

Application News

No. A439

Spectrophotometric Analysis

Introduction of Performance Validation Software for Improved GLP/GMP Support

Presently, validation of analytical instruments is a requirement in various regulations and standards, however, conducting instrument validation manually is a cumbersome task. This software conforms to the JIS K0115 General Rules for Molecular Absorptometric

Analysis, as well as testing stipulated by the Japanese Pharmacopoeia (JP), European Pharmacopoeia (EP), and US Pharmacopoeia (USP). Thus, performance validation tasks involving condition settings and other complex operations are greatly facilitated with this software.

■ Validation

The manufacturing control and quality control in manufacturing sites for quality management of drugs and quasi drugs is referred to as GMP (Good Manufacturing Practice), and in Japan, according to the "Ministerial Ordinance Concerning Standards for Manufacturing Control and Quality Control of Drugs and Quasi-Drugs (Ministry of Health, Labour and Welfare Ordinance No. 179, December 24, 2004), it is stated that ["Validation" means to verify and document that the buildings and facilities of the manufacturing site, procedures, processes, and other procedures of the manufacturing control and quality control (hereinafter referred to as "manufacturing procedure, etc.") provide the anticipated results.] Regarding analytical equipment, it states that "Validation refers to the periodic inspection and maintenance (including calibration) which must be conducted according to previously documented procedures, and that such records of inspection and maintenance must be retained."

The software for this validation has been incorporated as standard in the UV-2600/2700. Table 1 shows the Performance Validation Software inspection items and which standards require which items of inspection. This software supports all of the necessary items.

■ Japanese Pharmacopoeia (JP)

Spectrophotometric wavelength and transmittance testing is described in the "Apparatus and Adjustment" section of the "General Tests, Processes and Apparatus – Ultraviolet-visible Spectrophotometry" of 16th Edition of the Japanese Pharmacopoeia.

Wavelength testing is to be conducted using either a commercially available wavelength calibration optical filter or a low-pressure mercury lamp or a deuterium discharge lamp. Among these, wavelength testing is typically conducted using the 2 bright lines (486.00 nm, 656.10 nm) of the deuterium discharge lamp provided as standard with the instrument. On the other hand, in the case of the low-pressure mercury lamp, 4 bright lines are used (253.65 nm, 365.02 nm, 435.84 nm and 546.07 nm), permitting wavelength testing over a wide range, including the ultraviolet region. Thus, we used the optional low-pressure mercury lamp unit for the UV-2600/2700. Fig. 1 shows an external view of the low-pressure mercury lamp unit installed in the UV-2600.

Of these, 9 items related to "instrument performance testing" of spectrophotometers are described in the General Rules for Molecular Absorptometric Analysis. However, there is no clear reference to the range of compliance. Even in the US Pharmacopoeia, while the required inspection items are discussed, there is no clear specification of the range of compliance.

On the other hand, the required tests and compliance ranges are both specified in the Japanese Pharmacopoeia and European Pharmacopoeia, International harmonization of Japanese, the United States and European drug regulations is currently in progress however, there is not yet any common validation requirements for spectrophotometers.

Table 1 Inspection Items of Performance Validation Software

Inspection Item	JIS	JP	EP	USP
Wavelength accuracy	○	○	○	○
Wavelength repeatability	○	○		
Photometric accuracy	○	○	○	○
Photometric repeatability	○	○		
Resolution	○		○	
Stray light	○		○	
Noise level	○			
Baseline flatness	○			
Baseline stability	○			

When bright lines are used, the acceptable wavelength range for wavelength accuracy* is within ± 0.3 nm, and for wavelength repeatability*, it is within ± 0.2 nm of the mean value. When the filter is used, the wavelength accuracy* must be within ± 0.5 nm, and the wavelength repeatability* must be within ± 0.2 nm of the mean value.

In addition, for transmittance and absorbance, testing must be conducted using a commercially available transmittance calibration filter.

