SHIMADZU
Excellence in Science

Application News

Ultra High Performance Liquid Chromatograph Nexera™ FV

# Online Dissolution Testing of Supplements

A. Tanabe, K. Matsuoka

## User Benefits

- Dissolution testing can be automated.
- The automatic sampling function enables testing with good reproducibility.
- Dissolution testing of supplements is possible in the same manner as with pharmaceuticals.

## ■ Introduction

Dissolution testing is conducted for formulation development, quality control, bioequivalence tests of generic drugs, etc. In dissolution testing, the dissolution properties of a drug product are checked under specific conditions for certain periods. Dissolution testing takes a lot of labor and time, since dissolution media must be sampled from multiple vessels every sampling time and analyzed.

Nexera FV is an HPLC system for online dissolution testing.

It can automate the processes from sampling dissolution media, HPLC analysis, up to report output. By automating tasks that used to be conducted manually by operators, the system realizes labor saving and throughput improvement. In addition, automating tasks prevent human errors from whole processes.

This article introduces the workflow of online dissolution testing of two kinds of supplements using Nexera FV.

## ■ Online Dissolution Testing with Nexera FV

Fig. 1 shows a comparative example of the workflow of conventional method and online dissolution testing with Nexera FV.

The Nexera FV system can automate sampling of dissolution medium at designated times, filtration, dilution, addition of internal standards, HPLC analysis, and report generation, processes which had been conducted manually. Furthermore, the HPLC analytical conditions can be set easily by the dedicated software of DT-Solution (Fig. 2), and a report summarizing multiple data such as the dissolution rate is prepared simultaneously with completion of the analysis (Fig. 3)

Nexera FV has two modes of analysis: direct injection mode to inject the dissolution media delivered from the dissolution tester directly to the HPLC, and fraction analysis mode to fractionate the dissolution media into vials and analyze them. The former is effective when an analysis is enough short to complete by the next sampling time. The latter is used in tests with short sampling intervals, and in cases where dilution or addition of internal standards is required.

Furthermore, the fraction analysis mode also makes it possible to conduct HPLC analyses between sampling intervals. As a result, the total time could be reduced by 30%.

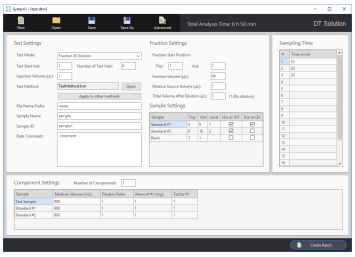


Fig. 2 DT-Solution Setting Window

Fig. 3 Multi Data Report*1 Window

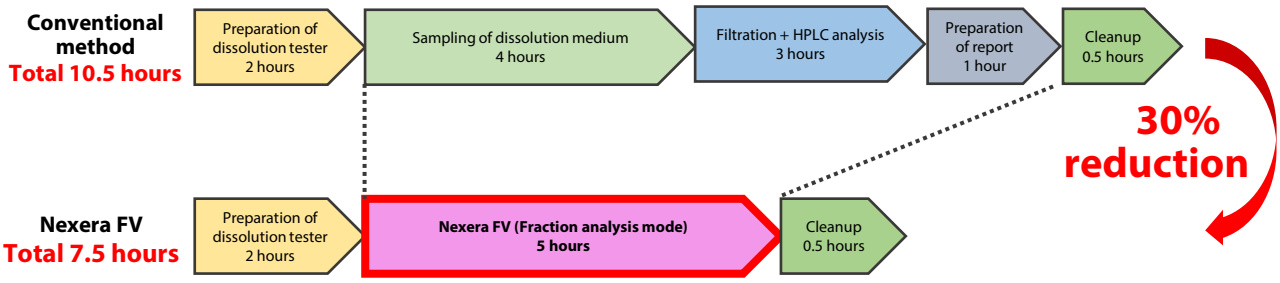



Fig. 1 Comparison of Workflow of Online Dissolution Testing*2

*1 Multi Data Report is an optional function of LabSolutions™DB/CS to generate reports automatically.
*2 The time above is an example of α-lipoic acid capsule with a dissolution test of 4 h and an HPLC analysis of 2.5 hours (50 analyses of 3 minutes each).

## ■ Dissolution Testing of Vitamin B2 (Riboflavin)

A dissolution test of riboflavin (labeled amount: 12 mg/tablet) was conducted using commercially-available vitamin B group tablets. Fig. 4 shows the chromatogram of vitamin B group tablet at a dissolution time of 60 min. Table 1 and Table 2 show the dissolution conditions and the HPLC conditions, respectively. Table 3 shows the results of the reproducibility of the retention time and area for riboflavin when using a 10 mg/L standard solution, Table 4 shows the dissolution rate at each sampling time, and Fig. 5 shows the dissolution curve.

