

ReadyToProcess WAVE 25

READYTOPROCESS

The single-use ReadyToProcess WAVE™ 25 bioreactor system is a reliable and intuitive cell culture device for working volumes up to 25 L (Fig 1). The system is based on a well-known rocking technology that provides mixing and aeration to the culture, which is maintained in an inflated, disposable Cellbag™ bioreactor cultivation chamber.

With ReadyToProcess WAVE 25, the rocking technology is enhanced with features such as advanced sensors and intelligent control strategies. System operation is easily managed via the intuitive and user-friendly UNICORN™ system control software. The system is designed for fast installation and convenient handling, and delivers a reliable and accurate performance suitable for research, process development, and manufacturing operations.

ReadyToProcess WAVE 25 offers the following benefits:

- Robust and reliable performance
- Intelligent and accurate process control
- Designed for ease of use
- Flexible operations with possibility of use in single or dual culture mode
- Suitable for manufacturing in a regulated environment

System benefits

Robust and reliable performance

ReadyToProcess WAVE 25 is designed to make hardware, consumables, and software work together in an integrated manner. The system has been tested in real applications for reliable performance. To decrease the risk for data losses, the system comprises two, mirrored, solid-state drives for data storage.

Intelligent and accurate process control

The system is equipped with advanced sensors and automated controllers for temperature, liquid handling, pH, and dissolved oxygen (DO). The system dynamics have been methodically characterized and the results used to create a library of predefined control parameters tailored for different cultivation volumes. The system can automatically determine suitable parameters for a run, a feature that facilitates fast start-up and enables accurate and stable control.



Fig 1. ReadyToProcess WAVE 25 bioreactor system can be operated in single or dual culture mode.

Easy to use

ReadyToProcess WAVE 25 system is designed for ease of use. Cellbag bioreactor attachment is straightforward and system operation is conveniently managed with the UNICORN software, with menus and process pictures that are easily accessible and interpretable. The harvest operation is facilitated by an innovative tilt function that omits the need for heavy lifting.

Flexible operations with possibility of use in single or dual culture mode

With ReadyToProcess WAVE 25, one or two cultures can be run at the same time with high control accuracy. When dual mode is used, two cultivations can be run using the same or different bag sizes and/or working volumes. Parameters such as temperature, pH, and DO can be controlled independently in the two Cellbag bioreactors. Separate pump units control liquid addition and/or removal, for example, to enable tailored feeding strategies, base addition, or perfusion in the two parallel cultures.



- 1 Graphical user interface**
- 2 Lid**
- 3 ReadyToProcess™ Pump 25**
Peristaltic pump
- 4 ReadyToProcess CBCU**
Gas flow/mix controller, pH and DO sensors
- 5 ReadyToProcess WAVE 25, rocker**
Rocker, temperature sensors, scale, embedded controller, and dual hard drives
- 6 Tray**
- 7 Client computer**
Installed with UNICORN software.
Can control up to three ReadyToProcess WAVE 25 systems

