

BOND RX System

FULLY AUTOMATED IHC AND ISH STAINING SYSTEM

BOND RX 7 USER MANUAL



CE

Advancing Cancer Diagnostics
Improving Lives

Leica
BIO SYSTEMS

Legal notices

This manual applies to BOND RX, BOND RX^m, and the BOND RX System Controller.



Not all processing modules are available in all regions.

Trademarks

Leica and the Leica logo are registered trademarks of Leica Microsystems IR GmbH and used under license. BOND, BOND RX, BOND RX^m, BOND RX-ADVANCE, Covertile, Bond Polymer Refine Detection, Bond Polymer Refine Red Detection, Parallel Automation, Compact Polymer, and Oracle are trademarks of Leica Biosystems Melbourne Pty Ltd ACN 008 582 401. Other trademarks are the property of their owners.

Copyright

Leica Biosystems Melbourne Pty Ltd owns the copyright on this document and any associated software. Under law, our written permission is required before either the documentation or the software is copied, reproduced, translated, or converted to electronic or other machine-readable form, in whole or in part.

Copyright © 2024 Leica Biosystems Melbourne Pty Ltd

Product identification

Doc. 49.7540.501 A04

Manufacturer



Leica Biosystems Melbourne Pty Ltd
495 Blackburn Road
Mount Waverley VIC 3149
Australia

Important information for all users

This manual contains important information on how to use BOND RX. For the latest information on Leica Biosystems products and services, visit www.leicabiosystems.com.

Due to a policy of continuous improvement, Leica Biosystems reserves the right to change specifications without notice.

The following terms are used in this document:

- Leica Biosystems—refers to Leica Biosystems Melbourne Pty Ltd.
- BOND RX system - the Leica Biosystems research platform that includes the BOND RX and BOND RX^m
- BOND RX - a type of automated IHC and ISH staining instrument
- BOND RX^m - a type of automated IHC and ISH staining instrument
- BOND RX software - the software application through which users can configure and operate the BOND RX and BOND RX^m system

Intended users

Persons operating a BOND RX Processing Module must have received sufficient training to ensure that it is used in accordance with this document and be fully aware of any potential hazards or hazardous procedures, before operating the processing module. Only trained staff are to remove any covers or parts from the processing module, and only if instructed within this manual.

Installation and repairs

Installation and repairs must only be carried out by qualified service personnel authorized by Leica Biosystems.

Warranty claims can be made only if the product has been used for the specified application and operated according to the instructions in this document. Damage resulting from inappropriate handling and/or misuse of the product will invalidate the warranty. Leica Biosystems cannot assume liability for any such damage.

Serious incident reporting

The occurrence of any serious incident(s) that either has led to, or may lead to, death of a patient or user, or the temporary or permanent deterioration in the state of health of a patient or user must be reported to a local Leica Biosystems representative and the relevant local Regulatory Authority.

Statement for User Data Security and Privacy

Leica Biosystems respects, and is committed to protecting, personal data security and privacy. The Leica Biosystems Privacy Notice below describes the personal data that we may collect, use and retain.

Privacy Notice

The licensee shall comply with all applicable data protection and privacy laws in processing personal data using the BOND RX System, including BOND RX-ADVANCE, without limitation, by making all required notifications to, and obtaining all required consents from, patients and other data subjects prior to processing their personal data.

Contacting Leica Biosystems

For service or support contact your local Leica Biosystems representative or see www.LeicaBiosystems.com.

Revision record

Rev.	Issued	Sections Affected	Detail
A04	August 2024	General cautions 13.1 Handheld Barcode Scanners 12.9 ID Imager	Added Proposition 65 caution Added Newland barcode scanner, removed symbol barcode scanner Removed ID imager re-initializing Minor corrections
A03	Dec 2020	All	Minor corrections
A02	Nov 2020	Front cover	Update to manual title.
A01	Sep 2020	All	New version for BOND RX systems running BOND RX 7 software. Based on existing BOND RX 6.0 User manual 21.7733.504 A01.

General warnings

Warnings are notifications of hazards that could lead to personal injury, or where there is the possibility of losing, damaging, or misidentifying patient samples. Follow all safety precautions to avoid personal injury, damage, loss or misidentification of patient samples, and damage to equipment.

Warnings use symbols with a black border and yellow background.

General BOND RX warnings appear below. Other warnings appear in the relevant sections in the manual.

Processing module operation



To avoid contamination of reagents and slides, the processing module should be operated in a clean environment as free as possible from dust and particulate matter.



To ensure correct operation of the processing module, place each bulk reagent container into its correct station in the cavity, as indicated by the color-coded name labels. Failure to do so may compromise staining.

For further details, see [2.2.7 Bulk Containers Cavity](#)



Check bulk container levels and fill or empty, as appropriate, at the start of each day (more frequently if required – see [12.2.1 Checking Container Levels](#)). Failure to do so can result in staining runs being interrupted to remove containers, which can compromise staining.



For BOND RX^m, if a bulk container needs filling during processing always check the **Protocol status** screen and confirm that the container is not being used, or is not about to be used. Failure to do so may compromise slides being processed. Return the container immediately after filling – see [12.2.2.5 During Runs](#). To avoid this situation check bulk container levels in between each protocol – see [12.2.1 Checking Container Levels](#)).

BOND RX bulk containers do not need to be removed for filling – see [12.2.2.1 Refill Bulk Reagent – BOND RX](#). To avoid this situation check bulk container levels daily (more frequently if required – see [12.2.1 Checking Container Levels](#)).



BOND RX does not require network access to function and perform its intended use. To prevent malicious or unauthorized access, install BOND RX without any connection to your network / infrastructure.

If you would like network connection, the preferred method is to connect BOND RX to a firewalled Virtual Local Area Network (VLAN). Alternatively, you can implement and validate your own network security mechanisms in accordance with your standard operating procedures.

For more information, refer to the BOND Information Systems Guide.



A malware infection on a BOND RX controller could lead to unexpected behaviors in operation, including disabling processing modules. Please take care to ensure your USB storage devices are virus free before connecting them to the BOND RX controller. Further, Leica Biosystems does not pre-install an anti-virus solution; we recommend that you install your own enterprise anti-virus product.

For more information, refer to the BOND Information Systems Guide.

Controls



Adequate laboratory control measures **MUST** be established and maintained to ensure an appropriate staining result for each slide. Leica Biosystems strongly recommends placing appropriate control tissue on the same slides as test tissue.

Chemical Hazards



Some of the reagents used in immunohistochemistry and in situ hybridization are hazardous. Ensure you have received adequate training for this procedure before continuing:

- Wear latex or nitrile gloves, safety glasses, and other suitable protective clothing when handling reagents or cleaning the processing module.
- Handle and dispose of reagents and condensate in accordance with all procedures and government regulations that apply at the laboratory site.



Reagent containers can tip during transit, leaving reagent residue around the cap. Always wear approved eye protection, gloves and protective clothing when opening reagent containers.



Potentially hazardous reagents can collect around the slide staining assemblies and contaminate the slide trays. Always wear approved protective clothing and gloves when handling slide trays.



Some of the reagents used on BOND Processing Modules are flammable:

- Do not place a flame or ignition source near the processing modules.
- Ensure all bulk container caps are properly sealed after refilling or emptying.



The processing modules have heaters and heated surfaces that can be ignition dangers if flammable materials are placed in close proximity:

- Do not place flammable materials on or near heaters.
- Do not place flammable materials on any hot surfaces on the processing module.
- Ensure all bulk container caps are properly sealed after refilling or emptying.

Mechanical Hazards



Take care when closing the processing module lid, ensuring hands are kept clear to avoid injury.



During operation the main robot, the aspirating probe, syringe pumps and bulk fluid robots (BOND RX) can move without warning and with a speed that can cause injury.

- Do not attempt to open the processing module lid while a run is in progress.
- Do not attempt to by-pass the interlocks that prevent operation of the processing module when the lid is opened.
- Ensure syringe pump covers are in place during operation.



Avoid contact with slide staining assemblies and their surrounds. These may be hot and cause severe burns. Allow twenty minutes after the cessation of operation for the slide staining assemblies and their surrounds to cool.



Contact customer support to relocate the processing module over a large distance or to transport for repair or disposal. The processing module is heavy and is not designed to be moved by a single user.



Ensure that the syringe door is closed (BOND RX^m) or the syringe cover is fitted (BOND RX) during normal operation. If a syringe or a syringe fitting becomes loose, reagent under pressure can spray from the syringe.



Contact customer support immediately if the main robot and/or bulk fluid robots continue to operate for more than five seconds after the processing module lid has been opened.



Do not move the main robot arm while the processing module is switched on. The robot can become misaligned, resulting in poor staining.

If the robot has been moved: power down the processing module, wait 30 seconds and then reinitialize.



Always switch the processing module off when performing cleaning or maintenance tasks (except for automated cleaning tasks, such as cleaning the aspirating probe).



The BOND RX bulk fluid robots move along the slide staining assemblies to allow users access for cleaning. Only operators who have been warned of the potential hazards and have received adequate training should carry out this procedure.



The slide staining assemblies contain moving parts that can cause serious injury. Keep fingers clear of the slide staining assembly opening when the processing module is in operation.

Before attempting to manually unlock the slide staining assemblies: turn the processing module power switch off, turn the mains power off, and disconnect the mains power supply plug at the wall.



The syringe pump module (BOND RX) is heavy and can fall forward when released. Only operators who have been warned of the potential hazards and have received adequate training should carry out this procedure.



Do not use the two black handles on the back cover of the BOND RX to lift the processing module.

Electrical hazards



Do not remove the processing module covers or attempt to access internal components. Dangerous voltages are present inside the BOND Processing Module and only qualified service technicians approved by Leica Biosystems should perform these tasks.



Do not change the processing module's operating voltage. Severe damage can occur if the processing module is connected to an incorrect power supply voltage. Contact customer support to have the setting changed.



The processing module must be connected to an earthed mains power outlet socket, and be positioned so that personnel are easily able to disconnect the mains power cable without having to move the processing module.



Do not bypass or short-circuit fuses.

Turn off the processing module and disconnect the power cord before changing fuses. Replace fuses only with standard parts and if fuses blow repeatedly, contact customer support.

General cautions

Cautions are notifications of hazards that could lead to damage to the BOND RX equipment or other adverse consequences that do not endanger people.

Cautions use symbols with a black border and white background.

General BOND RX cautions appear below. Other cautions appear in relevant sections in the manual.

Installation hazards



Do not block the ventilation openings located on the back cover of the processing module. Also, do not cover the ventilation openings located on the syringe door (BOND RX^m).

Operational hazards



Position all parts of the slide label within all slide edges. An exposed sticky surface can cause the slide label (and slide) to stick to the Covertile or other equipment and damage the slide.



Do not remove the small liquid level sensor cap from a bulk container (BOND RX™) as it can be damaged. Empty and refill bulk containers through the large fill/empty cap only.



Clean all removable components by hand only. To avoid damage, do not wash any component in an automatic dishwashing machine. Do not clean any part with solvents, harsh or abrasive cleaning fluids, or harsh or abrasive cloths.



Do not use Q-tips or other cotton tipped applicators to clean inside the wash block holes or the slide staining assembly wicking posts, as the cotton tip can come off and cause a blockage.



Do not force bulk containers back into position, as this can damage the container and liquid sensor.



Do not use damaged slides. Ensure all slides are correctly aligned on the slide trays, and that all Covertiles are correctly positioned (see [2.6.2 BOND Universal Covertiles](#)), before loading into the processing module.



Ensure the syringe module (BOND RX) is fully closed before starting a run or initializing the processing module (see [12.4.1 Manually Unlocking Slide Staining Assemblies](#)). Failure to do so can result in damage to the syringes during operation.



Ensure the bulk fluid robots (BOND RX) are in the home position at the rear of the processing module, and not positioned along the slide staining assemblies before cleaning or removing the top plate.



This product can expose you to chemicals including lead, which are known to the State of California to cause cancer, birth defects, or other reproductive harm. For more information, go to <https://www.P65Warnings.ca.gov>.

Reagent hazards



Unsatisfactory staining results and potential damage to the processing module can occur if incompatible solutions are allowed to come in contact with each other. Contact Leica Biosystems to determine whether the solutions are compatible.



Do not use xylene, chloroform, acetone, strong acids (e.g. 20% HCl), strong alkalis (e.g. 20% NaOH) on BOND Processing Modules. If any of these chemicals spill on or near a BOND Processing Module, clean the spill immediately with 70% alcohol to prevent damage to the processing module covers.



Use only BOND Dewax Solution on BOND instruments. Do not use xylene, xylene substitutes and other reagents that can degrade parts of the BOND RX system and cause fluid leakage.

Regulatory notices

Intended use



The BOND RX System provides the ability to automatically stain slides according to specific protocols in a research laboratory. The product is for research use only and not for use in diagnostic procedures. The product is to be operated by a trained laboratory technician or scientist within the research laboratory.

FCC compliance

This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 subpart B of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at his own expense.

To maintain compliance use only the cables supplied with the instrument.



WARNING: Any changes or modifications not expressly approved by Leica Biosystems could void the user's authority to operate this equipment.

CE marking



The CE mark on the equipment indicates compliance with the Electromagnetic Compatibility Directive (2014/30/EU), Low Voltage Directive (2014/35/EU) and Restriction on the Use of Certain Hazardous Substances in Electrical and Electronic Equipment (2011/65/EU).

Instructions for equipment for research use only

The electromagnetic environment should be evaluated prior to operation of the device.

Do not use this device in close proximity to sources of strong electromagnetic radiation (e.g. unshielded intentional RF sources) and/or magnetic fields, as these can interfere with the proper operation.

Classification of equipment under CISPR 11 (EN 55011)

This equipment is classified as Group 1 Class A under CISPR 11 (EN 55011). The explanation for group and class is described below.

Group 1 - This is applicable for all equipment which is not classified as group 2 equipment.

Group 2 - This is applicable for all ISM RF equipment in which radio - frequency energy in the frequency range 9 kHz to 400 GHz is intentionally generated and used or only used, in the form electromagnetic radiation, inductive and/ or capacitive coupling, for the treatment of material or inspection/ analysis purposes.

Class A - This is applicable for all equipment suitable for use in all establishments other than domestic and those directly connected to a low voltage power supply network which supplies buildings used for domestic purposes.

Class B - This is applicable for all equipment suitable for use in domestic establishments and in establishments directly connected to a low voltage power supply network which supplies buildings used for domestic purposes.

Definitions

ISM: Industrial, Scientific and Medical

RF: Radio Frequency

Glossary of symbols

This section describes the regulatory and safety symbols used in the product labeling.

Regulatory symbols

Explanation of the regulatory symbols used for BOND RX.











This glossary provides images of the symbols as presented in the relevant standards, however, some of the symbols may vary in color.

The following is a list of symbols used on the product labeling and their meaning.

ISO 15223-1


Medical devices – symbols to be used with medical device labels, labeling and information to be supplied – Part 1: General requirements.




Symbol	Standard/ Regulation	Reference	Description
	ISO 15223-1	5.1.1	Manufacturer Indicates the medical device manufacturer.
	ISO 15223-1	5.1.2	Authorized representative in the European community Indicates the Authorized representative in the European Community.
	ISO 15223-1	5.1.3	Date of manufacture Indicates the date when the medical device was manufactured.
	ISO 15223-1	5.1.4	Use by (expiration date) Indicates the date after which the medical device is not to be used.
	ISO 15223-1	5.1.5	Batch code Indicates the manufacturer's batch code so that the batch or lot can be identified.
	ISO 15223-1	5.1.6	Catalog number / Reference number Indicates the manufacturer's catalog number so that the medical device can be identified.
	ISO 15223-1	5.1.7	Serial number Indicates the manufacturer's serial number so that a specific medical device can be identified.

Symbol	Standard/ Regulation	Reference	Description
	ISO 15223-1	5.1.8	Importer Indicates the entity importing the medical device into the European Union.
	ISO 15223-1	5.1.9	Distributor Indicates the entity distributing the medical device into the locale.
	ISO 15223-1	5.3.1	Fragile: handle with care Indicates a medical device that can be broken or damaged if not handled carefully.
	ISO 15223-1	5.3.4	Keep away from rain Indicates that the transport package shall be kept away from rain and in dry conditions.
	ISO 15223-1	5.3.7	Temperature limit Indicates the temperature limits to which the medical device can be safely exposed.
	ISO 15223-1	5.4.2	Do not re-use Indicates a medical device that is intended for one use, or for use on a single patient during a single procedure.
	ISO 15223-1	5.4.3	Consult instructions for use Indicates the need for the user to consult the instructions for use.
	ISO 15223-1	5.4.4	Caution Indicates the need for the user to consult the instructions for use for important cautionary information such as warnings and precautions that cannot, for a variety of reasons, be presented on the medical device itself.

ISO 7000


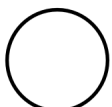
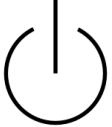
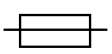


Graphical symbols for use on equipment – Registered symbols.




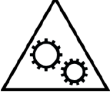

Symbol	Standard/ Regulation	Reference	Description
	ISO 7000	1135	Recycle Indicates that the marked item or its material is part of a recovery or recycling process.

Symbol	Standard/ Regulation	Reference	Description
	ISO 7000	1640	Technical manual: manual for service Identifies the location where the handbook is stored or to identify information that relates to the servicing instructions for the equipment. To indicate that the service manual or handbook should be considered when servicing the device close to where the symbol is placed.
	ISO 7000	2594	Ventilation open Identifies the control that allows outside air into interior environment.
	ISO 7000	3650	USB Identifies a port or plug as meeting the generic requirements of the Universal Serial Bus (USB). To indicate that the device is plugged into a USB port or is compatible with a USB port.



IEC 60417


Graphical symbols for use on equipment.




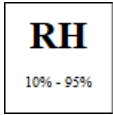

Symbol	Standard/ Regulation	Reference	Description
	IEC 60417	5007	On Indicates connection to the mains, at least for mains switches or their positions, and all those cases where safety is involved
	IEC 60417	5008	Off Indicates disconnection from the mains, at least for mains switches or their positions, and all those cases where safety is involved
	IEC 60417	5009	Stand-by Identifies the switch or switch position by means of which part of the equipment is switched on in order to bring it into the standby condition
	IEC 60417	5016	Fuse Identifies fuse boxes or their location.
	IEC 60417	5019	Protective earth: protective ground A terminal that is intended for connection to an external conductor for protection against electric shock in case of a fault, or the terminal of a protective earth (ground) electrode.
	IEC 60417	5032	Single phase alternating current Indicates on the rating plate that the equipment is suitable for alternating current only; to identify relevant terminals.

Symbol	Standard/ Regulation	Reference	Description
	IEC 60417	5134	Electrostatic Sensitive Devices Packages containing electrostatic sensitive devices, or a device or a connector that has not been tested for immunity to electrostatic discharge.
	IEC 60417	5988	Computer network Identifies the computer network itself or to indicate the connecting terminals of the computer network.
	IEC 60417	6040	Warning: Ultraviolet radiation Alert for the presence of UV light within the product's enclosure that may be of sufficient magnitude to constitute a risk to the operator. Turn off the UV lamp before opening. Use UV radiation eye and skin protection during servicing.
	IEC 60417	6057	Caution: moving parts An instructional safeguard to keep away from moving parts.
	IEC 60417	6222	Information: general Identifies the control to examine the status of the equipment, e.g. multifunctional copying machines

Other symbols and markings

Symbol	Standard/ Regulation	Description
	N/A	Research Use Only This product is intended for research use only and not for use in diagnostic procedures.
	The instrument Declaration of Conformity lists the Directives with which the system complies	European Conformity The instrument Declaration of Conformity lists the Directives with which the system complies.

Symbol	Standard/ Regulation	Description
	Directive 2012/19/EC EU: waste electrical and electronic equipment (WEEE)	<p>Waste Electrical and Electronic Equipment (WEEE) Directive The electronic product should not be discarded as unsorted waste but must be sent to separate collection facilities for recovery and recycling.</p> <p>The presence of this label indicates that:</p> <ul style="list-style-type: none"> • The device was put on the European Market after August 13, 2005. • The device is not to be disposed of via the municipal waste collection system of any member state of the European Union. <p>Customers must understand and follow all laws regarding the correct decontamination and safe disposal of electrical equipment.</p>
	AS/NZS 4417.1	<p>Regulatory Compliance Mark (RCM) Indicates compliance with the Australian Communications Media Authority (ACMA) requirements (safety and EMC) for Australia and New Zealand.</p>
	People's Republic of China Electronic Industry Standard SJ/T11364	<p>Restriction of Hazardous Substances (RoHS 2) Indicates that this electronic information product contains certain toxic or hazardous elements, and can be used safely during its environmental protection use period. The number in the middle of the logo indicates the environmental protection use period (in years) for the product. The outer circle indicates that the product can be recycled. The logo also signifies that the product should be recycled immediately after its environmental protection use period has expired. The date on the label indicates the date of manufacture.</p>
	People's Republic of China Electronic Industry Standard SJ/T11364	<p>Restriction of Hazardous Substances (RoHS 2) Indicates that this electronic information product does not contain any hazardous substances, or they do not exceed the concentration limits specified in GB/T 26572. It is a green environmentally friendly product that can be recycled.</p>
	Title 47 United States Code of Federal Regulations Part 15	<p>Federal Communications Commission (FCC) This product has been tested and found to comply with the limits pursuant to part 15 of the FCC Rules.</p>
	N/A	<p>Underwriters Laboratory (UL) certification mark Underwriter Laboratories have certified that the listed products comply with both US and Canadian safety requirements.</p>




Symbol	Standard/ Regulation	Description
	CSA International	Listed device with CSA Group testing agency CSA Group have certified that the listed products comply with both US and Canadian safety requirements.
	N/A	Listed device with Intertek testing agency Intertek Testing Agency have certified that the listed products comply with both US and Canadian safety requirements.
	Ordinance on In Vitro Diagnostic Medical Devices (IVDO) of 4 May 2022.	Swiss Authorised Representative Indicates the Swiss Authorised representative.
	N/A	Relative humidity range Indicate the acceptable upper and lower limits of relative humidity for transport and storage. This symbol is accompanied by the applicable relative humidity limits.
	N/A	Unconnected port This product has an unconnected port on the syringe pump.

Safety symbols

Explanation of the safety symbols used for BOND RX.

ISO 7010

Graphical symbols – Safety colors and safety signs – Registered safety signs.

Symbol	Standard/ Regulation	Reference	Description
	ISO 7010	W001	General warning Indicates the need for the user to consult the instructions for use for important cautionary information such as warnings and precautions that cannot, for a variety of reasons, be presented on the medical device itself.
	ISO 7010	W004	Warning: laser beam Laser hazard. Potential for severe eye damage. Avoid direct eye contact with laser beams.
	ISO 7010	W009	Warning: biological hazard Biological hazard. Potential exposure to a biological hazard. Follow directions in the accompanying documentation to avoid exposure.










Symbol	Standard/ Regulation	Reference	Description
	ISO 7010	W012	Caution: risk of electric shock Electrical hazard. Potential risk of electric shock. Follow directions in the accompanying documentation to avoid damage to persons or equipment.
	ISO 7010	W016	Warning: toxic material Toxic hazard. Potential danger of severe health impacts if proper chemical handling procedures are not followed. Use gloves and protective eye wear when handling reagents.
	ISO 7010	W017	Warning: hot surface Heat hazard. Hot surfaces will cause burns if touched. Avoid touching parts identified with this symbol.
	ISO 7010	W020	Warning: Overhead obstacle Overhead obstacle. Take care to avoid being struck by or walking into an overhead obstacle.
	ISO 7010	W021	Warning: flammable material Flammable hazard. Flammable materials may ignite if proper precautions are not followed.
	ISO 7010	W022	Warning: Sharp element Sharp element. Take care to avoid injury from sharp elements (e.g. needles, blades).
	ISO 7010	W023	Warning: corrosive substance Chemical hazard from a corrosive substance. There is a danger of severe health impacts if proper precautions are not followed. Always wear protective clothing and gloves. Immediately clean up spills using standard laboratory practice.
	ISO 7010	W024	Warning: crushing of hands Crush hazard. Hands or body parts can be crushed by a closing motion of mechanical parts of equipment.
	ISO 7010	W072	Warning: Environmental hazard Environmental hazard. Substance or mixture that can cause an environmental hazard.

