



EzDrop 1000/1000C

Micro-Volume / Cuvette Spectrophotometer BRED-1000 / BRED-1000C

Operation Manual

Ver 1.2

Table of Contents

1. Saf	ety Precautions	1	
1.1	Intended Use	1	
1.2	General Instrument Safety	1	
1.3	Chemical Waste Safety	2	
2. Ge	neral Description	3	
2.1	Features	3	
2.2	Product Overview	4	
3. Ge	tting Started	6	
3.1	Unpacking	6	
3.2	Initial Operation	7	
3.3	Main Menu	8	
3.4	Detection Arm Opening/Closing	10	
3.5	Adjusting the Path Length Selector	11	
3.6	Basic Operation	13	
4. 🔖	Nucleic Acid	17	
4.1	Overview of Screen Features	17	
4.2	Measuring Nucleic Acid Sample	20	
4.3	Calculation	22	
5. 🖉	Protein A280	23	
5.1	Overview of Screen Features	23	
5.2	Measuring Protein Sample	26	
5.3	Calculation	28	
6. 🌌	Protein Assay	29	
6.1	Overview of Screen Features	29	
6.2	Measuring Protein Assay Sample	34	
6.3	Calculation	38	
7. 员	OD600	39	
7.1	Overview of Screen Features	39	
7.2	Measuring OD600 Sample	42	
7.3	Calculation	44	
8. 📠	Kinetics (1000C only)	45	
8.1	Overview of Screen Features	45	
8.2	Measuring Kinetics Sample	50	

	9.		Microarray	52	
		9.1	Overview of Screen Features	52	
		9.2	Measuring Microarray Sample	57	
_		9.3	Calculation	60	
	10	. 🤺	Labeled Protein	63	
		10.	1 Overview of Screen Features	63	
		10.2	2 Measuring Labeled Protein Sample	68	
_		10.3	3 Calculation	71	
	11.	Ľ	Standard Curve	74	
		11.	1 Overview of Screen Features	74	
		11.2	2 Measuring User-defined Standard Curve Sample	79	
_		11.3	3 Calculation	82	
	12	. •••••	UV-Vis	83	
		12.	1 Overview of Screen Features	83	
		12.2	2 Measuring UV-Vis Sample	87	
	13	F(x)	Factor	89	
		13.	1 Overview of Screen Features	89	
		13.2	2 Measuring Factor Sample	93	
		13.3	3 Calculation	95	
	14	•	System Menu	96	
		14.	1 👼 Date & Time	96	
		14.2	2 📣 Beep Sound	96	
		14.3	3 🔅 Brightness	96	
		14.4	4 🏟 Indicator Light	96	
		14.		97	
		14.0	5 🔢 Self-Test	97	
		14.	About B III Operation Manual	97 07	
		14.0		97	
		14.	10 X Service	98	
	15		O History	99	
		15.	1 Duplicating Protocol Setting	100	
		15.2	2 Copying a Result	100	
		15.3	3 Viewing a Result	100	

16.1 Creating a New User Folder	
5	101
16.2 Deleting a User Folder	102
16.3 Viewing a User Folder	102
16.4 Editing a User Folder	102
16.5 Using a Flash Drive as a User Folder	102
I7. PC Connection	103
8. Maintenance	106
18.1 Cleaning the EzDrop 1000/1000C	106
18.2 Cleaning the Quartz Glass of Sample Window/Cover	106
18.3 Cleaning the Cuvette Holder (1000C only)	107
18.4 Re-Coating of Hydrophobic Layer	107
18.5 Annual Maintenance	108
18.6 Replacement	108
19. Troubleshooting	109
19.1 Error Messages	110
Appendix A: Technical Specifications	113
Appendix B: CE & UKCA Declaration	115
Appendix C: Order Information	116

1 Safety Precautions

Before using the EzDrop 1000/1000C for the first time, please read this entire Operation Manual carefully. To guarantee problem free, safe operation of the EzDrop 1000/1000C, it is essential to observe the following section.

1.1 Intended Use

EzDrop 1000/1000C is a Micro-Volume / Cuvette Spectrophotometer with a wide wavelength of 190 ~ 1000 nm. It can measure and quantify liquid samples, such as nucleic acid or proteins, in a specific wavelength. This instrument is intended to be used by trained personnel to perform solution analyses. In this manual, we assume that the user has knowledge of basic laboratory procedures and spectroscopic analysis.

1.2 General Instrument Safety

PHYSICAL INJURY HAZARD.

Using the instrument in a manner not specified by Blue-Ray Biotech may result in personal injury or damage to the instrument.

1.2.1 Transportation and Storage

This instrument should be transported and stored in an environment with a temperature of -10 \sim 60 °C, relative humidity 20 \sim 80%.

1.2.2 Installation and Operation

- 1. Do not use the instrument in a potentially explosive environment or with potentially explosive chemicals.
- 2. Avoid placing the instrument in direct sunlight.
- 3. Install the instrument in a location free of excessive dust.
- 4. Install the instrument in a room with a temperature of $15 \sim 30$ °C, relative humidity $20 \sim 80$ %.
- 5. Choose a flat, stable surface capable of bearing the weight of the instrument.
- 6. Make sure the power source conforms to the required power supply specifications.
- 7. To avoid electric shock, make sure the instrument is plugged into a grounded electrical outlet.

- 8. Do not allow water or any foreign objects to enter the various openings of the instrument.
- 9. If the instrument is not in use, please keep the detection arm closed.

1.2.3 Cleaning, Decontaminating, and Servicing the Instrument

Before using a cleaning or decontamination method other than those recommended by the manufacturer, verify with the manufacturer that the proposed method will not damage the equipment.

Switch off and unplug the instrument before cleaning, servicing, or replacing the fuses.

Repairs should be carried out by authorized service personnel only.

1.2.4 Instructions for Removal from Use, Transportation, or Disposal

Do not dispose of this product as unsorted municipal waste. Follow local municipal waste ordinances for proper disposal provisions to reduce the environmental impact of waste electrical and electronic equipment (WEEE).

European Union Customers: Call your local Blue-Ray Biotech Distributor's Customer Service office for equipment pick-up and recycling.

1.3 Chemical Waste Safety

1.3.1 Chemical Waste Hazard

HAZARDOUS WASTE.

Refer to Material Safety Data Sheets and local regulations for handling and disposal.



2 General Description

The EzDrop 1000/1000C is a Micro-Volume / Cuvette Spectrophotometer that provides accurate results in only 3 seconds in an intuitive operational experience. It enables the measuring of samples from 190 ~ 1000 nm, a broad range wavelength which offers flexibility for experiment.

2.1 Features

- Large LCD touch panel enhances visibility and ease-of-operation.
- Robust and modern outlook design.
- Simple and easy-to-use graphical interface.
- Multiple built-in applications.
- Dual detection mode, Micro-Volume and Cuvette (1000C only)
- Heater & stirring function (1000C only)
- Fast measuring time, in 3 seconds.
- Wide wavelength range, 190 ~ 1000 nm.
- Quartz sample window, which protects the optical analysis system.
- A Nano hydrophobic coating layer on the quartz sample window/cover.
- Assist light to avoid sample pipetting error.
- Cushioned detecting arm to protect it from shock and impact during closure.
- Auto measurement function to increase experiment efficiency.
- To automatically create operating history.
- PC connection for flexible data management.



Figure 1. Front View

Table 1. Detailed Description for Front View

Part Name	Function
Detection Arm	Detection arm with cushioning design to reduce closing impact. The detection arm must be closed when performing sample measurements.
Cuvette Holder (1000C only)	Use a cuvette corresponding to the detection wavelength. Cuvette specification: Width: 12.5 mm, Length: 12.5 mm, Height: 45 mm, Beam height: 8.5 mm. Cuvette holder with heater (37 ~ 45 °C) and stir function (1 ~ 8 speed corresponding to 150 ~ 850 rpm).
Path Length Selector	The path length (0.5 / 0.05 mm) of EzDrop 1000/1000C can be adjusted manually according to the absorbance (concentration) range difference by the path length selector.
7" Touch Panel	It is a high-resolution color LCD display (1280×800) with capacitive touch panel. It displays the current status of the system and allows the user to operate the instrument.



Table 2. Detailed Description for Back View

Part Name	Function
Power Switch	Power On/Off switch.
Power Adaptor Socket	Connect instrument to the AC power supply.
USB-B Port	For PC connection
Product Label	Indicates the product name, model, serial number, power specification, and other important information.

3 Getting Started

3.1 Unpacking

Once you open the EzDrop 1000/1000C package, remove all the EPE foam, a piece of which is under the detection arm (Figure 3), and the plastic bag. Confirm that all the following items are included:

- EzDrop 1000/1000C x 1
- Quick Operation Guide x 1
- Power Adapter x 1
- Power Cord x 1
- Calibration Report x 1
- USB-B Cable x 1
- 10 mm Quartz Cuvette w/ cover, stirrer bar in accessory box x 1 (1000C only)

If any items are missing, damaged, or any incorrect items are included in the package, please contact your local Blue-Ray Biotech distributor or sales representative immediately.



Figure 3. Take Out the EPE Foam under the Detection Arm



3.2 Initial Operation

Place the instrument on a steady and flat table. Keep the front and rear of the device at least 10 cm from the wall or other devices. Check that the power source is compatible with your device's fuse rating input. Connect the power cord and power adaptor, plug it into the power adaptor socket at the back of the instrument (Figure 2).

Switch on the instrument using the power switch at the back of the instrument (Figure 2). The LCD display will show the boot screen, start initiation progress, and then the "**EzDrop**" will be displayed (Figure 4). Please **DO NOT** open the detection arm until system diagnosis is completed. Tap the "**EzDrop**" icon to log into the **Main Menu** (Figure 5) and start operation.



Figure 4. EzDrop 1000/1000C Home Page

Note

1. The power adapter is foolproof, which requires more force to plug in and out.

2. Switch off the instrument when not in use.

3.3 Main Menu

On the **Main Menu**, there are some information indicating the status of the EzDrop 1000/1000C. Please refer to the following Figure 5 and Table 3 & Table 4 for the detailed description.



Figure 5. Main Menu Overview

Table 3. Detailed Description for the 10 Applications on Main Menu

lcon	Application	Description
A COL	Nucleic Acid	To measure the concentration and purity of nucleic acid samples at 260 nm, such as DNA, RNA, and Oligo.
280 A280	Protein A280	To measure the concentration and purity of protein samples at 280 nm.
<u>a</u>	Protein Assay	To measure the concentration of protein samples on BCA (562 nm), Bradford (595 nm), Lowry (650 nm), and Pierce 660 (660 nm).
OD 600	OD600	To measure the concentration of microbial cells at 600 nm.



10	Kinetics (1000C only)	To monitor the time-based continuous absorbance change of the sample in the cuvette. Optional heater (37 \sim 45 °C) and stirring (1 \sim 8 speeds corresponding to 150 \sim 850 rpm) functions are available.
	Microarray	To measure the concentration of nucleic acid / fluorescent dye conjugation at a specific wavelength.
-/	Labeled Protein	To measure the concentration of protein / fluorescent dye conjugation at a specific wavelength.
~	Standard Curve	To measure concentration of unknown samples at a specific wavelength based on the developed standard curve.
ali fali fali	UV-Vis	To measure absorbance on a specific wavelength between 190 ~ 1000 nm.
F(x)	Factor	User-defined analysis method to measure unknown samples.

Table 4. Detailed Description for the 3 Tools & Setting on Main Menu

Icon	Application	Description
¢	System	System setting.
-C)	History	Duplicate protocol setting / View previous result.
	User	User folder management.
O UN		

3.4 Detection Arm Opening/Closing

• Open the detection arm

Use your index finger and thumb to pinch the front edge of the detection arm and lift to the end as shown in Figure 6.



Figure 6. Opening the Detection Arm

• Close the detection arm

Use your index finger and thumb to pinch the front edge of the detection arm, and let the detection arm down gently to the correct position as shown in Figure 7. The cushion design on the detection arm reduces impact even when letting the detection arm drop.

Note

If the instrument is not in use, please keep the detection arm closed.





Figure 7. Closing the Detection Arm

3.5 Adjusting the Path Length Selector

The path length of EzDrop 1000/1000C for micro-volume detection is adjusted manually. The measurement range of **0.5 mm Path Length** is **0.04** ~ **30** A, and the range of **0.05 mm Path Length** is **20** ~ **400** A (Table 5).

Before starting to do the sample measurement, check if the light path length selector is in the right position. When the path length selector is in the vertical position to the detection arm (Figure 8), it represents the 0.5 mm Path Length. In the horizontal position (Figure 9), it represents the 0.05 mm Path Length. Hold the knob of the path length selector by index finger to adjust path length between 0.5 mm and 0.05 mm.

The EzDrop 1000C includes a cuvette holder for more applications (Figure 10 & 11), such as kinetics and cell culture. Users can use 10, 5, 2, 1, 0.5, 0.2, 0.125, 0.1 mm quartz glass or plastic cuvettes for detection. The measurement range is from **0.002** ~ **1.5 A**. The cuvette system has another 2 functions:

- Heater (37 ~ 45°C)
- Stirring (1 ~ 8 speed corresponding to 150 ~ 850 rpm)

	1. A path length of 0.5 mm should be selected for cuvette mode.
	2. Use a cuvette corresponding to the detection wavelength.
	3. Cuvette specification:
ote	Width: 12.5 mm, Length: 12.5 mm, Height: 45 mm, Beam height: 8.5 mm.
	4. The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).

5. Avoid using cuvettes with scratches or fingerprints on the optical path surface, it may affect the detection accuracy. Use lint-free paper to wipe the optical path surface before inserting it into the cuvette holder.