Regarding the acceptable ranges of transmittance or absorbance, for photometric accuracy*, it must be within the upper limit and low lower limit plus 1 %, respectively, of the relative accuracy indicated in the transmittance calibration filter test results document. For photometric repeatability*, when the absorbance is 0.500 or less, it must be within ± 0.002 of the mean value, and when the absorbance exceeds 0.500, it must be within ± 0.004 of the mean value. Also, if multiple

transmittance calibration filters having different transmittances are used together, it is advisable to verify the linearity.

Setting the various test conditions for these validation items, conducting the measurements, and then manually calculating the measurement values is extremely complex work. That is what makes the Performance Validation Software so useful. With this software, all that is required is to select the test item, and validation is easily conducted.

■ European Pharmacopoeia (EP)

In the "Absorption spectrophotometry, ultraviolet and visible" section of the 7th Edition of the European Pharmacopoeia, there are requirements related to wavelength accuracy*, photometric accuracy*, resolution*, and stray light* for ultraviolet-visible spectrophotometers. There are no items of resolution* or stray light* in the Japanese Pharmacopoeia. Additionally, there is a description regarding slit width setting and the optical path length of the measurement cell.

Regarding wavelength accuracy*, this can be checked using any of holmium perchlorate solution absorption bands, or the bright lines of a hydrogen-discharge tube, deuterium-discharge tube, or low-pressure mercury lamp, whichever is appropriate. The acceptable range is within ± 1 nm in the ultraviolet region, and ± 3 nm in the visible region.

■ The United States Pharmacopoeia (USP)

In the "spectrophotometry and light-scattering" section of the United States Pharmacopoeia, there are requirements related to wavelength accuracy* and photometric accuracy* for ultraviolet-visible spectrophotometers.

First, regarding wavelength accuracy*, it is stated that this can be checked using the bright lines of a low-pressure mercury lamp, the bright lines of a hydrogen-discharge tube, or an appropriate didymium or holmium, etc. glass filter having absorption in the ultraviolet region.

It is stated that photometric accuracy* can be checked using a standard glass filter or a solution of known transmittance, such as potassium dichromate.

However, specific acceptable ranges are not mentioned for these items. In the Performance Validation Software, the strictest values within the acceptable ranges of other regulations and standards are set as recommended values.

Note) The descriptions associated with the regulations and specifications are those that were current as of September, 2011.

Note) Items indicated with an asterisk (*) are the names of items that are supported in the Performance Validation Software.

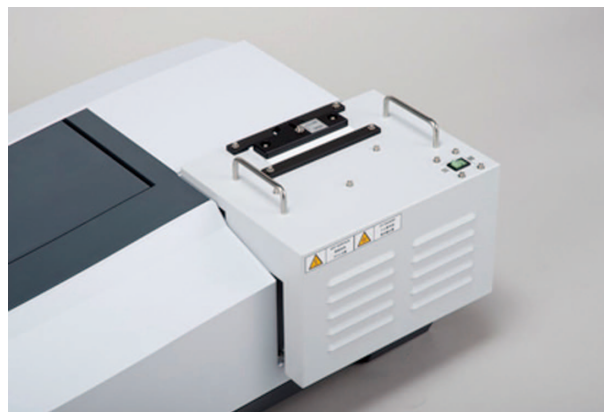


Fig. 1 Low-Pressure Mercury Lamp Unit

It is stated that photometric accuracy* can be checked using a suitable absorption filter or potassium dichromate solution. The acceptable range is within ± 0.010 of the absorbance.

Regarding stray light*, this can be checked using an absorption filter or 12 g/L potassium chloride solution. At 198 nm, absorbance must be 2.0 or greater (with transmittance, 1 % or less).

Resolution* is to be checked using hexane solution containing 0.02 % (V/V) of toluene. The ratio of the maximum absorption value at 269 nm and the minimum absorption value at 266 nm must be checked.

The Performance Validation Software supports both the stray light and resolution items of the European Pharmacopoeia, permitting easy validation of all the specified items.