Table of contents

1	Introduction	27
1.1	System Overview	27
1.2	Research Use Only	28
1.3	Legacy Research Systems	28
1.4	Getting Help	29
1.5	First Steps	30
1.6	Running a Protocol – Workflows	31
1.6.1	BOND RX and BOND RX ^m	31
2	Hardware	33
2.1	The BOND RX System	34
2.1.1	BOND Ancillary Products	34
2.2	BOND RX and BOND RX ^m Processing Modules	35
2.2.1	Main Components	35
2.2.2	Processing Module Initialization	41
2.2.3	Lid	41
2.2.4	Main Robot and ID Imager	42
2.2.5	Slide Staining Assemblies	43
2.2.6	Front Cover	44
2.2.7	Bulk Containers Cavity	47
2.2.8	Aspirating Probe	52
2.2.9	Wash Block and Mixing Station	53
2.2.10	Bulk Fluid Robots (BOND RX only)	54
2.2.11	Syringes	55
2.2.12	Power Switch	57
2.2.13	Back Cover	58
2.3	BOND RX Controller and Terminals	60
2.4	Handheld Barcode Scanner	61
2.4.1	Using the Handheld Barcode Scanner	61
2.5	Slide Labeler	62
2.6	Ancillary Equipment	63
2.6.1	Slides	63
2.6.2	BOND Universal Covertiles	64

2.6.3	Slide Trays	65
2.6.4	Reagent Trays	65
2.6.5	Reagent Systems and Containers	66
2.7	Relocating a Processing Module	67
2.8	Instrument Decommissioning and Disposal	67
3	Software Overview (on BOND RX Controller)	68
3.1	System Architecture	69
3.1.1	Single-Seat Configuration	69
3.1.2	BOND RX-ADVANCE	70
3.2	Starting and Shutting Down the BOND RX Software	72
3.2.1	Starting the BOND RX software	72
3.2.2	Shut down the BOND RX software	74
3.3	User Roles	74
3.4	Research Client Interface Overview	75
3.4.1	Function Bar	75
3.4.2	Processing Module Tabs	76
3.4.3	Sorting Tables	77
3.4.4	Date Format	77
3.5	BOND RX-ADVANCE Dashboard	78
3.5.1	Slide Staining Assembly Status	79
3.6	Notifications, Warnings, and Alarms	80
3.7	Reports	81
3.8	Help	81
3.9	About BOND RX	82
3.9.1	Service Log	83
3.10	BOND RX Data Definitions	84
3.10.1	Data Definitions Updates	84
3.11	Software Updates	84
4	Quick Start	85
4.1	BOND RX and BOND RX ^m	85
4.1.1	Preliminary Checks and Startup	86
4.1.2	Protocol and Reagent Checks	86
4.1.3	Setting Up Slides	87

4.1.4	Loading the Reagents	92
4.1.5	Running the Protocol	95
4.1.6	Finishing	96
5	Status Screens (on BOND RX Controller)	97
5.1	System Status Screen	98
5.1.1	Processing Module Tabs	99
5.1.2	Hardware Status	100
5.1.3	Reagent Status	102
5.1.4	Slide Information	111
5.1.5	On-Board Slide Identification	113
5.1.6	Run Progress Indicator	116
5.1.7	Starting or Stopping a Run	120
5.1.8	Delayed Start	122
5.2	Protocol Status Screen	123
5.3	Maintenance Screen	124
5.3.1	Maintenance Report	125
6	Slide Setup (on BOND RX Controller)	127
6.1	Slide Setup Screen	128
6.2	Working with Controls	129
6.2.1	Control Tissue	129
6.2.2	Control Reagent	129
6.3	Working with Studies	130
6.3.1	Study Controls and Active Study Information	130
6.3.2	Study Identification	131
6.3.3	Adding a Study	132
6.3.4	Study Duplication, Resurrection and Expiration	133
6.3.5	Editing a Study	134
6.3.6	Copying a Study	134
6.3.7	Daily Study Option	135
6.3.8	Study Report	135
6.4	Manage Researchers	136
6.5	Working with Slides	136
6.5.1	Description of Slide Fields and Controls	137

6.5.2	Creating a Slide	138
6.5.3	Copying a Slide	140
6.5.4	Editing a Slide	141
6.5.5	Deleting a Slide	141
6.5.6	Manually Identifying a Slide	141
6.5.7	Adding a Panel of Slides	142
6.5.8	Dispense Volumes and Tissue Position on Slides	142
6.6	Slide Labeling	143
6.6.1	Printing Labels and Applying to Slides	144
6.6.2	Slide IDs and Label IDs	146
6.7	Slide Setup Summary Report	146
6.8	Impromptu Slide and Study Creation	147
6.8.1	Creating New Studies and/or Slides After Imaging	148
6.8.2	On-Board Slide Identification Options	150
6.9	Slide Compatibility	151
6.9.1	Protocol Compatibility	152
7	Protocols (on BOND RX Controller)	154
7.1	Protocol Types	155
7.1.1	Staining Modes	155
7.1.2	Protocol Sequences	157
7.2	Protocol Setup Screen	159
7.2.1	Protocol Details	160
7.3	Creating New Protocols	162
7.4	Editing User Protocols	163
7.4.1	Editing Protocol Steps	163
7.4.2	Adding and Removing Protocol Steps	164
7.4.3	Protocol Rules	168
7.4.4	Multiple Processing Module Types and Protocol Versions	170
7.4.5	Deleting Protocols	171
7.5	Protocol Reports	171
7.6	Predefined Protocols	172
7.6.1	Staining Protocols	172
7.6.2	Template Protocols	174
7.6.3	Prestaining Protocols	174

8	Reagent Management (on BOND RX Controller)	176
8.1	Reagent Management Overview	177
8.1.1	General Information	178
8.2	Reagent Setup Screen	180
8.2.1	Adding or Editing a Reagent	182
8.2.2	Deleting a Reagent	184
8.3	Reagent Inventory Screen	184
8.3.1	Determining Reagent Volume	186
8.3.2	Reagent or Reagent System Details	187
8.3.3	Registering Reagents and Reagent Systems	189
8.3.4	Inventory Details Report	193
8.3.5	Reagent Usage Report	194
8.4	Reagents for Research	195
8.4.1	Research Reagent Systems	195
8.4.2	Refilling Research Reagent Systems	199
8.4.3	Mixed Reagents with Research Reagent Systems	200
8.5	Reagent Panels Screen	201
8.5.1	Creating a Panel	202
8.5.2	Viewing or Editing Panel Details	203
8.5.3	Removing a Panel	203
9	Slide History (on BOND RX Controller)	204
9.1	Slide History Screen	205
9.2	Slide Selection	206
9.3	Slide Properties and Slide Rerun	207
9.3.1	Rerunning Slides	207
9.4	Run Events Report	208
9.5	Run Details Report	208
9.6	Study Report	210
9.7	Protocol Report	211
9.8	Slides Summary	211
9.9	Export Data	212
9.10	Brief Slide History	214

10 Administration Client (on BOND RX Controller)	215
10.1 Users	216
10.2 LIS	218
10.3 Labels	220
10.3.1 Create, Edit, and Activate Label Templates	223
10.3.2 Information Types	224
10.4 BXD	228
10.4.1 BXD Updates	229
10.4.2 Audit Trail	229
10.5 Settings	230
10.5.1 Laboratory Settings	231
10.5.2 Study and Slide Settings	232
10.5.3 Database Backups	233
10.6 Hardware	234
10.6.1 Processing Modules	234
10.6.2 Pods	237
10.6.3 Slide Labelers	239
11 LIS Integration Package (on BOND RX Controller)	252
11.1 LIS Terminology	253
11.2 Additional Software Features	253
11.2.1 LIS Status Icon	254
11.2.2 LIS Studies	254
11.2.3 LIS Slides	255
11.2.4 Public Marker Names	255
11.2.5 Priority Slides	256
11.2.6 LIS Slide Data Fields	256
11.3 LIS Connection and Initialization	257
11.4 LIS Notifications	257
11.5 Study and Slide Data Requirements	258
11.5.1 Study Data	259
11.5.2 Slide Data	259
11.6 Sending Slide Data back to the LIS	260
11.7 Slide Labels	260
11.8 Workflows	261

12	Cleaning and Maintenance (BOND RX and BOND RX^m)	262
12.1	Cleaning and Maintenance Schedule	264
12.1.1	Cleaning and Maintenance Checklists	265
12.2	Bulk Containers	267
12.2.1	Checking Container Levels	267
12.2.2	Replenishing or Emptying Bulk Containers	268
12.2.3	Cleaning Bulk Containers	272
12.2.4	External Waste Container (BOND RX ^m only)	273
12.3	Covertiles	275
12.3.1	Remove DAB Residue (Optional)	275
12.3.2	Standard Cleaning (Mandatory)	275
12.4	Slide Staining Assembly	275
12.4.1	Manually Unlocking Slide Staining Assemblies	278
12.5	Restart Processing Module	281
12.6	Aspirating Probe	282
12.6.1	Cleaning the Aspirating Probe	282
12.6.2	Running an Aspirating Probe Clean	283
12.7	Wash Block and Mixing Station	283
12.8	Covers, Doors and Lid	284
12.9	ID Imager	285
12.10	Drip Trays	285
12.10.1	BOND RX Bulk Container Drip Trays	285
12.10.2	BOND RX Processing Module Drip Tray	287
12.10.3	BOND RX ^m Bulk Container Drip Tray	287
12.11	Slide Trays	288
12.12	Bulk Fluid Robot Probes (BOND RX only)	288
12.12.1	Cleaning the Bulk Fluid Robot Probes	288
12.13	Syringes	289
12.13.1	Replacing BOND RX Syringes	290
12.13.2	Replacing BOND RX ^m 9-Port Syringe	291
12.14	Power Supply Fuses	292
13	Cleaning and Maintenance (Miscellaneous)	294
13.1	Handheld Barcode Scanners	294
13.1.1	Honeywell Barcode Scanner	294

13.1.2	Zebra DS2208 Barcode Scanner	297
13.1.3	Newland NLS-HR2000 Barcode Scanner	300
13.2	Slide Labeler	304
14	Using BOND Reagents	305
14.1	Principle of the Procedure	305
14.1.1	BOND Detection Systems	306
14.2	Specimen Preparation	308
14.2.1	Materials Required	308
14.2.2	Tissue Preparation	309
14.2.3	Dewaxing and Baking	310
14.2.4	Epitope Retrieval	310
14.3	Quality Control	311
14.3.1	Assay Verification	311
14.3.2	Tissue Controls	312
14.3.3	Negative Reagent Control for IHC	312
14.3.4	Reagent Controls for ISH	313
14.3.5	The Benefits of Quality Control	314
14.4	Interpretation of Staining	314
14.4.1	Positive Tissue Control	315
14.4.2	Negative Tissue Control	315
14.4.3	Test Tissue	315
14.5	General Limitations	315
14.6	References	317
15	System Management (on BOND RX Controller)	318
15.1	BOND System Manager	318
15.1.1	Overview	318
15.1.2	BOND System Manager Window	319
15.1.3	Stopping Services	320
15.1.4	Starting Services	320
15.2	Hard Disk Redundancy	322
16	BOND RX-ADVANCE Operations	323
16.1	Restarting the BOND RX-ADVANCE System	323
16.2	Switching to the Secondary Controller	324

17	Slide Label Printer Replacement	329
17.1	Replace a Cognitive Cxi Printer on a Single Seat System	329
17.2	Replace a Cognitive Cxi Printer on BOND RX-ADVANCE System	330
17.3	Replace a Zebra Printer with a Cognitive Cxi Printer on a Single-seat System	334
18	Specifications	335
18.1	System Specifications	335
18.2	Physical Specifications	336
18.3	Electrical Power and UPS Requirements	336
18.4	Environmental Specifications	336
18.5	Operating Specifications	337
18.6	Microscope Slides	338
18.7	Transport and Storage	338
	Index	339

1

Introduction

1.1 System Overview

The Leica Biosystems BOND RX and BOND RX^m research systems (referred to as the BOND RX system within and includes the BOND RX and BOND RX^m) are designed to balance the need of translational researchers for customization and standardization. Researchers commonly experiment with a variety of detection techniques on the BOND RX system, including but not limited to: IHC, IF, ISH, mRNA ISH, FISH, TUNEL, etc.

BOND RX and BOND RX^m systems are built specifically to extend the functionality of the clinical BOND-III and BOND-MAX respectively. The BOND RX system is commonly used in pharmaceutical companies, contract research organizations, and academic medical centers. The customization flexibility of the BOND RX system is enabled through “Research” detection systems that allow users to build the detection chemistry of their choice.

There are two processing module (PM) types:

- BOND RX and BOND RX^m – each with a 30-slide capacity. Three runs of up to ten slides each can be processed simultaneously, using different staining protocols if required, with each run started separately to provide continuous processing. One or more of the runs may be set up for multiplex staining, while another may be processing a DAB or Red single stain.

The BOND RX software makes setting up and staining slides easy. Use rigorously tested protocols supplied with the system, or create your own. Select from a wide range of BOND ready-to-use reagents, or use any other antibodies or probes, pairing these with a range of high quality BOND detection systems. After you create your virtual slides in the software – or import them from a Laboratory Information System (LIS) – print the labels (or use the LIS-printed labels), attach them to the slides, and then load the slides onto the processing module. The BOND RX system does the rest, consistently and reliably producing high-quality stains.



Protocols and reagent products supplied by Leica Biosystems will be displayed in the software as being provided by Leica Microsystems.

BOND RX system features include:

- High throughput
- Flexibility
- Safety
- Automated IHC staining and counterstaining
- Automated ISH staining and counterstaining
- Automated dewaxing, baking, and retrieval
- Automated multiplex staining

We trust that the Leica Biosystems BOND RX and/or BOND RX™ will be an effective partner for you wherever your research takes you, allowing you to develop your scientific insights on a platform that is sensitive, consistent, reliable and efficient.

See sections:

- [1.2 Research Use Only](#)
- [1.3 Legacy Research Systems](#)
- [1.4 Getting Help](#)
- [1.5 First Steps](#)
- [1.6 Running a Protocol – Workflows](#)

1.2 Research Use Only

BOND RX processing modules, of any type, must be used for research purposes only. Under no circumstances must any part of the BOND RX system be used for any diagnostic purpose.

All functionality in the BOND RX software, including that which is also available in standard BOND IVD systems, is equally rendered suitable for research use only.

1.3 Legacy Research Systems

The BOND RX system supersedes the BOND Research system, commonly referred to as the BOND Research Dongle. The BOND RX system is incompatible with this legacy research system; the legacy research system cannot be connected to the BOND RX Controller. Instructions in this manual that refer to the BOND RX processing module apply equally to BOND-III processing modules, as it pertains to hardware and core technology. Instructions in this manual that refer to the BOND RX™ processing module apply equally to the BOND-MAX processing modules, as it pertains to hardware and core technology.

1.4 Getting Help

The BOND RX user manual (this manual) is installed in PDF format on all controllers (single-seat) and terminals (BOND RX-ADVANCE). It is also on a USB supplied with the system.

You can view this user manual by clicking the **Help** icon  on the function bar in both BOND RX software clients, or alternatively opening it from the desktop icon.

For problems with the BOND RX system, contact your local Leica Biosystems representative or see www.leicabiosystems.com.

1.5 First Steps

For users new to the BOND RX system, this section describes where to find information in the user manual in order to get a full working knowledge of the product.

Step	Description	Manual Section
1	Installation and Commissioning Hardware set up, software installed, system checked. Performed by representatives of Leica Biosystems or authorized distributor.	–
2	Read the Safety Section Become familiar with the safety requirements for the BOND RX system.	General warnings and General cautions
3	Know Your Hardware Become familiar with the names and uses of the BOND RX hardware.	2 Hardware
4	Know Your Software Gain a general understanding of the software and how to use it.	3 Software Overview (on BOND RX Controller)
5	Check Protocols and Reagents Reagents and protocols may have been set up during installation: <ul style="list-style-type: none"> • Check that the protocols you want to run have been set up. • Check that reagents required at your site have been set up. 	7 Protocols (on BOND RX Controller) 8 Reagent Management (on BOND RX Controller)
6	Running a Protocol For a very brief overview. For a more detailed overview.	1.6 Running a Protocol – Workflows 4 Quick Start
7	Advanced As required, gain a more in-depth understanding of the software.	5 Status Screens (on BOND RX Controller) to 9 Slide History (on BOND RX Controller)
8	Working with an LIS An optional package allows connection to a laboratory information system.	11 LIS Integration Package (on BOND RX Controller)
9	Looking After Your BOND RX System	12 Cleaning and Maintenance (BOND RX and BOND RX ^m)

1.6 Running a Protocol – Workflows

1.6.1 BOND RX and BOND RX^m



WARNING: To avoid contamination of reagents and slides, the processing module should be operated in a clean environment as free as possible from dust and particulate matter.

The following is an overview of the standard steps involved in staining a tray of slides. With different option settings other workflows are possible.

1.6.1.1 Initial Checks and Startup

- 1 Ensure that the processing module is clean and that all maintenance tasks are up to date ([12.1 Cleaning and Maintenance Schedule](#)). Daily prerun tasks are:
 - a Check bulk waste containers are no more than half full.
 - b Check bulk reagent containers. Refill if required.
- 2 Check wash blocks and mixing station – clean or replace if necessary.
- 3 Check that the slide labeler has labels and print ribbon, and is turned on.
- 4 Turn on the processing module, controller (and terminal for BOND RX-ADVANCE) and open the BOND RX research client.

1.6.1.2 Configure Reagents

- 1 Create reagents in the system if required ([8.2.1 Adding or Editing a Reagent](#)).
- 2 Register reagent containers ([8.3.3 Registering Reagents and Reagent Systems](#)).


1.6.1.3 Configure Protocols

- 1 Create new protocols if required ([7.3 Creating New Protocols](#)).

1.6.1.4 Configure Slides

- 1 Create studies in the software ([6.3.3 Adding a Study](#)).
- 2 Create slides in the software ([6.5.2 Creating a Slide](#)).
- 3 Print slide labels and apply to the slides ([6.6.1 Printing Labels and Applying to Slides](#)).
- 4 Place slides and Covertiles on slide trays ([4.1.3 Setting Up Slides](#)).

1.6.1.5 Load the Processing Module and Start Run

- 1 Insert the slide trays into the processing module (4.1.3.5 Loading Slides).
- 2 Load the detection system and reagent trays into the processing module (4.1.4 Loading the Reagents).
- 3 Press the Load/Unload buttons on the processing module to lock the slide trays.
- 4 On the **System status** screen check that all slides have been identified – manually identify slides that were not automatically identified (5.1.5.2 On-Board Manual Slide Identification).
- 5 View and rectify any alert indications on the **System status** screen.
- 6 Click the  button to start the run.

1.6.1.6 Monitor Run

- 1 Monitor run progress on the **System status** screen (5.1 System Status Screen) or BOND dashboard (3.5 BOND RX-ADVANCE Dashboard). View and rectify any notifications.

1.6.1.7 Unload Slides and Reagents

- 1 When the run is finished, remove the detection system and reagent trays and store the reagents (4.1.6 Finishing).



When a processing module is not being used, remove the ER1 and ER2 bulk containers and store at +2 to +8 °C (+36 to +46 °F). Also see 2.2.7 Bulk Containers Cavity.

- 2 Press the Load/Unload buttons on the processing module to unlock the slide trays, and remove the trays.
- 3 Remove Covertiles and clean (12.3 Covertiles).



Do not leave slides sitting in the trays while cleaning the Covertiles.

- 4 Remove the slides.
- 5 Clean any spills or marks on the slide staining assemblies (12.4 Slide Staining Assembly), on other parts of the processing module, or on slide or reagent trays.

1.6.1.8 Hydration on the BOND RX^m and BOND RX System

On completion of the staining process, slides will be hydrated until you remove them. On BOND RX^m and BOND RX, the slides within the Slide Tray will be periodically hydrated with the specified hydration fluid until the Slide Trays are raised. Make sure you remove trays promptly from the processing module after raising the Slide Tray.

2 Hardware

This section is designed to tell you:

- Names of the pieces of equipment in the BOND RX system
- Functions of these items, and how they relate to the system as a whole
- Where to find further information, for example, operational procedures and maintenance procedures related to the equipment.

Details of how to set up and connect components are not included with the hardware descriptions, as the system should be set up and tested for you. If you need to replace or re-connect components, details are included in [12 Cleaning and Maintenance \(BOND RX and BOND RX^m\)](#).

Where appropriate, information about the BOND RX and BOND RX^m Processing Modules is divided into separate sections to find relevant information faster.

See sections:

- [2.1 The BOND RX System](#)
- [2.2 BOND RX and BOND RX^m Processing Modules](#)
- [2.3 BOND RX Controller and Terminals](#)
- [2.4 Handheld Barcode Scanner](#)
- [2.5 Slide Labeler](#)
- [2.6 Ancillary Equipment](#)
- [2.7 Relocating a Processing Module](#)
- [2.8 Instrument Decommissioning and Disposal](#)

2.1 The BOND RX System

The BOND RX system consists of the following major components:

- One or more processing modules (see [2.2 BOND RX and BOND RX^m Processing Modules](#))
- A BOND RX controller or a BOND RX-ADVANCE controller (see [2.3 BOND RX Controller and Terminals](#))
BOND RX-ADVANCE installations have terminals as well as the controller, and may include a secondary (backup) controller
- One or more handheld barcode scanners (see [2.4 Handheld Barcode Scanner](#))
- One or more slide label printers (see [2.5 Slide Labeler](#))

Each new BOND RX or BOND RX^m Processing Module is supplied with:

- 4 slide trays (see [2.6.3 Slide Trays](#))
- 4 reagent trays (see [2.6.4 Reagent Trays](#))
- 1 mixing station (see [2.2.9 Wash Block and Mixing Station](#))
- 1 hex key for syringe pump replacement
- 1 Ethernet cable

For BOND RX or BOND RX^m Processing Modules, you will also need:

- Covertiles (see [2.6.2 BOND Universal Covertiles](#))
- BOND detection systems, BOND research reagent systems and BOND ready-to-use reagents or concentrates and/or open reagent containers (see [2.6.5 Reagent Systems and Containers](#))

Refer to www.leicabiosystems.com for a complete and up-to-date list of consumable items and spare parts.

See also [3.1 System Architecture](#).

2.1.1 BOND Ancillary Products

BOND ancillary products are designed specifically for the BOND RX system and their use helps ensure optimal staining results. Using BOND ancillary products also helps to maintain the processing module in top condition and prevent damage.



The following products should *always* be used on the BOND RX system, and *never* substituted with other products:

Ancillary Reagents

- BOND Wash Solution
- BOND Epitope Retrieval Solution (1 & 2)
- BOND Dewax Solution

BOND RX or BOND RX^m Consumables

- BOND Research Reagent Systems
- BOND Plus slides and Apex BOND slides (or glass slides conforming to the specifications listed in [2.6.1 Slides](#))
- BOND Universal Covertiles
- BOND Open Containers (7 mL and 30 mL)
- BOND Titration Containers and Inserts (6 mL)
- BOND Mixing Vial
- BOND Slide Label and Print Ribbon Kit

2.2 BOND RX and BOND RX^m Processing Modules

The processing module (PM) is the BOND RX system's staining platform. A single-seat BOND RX system can have up to 5 processing modules, and a BOND RX-ADVANCE system can have up to 30, in any mix of processing module types.



WARNING: The processing module must be connected to an earthed mains power outlet socket, and be positioned so that personnel are easily able to disconnect the mains power cable without having to move the processing module.

- [2.2.1 Main Components](#)
- [2.2.2 Processing Module Initialization](#)
- [2.2.3 Lid](#)
- [2.2.4 Main Robot and ID Imager](#)
- [2.2.5 Slide Staining Assemblies](#)
- [2.2.6 Front Cover](#)
- [2.2.7 Bulk Containers Cavity](#)
- [2.2.8 Aspirating Probe](#)
- [2.2.9 Wash Block and Mixing Station](#)
- [2.2.10 Bulk Fluid Robots \(BOND RX only\)](#)
- [2.2.11 Syringes](#)
- [2.2.12 Power Switch](#)
- [2.2.13 Back Cover](#)

2.2.1 Main Components

See the main components for BOND RX and BOND RX^m:

- [2.2.1.1 BOND RX](#)
- [2.2.1.2 BOND RX^m](#)

2.2.1.1 BOND RX

The following photos show the main processing module components for the BOND RX. The current model is shown – earlier models differ in appearance, however the main components are the same.

A description of the back cover is given at [2.2.13 Back Cover](#).

Figure 2-1: Front view of the previous (left) and current (right) BOND RX Processing Module



Legend

- | | |
|-----------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 Lid
2.2.3 Lid</p> <p>2 Main Robot Arm
2.2.4 Main Robot and ID Imager</p> | <p>3 Front Cover
2.2.6 Front Cover</p> <p>4 Bulk Containers Cavity
2.2.7 Bulk Containers Cavity</p> |
|-----------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------|

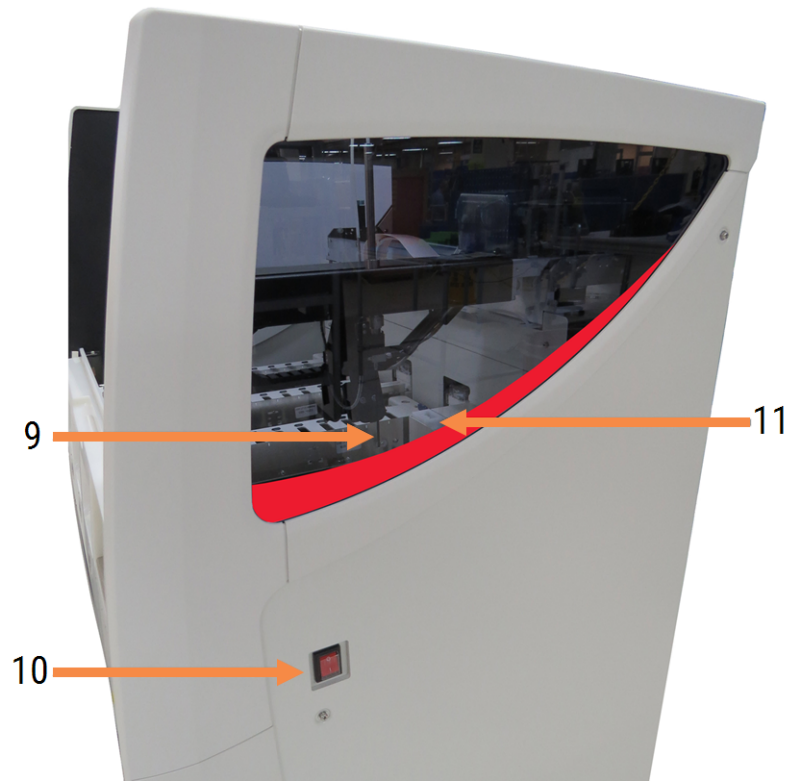
Figure 2-2: The front of the BOND RX Processing Module



Legend

- | | |
|----------------------------------------------------------------|------------------------------------------------|
| 5 Bulk Fluid Robots
2.2.10 Bulk Fluid Robots (BOND RX only) | 7 Syringes
2.2.11 Syringes |
| 6 Slide Staining Assemblies
2.2.5 Slide Staining Assemblies | 8 Reagent Platform
2.2.6.5 Reagent Platform |

Figure 2-3: The BOND RX processing module viewed from the right side



Legend

- 9 Aspirating Probe
2.2.8 Aspirating Probe
- 10 Power Switch
2.2.12 Power Switch

- 11 Wash Block and Mixing Station
2.2.9 Wash Block and Mixing Station

2.2.1.2 BOND RX^m

The following photos show the main components of the BOND RX^m Processing Module.