Table 5. Path Length Operation Guide

Path Length Operation Guide				
Path Length	Micro-Volume 0.5 mm	Micro-Volume 0.05 mm	Cuvette 10, 5, 2, 1, 0.5, 0.2, 0.125, 0.1 mm (1000C only)	
Position	Figure 8. 0.5 mm Path Length	Figure 9. 0.05 mm Path Length	Light Path	



Figure 11. The Light Path Arrow and Beam Height of Cuvette Holder (1000C only)



3.6 Basic Operation

3.6.1 Application Screen

The application screen can be separated into 3 parts; there are different display interfaces according to different applications. ×

- Information Tab (Figure 12 & Table 6) •
- Data Information Report Area •
- Functions Icons (Table 7) •



Figure 12. Application Screen

Table 6. Detailed Description for the Information Tab Page

Tab Page	Description
Setting	The page only exists in Kinetics (1000C only), Microarray, Labeled Protein, Standard Curve, UV-Vis and Factor applications. User need to enter protocol settings before measurement.
Standard	This page will only display standard data, and only exists in Protein Assay and Standard Curve . A standard curve needs to be established for sample measurement.
Dye List	The page only exists in Microarray and Labeled Protein applications. Display built-in dye information, users can also add new dye information.

Data	The page for the detailed sample data information and setting.
Table	The page of the total samples data report.
Graph	The page for graph results. The horizontal axis represents the Wavelength (nm) and the vertical axis shows Absorbance (10 mm) .
Trend	The page only exists in the Kinetics application (1000C only). The horizontal axis represents the Time and the vertical axis shows Absorbance (10 mm) .
Table 7. Detailed Descrip	otion for the Function Icons

Table 7. Detailed Description for the Function Icons

lcon	Function	Description
	Blank	To establish a reference absorbance.
	Auto Run: On	The instruction of Auto Run function is on.
	Auto Run: Off	The instruction of Auto Run function is off.
Measure		To do sample/standard measurement.
	Delete	To delete sample measurement data. Note: Standard measurement data cannot be deleted but can be overwritten.
	Save Result	To save the result.
	Back	Return to the previous page.
STOP Stop		To abort the measurement.

3.6.2 Basic Measurement Operation

- 1. Choose an application.
- Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be 20 ~ 400 A, it can be switched to 0.05 mm.

Note	A path length of 0.5 mm should be selected for cuvette mode (1000C only).	
3. Sel info	ect the correct Method / Path Length / Baseline Correction or others required bormation in Setting Tab Page based on the experiment protocol.	

- 4. Use lint-free paper to clean the sample window/cover.
- 5. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

	The liquid level in the cuvette should be higher than the instrument optical path, the beam height
Note	of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the
	arrow on the cuvette holder (Figure 11).

- 6. Lower the detection arm and tap **Blank**.
- 7. Lift the detection arm, then clean the sample window/cover with lint-free paper.
- 8. Enter the sample name (optional) or use the auto-numbering system. Then, pipette $1 \sim 2 \mu L$ of sample solution onto sample window, or insert a sample in a cuvette into the cuvette holder (1000C only).
- 9. Lower the detection arm and tap \checkmark Measure.

Note

If there are many samples, turn on Auto Run \rightarrow \rightarrow , measurement will be made automatically after the detection arm has been lowered and sample naming will be sequential.

10. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.

11. Tap Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up \bigcirc . Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer <u>Section 16.1 Creating a New User Folder</u>.

Lab: Blue-Ray Biotech	2022-12-15 09:59	
Use	er Menu	
	User Folder	
Flash Drive Public	TEST Public Folder	
	Customer USB Memory Device	
	Function Icons	
> ⊗ New User	Back	



4 🦠 Nucleic Acid

This application will measure the samples absorbance at 260 nm, which is the peak of nucleic acid absorbing UV light, to calculate the concentration. The unit is $ng/\mu L$. The purity of nucleic acid samples can be estimated by two absorbance ratios, A260/A280 and A260/A230.

4.1 Overview of Screen Features

The information tab bar has 3 tab pages:

- Data Tab Page
- Table Tab Page
- Graph Tab Page.

4.1.1 Data Tab Page

On the data tab page of the Nucleic Acid application (Figure 13), see the table below for details (Table 8).



Figure 13. Data Tab Page of Nucleic Acid Application

Table 8. Detailed Description for the Data Tab Page of Nucleic Acid Application

ltem	Description
[conc.]	The concentration is calculated from absorbance at 260 nm, and the unit is ng/µL .
A260	Displays the absorbance at 260 nm, which is normalized to a 10 mm Path Length equivalent.
A260/A280	Displays the ratio of the absorbance at 260 nm and 280 nm. If a ratio is detected out of range, an alert icon <u>N</u> will pop up. dsDNA < 1.75, RNA < 2.0, ssDNA < 1.75.
A260/A230	Displays the ratio of the absorbance at 260 nm and 230 nm.
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.
Method	Includes sample types, like dsDNA, ssDNA, RNA, miRNA, Oligo and Custom Factor (15.0 ~ 150 ng*cm/ul) . The default is "dsDNA".
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only).
Baseline Correction	The wavelength for bichromatic normalization is 340 nm . This is an optional function and the default is "ON".

4.1.2 Table Tab Page

The table tab page will show all the data results (Figure 14). If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. Users can also tap the **Sample** column to edit the sample name.

Lab	: Blue	-Ray Biotech		and the second second			2023-02-	09 10:41
				Nucleic Acid:	New			
		Data		Table		Gr	aph	
#	[×]	Sample	ng/µL	A260/A280	A260/A230	A260	A280	A23
1		Blank_1	0.000	0.00	0.00	0.0000	0.0000	0.000
2	[x]	Sample_1	0.123	4.20	1.75	1.0000	1.0000	2.200
_								
	L	=		~				
	Bla	nk Auto Ru	n Me	asure	Delete	Save Result	Bad	:k

Figure 14. Table Tab Page of Nucleic Acid Application

4.1.3 Graph Tab Page

Select more than **1** sample data in **[/]** column of the **Graph Tab Page** to overlay the graph (Figure 15). It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.



Figure 15. Graph Tab Page of Nucleic Acid Application

4.2 Measuring Nucleic Acid Sample

- 1. On the main menu, tap 🍫 to enter Nucleic Acid application.
- 2. Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be $20 \sim 400 \text{ A}$, it can be switched to 0.05 mm.

Note A path length of **0.5 mm** should be selected for cuvette mode (1000C only).

On the Data Tab Page, select the correct Method type or enter Custom Factor (15.0 ~ 150 ng*cm/ul) based on the experiment protocol. The default selection is dsDNA.



- 4. Select the correct **Path Length** based on the experiment protocol. For micro-volume detection, the instrument will detect the current path length selector position automatically. For cuvette mode detection, it should be selected manually (1000C only).
- 5. The **Baseline Correction** (340 nm) function is optional, and can be turned on/off anytime. The default is "ON".
- 6. Use lint-free paper to clean the sample window/cover.
- 7. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).

8. Lower the detection arm and tap **Blank**.

Note

9. Lift the detection arm, then clean the sample window/cover with lint-free paper.

- 10. Enter the sample name in the **Name** bar (optional) or it will be named by auto-numbering. Then, pipette $1 \sim 2 \mu L$ of the sample solution onto the sample window, or insert a sample in a cuvette into the cuvette holder (1000C only).
- 11. Lower the detection arm and tap Measure.

NoteIf there are many samples, turn on Auto Run \rightarrow \rightarrow \rightarrow , measurements will be made
automatically after the detection arm has been lowered and sample naming will be sequential.

- 12. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.
- 13. Tap Bave Results to system or flash drive.
 - a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up \checkmark . Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer <u>Section 16.1 Creating a New User Folder</u>.



4.3 Calculation

In the Nucleic Acid application, a modified Beer-Lambert equation is used to calculate concentrations with absorbance and factor as follows, with or without baseline correction:

Without baseline correction:

 $c = A260 \times \epsilon$

With baseline correction

 $c = (A260 - A_{Baseline}) \times \epsilon$

c = the nucleic acid concentration in ng/µl

A260 = the absorbance at 260 nm (10 mm Path Length)

ABaseline = the absorbance at baseline wavelength (10 mm Path Length)

 ϵ = the extinction coefficient of nucleic acid in ng*cm/µl

The general extinction coefficient used in the calculation of nucleic acid are shown in Table 9.

Table 9. Extinction	Coefficient of	f Nucleic Acids
---------------------	----------------	-----------------

Туре	Factor (ng*cm/µl)
dsDNA	50
ssDNA	33
RNA	40
miRNA	33
Oligo	33
Custom	15.0 ~ 150

5 🚆 Protein A280

This application will measure the samples absorbance at 280 nm, which is the peak of purified protein absorbing UV light, to calculate the concentration. The unit of protein concentration is **mg/mL**. The purity of homogenous protein can be estimated by absorbance ratios of A260/280.

5.1 Overview of Screen Features

The information tab bar has 3 tab pages:

- Data Tab Page
- Table Tab Page
- Graph Tab Page.

5.1.1 Data Tab Page

On the data tab page of Protein A280 application (Figure 16), see the table below for details (Table 10).



Figure 16. Data Tab Page of Protein A280 Application

Table 10. Detailed Description for the Data Tab of Protein Assay Application

Item	Description
[conc.]	The concentration is calculated from absorbance at 280 nm, and the unit is mg/mL.
A280	Display the absorbance at 280 nm, which is normalized to a 10 mm Path Length equivalent.
A260/A280	Displays the ratio of the absorbance at 260 nm and 280 nm. If a ratio of A260/A280 > 0.6, an alert icon 2 will pop up.
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.
Method	It includes sample types. like BSA, IgG Mouse, IgG Human, IgE Human, Lysozyme, SA Mouse, SA Human, 1A=1mg/mL and Custom Factor (0.001 ~ 100 g*cm/l). The default is "BSA".
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only).
Baseline Correction	The wavelength for bichromatic normalization is 340 nm . This is an optional function and the default is "ON".



5.1.2 Table Tab Page

The table tab page will show all the data results (Figure 17). If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. User can also tap the **Sample** column to edit the sample name.

Lab	: Blue	e-Ray Biotech		and the second	kowe		2023-02	-08 16:02	
				Protein A28	0: New				
		Data		Table	;		Graph	<u>A</u>	
#	[x]	Sample	mg/mL	A280	A260/A280	Method	Factor	λ (Co i	
1		Blank_1	0.000	0.0000	0.00	Custom	1	340n	
2	[x]	Sample_1	-0.002	-0.0015	-10.75	Custom	1	340n	
6									
	-	=	4	~)	
	Bla	ak Auto P	un Mo	asure	Delete	Save Pesult	Ra	ck	

Figure 17. Table Tab Page of Protein A280 Application

5.1.3 Graph Tab Page

Select more than **1** sample data in [$\sqrt{}$] column of the **Graph Tab Page** to overlay the graph (Figure 18). It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.

Lab: Blue-Ra	y Biotech			202	23-02-08 16:02
			280: New		
	Data	Tab	ble	Graph	
1.00 0.75 0.00 90000990 0.25 0.00 220	257 295 Waveleng	332 th (nm)	# [/] 370	Sample	mg/mL
Blank	Auto Run	Measure	Delete	Save Result	Back

Figure 18. Graph Tab Page of Protein A280 Application

5.2 Measuring Protein Sample

- 1. On the main menu, tap $\frac{22}{220}$ to enter **Protein A280** application.
- 2. Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be $20 \sim 400 \text{ A}$, it can be switched to 0.05 mm.

Note A path length of **0.5 mm** should be selected for cuvette mode (1000C only).

- On the Data Tab Page, select the correct Method type or enter Custom Factor (0.001 ~ 100 g*cm/l) based on the experiment protocol. The default selection is "BSA".
- 4. Select the correct **Path Length** based on the experiment protocol. For micro-volume detection, the instrument will detect the current path length selector position automatically. For cuvette mode detection, it should be selected manually (1000C only).
- 5. The **Baseline Correction** (340 nm) function is optional, and can be turned on/off anytime. The default is "ON".
- 6. Use lint-free paper to clean the sample window/cover.
- 7. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

Note The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).

- 8. Lower the detection arm and tap _ Blank.
- 9. Lift the detection arm, then clean the sample window/cover with lint-free paper.
- 10. Enter the sample name in the **Name** bar (optional) or it will be named by auto-numbering.. Then, pipette $1 \sim 2 \mu L$ of the sample solution onto the sample window, or insert a sample in a cuvette into the cuvette holder (1000C only).

11. Lower the detection arm and tap 🦯 Measure.

Note

If there are many samples, turn on Auto Run \rightarrow \rightarrow \rightarrow , measurement will be made automatically after the detection arm has been lowered and sample naming will be sequential.



- 12. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.
- 13. Tap Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up \bigcirc . Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer <u>Section 16.1 Creating a New User Folder</u>.



5.3 Calculation

In the Protein A280 protocol, a modified Beer-Lambert equation is used to calculate concentrations with absorbance and factor as follows, with or without baseline correction:

Without baseline correction:

 $c = A280 \times \epsilon$

With baseline correction

 $c = (A280 - A_{Baseline}) \times \epsilon$

c = the purified protein concentration in mg/mL

A280 = the absorbance at 280 nm (10 mm Path Length)

A_{Baseline} = the absorbance at baseline wavelength (10 mm Path Length)

 ϵ = the factor of purified protein factor in g*cm/l

The extinction coefficient used in the calculation of purified protein are shown in Table 11.

Туре	g*cm/l	l/g*cm
BSA	1.499	0.667
lgG Mouse	0.714	1.4
lgG Human	0.735	1.36
lgE Human	0.654	1.53
Lysozyme	0.379	2.64
SA Mouse	1.493	0.67
SA Human	1.718	0.582
1A=1mg/mL	1	N/A
Custom	0.001 ~ 100	N/A

Table 11. Extinction Coefficient of Purified Proteins



6 🎤 Protein Assay

The Protein Assay application will measure the absorbance of homogeneous proteins at specific wavelengths based on protein assay reagents. The unit of this application is **mg/mL**.

6.1 Overview of Screen Features

The information tab bar has 4 tab pages:

- Standard Tab Page
- Data Tab Page
- Table Tab Page
- Graph Tab Page

6.1.1 Standard Tab Page

The standard tab page (Figure 19) is the page to measure standard samples absorbance at specific wavelength to establish a standard curve. It only shows the standard data result. See the table below for details (Table 12).