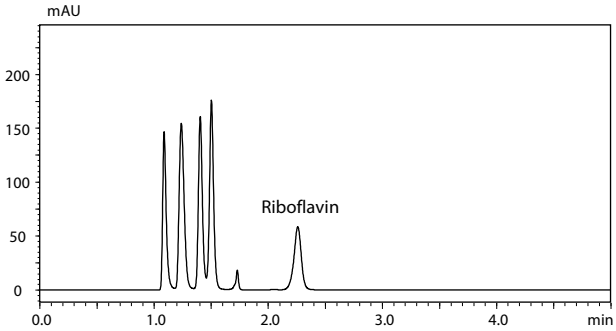



Fig. 4 Chromatogram of Vitamin B Group Tablet

Table 1 Dissolution Conditions (Riboflavin)

| | | |
|---|---|---|
| System | : | NTR-6600AST (TOYAMA SANGYO CO., LTD.) |
| Dissolution method | : | Paddle |
| Dissolution media | : | Water |
| Media volume | : | 900 mL |
| Rotation speed | : | 100 rpm |
| Bath temperature | : | 37 ˚C |
| Total time | : | 90 min |
| Sampling time | : | 5, 10, 15, 20, 30, 45 and 60 min |

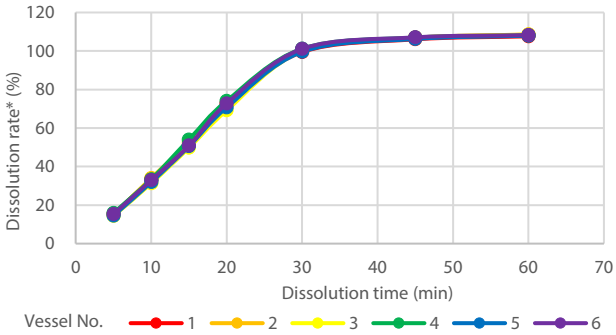
Table 2 HPLC Conditions (Riboflavin)

| | | |
|---|---|---|
| Column | : | Shim-pack™ VP-ODS*1 (150 mm × 4.6 mm I.D., 5 µm) |
| Mobile phase | : | 10 mmol/L (Sodium) phosphate buffer (pH2.6) / acetonitrile = 80 : 20 |
| Flow rate | : | 1.0 mL/min |
| Column temp. | : | 40 ˚C |
| Injection vol. | : | 10 µL |
| Vial | : | Shimadzu Vial, LC, 1.1 mL, Glass*2 |
| Detection | : | UV 210 nm |

*1 P/N: 228-34937-91 *2 P/N: 228-21283-91

Table 3 Reproducibility of Retention Time and Area for Riboflavin (10 mg/L, n=6)

| | Retention time (min) | Area |
|---|---|---|
| 1st | 2.261 | 182371 |
| 2nd | 2.260 | 182342 |
| 3rd | 2.260 | 182192 |
| 4th | 2.260 | 182266 |
| 5th | 2.261 | 182251 |
| 6th | 2.262 | 182250 |
| Average | 2.261 | 182279 |
| %RSD | 0.0231 | 0.0364 |




* Dissolution rate (%) = Concentration (mg/L) × Media volume 0.9 (L) / Labeled amount 12 (mg) × 100

Fig. 5 Dissolution Curve of Riboflavin

Table 4 Dissolution Rate of Riboflavin (%)

| Vessel No. \ Time (min) | 5 | 10 | 15 | 20 | 30 | 45 | 60 |
|---|---|---|---|---|---|---|---|
| 1 | 15.2 | 33.4 | 50.8 | 72.2 | 99.5 | 106.1 | 107.7 |
| 2 | 15.1 | 34.0 | 52.5 | 73.0 | 100.8 | 106.9 | 108.7 |
| 3 | 14.3 | 31.3 | 49.7 | 69.2 | 100.0 | 106.9 | 108.4 |
| 4 | 15.8 | 33.4 | 54.0 | 74.0 | 101.1 | 106.9 | 108.0 |
| 5 | 14.6 | 32.0 | 50.6 | 70.8 | 99.6 | 106.3 | 108.0 |
| 6 | 15.3 | 32.9 | 50.9 | 72.7 | 101.1 | 107.0 | 108.2 |
| Average | 15.0 | 32.8 | 51.4 | 72.0 | 100.3 | 106.7 | 108.2 |
| %RSD | 3.2 | 2.8 | 2.7 | 2.2 | 0.7 | 0.3 | 0.3 |

## ■ Dissolution Testing of *α*-Lipoic Acid

A dissolution test of *α*-lipoic acid (labeled amount: 105 mg/capsule) was conducted using commercial *α*-lipoic acid capsules. A sinker was used in the dissolution process. Fig. 6 shows the chromatogram of *α*-lipoic acid capsule at a dissolution time of 240 min. Table 5 and Table 6 show the dissolution conditions and the HPLC conditions, respectively. Table 7 shows the results of the reproducibility of the retention time and area for α-lipoic acid when using a 100 mg/L standard solution, Table 8 shows the dissolution rate at each sampling time, and Fig. 7 shows the dissolution curve.

