Selected Article

Microsemi Series Now Complete

Soichi Oue, Takaaki Shindo, Itsuo Ito, Hiroki Nagaoka, Akiko Sakato

The Microsemi Series is a full model change of the LC-550 Automatic Blood Cell Counter and the LC-178CRP Automatic Blood Cell and CRP Counter, now significantly easier to use, with addition of a touch panel, and timer and quality control functions.
Some examples of new technologies used are:
1) Cyanide-free lyse(reagent for erythrocyte lysis.
2) Reagent container and cooling unit permitting onboard use of CRP reagent (if a reagent is set to an instrument once it can be used while remaining installed in the instrument during the effective period of the reagent).
3) Reagent factors with reduced information for manual input.
4) A piercing probe with a new structure able to penetrate caps of sample tubes.
Another improvement is a significant increase in the memory capacity of the instrument. It is able to record not only measurement data, but also self-diagnosis results at startup, and maintenance information such as detailed operation histories, permitting the user to quickly analyze the cause of any problems occurring with the instrument. This report focuses on the new technologies mentioned above.

Introduction: Microsemi Series Overview

Development of the Microsemi series began in 2003, with the objective of a full model change of the small blood cell counter. The name of the series originates from several parts: 'Micros', the brand name of the HORIBA Group's small blood cell counter, and 'emi', composed of the first letters of easy operation, maintenance free, and information technology. The current line-up consists of the three models listed below.
The first of the series is the Microsemi LC-660 (LC-660) (Figure 1(a)), released in 2006. The LC-660 is the standard small hematology analyzer model able to classify three types of white blood cells, and having features common to the entire series, for example, micro sampling compatible with collection of extremely small amounts of

Figure 1 Microsemi Series (a) LC-660 (b) LC-667 (c) LC-661

blood, a highly reliable measurement system using multiple counts, user-friendly operation with a color touch screen, and a timer function for start, cleaning, and shutdown.
The second part of the series is the Microsemi LC-667CRP (LC-667) (Figure 1(b)), released in 2008. The LC-667 is able to simultaneously count the blood cell count (including classification of three types of white blood cells) and measure C-reactive protein (CRP) concentration with a mere 18-µL sample. White blood cell count and CRP are often used as the primary inflammation markers in fever screenings[1], and this instrument has been introduced in large numbers in pediatric facilities in particular, because of its ability to treat very small samples.
The third part of the series is the Microsemi LC-661 (LC-661) (Figure 1(c)), released in 2009. In addition to the functions of the LC-660, the LC-661 has a cap piercing function for sample tubes to reduce the risk of infection when opening caps of vacuum sample tubes.
The technologies used in the Microsemi series are introduced below.

Cyanide-Free Lyse (Reagent for Erythrocyte Lysis): LC-660, LC-667, LC-661

Lyse has the effect of destroying cell membranes of the red blood cells making up the majority of the socap components in the blood. It is the reagents used when measuring white blood cells, which exist in fewer quantities compared to red blood cells. At the same time, It is also the reagents used when measuring the amount of hemoglobin in red blood cells.
When hemoglobin is measured, a colorimetric method is used, which uses absorption in the optical wavelength range. Absorption in the optical wavelength range originates from the heme (iron porphyrin complex) contained in hemoglobin, but the wavelength spectrum that is absorbed varies slightly based on the heme condition (oxidation or reduction). These variances affect the measurements of hemoglobin amount, and thus in conventional methods, cyanogen (CN) is bonded to heme , and the heme condition has stabilized, then the sample is measures (Cyanmethemoglobin method). The Microsemi series uses Cyanide-free lyse to improve user friendliness and to reduce environmental load.
The hemoglobin treated using methods that do not use cyanogen (cyanide-free method) tends to be less stable than the hemoglobin treated using conventional methods. To overcome the above disadvantage, the Microsemi series analyzes the absorption wavelength of the hemoglobin and selects a wavelength range that has fewer variances. The Microsemi series also has a diluent tank with a preheating function to account for the effects of temperature. The Microsemi series thus has the same or higher precision than conventional methods, even if cyanide-free lyse agents is used.