	Lab: Bl	ue-Ray Biot	ech	2023-02-09 11:				1:05			
					Protein Assay: New				— Application		
		Standard				Table		Graph	Information Tab		
		mg/mL	Abs	Avg. Abs	[/]	Method	BC	A			
	Std1.1	0.01	0.0267	0.0267	[•]	Path Length	0.5m	nm ng/mt	Selected Path Length		
	Std2.1	0.05	0.0076	0.0076	[1]	Baseline C	orrection	750nm	Correction Wavelength		
	Std3.1	0.08	0.0274	0.0274	[1]				(Optional)		
Standard Data	Std4.1					Curve Type	Linea	ar	Standard Curve Type		
Information	Std5 1					Replicates	1		Number of Replicates		
	Stub.	State 1				— 🧹 Generate Standard C					
	Std6.1										
	Std7.1	Std7.1				y=-0.0164x+0.0213, R ² =0.0026					
	Std8.1										
Function Icons –		_		~							
	В	lank	Auto Run	Measu	ure	Delete	Save Result	Back			

Figure 19. Standard Tab Page of Protein Assay Application

Item	Description
mg/mL	This is the standard concentration column. The value is inserted by the users. The range is from 0.001 ~ 9.999 .
Abs	The absorbance measured at different wavelengths according to the protein assay type.
Avg. Abs	The average absorbance of the standard repetition. It is calculated automatically.
Method	It includes 4 protein assay types, like BCA Assay (562 nm), Braford Assay (595 nm), Lowry Assay (650 nm) and Pierce 660 Assay (660 nm). The default is "BCA Assay (562 nm)".
Path Length	For micro-volume detection, it can only be carried out at 0.5 mm Path Length . For cuvette mode detection, it should be selected manually (1000C only).
Baseline Correction	The wavelength for bichromatic normalization is different from protein assays. Default values for the baseline correction are depending on selected protein assay type. The baseline correction for BCA Assay , Braford Assay and Pierce 660 Assay is 750 nm, Lowry Assay is 405 nm . This is an optional function and the default is "ON".
Curve Type	The standard curve types the user can select: linear, interpolation, and 2nd order polynomial.
Repetition	The number of standard repetitions. The default value is "1" and the maximum is 3.
Generate Standard Curve	Select the standard data by tapping the $[]$ column to establish the standard curve and the formula will be display in the line below.

Table 12. Detailed Description for the Standard Tab Page of Protein Assay Application



6.1.2 Data Tab Page

On the data tab page of Protein Assay application (Figure 20), see the table below for details (Table 13). This page only shows the sample data, the standard data and formula is shown in standard tab page.



Figure 20. Data Tab Page of Protein Assay Application

Table 13. Detailed Description for the Data Tab Page of Protein Assay Application

Item	Description					
[conc.]	The detected absorbance value will be brought into the established standard curve formula to calculate the sample concentration. The unit is mg/mL .					
A562/ A595/ A650/ A660	Displays the absorbance at protein assay kit-requested wavelength. BCA Assay (562 nm), Braford Assay (595 nm), Lowry Assay (650 nm) and Pierce 660 Assay (660 nm). The absorbance is normalized to 10 mm Path Length equivalent.					
Dilution Factor	Set a dilution factor for manual diluted samples. The default is "1" and the range is from 1 ~ 1,000,000 . It should be entered before measurement.					
Recalculated [conc.]	[conc.] multiplied by dilution factor. The unit is mg/mL .					
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.					

Method	It includes 4 protein assay types, such as BCA Assay (562 nm), Braford Assay (595 nm), Lowry Assay (650 nm) and Pierce 660 Assay (660 nm). The default is "BCA Assay (562 nm)". On the data tab page, the method setting is the same as the standard tab page.
Path Length	Micro-volume detection can only be carried out at 0.5 mm Path Length . For cuvette mode detection, it should be selected manually (1000C only). The path length setting on the data tab page is the same as the standard tab page.
Baseline Correction	The wavelength for bichromatic normalization is different from protein assays. Default values for the baseline correction are dependent on the protein assay type selected. The baseline correction for BCA Assay , Braford Assay and Pierce 660 Assay is 750 nm, Lowry Assay is 405 nm. This is an optional function and the default is "ON". The baseline correction setting on the data tab page is the same as the standard tab page.

6.1.3 Table Tab Page

The table tab page will show only the sample data result (Figure 21). The standard data is NOT included on this page. If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. Users can also tap the **Sample** column to edit the sample name.

La	b: Blue-Ra	ay Biotech					2023-0	2-09 11:05		
				Protein Assay: Ne	w					
	Star	dard	Data		Table			Graph		
#	# [x]	Sample	mg/mL	Dilution Factor	Recalc	Method	A562	λ (Corr.		
	L			<u>~</u> [5		
	Blank	Auto Ru	n Me	asure De	lete	Save Result	В	ack		

Figure 21. Table Tab Page of Protein Assay Application
6.1.4 Graph Tab Page

In the Protein Assay application, there are 2 graph types: the **Spectrum View** and **Standard Curve View** (Figure 22). Select more than **1** sample data in $[\sqrt{}]$ column of the **Graph Tab Page** to overlay the graph. It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.

Lab: Blue-Ray B	liotech				2023-02-09 11:05
Standard	d	Data	Table		Graph
1.00			# [/] Sample	mg/mL
(0.75 E E O					
orbance (10 50					
sq 9 0.25					
0.00	487 575 Waveleng	662 th (nm)	750		
Spectrum V	View	Std. Curve View			
				6	-
Blank	Auto Run	Measure	Delete	Save Result	Back

Figure 22. Graph Tab Page of Protein Assay Application

6.2 Measuring Protein Assay Sample

- 1. On the main menu, tap 🖉 to enter **Protein Assay** application.
- 2. Lift the detection arm and switch the path length selector to **0.5 mm**.
- 3. On the **Standard Tab Page**, select the correct **Method** type based on the experiment protocol. The default selection is "BCA".

Method		
Туре	Wavelength	
BCA	562nm	
Bradford	595nm	
Lowry	650nm	
Pierce 660	660nm	
✓		

- 4. Select the correct **Path Length** based on the experiment protocol. For micro-volume detection, it can only be carried out at **0.5 mm Path Length**. For cuvette mode detection, it should be selected manually (1000C only).
- 5. The **Baseline Correction** function is optional, and can be turned on/off anytime. Default values for the baseline correction are dependent on the protein assay type selected. The default is "ON".
- 6. Select the correct **Curve Type** and **Replicates** $(1 \sim 3)$ based on the experiment protocol.

	a. Linear curve type requires minimum 2 DIFFERENT concentrations to establish standard curve.
Note	b. Interpolation curve type requires minimum 2 DIFFERENT concentrations to establish standard curve.
	c. 2nd order polynominal curve type requires minimum 3 DIFFERENT concentrations to establish standard curve.

7. Tap on a cell in the mg/mL column to enter the concentration of standard samples.

Lab: Blue-Ray Bio	tech					2023-02-09	9 11:05
Standard		Data		Table		Graph	
mg/mL	Abs	Avg. Abs	[/]	Method	B	CA	V
Std1.1				Path Length	0.9	Smm	
Std2.1				Baseline G	0.01 - Correction	^{8 mg/mL} 750nm	•
Std3.1				Curve Type	Lir	ear	•
Std5.1				Replicates		1	•
Std6.1				Generate S	Standard Curve		
Std7.1							
Std8.1							
					B	•	
Blank	Auto Run	Measu	ire	Delete	Save Result	Back	



- 8. Use lint-free paper to clean the sample window/cover.
- 9. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

Note The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).

- 10. Lower the detection arm and tap **Blank**.
- 11. Lift the detection arm, then clean the sample window/cover with lint-free paper.
- 12. Pipette a $1 \sim 2 \mu L$ standard sample solution onto the sample window, or insert the standard sample cuvette into the cuvette holder (1000C only).
- 13. Tap on a cell in the **Abs**. column and tap Measure to establish standard data. If the number of replicates is higher than 1, it will automatically jump to the next cell. However, if the number of repetitions of this concentration has been measured and finished, it will not automatically jump to the next concentration.



Note

If there are many standard samples, turn on Auto Run \rightarrow \rightarrow measurement will be made automatically after the detection arm has been lowered and sample naming will be sequential.

14. Select the standard data by tapping the [/] column and check **Generate Standard Curve** to establish the standard curve and the formula will be display in the line below.

ab: Blu	e-Ray Biote	ech					2023-02-0	9 11:0!
	Standard		Data		Table		Graph	
	mg/mL	Abs	Avg. Abs	[/]	Method	B	CA	
Std1.1	0.01	0.0267	0.0267	[1]	Path Length	0.5	Smm	
Std2.1	0.05	0.0076	0.0076	[1]	Baseline	0.01 - Correction	8 mg/mt 750nm	•
5td3.1	0.08	0.0274	0.0274	[1]				
Std4.1					Curve Type	Lin	lear	
Std5.1				1	Replicates		1	
Std6.1				((Construction of the second sec	Standard Curve		
Std7.1						17 02-0 0026		
Std8.1					C	15, K-0.0020		
,								
						E	•	

Note

Note

If user need to rebuild the standard curve, uncheck **Generate Standard Curve** and remeasure standard sample or input new standard data.

- 15. Jump to the Data Tab Page to measure protein samples.
- 16. Enter the sample name in the **Name** bar (optional) or it will be named by auto-numbering. Then, pipette $1 \sim 2 \mu L$ of the sample solution onto the sample window, or insert a sample in a cuvette into the cuvette holder (1000C only). If the protein sample has been diluted, users can enter **Dilution Factor**, the actual concentration will be automatically calculated by multiplying the measured concentration [conc.] by the dilution factor and the result will be displayed in **Recalculated [conc.]**.
- 17. Lower the detection arm and tap Measure.
 - If there are many samples, turn on Auto Run → → → , measurement will be made automatically after the detection arm has been lowered and sample naming will be sequential.
 - 2. It is suggested to clean the sample window/cover when exchanging different concentrations of samples.
- 18. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.
- 19. Tap Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will **pop up** is and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.



b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer <u>Section 16.1 Creating a New User Folder</u>.

User Menu Flash Drive Public TEST User Folder Public Folder Customer USB Memory Device	5
Flash Drive Public TEST User Folder Public Customer USB Memory Device	
Flash Drive Public TEST Public Folder Customer USB Memory Device	
Customer USB Memory Device	
Function Icons	
Rew User Back	

6.3 Calculation

For protein quantification, the concentration can be calculated by measuring the final absorbance of colorimetric samples and standards.

The BCA Assay is based on the reduction of Cu^{2+} by alkaline in the protein. This has a peak absorbance at 562 nm and has a baseline correction at 750 nm.

The Bradford Assay is based on the protein complex with Coomassie blue dye. This measures the absorbance at 595 nm and has a baseline correction at 750 nm.

The Lowry Assay is based on the protein complex with copper. This has a peak absorbance at 650 nm and has a baseline correction at 405 nm.

The Pierce 660 Assay is based on the binding of a proprietary dye-metal complex to protein in acidic conditions that causes a shift in the dye's absorption maximum. This has a peak absorbance at 660 nm and has a baseline correction at 750 nm.

Note
 The assay kits describe the detailed protocols. Please set the protocol according to the kits' instructions.
 If users have other protein assay kits needing to establish a standard curve, please refer to Section 11 Standard Curve Application.



7 🔛 OD600

This application will measure microbial cell samples absorbance at 600 nm, which can be used for monitoring the samples growth rate. The range of the light path length is shown in the unit of absorbance. The absorbance also can be calculated to concentration with a conversion factor, which is an optional function in this application. If the user inserts the conversion factor, the unit of concentration is represented in **cells/mL**.

7.1 Overview of Screen Features

The information tab bar has 3 tab pages:

- Data Tab Page
- Table Tab Page
- Graph Tab Page

7.1.1 Data Tab Page

On the data tab page of OD600 application (Figure 23), see the table below for details (Table 14).



Figure 23. Data Tab Page of OD600 Application

Table 14. Detailed Description for the Data Tab Page of OD600 Application

Item	Description			
[conc.]	The concentration is calculated from absorbance at 600 nm, and the unit is cells/mL . This is an optional function.			
A600	Displays the absorbance at 600 nm, which is normalized to a 10 mm Pat Length equivalent.			
Dilution Factor	Set a dilution factor for manual diluted samples. The default is "1 " and the range is from 1 ~ 1,000,000 . It should be entered before measurement.			
Recalculated [conc.]	[conc.] multiplied by dilution factor. The unit is cells/mL .			
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.			
Factor	A self-defined conversion factor from A600 to concentration (1*10⁸ cells/ mL). This is an optional function; the default is "OFF" and the range is from 1 ~ 100.00 . It should be entered before measurement.			
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only).			
Baseline Correction	Self-defined wavelength for bichromatic normalization. This is an option function and the default is "OFF".			



7.1.2 Table Tab Page

The table tab page will show all the data results (Figure 24). If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. Users can also tap the **Sample** column to edit the sample name.



Figure 24. Table Tab Page of OD600 Application

7.1.3 Graph Tab Page

Select more than **1** sample data in [$\sqrt{}$] column of the **Graph Tab Page** to overlay the graph (Figure 25). It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.



Figure 25. Graph Tab Page of OD600 Application

7.2 Measuring OD600 Sample

- 1. On the main menu, tap 🔀 to enter **OD600** application.
- 2. Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be $20 \sim 400$ A, it can be switched to 0.05 mm.

3. On the **Data Tab Page**, enter the **Factor** to convert from absorbance at 600 nm to 1*10⁸ cells/mL. This factor is optional, and the default is "OFF".

Conversion Factor			۱	
			0.01 ~ 100.00 or Emp	sty
7	8		9	\leftarrow
	_			
4	5		6	~
1	2		3	
	_			
0				~

- 4. The **Baseline Correction** function is optional, and can be turned on/off anytime. The default is "OFF".
- 5. Use lint-free paper to clean the sample window/cover.
- 6. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

Note

The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).

7. Lower the detection arm and tap 📙 Blank.

- 8. Lift the detection arm, then clean the sample window/cover with lint-free paper.
- 9. Enter the sample name (optional) or use the auto-numbering system.

- 10. Set a **Dilution Factor** if the sample has been diluted, the default is "1". Then, pipette $1 \sim 2$ μ L of the sample solution onto the sample window, or insert a sample in a cuvette into the cuvette holder (1000C only).
- 11. Lower the detection arm and tap Measure.