System setup and integration of subunits

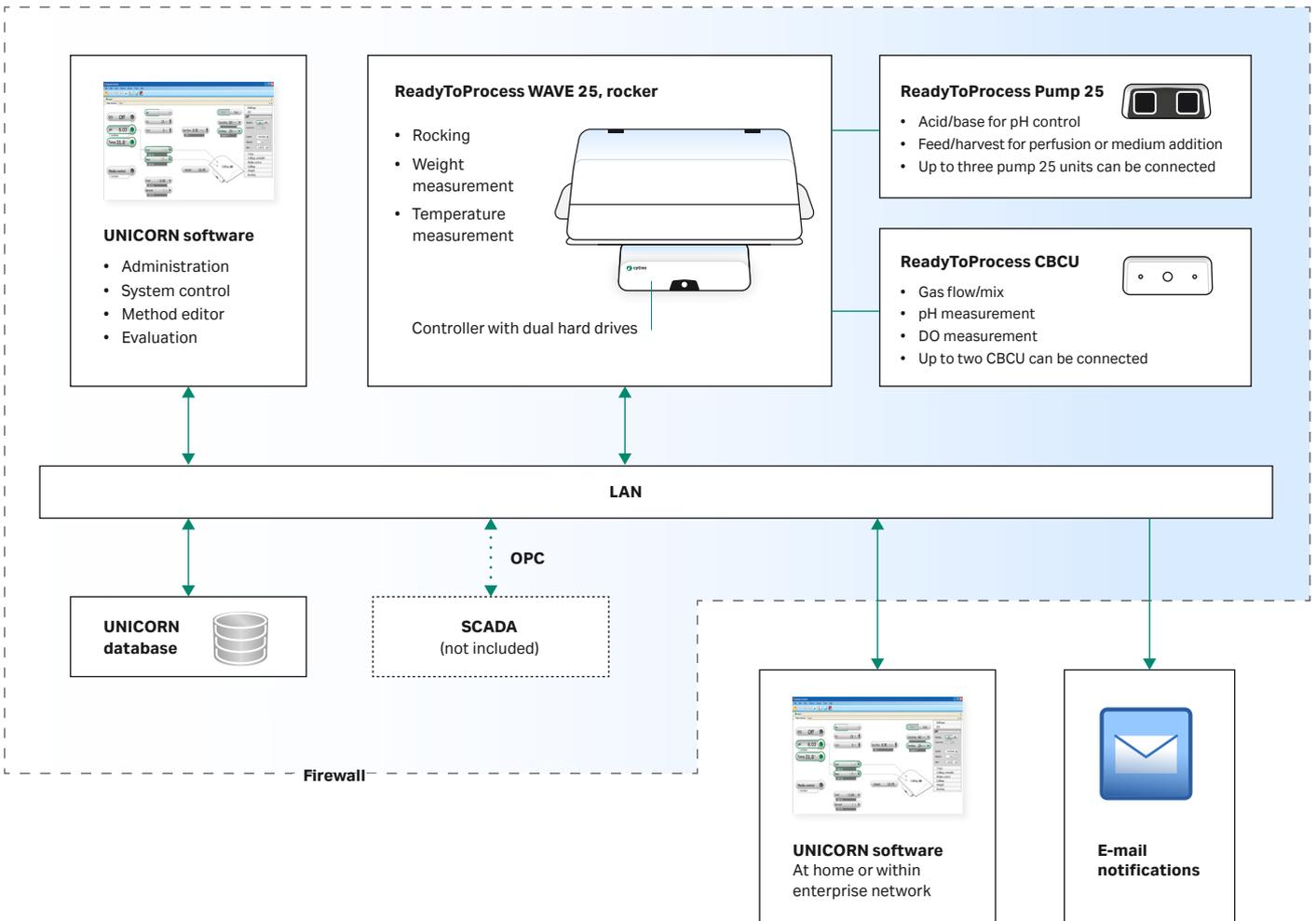


Fig 2. ReadyToProcess WAVE 25 system overview. The System Control module on the client computer is used to start and monitor the cultivation process. One UNICORN client can control up to three ReadyToProcess WAVE 25 systems simultaneously. SCADA = supervisory control and data acquisition, OPC = open platform communications, LAN = local area network.

Suitable for manufacturing in a regulated environment

ReadyToProcess WAVE 25 is designed to meet the demands and standards that are required in a regulated environment. The documentation is extensive and includes material certificates, system specifications, installation and operational qualification (IQ/OQ) protocols, and a detailed user manual. The UNICORN software is suitable for use in a manner that complies with 21 CFR Part 11 and Good Automated Manufacturing Practice (GAMP™)5.

System overview

ReadyToProcess WAVE 25 system components

The ReadyToProcess WAVE 25 system consists of a rocker, CBCU gas mixers, and pumps, all operated by the UNICORN software installed on a client computer. The rocker is the main unit and is used together with a tray and a disposable Cellbag bioreactor. When operated in dual culture mode, culturing can be performed in two separate Cellbag bioreactors simultaneously, although controlled separately. The rocker has multiple functions, amongst them temperature control, culture mixing, and weight measurement. The CBCU contains a gas mixer that delivers gas of a defined composition to the culture and is used together with the optical sensors of the Cellbag bioreactor for online control of culture pH and DO. Liquid is delivered accurately to the culture by ReadyToProcess Pump 25. The pump unit has a flow range covering multiple applications, from additions of feed and base to perfusion culturing. Up to two CBCU gas mixers and up to three pump units can be connected to ReadyToProcess WAVE 25. An overview of the ReadyToProcess WAVE 25 system is shown in Figure 2. The individual components are described in detail below.

