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LM1010 High Performance Liquid Chromatography

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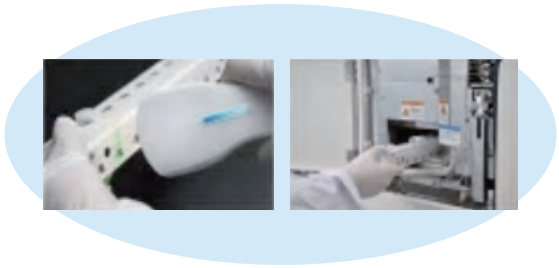
1. Introduction

The LM1010 system, developed in response to requests from those working in clinical sites[1], is a High Performance Liquid Chromatography (HPLC-UV System) for analyzing biological samples in hospitals, laboratories and research institutes. A general medical device notification for the system was submitted to the Japanese Pharmaceuticals and Medical Devices Agency (PMDA) in February 2020 (marketing notification number in Japan: 22B3X10009000003) [2] (Figure 1).

Operating the HPLC system typically requires expert knowledge and experience to establish proper sample pretreatment methods, analytical conditions, and operating procedures. Moreover, the equipment generally requires regular performance evaluation and periodic maintenance to acquire accurate results. These accumulating tasks may become a workload for operators.



Proprietary name: LM1010 High Performance Liquid Chromatography
General name: High Performance Liquid Chromatography analytical equipment
Marketing notification number in Japan: 22B3X10009000003
General medical device (specially-designated medical device requiring maintenance)

Fig. 1 LM1010 High Performance Liquid Chromatography

2. Key Features of LM1010 System

LM1010 system supports users by reducing the burden of the complex tasks involved in HPLC analysis—from establishing proper sample pretreatment methods, analytical conditions and operating procedures to routine quality control and periodic maintenance—and provides stable results not affected by the experience or knowledge of the user. The system's key features are presented here.

(1) Efficiency of operations

The goal was to optimize routine device operation by simplifying each operating step from system setup to sample analysis and result acquisition as much as possible. In addition, the consumables and general reagents needed for analysis are bundled as a set (Figure 2), and a common flow of analysis is provided (Figure 3) to increase efficiency.

(2) Routine quality control

Once booted, the system can perform everything from automatic conditioning to sample analysis for quality control (QC) with a single click, enabling system optimization for analysis and quality control.

(3) Adding analysis items

Since the system is based on liquid chromatography, the user can consider adding analysis items, a function made available at the request of users.

Fig. 2 Set of ready-to-use general reagents

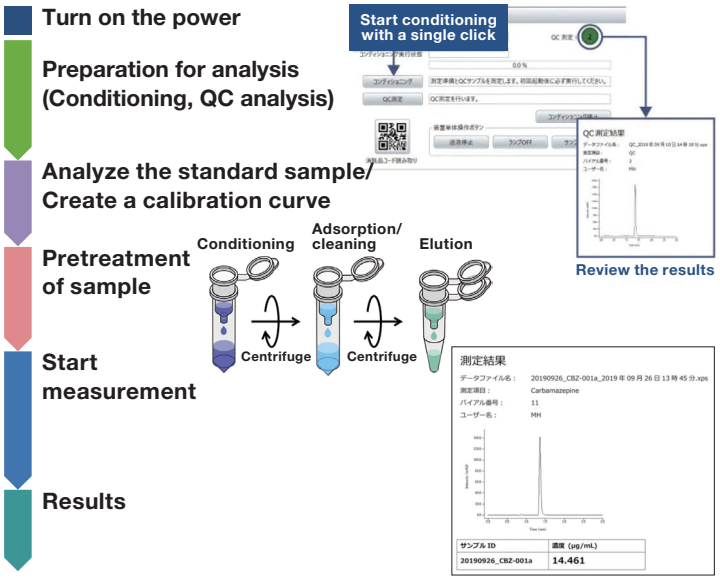


Fig. 3 Flow of analysis

3. Procedures

To use LM1010, load the mobile phase and cleaning fluid, boot the system, and click *Conditioning* on the operating screen to analyze a QC sample for LM1010. A passing result obtained in the QC sample analysis indicates that the system is operating normally and can begin analyzing samples. The system may not be operating normally if a failing result is obtained in the QC sample analysis, so the operator cannot proceed to sample analysis. This concludes preparations for analysis. Once preparations have concluded successfully, select the analysis item to perform and analyze the standard solution. A calibration curve will be automatically created. Next, pretreat the samples as directed in the procedures provided for the analysis item in question, then load and analyze the samples in the LM1010 to obtain the required data. The measured values for the samples are described in the report (Figure 3).