Note	If there are many samples, turn on Auto Run \rightarrow \rightarrow , measurements will be made automatically after the detection arm has been lowered and sample naming will be sequential.
------	--

- 12. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.
- 13. Tap 💾 Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up (). Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. ave result in system:

Choose an existing user folder or create a **New User**. For the details, please refer **Section 16.1 Creating a New User Folder**.



7.3 Calculation

The principle of OD600 is measuring the light scatter of the particles in the sample solution. The absorbance will differ from different spectrophotometer systems. A modified Beer-Lambert equation is used to calculate the concentration (optional).

 $c = A600 \times cf$

c = concentration of sample suspension solution in cells/mL

A600 = the absorbance at 600 nm (10 mm equivalent)

cf = the cell number conversion factor, which is represented in the unit of 1×10^8 cells/mL

Note The cell number conversion factor is an optional function in EzDrop. Users can enter a self-defined number to calculate the concentration if needed.



8 Kinetics (1000C only)

This application is only used in cuvette mode of EzDrop 1000C. Make a time-based measurement at a user-defined wavelength, with the heater ($37 \sim 45^{\circ}$ C) and stirring ($1 \sim 8$ speed corresponding to 150 ~ 850 rpm) function capable of being optionally matched. Up to 2 analysis wavelengths can be selected, and up to 4 stages can be set.

8.1 Overview of Screen Features

The information tab bar has 5 tab pages:

- Setting Tab Page
- Data Tab Page
- Table Tab Page
- Graph Tab Page
- Trend Tab Page

8.1.1 Setting Tab Page

This is the first tab page when users enter the Kinetics application. Set the required parameters on this page before measurement (Figure 26), see the table below for details (Table 15).



Figure 26. Setting Tab Page of Kinetics Application

ltem			Description		
Method	 Save Method: After all parameters have been set, the method can be stored. Load Method: Chose the saved method and use this method to do the measurement. Delete Method: Delete saved method. 				
Path Length	Only cuvette mode detection can be available in the Kinetics application, the default setting is Cuvette 10 mm . Others should be selected manually.			cs application, elected	
Analysis λ1	User-defined wavel must enter to activa	ength (19 te the fun	0 ~ 1000 nm) for m ction icons. (Require	easuring ed)	samples. User
Analysis λ2	User-defined second wavelength (190 ~ 1000 nm) for measuring samp (Optional)		suring samples.		
Baseline correction	Self-defined wavelength for bichromatic normalization. This is an optional function and the default is "OFF".				
Heater	User can set the heater temperature from 37 ~ 45°C for sample measurement.				
	Users can select the stirrer speed, which has 8 speed settings corresponding to 150 ~ 850 rpm.				
	Stirrer speed	rpm	Stirrer speed	rpm	
Stir	1	150	5	550	
	2	230	6	650	
	3	350	7	750	
	4	450	8	850	
Delay	A delay time before a stage starts. Stage 1: 0 ~ 3600 s ; Stage 2 ~ 4: 10 ~ 3600 s.				
Interval	Length of time for each measurement. The setting range is from 10 ~ 3600 s .				
Interval #	The number of measurements of interval time. The setting range is from 1 ~ 150 .				
Duration	Total time required to The formula is Delay It will display 00:00 The total duration	for this sta y + Interva :00 (Hour of four sta	age. al * Interval #. : Minute: Second) ages is 23:59:59.		

Table 15. Detailed Description for the Setting Tab Page of Kinetics Application



8.1.2 Data Tab Page

On the data tab page of Kinetics application (Figure 27), see the table below for details (Table 16).



Figure 27. Data Tab Page of Kinetics Application

Table 16. Detailed Description for the Data Tab Page of Kinetics Application

Item	Description	
# (Data Table)	Number of measurements.	
Stage (Data Table) Number of stage.		
Time (Data Table)	Time of measurement.	
A? (Data Table)	The "?" will turn to the user-defined Analysis $\lambda 1$ after going blank. After measuring samples, it will show the absorbance of the samples in user-defined Analysis $\lambda 1$, which is normalized to a 10 mm Path Length equivalent.	
A? (Data Table)	The "?" will turn to the user-defined Analysis $\lambda 2$ after going blank. After measuring samples, it will show the absorbance of the samples in user-defined Analysis $\lambda 2$, which is normalized to a 10 mm Path Length equivalent.	

Heater	Displays the current heating block temperature, not the temperature of the liquid sample in the cuvette. If there is a tick in the box, it means the heater is on. Note: If the liquid sample in the cuvette reaches the set temperature, wait for 15 minutes after the heater reaches the set temperature.
Stir	Display the current stirrer speed. If there is a tick in the box, it means the stirrer is on.
Stage	After the measurement starts, the stage under measurement will be displayed here.
Time	After the measurement starts, the elapsed time will be displayed here. The unit is second (s).
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.

8.1.3 Table Tab Page

The table tab page will show all the data results (Figure 28). If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. Users can also tap the **Sample** column to edit the sample name.



Figure 28. Table Tab Page of Kinetics Application

8.1.4 Graph Tab Page

The graph tab page show the Absorbance (10 mm) Vs Wavelength (nm). Select more than 1 sample data in $[\sqrt{}]$ column of the Graph Tab Page to overlay the graph (Figure 29). It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin. $\frac{Lab: Blue-Ray Biotech}{Setting} Date Table$

Lab: Blue-Ray B	iotech	Kinotic	c. Now		2022-12	-15 09:59
Setting	Data	Tab	le	Graph	Tren	d
1.00						
			# [/]	Stage Time	A?	A?
0.75 (
orbance (10 95						
Sq V 0.25						
0.00	392 595 Wavelength (nm	797 1000 i)				
Blank	Stop	Measure	Dele	te Save R	esult Ba	ck

Figure 29. Graph Tab Page of Kinetics Application

8.1.5 Trend Tab Page

The trend tab page show the **Absorbance (10 mm) Vs Time** to observe the absorbance value over time (Figure 30). Users can set the **Start Time** and **End Time** of the X axis. The end time can only be set as the last measurement time point of this protocol.

Lab: Blue-Ray Bio	tech				2022-12-15 09:59
Setting	Data	Tab	le	Graph	Trend
Start 1.00 0.75 (mm01) 9 0.50 0.50 0.25	00:00:00		End	00:01:00	
0,00		004 T	00:30 me		00:01:00
Blank	Stop	Measure	Delete	Save Result	Back

Figure 30. Trend Tab Page of Kinetics Application

8.2 Measuring Kinetics Sample

- 1. On the main menu, tap 1 to enter Kinetics application.
- 2. Lift the detection arm and switch the path length selector to **0.5 mm**.

Note	A path length of 0.5 mm should be selected for cuvette mode.
3. On	the Setting Tab Page,
a.	Load a saved protocol: Tap Method bar, chose a saved method and then tap + Load Method to load the protocol. If the heater or stir function is required, check the heater box to preheat.
b.	Create a new protocol:
	I. Select the correct Path Length based on the experiment protocol.
	II. Enter the Analysis $\lambda 1$ (required) and Analysis $\lambda 2$ (optional) based on the experiment protocol.
	III. The Baseline Correction function is optional, and can be turned on/off anytime. The default is "OFF".
	IV. If the heater or stir function is required, enter the temperature range 37 ~ 45°C and stirrer speed 1 ~ 8. After the temperature setting is completed, the system will automatically start heating.
Note	The recommended stirrer bar size used in a 10 mm Cuvette is a length of 6 mm, Φ 3 mm.
	V. Set the Delay, Interval and Interval # of each stage.
	VI. If the user would like to save this protocol, tap Method bar, 🛃 Save Method and name it.
4. Use	lint-free paper to clean the sample window/cover.
5. Inse	ert the blank cuvette into the cuvette holder.
Note	The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).

6. Lower the detection arm and tap **Blank**.

Note If the heater is on, wait for the heater to reach the set temperature, the message will pop up "Heater Ready". Or the warning message will show "Please wait for the heater to reach the set temperature" if it is still heating.

- 7. Lift the detection arm, take out the blank cuvette.
- 8. Enter the sample name in the Name bar (optional) or it will be named by auto-numbering.
- 9. Insert a sample in a cuvette into the cuvette holder. Lower the detection arm and tap Measure.

Note	If user would like to abort this measurement, tap 💿 Stop to end the meauement. Once stop
Note	the measurement, it cannot be resumed.

- 10. At the end of experiment, the message "**Finish**" will pop up. Lift the detection arm to remove the sample cuvette.
- 11. Tap 💾 Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up \bigcirc . Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer **Section 16.1 Creating a New User Folder**.

Lab: Blue-Ray Biotech			2022-12-15 09:59
	ι	Jser Menu	X
6 ³	Ð	\$	— User Folder
Flash Drive	Public	TEST	— Public Folder
			Customer USB Memory Device
			— Function Icons
	O New User		Sack

9 **III** Microarray

The Microarray application is used to measure the dye-labeled nucleic acid samples with up to 2 fluorescent dyes. It can measure the concentration of nucleic acid at 260 nm in units of $ng/\mu L$ and the concentration of fluorescent dye at dye-specific wavelength in units of μM .

9.1 Overview of Screen Features

The information tab bar has 5 tab pages:

- Dye List Tab Page
- Setting Tab Page
- Data Tab Page
- Table Tab Page
- Graph Tab Page

9.1.1 Dye List Tab Page

On the dye list tab page of the Microarray application (Figure 31), there are 21 common dyes built in to choose from. If the dye to be analyzed is not listed, users can add dye information and save it by tapping "**Add new dye**". See the table below for details (Table 17).

Lab:	Blue-Ray Biotech	Micr	oarray: New		2023-02-09 11:05	
	Dye List So	etting	Data	Table	Graph	
#	Dye	Wavelength (n	m) Coeffi	icient (1/M°cm)	A260 Correction	
15	Oyster-500	503		78,000	0.29	
16	Oyster-550	553		150,000	0.05	
17	Oyster-556	560		155,000	0.03	
18	Oyster-645	649		220,000	0.05	
19	Oyster-650	653		200,000	0.04	
20	Oyster-656	660		200,000	0.04	
21	Texas Red	593		85,000	0.23	
22	(Add new dye)					
		~				
	Blank Auto Ru	in Measure	Dele	ete Save F	Result Back	

Figure 31. Dye List Tab Page of Microarray Application



Table 17. Detailed Description for the Dye List Tab Page of Microarray Application

ltem	Description
[conc.]	Dye name.
Wavelength (nm)	Displays the absorbance of maximum dye. The range is from 190 ~ 1000 nm .
Coefficient (1/M*cm)	Molar extinction coefficient of dye, the unit is 1/M*cm and the range is from 1 ~ 1,000,000 .
A260 Correction	Dye-dependent correction factor at 260 nm.
Add new dye	Users can input dye information by themselves and it can be saved/ deleted.
912 Setting	Tab Page

9.1.2 Setting Tab Page

This is the first tab page when users enter the Microarray application (Figure 32). Set the required parameters on this page before measurement. See the table below for details (Table 18).



Figure 32. Setting Tab Page of Microarray Application

Table 18. Detailed Description for the Setting Tab Page of Microarray Application

Item	Description
Method	It includes sample types, like dsDNA, ssDNA, RNA, miRNA, Oligo and Custom Factor (15.0 ~ 150). The default is "dsDNA".
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only).
Baseline correction	The wavelength for bichromatic normalization is 750 nm . This is an optional function and the default is "ON".
Dye 1	Analysis dye 1. The user should choose at least 1 dye before measurement.
Dye 2	Analysis dye 2. This application can analyze up to 2 dyes and dye 2 is optional.

9.1.3 Data Tab Page

On the data tab page of the Microarray application (Figure 33), see the table below for details (Table 19).



Figure 33. Data Tab Page of Microarray Application

Table 19. Detailed Description for the Data Tab Page of Microarray Application

Item	Description
[conc.]	The concentration of a nucleic acid sample is calculated from absorbance at 260 nm, and the unit is ng/µL .
A260	Displays the absorbance at 260 nm, which is normalized to a 10 mm Path Length equivalent.
A260/A280	Displays the ratio of the absorbance at 260 nm and 280 nm. If a ratio is detected out of range, an alert icon (1) will pop up. dsDNA < 1.75, RNA < 2.0, ssDNA < 1.75.
Dye 1	Displays the concentration of dye1, and the unit is μ M.
Dye 2	Displays the concentration of dye2, and the unit is μM .
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.
Method	It includes sample types, like dsDNA , ssDNA , RNA , miRNA , Oligo and Custom Factor (15.0 ~ 150) . The default is "dsDNA". The method setting on the data tab page is the same as the setting tab page.
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only). The path length setting on the data tab page is the same as the setting tab page.

9.1.4 Table Tab Page

The table tab page will show all the data results (Figure 34). If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. Users can also tap the **Sample** column to edit the sample name.

Lab: Blue	e-Ray Biotech					2	023-02-09 11:05	
			Microarra	y: New				
Dye	List	Setting	Data	a			Graph	
# [x]	Sample	ng/μL	A260/A280	A260	A280	Dye 1	Dye 1 (μM)	
Ŀ	=						•	
Bla	nk Auto	Run I	Measure	Delete	Save	Result	Back	

Figure 34. Table Tab Page of Microarray Application

9.1.5 Graph Tab Page

Select more than **1** sample data in [$\sqrt{}$] column of the **Graph Tab Page** to overlay the graph (Figure 35). It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.

Lab: Blue-Ray Bi	otech	and the second			2023-02-09 11:05
		Microarr	ay: New		
Dye List	Setting	Da	ta	Table	Graph
1.00				# [√] Sar	mple ng/µL
0.75 E					
orbance (10 99					
0.25					
0.00	330 4 Waveler	70 610 ngth (nm)	750		
		~	前		
Blank	Auto Run	Measure	Delete	Save Result	Back

Figure 35. Graph Tab Page of Microarray Application



9.2 Measuring Microarray Sample

- 1. On the main menu, tap **to enter Microarray** application.
- Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be 20 ~ 400 A, it can be switched to 0.05 mm.

 On the Setting Tab Page, select the correct Method type or enter Custom Factor (15.0~150 ng*cm/ul) based on the experiment protocol. The default selection is "dsDNA".