ReadyToProcess WAVE 25, rocker

The rocker is the main hardware unit of the system and provides mixing through the rocking, reliable temperature measurement from integrated sensors, and accurate weight measurement from integrated load cells. The ergonomic design makes handling convenient and easy. The ability to handle the bioreactor in a tilt position facilitates sampling and harvest (Fig 3). The minimal footprint facilitates placement when space is limited.

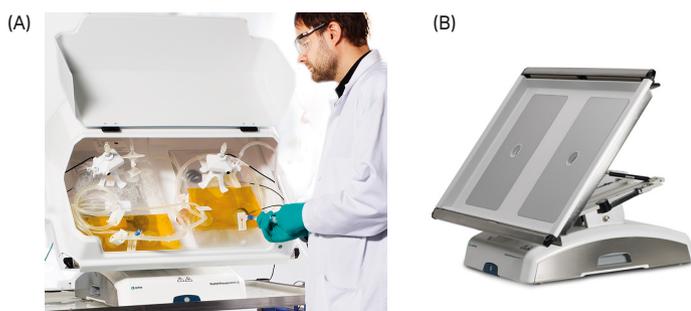


Fig 3. (A) The ergonomic design makes activities such as sampling and harvest convenient and easy. (B) ReadyToProcess WAVE 25 in tilt position.

Trays and lids

Trays are available in three different sizes for culturing of up to 5 L, 10 L, and 25 L, respectively. The trays are easily attached to the rocker when in the tilt position. A snap lock mechanism allows Cellbag bioreactors to be correctly installed and rapidly changed (Fig 4). To protect light-sensitive components of the culture medium and to prolong the life of the optical sensors, lids are available for all tray sizes.



Fig 4. The tray has an easy lock mechanism for convenient and proper attachment of Cellbag bioreactors.

Temperature control

Efficient and evenly distributed heating is provided by the tray heater plate. The temperature control is managed by sensors that are integrated in the rocker base unit to enable reliable temperature measurement and to minimize the need for calibration when changing trays (Fig 5). To minimize the risk for overheating, heating is only enabled when the rocker is in motion. For an accurate, stable, and fast temperature control, the heater power output is automatically adjusted based on the Cellbag bioreactor size and content weight. Temperature control in two separate Cellbag bioreactor cultures when using the dual functionality is shown in Figure 6.



Fig 5. Integrated temperature sensors for convenient handling and reliable temperature control.

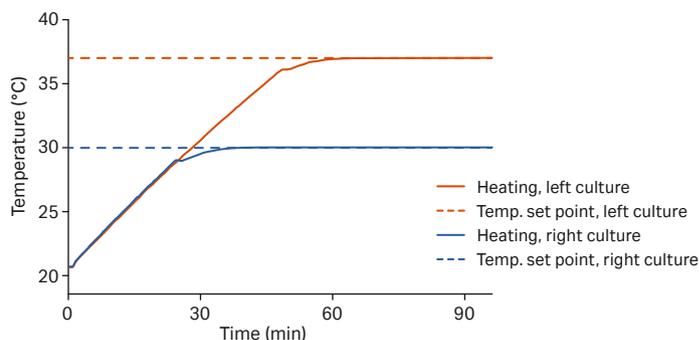


Fig 6. Temperature is accurately controlled independently in two separate 5 L cultures using the dual functionality (ambient temperature 22°C).