■ Example of Instrument Validation

Fig. 2 shows the Performance Validation Software screen. The screen is divided broadly into 4 panes.

(1) File Information Pane

This is where information is entered for automatic saving of files following measurement. Testing cannot start unless the required items are entered to prevent omissions of necessary items. Also, measured data is saved without fail, so there is no forgetting to record data.

(2) Inspection Items Pane

This is where inspection items are verified. The items are displayed in an easy-to-understand list. Pressing the Conditions setting button in the toolbar displays the Inspection Conditions setting screen, where items to be displayed can be added or removed. Fig. 3 shows the screen for setting the inspection conditions. The items required to be inspected can easily be selected using the checkboxes. Displaying the detailed conditions makes it easy to modify the acceptable ranges and perform settings according to recommended values.

(3) System State Pane

This pane displays the remaining inspection time and inspection progress status (inspection log).

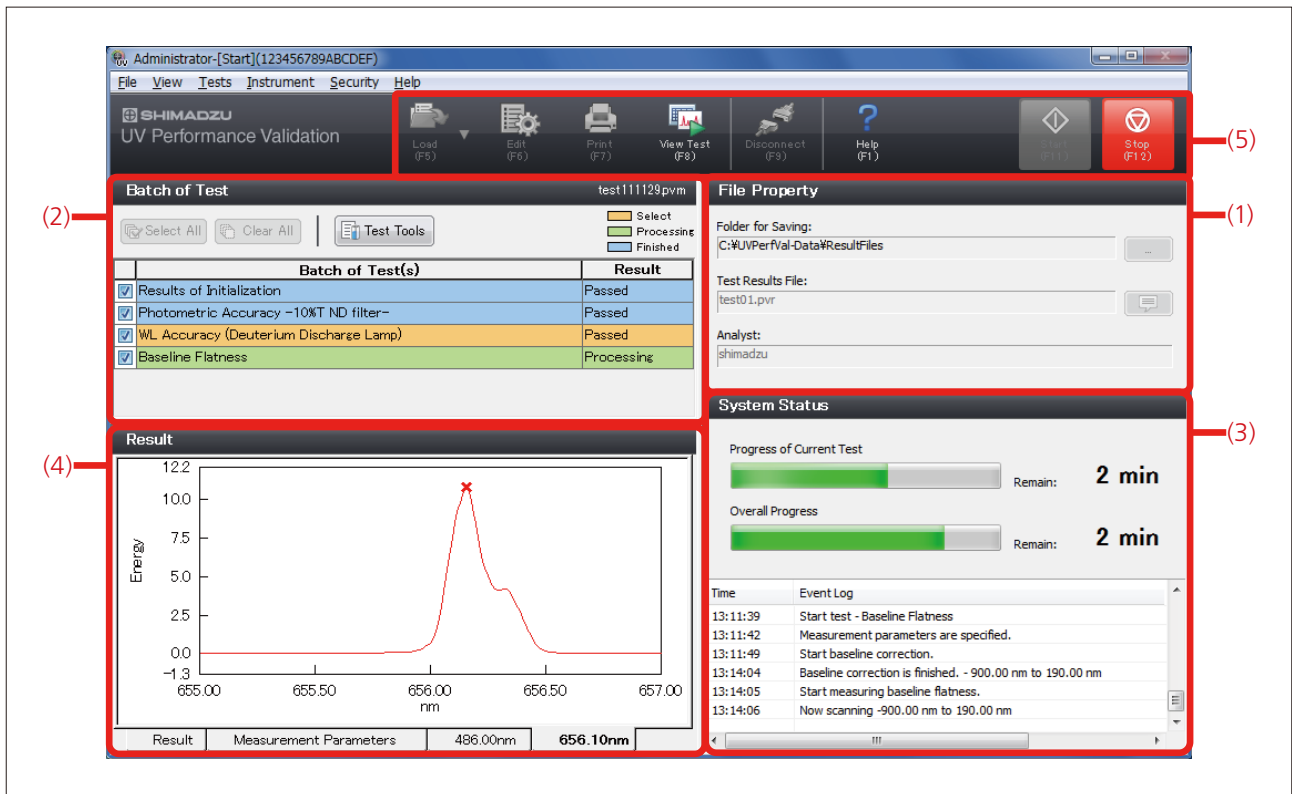


Fig. 2 Performance Validation Software Screen

(4) Inspection Results Pane

