SHIMADZU
Excellence in Science

Application News

No.A465

Spectrophotometric Analysis

Identification Testing Using Matching Calculation – Method Using Easy Macro

■ Introduction

There are several ways to evaluate the degree of similarity between two samples, one of which is by use of a matching calculation. The matching calculation is a way to display a number that represents the degree of similarity obtained by comparing the infrared spectra of two samples.

Here, we introduce a matching calculation Easy Macro which was used to determine the degree of similarity between an infrared spectrum we obtained through sample measurement and an existing infrared spectrum.

■ Degree of Similarity

The matching calculation is conducted by comparing the two infrared spectra. In the actual calculation, the absorbance values at each wavenumber in the two infrared spectra are plotted two-dimensionally to determine the similarity. The correlation coefficient is used for evaluating the degree of similarity. The closer the correlation coefficient is to 1, the greater the similarity of the infrared spectra, and a correlation coefficient of 1 indicates an exact match between the two infrared spectra.

Thus, the degree of similarity expresses the similarity of two spectra using a numerical value. Reducing the correlation between two spectra to a numerical value is a way of managing identification testing and product control in various fields, including the pharmaceutical and food additive fields.

■ Easy Macro

LabSolutions IR now includes a new Easy Macro feature. This Easy Macro feature automates the programming process using drag-and-drop macro components.

Here, we describe a case in which we measured the infrared spectrum of the sample of interest, and then calculated the degree of similarity with respect to an existing reference sample spectrum.

The entire process from startup of the program to calculation of the degree of similarity and printout of the results is shown in the flowchart of Fig. 1. The degree of similarity between a measured spectrum and an existing spectrum is calculated using this procedure.
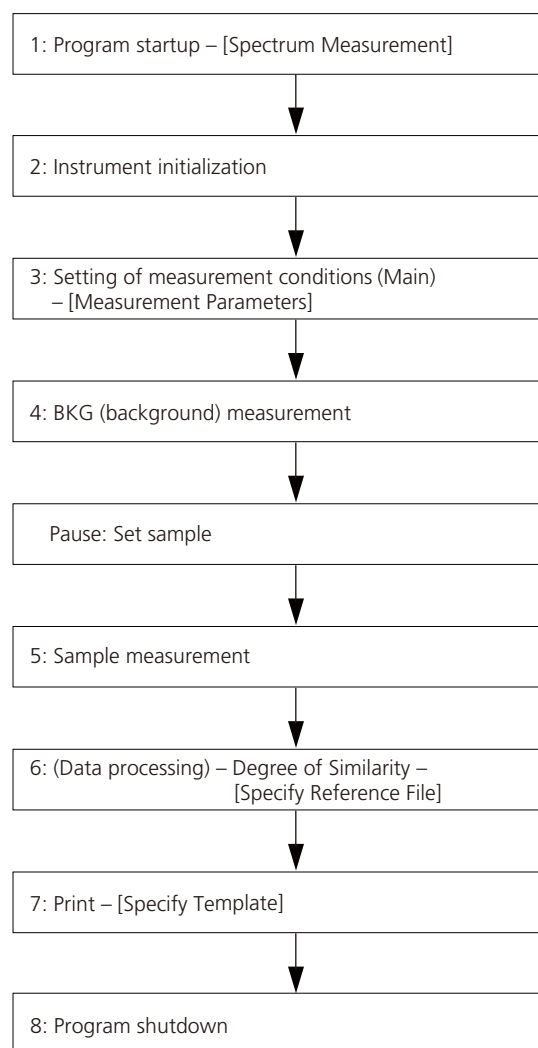
| 1: Program startup – [Spectrum Measurement] |
| 2: Instrument initialization |
| 3: Setting of measurement conditions (Main) – [Measurement Parameters] |
| 4: BKG (background) measurement |
| Pause: Set sample |
| 5: Sample measurement |
| 6: (Data processing) – Degree of Similarity – [Specify Reference File] |
| 7: Print – [Specify Template] |
| 8: Program shutdown |

Fig. 1 Flowchart of Similarity Calculation

■ Creating a Macro

Creating a macro begins with starting up the Easy Macro editing tool in LabSolutions IR. This tool is used to create the macro sequence (automatic execution procedure).

Fig. 2 shows the macro sequence editing screen. To create a macro, macro items are selected from the macro item list shown on the left side, and moved to the right side by drag-and-drop or double-clicking.