Onboard Use of CRP Reagent: LC-667

Teasting instruments are becoming even more automated and systematized, and it is not enough for instruments to simply take measurements - they must also be easy to operate and routinely maintain.
We used an all-in-one unit containing three reagents for the LC-667 CRP (CRP Unit 50) (Figure 2(b)). The reagent (Buloimmu Kit CRP (Figure 2(a)) for the LC-178CRP conventional model has three separate containers for the reagents, and we therefore improved ease of use. Having an all-in-one unit for the three reagents reduced the time needed for installing and removing containers, however reagents still had to be removed and placed in a refrigerator unit at instrument startup and shutdown. To solve this problem, a change to onboard*1 reagents was planned to allow the reagent to be left in the instrument, even after shutdown.

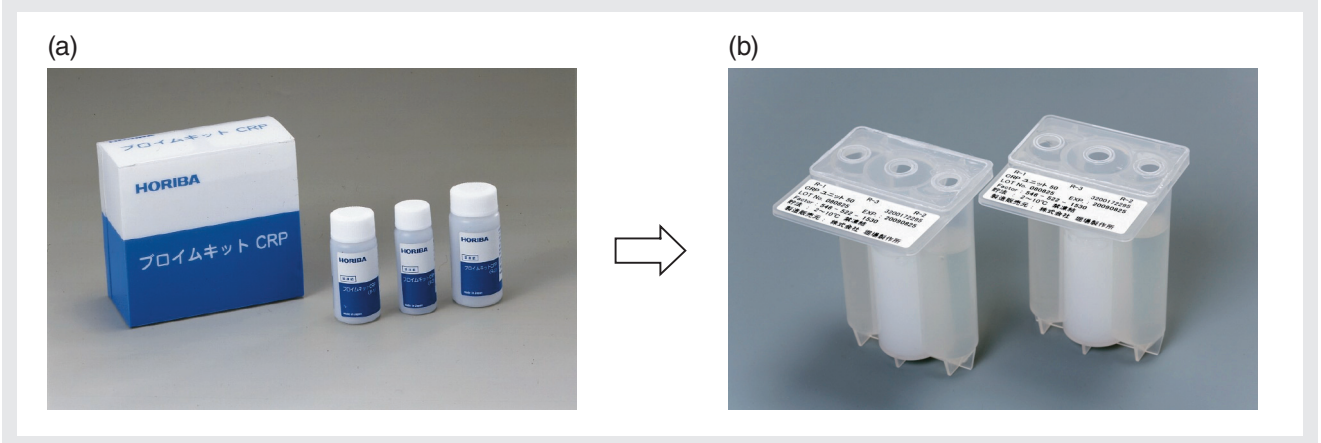

Figure 2 CRP Measuring Reagents (a) Buloimmukit CRP (b) CRP Unit 50

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To achieve the change to onboard reagents, stable reagents were required, and to this end, we first developed a reagent cooling unit free of the effects of outside air temperature. In addition, we investigated the following two points as a means of controlling variances in reagent concentration (due to evaporation, or condensation).
(1) The diameter of reagent container opening
(2) Sealing cap when reagent is not being used

(1), We set the diameter of reagent container opening to an inside diameter of 7 mm, aiming to minimize the effects of evaporation and condensation, and taking into account the ease of reagent fluid intake operations through a probe. (2) For the sealing cap, we investigated various conditions such as cap shape and the force pressing on the container. As a result, we were able to minimize concentration variances in the assumed environment in both dry (30 °C, 20% RH) and humid (30 °C, 85% RH) conditions, and were able to establish stable reagent performance for at least two months.
Based on these investigations, we were able to employ onboard reagents, significantly reducing the effort required by our customers.

*1: 'On board': Once a reagent is set in the instrument it can remain in the instrument during its effective life.

