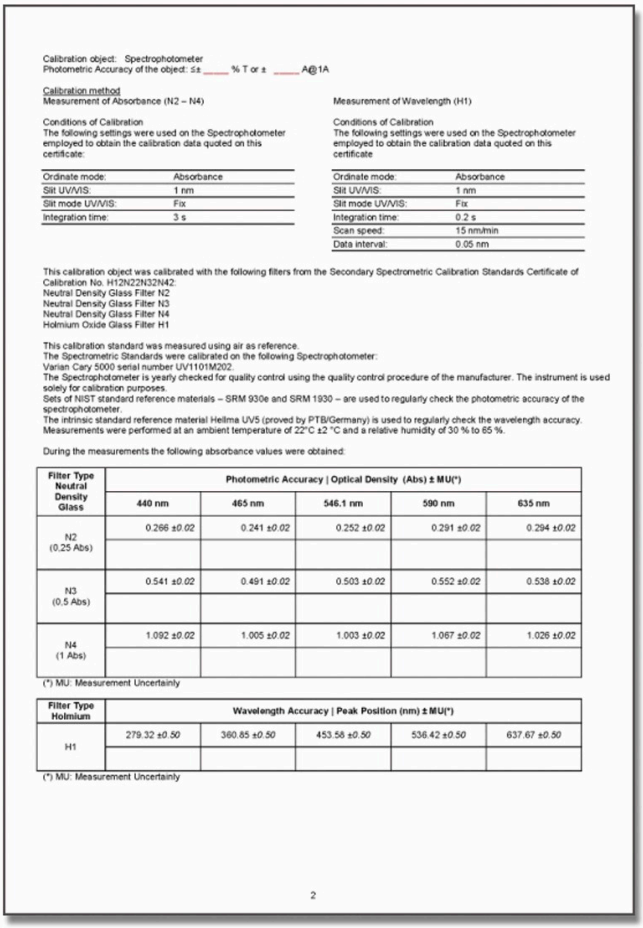
4. Recertification

As it is the case for all measuring devices, the reference materials used to verify spectrophotometers must also be checked and recertified at regular intervals (see for example ISO 9001:2015 “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

The length of intervals between the recertification of reference materials depends on how frequently materials are used, the wear associated with this, accuracy requirements, as well as the company’s internal auditing requirements. Therefore, only users themselves can determine these.

5. Works Calibration Form

Together with the EMC-SET-100, EMCLAB Instruments provides a blank calibration form to enable the user to issue a works calibration certificate. To duplicate the calibration form or to issue a calibration certificate with printed instead of handwritten values, you can contact us to get the form as a file by Email.



Calibration object: Spectrophotometer
Photometric Accuracy of the object: ≤± _____ % T or ± _____ A@1A

Calibration method
Measurement of Absorbance (N2 – N4)

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

Ordinate mode:	Absorbance
Slit UV/VIS:	1 nm
Slit mode UV/VIS:	Fix
Integration time:	3 s

Measurement of Wavelength (H1)

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

Ordinate mode:	Absorbance
Slit UV/VIS:	1 nm
Slit mode UV/VIS:	Fix
Integration time:	0.2 s
Scan speed:	15 nm/min
Data interval:	0.05 nm

This calibration object was calibrated with the following filters from the Secondary Spectrometric Calibration Standards Certificate of Calibration No. H12N22N32N42:
Neutral Density Glass Filter N2
Neutral Density Glass Filter N3
Neutral Density Glass Filter N4
Holmium Oxide Glass Filter H1

This calibration standard was measured using air as reference.
The Spectrometric Standards were calibrated on the following Spectrophotometer:
Varian Cary 5000 serial number UV1101M202.
The Spectrophotometer is yearly checked for quality control using the quality control procedure of the manufacturer. The instrument is used solely for calibration purposes.
Sets of NIST standard reference materials – SRM 930e and SRM 1930 – are used to regularly check the photometric accuracy of the spectrophotometer.
The intrinsic standard reference material Hellma UV5 (proved by PTB/Germany) is used to regularly check the wavelength accuracy.
Measurements were performed at an ambient temperature of 22°C ±2 °C and a relative humidity of 30 % to 65 %.

During the measurements the following absorbance values were obtained:

Filter Type Neutral Density Glass	Photometric Accuracy | Optical Density (Abs) ± MU(*)				
	440 nm	465 nm	546.1 nm	590 nm	635 nm
N2 (0,25 Abs)	0.266 ±0.02	0.241 ±0.02	0.252 ±0.02	0.291 ±0.02	0.294 ±0.02
N3 (0,5 Abs)	0.541 ±0.02	0.491 ±0.02	0.503 ±0.02	0.552 ±0.02	0.538 ±0.02
N4 (1 Abs)	1.092 ±0.02	1.005 ±0.02	1.003 ±0.02	1.067 ±0.02	1.026 ±0.02

(*) MU: Measurement Uncertainly

Filter Type Holmium	Wavelength Accuracy | Peak Position (nm) ± MU(*)				
H1	279.32 ±0.50	360.85 ±0.50	453.58 ±0.50	536.42 ±0.50	637.67 ±0.50

(*) MU: Measurement Uncertainly

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Fig. 6 Blank Calibration Form

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