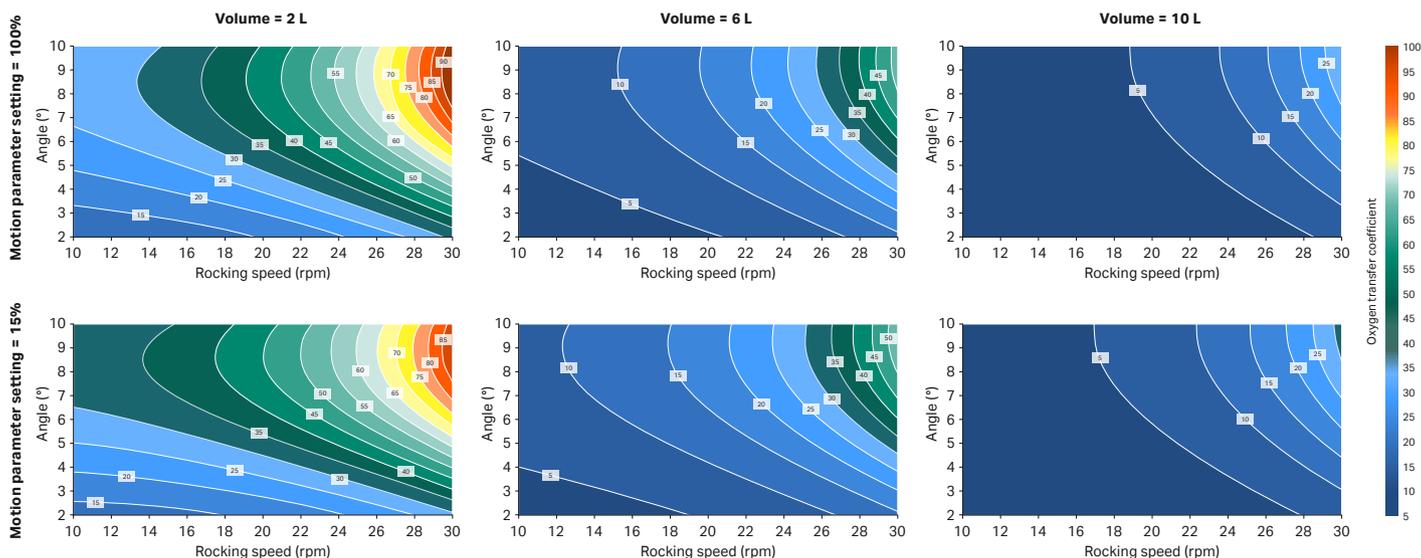


Fig 7. 4D contour plot of the oxygen transfer coefficient at the highest (100%) and lowest (15%) rocking motion parameter settings.

Mixing and gas transfer

The adjustable rocking parameters are speed, angle, and motion. The parameter settings, in combination with the cell culture volume, affect the mixing and gas transfer rate in the Cellbag bioreactor. The speed parameter determines the number of rocking cycles per minute, and the angle parameter relates to the tray's degree of tilting at the turning points. The motion parameter determines the acceleration profile. The lowest motion parameter setting, 15%, gives an almost constant speed throughout the rocking cycle, whereas the highest setting, 100%, gives a faster speed in the middle of the cycle and slower at the turning points. Whereas oxygen transfer increases with increasing rocking speed and angle, the rocking motion pattern only has a minor effect on the oxygen transfer rate (Fig 7).

Weight measurement

Load cells integrated in the rocker provide accurate and continuous weight measurement of the culture and eliminate the need for external scales. In Dual mode, separate weight measurements are provided for each Cellbag bioreactor. The load cells communicate with the system control function for proper media handling and tailored regulation of pH and temperature. The weight measurement functionality is also used for automatic calibration of the pumps in perfusion cultures. An adjustable foot enables equal weight distribution between the load cells also on nonlevel surfaces, for increased precision of the weight measurement.

Data storage and communication

Data and control parameters from the cell culture run are stored on two independent, mirrored, solid-state drives integrated in the rocker. Thus, once cultivation has been started and data from the instrument modules have been collected, the run can continue without the need for a network connection. The use of two solid-state drives enables culturing without losing any data even if only one drive is functional. After a run, data is stored in a UNICORN database on an external computer. For facilitated integration into a larger manufacturing operation, the rocker also contains software for enabling direct communication with the system via an open platform communication (OPC) link.

ReadyToProcess CBCU

The ReadyToProcess CBCU gas mixer is a compact unit with multiple functions. The unit is primarily used for providing gas to the culture and for monitoring of pH and DO. The unit contains a mass flow controller, sensors for gas pressure and O₂ and CO₂ concentrations, and transmitters for pH and DO (Table 1).

Table 1. ReadyToProcess CBCU gas mixer overview

Feature	Description
CO ₂ /O ₂ /air mix controller	Depending on the configuration, air is mixed with CO ₂ and/or O ₂ according to the set-points. Nitrogen may be used instead of air for maintaining a low oxygen environment for near-anaerobic applications.
Gas flow controller	The gas-mix flow is measured and controlled by a mass flow controller (MFC). A correct volumetric flow is achieved by compensation for CO ₂ concentration. Quick filling of gas into the Cellbag bioreactor, enabled by the fast-fill function, significantly reduces start-up time. An alarm will inform the user if pressure sensors detect under or overpressure at the gas inlets or within the Cellbag bioreactor.
pH measurement	The pH is measured with optical pH sensors preinstalled in Cellbag bioreactors of pHOPT type. The sensor is connected to the CBCU via an optical fiber cable.
DO measurement	The DO level is measured with optical DO sensors preinstalled in Cellbag bioreactors of DOOPT type. The sensor is connected to the CBCU via an optical fiber cable.