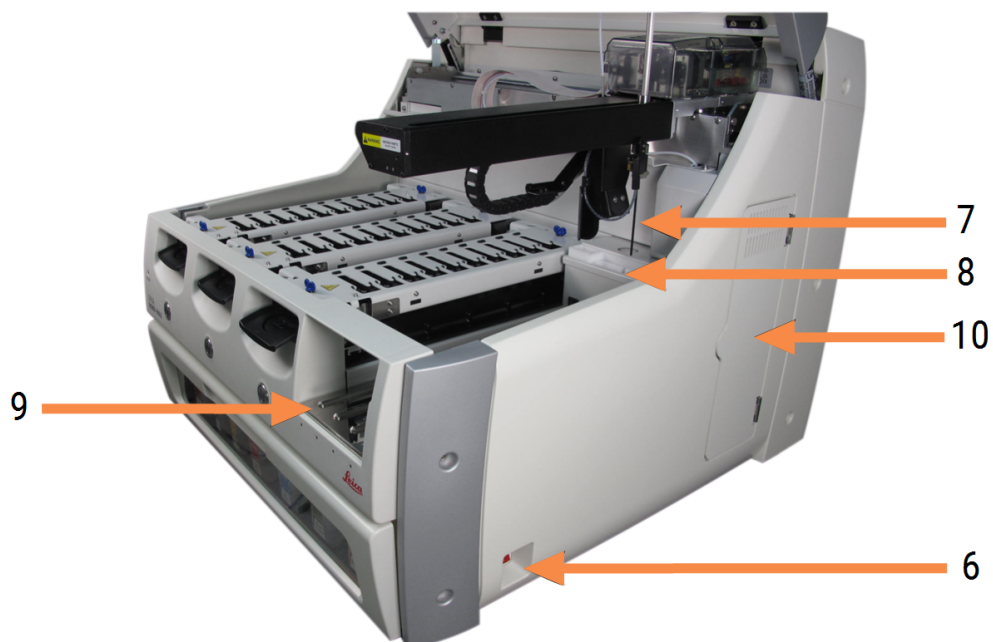
Figure 2-4: Front view of the BOND RX^m Processing Module



Legend

- | | |
|----------------------------------------------------------------|----------------------------------------------------------|
| 1 Lid
2.2.3 Lid | 4 Front Cover
2.2.6 Front Cover |
| 2 Robot Arm
2.2.4 Main Robot and ID Imager | 5 Bulk Containers Cavity
2.2.7 Bulk Containers Cavity |
| 3 Slide Staining Assemblies
2.2.5 Slide Staining Assemblies | |

Figure 2-5: The BOND RX™ Processing Module viewed from the right side



Legend

6	Power Switch 2.2.12 Power Switch	9	Reagent Platform 2.2.6.5 Reagent Platform
7	Aspirating Probe 2.2.8 Aspirating Probe	10	Syringe (see below) 2.2.11 Syringes
8	Wash Block & Mixing Station 2.2.9 Wash Block and Mixing Station		

A description of the back cover is given at [2.2.13 Back Cover](#).

Figure 2-6: Syringe behind hinged door



2.2.2 Processing Module Initialization

When you turn the processing module on, the BOND RX system performs internal checks, primes the fluidics system and moves the robots to their home positions. The main robot moves to the back left corner of the processing module and the three bulk fluid robots (BOND RX only) move to the rear of the processing module.

The slide staining assemblies initialize and return to their unlocked position. The initialization process halts if a fault is found or if the module is in a state unsuitable for processing.

Before attempting to initialize a processing module, check the following items:

- The lid is closed
- The front door is closed (BOND RX^m only)
- The bulk waste containers are less than half full
- The bulk reagent containers have adequate reagent
- The mixing station is in place
- The mixing station vials are empty and clean
- The top plates of the slide staining assemblies (SSAs) are in the closed position.

The power LED on the front of the processing module turns green and the BOND RX software indicates that the module is connected. When initialization is complete an icon of the three slide trays appears on the processing module tab (see [5.1.1 Processing Module Tabs](#)). Do not attempt to use a processing module until it is fully initialized.

2.2.3 Lid

The lid is designed to be closed during operation, and is protected with interlocks.



WARNING: Take care when closing the processing module lid, ensuring hands are kept clear to avoid injury.



WARNING: During operation the main robot, the aspirating probe and bulk fluid robots (BOND RX only) can move without warning and with a speed that can cause injury.

Do not attempt to open the processing module lid while a run is in progress.

Do not attempt to by-pass the interlocks that prevent operation of the processing module when the lid is opened.



WARNING: Contact customer support immediately if the main robot and/or bulk fluid robots continue to operate for more than approximately 5 seconds after the processing module lid has been opened.

2.2.4 Main Robot and ID Imager

The main robot positions the aspirating probe to aspirate and dispense reagents. The robot arm holds the ID imager, which is used to identify the slides and reagents loaded in the processing module.

Figure 2-7: Photo of the main robot with the ID imager indicated by the arrow



WARNING: Do not move the main robot arm while the processing module is switched on. The robot can become misaligned, resulting in poor staining.

If the robot has been moved: power down the processing module, wait 30 seconds and then reinitialize.

For slides, the BOND RX system scans each slide label for identification purposes (see [5.1.5.1 Automatic Slide Identification](#)).

- The ID imager window should be periodically cleaned.
See [12.9 ID Imager](#) for instructions.
- If the aspirating probe is broken or bent, contact Customer Support.

2.2.5 Slide Staining Assemblies




WARNING: Avoid contact with slide staining assemblies and their surrounds. These may be very hot and cause severe burns. Allow twenty minutes after the cessation of operation for the slide staining assemblies and their surrounds to cool.



WARNING: Potentially hazardous reagents can collect around the slide staining assemblies and contaminate the slide trays. Always wear approved protective clothing and gloves when handling slide trays.

Slides are processed in the slide staining assemblies. Each processing module contains three slide staining assemblies. To begin a run, an operator inserts a slide tray through the front cover (described in [2.2.6 Front Cover](#)), then presses the load button. The BOND RX system will capture images of the slides. If the slides are compatible (refer to [6.9 Slide Compatibility](#)) and all reagents are present, the user can then start the run. For more information about entering slide details and loading slides, see [6 Slide Setup \(on BOND RX Controller\)](#).

Before processing begins, the BOND RX system locks the slides into the slide staining assembly. If you need to remove a slide tray while the BOND RX system is processing its slides, you must first abandon the run. Click  below the tray on the **System status** screen (see [5.1.7 Starting or Stopping a Run](#)) and then unlock the slide staining assembly.

For cleaning and routine maintenance of the slide staining assembly, see [12.4 Slide Staining Assembly](#).

Slide Staining Assembly Heaters



WARNING: Heaters and heated surfaces on the processing module can be ignition dangers:

- Do not place flammable materials on or near the heaters.
- Do not place flammable materials on any hot surfaces on the processing module.
- Ensure all bulk container caps are properly sealed after refilling or emptying.

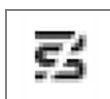


WARNING: Some of the reagents used on BOND RX and BOND RX^m processing modules are flammable:

- Do not place a flame or ignition source near the processing modules.
- Ensure all bulk container caps are properly sealed after refilling or emptying.

The BOND RX and BOND RX^m Processing Modules have a heating element at each slide position. Each of these elements is independently monitored and is marked as faulty if a temperature error occurs (see [Figure 2-8](#)). Contact customer support if a faulty heater is indicated.

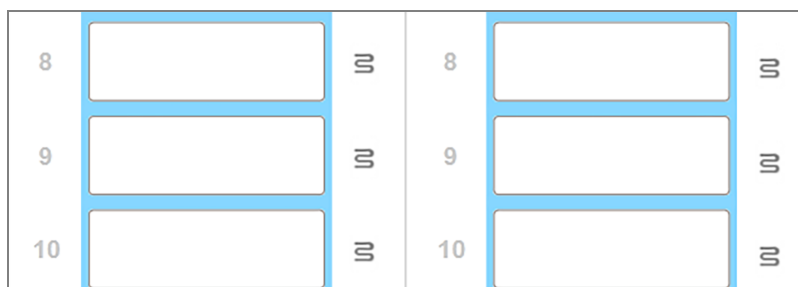
Figure 2-8: Individual heater error



You should not attempt to run a slide that requires heating at a position marked as faulty. If a heater malfunctions during a run then the slide at that position may not have processed correctly.

If the heater malfunction is a potential safety risk, the processing module turns off all slide heaters, including the heater of any temperature-controlled slide currently being processed.

Figure 2-9: Gray heater symbols at each position indicate a complete heating shutdown



Once slide heating is shut down, you must turn off then restart the processing module to clear the heater lock. You can continue to use slide positions with faulty heaters so long as the slides processed there do not require heating.

2.2.6 Front Cover

The figures below show the front covers of the BOND RX and BOND RX^m.

Figure 2-10: BOND RX front cover



Legend

- | | |
|--------------------------------------------|------------------------------------------------------|
| 1 Front Cover
2.2.6.1 Power LED | 4 Reagent Platform
2.2.6.5 Reagent Platform |
| 2 Slide Tray Bay
2.2.6.2 Slide Tray Bay | 5 Reagent Tray LED
2.2.6.6 Reagent Tray LED |
| 3 Slide Tray LED
2.2.6.3 Slide Tray LED | 6 Load / Unload Button
2.2.6.4 Load/Unload Button |

Figure 2-11: BOND RX^m front cover

Legend

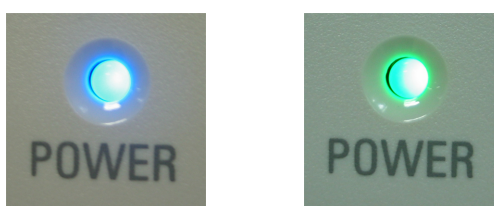
- | | |
|--------------------------------------------|------------------------------------------------------|
| 1 Front Cover
2.2.6.1 Power LED | 4 Reagent Platform
2.2.6.5 Reagent Platform |
| 2 Slide Tray Bay
2.2.6.2 Slide Tray Bay | 5 Reagent Tray LED
2.2.6.6 Reagent Tray LED |
| 3 Slide Tray LED
2.2.6.3 Slide Tray LED | 6 Load / Unload Button
2.2.6.4 Load/Unload Button |

2.2.6.1 Power LED

This operates as follows:

- **Off** – no power
- **Blue** (current model) or **Orange** (previous models) – power on, but processing module software has not yet started
- **Green** – power on, system operating.

Figure 2-12: Power LED colors (blue, green) on BOND RX^m Processing Module



2.2.6.2 Slide Tray Bay

There are three bays (one for each slide staining assembly) where slide trays are inserted. When a slide tray is inserted, press the Load/Unload button to lock it into the slide staining assembly. After a tray is locked, the robot arm moves the ID imager over the slides in the tray to automatically identify the slides.

2.2.6.3 Slide Tray LED

Multi-color LEDs on the front cover under each slide staining assembly indicate the state of the slide tray. On BOND RX^m Processing Modules, the slide tray LEDs are incorporated into the Load/Unload buttons. On these processing modules the LED turns blue for a few seconds when you press it.

Slide staining assembly LED color indicators are as follows:

- **Off** – there is no slide tray present, or the slide tray is unlocked.
- **Steady orange** – the tray is loaded and locked but processing has not commenced.

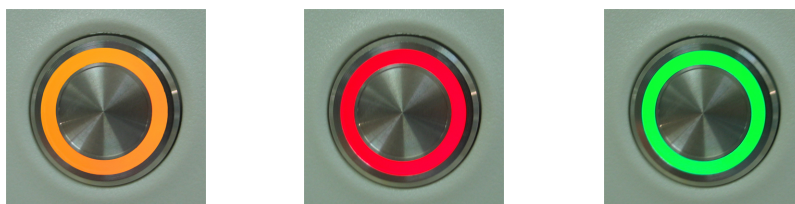
The tray can be safely unlocked and removed with the Load/Unload button.

- **Steady red** – the slides in the tray are being processed.

The tray is locked and cannot be unlocked with the Load/Unload button. To unload you need to first abandon the run in the software.

- **Flashing green** – processing has finished without notifications. Unlock with the Load/Unload button.
- **Flashing red** – the run has been rejected, or processing has completed with notifications. Unlock with the Load/Unload button.

Figure 2-13: Slide tray LED colors (orange, red, green) on BOND RX^m Processing Module



2.2.6.4 Load/Unload Button

Pressing a Load/Unload button does the following:

- If a tray is not loaded, nothing will happen.
- If a tray is loaded and not locked, the BOND RX or BOND RX^m will lock the tray and, when the robot arm is available, the ID imager will identify the slide IDs.
- If a tray is locked and the run has not started, the BOND RX or BOND RX^m will unlock the tray.
- If a tray is locked and the run is finished, the BOND RX or BOND RX^m will unlock the tray.
- If a tray is locked and a run is in progress, the Load/Unload button has no effect. You cannot unlock a tray until a run using that tray is finished or abandoned.

If a slide staining assembly is hot you cannot lock or unlock a tray – wait until the assembly has cooled.

2.2.6.5 Reagent Platform

This is where reagent trays are placed, containing detection systems, 7 mL and 30 mL reagent containers, and/or 6 mL titration containers. Each tray can hold up to nine reagents, and the reagent platform can hold four reagent trays.

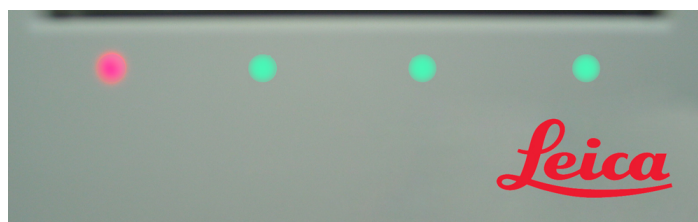
To load a reagent tray, slide the tray onto the platform and into the locking mechanism (see [4.1.4 Loading the Reagents](#)). When the robot arm is available the BOND RX system will identify the reagents in each reagent position.

2.2.6.6 Reagent Tray LED

Below each tray position there is a bi-color LED that functions as follows:

- **Off** – a tray has not been detected.
If a tray is inserted and the LED is off, check that the tray is inserted correctly.
- **Steady red** – a reagent on the tray is required within the next two minutes.
The tray is locked and cannot be removed.
- **Steady green** – none of the reagents on this tray are required within the next two minutes.
The tray is unlocked and may be temporarily removed.

Figure 2-14: Reagent tray LED colors (red, green) on BOND RX^m Processing Module



2.2.7 Bulk Containers Cavity

Bulk reagent and waste containers are located below the front cover in both the BOND RX and BOND RX^m. The BOND RX^m also has an external container for standard waste.

See [12.2 Bulk Containers](#) for bulk container filling, emptying, and maintenance instructions.



WARNING: To ensure correct processing module operation, place each bulk reagent container into its correct station in the cavity, as indicated by the color-coded name labels.

For BOND RX, see [Figure 2-15](#); for BOND RX^m, see [Figure 2-17](#).

Failure to do so may compromise staining.



WARNING: Some of the reagents used on BOND RX and BOND RX^m Processing Modules are flammable:

- Do not place a flame or ignition source near the processing modules.
- Ensure all bulk container caps are properly sealed after refilling or emptying.

- [2.2.7.1 BOND RX](#)
- [2.2.7.2 BOND RX^m](#)

2.2.7.1 BOND RX

The previous BOND RX has two transparent cabinet doors that allow easy access to all bulk containers. Hold the railing at the top of the doors when opening.

All waste from the slide staining assemblies is sent to the hazardous waste container. Waste from the wash block is sent to the standard or hazardous waste containers depending on the status of reagent in the waste (you must set reagents that you create to be hazardous if appropriate – see [8.2.1 Adding or Editing a Reagent](#)).

Weight sensors for each bulk reagent and waste container warn the user when the reagent level is low or the waste level is too high. Each bulk container's status is visually indicated by the [Bulk Container Lighting System \(BOND RX\)](#) (on page 49). Note that this system is not fitted to the previous BOND RX; you can instead use the on-screen icons (see [5.1.3.7 Bulk Container Status](#)).

The BOND RX has space for the following containers, in the shelves indicated in [Figure 2-15](#), moving from left to right:

Station	Container	Position	Size (L)	Color	Reagent
8	ER1	Upper shelf	2	Purple	BOND Epitope Retrieval Solution 1*
9	ER2		2	Light Purple	BOND Epitope Retrieval Solution 2*
1	Dewax Solution	Lower shelf	5	Red	BOND Dewax Solution*
2	Deionized Water		5	Blue	Deionized Water
3	Wash Buffer		5	Green	BOND Wash Solution*
4	Alcohol		5	Orange	Alcohol (reagent grade)
5	Bulk Waste		5	Gray	Standard Waste
6	Bulk Waste		5	Gray	Standard Waste
7	Hazardous Waste		5	Brown	Hazardous Waste

* Use only BOND reagents – do not substitute with alternative products.

If your laboratory does not use the epitope retrieval and/or dewax reagent containers, these can be disabled in the administration client – see [10.6.1.1 Disabling Bulk Reagent Containers](#).

Figure 2-15: BOND RX bulk reagent containers in position



Bulk Container Lighting System (BOND RX)

BOND RX Processing Modules are fitted with a bulk container lighting system, as shown in [Figure 2-16](#) below.

Figure 2-16: Bulk container lighting system



The bulk container lighting system helps you to see the liquid level in each container, and the lights are a static white color during normal operation.

The lights also indicate the current status of each bulk container:

- When a bulk supply container is almost empty, or a waste container is almost full, its white lighting pulses.
- When a bulk supply container is empty or a waste container is full, and this affects the current run, its lighting pulses red.
- When a bulk container is removed, its backlight switches off and its label lighting on the processing module cavity pulses white.



The bulk container lighting system will operate only with BOND RX 6.0 or later software.

Also refer to [5.1.3.7 Bulk Container Status](#) for details of how the bulk containers are shown on the **System status** screen.

2.2.7.2 BOND RX^m

BOND RX^m has a single downwards-opening door for access to bulk containers. The door has a transparent panel allowing you to view reagent levels in the bulk containers (which are also translucent).

The door is held by magnetic latches.



The bulk container cavity door must remain closed during staining runs. If the door is opened, an alert indication will appear on the system status screen (see [5.1.2 Hardware Status](#)) and any current runs may pause.

Waste from the processing module is sent to the standard or hazardous waste containers depending on the status of reagent in the waste (you must set reagents that you create to be hazardous if appropriate – see [8.2.1 Adding or Editing a Reagent](#)).

BOND RX^m bulk reagent containers have liquid level sensors to warn when the reagent level is low; waste containers also have liquid level sensors to warn when the waste level is too high. See [12.2 Bulk Containers](#) for refilling and emptying directions.

BOND RX^m has space for the following containers, in order from left to right:

Station	Container	Size (L)	Color	Reagent
1	Hazardous Waste	2	Brown	Hazardous Waste
2	ER1	1	Purple	BOND Epitope Retrieval Solution 1*
3	ER2	1	Light Purple	BOND Epitope Retrieval Solution 2*
4	Dewax Solution	2	Red	BOND Dewax Solution*
5	Deionized Water	2	Blue	Deionized Water
6	Wash Buffer	2	Green	BOND Wash Solution*
7	Alcohol	2	Orange	Alcohol (reagent grade)

*Use only BOND reagents – do not substitute with alternative products.

The epitope retrieval and/or dewaxing reagent containers can be removed from the processing module if not used – see [10.6.1.1 Disabling Bulk Reagent Containers](#).

Figure 2-17: BOND RX^m bulk reagents in position

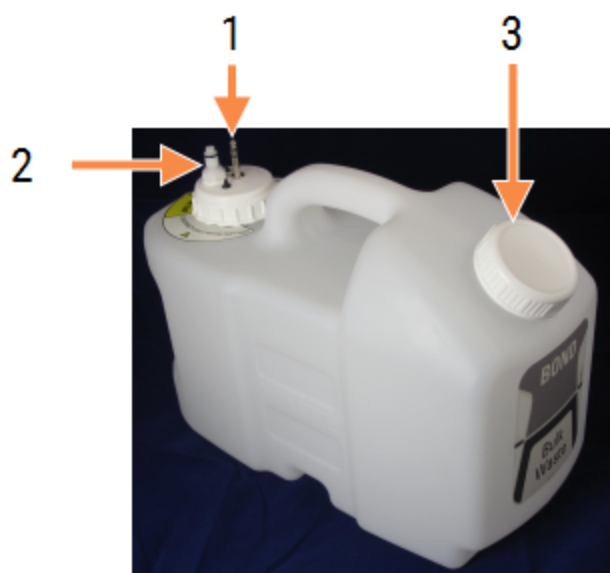


External Waste Container

A nine-liter external standard waste container is included with the BOND RX^m.

The supplied container has two caps – one for the connectors and a second for emptying the waste. Never remove the connector cap on this container.

Figure 2-18: BOND RX^m external waste container



Legend

- | | |
|---|--------------------------------|
| 1 | Sensor connector |
| 2 | Fluid connector |
| 3 | Opening for emptying container |

The fluid line connects to a push-fit connector at the bottom right of the processing module back cover. The liquid level sensor connects to a three-pin connector at the upper left of the rear cover (see [Figure 2-26](#)).

See [12.2.4 External Waste Container \(BOND RX^m only\)](#) for emptying and maintenance instructions for the external container.



CAUTION: Always disconnect the sensor and fluid connectors (in this order) before emptying an external waste container. Do not attempt to pour fluid from a container while the cable and tube are still attached.



WARNING: Some of the reagents used in immunohistochemistry and in situ hybridization are hazardous. Ensure you have received adequate training for this procedure before continuing:

- 1 Wear latex or nitrile gloves, safety glasses, and other suitable protective clothing when handling reagents or cleaning the processing module.
- 2 Handle and dispose of reagents and condensate in accordance with all relevant procedures and government regulations that apply at the laboratory site.



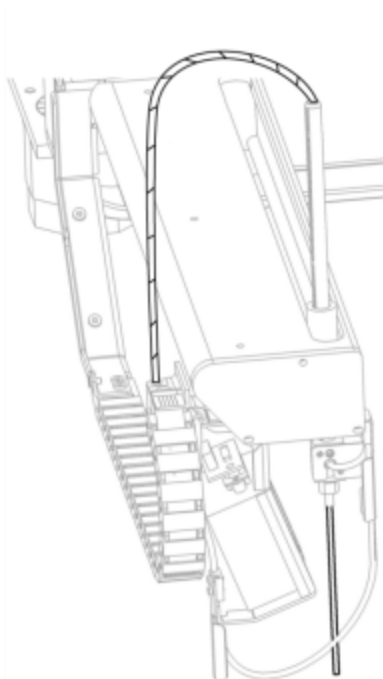
WARNING: Some of the reagents used on BOND RX and BOND RX^m Processing Modules are flammable:

- Do not place a flame or ignition source near the processing modules.
- Ensure all bulk container caps are properly sealed after refilling or emptying.

2.2.8 Aspirating Probe

The aspirating probe aspirates reagents from containers, delivers reagents to the slides in the slide staining assemblies, and mixes chromogens in the mixing station. It contains a liquid level sensor to detect reagent level (refer to [8.3.1 Determining Reagent Volume](#)).

Figure 2-19: Aspirating probe in the robot arm

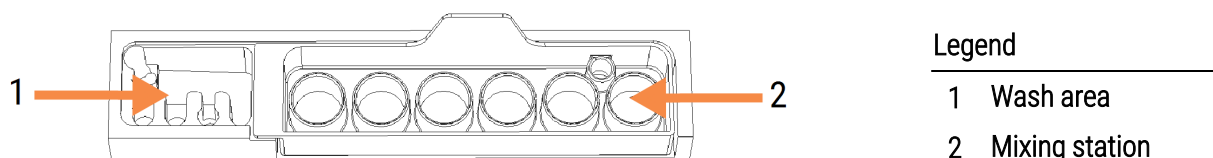


There is a residual volume in each container that the probe is unable to reach. This volume is referred to as the “dead volume”. Dead volume is different for each type of container (see [18.5 Operating Specifications](#) in [18 Specifications](#) for dead volume values).

See [12.6 Aspirating Probe](#) for maintenance instructions for the aspirating probe.

2.2.9 Wash Block and Mixing Station

Figure 2-20: Wash block with mixing station inserted



The left-hand wash area includes small holes for washing the aspirating probe.

The right-hand part of the wash block holds the mixing station, which consists of six cavities. These are mixing vials for short-life reagents that must be mixed just before use. The mixing of reagents is determined by the software, depending on the reagent type.



The BOND RX software tracks the state of the mixing station and does not initialize the BOND RX or BOND RX^m if the station's tracked state is other than clean and empty (see [5.1.2 Hardware Status](#)). If notified during initialization that the mixing station is dirty or has liquid in it, ensure that the station is clean and empty before clicking **OK** in the notification dialog. If you continue with a dirty and/or non-empty mixing station, reagents could be contaminated or the mixing vials could overflow.

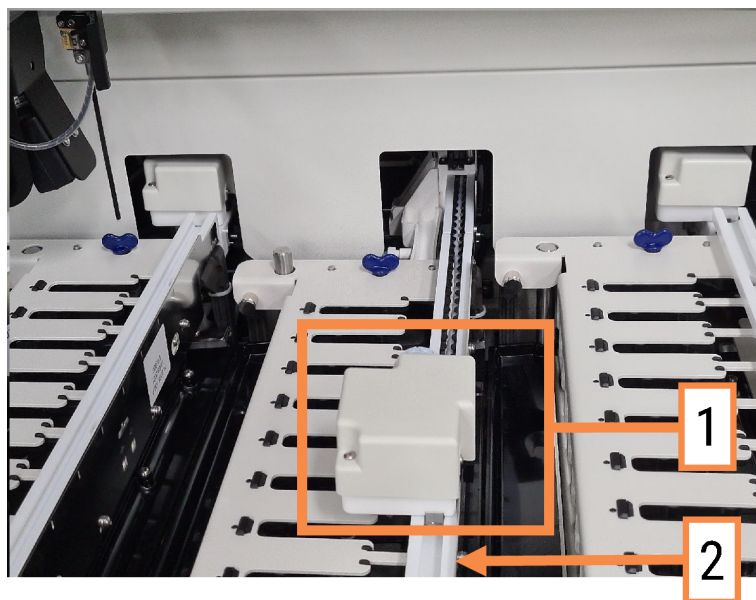


During initialization the BOND RX system scans a label on the mixing station to check that it is present. If the BOND RX software cannot detect this ID, then a message will prompt you to confirm that a mixing station is present.

See [12.7 Wash Block and Mixing Station](#) for mixing station maintenance instructions.

2.2.10 Bulk Fluid Robots (BOND RX only)

Figure 2-21: The BOND RX bulk fluid robot (1) moves along a guide rail (2) on each slide staining assembly



WARNING: Contact customer support immediately if the main robot and/or bulk fluid robots continue to operate for more than 5 seconds after the processing module lid has been opened.

The BOND RX Processing Module has three bulk fluid robots that move along a guide rail on each slide staining assembly and dispense reagents to all slides present. The robots deliver only bulk reagents, while the aspirating probe delivers reagents from containers in the reagent platform and some bulk reagents. Each bulk fluid robot has a wash block to rinse and clean its dispensing probe.