- 4. Select the correct **Path Length** based on the experiment protocol. For micro-volume detection, the instrument will detect the current path length selector position automatically. For cuvette mode detection, it should be selected manually (1000C only).
- 5. The **Baseline Correction** (750 nm) function is optional, and can be turned on/off anytime. The default is "ON".
- 6. Select at least 1 dye to be analyzed, and a maximum of 2 dyes.

	Dye 1			Dye 1	
Alexa Fluor 350	Alexa Fluor 488	Alexa Fluor 532	Alexa Fluor 350	Alexa Fluor 488	Alexa Fluor 532
Alexa Fluor 546	Alexa Fluor 555	Alexa Fluor 568	Alexa Fluor 546	Alexa Fluor 555	Alexa Fluor 568
Alexa Fluor 594	Alexa Fluor 647	Alexa Fluor 660	Alexa Fluor 594	Alexa Fluor 647	Alexa Fluor 660
Alexa Fluor 680	Cy3	Су3.5	Alexa Fluor 680	СуЗ	Cy3.5
Cy5	Cy5.5	Oyster-500	Cy5	Cy5.5	Oyster-500
×		 Image: A set of the set of the	×		 Image: A set of the set of the

7. If the dye to be analyzed is not listed, tap the **Dye List Tab Page** and scroll to the bottom, click "**Add new dye**" to add dye information.

D	ye
Dye	•
Wavelength (nm)	•
Coefficient (1/M*cm)	•
A260 Correction	•
×	~



- 8. Use lint-free paper to clean the sample window/cover.
- 9. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).



- 10. Lower the detection arm and tap _____ Blank.
- 11. Lift the detection arm, then clean the sample window/cover with lint-free paper.
- 12. Enter the sample name in the **Name** bar (optional) or it will be named by auto-numbering. Then, pipette $1 \sim 2 \mu L$ of the sample solution onto the sample window, or insert a sample in a cuvette into the cuvette holder (1000C only).
- 13. Lower the detection arm and tap Measure.

Note

If there are many samples, turn on Auto Run \rightarrow \rightarrow \rightarrow , measurement will be made automatically after the detection arm has been lowered and sample naming will be sequential.

- 14. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.
- 15. Tap Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up \bigcirc . Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.



b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer <u>Section 16.1 Creating a New User Folder</u>.

Lab: Blue-Ray Biotech	2022-12-15 09:59
User Me	nu
Flash Drive Public TEST	User Folder
	Public Folder
	Customer USB Memory Device
	Function Icons
New User	Back
	G
	19
G	

9.3 Calculation

In the Microarray application, a modified Beer-Lambert equation is used to calculate concentrations with absorbance and factor as follows, with or without baseline correction:

• Dye-labeled Nucleic Acid concentration

Without baseline correction:

 $c = [(A260 - dye1 - factor * A_{max,dye1} - dye2 - factor * A_{max,dye2})] \times \epsilon$

With baseline correction

 $c = ((A260 - A_{Baseline}) - [dye1 - factor * (A_{max,dye1} - A_{Baseline})] - [dye2 - factor * (A_{max,dye2} - A_{Baseline})]) \times \epsilon$

c = the nucleic acid concentration in $ng/\mu L$

A260 = the absorbance at 260 nm (10 mm Path Length)

ABaseline = the absorbance at baseline wavelength (10 mm Path Length)

Amax,dye = the absorbance at dye peak wavelength (10 mm Path Length)

dye-factor = the dye correction factor at 260 nm

 ϵ = the extinction coefficient factor of nucleic acid in ng*cm/µL

Dye concentration

Without baseline correction:

 $c = A_{max,dye} / (\epsilon_{dye} * 10^{-6})$

With baseline correction

 $c = (A_{max,dye} - A_{Baseline}) / (\epsilon_{dye} * 10^{-6})$

c = the dye concentration in μM A_{max,dye} = the absorbance at dye peak wavelength (10 mm Path Length) A_{Baseline} = the absorbance at baseline wavelength (10 mm Path Length) ε_{dye} = the extinction coefficient factor of dye in 1/M*cm

The general extinction coefficient used in the calculation of nucleic acid are shown in Table 20.



Table 20. Extinction Coefficient of Nucleic Acids

Туре	Factor (ng*cm/µl)
dsDNA	50
ssDNA	33
RNA	40
miRNA	33
Oligo	33
Custom	15.0 ~ 150

Table 21. Dye List and Detailed Information

Dye Type	Dye Wavelength (nm)	Extinction Coefficient (1/M*cm)	A260 Correction Factor
Alexa Fluor 350	345	18,400	0.25
Alexa Fluor 488	492	62,000	0.3
Alexa Fluor 532	525	82,300	0.24
Alexa Fluor 546	555	104,000	0.21
Alexa Fluor 555	555	150,000	0.04
Alexa Fluor 568	576	93,000	0.45
Alexa Fluor 594	588	80,400	0.43
Alexa Fluor 647	650	239,000	0
Alexa Fluor 660	660	107,000	0
Alexa Fluor 680	680	164,000	0
СуЗ	550	150,000	0.08
Cy3.5	581	150,000	0.08
Cy5	649	250,000	0.05
Cy5.5	675	250,000	0.05
Oyster-500	503	78,000	0.29
Oyster-550	553	150,000	0.05
Oyster-556	560	155,000	0.03
Oyster-645	649	220,000	0.05
Oyster-650	653	200,000	0.04
Oyster-656	660	200,000	0.04
Texas Red	593	85,000	0.23
Add new dye	190 ~ 1000	0 ~ 1,000,000	0 ~ 0.999

10 者 Labeled Protein

The Labeled Protein application is used to measure the dye-labeled protein samples with up to 2 fluorescent dyes. It can measure the concentration of proteins at 280 nm in units of **mg/mL** and the concentration of fluorescent dye at a dye-specific wavelength in units of μ M.

10.1 Overview of Screen Features

The information tab bar has 5 tab pages:

- Dye List Tab Page
- Setting Tab Page
- Data Tab Page
- Table Tab Page
- Graph Tab Page

10.1.1 Dye List Tab Page

On the dye list tab page of Labeled Protein application (Figure 36), it has 21 common dyes built-in to choose from. If the dye to be analyzed is not listed, users can add dye information and save it by tapping "Add new dye". See the table below for details (Table 22).

Lat	o: Blue-Ray Biotech					2023-02-04 10:41	
		Lab	eled Protein				
	Dye List	Setting	Data	a Table		Graph	
#	Dye	Wavelength	(nm) Co	pefficient (1/N	/l*cm)	A280 Correction	
16	5 Pacific Blue	409		30,000		0.20	
17	Pacific Orange	397		24,500		0.60	
18	pHrodo Green	505		75,000		0.20	
19	pHrodo Red	560		65,000		0.12	
20	r-PE	566		200,000		0.18	
21	I Texas Red	595		80,000		0.18	
22	2 (Add new dye)						
	Blank Auto	Run Meas	ure	Delete	Save Re	esult Back	

Figure 36. Dye List Tab Page of Labeled Protein Application

Table 22. Detailed Description for the Dye List Tab Page of Labeled Protein Application

ltem	Description
Dye	Dye name.
Wavelength (nm)	Displays the absorbance of maximum dye. The range is from 190 ~ 1000 nm
Coefficient (1/M*cm)	Molar extinction coefficient of dye, the unit is 1/M*cm and the range is from 1 ~ 1,000,000 .
A280 Correction	Dye-dependent correction factor at 280 nm.
Add new dye	Users can input dye information by themselves and it can be saved/ deleted.

10.1.2 Setting Tab Page

This is the first tab page when users enter the Labeled Protein application (Figure 37). Set the required parameters on this page before measurement. See the table below for details (Table 23).



Figure 37. Setting Tab Page of Labeled Protein Application



Table 23. De	tailed Description	for the Setting	Tab Page of I	Labeled Protein	Application
--------------	--------------------	-----------------	---------------	-----------------	-------------

ltem	Description
Method	It includes sample types, like BSA, IgG Mouse, IgG Human, IgE Human, Lysozyme, SA Mouse, SA Human, 1A=1mg/ml and Custom Factor (0.001 ~ 100). The default is "BSA".
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only).
Baseline correction	The wavelength for bichromatic normalization is 750 nm. This is an optional function and the default is "ON".
Dye 1	Analysis dye 1. The user should choose at least 1 dye before measurement.
Dye 2	Analysis dye 2. This application can analyze up to 2 dyes and dye 2 is optional.

10.1.3 Data Tab Page

On the data tab page of the Labeled Protein application (Figure 38), see the table below for details (Table 24).



Figure 38. Data Tab Page of Labeled Protein Application

Table 24. Detailed Description for the Data Tab Page of Labeled Protein Application

Description
The concentration of a protein sample is calculated from absorbance at 280 nm, and the unit is ng/µL .
Displays the absorbance at 280 nm, which is normalized to a 10 mm Path Length equivalent.
Displays the ratio of the absorbance at 260 nm and 280 nm. When the ratio of A260/280 > 0.6, an alert icon 2 will pop up.
Displays the concentration of dye1, and the unit is μ M.
Displays the concentration of dye2, and the unit is μM .
The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.
Includes sample types like BSA, IgG Mouse, IgG Human, IgE Human, Lysozyme, SA Mouse, SA Human, 1A=1mg/ml and Custom Factor (0.001 ~ 100). The default is "BSA". The method setting on the data tab page is the same as the setting tab page.
For micro-volume detection, the instrument will detect the current path length selector position automatically and shown the path length here. For cuvette mode detection, it should be selected manually (1000C only). The path length setting on the data tab page is the same as the setting tab page.
tab page.



10.1.4 Table Tab Page

The table tab page will show all the data results (Figure 39). If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. Users can also tap the **Sample** column to edit the sample name.



Figure 39. Table Tab Page of Labeled Protein Application

10.1.5 Graph Tab Page

Select more than **1** sample data in [$\sqrt{}$] column of the **Graph Tab Page** to overlay the graph (Figure 40). It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.

Lab: Blue-Ray Bio	tech	Labeled Pro			20	023-02-09 11:
Dye List	Setting	Dal	ta	Table		Graph
1.00				# [/]	Sample	mg/n
0.75 [.] F						
ance (10mr 0.20						
Absorb 0.25						
0.00 190	330 4	70 610	750			

Figure 40. Graph Tab Page of Labeled Protein Application

10.2 Measuring Labeled Protein Sample

- 1. On the main menu, tap 📌 to enter Labeled Protein application.
- 2. Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be $20 \sim 400$ A, it can be switched to 0.05 mm.

On the Setting Tab Page, select the correct Method type or enter Custom Factor (0.001 ~ 100 g*cm/l) based on the experiment protocol. The default selection is "BSA".

Method	
Туре	Factor (g*cm/l)
BSA	1.499
lgG Mouse	0.714
IgG Human	0.735
IgE Human	0.654

- 4. Select the correct **Path Length** based on the experiment protocol. For micro-volume detection, the instrument will detect the current path length selector position automatically. For cuvette mode detection, it should be selected manually (1000C only).
- 5. The **Baseline Correction** (750 nm) function is optional, and can be turned on/off anytime. The default is "ON".
- 6. Select at least 1 dye to be analyzed, and a maximum of 2 dyes.


7. If the dye to be analyzed is not listed, tap the **Dye List Tab** Page and scroll to the bottom, click "**Add new dye**" to add dye information.

	Dye
Dye	•
Wavelength (nm)	•
Coefficient (1/M*cm)	•
A280 Correction	•
×	~

- 8. Use lint-free paper to clean the sample window/cover.
- 9. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

Note	The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).
------	---

- 10. Lower the detection arm and tap **Blank**.
- 11. Lift the detection arm, then clean the sample window/cover with lint-free paper.
- 12. Enter the sample name in the **Name** bar (optional) or it will be named by auto-numbering. Then, pipette $1 \sim 2 \mu L$ of the sample solution onto the sample window, or insert a sample in a cuvette into the cuvette holder (1000C only).
- 13. Lower the detection arm and tap Measure.

Note

If there are many samples, turn on Auto Run \rightarrow \rightarrow \rightarrow , the measurement starts automatically after you lower the detection arm, and the sample name will sort in order.

- 14. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover by ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.
- 15. Tap 💾 Save Results to system or flash drive.
 - a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up \bigotimes . Tap it and enter the file name. Once

the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer <u>Section 16.1 Creating a New User Folder</u>.



10.3 Calculation

In the Labeled Protein application, a modified Beer-Lambert equation is used to calculate concentrations with absorbance and factor as follows, with or without baseline correction:

• Dye-labeled protein concentration

Without baseline correction:

 $c = [(A280 - dye1 - factor * A_{max,dye1} - dye2 - factor * A_{max,dye2})] \times \epsilon$

With baseline correction

 $c = ((A280 - A_{\text{Baseline}}) - [dye1 - factor * (A_{\text{max,dye1}} - A_{\text{Baseline}})] - [dye2 - factor * (A_{\text{max,dye2}} - A_{\text{Baseline}})]) \times \epsilon$

c = the protein concentration in mg/mL A280 = the absorbance at 280 nm (10 mm Path Length) A_{Baseline} = the absorbance at baseline wavelength (10 mm Path Length) A_{max,dye} = the absorbance at dye peak wavelength (10 mm Path Length) dye-factor = the dye correction factor at 280 nm ε = the extinction coefficient factor of protein in g*cm/l

Dye concentration

Without background correction:

 $C = A_{max,dye} / (\varepsilon_{dye} * 10^{-6})$

With background correction

 $c = (A_{max,dye} - A_{Baseline}) / (\epsilon_{dye} * 10^{-6})$

 $c = the dye concentration in \mu M$

A_{max,dye} = the absorbance at dye peak wavelength (10 mm Path Length)

ABaseline = the absorbance at baseline wavelength (10 mm Path Length)

 ϵ_{dye} = the extinction coefficient factor of dye in 1/M*cm

The general extinction coefficient factors used in the calculation of protein are shown in Table 25.