CRP Reagent Factor: LC-667

The CRP reagent for this instrument uses the principle of latex immunoturbidimetric method. This method utilizes the fact that the turbidity of the reaction solution varies based on the CRP protein concentration in the sample, measures the turbidity around 660 nm as the variance in absorbance, and converts it into concentration. Furthermore, the reactivity of the reagent differs with manufacturing lot, and thus a calibration curve needs to be created for each reagent lot.
The large instruments generally used in laboratories centers and medium-sized and larger hospitals measure the reference solution of multiple concentrations called calibrators immediately before every sample measurements and create the calibration curve based on those results. This technique has high accuracy, however its disadvantage is the consumption of a large volume of reagents, and thus these types of instruments are not practical for general practitioners, the target users of our instrument. The LC-667 has a mechanism that the calibration curve is set for each lot within the effective period of the reagent. When a customer uses a reagent from a new lot, the calibration curve factor for the reagent is input into the instrument first, and the same calibration curve can then be used for approximately 100 measurements (or two months after the reagent container is opened).
On the conventional model LC-178CRP, three-digit reagent factors is input for the three types of reagents using three buttons (↑, ↓, as Enter), as calibration curve information input. However, in some cases, depending on the reagent lot, it is difficult to cover the entire measurement range (0-20 mg/dL), and the calibration curve system needed fundamental improvement.
Higher-order parameters needed to be input in order to improve the accuracy of the calibration curve, however this would increase the burden on customers, causing input mistakes. Improving accuracy while maintaining the ease of inputting calibration curve information was a major issue.
We proposed the new Magic Number Table Calibration Curve Input System (Figure 3) to solve these problems. In this system, the reagent parameter information that needs to be input is entered in a database as a table in the instrument beforehand, and each parameter is expressed as a 1-digit number. For example, if there are five 3-digit reagent parameters that need to be input with a conventional system, 15 characters (3×5) must be input. However, with the new system, only 5 characters need to be input. By using this system with the LC-667, complicated calibration curves can be created by inputting a 10-digit number. As a result, we have improved both accuracy and ease of use.

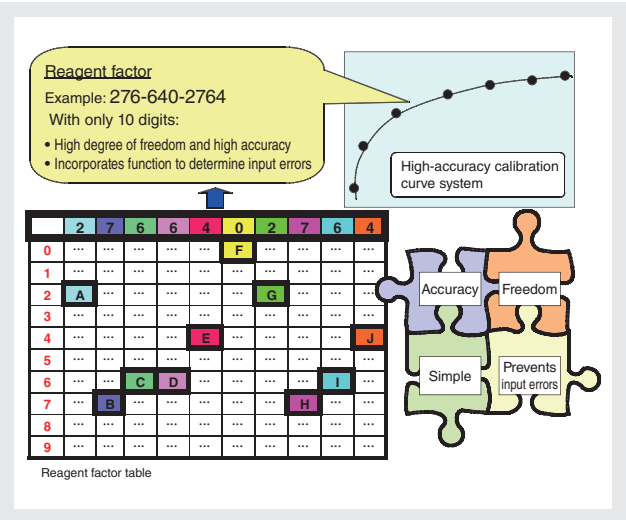

Figure 3 Magic Number Table Calibration Curve Input System

Piercing Probe: LC-661

The cap piercing function is implemented with a needle with a sharp end which penetrates the cap of a vacuum sample tube set on the instrument, and aspirates blood from the sample tube. This function eliminate the need for customers to open the caps of vacuum sample tubes, not only reducing time, but also proving extremely useful in reducing the risk of infections from patient samples. This probe is also installed on our large automatic hematology analyzer, and is an essential function for medium-sized and larger hospitals using large numbers of vacuum sample tubes. Even in the general practitioner market, the main target market for the Microsemi series, the demand for the cap piercing function is increasing, and we have created a line-up with this function implemented.

The cap piercing mechanism used on our large automatic hematology analyzer has a relatively thick needle (referred to as a 'piercing needle') that penetrates the sample tube, and a probe (referred to as a 'sampling probe') aspirating blood from inside the needle and placing it in a sample tube.

An inherent disadvantage of this system is that the piercing needle cuts off part of the cap when penetrating the sample tube, with the rubber residue entering the hydraulic or counting system inside the instrument. To prevent this, we used filters in our large automatic hematology analyzers, however this requires maintenance work, and is not practical for the general practitioner market, the main market of the Microsemi series. Implementing the piercing needle and sampling probe as separate mechanisms increases the size of the instrument, again presenting problems in the market. On the LC-661, we therefore uses a piercing probe that functions both as a piercing needle and a sampling probe.

The outside diameter of the piercing probe is the same as the conventional sampling probe, which is less than half the diameter of the piercing needle installed on our large automatic hematology analyzer. The end of the probe is sharp, and thus able to penetrate the caps of vacuum sample. We also moved the blood aspiration inlet to the side of the piercing probe (Figure 4).

The rubber residue caused by penetrating the caps of vacuum sample tubes are primarily due to the following two phenomena.