2.2.10.1 Manually Returning a Bulk Fluid Robot to the Home Position

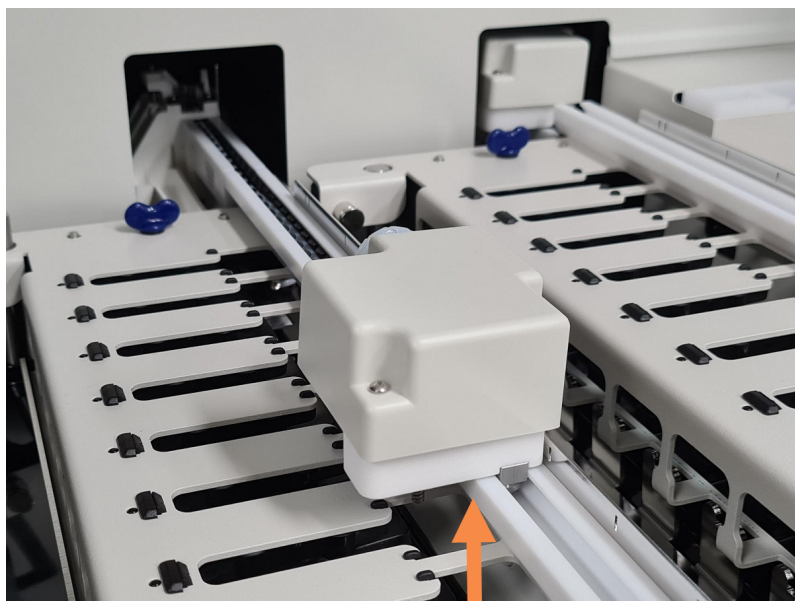
If a bulk fluid robot stops working and is positioned along the slide staining assembly, press the Load/Unload button to return it to its home position. If it remains on the slide staining assembly complete the following steps to manually return it to the home position and retrieve any slides in the slide staining assembly.

- 1 Ensure that the processing module is idle with no runs scheduled or processing, then turn it off.
- 2 Gently lift the dispense block on the bulk fluid robot (see [Figure 2-22](#)) until the probe clears the top plate.

- 3 Push the robot along the rail to the rear of the slide staining assembly. Use a slow, steady motion – do not push too fast.

Push until the robot is just clear of the top plate rail – **do not** push it back as far as it will go.

Figure 2-22: Lift the dispense block



- 4 When the robot is clear of the top plate, close the lid and turn the processing module back on. The slide staining assembly should unlock as part of the initialization routine.

If the slide staining assembly does not unlock, see [12.4.1 Manually Unlocking Slide Staining Assemblies](#) for instructions on how to retrieve the slide trays.

- 5 Retrieve the slide tray and slides.

2.2.11 Syringes

The syringes aspirate and dispense the precise reagent fluid volumes required by the BOND RX system. See [12.13 Syringes](#) for syringe maintenance instructions.

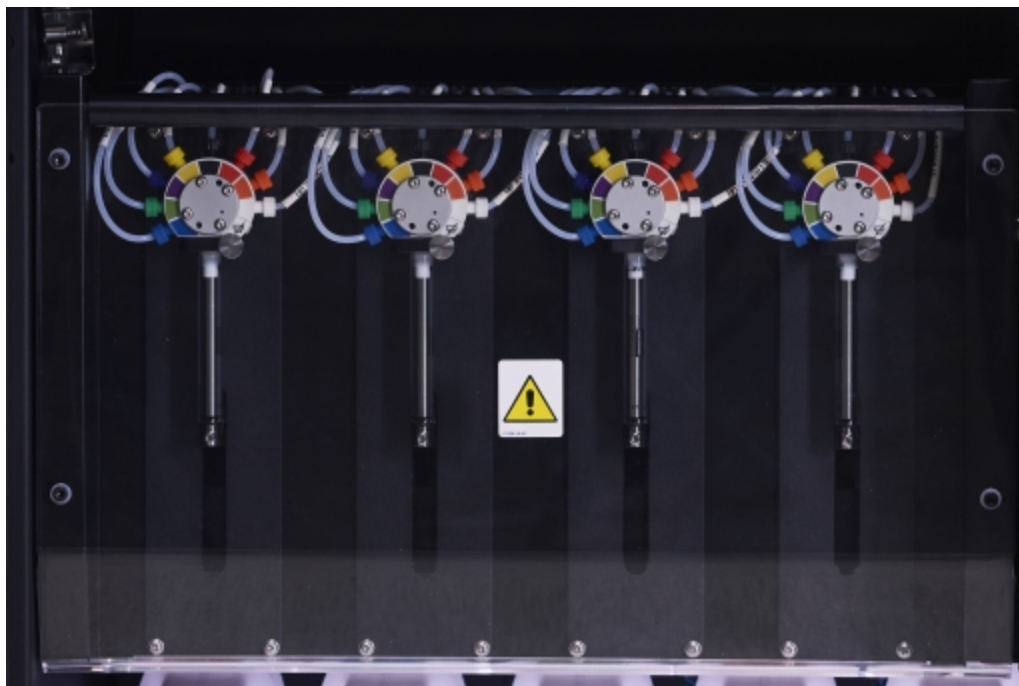


WARNING: Ensure that the syringe door is closed (BOND RX™) or the syringe cover is fitted (BOND RX) during normal operation. If a syringe or a syringe fitting becomes loose, reagent under pressure can spray from the syringe.

2.2.11.1 BOND RX

The BOND RX has four syringe pumps, located below the front cover. The first three syringe pumps, from left to right, are used by the bulk fluid robots on SSA1, SSA2 and SSA3 above. The fourth, main syringe pump, is used by the aspirating probe.

Figure 2-23: BOND RX syringes



CAUTION: Ensure the syringe module is fully closed before starting a run or initializing the processing module (see [12.4.1 Manually Unlocking Slide Staining Assemblies](#)). Failure to do so can result in damage to the syringes during operation.

2.2.11.2 BOND RX^m

The BOND RX^m has a single syringe pump located in a compartment on the right side of the processing module. This is a 9-port syringe valve (one port is not used) with a screw-in syringe barrel and a small clamp.

Figure 2-24: BOND RX^m 9-port syringe



To check the condition of the syringe unit, open the door by pressing and releasing at the rounded tab in the front middle of the door.



WARNING: Always wear protective clothing and gloves.

Check regularly during initialization and replace when required or prompted – see [12.13 Syringes](#).

2.2.12 Power Switch

This is a single rocker switch located on the right cover of the processing module. This is used to turn the processing module on and off.

- For the power switch location on the BOND RX, see [Figure 2-3](#).
- For the power switch location on the BOND RX^m, see [Figure 2-5](#).

2.2.13 Back Cover



WARNING: Do not remove the processing module covers or attempt to access internal components. Dangerous voltages are present inside BOND RX Processing Modules and only qualified service technicians approved by Leica Biosystems should perform these tasks.

2.2.13.1 BOND RX

Figure 2-25 shows the back cover of the BOND RX Processing Module.

Figure 2-25: BOND RX back cover



Legend

- | | | | |
|---|---------------------------------------------------------------------------------------------------------------------------------------|---|------------------------|
| 1 | Circuit Breakers (Legacy processing modules only) | 3 | Mains Power Connection |
| 2 | Fuses | 4 | Ethernet Connection |
| | <ul style="list-style-type: none"> • Legacy processing modules—4 fuses • Alternate processing modules—2 fuses | | |

See [12.14 Power Supply Fuses](#) for instructions to replace fuses.

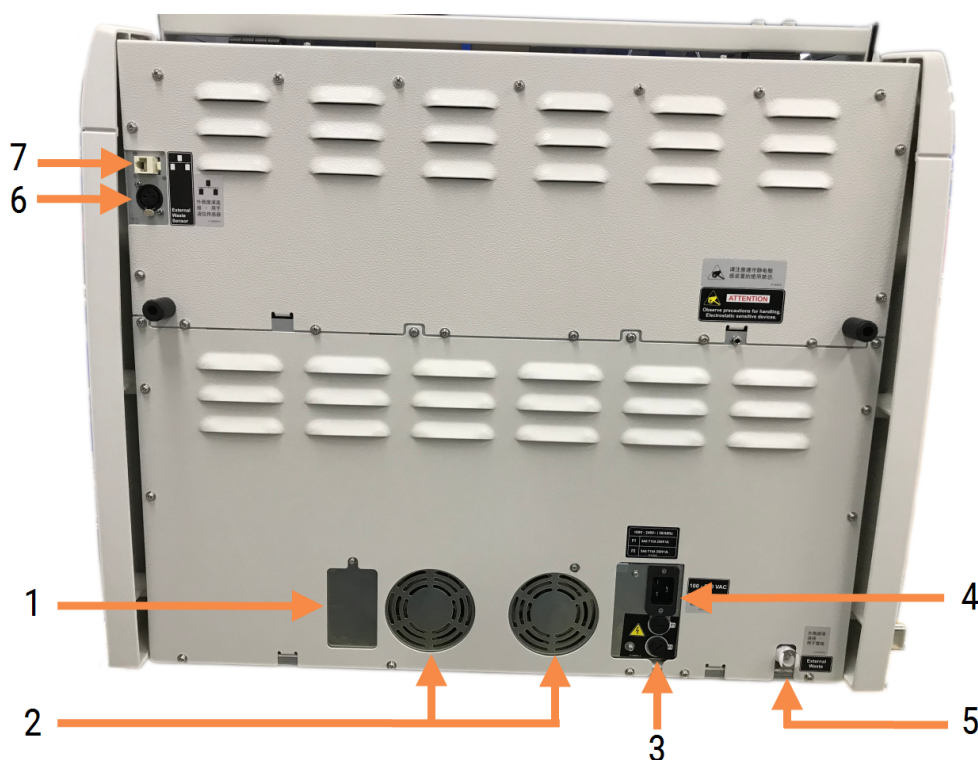


WARNING: Do not use the two black handles on the back cover of the BOND RX to lift the processing module.

2.2.13.2 BOND RX^m

Figure 2-26 shows the back cover of the BOND RX^m Processing Module.

Figure 2-26: BOND RX^m back cover



Legend

- | | | | |
|---|---------------------------------------------------------------------------------------------------------------------------------------------|---|---------------------------------------------------------------------------------------------------------------------------------------|
| 1 | Circuit Breakers (Legacy processing modules only) | 5 | External Waste Connection – for tubing (see 12.2.4 External Waste Container (BOND RX^m only)) |
| 2 | Power Supply Fans | 6 | External Waste Connection – for liquid level sensor (see 12.2.4 External Waste Container (BOND RX^m only)) |
| 3 | Fuses <ul style="list-style-type: none"> • Legacy processing modules—4 fuses • Alternate processing modules—2 fuses | 7 | Ethernet Connection |
| 4 | Mains Power Connection | | |

See [12.14 Power Supply Fuses](#) for instructions to replace fuses.

2.2.13.3 Disconnecting the Processing Module

To disconnect a BOND RX or BOND RX^m Processing Module from the mains power supply, do the following:

- 1 Switch off the power using the switch at the right side of the processing module.
- 2 Trace the power cable from the processing module mains power connection (item 3 in [Figure 2-25](#) and item 4 in [Figure 2-26](#)) to the wall. Switch off the mains power supply at the wall socket.
- 3 Disconnect the plug from the back of the processing module.

2.3 BOND RX Controller and Terminals

All BOND RX systems include a BOND RX controller, where all software processing is carried out. In single-seat installations (see [3.1.1 Single-Seat Configuration](#)), one controller with a keyboard, mouse and monitor, is used to run the client software. Single-seat installations are adequate for running five or fewer processing modules.

Laboratories with BOND RX-ADVANCE installations (see [3.1.2 BOND RX-ADVANCE](#)), with more than five processing modules, have, in addition, BOND terminals. In these installations most user interaction with the BOND software occurs at the terminals, each of which can control any or all processing modules. It is also possible to control the same processing module(s) from more than one terminal.

The BOND controller continues to carry out all software processing. Controllers in BOND RX-ADVANCE installations have a higher specification than those used in single-seat installations and include multiple levels of redundancy to ensure excellent reliability.

Some BOND RX-ADVANCE installations include a secondary (backup) controller. This controller records all processes on the primary controller, and can be switched to in the event that the primary controller malfunctions. Ideally, secondary controllers should not be located nearby the primary controller, to decrease the likelihood of both controllers being damaged by a localized event.

A slide label printer and handheld barcode scanner are connected to the controller in single-seat installations, or to each terminal in BOND RX-ADVANCE installations.



CAUTION: The operating system and software on the BOND RX controller are designed to provide optimal control over the BOND RX system. To avoid any possibility of delays or interference with system control, do not install any additional software on the BOND RX controller or terminal.

2.4 Handheld Barcode Scanner

Figure 2-27: The handheld barcode scanner



USB handheld barcode scanners are attached to the controller (single-seat installations) or to terminals (BOND RX-ADVANCE installations). They are used to register reagents, and can also be used to identify slides (see [6.5.6 Manually Identifying a Slide](#)).



Creating 1D and OCR barcodes is not supported in BOND RX version 7 onwards.

The handheld barcode scanner should be installed and operational when your BOND RX system is installed. See [13.1 Handheld Barcode Scanners](#) for maintenance and configuration instructions.

2.4.1 Using the Handheld Barcode Scanner

To read a barcode, point the scanner at it and press the trigger. Align so that the red line extends the full length of the barcode. The scanner beeps and the indicator turns green when a barcode is recognized. If a barcode is not recognized the scanner beeps and the indicator turns red.



Do not hold barcodes too close to the scanner. If the scanner does not recognize a barcode, try moving the barcode further away or scan the barcode at a 45° angle (to prevent feedback to the scanner).

When the scanner is placed in its stand then it is in hands-free use, and you do not need to press the trigger when reading a barcode.

2.5 Slide Labeler

Single-seat BOND RX systems include one slide label printer (called a “slide labeler”) connected to the controller. In BOND RX-ADVANCE installations a separate slide labeler is connected to each terminal.

The slide labeler prints stick-on labels to attach to slides for identification. All labels include a unique slide ID rendered as a 2D barcode (see [10.5.2 Study and Slide Settings](#)). The BOND RX system uses the IDs to automatically identify slides when they are loaded onto processing modules. You can configure other information, as well as the IDs, to appear on labels – see [10.3 Labels](#).

Some laboratories use slide labels printed from their LIS, however the BOND RX slide labeler is still included in these systems for any slides created with the BOND RX research client.

The slide labeler is set up as part of the standard BOND RX installation. If you replace a slide labeler, configure this in the administration client **Hardware** screen (see [10.6.3 Slide Labelers](#)). Use the documents supplied with the labeler for information on label and ribbon replacement, and cleaning.



WARNING: Use only BOND slide labels and printing ribbon. These labels must remain attached and legible during processing on BOND RX processing modules.

2.6 Ancillary Equipment

This section describes the ancillary equipment used with the BOND RX system.

- [2.6.1 Slides](#)
- [2.6.2 BOND Universal Covertiles](#)
- [2.6.3 Slide Trays](#)
- [2.6.4 Reagent Trays](#)
- [2.6.5 Reagent Systems and Containers](#)

2.6.1 Slides

Use only glass slides of correct size on BOND RX and BOND RX^m Processing Modules. Slides of the wrong size may not sit properly in the slide trays, and Covertiles will not sit properly on them. Both of these could affect staining quality.

Leica Biosystems recommends Leica BOND Plus slides and Apex BOND slides, which are designed for use on the BOND RX system. As well as being the optimal size for BOND slide trays and Covertiles, these positively charged slides are marked to show the areas where tissue should be placed for 100 µL and 150 µL dispenses (see [6.5.8 Dispense Volumes and Tissue Position on Slides](#)).

If you use your own slides, they must conform to the following specifications:

Dimensions	Width: 24.64–26.0 mm (0.97–1.02 in) Length: 74.9–76.0 mm (2.95–2.99 in) Thickness: 0.8–1.3 mm (0.03–0.05 in)
Label area	Width: 24.64–26.0 mm (0.97–1.02 in) Length: 16.9–21.0 mm (0.67–0.83 in)
Material	Glass, ISO 8037/1



CAUTION: Do not use damaged slides. Ensure all slides are correctly aligned on the slide trays before loading into the processing module.



CAUTION: Do not use slides with rounded or clipped corners. These slides may fall through the slide tray, and could alter fluid flow under the Covertiles, affecting staining quality.

2.6.2 BOND Universal Covertiles

BOND Universal Covertiles are transparent plastic covers that sit over slides during staining. Capillary action draws reagent that has been dispensed to slides between the Covertiles and slides, ensuring gentle, uniform tissue coverage. The Covertiles minimize the volumes of reagent required, and protect slides from drying between applications. Covertiles are an essential part of the BOND RX staining system and must always be used.

Place Covertiles on slides after placing slides in slide trays (see [4.1.3.5 Loading Slides](#)). Ensure the Covertiles are properly positioned, with the key in the neck of each Covertile (circled in photograph, right) fitting into the slot in the slide tray.

There are two Covertile designs – they can be used interchangeably. The new design includes features (the word **Leica**, a small circular mark and a projection at the top left) that make it more obvious when a Covertile has been incorrectly placed on a slide.



Figure 2-28: A BOND Universal Covertile (original design)

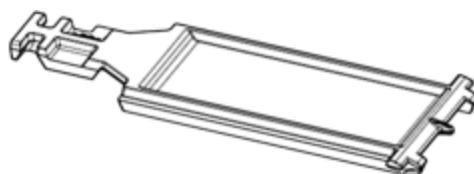
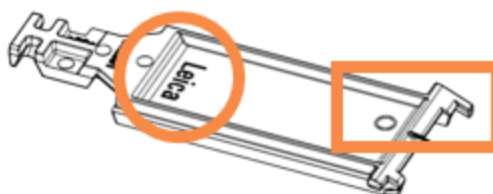


Figure 2-29: A BOND Universal Covertile (new design)



Covertiles can be reused up to 25 times provided they are not heavily discolored or damaged, and provided they are cleaned properly (see [12.3 Covertiles](#)). Discard damaged Covertiles.

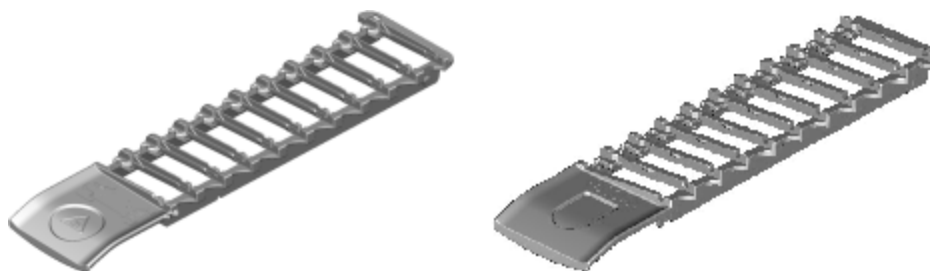
Some assays require the use of new (unused) Covertiles. Check the relevant assay Instructions For Use (IFU) beforehand.

2.6.3 Slide Trays

Use the slide trays to hold slides and Covertiles in position when you load them into the BOND RX or BOND RX^m Processing Module. Each tray can hold ten slides.

There are two slide tray designs – they can be used interchangeably.

Figure 2-30: Slide tray (new design (left) and old design (right))

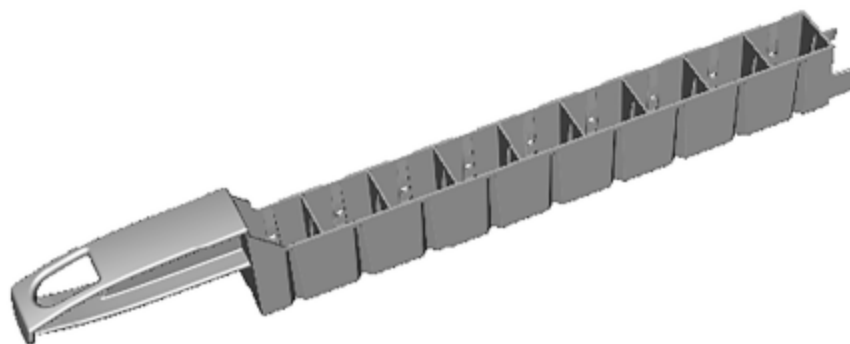


For instructions on loading slides and Covertiles into the processing module, see [4.1.3.5 Loading Slides](#).

2.6.4 Reagent Trays

Reagent trays hold 7 mL and 30 mL BOND reagent containers, and 6 mL BOND titration containers. The trays are loaded onto the processing module in the reagent platform (see [2.2.6.5 Reagent Platform](#)).

Figure 2-31: Reagent tray



Container positions within reagent trays are numbered from the end furthest from the handle (position 1) to the position nearest the handle (position 9).

For the BOND RX system, researchers are able to purchase research reagent systems to utilize open research detection capabilities.

For instructions on loading reagents into the processing module, see [4.1.4 Loading the Reagents](#).

2.6.5 Reagent Systems and Containers

A range of reagent container types can be used in reagent trays.

2.6.5.1 Reagent Systems

Reagent systems are predefined sets of reagents in a reagent tray. The BOND RX system can use three types of reagent systems:

- User-configured BOND Research reagent systems
- BOND detection systems
- BOND cleaning systems

See [8.1 Reagent Management Overview](#) for further details on each of these.

Non-research reagent systems are treated as wholes, e.g. entire systems are registered and not the individual reagent containers that make up the systems. The reagent containers are sealed into the tray, and should not be removed or rearranged. For research reagent systems the component containers must be registered independently, but once this is done the systems are treated as units. When a reagent system is exhausted or expired, discard the complete tray and containers.

2.6.5.2 BOND Ready-To-Use Reagents

BOND ready-to-use reagents use containers that fit into the reagent trays. These reagents are provided in concentrations optimized for the BOND RX system, so require only registration and opening before use. Not all BOND ready-to-use reagents that are available on the BOND Clinical System are available on the BOND RX system. Please check the reagent inventory screen for availability prior to purchasing.

The containers hold different volumes of reagent, from 3.75 mL up to 30 mL, depending on the reagent type.

2.6.5.3 Open Containers

Open containers are empty, clean, containers for holding a user-supplied reagent (for example a primary antibody). They are available in 7 mL and 30 mL sizes. Open containers can be used with one reagent only, and can be refilled so that each container delivers a maximum of 40 mL of reagent (see [8.3.2.4 Refilling an Open Reagent Container](#)).

Only BOND open containers should be used on the BOND RX system – do not attempt to use other containers (except titration containers) for user-supplied reagents.

2.6.5.4 Titration Containers

Special purpose titration containers are also available (see [14.2.1.4 Titration Kit](#)). These include a 6 mL removable insert so that the reagent in a container can be easily changed, for example during concentration optimization. Like open containers, each titration container can be refilled and used to deliver up to 40 mL of reagent. Five inserts are provided per container in the BOND titration kit, available from Leica Biosystems.

The kits can be re-used for different antibodies and are designed with minimal dead volume to preserve reagent.

2.7 Relocating a Processing Module



WARNING: Contact customer support to relocate the processing module over a large distance or to transport for repair or disposal. The processing module is heavy and is not designed to be moved by the user.



CAUTION: Do not block the ventilation openings located on the back cover of the processing module. Also, do not cover the ventilation openings located on the syringe door (BOND RX^m).

If relocating a BOND RX Processing Module a short distance, consider the following points before proceeding:

- Ensure the flooring is able to withstand the weight of the processing module, see [18.2 Physical Specifications](#) in [18 Specifications](#) for dimensions, and consult with local requirements before moving.
- Evaluate the electromagnetic environment prior to operation of the processing module for interference.
- Do not use a BOND RX Processing Module in close proximity to sources of strong electromagnetic radiation. For example unshielded intentional RF sources, which may interfere with proper operation.
- Do not lift a BOND RX Processing Module with a forklift.
- Use only the power cord supplied, and ensure that the operator can access the power connection that the cord is plugged into.
- Ensure the power cord and Ethernet cable are disconnected before moving.
- Ensure adequate ventilation.
- Empty waste containers before moving.
- Ensure you unlock all four wheels on the BOND RX Processing Module (or trolley, for a BOND RX^m) before moving, and re-lock when in the new location.

2.8 Instrument Decommissioning and Disposal

The instrument, including parts and associated accessories used, must be disposed of according to applicable local procedures and regulations. Dispose of any reagents used with the instrument in accordance with the recommendations of the reagent manufacturer.

Clean and decontaminate in accordance with local procedures and regulations before returning or disposing of the instrument or parts and accessories.

In the EU, all electronic waste must be disposed of in accordance with Waste Electrical and Electronic Equipment (2012/19/EU). In regions outside of the EU, follow local procedures and regulations for the disposal of electronic waste.

If you need assistance, contact your local Leica Biosystems representative.

3

Software Overview (on BOND RX Controller)

This chapter is designed to help you become familiar with the general features of the BOND RX software. For instructions to use the software to run the processing modules and manage slides, studies and reagents, see the relevant chapters. See [10 Administration Client \(on BOND RX Controller\)](#) for instructions for the administration client.

- [3.1 System Architecture](#)
- [3.2 Starting and Shutting Down the BOND RX Software](#)
- [3.3 User Roles](#)
- [3.4 Research Client Interface Overview](#)
- [3.5 BOND RX-ADVANCE Dashboard](#)
- [3.6 Notifications, Warnings, and Alarms](#)
- [3.7 Reports](#)
- [3.8 Help](#)
- [3.9 About BOND RX](#)
- [3.10 BOND RX Data Definitions](#)
- [3.11 Software Updates](#)

3.1 System Architecture

Users interact with the BOND RX software through two “clients” – in effect, two separate programs. These are the research client (or simply “the client”) and the administration client. The research client is for everyday operation – to set up reagents, protocols, and studies and slides in preparation for processing, and then to monitor and control runs on the processing module. The administration client is used to configure advanced settings that are rarely changed after initial setup. These include slide label configurations, hardware connections, and user accounts (see [10 Administration Client \(on BOND RX Controller\)](#)).

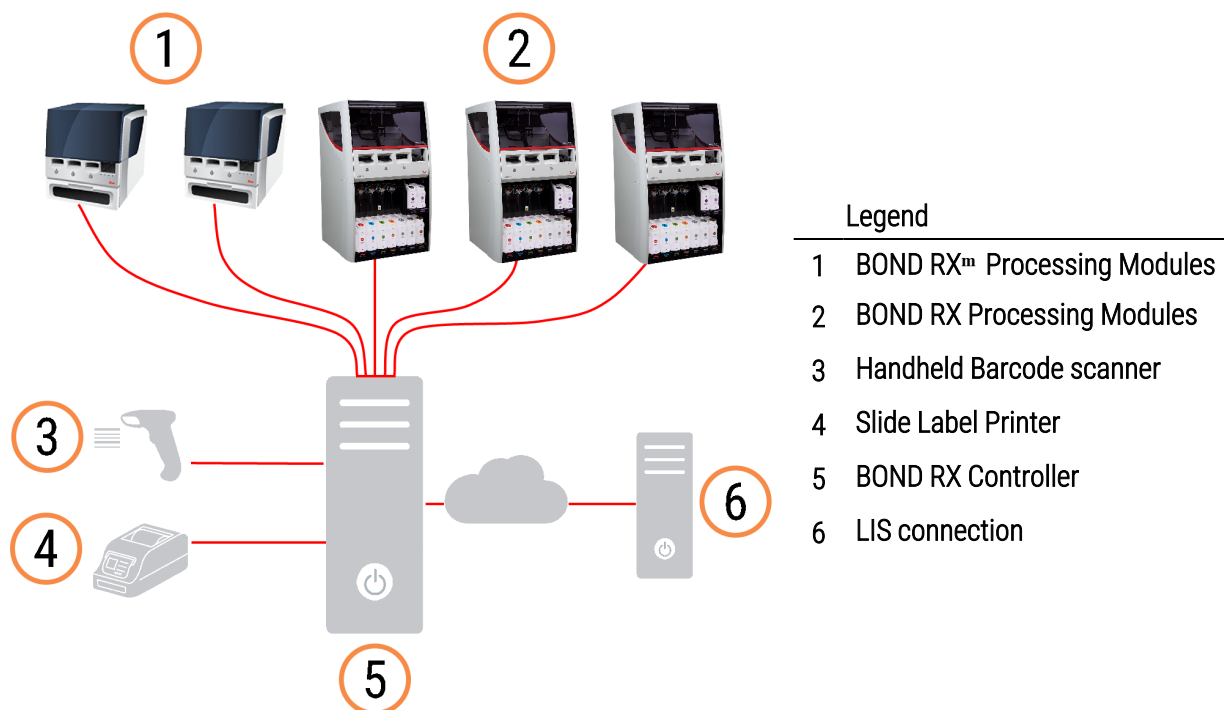
- [3.1.1 Single-Seat Configuration](#)
- [3.1.2 BOND RX-ADVANCE](#)

3.1.1 Single-Seat Configuration

Single-seat installations have just one “BOND RX controller”, which is the single point for user interaction with the BOND RX software (and through that, control of the processing modules). The BOND RX controller carries out all software processing for the system and maintains the system database, where study and slide information is held. It has a keyboard, mouse and monitor, and slide label printer and scanner attached.