Table 25. Extinction Coefficient of Protein

		1/9 Cili	
BSA	1.499	0.667	
lgG Mouse	0.714	1.4	
lgG Human	0.735	1.36	
lgE Human	0.654	1.53	
Lysozyme	0.379	2.64	
SA Mouse	1.493	0.67	
SA Human	1.718	0.582	
1A=1mg/mL	1	N/A	
Custom	0.001 ~ 100	N/A	

Table 26. Dye List and Detailed Information

Dye Type	Dye Wavelength (nm)	ye Wavelength Extinction (nm) Coefficient (1/M*cm)		
Alexa Fluor 350	346	19,000	0.19	
Alexa Fluor 405	401	34,500	0.70	
Alexa Fluor 488	494	71,000	0.11	
Alexa Fluor 532	530	81,000	0.09	
Alexa Fluor 546	554	104,000	0.12	
Alexa Fluor 555	555	150,000	0.08	
Alexa Fluor 568	577	91,300	0.46	
Alexa Fluor 594	590	73,000	0.56	
Alexa Fluor 647	650	239,000	0.03	
Alexa Fluor 680	679	184,000	0.05	
СуЗ	Cy3 550 15		0.05	
Cy5	649	649 250,000 0.		
DyLight 649	654	654 250,000 0.0		
DyLight 488	493	70,000	0.15	
FITC	495	68,000	0.30	
Pacific Blue	409	30,000	0.20	
Pacific Orange	397	245,000	0.60	
pHrodo Green	505	75,000	0.20	
pHrodo Red	560	65,000	0.12	
r-PE	566	200,000	0.18	
Texas Red	595	80,000	0.18	
Add new dye	190 ~ 1000	0 ~ 1,000,000	0 ~ 0.999	

11 🗾 Standard Curve

In the Standard Curve application, users can use self-defined wavelength to establish a standard curve for sample measuring. The correction wavelength is also a self-defined function (optional).

11.1 Overview of Screen Features

The information tab bar has 5 tab pages:

- Setting Tab Page
- Standard Tab Page
- Data Tab Page
- Table Tab Page
- Graph Tab Page

11.1.1 Setting Tab Page

This is the first tab page when users enter the Standard Curve application (Figure 41). Set the required parameters on this page before measurement. See the table below for details (Table 27).



Figure 41. Setting Tab Page of Standard Curve Application



Table 27. Detailed Description for the Setting Tab Page of Standard Curve Application

ltem	Description
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only).
Analysis Wavelength	Self-defined wavelength (190 ~ 1000 nm) for measuring samples. This is required setting to activate the function icons.
Unit	User can enter their self-defined units. This is a required setting to activate the function icons.
Baseline Correction	Self-defined wavelength for bichromatic normalization. This is an optional function and the default is "OFF".
Graph_Minimum	The minimum value of the X-axis of the user-defined graph, the range is from 190 ~ 1000 nm . The default is "190 nm".
Graph_Maximum	The maximum value of the X-axis of the user-defined graph, the range is from 190 ~ 1000 nm . The default is "1000 nm".

11.1.2 Standard Tab Page

The standard tab page (Figure 42) is the page to measure standard samples absorbance at specific wavelength to establish a standard curve. It only shows the standard data result. See the table below for details (Table 28).



Figure 42. Standard Tab Page of Standard Curve Application

Item	Description
mg/mL	This is the standard concentration column. The value is inserted by the users. The range is from 0.001 ~ 99999 .
Abs	The absorbance measured at different wavelengths according to the protein method.
Avg. Abs	The average absorbance of the standard repetition. It is calculated automatically.
Curve Type	The standard curve types the user can select: linear, interpolation, and 2^{nd} order polynomial.
Repetition	The number of standard repetitions. The default value is "1" and the maximum is 3.
Generate Standard Curve	Select the standard data by tapping the $[]$ column to establish the standard curve and the formula will be display in the line below.

Table 28. Detailed Description for the Standard Tab Page of Standard Curve Application

11.1.3 Data Tab Page

On the data tab page of Standard Curve application (Figure 43), see the table below for details (Table 29). This page only shows the sample data, and does not show standard data.



Figure 43. Data Tab Page of Standard Curve Application

Table 29. Detailed Description for the Data Tab Page of Standard Curve Application

Item	Description
[conc.]	The detected absorbance value will be brought into the established standard curve formula to calculate the sample concentration. The unit is set by user.
A?	Displays the absorbance at a set wavelength. The absorbance is normalized to a 10 mm Path Length equivalent.
Dilution Factor	Set a dilution factor for manual diluted samples. The default is "1" and the range is from 1 ~ 1,000,000. It should be entered before measurement.
Recalculated [conc.]	[conc.] multiplied by dilution factor. The unit is set by user.
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.
Path Length	For micro-volume detection, it can only be carried out at 0.5 mm Path Length . For cuvette mode detection, it should be selected manually (1000C only). The path length setting on the data tab page is the same as the setting tab page.

11.1.4 Table Tab Page

The table tab page will show only the sample data result (Figure 44). The standard data is NOT included on this page. The table tab page will show all the data results. If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. Users can also tap the **Sample** column to edit the sample name.

ab: Blue	e-Ray Biotech					2023-02-09	11:05
Set	tting	Standard	Data	Та	ble	Graph	
# [x]	Sample	[conc.]	Dilution Fa	ctor Recalc	Unit	λ (Analysis)	
L							
Bla	nk Au	to Run I	Measure	Delete	Save Result	Back	

Figure 44. Table Tab Page of Standard Curve Application

11.1.5 Graph Tab Page

In the Standard Curve application, there are 2 graph types: the **Spectrum View** and **Standard Curve View** (Figure 45). It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.

Lab: Blue-Ray B	liotech	and the first of	and the second sec		20	23-02-09 11:05
		Standard				
Setting	Standard	D	ata	Table		Graph
1.00		*	ŧ [•]	Sample	[conc.]	Abs
(mm 0.75						
orbance (
0.25						
0.00	92 595 79 Wavelength (nm)	97 1000				
Spectrum	View Std. C	urve View				
					5	-
Blank	Auto Run	Measure	Dele	te Save	Result	Back

Figure 45. Graph Tab Page of Standard Curve Application



11.2 Measuring User-defined Standard Curve Sample

- 1. On the main menu, tap 📈 to enter **Standard Curve** application.
- 2. Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be $20 \sim 400$ A, it can be switched to 0.05 mm.

Note A path length of **0.5 mm** should be selected for cuvette mode (1000C only).

- 3. On the **Setting Tab Page**, select the correct **Path Length** based on the experiment protocol. For micro-volume detection, the instrument will detect the current path length selector position automatically. For cuvette mode detection, it should be selected manually (1000C only).
- 4. Enter the Analysis Wavelength and Unit based on the experiment protocol.
- 5. The **Baseline Correction** function is optional, and can be turned on/off anytime. The default is "OFF".
- 6. Set the X axis minimum and maximum for the graph if required.
- 7. Jump to Standard Tab Page to establish standard curve data.
- 8. Select the correct **Curve Type** and **Replicates** $(1 \sim 3)$ based on the experiment protocol.

	a. Linear curve type requires minimum 2 DIFFERENT concentrations to establish standard curve.
Note	b. Interpolation curve type requires minimum 2 DIFFERENT concentrations to establish standard curve.
	c. 2nd order polynominal curve type requires minimum 3 DIFFERENT concentrations to establish standard curve.

9. Tap on a cell in the **mg/mL** column to enter the concentration of standard sample.



- 10. Use lint-free paper to clean the sample window/cover.
- 11. For Micro-Volume mode detection, pipette a $1 \sim 2 \mu L$ blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

Note The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).

- 12. Lower the detection arm and tap **Blank**.
- 13. Lift the detection arm, then clean the sample window/cover with lint-free paper.
- 14. Pipette $1 \sim 2 \mu L$ of standard sample solution onto the sample window, or insert a standard sample cuvette into the cuvette holder (1000C only).
- 15. Tap on a cell in the **Abs**. column and tap **Measure** to establish standard data. If the number of replicates is higher than 1, it will automatically jump to the next cell. However, if the number of repetitions of this concentration has been measured and finished, it will not automatically jump to the next concentration.



Note

If there are many standard samples, turn on Auto Run \rightarrow \rightarrow \rightarrow , measurements will be made automatically after the detection arm has been lowered and sample naming will be sequential.

16. Select the standard data by tapping the $[\sqrt{}]$ column and check **Generate Standard Curve** to establish the standard curve and the formula will be display in the line below.

11 Standard Curve



Note

If user need to rebuild the standard curve, uncheck **Generate Standard Curve** and remeasure standard sample or input new standard data.

17. Jump to the Data Tab Page to measure samples.

- 18. Enter the sample name in the **Name** bar (optional) or it will be named by auto-numbering . Then, pipette $1 \sim 2 \mu L$ sample solution onto the sample window, or insert a sample in a cuvette into the cuvette holder (1000C only). If the protein sample has been diluted, users can enter **Dilution Factor**, the actual concentration will be automatically calculated by multiplying the measured concentration [conc.] by the dilution factor and the result will be displayed in **Recalculated [conc.]**.
- 19. Lower the detection arm and tap Measure.

1. If there are many sample solution, turn on Auto Run \rightarrow \rightarrow \rightarrow , measurements will be made automatically after the detection arm has been lowered and sample naming will be sequential.

Note

2. It is suggested to clean the sample window/cover window when exchanging different concentrations of samples.

20. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.

21. Tap Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up \bigcirc . Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer <u>Section 16.1 Creating a New User Folder</u>.



11.3 Calculation

In the **Standard Curve** application, the concentration is calculated by the absorbance values measuring in user-defined wavelength and the established standard curve. The standard curve can be in types of linear, interpolation, or 2nd order polynomial.



12 •••• UV-Vis

The UV-Vis application can function as a traditional spectrophotometer, and it can do whole wavelength scan of unknown samples ranging from 190 ~ 1000 nm. Up to 8 wavelengths can be entered and displayed on the screen, and the absorbance of the entire wavelength data can be obtained from the output csv files.

12.1 Overview of Screen Features

The information tab bar has 4 tab pages:

- Setting Tab Page
- Data Tab Page
- Table Tab Page
- Graph Tab Page

12.1.1 Setting Tab Page

This is the first tab page when user enter the UV-Vis application. In the setting tab page (Figure 46), there are 8 user-defined wavelengths and the setting wavelength range is from 190 nm to 1000 nm. At least 1 user-defined wavelength is required to activate the function icons. See the table below for details (Table 30).



Figure 46. Setting Tab Page of UV-Vis Application

Table 30. Detailed Description for the Setting Tab Page of UV-Vis Application

ltem	Description
Wavelength	User-defined analysis wavelength (190 ~ 1000 nm) for measuring samples. This is a required setting to activate the function icons.
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only).
Baseline Correction	Self-defined wavelength for bichromatic normalization. This is an optional function and the default is "OFF".
Graph_Minimum	The minimum value of the X-axis of the user-defined graph, the range is from 190 ~ 1000 nm . The default is "190 nm".
Graph_Maximum	The maximum value of the X-axis of the user-defined graph, the range is from 190 ~ 1000 nm . The default is "1000 nm".

12.1.2 Data Tab Page

On the data tab page of UV-Vis application (Figure 47), see the table below for details (Table 31).



Figure 47. Data Tab Page of UV-Vis Application

Table 31. Detailed Description for the Data Tab Page of UV-Vis Application

Item	Description
Α?	The "?" will turn to the user-defined wavelength after going blank. After measuring samples, it will show the absorbance of the samples in a user-defined wavelength, which is normalized to a 10 mm Path Length equivalent.
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only). In the data tab page, the settings for the path length are the same as in the setting tab page.
	S

12.1.3 Table Tab Page

The table tab page will show all the data results (Figure 48). If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. Users can also tap the **Sample** column to edit the sample name.



Figure 48. Table Tab Page of UV-Vis Application

Sector BLUE-RAY

86

12.1.4 Graph Tab Page

It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.

Lab: Blue-Ray I	Biotech		Section of the sectio		2023-02-09 10:41
Catting		UV-Vi	s: New		Cranh
Setting		Data	Table		Graph
1.00				# [/]	Sample
0.75 C					
lomn					
) 92 0.50					
orba					
sqF 0.25					
0.00+ 190	392	595	797	1000	
	wa	velength (nm)			
Blank	Auto Run	Measure	Delete	Save Result	Back

Figure 49. Graph Tab Page of UV-Vis Application

12.2 Measuring UV-Vis Sample

- 1. On the main menu, tap ()) to enter UV-Vis application.
- Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be 20 ~ 400 A, it can be switched to 0.05 mm.

Note	A path length of 0.5 mm should be selected for cuvette mode (1000C only).
NOLE	reputit tengen of 0.0 min should be selected for euverie mode (10000 only).

- 3. On the **Setting Tab Page**, enter up to **8 analysis wavelengths** based on the experiment protocol.
- Select the correct Path Length based on the experiment protocol. For micro-volume detection, the instrument will detect the current path length selector position automatically. For cuvette mode detection, it should be selected manually (1000C only).
- 5. The **Baseline Correction** function is optional, and can be turned on/off anytime. The default is "OFF".
- 6. Set the X axis minimum and maximum of graph if required.
- 7. Use lint-free paper to clean the sample window/cover.
- 8. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check that there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

Note

The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).

- 9. Lower the detection arm and tap **Blank**.
- 10. Lift the detection arm, then clean the sample window/cover with lint-free paper.
- 11. Enter the sample name in the **Name** bar (optional) or use the auto-naming system. Then, pipette $1 \sim 2 \mu L$ sample solution onto the sample window, or insert a sample in a cuvette into the cuvette holder (1000C only).

12. Lower the detection arm and tap Measure.

Note

If there are many samples, turn on Auto Run \rightarrow \rightarrow \rightarrow , measurements will be made automatically after the detection arm has been lowered and sample naming will be sequential.

- 13. At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.
- 14. Tap Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up \bigcirc . Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer Section 16.1 Creating a New User Folder.



13 **F** Factor

In this application, the user can measure a sample at a user-defined wavelength. The user can also enter custom units, conversion factors (optional) and correction wavelengths (optional) to calculate sample concentration.

13.1 Overview of Screen Features

The information tab bar has 4 tab pages:

- Setting Tab Page
- Data Tab Page
- Table Tab Page
- Graph Tab Page

13.1.1 Setting Tab Page

On the setting tab page (Figure 50), there are 5 customized options, **Factor, Unit, Ratio** (λ 1/ λ 2), **Baseline Correction** and the **Graph Range**, offered for users. The function icons will be activated after entering the **Analysis Wavelength**. See the table below for details (Table 32).