The macro items are organized into groups, including program startup/shutdown, measurement, file operations, quantitation, and spectrum search. Fig. 3 shows an example (Degree of Similarity) of a macro item in detail. The macro items include operation parameters. When the macro item is selected, it is also necessary to set the appropriate operating parameters. For example, in the case of the Degree of Similarity macro, a reference spectrum file must be specified as shown in Fig. 3. In addition, this macro item includes the [Display prompt before executing] and [Pause after executing] options. By selecting the [Display prompt before executing] checkbox, the macro can be paused before it is run in order to edit the parameter settings. In the case of Degree of Similarity, when the prompt is displayed before the macro is executed, there is an opportunity to specify a spectrum file to be loaded other than that currently specified in the panel shown in Fig. 4.
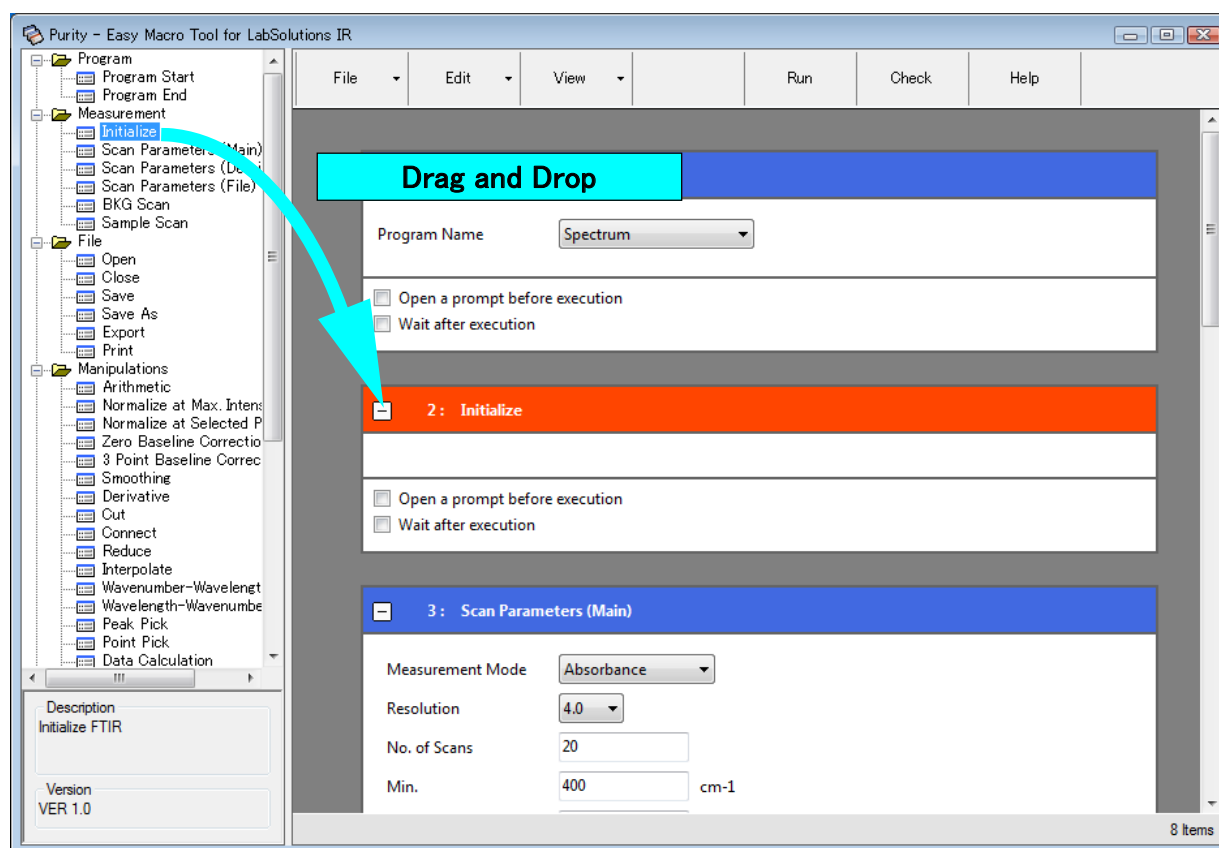