4. Application Example: Determining Blood Concentrations of Antimicrobial Agents in the Treatment of Infections

Blood concentrations of the antimicrobials used for treating infections shown in Table 1 can be determined when required at the rate of about 30 minutes per sample. Therapeutic drug monitoring (TDM) is recommended for antimicrobials with a narrow target therapeutic blood concentration window.

- The following text is excerpted from Reference 3.

“Therapeutic drug monitoring (TDM) starts from the initial treatment dosage design, measuring blood concentrations of drug after the start of treatment, and revising the dosage to ensure that treatment is safe and effective. TDM is used for drugs with a narrow target blood concentration window, such as glycopeptide and aminoglycoside antibiotics, and the antifungal agent voriconazole.”

Table 1 Antimicrobials determinable with the LM1010 system (as of September 2021)

Drug name	Category
Vancomycin	Anti-MRSA drug
Linezolid	Anti-MRSA drug
Voriconazole	Antifungal
Favipiravir	Antiviral

The results of analyses of vancomycin and voriconazole are shown below as examples.

4-1. Analysis of the glycopeptide antimicrobial vancomycin

A calibration curve was made with vancomycin standard solution (Figure 4). Next, standard serum (commercially purchased) spiked with vancomycin at concentrations of 1, 50, and 100 µg/mL were analyzed as pseudo-samples (Figure 5).

The results are data determined by analyzing these samples as directed in the provided operating procedures (Technical Report: AS/LM-012 Procedural Manual[4]) using the set of general reagents shown in Figure 2. Accuracy for the 1 to 100 µg/mL vancomycin-spiked sera was 94.4% to 95.5%, and precision was 1.25% to 6.58% (Table 2).

The correlation between the concentrations determined by LM1010 in the pseudo-samples (µg/mL, vertical axis) and vancomycin-spiked serum (µg/mL, horizontal axis) is shown in Figure 6. The coefficient of determination (R^2) was excellent, at 0.999.

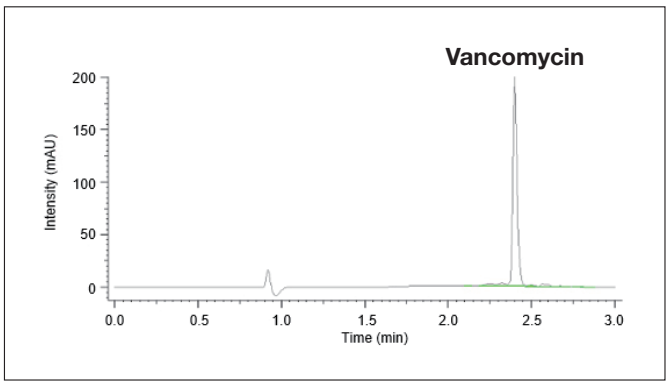


Fig. 4 Chromatogram of 100 µg/mL vancomycin standard solution

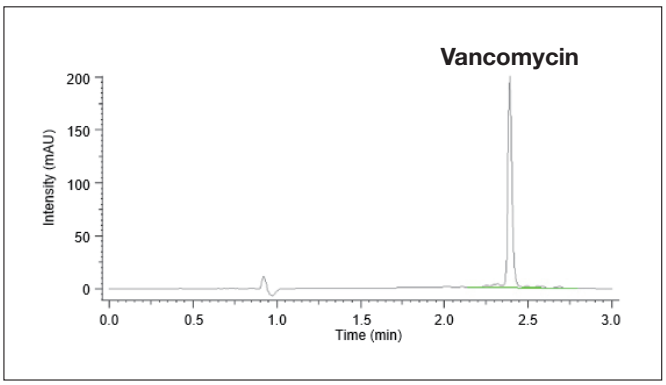


Fig. 5 Chromatogram of 100 µg/mL vancomycin-spiked serum (All steps from pretreatment to analysis of the pseudo-samples were performed as directed in the operating procedures.)