Three CBCU configurations are available:

- CO₂, O₂, and pH
- CO₂, O₂, and DO
- CO₂, O₂, pH, and DO

The most suitable CBCU configuration will depend on the specific application.

ReadyToProcess Pump 25

The ReadyToProcess Pump 25 is a peristaltic unit incorporating two roller pumps for feed, harvest, and pH control. The pump design makes tubing installation convenient and easy. Tubing sizes from 0.5 mm (1/50") to 4.8 mm (3/16") internal diameter can be used to support flow rates from 0.07 to 100 mL/min. Flow rate is regulated by weight feedback from the scale and is controlled by automatic adjustments of the pump speed. Manual adjustment of the flow rate is also possible.

Up to three pump units with all together six pump heads can be connected to the rocker. Up to six pump roles can be selected from a set of 12 different possible options (*left: acid, base, feed1, feed2, feed3, or harvest; and right: acid, base, feed1, feed2, feed3, or harvest*) for the dual mode and seven different possible options (*acid, base, feed1, feed2, feed3, feed4, or harvest*) for the single mode.

Media handling and perfusion using calibrated pumps

Calibration of pumps can be performed manually or automatically during an ongoing perfusion process. Automatic calibration is possible for flow rates above 3.5 L/d and is easily managed by entering the tubing diameter in the UNICORN control software.

Automated pH control

To minimize fluctuations in pH during adjustments with CO₂/base or acid/base, the flow rate of the acid/base pumps needs to be rigorously controlled. The ReadyToProcess WAVE 25 system uses combined information from the load cells, the current process pH value, the pump tubing diameter, and the acid/base molarity for calculation of the needed flow rate during the run.

Automation of process parameters in pH and DO control

The UNICORN software automatically sets the desired PID* control parameters based on Cellbag bioreactor size and gas flow set-point. The system also adapts the parameters during the run, for optimization to reach a new set-point or to maintain a current set-point.

* Proportional-integral-derivative

UNICORN software

UNICORN system control software consists of four modules: **Administration**, **Method Editor**, **System Control**, and **Evaluation**. A comprehensive help tool is also included.

Administration

The **Administration** module is used to administer all functions of the UNICORN software. Available functions include user and email setup, controlling access to groups and network users, defining and editing system properties, database management, and logging records of usage and activity.

Method editor

Instructions to control a bioreactor run can be defined in a method. The **Method Editor** module is used to create, edit, save, and work with methods. An existing method can be changed to simplify the editing process. Individual changes can be saved for later use on systems having the same instrument and component configuration.

System control

The **System Control** module is used to connect to the system as well as to start, view, and control a run (Fig 8). Default parameter values for an instrument can be viewed and edited in the **System Settings** dialog before a run is started or during an ongoing run using manual instructions. It is also possible to connect to other systems.

The user-friendly and intuitive **Process Picture** pane enables manual interaction with the system and provides status of the run parameters. Data is shown in the process picture but can also be viewed as curves in the **Charts** tab. Curves and information about the run are saved in a database, which can be opened in the **Evaluation** module. The default view shows the curves most commonly used. The user may customize which curves to display and the color and style of the displayed curves.

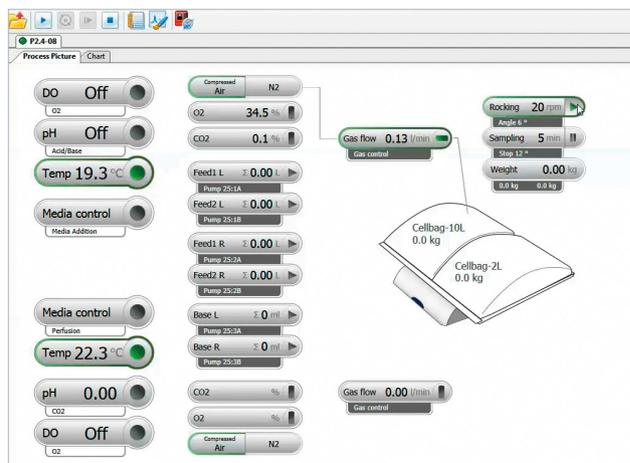


Fig 8. Monitoring and controlling all aspects of the culture is managed through the user-friendly interface.