(1) The rim of the piercing needle opening gouges through the cap

(2) The cap is peeled off due to the friction between piercing needle and cap

On the LC-661, sharpening the end of the piercing probe and moving the blood aspiration inlet to the side of the piercing probe control (1) above. Furthermore, by reducing the outside diameter, (2) can be significantly reduced. In addition, penetration of the caps of sample tubes and blood aspiration are performed by one unit, and thus only one mechanism is required, contributing to down sizing of instrument.

A requirement for the cap piercing function is the ability to release the pressure inside the vacuum sample tube. After a patient blood sample is aspirated, the vacuum sample tube is in a sealed state, however the pressure inside may vary. If the volume of a patient blood sample is less than the prescribed amount for the vacuum sample tube, pressure inside the vacuum sample tube will become negative. On the other hand, the pressure in the vacuum sample tube may also be positive depending on differences in the patient's blood temperature, the storage temperature, and the instrument temperature. If the vacuum sample tube is set in the instrument, the amount of blood that the instrument aspirates is affected by the pressure inside the vacuum sample tube. This problem is resolved by releasing the pressure inside the vacuum sample tube when the cap is pierced.

The LC-661 piercing needle appears to be in one piece, however the interior is of a double structure, with each part having an independent opening. One part is for aspirating blood, as mentioned previously, and the other (the release outlet) is for releasing the pressure inside the vacuum sample tube. This release outlet passes through an air syringe inside the instrument and is connected to a waste discharge line. Any blood spatters resulting from pressure release (if pressure in the vacuum sample tube is positive) can therefore be treated inside the instrument, reducing the likelihood of customer blood infections during maintenance.

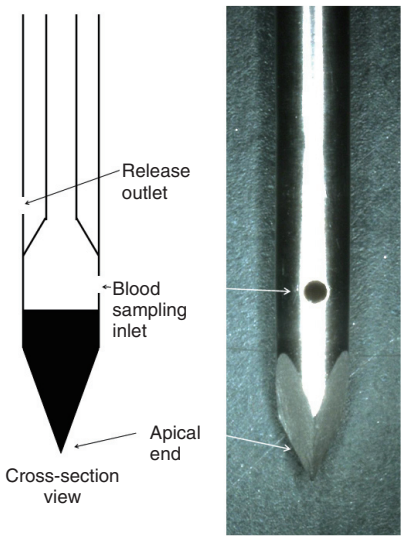

Figure 4 Piercing Probe Structure

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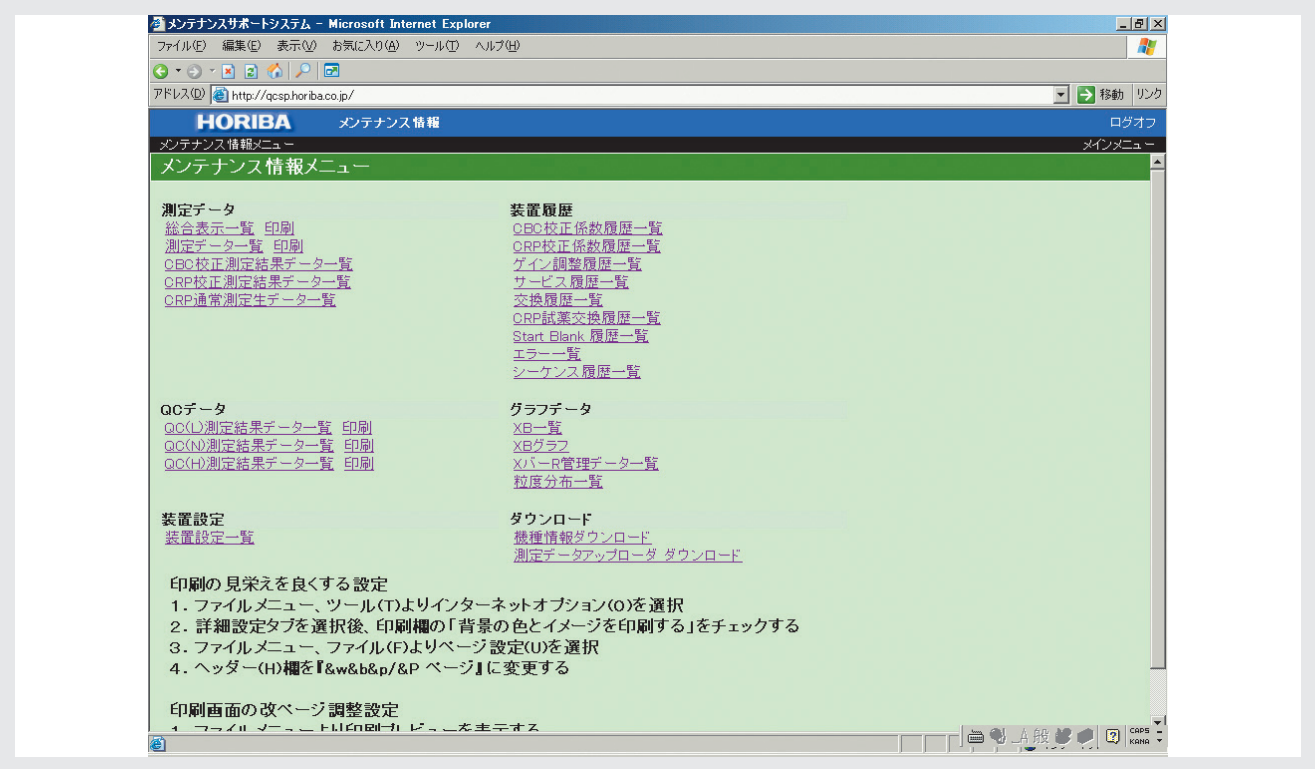