There is a limit of five processing modules in a single-seat installation. If you need more processing modules, upgrade to BOND RX-ADVANCE.

Figure 3-1: Diagram of a single-seat installation



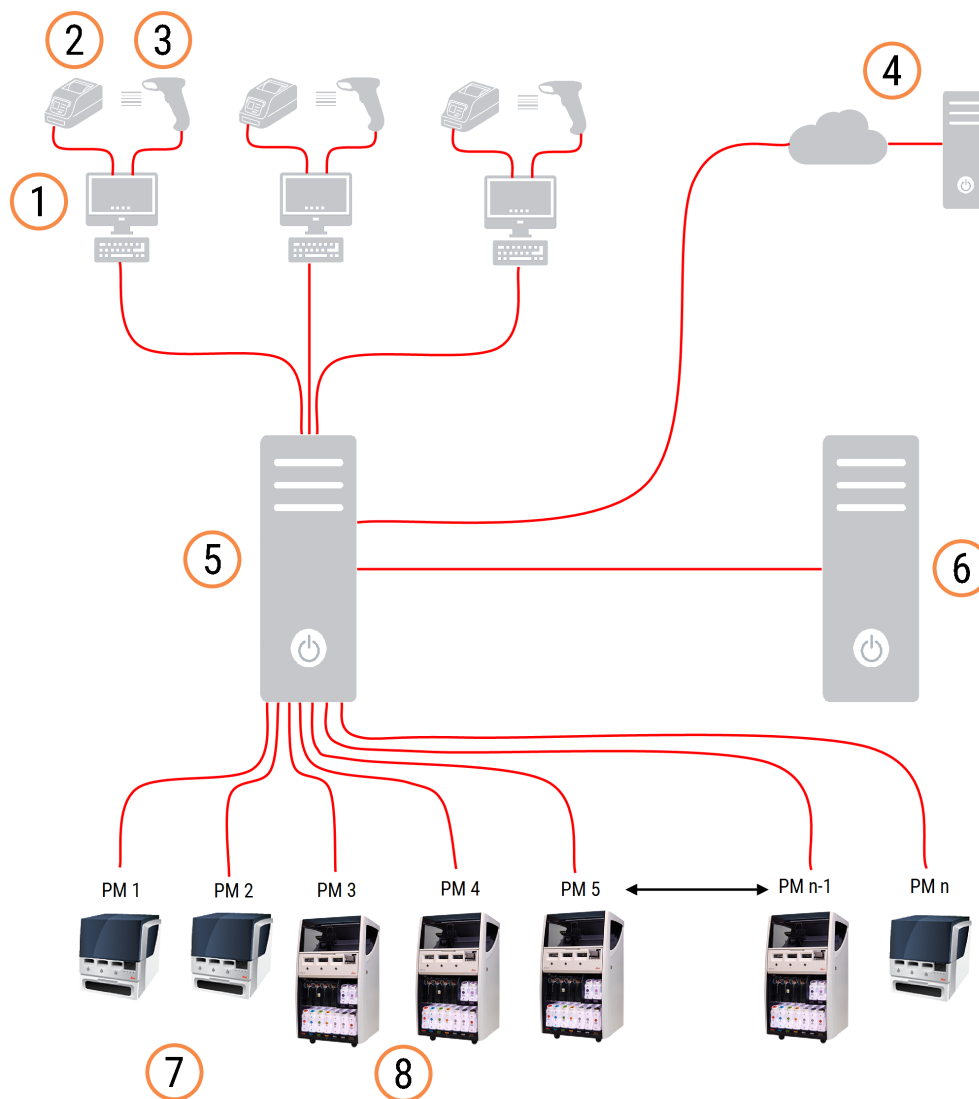
3.1.2 BOND RX-ADVANCE

BOND RX installations with more than five processing modules are configured as multi-seat BOND RX-ADVANCE installations. The BOND RX controller continues to perform all software processing for the entire system, but most input comes from BOND RX-ADVANCE terminals located near processing module work cells (called “pods” in the BOND RX software) that they control. Pods are defined in the administration client.

A monitor connected to the controller shows the “BOND RX Dashboard”, which gives a summary of the real-time status of every processing module in the system (see [3.5 BOND RX-ADVANCE Dashboard](#)). The dashboard can also be connected to a dedicated terminal if requested. The administration client can be run from any terminal.

Some laboratories may have a secondary controller, which backs up all BOND RX data in real time and can be switched to, in the case that the primary controller malfunctions. For details of how to do this, see [16.2 Switching to the Secondary Controller](#).

Figure 3-2: Diagram of a BOND RX-ADVANCE installation – the BOND RX-ADVANCE terminals control the processing modules in the pods, via the BOND RX-ADVANCE controller.



Legend

- | | |
|--------------------------------------|-------------------------------------------|
| 1 BOND RX-ADVANCE terminals | 6 BOND RX-ADVANCE Secondary Controller |
| 2 Slide Label Printers | 7 BOND RX [™] Processing Modules |
| 3 Barcode Scanners | 8 BOND RX Processing Modules |
| 4 LIS connection | |
| 5 BOND RX-ADVANCE Primary Controller | |

3.2 Starting and Shutting Down the BOND RX Software

3.2.1 Starting the BOND RX software

You can start the BOND RX software before or after starting any connected processing modules. To start the software:

- 1 **Single-seat:** if necessary, start the BOND RX controller and log on to Windows® as user “BONDUser”. When the system is new, no initial password is configured. However, if a password has been configured, see the laboratory manager for the details.

BOND RX-ADVANCE: if necessary, start the BOND RX-ADVANCE controller. The dashboard should open automatically (if not, double-click the **BONDDashboard** shortcut on the Windows desktop. Press <F11> to set Internet Explorer to full screen mode).

Start the terminal you need and log on to Windows as user “BONDUser”.

- 2 Double-click the appropriate desktop icon to start the research client or administration client (or both – they can run concurrently).
- 3 Enter your BOND RX username and password.

If you are opening the research client in a BOND RX-ADVANCE system you can select the pod to connect to.



The BOND RX-ADVANCE research client remembers the last pod selected.

You can change your password on the logon dialog at any time. Follow laboratory procedures for frequency of password changes and password strength. The BOND RX software requires that passwords are 4–14 characters and include at least one number.

- 4 Click **Log on**.

The system displays the research client screen or the administration client screen as selected. The title bar displays the username of the user that is currently logged on. If you take over from another user you should log off that user and log on again with your own username. For BOND RX-ADVANCE, the title bar also displays the currently selected pod.



WARNING: Because the BOND RX software is controlling important hardware and storing sensitive data, do not run other applications on the BOND RX controller – this will invalidate the BOND RX System warranty. Do not use the BOND RX controller for general purpose computing.

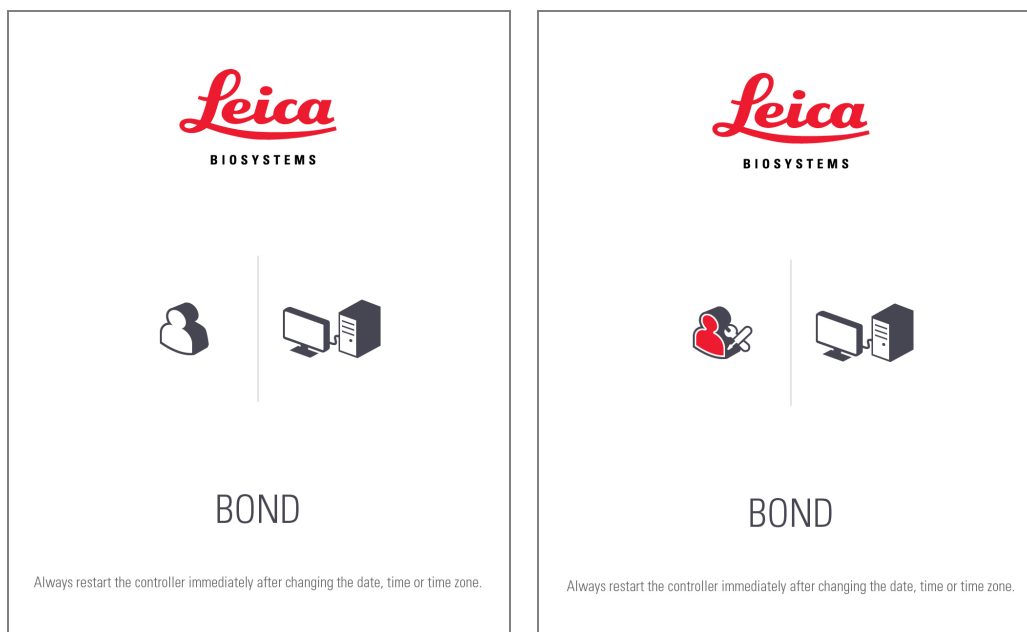
Desktop Backgrounds

Different Windows desktop backgrounds are used to distinguish between the type of Windows user currently logged on, and the role of the currently connected controller or terminal.

Single-seat

Normally, you would see the “Controller BONDUser” background, but if a service engineer is on site, you may see the “Controller BONDSERVICE” background. See [Figure 3-3](#).

Figure 3-3: BOND RX desktop backgrounds: “Controller BONDUser” and “Controller BONDSERVICE”



BOND RX-ADVANCE

On the BOND RX-ADVANCE desktop backgrounds, the connected controller’s or terminal’s icon changes according to its role. See the examples in [Figure 3-4](#).

Figure 3-4: Terminal, Standalone Controller, Primary Controller and Secondary Controller icons




You will also see different icons that represent the type of user. See [Figure 3-5](#).

Figure 3-5: BONDUser, BONDService, BONDControl and BONDDashboard icons



3.2.2 Shut down the BOND RX software

To shut down the research client or administration client, click the **Log out** icon  on the function bar.

You can shut down the research client while a run is in progress if you need to change users. Do not leave the processing module running without the client open for any length of time, however, as you will not see any alarms or warnings.

Never shut down the BOND RX controller during a run. If closing down the BOND RX system completely, you can shut down the software before or after turning off the processing modules.

3.3 User Roles

There are three user roles in the BOND RX system:

- **Operator:** can update reagent inventory, create studies and slides, start and control staining runs, create and edit researchers, and generate reports.
- **Supervisor:** create and edit protocols, reagents, and panels.
- **Administrator:** has access to the administration client to manage BOND RX users and configure system-wide settings.

Users can have multiple roles. Supervisors automatically get operator roles. Only users with the administrator role can run the administration client, and only users with operator or supervisor roles can run the research client.

Users are created and their roles set on the administration client **Users** screen (see [10.1 Users](#)).



The username of the currently logged-in user is displayed in the client window's title bar.

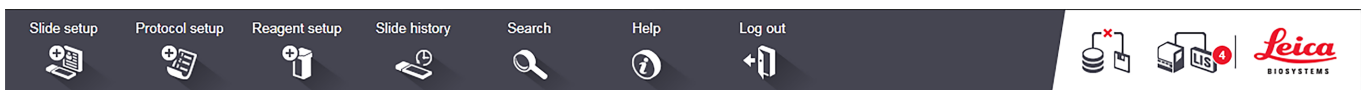
3.4 Research Client Interface Overview

At the top and at the left of the research client screen there are features that are common to all pages of the software. This section describes these features, and also describes general features of the software.




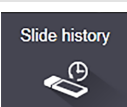
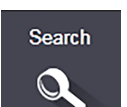
- [3.4.1 Function Bar](#)
- [3.4.2 Processing Module Tabs](#)
- [3.4.3 Sorting Tables](#)
- [3.4.4 Date Format](#)







3.4.1 Function Bar

The function bar is located at the top of the BOND RX software screen, and provides quick access to the main sections of the BOND RX software.



Click on an icon on the function bar to go to a screen or perform a specific function as described in the following table.

Icon	Screen displayed (or function performed)	Purpose
	Slide setup	Create studies and set up slides in the BOND RX software. For more information see 6 Slide Setup (on BOND RX Controller) .
	Protocol setup	Edit and manage your protocols. For more information refer to 7 Protocols (on BOND RX Controller) .
	Reagent setup, Reagent inventory, and Reagent panels (3 tabs)	Configure new reagents, manage reagent inventory, and create reagent panels (sets of markers used to speed up slide creation). For more information refer to 8 Reagent Management (on BOND RX Controller) .
	Slide history	Display details of slides that have been run on the BOND RX system, view details of individual slides, runs and studies, and generate a wide range of reports. For more information refer to 9 Slide History (on BOND RX Controller) .
	Search	Identify slides, reagent containers and reagent systems by scanning the barcode or manually entering the slide ID or reagent ID. A unified search dialog is used where the search content (slide or reagent) is automatically identified by the system. See 6.5.6 Manually Identifying a Slide or 8.1.1.3 Reagent Identification for further information.

Icon	Screen displayed (or function performed)	Purpose
	Help	Open this user manual.
	Log out	Log out of the client.
	Backup failed	A database backup has failed to complete successfully. For more information refer to 10.5.3 Database Backups .
	LIS not connected	An LIS module is installed, but is currently not connected to the LIS. For more information refer to 11.3 LIS Connection and Initialization .
	LIS connected	An LIS module is installed and is currently connected to the LIS. For more information refer to 11.3 LIS Connection and Initialization .
	LIS notifications	The number of outstanding LIS notifications. For more information refer to 11.4 LIS Notifications .

At the top right of the screen is the Leica Biosystems logo. Click the logo to display the **About BOND RX** dialog. See [3.9 About BOND RX](#).

At the top right of the screen, warning and status icons may be displayed. See [11 LIS Integration Package \(on BOND RX Controller\)](#) and [10.4.2 Audit Trail](#).

3.4.2 Processing Module Tabs

Tabs on the left-hand side of the interface open **System status**, **Protocol status** and **Maintenance** screens for each of the processing modules in the pod that the client is connected to. The tabs themselves display some information about the current state of each processing module (see [5.1.1 Processing Module Tabs](#)).

Figure 3-6: Processing Module Tab (BOND RX^m)



System status screens show the state of each processing module, while the **Protocol status** screens show the progress of protocols being run. The **Maintenance** screen has commands for a range of maintenance operations.

3.4.3 Sorting Tables

Many screens in the BOND RX software display data in tables. Click on a column heading to sort by the values in that column. An upward triangle appears beside the heading to indicate the table is being sorted in ascending order (0-9 A-Z). Click again to sort in descending order; the triangle points down.

To sort on two columns, click the first column you want to sort by, then hold the <Shift> key and click on the second column. The order of values in the first column does not change, but where there are multiple rows with the same column-one value, the rows are ordered by the values in the second column.

You can also resize column widths and drag columns to new positions in the table.

Any changes you make to table sorting, also column widths and positions, are retained until you log out.

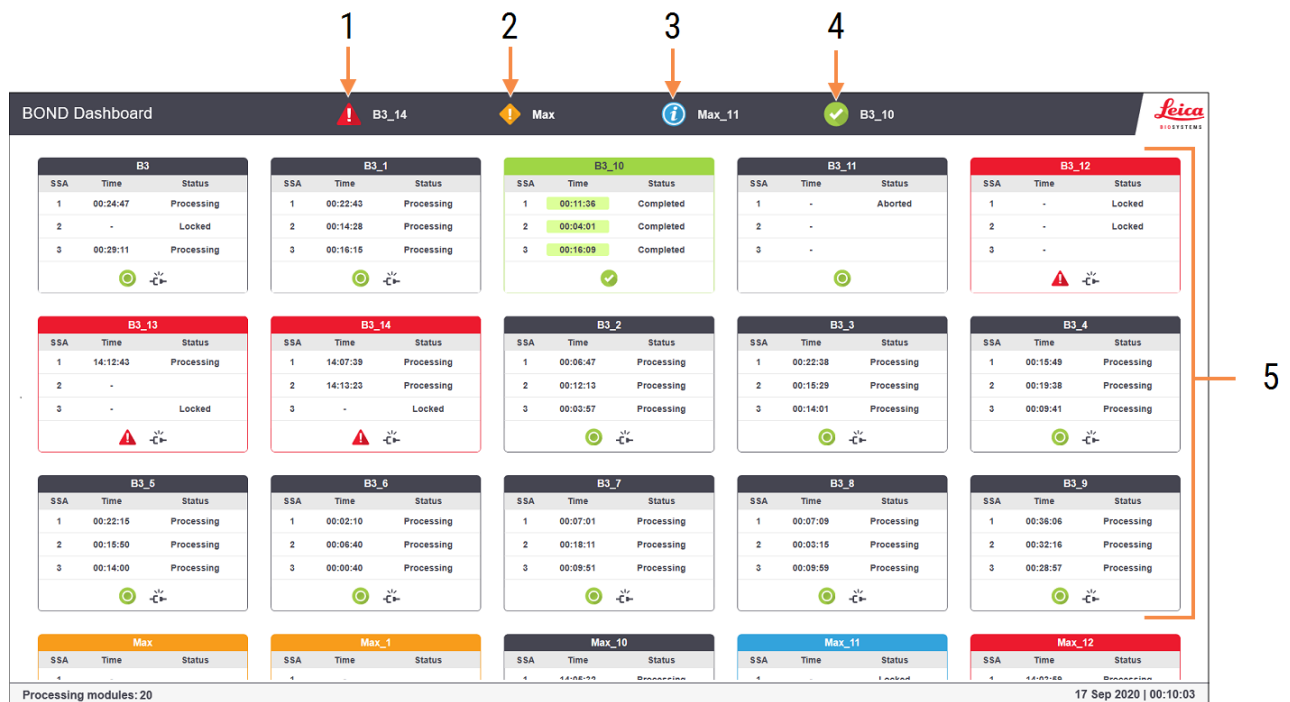
3.4.4 Date Format

For single-seat installations, dates and times in the software and reports use the formats set in the BOND RX controller operating system. For BOND RX-ADVANCE installations, the formats set in the terminals are used. Short and long date formats should have maximum lengths of 12 and 28 characters respectively.

3.5 BOND RX-ADVANCE Dashboard

For BOND RX-ADVANCE installations the BOND RX dashboard is displayed on a monitor connected to the controller or terminal. It gives a real-time status summary for all the processing modules in the system.

Figure 3-7: The BOND RX Dashboard









Legend

- | | |
|-----------------------------------------|-----------------------------------------------------------------------------------|
| 1 Processing modules with alarms | 4 Processing modules with finished runs |
| 2 Processing modules with warnings | 5 Individual processing module panes, showing status of slide staining assemblies |
| 3 Processing modules with notifications | |

At the top of the screen are four icons showing processing modules with (from left to right) alarms, warnings, notifications, and finished runs. If there is more than one processing module in a category, the icons cycle through them in sequence.

Beneath the top row are panes for each processing module in the system, ordered alphabetically by name (set in the administration client). The panes show the status of each of the three slide staining assemblies on the processing modules, plus any general status indicators applying to the modules as a whole.

Dashboard icons

Icon	Description
	The processing module has an alarm.
	The processing module has a warning.
	The processing module is functioning normally. The timestamp has a white background (<code>00:14:28</code>).
	The run has successfully completed on at least one tray on the processing module, and the tray is ready to unload. The timestamp has a green background (<code>00:11:36</code>).
	The processing module has a notification.
	The processing module has been disconnected.

Processing modules with warnings, notifications or finished runs appear both in the appropriate position at the top of the display, and as individual panes in the alphabetical listing below.

3.5.1 Slide Staining Assembly Status

The status of each slide staining assembly is shown on the processing module panes. There are three status categories:

- **Locked** – shown when the slide tray is locked. No time is shown.
- **Processing** – processing has started on the tray. The **Time** column shows the time left to run, in hours, minutes and seconds.
- **Completed** – processing has finished. The **Time** column shows the time since the run finished, in hours, minutes and seconds, and has a green background.

If no tray is locked the row is blank.

You cannot interact with the dashboard. If the dashboard displays a message that a PM needs attention, you need to interact via the BOND RX-ADVANCE terminal.

3.6 Notifications, Warnings, and Alarms

The BOND RX system has three alert levels: notification, warning, and alarm. Each alert is indicated by an icon that appears on the **System status** screen over or adjacent to the item subject to the alert message. A corresponding alert icon may also appear on the processing module tab to provide an indication irrespective of the currently visible screen (refer to [5.1.1 Processing Module Tabs](#)). In BOND RX-ADVANCE, alerts also appear on the dashboard (see [3.5 BOND RX-ADVANCE Dashboard](#)).

Right-clicking an alert icon and selecting **Attention message** launches a dialog that details the alert condition.

The three alert levels and their associated icons are described below.



Steady

Notification

Provides information about a condition that may require action now or later, in order to start a run or to avoid a later delay in processing.



Steady

Warning

Action is required now, possibly to avoid a delay in processing. Delays in processing can compromise staining.



Flashing

Alarm

Action is urgently required. If the processing module was processing slides it has been paused and cannot resume until you rectify the alert condition. Delays in processing can compromise staining.



WARNING: Always read warning and alarm messages as soon as you see the icons (especially when a run is in progress). Quick response may avoid compromising slide staining.

It is also advisable to act on notifications that occur during runs as soon as possible.

3.7 Reports

The BOND RX software generates a number of reports. These open into a “BOND RX Report Viewer” in a new window. General information such as time, place and the processing module that the report refers to is provided in report headings. Report page footers show the time and date each report was generated, and page numbers.

Some reports, especially those containing study, slide or reagent information, may take minutes to generate, particularly in laboratories with many processing modules and/or high turnover.

The BOND RX report viewer has a small range of navigation, viewing and output options. As well as opening a standard print dialog to select and configure a printer, or to select which pages to print, you can export reports in a range of formats, including PDF, XLS, CSV and text.

You can use various keyboard shortcuts for navigation, such as **Page Up**, **Page Down**, **Home** (first page) and **End** (last page). Other functions are also available via keyboard shortcuts, for example **Ctrl-F** displays the Search dialog, **Ctrl-S** opens the Save dialog, and **Ctrl-P** opens the Print dialog.

BOND RX reports are documented in the following sections:

- [5.3.1 Maintenance Report](#)
- [6.7 Slide Setup Summary Report](#)
- [7.5 Protocol Reports](#)
- [8.3.4 Inventory Details Report](#)
- [8.3.5 Reagent Usage Report](#)
- [9.4 Run Events Report](#)
- [9.5 Run Details Report](#)
- [9.6 Study Report](#)
- [9.8 Slides Summary](#)
- [9.10 Brief Slide History](#)
- [3.9.1 Service Log](#)

It is also possible to export slide information in a CSV (comma-separated values) file format. See [9.9 Export Data](#).

3.8 Help

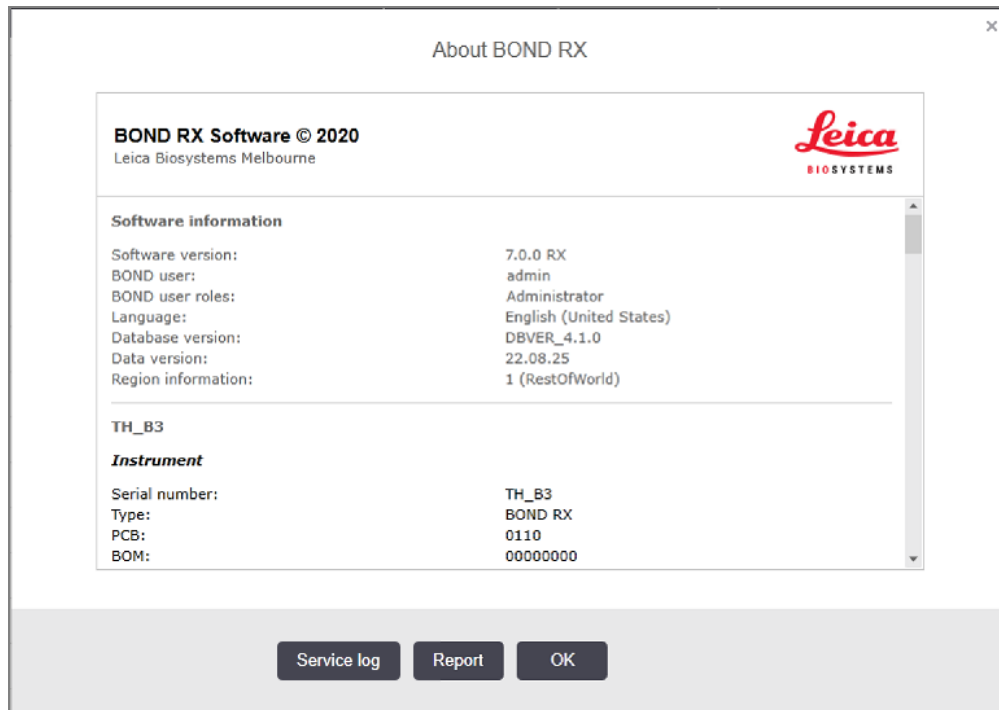


The **Help** icon on the function bar in both the research and administration clients opens this user manual.

3.9 About BOND RX

Click the Leica Biosystems logo at the top right of the screen to view the **About BOND RX** dialog, which lists system information.

Figure 3-8: About BOND RX dialog



Much of the information in the **About BOND RX** dialog is principally of interest to service personnel, however laboratory staff may find the initial information group useful, especially during discussions with customer support.