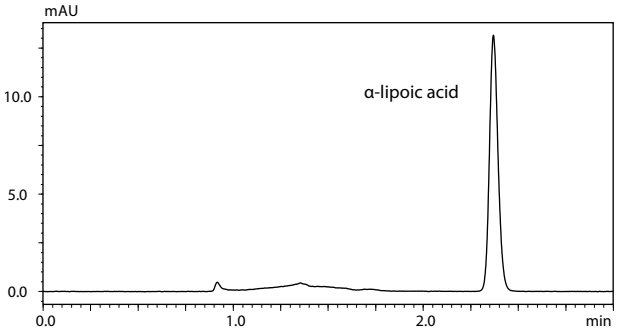



Fig. 6 Chromatogram of *α*-Lipoic Acid Capsule

Table 5 Dissolution Conditions (*α*-Lipoic Acid)

| | |
|---|---|
| System | : NTR-6600AST (TOYAMA SANGYO CO., LTD.) |
| Dissolution method | : Paddle |
| Dissolution media | : Water |
| Media volume | : 900 mL |
| Rotation speed | : 100 rpm |
| Bath temperature | : 37 ˚C |
| Total time | : 240 min |
| Sampling time | : 5, 10, 20, 30, 50, 90, 180 and 240 min |

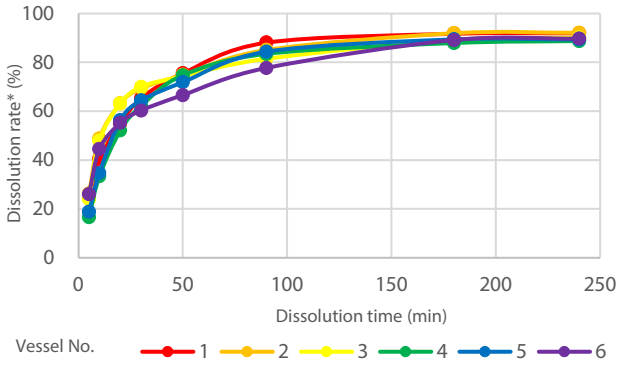
Table 6 HPLC Conditions (*α*-Lipoic Acid)

| | |
|---|---|
| Column | : Shim-pack VP-ODS*1 (150 mm × 4.6 mm I.D., 5 µm) |
| Mobile phase | : 10 mmol/L (Sodium) phosphate buffer (pH2.6) / acetonitrile = 30 : 70 |
| Flow rate | : 1.0 mL/min |
| Column temp. | : 40 ˚C |
| Injection vol. | : 10 µL |
| Vial | : Shimadzu Vial, LC, 1.1 mL, Glass*2 |
| Detection | : UV 333 nm |

*1 P/N: S228-41600-91 *2 P/N: 228-31537-91

Table 7 Reproducibility of Retention Time and Area for *α*-Lipoic Acid (100 mg/L, n=6)

| | Retention time (min) | Area |
|---|---|---|
| 1st | 2.368 | 39576 |
| 2nd | 2.370 | 39533 |
| 3rd | 2.369 | 39550 |
| 4th | 2.368 | 39719 |
| 5th | 2.368 | 39463 |
| 6th | 2.369 | 39566 |
| Average | 2.369 | 39568 |
| %RSD | 0.027 | 0.21 |




* Dissolution rate (%) = Concentration (mg/L) × Media volume 0.9 (L) / Labeled amount 105 (mg) × 100

Fig. 7 Dissolution Curve of *α*-Lipoic Acid

## ■ Conclusion

This article introduced online dissolution testing of two kinds of supplements using Nexera FV. As a result, good separation and reproducibility can be obtained, and confirmed that online dissolution testing of supplement was conducted easily.

Table 8 Dissolution Rate of *α*-Lipoic Acid (%)

| Vessel No. \ Time (min) | 5 | 10 | 20 | 30 | 50 | 90 | 180 | 240 |
|---|---|---|---|---|---|---|---|---|
| 1 | 24.3 | 40.6 | 54.9 | 65.3 | 75.5 | 88.1 | 91.7 | 91.9 |
| 2 | 26.2 | 48.8 | 63.2 | 69.3 | 74.8 | 85.0 | 91.9 | 92.1 |
| 3 | 24.0 | 47.9 | 63.0 | 69.9 | 74.3 | 81.4 | 89.1 | 90.1 |
| 4 | 16.6 | 33.4 | 52.0 | 62.1 | 74.6 | 83.4 | 87.8 | 88.6 |
| 5 | 18.8 | 34.7 | 56.3 | 64.4 | 71.8 | 84.3 | 89.4 | 89.3 |
| 6 | 18.8 | 34.7 | 56.3 | 64.4 | 71.8 | 84.3 | 89.4 | 89.3 |
| Average | 21.4 | 40.0 | 57.6 | 65.9 | 73.8 | 84.4 | 89.9 | 90.2 |
| %RSD | 16.5 | 15.8 | 7.2 | 4.3 | 2.0 | 2.4 | 1.6 | 1.5 |



01-00031-EN First Edition: Oct. 2021

SHIMADZU
Shimadzu Corporation
Analytical & Measuring Instruments Division
Global Application Development Center

www.shimadzu.com/an/