Evaluation

In the **Evaluation** module, content of the result files can be viewed, analyzed, and compiled as reports. Reports can be customized, saved, and printed.

Alarms, notifications, and data logging

The UNICORN software allows easy access to all operations, data, and alarm conditions. A dynamic, graphical user interface shows the real-time status of the run, while data are automatically saved. From the UNICORN software, the system is easily configured to trigger an alarm when certain conditions are met. Individual deviation alarms can be set for all essential parameters (heating, gas flow, weight, O₂, CO₂, pH, and DO). The regulation of alarm sensitivity and delay is defined by the user. If such an alarm condition is triggered, the part of the system generating the alarm is highlighted in the graphical user interface and an alarm dialog is displayed. The alarm dialog displays information about the alarm, such as date and time of occurrence, as well as a help text describing the cause of the alarm and how to solve the problem. The system is easily configured to send email notifications of alarms and errors.

Regulatory readiness

The ReadyToProcess WAVE 25 system is suitable for biomanufacturing of regulated products under various Quality Management Systems. The use of UNICORN software in a 21 CFR Part 11 and GAMP 5 compliant manner enables use of the system in a regulated environment. Individual user access permissions can be set and individual users are password-protected. Active processes can be locked to enable unattended operations without the risk of unauthorized interference. All records are stored in a single, unalterable database, including results and extended run documentation. Additional validation support includes comprehensive documentation on control system validation as well as IQ/OQ services. The IQ/OQ offers proven test procedures, verifying that the equipment has been installed in accordance with system drawings and specifications. The IQ/OQ also assures that the system operates as specified in the design, satisfying functional requirements. The IQ/OQ protocols enable quality assurance and allow regulatory reviewers to verify that all functional testing of the quality critical equipment and components, including the requirements of 21 CFR Part 11, has been performed and documented.

Available validation support documentation includes:

- Detailed description of the development model used for UNICORN software
- 21 CFR Part 11 system assessment in checklist format
- Audit report and 21 CFR Part 11 conclusion on functionality by an external, independent expert

Networking capabilities

UNICORN 7 operates in Windows® 7 and Windows 10 environments and the network ability allows real-time control from a remote or local computer. Communication is Ethernet-based and each instrument is controlled by a built-in instrument server. One database can be connected to 32 systems and up to three instruments can be controlled simultaneously from one UNICORN client. Results are saved locally, in the rocker, during the run and then stored on the database server. Because results and instructions are saved locally on the embedded drives, a run can continue even in the event of a network communication failure.

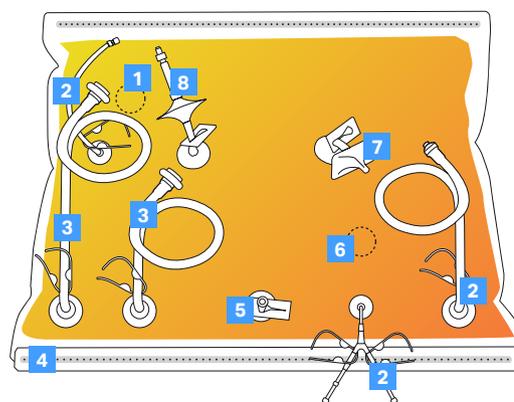
UNICORN software can be integrated in a supervisory control and data acquisition (SCADA) system such as the Emerson DeltaV™ control system using the UNICORN OPC server. Please contact your local representative for further information.

Cellbag bioreactors

Presterilized Cellbag bioreactors are single-use bags for the noninvasive mixing of culture medium and cells during cultivation (Fig 9). Cellbag bioreactors require no sterilization or cleaning steps. The disposable bioreactors provide a suitable environment for cell growth while minimizing the cross-contamination risk.

The bags are manufactured from multilayer, laminated, clear USP Class VI plastics and are easily connected to the full suite of ReadyToProcess cell culture, purification, and fluid handling products. Table 2 shows possible combinations of Cellbag bioreactors and trays when culturing in single or dual mode.