Figure 5 Maintenance Support System

Maintenance Information Recording Function: LC-660, LC-667, LC-661

With the conventional LC-550 and LC-178CRP models, information on the status of the instrument in the event of a problem at a customer's facility was transmitted from the customer to the distributor, then to HORIBA Techno Service, and finally to the HORIBA , Ltd. This complex process of transmission sometimes resulted in problems with the accuracy of the information obtained. Furthermore, the information was often out-of-date..

With the Microsemi series, both customer instrument measurement results and exclusive maintenance information are recorded in the instrument itself, and the instrument has a function that allows users to download the information to a compact flash card, or send it via the Internet. Distributor personnel or sales personnel upload this information to our exclusive maintenance support system, and team members able to access the system can view the information (Figure 5). The Microsemi series therefore differs from the conventional system in that the reported information is transmitted via fewer people, facilitating an accurate understanding of instrument status, and ensuring a rapid solution to the problem.

Maintenance information, includes instrument setting information, operating history, error history, and instrument status information available during measurement and at startup, as well as interim calculation values for measurement results. Comprehensive analysis of all this information makes it possible to analyze the cause of the problem remotely from the machine, providing a system that allows us to quickly present research results and issue technical advice to site personnel and customers in the field.

Example of Utilizing Maintenance Information

In one example, the count value during measurement was not constant (variance flag "$"), and this frequently occurred a number of times. There had previously been examples where this was the result of noise or the contamination of the counting unit, however by analyzing the maintenance information, we found that temperature was low during measurement. The low temperature slowed down the hemolytic reaction speed, causing the problem. We requested the customer to improve the installation environment where the instrument was installed, and the problem was resolved.

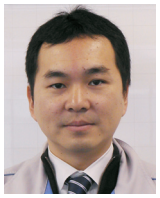
Conclusion

We introduced five new technologies in this paper, however the Microsemi series incorporates too many other technologies and innovations to present here. Many of the functions make the instruments easier for the customer to use, and we plan to continue in this direction in the future. Also, the maintenance information recording function not only helps analyze causes of problems when they occur, but helps predict problems and provides a basic data that can be used to develop new products.

The Microsemi series consists of the three current models. While this paper is titled "Microsemi Series Now Complete", this is only the first segment, and we plan to develop our technology in various other directions such as veterinary instruments.

Reference

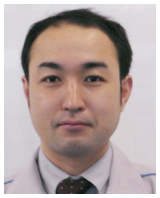
[1] Toru Inaba and Naohisa. Fujita Deducing Clinical Conditions Based on Fever Types and Clinical Profiles: Screening Examinations. Clinical Pathology Review, Special Edition No. 143, 2009;35-40.



Soichi Oue
Medical Development Dept.
Application Development Center
HORIBA Ltd.

Takaaki Shindo
Medical Development Dept.
Application Development Center
HORIBA Ltd.

Itsuo Ito
Medical Development Dept.
Application Development Center
HORIBA Ltd.



Hiroki Nagaoka
Medical Development Dept.
Application Development Center
HORIBA Ltd.

Akiko Sakato
Medical Development Dept.
Application Development Center
HORIBA Ltd.