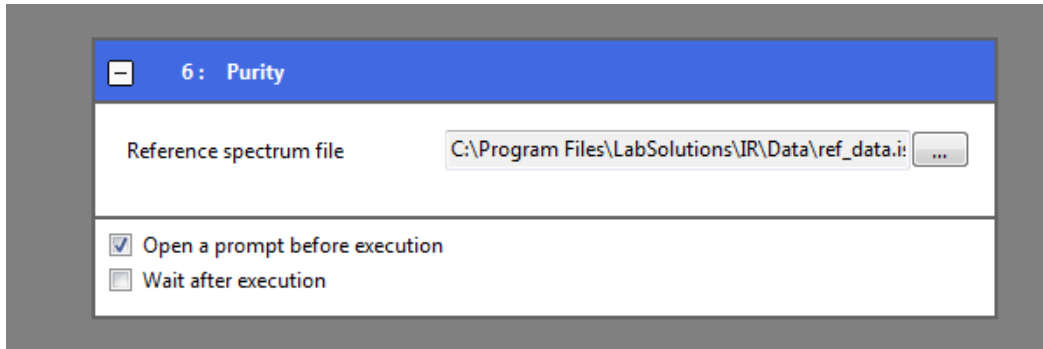
Fig. 2 Easy Macro Editor Screen

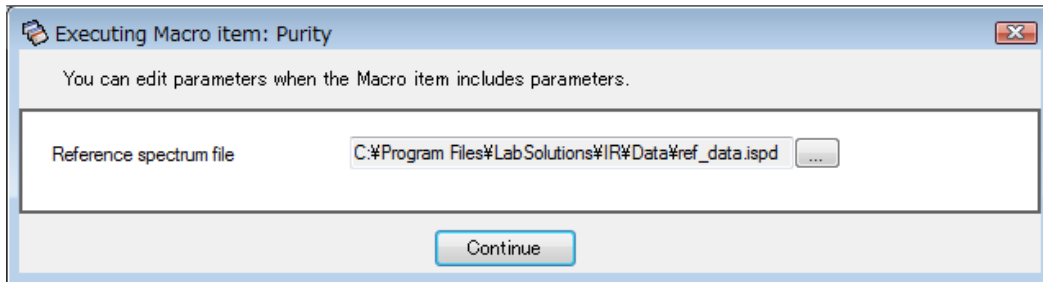**Fig. 3 Example of Macro Item Detail (Degree of Similarity)**

Fig. 4 Selection of Reference Spectrum File for Degree of Similarity

■ Control Statement

Macro items other than those listed in the regular menu are also included in the software as control statements. Insertion of control statements permits the repetition of operations and the display of messages, etc. An example of a displayed message is shown in Fig. 5. Thus, by creating messages related to the operation procedure, even those who are unfamiliar with details of that macro can confidently execute the procedure guided by the on-screen messages.
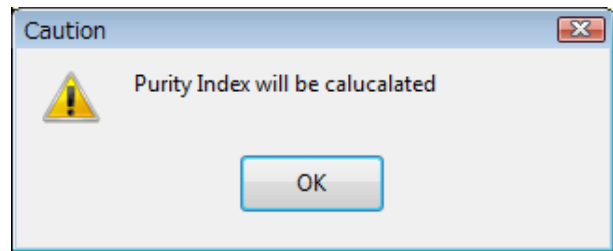

Fig. 5 Example of Message

■ Verifying the Easy Macro

A macro verification feature is also included among the Easy Macro editing tools. By clicking [Check] in the menu shown in Fig. 6, a macro check function is initiated to identify any problems with the macro sequence (such as data processing occurring before the program is started). It also checks to verify that the necessary parameters have been set for each of the relevant macro items. If there are no problems found in the macro, the message shown in Fig. 7 is displayed.
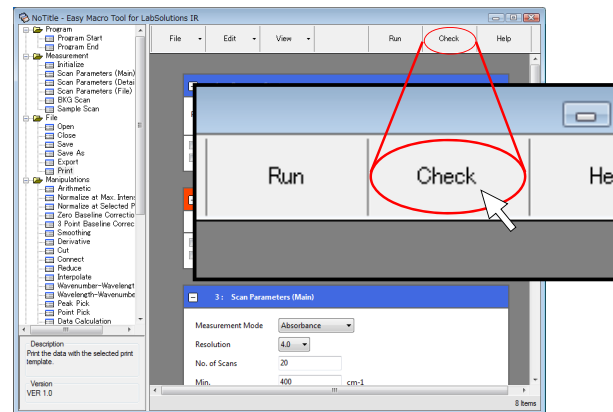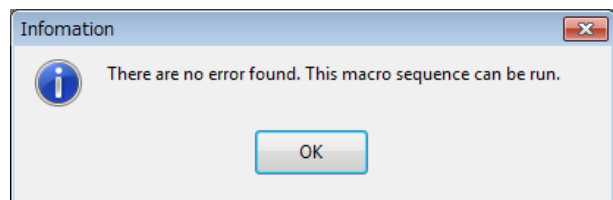

Fig. 6 Macro Check Menu Item

Fig. 7 Macro Check Message

Running a Macro

Created macros can be added to the LabSolutions IR launcher menu and measurement program tool bar. This makes it easy to run a macro.