The data of the inspection currently in progress or the results of selected inspection items are displayed. Also, switching between tabs at the bottom of the screen allows viewing of measurement data and conditions.

(5) Toolbar

Functions that are used often are displayed as tool buttons.

After verifying the items to be included in the inspection, the inspection is started by pressing the Start button. Since the instrument conditions and other settings are conducted automatically by the software, the operation is nearly effortless for the analyst. All that is required is to check that the sample compartment is empty and to insert or remove filters according to the on-screen directions.

When the inspection is completed, a pass/fail assessment result is displayed for each inspection item in pane (2). To print all of the inspection results, all that is required is to press the Print button. The printout contains the details and the pass/fail assessment result for each of the inspection items. Moreover, the measurement results are automatically saved as an electronic file. Printout examples of wavelength accuracy (low-pressure mercury lamp) and resolution (toluene/hexane method) inspection results newly supported in the Performance Validation Software are shown in Fig. 4 and Fig. 5.

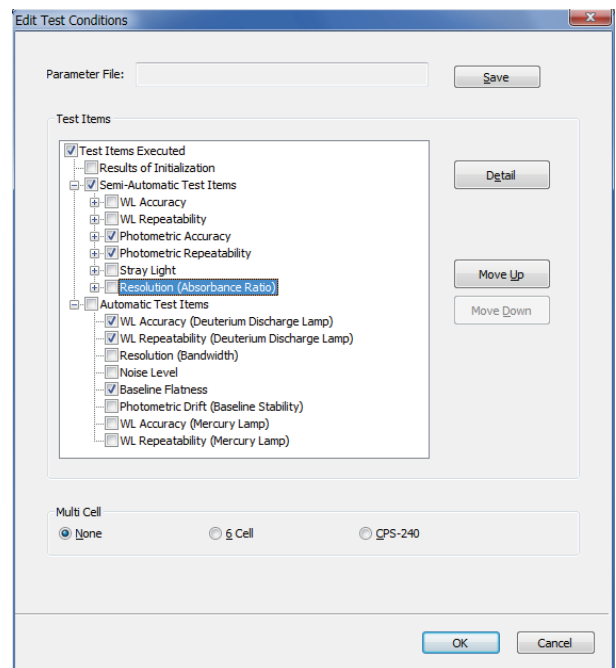
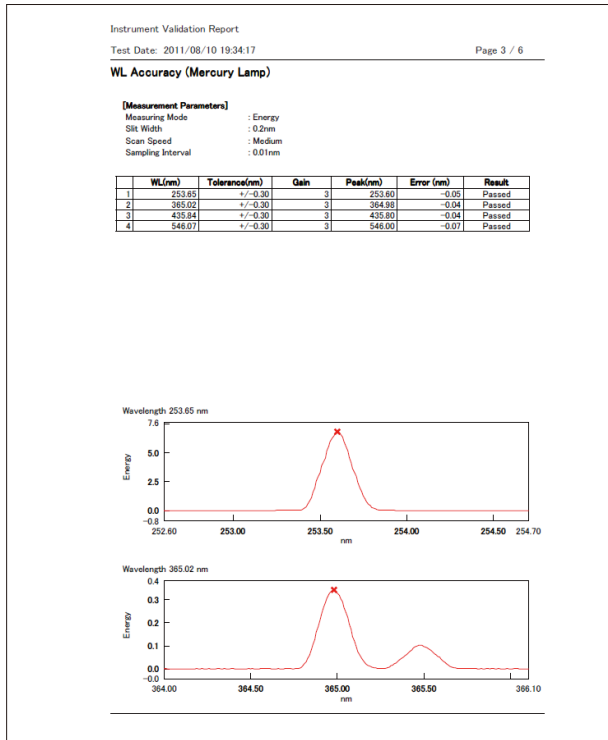
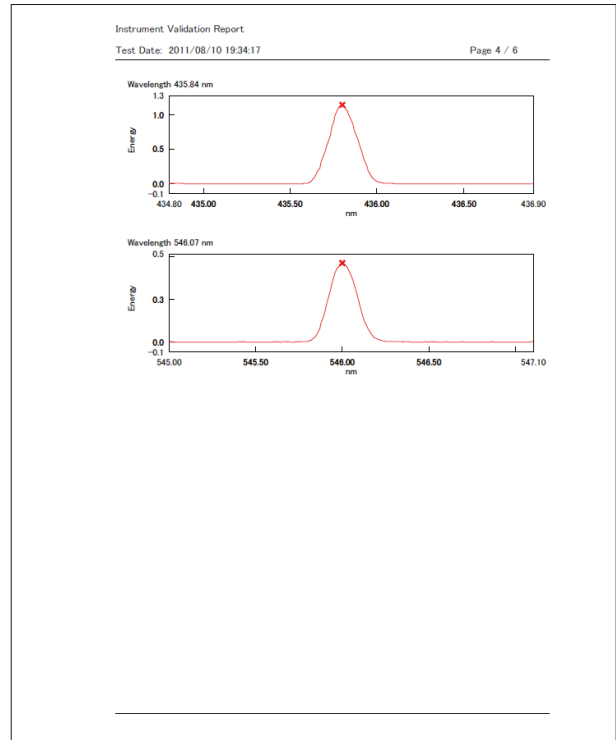