Cellbag bioreactors are available in two films: Bioclear™ 10 and the low-antioxidant Bioclear 11. For more information about Cellbag bioreactors, see data file 28-9511-36.



- | | | | |
|---|-----------------------------------|---|-----------------------------------|
| 1 | Optical DO sensor embedded in bag | 5 | Needleless sampling port |
| 2 | Optional tubing | 6 | Optical pH sensor embedded in bag |
| 3 | Inoculation/harvest lines | 7 | Air inlet filter |
| 4 | Cellbag rod | 8 | Air outlet filter |

Fig 9. Presentation of typical Cellbag bioreactor fittings.

Table 2. Possible combinations of Cellbag bioreactors and trays when culturing in single or dual mode

Cellbag bioreactor size (L)*	Bioreactor culture volume (L)	Tray	
		Single culture mode	Dual culture mode
2	0.3 [†] to 1	Tray 10, Tray 20	Tray 20
10	0.5 to 5	Tray 10, Tray 20	Tray 20
20	1 to 10	Tray 20	Not applicable
22	1 to 10	Tray 50	Tray 50
50	5 to 25	Tray 50	Not applicable

* All Cellbag bioreactor sizes are available with internal perfusion filter for perfusion cultures and/or can be equipped with single use optical pH (pHOPT) and DO (DOOPT II) sensors.

[†] Cellbag bioreactors with optical sensors require 300 mL minimal working volume. Depending on application and configuration, it might be possible to cultivate below the recommended minimum volume. However, it is highly recommended to stay above this volume for applications that require high agitation and pH and DO control. The temperature, pH, and DO sensors need to be submerged in liquid throughout the complete rocking cycle to function correctly.

System specifications

System specifications are listed in Table 3.

Table 3. ReadyToProcess WAVE 25 system specifications

General system specifications

Control system	UNICORN 6.3.2 or later version	
Dimensions, (W × H × D)		
Rocker	404 × 205 × 560 mm	
Tray 10	475 × 60 × 430 mm	
Tray 20	740 × 70 × 480 mm	
Tray 50	800 × 70 × 610 mm	
Lid 10	475 × 230 × 430 mm	
Lid 20	740 × 245 × 480 mm	
Lid 50	800 × 260 × 610 mm	
CBCU	276 × 117 × 360 mm	
Pump 25	275 × 115 × 280 mm	
Weight		
Rocker	24.0 kg	
Tray 10	4.5 kg	
Tray 20	7.3 kg	
Tray 50	9.5 kg	
Lid 10	1.7 kg	
Lid 20	3.3 kg	
Lid 50	3.9 kg	
CBCU	4.8 kg	
Pump 25	3.8 kg	
Power supply	100 to 240 V, ~ 50 to 60 Hz	
Power consumption	1500 VA	
Enclosure protective class	IP 21	
Gas supply (per CBCU)	Normal use	Fast fill
External air supply, 1.0 to 1.5 bar	1.3 L/min	3.5 L/min
External CO ₂ supply, 1.0 to 1.5 bar	0.2 L/min	0.5 L/min
External O ₂ supply, 1.0 to 1.5 bar	0.7 L/min	1.7 L/min

Environmental aspects

Operating ambient temperature range	15°C to 32°C
Operating humidity range	20% to 80% relative humidity (noncondensing)

ReadyToProcess WAVE 25, rocker

Rocking speed control range ¹	2 to 40 rpm
Rocking angle control range ¹	2° to 12°
Rocking motion control range	15% to 100%
Medium weight control range	0.2 to 25 kg
Scale, absolute accuracy	± (0.050 + 1% of load) kg
Scale, left/right absolute accuracy (dual mode)	± (0.1 + 6% of load) kg
Temperature sensor	Pt100 Class A
Temperature measurement range	2°C to 50°C
Temperature measurement accuracy	± 0.3°C in the range of 15°C to 50°C within ± 5°C from calibration temperature
Temperature control range	(ambient temperature 5°C) to 40°C
Temperature control accuracy (excl. measurement error)	± 0.2°C
Temperature set point difference (dual mode)	Max. 10°C at ambient temperature (21°C) Set point difference reduced by 1°C for each °C increase in ambient temperature (e.g., at ambient 25°C, max. set point difference is 6°C)

¹ When cultivating in a 50 L Cellbag bioreactor at maximum working volume of 25 L, rocking speed and angle multiplied should not exceed 240 rpm degrees. For example, if the rocking angle is set to 12 degrees, the rocking speed should not be set higher than 20 rpm.