The screen at left in Fig. 8 shows examples of created Easy Macro shortcuts added to the LabSolutions IR launcher macro execution screen. The macro that was added here has a red frame around it. Just clicking on a macro in this list will run the macro. Easy Macro files can also be listed on the desktop as shortcuts. Because Easy Macro files are associated with the LabSolutions IR program, just double-clicking any of the macro files or macro shortcuts will execute the macro. The screen at right in Fig. 8 shows an example of a macro shortcut added to the desktop.
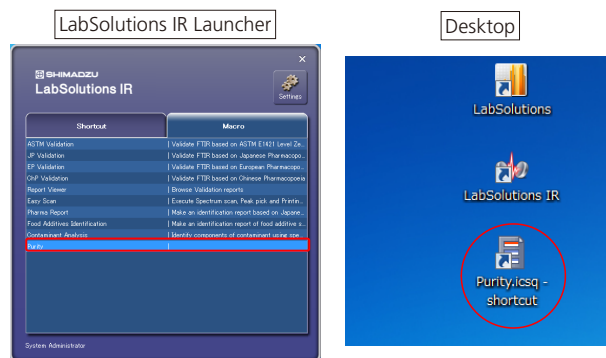

Fig. 8 Examples of Added Macro Programs

Degree of Similarity Calculation Results

After measuring the infrared absorption spectrum of glycine, as specified in the Japanese Pharmacopoeia, we used the Easy Macro to compare it with an existing spectrum and calculate the degree of similarity. An example of the printout results is shown in Fig. 9.

The overlaid target and reference spectra are shown in the upper part of Fig. 9. The middle part of the printout shows the degree of similarity regression graph. This graph is a curve representing the correlation between the respective intensities of reference data and target data. The lower part of the printout shows the degree of similarity parameters, and the calculated result (correlation coefficient). The calculated result for the degree of similarity was 0.9978, indicating good agreement.

Thus, by following the on-screen directions, the operation can easily be executed right through to the final printout.

Conclusion

Here, we calculated a similarity index using the Easy Macro feature. The similarity index is a measure of how similar the FTIR spectra of two samples are, and may be suitable as a method for evaluating the degree of similarity between two spectra.

We also conducted automated data processing using the Easy Macro feature. With this feature, creating macro programs, including specialized data processing such as Kubelka-Munk conversion, and printing out the results using a template can now all be conducted automatically.

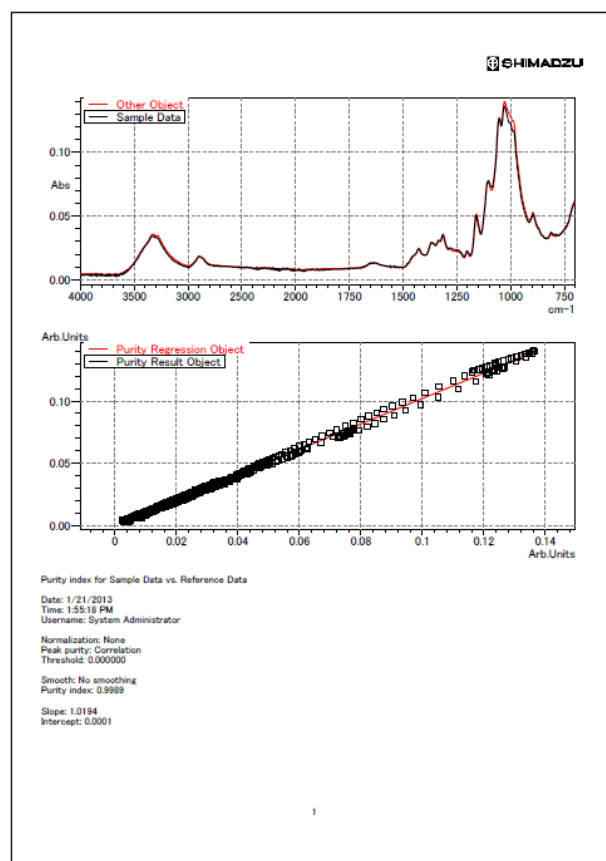The Easy Macro feature is extremely useful not only for calculating the degree of similarity, but for automating a wide range of procedures.

Fig. 9 Output Example of Degree of Similarity

SHIMADZU

Shimadzu Corporation

www.shimadzu.com/an/