Figure 50. Setting Tab Page of Factor Application.

Table 32. Detailed Description for the Setting Tab Page of Factor Application

ltem	Description
Analysis Wavelength	User-defined wavelength for measuring samples. User has to enter to activate the function icons. (Required)
Factor	User can enter a self-defined conversion factor (0.01 \sim 100) according to the relationship between sample absorbance and units. (Optional)
Unit	User can enter self-defined units. (Optional)
Ratio (λ1/λ2)	User-defined wavelengths used to calculate ratio.
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and display the path length here. For cuvette mode detection, it should be selected manually (1000C only).
Baseline Correction	Self-defined wavelength for bichromatic normalization. This is an optional function and the default is "OFF".

13.1.2 Data Tab Page

On the data tab page of factor application (Figure 51), see the table below for details (Table 33).



Figure 51. Data Tab Page of Factor Application.

Table 33. Detailed Description for the Data	a Tab Page of Factor Application
---	----------------------------------

Item	Description
[conc.]	The concentration is calculated from absorbance at user-defined wavelength. The unit is also user-defined, but only shown on the custom setting page. This is an optional function.
A?	The "?" will turn to the user-defined wavelength after going blank. After measuring samples, it will show the absorbance of the samples in the user-defined wavelength, which is normalized to a 10 mm Path Length equivalent.
Ratio	Displays the ratio ($\lambda 1/\lambda 2$) of the absorbance at user-defined wavelength.
Name	The sample name can be entered here. The default is "Sample". Note: "Blank, Sample" cannot be used as a sample name.
Path Length	For micro-volume detection, the instrument will detect the current path length selector position automatically and shown the path length here. For cuvette mode detection, it should be selected manually (1000C only). In the data tab page, the settings for the path length are the same as in the setting tab page.

13.1.3 Table Tab Page

The table tab page will show all the data results (Figure 52). If the user needs to delete data, tap the **[x]** column, the **Delete** will become functional. User also can tap the **Sample** column to edit the sample name.

La	b: Blue-Ra	ay Biotech					2023-	02-09 10:
			Factor: New					
	Set	ting	Data	•	Tabl	e	Grapi	• X
#	[x]	Sample	[conc.]	Unit	λ (Analysis)	Abs	λ 1/ λ2	Ratio
						F	7	•
	Blank	Auto P	un M	leasure	Delete	Save	Posult	Back

Figure 52. Table Tab Page of Factor Application.

13.1.4 Graph Tab Page

It can be zoomed in or out by spreading two fingers or bringing them together on the screen. The axis can be moved by dragging the graph. Go back to the default by a long touch on the coordinate origin.

Lab: Blue-Ray Biote	ch		24.00		2023-02-09 10:41	
				_	$\langle \cdot \rangle$	
Setting		Data	Table		Graph	
1.00			# [/]	Sample	[conc.]	5
0.75 E						
orbance (10 00						
0.25						
0.00	92 595 Wavelengt	797 th (nm)	1000			
					•	
Blank	Auto Rup	Moasuro	Delete S:	No Posult	Back	

Figure 53. Graph Tab Page of Factor Application.



13.2 Measuring Factor Sample

- 1. On the main menu, tap F(x) to enter Factor application.
- 2. Lift the detection arm and switch the path length selector to 0.5 mm for measuring an unknown sample. If the sample absorbance is predicted to be $20 \sim 400$ A, it can be switched to 0.05 mm.

Note A path length of **0.5 mm** should be selected for cuvette mode (1000C only).

- 3. On the Setting Tab Page, enter Analysis Wavelength (required), Factor, Unit, Ratio ($\lambda 1/\lambda 2$) based on the experiment protocol.
- Select the correct Path Length based on the experiment protocol. For micro-volume detection, the instrument will detect the current path length selector position automatically. For cuvette mode detection, it should be selected manually (1000C only).
- 5. The **Baseline Correction** function is optional, and can be turned on/off anytime. The default is "OFF".
- 6. Set the X axis minimum and maximum for the graph if required.
- 7. Use lint-free paper to clean the sample window/cover.
- 8. For micro-volume detection, pipette a $1 \sim 2$ uL blanking solution onto the sample window and check there is no cuvette in the cuvette holder (1000C only). For cuvette mode detection, insert the blanking cuvette into the cuvette holder (1000C only).

Note The liquid level in the cuvette should be higher than the instrument optical path, the beam height of which is 8.5 mm. The light-transmitting surface of the cuvette should be in the direction of the arrow on the cuvette holder (Figure 11).
9. Lower the detection arm and tap Blank.
10. Lift the detection arm, then clean the sample window/cover by lint-free paper.

- 11. Enter the sample name in the **Name** bar (optional) or the naming system is auto-numbering. Then, pipette $1 \sim 2 \mu L$ sample solution onto the sample window, or insert the sample cuvette into the cuvette holder (1000C only).
- 12. Lower the detection arm and tap Measure.

Note

If there are many samples, turn on Auto Run \rightarrow \rightarrow \rightarrow , measurements will be made automatically after the detection arm has been lowered and sample naming will be sequential.

- At the end of experiment, wipe away the sample with lint-free paper and clean the sample window/cover with ddH₂O, or remove the sample cuvette (1000C only). For the details, please refer <u>Section 18.2.1 Daily Cleaning</u>.
- 14. Tap Save Results to system or flash drive.

a. Save results to flash drive:

Insert the flash drive with **FAT/FAT32** formatted into the USB-A port on the front of the EzDrop; the flash drive icon will pop up () . Tap it and enter the file name. Once the result is saved successfully, the message, "**Report has been saved**" will pop up. For details, please refer <u>Section 16.5 Using a Flash Drive as a User Folder</u>.

b. Save result in system:

Choose an existing user folder or create a **New User**. For the details, please refer <u>Section 16.1 Creating a New User Folder</u>.



13.3 Calculation

In the **Factor** application, a modified Beer-Lambert equation is used to calculate concentrations with user-defined measuring absorbance and factor as follows, with or without baseline correction (optional):

Without background correction:

 $c = A \times \epsilon$

With background correction

 $c = (A - A_{Baseline}) \times \epsilon$

c = the purified protein concentration in the user-defined factor

A = the user-defined absorbance at analysis wavelength

A_{Baseline} = the user-defined absorbance at baseline wavelength

 ϵ = the user-defined extinction coefficient factor

14 System Menu

Tap on theSystem icon on the main menu to enter the System Menu as shown in Figure54 below. Users can adjust several parameters or settings for the EzDrop 1000/1000C.

Lab: Blue-Ray Biotech	n			2023-02-09 11:05
鼸	((ا	- ` ¢-	·ੱਸੇਂ 🔹	
Date & Time	Beep Sound	Brightness	Indicator Light	Storage
	(i)	?		
Self-Test	About	Operation Manual		\sim
20		X		◆
Admin		Service		Back

Figure 54. System Menu Overview

14.1 🗒 Date & Time

Users can change the date and time setting of the EzDrop 1000/1000C from here.

14.2 (I) Beep Sound

Users can turn **ON** or **OFF** the system buzzer from here.

14.3 🔅 Brightness

Users can adjust the brightness of the display panel according to your environment's lighting condition.

14.4 🔆 Indicator Light

Users can turn **ON** or **OFF** the LED auxiliary light beside the sample window from here.



14.5 🔒 Storage

This function shows the information of total and remaining memory on an SD card in the EzDrop 1000/1000C.

14.6 🔋 Self-Test

Users can do a system self-test of the EzDrop 1000/1000C. Keep the detection arm closed and do not lift the detection arm during testing.

If the Self-Test fails, the following system information message will appear: "The flash intensity is out of range, please check and clean the window on the pathlength selector. If this message keeps recurring, please contact your distributor for technical support."

14.7 (i) About

Users can check the basic information of EzDrop 1000/1000C with this icon, including the System Version, Initialization Date and Calibration Date.

14.8 🗵 Operation Manual

Users can scan QR code to download operation manual.

14.9 **Admin**

The default Administrator password is "**0000**". The administrator has rights to alter settings as described below.



Figure 55. Administrator Menu Overview

14.9.1 😁 Reset System

Reset the EzDrop 1000/1000C to the default setting.

14.9.2 🦓 Manage User

Enter to the **User Menu** and the administrator has rights to **Delete User** or **Clear All**.

14.9.3 冷 Back Up to Flash Drive

When inserting the flash drive, it will change to a blue color \Re . It can be used to back up all user data.

14.9.4 Name Lab Name

Change the Lab Name, which will be on the left-up corner of every page.

14.9.5 🔮 PIN

Change the admin password; up to 8 numbers. Once setup is complete, go back to the main menu and tap the **EzDrop** icon to update your password.

14.9.6 🔍 History Data

The administrator has the rights to delete/copy/view History Files.



Figure 56. History Data Overview



Only authorized service personnel have the password to enter Service Mode and perform necessary maintenance and repairs.



15 🕓 History

Both saved and unsaved results will be recorded here, including the results stored in the flash drive.

Tap on the **W** History icon on the main menu to enter the History list information screen. If all the results can't be shown on one page, you can slide up or down on the screen to check the rest. The maximum number of records is **300**. If this number is exceeded, the latest record will replace the oldest one. A sample screen is shown in Figure 57 below.

Lab: Blue-Ray Biotech			2022-12-15 09:59	
	Hist	ory		
Date & Time 🛛 🛡	Protocol	User	File	
2022-10-04 10:20:41	Nucleic Acid	TEST	0000	— Lock Symbol
+		Ē		
		Ŀα	• • •	
New Protocol	Сору	View	Back	

Figure 57. History List Overview

The lock symbol a on the right corner of the user column indicates that the result is saved in a password protected user folder. If the result in the user folder has been deleted, the record will still be shown in history list.

If the user does not save the result after executing the application, the user column will display **"AutoSave"**, and the file column will show a system-defined name.

If the user saves the result to a flash drive, the user column will display "Flash Drive," and the name displayed in the file column will be user-defined.

The function icons on the lower part of the screen allow users to duplicate the result settings to a **New Protocol** or **Copy**/ **New** the results.

15.1 Duplicating Protocol Setting

Select the saved results with the protocol setting you want to duplicate. Tap on the **New Protocol** to duplicate the protocol settings. The new protocol will have the same settings as the original, but modification is still possible.

15.2 Copying a Result

Select a result and tap **Copy** to copy a result. If the result is from a passwordprotected user folder, you'll be requested to input the password. Input the password and tap on **v** to confirm the password or tap on **x** to abort the operation. If the password is entered correctly, the report will then open. If the password is entered incorrectly, the following warning message will pop up: **"This is not the correct password, please try again"**. Tap on **v** to return to the password input screen and enter the correct password.

15.3 Viewing a Result

To view the details of a result, tap on the record to select and highlight it. You can then tap for the second time or tap on the **Wiew** to open it. If the result is from a passwordprotected user folder, you'll be requested to input the password. Input the password and tap on **v** to confirm the password or tap on **v** to abort the operation. If the password is entered correctly, the report will then open. If the password is entered incorrectly, the following warning message will pop up: **"This is not the correct password, please try again**". Tap on **v** to return to the password input screen and enter the correct password.



16 User Menu

Tap on the **User** on the main menu to enter the **User Menu**. A sample screen is shown in Figure 58 below.



Figure 58. User Folder Overview

All results can be stored inside the user folders. User Menu contains a public folder which is unlocked so users can view results freely. There are 10 user folders that can be full displayed on one page. If there are more than 10 user folders created in the system, user can scroll up or down the screen to see the other pages.

Users can utilize the function icons on the lower part of the screen to **create**, **delete**, **edit** and **open** the user folders. The **Back** icon is used to return to the main menu. The detail icon function is described below.

16.1 Creating a New User Folder

Tap on the icon **New User** to create a new User folder. Input the user folder name and chose one of the head shot icon (8 different icons are available (Figure 59)), and then enter the password (optional). If the user folder has a password, the lock symbol **f** on the lower right corner of the folder icon indicates that the folder is password-protected (Figure 58).



Figure 59. Head Shot Icon

16.2 Deleting a User Folder

Tap on the folder icon to select and highlight the folder, then tap on the **Delete** to delete it. You are required to enter the password if the folder is password-protected. The screen will prompt: "**Are you sure you want to delete the folder with a report inside?**" Tap on to confirm. Tap on **X** to cancel.

16.3 Viewing a User Folder

Tap on the folder icon to select and highlight it. Then, you can then tap for a second time or tap on the Open to open it. If the folder is password-protected, you'll be requested to input the password. Input the password and tap on to confirm the password or tap on to cancel. If the password is entered correctly, the folder will then be opened. If the password is entered incorrectly, the following warning message will pop up: "This is not the correct password, please try again". Tap on enter the correct password.

16.4 Editing a User Folder

To edit the properties of a user folder, tap on the folder icon to select and highlight the folder, then tap on the **Edit** to edit it. Users can change the folder name and password (optional) or change the head shot icon. Tap on **v** to store and finish editing.

16.5 Using a Flash Drive as a User Folder

To use a **USB flash drive** as a **User Folder** to store the results, please insert the flash drive with **FAT** or **FAT32** format into the front USB-A port, and the icon is will pop up. (The icon loading time depends on the specification of the flash drive.). The saved file format will be .**csv** file. The user can open it with Excel.

User can also use the flash drive to transfer the results between the EzDrop 1000/1000C and a computer.



17 🛃 PC Connection

In addition to stand-alone operation, EzDrop can also be connected to a computer.

Download the PC software on Blue-Ray Biotech Website.

- EzDrop 1000 Micro-Volume Spectrophotometer > Resources > EzDrop 1000 PC software
- EzDrop 1000C Micro-Volume / Cuvette Spectrophotometer > Resources > EzDrop 1000C PC software
 - a. Unzip the PC software file and install the following 2 drivers:
 - Generic_USB_Driver_Setup_V1.0.6.exe
 - CP210xVCPInstaller_x64.exe
 - **b.** Connect the EzDrop 1000/1000C to PC via USB-B cable (Figure 2). Turn on the EzDrop 1000/1000C and wait to enter the standby screen (Figure 4).
 - c. Open the 🥏 EzDrop software.