The information contained in the initial group is as follows:

- Software version: the software release version number.
- BOND RX user: the username of the current user.
- BOND RX user roles: the user roles of the current user.
- Language: the current language.
- Database version: the version of database (refers to the database structure).
- Data version: the version of data loaded in the database.
- Region information: the world region the system is configured for (set during installation).

You can save the information in the dialog to a text file – click **Report** and select a location to save the file.

3.9.1 Service Log

In the administration client you can generate service log reports from the **About BOND RX** dialog. Typically this would be done at the request of a service representative. To create a service log:

- 1 Click **Service log** in the **About BOND RX** dialog (see [Figure 3-8](#)).
- 2 Select either:
 - the serial number of a specific processing module,
 - ***System*** to report on software or controller events in the BOND RX system, or
 - ***LIS*** for events related to the LIS system.
- 3 Select a time period for the report, or click **Last seven days**.
- 4 To generate the report, click **Generate**. The report appears in the report viewer – see [3.7 Reports](#).
- 5 To export the service log to a CSV file, click **Export Data**.

Service log ✕

Select processing module

Serial N°:

Name:

Type:

Time span

From:

To:

[Last seven days](#)

3.10 BOND RX Data Definitions

The BOND RX controller stores data definitions that contain all reagent and protocol details for the entire system. Default protocols and details of Leica Biosystems reagents and reagent systems are also included.

3.10.1 Data Definitions Updates

Leica Biosystems periodically distributes data definitions updates on the web site, e.g. to add newly released reagents. See [10.4 BXD](#) for instructions to update the data definitions.



When you update the data definitions, you must only use update files that have the **.bxd** file extension and are for the correct region.

Check your current data version in the **About BOND RX** dialog. To view this dialog, click the Leica Biosystems logo at the top right of the BOND RX software screen. Also see [3.9 About BOND RX](#).

3.11 Software Updates

Leica Biosystems may release software updates as the BOND RX system continues to develop. The updates may be to the main software or to the database that contains the default protocols, reagents and reagent systems.

The version number of the current software version can be found in the **About BOND RX** dialog (refer to [3.9 About BOND RX](#)). The data version is also displayed in the **About BOND RX** dialog.

4

Quick Start

This chapter is designed to take you on a guided tour of your first individual run with the BOND RX system. In it we create a sample study and configure and process four slides, testing with BOND ready-to-use primary antibodies *CD5, *CD3, *CD10, and *Bcl-6.

For BOND RX and BOND RX^m, the default protocol and detection system for these antibodies is *IHC Protocol F and BOND Polymer Refine Detection System (DS9800).

The procedures described are also valid for ISH probes and protocols (simply swap the antibody for a probe and replace IHC protocols with ISH protocols).

4.1 BOND RX and BOND RX^m

Before you start you should be familiar with the relevant sections of the [2 Hardware](#) and [3 Software Overview \(on BOND RX Controller\)](#) chapters of this manual.

- [4.1.1 Preliminary Checks and Startup](#)
- [4.1.2 Protocol and Reagent Checks](#)
- [4.1.3 Setting Up Slides](#)
- [4.1.4 Loading the Reagents](#)
- [4.1.5 Running the Protocol](#)
- [4.1.6 Finishing](#)

4.1.1 Preliminary Checks and Startup

Carry out the following steps before beginning a run:

- 1 Ensure that the processing module is clean and that all maintenance tasks are up to date (see [12.1 Cleaning and Maintenance Schedule](#)).

Daily pre-run tasks are:

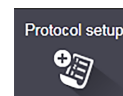
- a Check bulk waste containers are no more than half full; on the BOND RX^m, use the white horizontal line on the container label as a guide to the half-full level (see [Figure 12-3](#)).
 - b Check bulk reagent containers have an adequate volume of the correct reagent.
- 2 Check wash blocks and mixing station – clean or replace if necessary.
 - 3 Check that the slide labeler has an adequate supply of labels.
 - 4 If the processing module and controller (and terminal, for BOND RX-ADVANCE) are not on, turn them on now.
 - 5 When the controller or terminal is running, start the research client.
 - 6 Once the software has started, check the **Status** screens to ensure there are no processing module notifications. Rectify before attempting to run any slides.
 - 7 Power up the slide labeler.

4.1.2 Protocol and Reagent Checks

You should check that the protocols and reagents you are going to use in the run are set up in the software.

To check the protocols:

- 1 Select the **Protocol setup** icon (shown at the right) on the function bar.
- 2 Check that “*IHC Protocol F” is listed in the table.



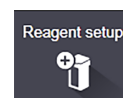
If the protocol is not listed select **All** in the **Preferred status** filter at the bottom of the screen (see [7.2 Protocol Setup Screen](#)).

- 3 Select the protocol in the table, click **Open**, and note the preferred detection system in the **Edit protocol properties** dialog; **BOND Polymer Refine Detection**.

Make sure that the protocol is selected as **Preferred** near the top of the dialog (you need to be logged on with a supervisor user role to make the protocol preferred, if it is not).

To check the reagents:

This check assumes that you have stock of the required antibodies and detection system, and that these have been registered in the BOND RX reagent inventory. See [8.3.3 Registering Reagents and Reagent Systems](#) for more information.



- 1 Select the **Reagent setup** icon (shown at the right) on the function bar.
- 2 On the **Setup** tab select **Primaries** as **Reagent type**, **Leica Microsystems** as **Supplier**, and **All** for **Preferred status** in the filters at the bottom of the screen.
- 3 Locate each of the antibodies that we need (*CD5, *CD3, *CD10, and *Bcl-6) and double-click to open the **Edit reagent properties** dialog:
 - a Click **Restore factory default protocols** (you need to be logged on with a supervisor user role to restore factory defaults). This ensures that the default staining protocol, *IHC Protocol F, and default pretreatment protocols are set.
 - b Ensure that the reagent is checked as **Preferred** (you need to be logged on with a supervisor user role to make the reagent preferred, if it is not).
 - c Click **Save**.
- 4 Now go to the **Inventory** tab and select **Reagent containers** as **Package type**, **Primaries** as **Reagent type**, **In stock** for **Inventory status**, **Leica Microsystems** for **Supplier** and **Preferred** for **Preferred status** in the filters at the bottom of the screen.

All the antibodies we need should appear with the volumes available.

Make sure that there is sufficient volume for each antibody.
- 5 On the same tab, select **BOND detection systems** as **Package type** and **In stock** for **Inventory status**. Check that the preferred detection system, **BOND Polymer Refine Detection** is listed in the table, and that there is enough volume (see [8.3.1.1 Reporting Volume for Detection Systems](#)).

4.1.3 Setting Up Slides

This section describes the processes of telling the BOND RX system the details it needs to stain the slides and of physically placing slides into the processing module.

The software operations in this section are carried out from the **Slide setup** screen. To display this screen, click the **Slide setup** icon on the function bar.



See subsections:

- [4.1.3.1 Entering Study Details](#)
- [4.1.3.2 Entering Slide Details](#)
- [4.1.3.3 Controls](#)
- [4.1.3.4 Labeling Slides](#)
- [4.1.3.5 Loading Slides](#)

4.1.3.1 Entering Study Details

First we must create a “study” in the software. For our example the study name is My Study, study ID 3688, with Smith the researcher.

- 1 Click **Add study** in the **Slide setup** screen. The software displays the **Add study** dialog.

Figure 4-1: The **Add study** dialog

- 2 Click in the **Study ID** field and type “3688”.
- 3 Click in the **Study name** field and type “My Study”.
- 4 Click **Manage researchers** to open the **Manage researchers** dialog. There, click **Add** to open the **Add researcher** dialog and type “Smith” in the **Name** field. Ensure that the **Preferred** box is checked. Click **Save**.
- 5 Select “Smith” and click **OK** in the **Manage researchers** dialog.
- 6 Select 150 µL dispense volume as the study default. For BOND RX™, this setting can be overridden during slide setup if you wish.
- 7 Select *Dewax or *Bake and Dewax in the **Preparation protocol** field to set a default preparation for slides in the study. This setting can be overridden during slide setup if you wish.
- 8 Click **OK** to close the **Add study** dialog – the table on the left of the **Slide setup** screen displays the new study.

For more information on working with studies, see [6.3 Working with Studies](#).

4.1.3.2 Entering Slide Details

At the next stage we create “slides” in the software for each of the four physical slides:

- 1 Select our new study ID 3688 in the study list on the left of the screen.
- 2 Click **Add slide** to display the **Add slide** dialog.

Figure 4-2: The **Add slide** dialog

- 3 Optionally, add a comment specific to this slide.
- 4 Ensure **Test tissue** is selected as the **tissue type**.
- 5 Select a dispense volume suitable for the processing module and tissue size (see [6.5.8 Dispense Volumes and Tissue Position on Slides](#)).

We will assume the slides will be processed on a BOND RX, so set dispense volume to 150 µL.

- 6 Select **Single** and **Routine** in **Staining mode**.
- 7 Click **IHC** to specify the IHC process.
- 8 Select ***CD5 (4C7)** from the **Marker** list.

In the **Protocols** tab, the software automatically enters the preparation protocol set for the study, and the default staining and retrieval protocols for *CD5.

9 For Single staining, you should generally leave the default of **Auto** for the Unique Product Identifiers (UPIs) on the left side of the dialog. However, if you want to select a specific lot number for a specific slide (e.g. for lot-to-lot validation), select from the drop-down list in the following fields:

- **Marker UPI** – UPI of the reagent container for the marker
- **Detection System UPI** – UPI of the Detection System.

For slides to be processed on the same run (on BOND RX^m and BOND RX), either the UPIs must be the same, or **Auto** must be selected.

10 Click **Add slide**.

The slide is added to the slide list on the right of the **Slide setup** screen. The **Add slide** dialog remains open.

11 Repeat steps 8–10 three times and select ***CD3 (LN10)**, ***CD10 (56C6)** and ***Bcl-6 (LN22)** as the marker in step 8.

12 After all slides have been added, click **Close** to close the **Add slide** dialog.

Review the details in the slide list.

Figure 4-3: Four slides configured in the **Slide setup** screen

1	00000002 *CD5	*IHC F *D *--- *H2(20)
2	00000003 *CD3	*IHC F *D *--- *H2(20)
3	00000004 *CD10	*IHC F *D *--- *H2(20)
4	00000005 *Bcl6	*IHC F *D *--- *H2(20)

If you need to change details for a slide, double-click the slide to open the **Slide properties** dialog, change the details as required, then click **OK**.

For more information on working with slides, see [6.5 Working with Slides](#).



You can use **panels** to quickly add a number of slides that you commonly use. For an explanation of panels and how to create and use them, see [8.5 Reagent Panels Screen](#).

4.1.3.3 Controls

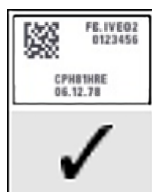
Always use controls on the BOND RX system. We strongly recommend placing appropriate control tissue on the same slides as test tissue. In addition to this, you can create a separate study specifically for control slides. See [6.2 Working with Controls](#) for further discussion.

4.1.3.4 Labeling Slides

You are now ready to print slide labels and attach them to the slides:

- 1 Click **Print labels** from the **Slide setup** screen.
- 2 In **Slide labels to print**, select the appropriate option, then click **Print**.
The labels are printed.
- 3 Ensure the frosted area of the slide (where the label will be applied) is dry, then apply the label with the slide ID or barcode aligned parallel with the end of the slide. The label should be right-side-up when the slide is held with the label at the top.

Figure 4-4: Correctly applied label



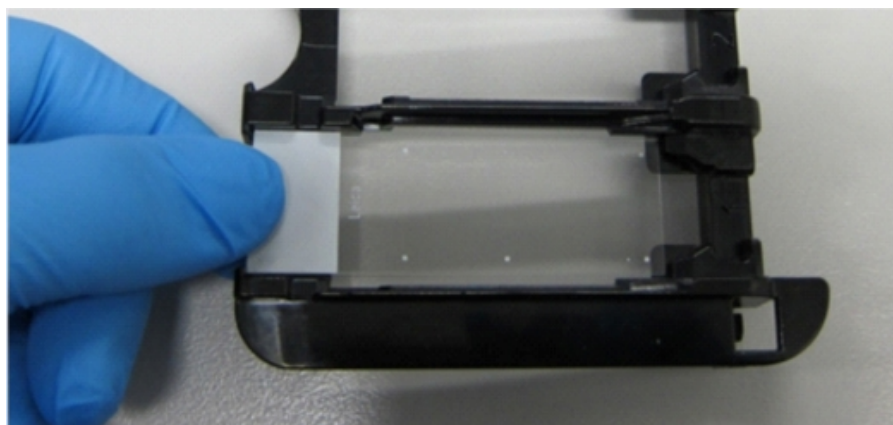
For more information, see [6.6 Slide Labeling](#).

4.1.3.5 Loading Slides

Load slides as follows:

- 1 Hold the slide by the label end with the sample uppermost.
- 2 Orient the slide over an empty position on the slide tray, with the label end of the slide over the indent in the side of the tray (see [Figure 4-5](#)). Place the slide down so it sits in the recessed position in the tray.

Figure 4-5: Positioning a slide in a slide tray



- 3 Hold a Covertile by the tail and lay it on the slide, fitting the key on the neck of the Covertile into the recess in the slide tray (circled in [Figure 4-6](#)). With new-design Covertiles the word "Leica" imprinted on the Covertile should read correctly, showing that the Covertile is the correct way up.

Figure 4-6: Positioning a Covertile on a slide



- 4 When all slides and Covertiles are loaded into the tray, lift the tray and rest the end on the entrance to an empty slide staining assembly. Slide the tray as far as it will go into the module. The tray should slide in easily and audibly click when it is in place.

4.1.4 Loading the Reagents

Now the detection system (BOND Polymer Refine) and marker containers (for *CD5, *CD3, *CD10 and *Bcl-6) must be placed onto the processing module.



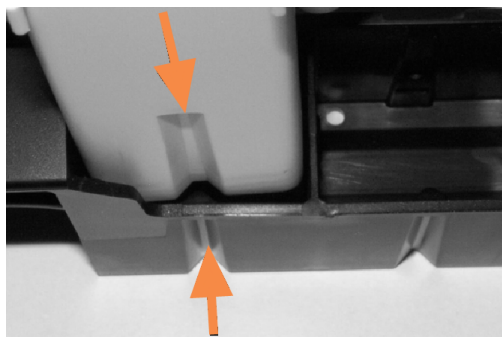
WARNING: Reagent containers can tip during transit, leaving reagent residue around the cap. Always wear approved eye protection, gloves and protective clothing when opening reagent containers.

To load reagents into the BOND RX or BOND RX^m Processing Module, do the following:

- 1 Place marker containers into reagent trays by aligning the grooves on the rear of the containers with the indents in the tray compartments. Press down until the containers click into place.

Marker containers can be placed in the spare compartments in detection system trays if you wish.

Figure 4-7: Reagent container in reagent tray

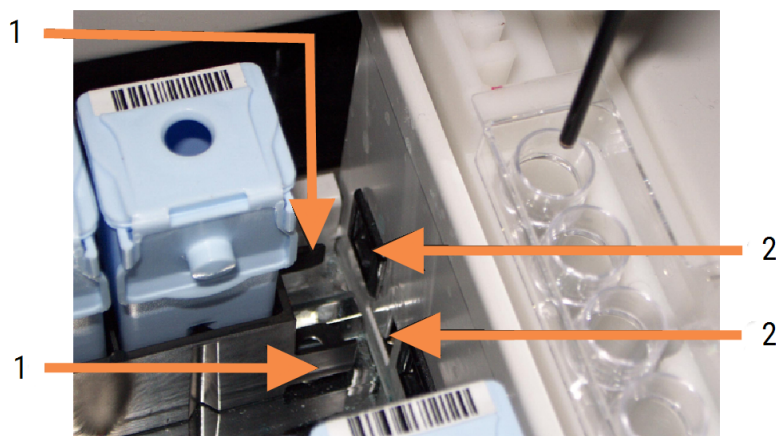


The arrows indicate the grooves in the reagent container and in the reagent tray.

- 2 Open all marker and detection system containers. Click open the lids and swing back until they clip into the tabs at the backs of the containers.
- 3 Ensure the top barcode labels are fully adhered to the containers – press down any labels that are lifting.
- 4 Wipe any moisture/condensation from the top barcode label.
- 5 Place the reagent trays on the reagent platform of the processing module. Use the guides on the platform to guide the trays correctly into the platform.

When the tray reaches the end of the platform it should engage the interlock. The tray LED turns green to indicate the tray is in position.

Figure 4-8: Inserting the reagent tray



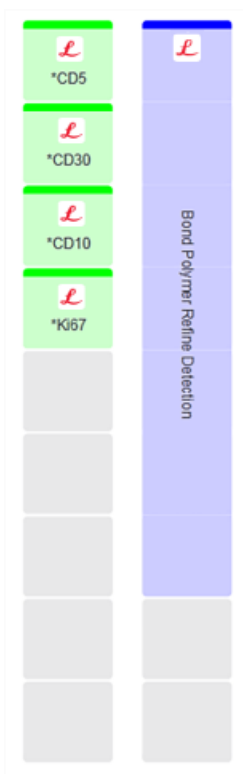
Legend

- 1 The tray's locking mechanism
- 2 Processing module's locking port

- In the software, click the processing module tab to display the **System status** screen.

The reagent column is displayed in a lighter color with a dark border to indicate that the tray is about to be imaged. The BOND RX system images the IDs on the reagents as soon as the main robot is available, then updates the icons for the reagents.

Figure 4-9: Reagent tray status as shown in the System status screen



If there are any problems with reagents, the software displays an attention icon on that screen. Right-click the icon to get more information (see [5.1.3.5 Fixing Reagent Problems](#)).



Reagent trays can be removed at any time while the tray LED is green. When a reagent in a tray will be required within 2 minutes the LED turns red, indicating that the tray is locked (see [2.2.6.5 Reagent Platform](#)).

4.1.5 Running the Protocol

With slides and reagents configured and loaded in the processing module, you are ready to start processing.

- 1 Ensure that the processing module lid is closed.
- 2 Press the Load/Unload button on the front cover beneath the loaded slide tray.

BOND RX or BOND RX^m locks the tray, and the slide tray LED should glow orange.




Listen as the slide tray locks – if there are any loud cracking or clicking sounds it is likely that Covertiles are out of position. In this case unlock the tray, remove, and check the slides and Covertiles.

- 3 As soon as the main robot is available, the BOND RX system images the slides.

If any of the required reagents are not available, the software displays an attention icon below the slide list. Right-click on the icon for more information.

- 4 Provided there are no unrecognized or incompatible slides, the slides are now ready for a staining run. The progress bar will be in the starting phase (refer to [5.1.6.2 Run Progress](#)) and the run status will be **Slides ready** (refer to [5.1.6.1 Run Status](#)).


Click  to begin running the protocol (or you can set the processing module to start later; see [5.1.8 Delayed Start](#)).

The system will schedule the run then the progress bar will switch to the processing phase and the run status will be **Proc (OK)**.



You should start only one run at a time, and then wait until that run has started/been scheduled before starting the next run. Wait for a short while after starting each run to confirm it has started successfully. If not, the run status is set to **Rejected/Slides ready**. See [5.1.6.1 Run Status](#).

While a run is being processed, the Load/Unload button for its slide staining assembly will not release the slide tray.

Click  below the tray on the **System status** screen to abandon the run (see [5.1.7 Starting or Stopping a Run](#)).

4.1.6 Finishing

When the processing run is finished, the processing module tab icon flashes (see [5.1.1 Processing Module Tabs](#)). If there were unexpected events during the run, the display text is red and the notification symbol will appear below the tray and on affected slides. If this happens, check the **System status** screen for attention icons and right-click on them to display information about the attention state. You should also inspect the Run Events Report (refer to [9.4 Run Events Report](#)) to see any other information about problems during the run.

When the run has finished:

- 1 Remove reagent trays.

Close reagent container lids firmly to prevent reagent evaporation, and immediately store the reagents as recommended on the label or reagent data sheet.

- 2 Generate the Run Event report (refer to [9.4 Run Events Report](#)).
- 3 Press the Load/Unload button and remove slide trays from the processing module.



Again listen for cracking or clicking sounds as the tray unloads. If you hear this inspect in and around the slide staining assembly for broken slides in the unexpected event that a misaligned slide has been crushed; if so contact customer support.

- 4 Place the slide tray on a flat, stable surface. Remove the Covertiles by holding down the label of the slide, then carefully putting pressure downwards on the neck of the Covertile to lift the end of the Covertile off the slide.



Do not slide the Covertile across the surface of the slide, as you may damage the tissue, making slide reading difficult.

- 5 Lift the Covertiles from the slides and clean them as described in [12.3 Covertiles](#).
- 6 Remove the slides and proceed with the next step in processing them according to your laboratory processes.

You can choose to rerun any slides (see [9.3 Slide Properties and Slide Rerun](#)).

This completes your first run on the BOND RX system.

5

Status Screens (on BOND RX Controller)

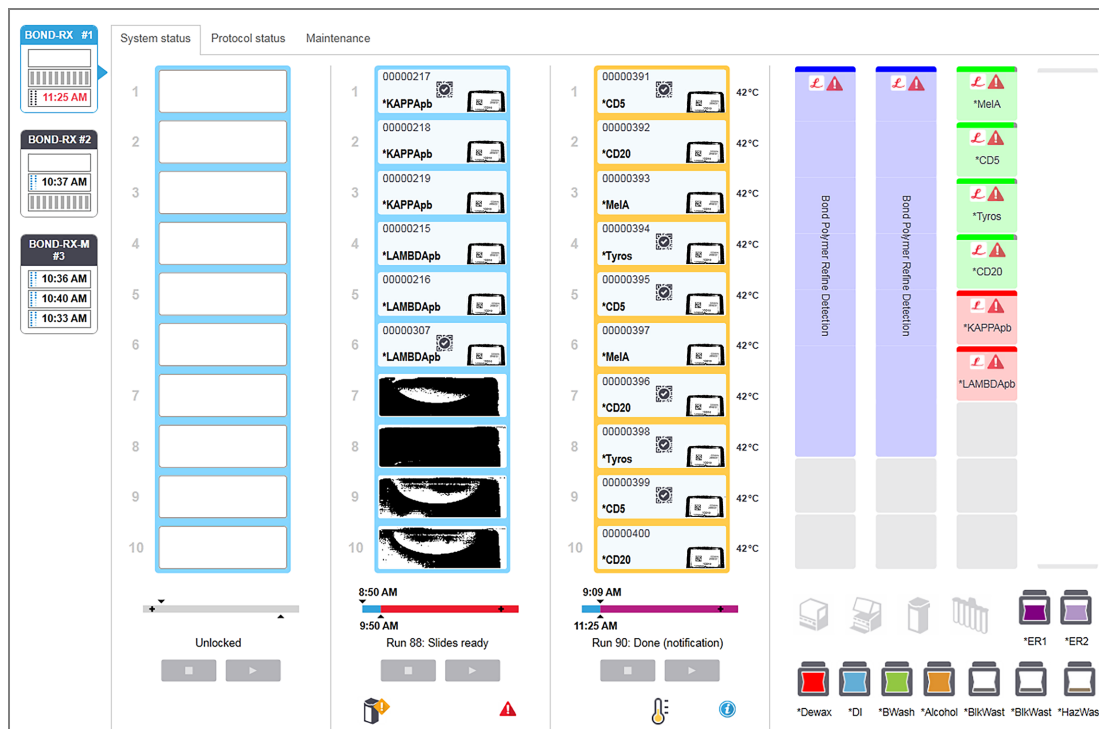
In the research client, each processing module has two status screens and a maintenance screen, selected from tabs at the top left of the window when a processing module has been selected from the left-hand tabs. The **System status** screen offers system control from a view that shows slide and reagent placement in the module. The **Protocol status** screen gives information on protocol progress for individual slides. The **Maintenance** screen has commands for a range of maintenance operations.

- [5.1 System Status Screen](#)
- [5.2 Protocol Status Screen](#)
- [5.3 Maintenance Screen](#)

5.1 System Status Screen

This screen allows you to control processing, and it displays the details of slide trays and reagents loaded, as well as displaying status of reagents, waste, and interlocks in the system.

Figure 5-1: The **System status** screen for a BOND RX Processing Module



The processing module tabs at the left of the status screens give a visual summary of the status of the associated processing module. Click on the tab to see the detailed status of the processing module.

For more information see:

- [5.1.1 Processing Module Tabs](#)
- [5.1.2 Hardware Status](#)
- [5.1.3 Reagent Status](#)
- [5.1.4 Slide Information](#)
- [5.1.5 On-Board Slide Identification](#)
- [5.1.6 Run Progress Indicator](#)
- [5.1.7 Starting or Stopping a Run](#)
- [5.1.8 Delayed Start](#)

5.1.1 Processing Module Tabs

The software displays a tab at the left of the screen for each processing module in the system (single seat) or in the pod that the client is connected to (BOND RX-ADVANCE). If there is not enough vertical space to display all the processing modules, scroll up and down using the arrow buttons that appear (up arrow shown at right).



Figure 5-2: Processing Module Tab (BOND RX)



Each tab shows the processing module name, and rectangular icons display the state of the module's slide staining assemblies (see below). To display the **System status** screen for a processing module, click on the tab. A blue outline and right-facing arrow appear around a processing module tab when it is selected (see above).

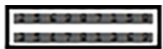
5.1.1.1 Slide Staining Assembly States

Below are examples of slide assembly states you may see on a processing module tab.

Before a Run:



Blank rectangle: no tray present or not locked.

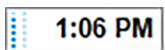


Animated ID numbers and solid bars: tray is being imaged.

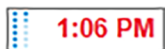


Icon of tray with slides: slide labels have been imaged and the tray is ready to run.

During a Run:

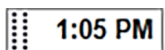


Time display in black with moving dots at left: tray is running with no unexpected events reported. The time displayed is the estimated completion time for the tray.



Time display in red with moving dots at left: tray is running with unexpected events reported. The time displayed is the estimated completion time for the tray.

After a Run:



Flashing time display in black, with static dots at left: the run finished at the time reported without any unexpected events.





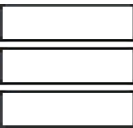



Flashing time display in red, with static dots at left: the run finished at the time reported – unexpected events occurred.






The run was abandoned.

5.1.1.2 Processing Module States

The software continuously monitors the system status and may display icons on the processing module tab as follows:

Icon	Meaning	Icon	Meaning
	The processing module is not connected.		Warning: The BOND RX software has detected an unexpected state.
	(Flashing) The processing module is initializing.		Alarm (flashing): To continue operation the processing module needs user intervention.
	The processing module is currently being serviced.		The processing module is undergoing a maintenance operation.