Table 2 Quantification results for vancomycin-spiked serum using LM1010 (mean)

Sample	Quantitative value (µg/mL)	Accuracy (%)	Precision (% RSD)
100 µg/mL standard solution	100	—	—
Blank serum (n=3)	0	—	—
1 µg/mL spiked serum (n=5)	0.946	94.6	6.58
50 µg/mL spiked serum (n=5)	47.748	95.5	1.25
100 µg/mL spiked serum (n=5)	94.419	94.4	2.69

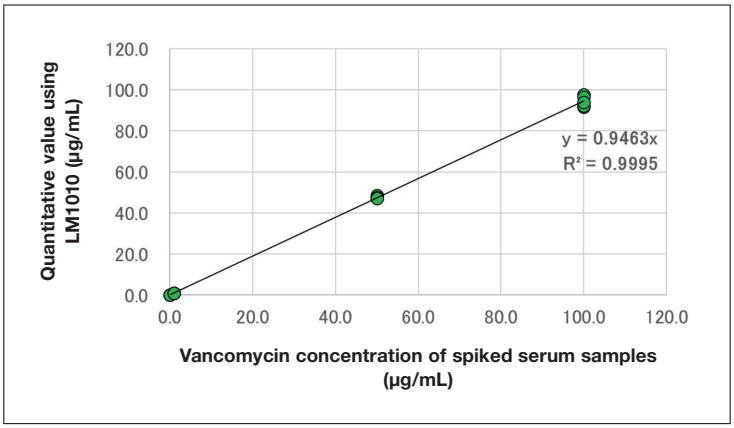


Fig. 6 Correlation between assay values determined by the LM1010 (µg/mL) and concentrations in vancomycin-spiked serum (µg/mL)

4-2. Analysis of the antifungal voriconazole

Voriconazole is the only antifungal drug targeted for TDM.

A calibration curve was made with voriconazole standard solution (Figure 7). Next, standard serum (commercially purchased) spiked with voriconazole at the concentrations of 1, 2, and 5 µg/mL were analyzed as pseudo-samples (Figure 8).

The results are data determined by analyzing these samples as directed in the provided operating procedures (Technical Report: AS/LM-004 Procedures[5]) using the set of general reagents shown in Figure 2. Accuracy for the 1 to 5 µg/mL voriconazole-spiked sera was 99.6% to 104.2%, and precision was 0.63% to 1.02% (Table 3).

The correlation between the concentrations determined by LM1010 in the pseudo-samples (µg/mL, vertical axis) and voriconazole-spiked serum (µg/mL, horizontal axis) is shown in Figure 9. The coefficient of determination (R^2) was excellent, at 0.998.

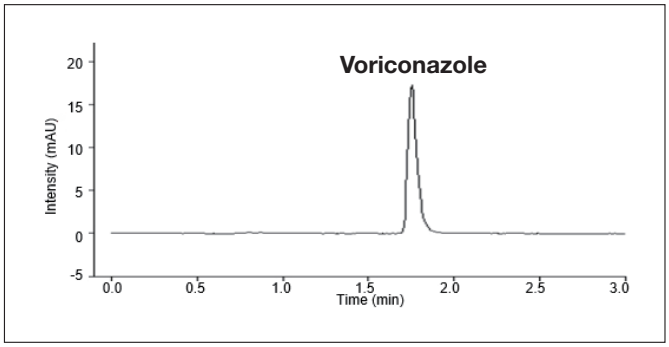


Fig. 7 Chromatogram of 5 µg/mL voriconazole standard solution

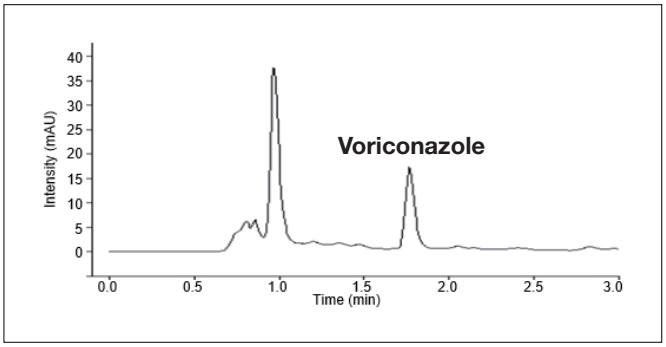


Fig. 8 Chromatogram of 5 µg/mL voriconazole-spiked serum (All steps from pretreatment to analysis of the pseudo-samples were performed as directed in the operating procedures.)