Figure 60. The Standby Page of EzDrop PC Software

- d. Click the **EzDrop** icon to connect.
- e. When the PC Software is connected to EzDrop 1000/1000C successfully, the connection status on the upper right corner will turn to " Connected", and it will enter to the Main Page (Figure 61). The touch screen of EzDrop 1000/1000C will turn to connecting picture (Figure 62). Please refer to the Table 3 & Table 34 for the detailed description.



Figure 61. The Main Page of EzDrop PC Software

Lab: Blue-Ray Biotech Ve	ersion: 3533C	2022-12-15 09:59
Ez	Drop	
Co	nnecting	

Figure 62. The Screen on EzDrop 1000/1000C When it is Connected to PC

Note

- 1. The PC software cannot read any reports of the EzDrop 1000/1000C, including dye lists or Kinetics methods. Both are regarded as independent.
- 2. When the EzDrop 1000/1000C is connected to the computer, the touch screen of the EzDrop 1000/1000C will be untouchable.


Table 34. Detailed Description for 3 Tools & Setting on Main Menu of PC Software	

lcon	Function	Description
¢	System	System setting. Only three functions can be used. For the details, please refer to the following section: 14.6 Self-Test 14.4 H Indicator Light 14.8 Operation Manual
Ĩ	View	View saved result.
i	About	Users can check the basic information of EzDrop 1000/1000C with this icon, including the System Version, Initialization Date and Calibration Date.

18 Maintenance

18.1 Cleaning the EzDrop 1000/1000C

Please avoid liquid spilling onto or into the instrument. Liquid may damage EzDrop 1000/1000C internal components. In addition, periodically use a dry cloth to wipe it clean of dust and other residues that come with the regular operation of the instrument.

18.2 Cleaning the Quartz Glass of Sample Window/Cover

18.2.1 Daily Cleaning

It is better to clean the quartz glass every time before starting and after finishing experiments. It is also recommended to clean the quartz glass between measurements.

- 1. Pipette 2 uL ddH₂O onto the sample window and lower the detection arm.
- 2. Wipe away the ddH₂O from sample window//cover with lint-free paper and clean the sample window/cover by ddH₂O

18.2.2 Decontamination Cleaning

If there is a contamination issue, 0.5% sodium hypochlorite solution (10-fold dilution of commercial bleach, freshly prepared) can be used to do the decontamination cleaning. Please follow the procedure below:

- 1. Pipette 2 uL 0.5% sodium hypochlorite solution (bleach) onto the sample window and lower the detection arm.
- 2. Wait for 2 minutes.
- 3. Wipe away the 0.5% sodium hypochlorite solution from sample window/cover with lint-free paper.
- 4. Pipette 2 uL ddH₂O onto the sample window and lower the detection arm.
- 5. Wait for 2 minutes.
- 6. Wipe away the ddH₂O from sample window/cover with new lint-free paper.



18.2.3 Deep Cleaning

If there has been a dried sample left on the quartz glass of the sample window or routine doing protein sample detection, use a lint-free paper dampened with **70% Ethanol** to lightly wipe the sample window/cover and then dry the surface with a new lint-free paper.

If it still cannot be cleaned, 0.5M HCl can be used for deep cleaning. Please follow the procedure below:

- 1. Pipette 2 uL 0.5M HCl onto the sample window and lower the detection arm.
- 2. Wait for 2 minutes.
- 3. Wipe away the 0.5M HCl from sample window/cover with lint-free paper.
- 4. Pipette 2 uL ddH₂O onto the sample window and lower the detection arm.
- 5. Wait for 2 minutes.
- 6. Wipe away the ddH₂O from sample window/cover with new lint-free paper.

18.3 Cleaning the Cuvette Holder (1000C only)

- Clean the dust in the cuvette holder with canned air.
- Please avoid liquid flowing into the cuvette holder; if the liquid goes into the cuvette holder, use a dry cloth to absorb the liquid immediately.
- For the cleaning and maintenance of the cuvette, please follow the manufacturer's instruction manual.

	 Use only a dry, soft, lint-free paper to clean the front screen. Do not use a spray bottle to apply water or any other solutions onto any surface of the instrument as the liquid may damage internal components.
Note	 Do NOT use Hydrofluoric Acid (HF) on the quartz glass as the fluoride ion will dissolve the hydrophobic coating.
	4. Please use 0.5M HCl / 0.5% sodium hypochlorite solution (bleach) carefully and do NOT spray it onto a metal part of the path length selector as it may corrode it and affect the path length.

18.4 Re-Coating of Hydrophobic Layer

If the sample on the sample window cannot form a liquid column, the quartz glass may need nano-hydrophobic layer recoating. Please contact your local Blue-Ray Biotech distributor for this service.

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108

18.5 Annual Maintenance

For the best performance, it is suggested to do annual maintenance on the EzDrop 1000/1000C. This includes flash calibration, light path confirmation and nano hydrophobic layer recoating. Please contact your local Blue-Ray Biotech distributor for the service.

115

18.6 Replacement

When the surface of quartz glass has been damaged, please contact your local Blue-Ray Biotech distributor for exchange service.

19 Troubleshooting

	Problem	Cause	Action
	The display is	Power is not reaching the system.	Check power source voltage.
	off even when the power is	Power cord is not plugged into the socket properly.	Reconnect the power cord.
	switched on	Faulty Power adaptor.	Return the EzDrop for service.
		The solutions are not homogenous and well-mixed prior to sampling.	Ensure all solutions are homogenous and well-mixed prior to sampling.
		Sample has air bubbles.	Remove air bubbles from sample.
		There are scratches on the surface of quartz glass.	Return the unit for service.
9	Can't reach sample accuracy	Quartz glass surface is contaminated.	Clean the quartz glass above and below with a suitable solution.
		Pulsed Xenon flash lamp problem.	Return the EzDrop for service.
		Optics module problem.	Return the EzDrop for service.
		Optical fiber problem.	Return the EzDrop for service.
		Mechanism alignment problem.	Return the EzDrop for service.
	Detection time is	Faulty electronic module.	Return the EzDrop for service.
	too long	Faulty optics module.	Return the EzDrop for service.
	Path length selector does not	Faulty path length selector mechanism.	Return the EzDrop for service.
	work	Path length selector sensor problem.	Return the EzDrop for service.
	No beep sound	Sound may currently be set to off.	Check Beeper setting in System Mode.
icons Faulty touch panel.		Faulty touch panel.	Return the EzDrop for service.

19.1 Error M	essages	
Error message appears	Refer to Section 19.1 Error Messages.	Check the nature of the error and take the suggested actions.
Detection arm will not close	Faulty detection arm mechanism.	Return the EzDrop for service.
	Foreign object between detection arm and the area inside the detection arm.	Remove the foreign object or matter.
Display is too dark or bright	Display brightness is not adjusted properly.	Adjust Display Brightness Potentiometer.
off	Faulty LCD panel.	Return the EzDrop for service.
The display goes	Faulty backlight.	Return the EzDrop for service.

19.1 Error Messages

19.1.1 EzDrop 1000/1000C Error Messages List

Message	Cause	Action
System Information No SD Card	Did not receive SD card signal in 1 second continuously.	 Insert the SD card. Return the EzDrop for service.
System Information The flash intensity is out of range, please check and clean the window on the pathlength selector. If this message keeps pop up, please contact your distributor for technical support	Automatically detected numerical anomalies. Insufficient light source intensity or excessive noise.	 Check the detection arm is closed while testing. Reboot the EzDrop. Return the EzDrop for service.
Error Message 3 Heater temperature detect abnormal. NTC error.	Heater temperature sensor problem.	Reboot the EzDrop.
Error Message 4 Heater can't reach set temperature. Can't reach set temperature in 3 m inutes.	Heater cannot reach set temperature in 3 minutes.	Reboot the EzDrop.
Error Message 5 ERR_ FW_ABNORMAL	MCU Electronic module Board problem.	Reboot the EzDrop.



Error Message 6 ERR_ HW_ABNORMAL	MCU Electronic module Board problem.	Reboot the EzDrop.
Error Message 7 ERR_ METER_NO_ANSWER	Spectrometer Electronic module Board problem.	Reboot the EzDrop.
Error Message 8 ERR_ METER_CALIBRATE	Spectrometer Electronic module Board problem.	Reboot the EzDrop.
Error Message 9 ERR_ UART_NO_ANSWER	ARM Electronic module Board problem.	Reboot the EzDrop.
Error Message 10 ERR_ UART_WRONG_ANSWER	ARM Electronic module Board problem.	Reboot the EzDrop.
Error Message 11 ERR_ UART_WRONG_COMMAND	ARM Electronic module Board problem.	Reboot the EzDrop.
Error Message 12 ERR_ UART_TRANSMIT_ OVERFLOW	ARM Electronic module Board problem.	Reboot the EzDrop.

If the same error message appears after rebooting the unit, please return the instrument for service.

19.1.2 PC Software Error Messages List

Message	Cause	Action
System Information The flash intensity is out of range, please check and clean the window on the pathlength selector. If this message keeps pop up, please contact your distributor for technical support	Automatically detected numerical anomalies. Insufficient light source intensity or excessive noise.	 Check the detection arm is closed while testing. Reboot the EzDrop. Return the EzDrop for service.
Error Message 4 Heater temperature detect abnormal. NTC error.	Heater temperature sensor problem.	Reboot the EzDrop.
Error Message 5 Heater can't reach set temperature. Can't reach set temperature in 3 minutes.	Heater cannot reach set temperature in 3 minutes.	Reboot the EzDrop.
Error Message 6 ERR_ FW_ABNORMAL	MCU Electronic module Board problem.	Reboot the EzDrop.

Error Message 7 ERR_ HW_ABNORMAL	MCU Electronic module Board problem.	Reboot the EzDrop.
Error Message 8 ERR_ METER_OPEN	Spectrometer Electronic module Board problem.	Reboot the EzDrop.
Error Message 9 ERR_ METER_READ	Spectrometer Electronic module Board problem.	Reboot the EzDrop.
Error Message 10 ERR_ METER_CONFIG	Spectrometer Electronic module Board problem.	Reboot the EzDrop.
Error Message 11 ERR_ METER_NO_ANSWER	Spectrometer Electronic module Board problem.	Reboot the EzDrop.
Error Message 12 ERR_ METER_CALIBRATE	Spectrometer Electronic module Board problem.	Reboot the EzDrop.
Error Message 13 ERR_ UART_NO_ANSWER	ARM Electronic module Board problem.	Reboot the EzDrop.
Error Message 14 ERR_UART_WRONG_ANSWER	ARM Electronic module Board problem.	Reboot the EzDrop.
Error Message 15 ERR_UART_WRONG_COMMAND	ARM Electronic module Board problem.	Reboot the EzDrop.
Error Message 16 ERR_UART_TRANSMIT_OVERFLOW	ARM Electronic module Board problem.	Reboot the EzDrop.

Appendix A: Technical Specifications

Micro-Volume Mode			
Minimum Sample Volume	1 µL		
Pathlength	0.5, 0.05 mm		
Light Source	Xenon flash lamp		
Detector Type	2048 element CMOS		
Wavelength Range	190 ~ 1000 nm		
Wavelength Accuracy	±1.0 nm		
Spectral Resolution	1.5 nm (FWHM at Hg 253.7 nm)		
Absorbance Precision	Raw: 0.0015 A (0.5 mm); 0.03 A (1 cm equivalent)		
Absorbance Accuracy	1.5% at 1.0 A at 300 nm		
Absorbance Range (1 cm equivalent)	0.04 ~ 400 A		
Detection Panage	dsDNA: 2 ~ 20000 ng/µL		
Detection Range	BSA: 0.06 ~ 600 mg/mL		
Measurement Time	< 3 seconds		
Cu	Cuvette Mode (1000C only)		
Pathlength	10, 5, 2, 1, 0.5, 0.2, 0.125, 0.1 mm		
Absorbance Range (1 cm equivalent)	0.002 ~ 1.5 A		
Detection Range	dsDNA: 0.3 ~ 75 ng/µL BSA: 0.003 ~ 2.25 mg/mL		
Stirring	8 Speeds (150 ~ 850 rpm)		
Temperature Control	37 ~ 45°C ±0.5 °C (Quartz Cuvette)		

Software		
Operating System	Custom Linux based OS	
PC Software Requirement	Windows® 7 and 10, 64 bit	
General		
Display	7" touch screen, 1280*800 high-definition color display	
Connectivity	USB-A port x1 (Data output); USB-B port x1 (PC connection)	
Footprint Dimensions (W \times D \times H)	206 x 333 x 166 mm (8.1 x 13.1 x 6.5 ln.)	
Weight	3.3 kg (7.3 lbs.)	
Glove Compatibility	All common lab gloves	
Internal Storage	32 GB flash memory	
Operating Voltage	Input: AC 100 ~ 240 V, 50/60 Hz; Output: DC 24 V, 2.08 A	
Certifications	CE, UKCA	

Specifications are subject to change without prior notice.



Appendix B: CE & UKCA Declaration

CE RR **BLUE-RAY BIOTECH CORP.** 5F., No. 2, Aly. 2, Siwei Ln., Zhongzheng Rd., Xindian Dist., New Taipei City 231, Taiwan (R.O.C.) Declaration of Conformity Product Name: Micro-Volume / Cuvette Spectrophotometer Model Names: EzDrop 1000/1000C All models comply with the following European & UK standards: EMC: EN IEC 61326-1. EN IEC 61326-2-6 BS EN IEC 61326-1. BS EN IEC 61326-2-6 Safety: EN 61010-1 and EN 61010-2-101 BS EN 61010-1. BS EN 61010-2-101 To the best of my knowledge and belief, these units conform to these standards. Name: Jimmy Kuo Position: Quality Assurance Manager Issue Date: 2023.03.10

Appendix C: Order Information

Cat. No.	Description
BRED-1000	EzDrop 1000 Micro-Volume Spectrophotometer
BRED-1000C	EzDrop 1000C Micro-Volume / Cuvette Spectrophotometer
T01-000182-00	Nano-hydrophobic coating maintenance kit
219-110000-00	Quartz cuvette w/ cover
207-000535-00	Stirrer bar 6 x Φ3 mm



For more information, visit <u>www.blue-raybio.com</u>

EzDrop 1000/1000C Product Webpage

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