5.1.2 Hardware Status

The icons at the lower right of the screen display a warning  or alarm  if there is a problem with some part of the BOND RX system, or information indicator  if there is a general notification for the system. Right-click on the icon to get more information.



General fault with the system, or a maintenance task reminder.



Appears when either the lid is opened or (BOND RX™ only) the bulk container door is opened during a staining run. These must be closed to operate the processing module.

If a staining run is not in progress, the information indicator  appears instead.



Missing or insufficient reagent.



The processing module has started initialization and has not yet attempted to scan the mixing station.



The mixing station was not detected during initialization. The station may not be present, or it may be present but the barcode was not recognized.

If necessary place a clean mixing station in the processing module. Right-click on the icon and follow the prompts to inform the system that the mixing station is in place.



The mixing station state at initialization is dirty (e.g. the station was dirty when the processing module was last closed down).

Ensure there is a clean mixing station in place then right-click on the icon and confirm.



Mixing station cleaning has failed.

You may still be able to proceed using remaining clean vials. Otherwise, you will need to restart the processing module to clear the notification.

If the notification persists it may indicate a fluidics problem – contact customer support.



There are no clean mixing vials available.

Wait for the processing module to clean some vials and then proceed as normal.

If vials are not cleaned you may need to restart the processing module. If the notification persists it may indicate a fluidics problem – contact customer support.

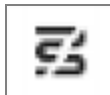


You can manually clean the mixing station if necessary. See [12.7 Wash Block and Mixing Station](#).

5.1.2.1 Heater Errors

Each of the BOND RX and BOND RX^m slide heaters is independently monitored and will be marked as faulty if a temperature error occurs (see [Figure 5-3](#)). Contact customer support if a faulty heater is indicated.

Figure 5-3: Individual heater error



You should not attempt to run a slide that requires heating at a position marked as faulty. If a heater malfunctions during a run it may compromise the slide at that position. If the heater malfunction is a safety risk, it may shut down all slide heating on the processing module (see [Figure 5-4](#)).

Figure 5-4: Gray heater symbols at each position indicate a complete heating shutdown



Once slide heating is shut down, you must turn off then restart the processing module to clear the heater lock. You can continue to use slide positions with faulty heaters so long as the slides processed there do not require heating.

5.1.2.2 Temperature Indication

When a slide staining assembly is above ambient temperature, a temperature indicator appears near the bottom of the **System status** screen.

The temperature indicator at the bottom of the screen shows that a slide staining assembly is either warm or hot.

Figure 5-5: Temperature indicator – warm (left) and hot (right)



The borders of the slide trays on the **System status** screen also change color to indicate temperature: blue when the tray is at ambient temperature, orange when it is warm and red when it is hot.

Figure 5-6: Temperature-indication borders of the slide trays: warm (left) and hot (right)



WARNING: Avoid contact with slide staining assemblies and their surrounds. These may be very hot and cause severe burns. Allow twenty minutes after the cessation of operation for the slide staining assemblies and their surrounds to cool.

5.1.3 Reagent Status

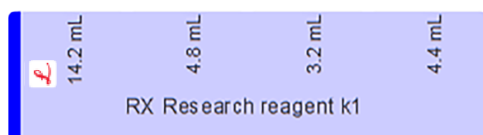
The right side of the **System status** screen displays the status of reagents detected. The sections below describe the icons used and how to fix some reagent problems indicated on the screen.

- [5.1.3.1 Reagent Systems](#)
- [5.1.3.2 Reagent Containers](#)
- [5.1.3.3 Reagent Levels](#)
- [5.1.3.4 Reagent Levels in Research Systems](#)
- [5.1.3.5 Fixing Reagent Problems](#)
- [5.1.3.6 Fixing Undetected Reagents](#)
- [5.1.3.7 Bulk Container Status](#)

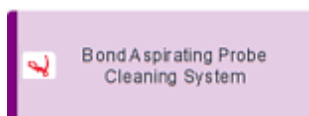
5.1.3.1 Reagent Systems



BOND detection system



BOND research reagent system

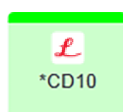


BOND cleaning system

5.1.3.2 Reagent Containers

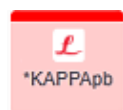


Reagent container icons have an asterisk (*) before BOND-supplied reagent names.



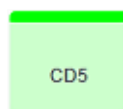
A BOND ready-to-use primary antibody.

Details for these reagents are automatically entered by the BOND RX software when you register them. The abbreviated name of the reagent is shown.



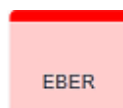
A BOND ready-to-use ISH probe.

Details for these reagents are automatically entered by the BOND RX software when you register them. The abbreviated name of the reagent is shown.



User-supplied primary antibody in a BOND open or titration container.

Details for these reagents must be entered manually in the **Reagent Setup** screen before registering, with lot number and expiration date required when registering. The abbreviated name of the reagent is shown.



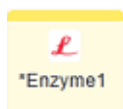
User-supplied ISH probe in a BOND open or titration container.

Details for these reagents must be entered manually in the **Reagent Setup** screen before registering, with lot number and expiration date required when registering. The abbreviated name of the reagent is shown.



BOND ready-to-use ancillaries.

Details for these reagents are automatically entered by the BOND RX software when you register them. The abbreviated name of the reagent is shown.



BOND enzyme in a BOND open or titration container.

BOND enzyme must be prepared by users and put into open containers, but reagent setup details are predefined in the BOND RX software. Only lot number and expiration date are required when registering.



User-supplied ancillary reagent in a BOND open or titration container.

Details for these reagents must be entered manually in the **Reagent Setup** screen before registering, with lot number and expiration date required when registering. The abbreviated name of the reagent is shown.



The software did not detect a reagent in this position.

If there is a reagent present, see [5.1.3.6 Fixing Undetected Reagents](#) for details on how to fix the problem. If the imager frequently fails to properly image IDs, clean the ID imager window (see [12.9 ID Imager](#)).



The BOND RX software detected a problem with this reagent. Right-click the Information symbol for further information.

It may be that the BOND RX software did not recognize the reagent. In that case use the handheld scanner to scan the reagent and add it to the inventory. If the ID is damaged, enter the ID manually. Refer to [8.3.3 Registering Reagents and Reagent Systems](#) for more information.



The BOND RX software detected a problem with this reagent or reagent system.

Right-click the notification symbol for further information.

5.1.3.3 Reagent Levels

The icons for reagent systems indicate only three volume levels on the **System status** screen:



Ready-to-use reagents and open container icons indicate reagent levels more precisely.

Figure 5-7: Examples of ready-to-use reagent levels shown on **System status** screen



To view more detailed reagent or reagent system inventory information, right-click on the icon and select **Inventory ...** from the pop-up menu. The **Reagent inventory details** screen appears. See [8.3.2 Reagent or Reagent System Details](#).

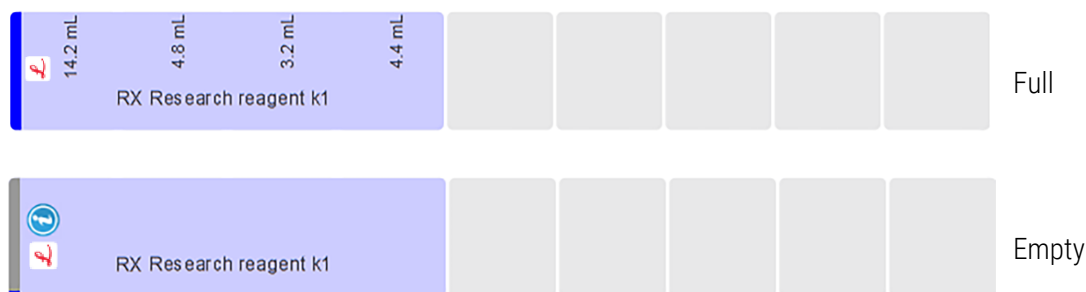
5.1.3.4 Reagent Levels in Research Systems

The icons for research reagent systems indicate either Full or Empty on the **System status** screen (see below). The icon always indicates Full, unless:

- the number of tests remaining in the system is less than one, and/or
- there is zero physical volume in one or more system components
- the user has marked the research system as Empty

The remaining volume for each system component is displayed, unless the system is determined to be Empty (see above), in which case no remaining volume will be displayed.

Figure 5-8: Examples of research reagent system levels shown on **System status** screen



5.1.3.5 Fixing Reagent Problems

If the BOND RX software detects a problem with a reagent required for processing, before a run starts, then the software will display an attention icon on a reagent container graphic below the slide tray on the **System status** screen. If the problem occurs during a run, the attention icon appears over the reagent hardware status icon, as described earlier in this section. To see more information about the problem, right-click on the attention icon.

If you need to replace or add reagent, remove the reagent tray containing the problem reagent, replace or add the required reagent to the tray, then reload the tray.




Note that if processing is already in progress, and reagent in a specific tray will be required within 2 minutes, you will not be able to remove that tray without abandoning the run. This is indicated by the indicator for that reagent tray glowing red.

5.1.3.6 Fixing Undetected Reagents

If a reagent is not detected or a kit is only partially detected, do the following:

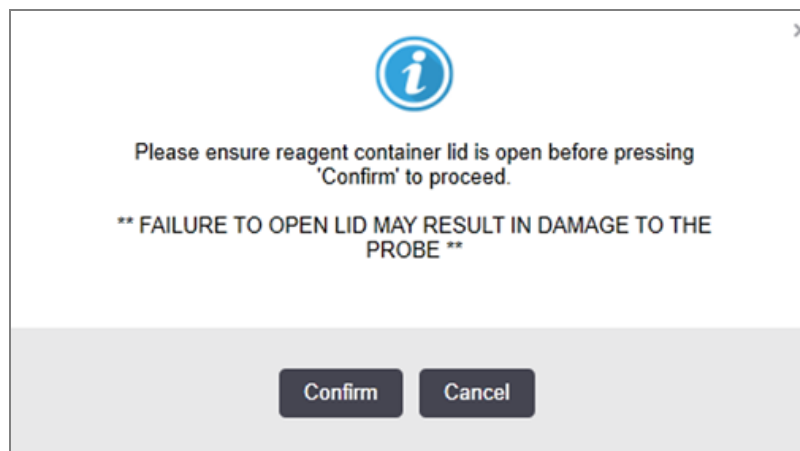
- 1 Check that:
 - The reagent container is correctly positioned in the reagent tray
 - The reagent container cap is opened and clipped to the back of the container
 - There is an undamaged reagent barcode ID across the top front of the container.

- 2 Check that the reagent is registered in the inventory.
 - If the reagent is not registered, then register it as described in [8.3.3 Registering Reagents and Reagent Systems](#).
- 3 At this point you can either:
 - a remove the reagent tray (then make a note of the Unique Pack Identifier (UPI) of the reagent concerned) and reinsert it to make the system automatically identify the reagent tray again, or
 - b if reinserting the reagent tray is not an effective solution, you can manually identify the reagent - right-click on the container icon on the **System status** screen and click **Select ...** from the submenu. Enter the UPI of the reagent you noted above, and click **OK**.

A symbol  appears on the image to identify a reagent that has been manually entered or partially auto-identified. The symbol (and the manually identified reagent or auto-identified kit) is removed if the reagent tray is removed.

If you manually enter a reagent UPI number, the following message is displayed:

Figure 5-9: Notification for manually entered reagent



Once the manually entered UPI number is identified by the system or the processing module has auto-identified the reagent, the following icon is displayed.

Figure 5-10: Manually entered or auto-identified reagent



If a kit has been only partially identified and the processing module has auto-identified some containers, the following message and icon are displayed.

Figure 5-11: Notification of auto-identified kit

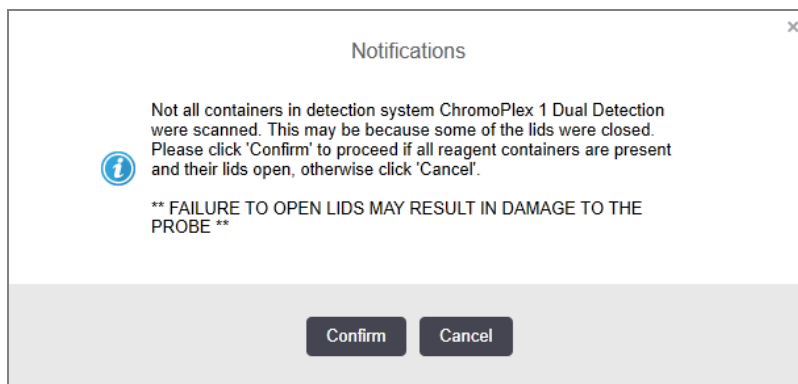


Figure 5-12: Auto-identified kit



5.1.3.7 Bulk Container Status

The bottom right of the **System status** screen displays icons for bulk waste and reagent containers. Each container is labeled and the colors match the installed containers. The positions of the bulk container icons on the System status screen reflect the physical positions of the corresponding bulk container cavities on the processing module.

Refer to [2.2.7 Bulk Containers Cavity](#) for the actual container configuration for each processing module type.

Figure 5-13: Bulk containers (BOND RX configuration)

Figure 5-14: Bulk containers (BOND RX^m configuration).

The right-most icon represents the external waste container

Below are descriptions of each of the above bulk container's contents.

Bulk container label	Bulk container contents
*Dewax	BOND Dewax Solution
*DI	Deionized Water
*BWash	BOND Wash Solution
*Alcohol	Alcohol (reagent grade)
*BlkWast	Bulk Waste
*HazWast	Hazardous Waste
*ER1	BOND Epitope Retrieval Solution 1
*ER2	BOND Epitope Retrieval Solution 2

BOND RX

The software displays the BOND RX bulk reagent and waste containers fluid levels. If the reagent supply is low, or the waste levels are high, there may be an audible alarm, pulsing bottle light (white or red) and a warning icon displayed on the status screen, depending on the severity of the problem. Right-click the icon to view the attention message and take any actions needed to fix the problem – see [12.2.2 Replenishing or Emptying Bulk Containers](#).



If the warning symbol appears, processing is paused until the problem is fixed.








The bulk container status on the **System status** screen is synchronized with the lighting system, as described in [Bulk Container Lighting System \(BOND RX\) \(on page 49\)](#).

The display on the BOND RX software shows an interpreted level in the bottle based on an estimate of how many more slides can be processed with the bulk reagents. The following images are used to indicate the bulk container states:

Bulk reagent container volume icons

Figure 5-15: Bulk reagent container volume icons

Level	State	Supply Bottles	Dewax	Alcohol	DI	Buffer	HEIR1	HEIR2	Label	Bottle
		GUI	Volume range						Lights	
-	-	Bottle removed	-	-					WHITE Flashing	Off
0	Pause running batch		0 - 150	0 - 150	0 - 150	0 - 150	0 - 100	0 - 100	RED Flashing	RED Flashing
1	Can't start batch		150 - 500	150 - 500	150 - 1000	150 - 1000	100 - 300	100 - 300	WHITE Flashing	WHITE Flashing
2	OK		500 - 750	500 - 750	1000 - 1500	1000 - 1500	300 - 500	300 - 500	WHITE	WHITE
3	OK		750 - 2500	750 - 2500	1500 - 3500	1500 - 3500	500 - 1500	500 - 1500	WHITE	WHITE
4	OK		2500 - 5000	2500 - 5000	3500 - 5000	3500 - 5000	1500 - 2000	1500 - 2000	WHITE	WHITE



Appears if the following occurs:

- reagent is running low and needs to be filled immediately
- container is missing
- insufficient volume to start a run

See [12.2.2 Replenishing or Emptying Bulk Containers](#).



or



Appears if a run has been paused because one of the following occurs:

- reagent is low and needs to be filled urgently (warning)
- container is missing and needed for processing (alarm)

See [12.2.2 Replenishing or Emptying Bulk Containers](#).

Waste container volume icons

Figure 5-16: Bulk waste container volume icons

Level	State	Waste Bottles	Std Waste	Haz Waste	Label	Bottle
		GUI	Volume range		Lights	
-	-	Bottle removed	-	-	WHITE Flashing	Off
0	OK		0 - 1100	0 - 1100	WHITE	Off
1	OK		1100 - 3000	1100 - 3000	WHITE	WHITE (1 strip only)
2	OK		3000 - 3900	3000 - 3900	WHITE	WHITE
3	Can't start batch		3900 - 4800	3900 - 4800	WHITE Flashing	WHITE Flashing
4	Pause running batch		4800 - 5000	4800 - 5000	RED Flashing	RED Flashing



Appears if the following occurs:

- waste is nearly full and needs to be emptied immediately

See [12.2.2 Replenishing or Emptying Bulk Containers](#).



or



Appears if a run has been paused because one of the following occurs:

- waste is full and needs to be emptied urgently (warning)
- container is missing and needed for processing (alarm)

See [12.2.2 Replenishing or Emptying Bulk Containers](#).

BOND RX^m

The software displays an attention icon (as above) over a bulk container when it detects a problem (for example, the volume in a reagent container is low, or the volume in a waste container is high). Right-click the notification icon for details.

5.1.4 Slide Information

The sections below describe the icons used to represent slide information on the **System status** screen. Options in the slide pop-up menu are also described.

- [5.1.4.1 Slide Icons](#)
- [5.1.4.2 Slide Tray Pop-up Menu](#)
- [5.1.4.3 Slide Event Notifications](#)
- [5.1.4.4 Fixing Incompatible Slide Setup](#)

5.1.4.1 Slide Icons

The **System status** screen displays a graphical representation of each of the three slide trays with an icon for each slide. The slide icons indicate the state of each slide.

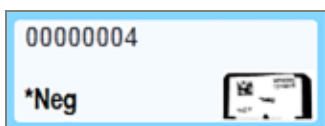
Your system uses 2D barcodes. The slide icons can be optionally configured to include captured images of the slide labels. Contact customer support if you wish to change the existing settings.

Examples of slide icons are shown in the following tables.

Slide Icons for Barcode Labels



No slide at this position, or slide imaged but system unable to identify



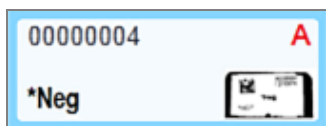
Single slide imaged and automatically identified
(refer to [5.1.5.1 Automatic Slide Identification](#))



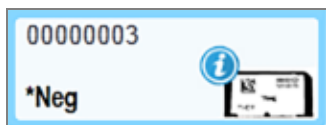
Multiplex slide imaged and automatically identified
(refer to [5.1.5.1 Automatic Slide Identification](#))



Slide imaged and manually identified – note the symbol (circled in red) on the slide
(refer to [5.1.5.2 On-Board Manual Slide Identification](#))



Slide is incompatible with one or more other slides on the tray
(refer to [5.1.4.4 Fixing Incompatible Slide Setup](#))



Slide processing with event notification
(refer to [5.1.4.3 Slide Event Notifications](#))

Double-click slides that have been recognized by the BOND RX system to open the **Slide properties** dialog for them. If the run has not been initiated you can edit slide details in the dialog, but you will then need to print a new label for the slide, unload the tray and apply the new label, and then reload.

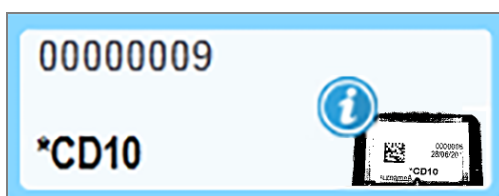
5.1.4.2 Slide Tray Pop-up Menu

Right-click on slides in the slide tray graphics on the **System status** screen for a number of options for the slide or tray.

Command	Description
Select manually...	Enabled if the slide was not automatically identified. Select to open the Slide identification dialog, which allows you to identify the slide with one configured in the system (see 5.1.5.2 On-Board Manual Slide Identification). This option can also be selected if you double-click on an unidentified slide.
Attention message...	View an attention message if the slide is showing an event notification (see 5.1.4.3 Slide Event Notifications).
Run events	Generate a Run Events Report for the run (see 9.4 Run Events Report).
Delayed start	Set a delayed start for the run (see 5.1.8 Delayed Start).

5.1.4.3 Slide Event Notifications

Figure 5-17: Slide with event notification



When an unexpected event occurs during processing an alert symbol appears on the slide icon. This notification does not necessarily indicate that staining was in any way unsatisfactory. When the notification symbol appears the system operator or laboratory supervisor must take the following extra steps to confirm that the slide is suitable for use.

- 1 Right-click on the slide and select **Run events** to generate the Run Events Report (refer to [9.4 Run Events Report](#)).
Any events that caused a notification are displayed in **Bold** text. The system operator or laboratory supervisor should carefully consider the notification events listed as these provide important details about the nature of the slide notification events.
- 2 Carefully inspect the stained tissue.
- 3 Carefully inspect any control slides.

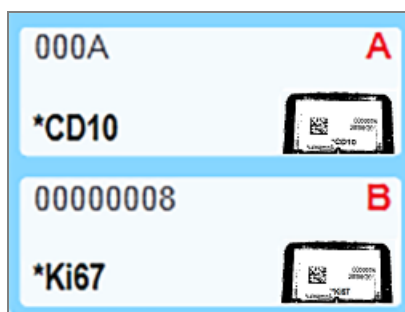
If unable to confirm the staining quality, consider rerunning the test.

Multiple notifications may be present within a single Run Events Report. If the run completes with status **Done (notification)**, ensure that the entire report is inspected. If the status is **Done (OK)**, there is no need to inspect the report.

5.1.4.4 Fixing Incompatible Slide Setup

If the BOND RX system detects an incompatible slide, it will assign letters in bold red lettering at the upper right of all slides in the tray. Slides with the same letter are compatible.

Figure 5-18: Incompatible slides



Remove the slide tray and remove the incompatible slides, or change slide properties (if there were errors in these) to make slides compatible. If you change slide properties you must reprint the labels for the changed slides and attach these before reloading the tray.

See [6.9 Slide Compatibility](#) for further details about slide compatibility.

5.1.5 On-Board Slide Identification

In the most common workflow, slides with labels from the BOND RX system or an LIS are loaded on the processing module and then automatically identified. Identification is by reading 2D barcodes on the labels. If a label is smudged or for some other reason cannot be read, you can manually identify it to the BOND RX software. Some workflows use manual identification as a matter of course (see [6.8 Impromptu Slide and Study Creation](#)).

5.1.5.1 Automatic Slide Identification

The BOND RX system is able to automatically identify standard BOND 2D barcode slide labels created using the BOND RX labeler (as described in [6.6 Slide Labeling](#)), and LIS-printed slides that use a recognizable barcode format (see [11.3 LIS Connection and Initialization](#)). When a slide tray is locked, the system attempts to identify each slide label and match it against a slide that has had a label printed. Where it is able to match the label to a printed slide, the slide is automatically identified and no further action is required.

The system captures an image of each label during the slide identification process. These images appear in the following reports to provide a permanent record of the slide matching:

- [9.4 Run Events Report](#)
- [9.5 Run Details Report](#)
- [9.6 Study Report](#)

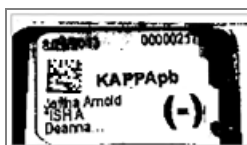
If the system was unable to identify the label then the slide must be manually identified using the manual slide identification procedure (see the next section).

5.1.5.2 On-Board Manual Slide Identification

On systems that are set up to take an image of each slide label, if automatic identification fails, slides can be manually identified while still loaded on the processing module. Use the following procedure to manually identify a loaded slide.

- 1 When the system is unable to automatically identify a slide, the System Status dialog displays an image of the label.

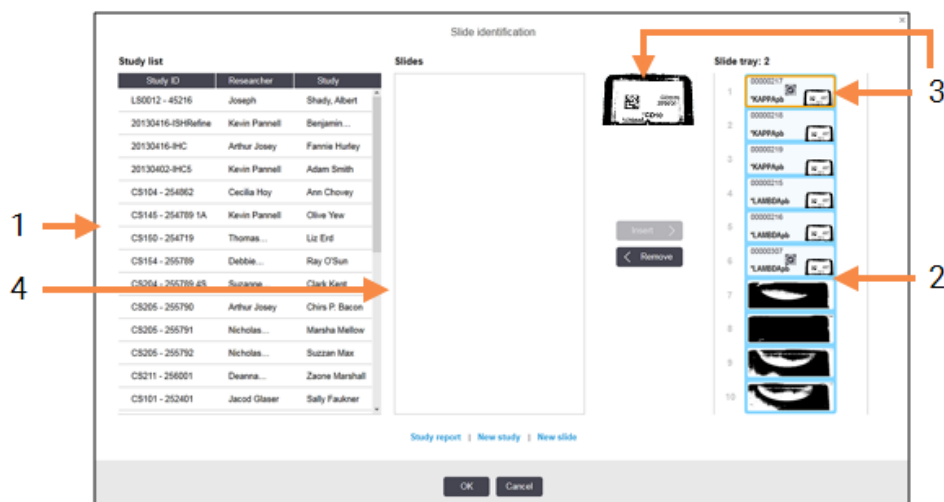
Figure 5-19: Slide not automatically identified



- 2 To launch the Slide Identification dialog do one of the following:
 - a Double-click the slide image; or
 - b Right-click on the image and select **Select manually** from the pop-up menu.

- The **Slide identification** dialog is displayed.

Figure 5-20: Slide identification dialog



The left-hand pane (item 1) lists all studies with unprocessed slides. Under default settings, only studies with slides for which labels have been printed appear (you can change this to include studies with slides for which labels have not been printed – see [6.8.2.2 External Slide Labels](#)).

Slide labels in the current slide staining assembly are shown in the right-hand pane (item 2).

The slide selected when the dialog was opened is highlighted (item 3) in the right-hand pane and displayed enlarged. Hold the cursor over the slide in the right-hand pane to see an even bigger enlargement of the image.

The center pane (item 4) shows slides configured for the study selected in the left-hand pane, where the slides have not yet been matched to any slides imaged on the processing module. Again, under default settings, only slides that have had labels printed appear, but this can be changed to show all slides configured for the study (see [6.8.2.2 External Slide Labels](#)).


It is possible to create new studies and slides at this point, with **New study** and **New slide**, if necessary (see [6.8 Impromptu Slide and Study Creation](#) for instructions). The instructions below assume that all required slides are already configured in the BOND RX software.

- Use the information visible in the selected label image, on the right, to determine the study that the slide belongs to. Select that study from the study pane (item 1).

The slide list (item 4) is populated with the unmatched slides configured for that study.

- Now match the unidentified slide to a slide in the slide list (item 4).

Select the slide and click **Insert**.

The slide is removed from the slide list and the image in the right-hand pane updates to show that the slide has been identified. A symbol  identifies the slide as having been manually selected.

The next unidentified slide label, if there are any, is now highlighted for identification.

- Match all the unidentified slides by repeating the steps above.


- 7 When all the slides in the tray have been identified click **OK** to close the dialog. If you click **Cancel** any slide identifications you may have made will be lost.
- 8 The **System status** screen now shows all slides in the tray with their slide details. The slides that were manually identified include an image of the label and the symbol  to show that the slide was manually selected.