Fig. 3 Screen for Setting of Inspection Items

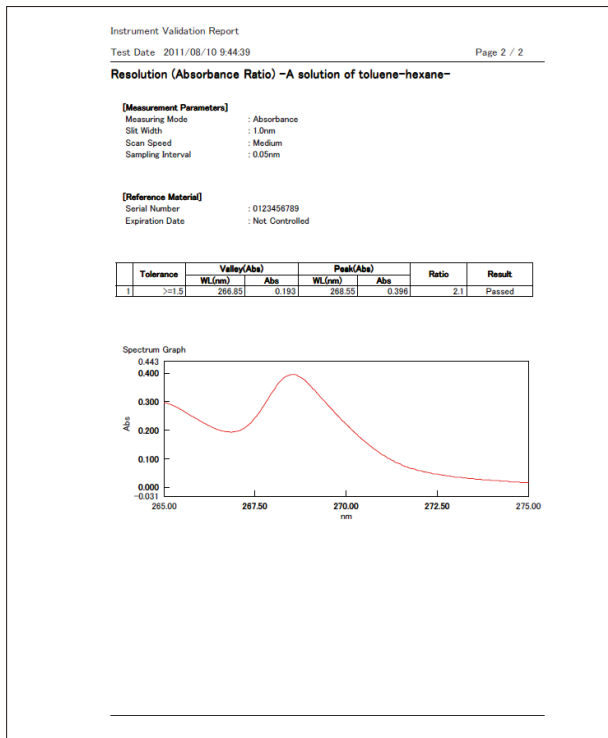


Wavelength Accuracy (Low-Pressure Mercury Lamp) First Page



Wavelength Accuracy (Low-Pressure Mercury Lamp) Second Page

Fig. 4 Printout Results (1)



Resolution (Toluene/Hexane Method)

Fig. 5 Printout Results (2)

Conclusion

Using this Performance Validation Software makes it easy to conduct an entire series of required regulation- and standards-related operations; from the setting of validation conditions, to data measurement, pass/fail judgment, results printout and file storage.