Table 3 Quantification results for voriconazole-spiked serum using LM1010 (mean)

Sample	Quantitative value (µg/mL)	Accuracy (%)	Precision (% RSD)
5 µg/mL standard solution	5.0	—	—
Blank serum	0.215	—	—
1 µg/mL spiked serum (n=5)	0.996	99.6	0.73
2 µg/mL spiked serum (n=5)	1.995	99.8	0.63
5 µg/mL spiked serum (n=5)	5.211	104.2	1.02

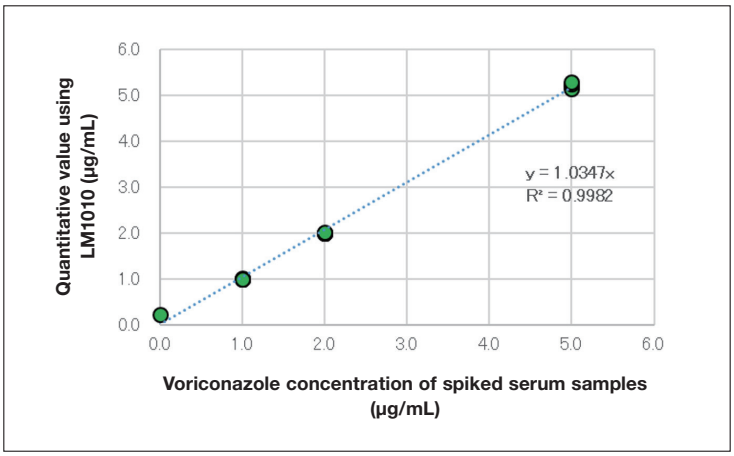


Fig. 9 Correlation between assay values determined by the LM1010 (µg/mL) and concentrations in voriconazole-spiked serum (µg/mL)

5. Use of LM1010 System

For drugs such as voriconazole that require blood drug concentration monitoring, the ability to quickly determine the concentration when required helps to optimize drug therapy.

- The following text is excerpted from Reference 3.

"Voriconazole concentration is an outsourced test in most medical institutions, and it takes more than one week from the start of administration to obtain the blood concentration results. Voriconazole can be used safely if genetic polymorphism analysis is performed in advance and the dosing regimen is decided based on the results. In addition, one of the key features of the LM1010 High Performance Liquid Chromatography is its ability to determine voriconazole concentrations. Medical institutions that own the LM1010 can perform same-day concentration analyses. So if the patients show signs of deterioration or development of liver injury, the clinician can check the blood levels immediately and decide on the same day to increase or decrease the voriconazole dose or change to another drug, instead of having to wait until 3-5 days for the blood levels, thereby optimizing treatment in a timely manner."

6. Conclusions

The LM1010 system can measure the drug concentration in a sample of serum (or plasma) in about 30 minutes after blood sampling. The ability to analyze samples when required, such as in emergencies or when prompt intervention is required, could help clinicians optimize drug therapy. The manufacturer will continue to look into adding analysis items and improving the system with an even greater focus on clinical needs.

Table 4 Other drugs determinable with LM1010 system (as of September 2021) (for reference)

Drug name	Category
Carbamazepine	Antiepileptic
Phenytoin	Antiepileptic
Lamotrigine	Antiepileptic
Disopyramide	Antiarrhythmic
Lidocaine	Antiarrhythmic
Mexiletine	Antiarrhythmic
Procainamide	Antiarrhythmic
Sotalol	Antiarrhythmic
Propafenone	Antiarrhythmic
Flecainide	Antiarrhythmic
Mycophenolate	Immunosuppressant
Pazopanib	Anticancer agent
Caffeine	Analgesic

References

1) Morikawa T, Development of a Practical HPLC System for In-hospital Analysis of Blood Concentration of Various Medicines (and Related Compounds), *The Hitachi Scientific Instrument News*, Vol. 63 No. 2 (2020) (in Japanese).

2) Product Literature for LM1010 High Performance Liquid Chromatography, July 2021 revision (Version 3) (in Japanese).

3) Matsumoto K, Use Genetic Analysis and Blood Drug Concentration Analysis to Empower Antimicrobial Stewardship (AS)!, *The Hitachi Scientific Instrument News*, **20** (2023).

4) Determining Vancomycin with the LM1010 High Performance Liquid Chromatography, Technical Report, AS/LM-012 (in Japanese).

5) Determining Voriconazole with the LM1010 High Performance Liquid Chromatography, Technical Report, AS/LM-004 (in Japanese).

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