ReadyToProcess CBCU

Gas flow control range	50 to 1000 mL/min
Total gas flow accuracy (reference flow – set point)	± (10 + 3% of read value) mL/min
Fast-fill flow	~ 3 L/min
CO ₂ control range	0% to 15% CO ₂
CO ₂ measurement accuracy at 5% CO ₂	± 0.5% CO ₂ when mixed only with air/N ₂
CO ₂ control accuracy (versus set point)	± 0.4% CO ₂
O ₂ control range	0% to 50% O ₂ when mixed with N ₂ , 21% to 50% O ₂ when mixed with air
O ₂ measurement accuracy	± (0.6% + 1% of read value) % O ₂ within 0% to 50% O ₂ when mixed only with air/N ₂
O ₂ control accuracy (versus set point)	± 0.6% O ₂
pH measurement range	pH 4.5 to 8.5
pH control range	pH 6.0 to 8.0
pH measurement accuracy	± 0.05 pH within ± 0.25 pH from offset calibration pH ± 0.1 pH within 0.25 to 0.5 pH from offset calibration
pH control accuracy (versus set point)	± 0.05 pH
DO measurement range	0% to 250% air saturation
DO measurement accuracy	± 5% air saturation (excl. atmospheric pressure variations)
DO control range	0% to 100% air saturation

ReadyToProcess Pump 25

Pump flow rate range	0.1 to 144 L/d (0.07 to 100 mL/min)
Pump flow accuracy	± (0.1 + 5% of read value) mL/min after calibration
Accumulated pumped volume accuracy	± 10% of measured volume
Supported tubing dimensions	i.d. 0.5 to 4.8 mm (1/50" to 3/16") wall thickness: 1.6 mm (1/16")

Applications

ReadyToProcess WAVE 25 is designed for a variety of cell culture applications involving mammalian, insect, and plant cells:

- Batch culture
- Fed-batch culture
- Perfusion culture
- Cultivation of adherent cells

Ordering information

Product	Description	Product code	Related literature	Product code
ReadyToProcess WAVE 25	Rocker	28988000	UNICORN 7.0.2 Manual Package	29191514
ReadyToProcess CBCU pH	Gas flow/mix and pH	29044213	UNICORN 7 software, data file	29135786
ReadyToProcess CBCU DO	Gas flow/mix and DO	29044216	UNICORN 7 Academia Package	29203853
ReadyToProcess CBCU Full	Gas flow/mix, pH, and DO	29044081	UNICORN 7 Process Development Package	29203854
ReadyToProcess Pump 25	Pump	29032003	UNICORN 7 Manufacturing Package	29203855
UNICORN 7 WrkStn pure-BP-exp	License	29128116	Disposable Cellbag bioreactors for WAVE Bioreactor™ systems, data file	28951136
UNICORN 7 Remote	License	29115426	Validation Support File UNICORN 7.0	28962650
UNICORN 7 Dry	License	29115427	ReadyToProcess WAVE 25, site preparation guide	29056702
UNICORN 7 Evaluation Classic	License	29115456	ReadyToProcess WAVE 25, operating instructions	29009597
UNICORN 7.0.2 DVD pack	Media (no license)	29195162	ReadyToProcess WAVE 25, system handbook	29009598
Tray 10	–	29044472		
Tray 20	–	29044473		
Tray 50	–	29044474		
Lid 10	–	29044475		
Lid 20	–	29044476		
Lid 50	–	29044477		
Filter heater*	–	29044471		
Bag sensor adaptor 2.5 m assembly†	Fiber cable for pH and DO control	28984189		
Tube kit CBCU‡	1	29112187		
Adjustable foot wrench‡	1	29112525		

* One delivered with the rocker, additional needed for dual functionality.

† Depending on configuration, one or two fiber cables are delivered with the CBCU unit.

‡ One delivered with the rocker.

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Cellbag bioreactors with integrated optical sensors are sold under a sublicense from Sartorius Stedim Biotech under US patent numbers 6,673,532, 7,041,493, and/or its foreign equivalents.

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