Figure 5-21: Manually identified slide prior to processing



- 9 Manually selected slides process normally.

An image of the slide appears in the following reports to provide a permanent record of the slide matching:

- [9.4 Run Events Report](#)
- [9.5 Run Details Report](#)
- [9.6 Study Report](#)

Systems NOT set up to take an image of each slide label

These slides can still be manually identified, but this may involve removing the slide tray.

Take note of the slide ID and slide position number (embossed on the slide tray below the neck of the Covertile) for the slide that was not automatically identified.

Reload the slide tray and double-click the corresponding slide position (counting down from the top position of the slide staining assembly in the **System status** screen).

5.1.6 Run Progress Indicator

Progress indicators sit below each of the slide tray graphics. They provide a quick visual indication of run status and progress.

- [5.1.6.1 Run Status](#)
- [5.1.6.2 Run Progress](#)
- [5.1.7.1 Stopping a Run](#)
- [5.1.8.1 Setting Delayed Start Time](#)

5.1.6.1 Run Status

The current run number and status is displayed at the bottom of each progress indicator. The possible run states are:

Run Status	Description
Unlocked	The slide tray is unlocked.
Locked	The slide tray is locked but it is not yet possible to start. This state usually occurs prior to the completion of slide imaging.
Slides ready	All slides in the slide staining assembly have been imaged.
Starting	The start button has been pressed and the system is performing pre-start checks and scheduling.
Rejected/Slides ready	The BOND RX system attempted to start the run but was unsuccessful. The most likely causes of rejection are missing reagents, low bulk reagent levels, or a full waste container. Generate a Run Events Report, resolve any problems that it indicates, then restart the run.
Scheduled	The run is scheduled but has not started processing. The run progress indicator indicates the scheduled start time.
Proc (OK)	The run is processing, no unexpected events have occurred.
Proc (notification)	The run is processing, unexpected events have occurred. Check the Run Events Report for details.
Abandoning	The run is being abandoned. This occurs when the operator presses the stop button.
Done (OK)	Processing is complete, no unexpected events occurred.
Done (notification)	Processing is complete, unexpected events occurred. Check the Run Events Report for details.

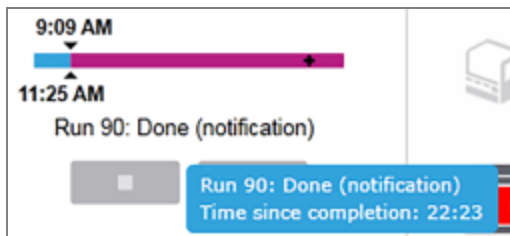
5.1.6.2 Run Progress

A progress bar below each slide tray graphic provides a visual display of run progress. The progress bar displays critical times, shows the current progress with respect to the critical times, and uses the following colors to represent the four stages of run progress:

- Blue – slide tray is locked, processing has not started
- Red – processing has not started and the starting time limit has been exceeded
- Green – processing
- Purple – run has been completed and is now being hydrated.

You can pause the mouse pointer in the run progress section to display the run status, such as “Time since locked”, “Time to completion” and “Time since completion” as shown in [Figure 5-22](#).

Figure 5-22: Run status display



Slides Ready – Starting

After slides have been imaged and the run is ready to start, and for a brief period after the start button has been pushed or a delayed start initiated, the bar displays the following items (refer to [Figure 5-23](#) for item numbers).

Figure 5-23: Run progress (starting)



Legend

- | | | | |
|---|-----------------------------------------------------------------------------------------------------------------------|---|----------------------------------------------------------|
| 1 | Time the tray was locked | 4 | The current progress |
| 2 | The acceptable starting period (blue bar) (see Acceptable Starting Period & Alarm (on page 118)) | 5 | The start time exceeded period (red bar) |
| 3 | The acceptable start time limit | 6 | The run status (see 5.1.6.1 Run Status) |

Acceptable Starting Period & Alarm

Always start processing as soon as possible after slide trays are locked. The slides are not hydrated during the “starting period” (between locking a tray and start of processing) so if this period is too long, for dewaxed slides, tissue can be damaged. The BOND RX software helps you monitor this by tracking the times since trays were locked and showing the acceptable maximum starting period for the slide type loaded (waxed or dewaxed). The acceptable starting periods are visually displayed as the blue bar in the “Slides ready” progress bar (see above). For dewaxed slides, if processing has not started some time after the acceptable starting period, there is an alarm.

Starting periods and the dewaxed slide alarm period are shown below. All times are from when trays are locked:

Acceptable Starting Period or Alarm	Time (min) from locking tray
Dewaxed slides acceptable starting period	15
Dewaxed slides time to alarm	25
Waxed slides acceptable starting period	60

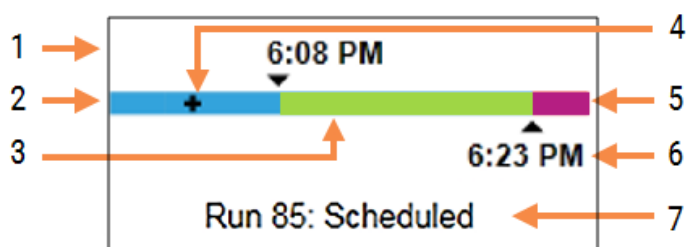
If processing has not started within the starting period you can remove trays to manually hydrate the slides. When you reinsert the tray the BOND RX software starts a new run, allocating a new run ID number and starting the period count again.

Acceptable start time limits apply only to immediate-start runs; they do not apply to delayed start runs.

Scheduled

After a run has been initiated with the start button or delayed start it is scheduled in the system. In the period between scheduling and processing beginning – which may be long in the case of a delayed start – the progress bar displays the following items (refer to [Figure 5-24](#) for item numbers).

Figure 5-24: Run progress (starting, with delayed start)



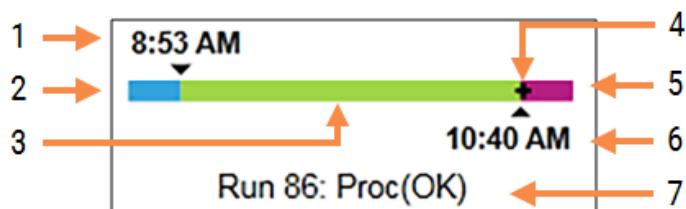
Legend

- | | |
|--------------------------------------|------------------------------------------------------------|
| 1 Time the run is scheduled to start | 5 Post processing hydration period (purple bar) |
| 2 Delay before the start (blue bar) | 6 The approximate run finish time |
| 3 Processing period (green bar) | 7 The run status (see 5.1.6.1 Run Status) |
| 4 The current progress | |

During Processing

During the processing phase the bar displays the following items (refer to [Figure 5-25](#) for item numbers).

Figure 5-25: Run progress (processing)



Legend


- | | | | |
|---|-----------------------------------------------------------------|---|----------------------------------------------------------|
| 1 | Scheduled start time | 5 | Post processing hydration period (purple bar) |
| 2 | The starting period – blue: start OK, red: start limit exceeded | 6 | The approximate time the run will finish |
| 3 | Processing period (green bar) | 7 | The run status (see 5.1.6.1 Run Status) |
| 4 | The current progress | | |

5.1.7 Starting or Stopping a Run

You begin a run by loading and locking a slide tray. The tray is imaged and the system checks the following to ensure it can run:

- All slides are compatible
- All reagents are available.

When the slides are imaged the run status is set to **Slides ready** (see [5.1.6.1 Run Status](#)) and the progress bar appears in the starting phase (refer to [5.1.6.2 Run Progress](#)). Once any slide incompatibilities have been resolved, all slides have been identified, and checks run to ensure that all required reagents are present, the run can be started.

- To start the run as soon as possible click . For delayed start right-click on the tray and select **Delayed start** from the pop-up menu; see further directions in [5.1.8 Delayed Start](#)
 - The run status is set to **Starting** as the pre-run checks and scheduling are completed.
The progress bar remains in the starting phase.
 - Once the scheduling is complete, the state changes to **Scheduled**.
The progress bar now appears in the processing phase. The scheduled start time is displayed and the starting condition (OK or time limit exceeded) is displayed at the left end of the bar.
 - When processing starts at the scheduled time, the state changes to **Proc (OK)**.
If the start time limit was exceeded the warning or alarm clears once processing actually starts. The starting section of the progress bar remains red however.
 - Note that the **Starting** and **Scheduled** states may take some time and it is possible the starting time limit is exceeded. If this is likely to occur you can unlock the slide tray and manually hydrate the slides before restarting. If you unlock a tray prior to processing commencing, the run is not considered abandoned and can be restarted.



You should start only one run at a time, and then wait until that run has started/been scheduled before starting the next run. Wait for a short while after starting each run to confirm it has started successfully. If not, the run status is set to **Rejected/Slides ready**. See [5.1.6.1 Run Status](#). You should then generate the Run Events report to identify why the run did not start (see [9.4 Run Events Report](#)).

5.1.7.1 Stopping a Run

After pressing the start button (or activating delayed start) up until processing actually begins – while the run is in **Starting** or **Scheduled** states – processing can be stopped for a run without having to abandon it. To cancel a processing request at this time unlock the slide tray on the processing module (the start and abandon buttons are disabled during this period). Slide information remains in the system and the run can be restarted later if you want. A single line is written to the **Slide history** list for the rejected run.

To abandon a run once processing has started click . The processing module will cease operation on the run after completing the current step. The status of the slides on the **Slide history** screen changes to **Done (notification)**.



Consider carefully before abandoning a run – abandoned runs cannot be restarted, and any slides for which processing has not been completed may be compromised.

5.1.8 Delayed Start

Runs with waxed slides can be scheduled to start at a specified future time (up to one week from the current time) on the BOND RX and BOND RX^m systems. Runs started overnight, for example, can be timed so that they finish shortly before start of work on the following day. Slides sit safely, still waxed, until processing begins, and the hydration period that follows processing is minimized.

On the BOND RX system there is also a prestaining protocol ***Frozen slide delay** (see [7.2 Protocol Setup Screen](#)). This protocol is specifically designed for use with unwaxed fresh and frozen sections on slides, and contains three steps of BOND Wash only.



Some non-Leica Biosystems reagents could deteriorate if kept for long periods on processing modules awaiting delayed starts. Check product data sheets for reagent use and storage information. As always, Leica Biosystems recommends placing control tissue on slides with test tissue.



If the scheduled end time is not suitable, use the **Load / Unload** button to raise and re-lower the SSA. After the slides have been re-scanned, the delayed start time can now be adjusted to reflect the desired end time.

5.1.8.1 Setting Delayed Start Time

To run a tray with delayed start, prepare slides as usual and lock the slide tray. When the run status is **Slides ready** select **Delayed start** from the tray's right-click pop-up menu on the **System status** screen.

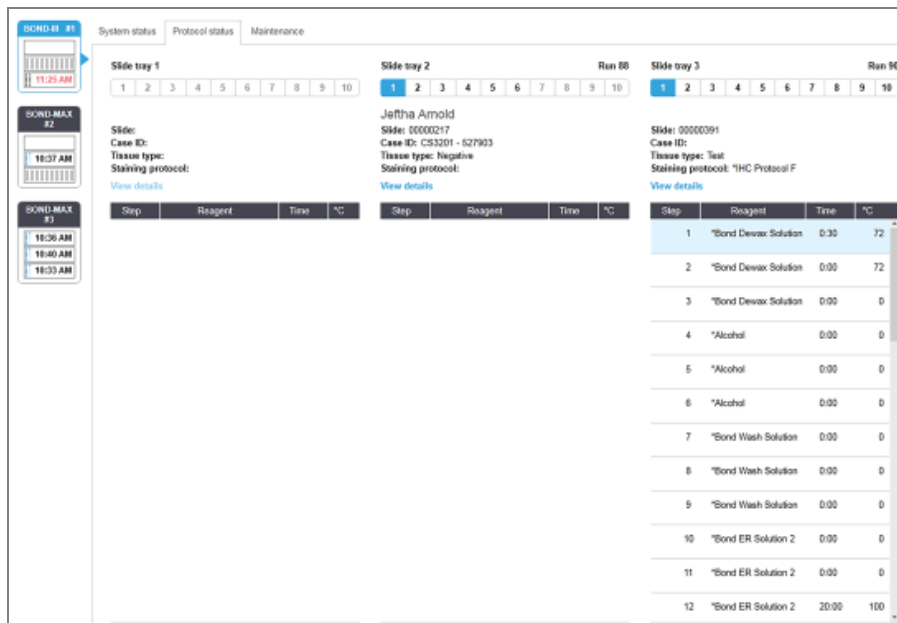
Set the date and time that you want the tray to start in the **Delayed start** dialog, and click **OK** (see [Using the Date & Time Selectors \(on page 207\)](#)). The system goes into **Starting** state as usual, and schedules the run in coordination with other operations. The tray then waits with status **Scheduled** until the set start time, when normal processing begins.

5.2 Protocol Status Screen

This screen displays detailed information about the status of individual slides.

To display the **Protocol status** screen, go to the **System status** screen and click the **Protocol status** tab.

Figure 5-26: The **Protocol status** screen



To see how a run is progressing on a slide, click the corresponding slide position button near the top of the screen. Option buttons corresponding to positions without a slide are dimmed, and you cannot select them.



If the study name is too long to fit in the available space (slide tray 1, 2 and 3) the name is shortened with “...” at the end. If you want to see the study’s full name in a pop-up field, hover the mouse pointer over the shortened name.

When you select a slide position, the software displays some slide details and the protocol progress. To view additional slide details click **View details** to launch the **Slide properties** dialog.

The protocol steps for the selected slide are displayed beneath the slide details. The current step is highlighted blue.

Completed steps show a check mark in a green circle or, if unexpected events occurred, a icon.

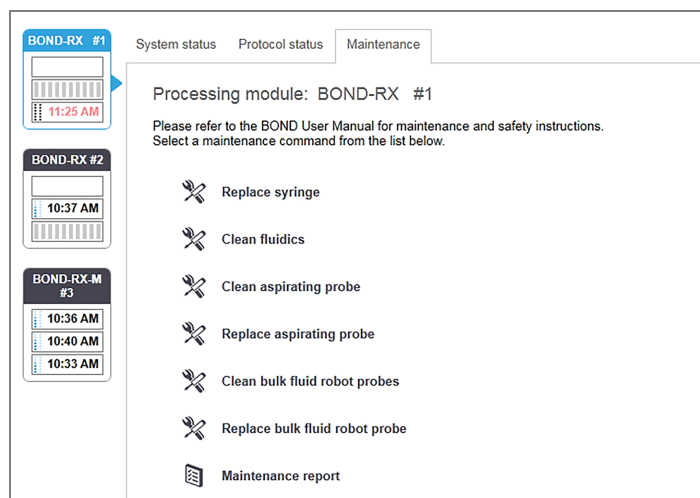
If all the required actions for the current step have been performed but there is a waiting period before the next step begins, the check mark or is gray. It remains gray until the next step starts, when it changes to the normal color.

You can view run events by right-clicking the step list and selecting **Run events** from the pop-up menu. You can also open the **Slide properties** dialog from the pop-up menu.

5.3 Maintenance Screen

To display the **Maintenance** screen, go to the **System status** screen and click the **Maintenance** tab.

Figure 5-27: The **Maintenance** screen



The **Maintenance** screen has command buttons for a range of maintenance tasks, listed below:

Command	Description
Replace syringe	Control the processing module while replacing the syringe or syringes. See 12.13 Syringes .
Clean fluidics	Prime the fluidics system. See Clean Fluidics (on page 281) .
Clean aspirating probe	Clean the aspirating probe with the BOND Aspirating Probe Cleaning System. See 12.6.1 Cleaning the Aspirating Probe .
Replace aspirating probe	Contact Customer Support.
Clean bulk fluid robot probes	Moves the bulk fluid robots (BOND RX only) into position so the probes can be wiped clean. See 12.12.1 Cleaning the Bulk Fluid Robot Probes
Replace bulk fluid robot probes	Contact Customer Support.
Maintenance report	Generate a maintenance report for the selected processing module. This command is always available. See 5.3.1 Maintenance Report

The **Maintenance** screen shows the name of the currently selected processing module and the associated maintenance command buttons. A series of dialog boxes will assist you to carry out the maintenance task you select.

Whenever a maintenance task is unavailable, for example when maintenance is already in progress, its command button is disabled. All command buttons (except **Maintenance report**) are disabled when the processing module is disconnected.

5.3.1 Maintenance Report

Maintenance report displays information about a specific processing module, for a time span that you choose.

- 1 In the research client, select the processing module's tab to display its **System status** screen.
- 2 Click the **Maintenance** tab, and then click the **Maintenance report** button.

Figure 5-28: Maintenance report dialog box

Select a processing module from the drop-down list, and then choose the time span you want, using the **From** and **To** date controls. Or, you can click **Last twelve months** to set the time span to this period.

Click **Generate** to generate the maintenance report.

The report is displayed in a new window. The top right of the report shows the information in the following table:

Field	Description
Facility	The name of the facility as entered in the Facility field on the administration client Laboratory settings screen – see 10.5.1 Laboratory Settings
Time period	The “From” and “To” dates for the period that the report covers
Processing module	The unique name of the processing module as entered in the Name field on the administration client Hardware configuration screen – see 10.6.1 Processing Modules
Serial No.	The unique serial number of the processing module

Points to note regarding the report are listed below:

- An attention icon will appear over the processing module icon on the System status screen (as in [5.1.2 Hardware Status](#)) with a right-click reminder notification when these maintenance tasks become due (at which time the estimated date will display with “Due now”).
- Estimated dates of next maintenance actions are based on the number of slides processed and/or the recommended time period between actions.
- If there is no event history for the report’s time period, a statement to this effect appears in place of a history table.
- The first date in a history table is either the start of the report period, or the processing module’s commissioning date, if this occurred later. The entries in the associated “Slides since last maintenance / replacement” columns always show 0 slides.
- The last date in a history table is the end of the report period.
- There are slide counts for each slide staining assembly, also a combined total slide count for all 3 assemblies. Slide counts reset to 0 after each successful maintenance action.
- There are individual slide counts for each slide staining assembly’s bulk fluid robot probe (BOND RX only).
- There are individual slide counts for each slide staining assembly’s syringe (BOND RX only).
- There is a separate slide count for the main syringe.

6

Slide Setup (on BOND RX Controller)

The standard workflow for creating slides for processing by the BOND RX system involves the following major steps:

- 1 Preparing the sections on the slides.
- 2 Creating a study for the slides in the BOND RX software (or the study may be imported from an LIS).
- 3 Adding or editing researcher details, if necessary.
- 4 Entering the details of the slides (or these may be imported from an LIS).
- 5 Creating control slides as per the laboratory's standard practices.
- 6 Labeling the slides (unless already labeled with LIS labels).
- 7 Loading the slides on slide trays, and placing the slide trays in the processing module.

Once your slides have started processing, the **Slide history** screen allows you to produce a range of slide, study and run reports. Refer to [9 Slide History \(on BOND RX Controller\)](#) for details.

If the standard workflow does not suit your laboratory, there are alternative workflows.

This chapter has the following sections:

- [6.1 Slide Setup Screen](#)
- [6.2 Working with Controls](#)
- [6.3 Working with Studies](#)
- [6.4 Manage Researchers](#)
- [6.5 Working with Slides](#)
- [6.6 Slide Labeling](#)
- [6.7 Slide Setup Summary Report](#)
- [6.8 Impromptu Slide and Study Creation](#)
- [6.9 Slide Compatibility](#)

6.1 Slide Setup Screen

The **Slide setup** screen displays studies and slides entered into the BOND RX system but not yet processed. For LIS-integrated systems it shows the studies and slides imported from the LIS. For non-LIS systems you create and, if necessary, edit studies and slides on this screen. Slides must belong to a study, so you must create a study before you can create slides.



To display the **Slide setup** screen, click the **Slide setup** icon on the function bar.

Figure 6-1: The Slide setup screen

The screenshot shows the Slide setup screen with a table of studies and slides. The table has columns for Study ID, Study name, Researcher name, and Slides. The right side of the screen shows a detailed view of a slide, including its ID, name, and a QR code.

Study ID	Study name	Researcher name	Slides
LS0012 - 45216	Shady, Albert	Joseph	1
20130416-ISHRefine	Benjamin Hightower	Kevin Pannell	10
20130416-IHC	Fannie Hurley	Arthur Josey	10
20130402-IHC5	Adam Smith	Kevin Pannell	10
CS104 - 254862	Ann Chovey	Cecilia Hoy	12
CS145 - 254789 1A	Olive Yew	Kevin Pannell	5
CS150 - 254719	Liz Erd	Thomas Matthews	1
CS154 - 255789	Ray O'Sun	Debbie Hanrahan	3
CS204 - 255789 4S	Clark Kent	Suzanne Rhinehart	3
CS205 - 255790	Chirs P. Bacon	Arthur Josey	1
CS205 - 255791	Marsha Mellow	Nicholas Monahan	10
CS205 - 255792	Suzzan Max	Nicholas Monahan	10
CS211 - 256001	Zaone Marshall	Deanna Hayman	7
CS101 - 252401	Sally Faulkner	Jacod Glaser	3
CS102 - 252413	James Donovan	Jacod Glaser	1
LS0012 - 45214	Barb, Akew	Jenny	1
CS3201 - 527890	Reeve Ewer	Jack Browne	11
CS3201 - 527891	Theresa Brown	Jack Browne	14
CS3201 - 527892	Tex Ryta	Arthur Josey	11

Positive tissue controls: 21
Negative tissue controls: 25

Total studies: 28
Total slides: 143

Slide details (1. 00000198): *GFAP, *IHC F *D *... *H2(20)

Figure 6-1 shows the **Slide setup** screen. The top right of the screen contains features for working with studies, the right of the screen also contains features for working with slides.

6.2 Working with Controls

Leica Biosystems recommends routine use of controls on the BOND RX system. Bear in mind that controls should be a test of the whole process. See [14.3 Quality Control](#) for further discussion.



To most adequately test the performance of the BOND RX system, Leica Biosystems strongly recommends placing appropriate control tissue on the same slide as test tissue.

While placement of control tissue with test tissue is strongly recommended, the BOND RX software also allows for slides with only control tissue, and reagent controls. Take care that slides with only control tissue are well marked to avoid confusion with test samples.

6.2.1 Control Tissue

Each slide must be entered into the BOND RX software as having one of the following tissue types:

- Test tissue
- Negative tissue
- Positive tissue

This is set in the **Add slide** dialog (see [6.5.2 Creating a Slide](#)). Any slide with patient test tissue should be set as “Test tissue”. Use the “Positive tissue” and “Negative tissue” control settings only for slides with only control tissue.

Whenever the tissue type is changed for a new slide in the **Add slide** dialog, the **Marker** field automatically clears, to help ensure that you select the correct marker for the tissue.

Slides with negative or positive tissue are marked with a “-” or “+” respectively in the **Slide setup** screen. On the **Slide history** screen, “Test”, “Negative” or “Positive” is displayed for each slide in the **Type** column.

So that the slides themselves stand out clearly as controls, we include “Tissue type” as one of the information fields in the default slide label templates. This prints a large “(+)” on positive tissue control labels, and “(-)” on negative tissue control labels. Nothing is printed in the field for test tissue. We recommend including this field in any other slide labels you configure (see [10.3 Labels](#)).

6.2.2 Control Reagent

Slides are set up with a control reagent by selecting the appropriate reagent as the marker, in place of standard antibodies or probes, during slide configuration.

For IHC, the BOND RX software includes a negative control reagent option. With IHC selected in the **Add slide** dialog, select ***Negative** from the **Marker** drop-down list. BOND RX delivers BOND Wash Solution for these steps.

For ISH, the BOND RX software includes negative and positive control reagents for RNA and DNA. These reagents need to be purchased, registered, and loaded onto the BOND RX system. Select the appropriate control probe from the **Marker** list.

Slides with control reagents are not specially marked other than by the marker name shown in the **Slide setup** screen and on the slide label if the marker field is included in the applicable slide label template.

6.3 Working with Studies

This section describes the features at the left of the **Slide setup** screen that allow you to work with studies. The subsections following the descriptive section give procedures for adding, editing, and deleting study details.

Sections below:

- [6.3.1 Study Controls and Active Study Information](#)
- [6.3.2 Study Identification](#)
- [6.3.3 Adding a Study](#)
- [6.3.4 Study Duplication, Resurrection and Expiration](#)
- [6.3.5 Editing a Study](#)
- [6.3.6 Copying a Study](#)
- [6.3.7 Daily Study Option](#)
- [6.3.8 Study Report](#)

6.3.1 Study Controls and Active Study Information

Click **Add study** to add details of a new study.

[6.3.3 Adding a Study](#) describes the process.

Click **Edit study** to edit details of an existing study.

[6.3.5 Editing a Study](#) describes the process.

Click **Delete study** to delete an existing .

[6.3.5.1 Deleting a Study](#) describes how to delete a study

Click **Copy study** to add a copy of a study and the slides for that study.

[6.3.6 Copying a Study](#) describes how to copy a study.

The **Edit**, **Delete**, and **Copy** commands can also be accessed in the pop-up menu if you right-click on a study.

Click **Study report** (below the study list) to view a report for the selected study (see [6.3.8 Study Report](#)).

The table below the buttons shows active study information as follows:

Study ID	The study identification. This can be any alphanumeric characters. Because this field can contain letters as well as numbers, clicking the table's Study ID column heading sorts this field as text – an identifier beginning with "10" will be sorted ahead of an identifier beginning with "2".
Study name	Identification of the study.
Researcher	Name of the researcher running the study.
Slides	The number of unprocessed slides configured for the selected study. Once processing starts on slides they are moved from the Slide setup screen to the Slide history screen, and this number updates accordingly.

A study with a red bar at the left-hand side indicates that it has one or more priority LIS slides (see [11.2.5 Priority Slides](#)).

Beneath the active study list there is a summary of all studies and slides as follows:

Positive tissue controls	The total number of positive tissue controls for all studies currently entered and not run.
Negative tissue controls	Total number of negative tissue controls for all studies currently entered and not run.
Total studies	The total number of active studies.
Total slides	The total number of slides for all studies currently entered and not run.

6.3.2 Study Identification

The BOND RX system uses two primary study identifiers: the study ID and study number (**Study ID** and **Study No.** respectively, in the software).

- **Study ID:** a user-entered study ID, using the laboratory's identification scheme. For studies created in the BOND RX system the study ID is entered in the **Add study** dialog when studies are created. For LIS-ip systems the study ID is received from the LIS (where it may be known as the "accession number", or by another term).
- **Study No.:** a unique identifying number that the BOND RX system automatically assigns to every study in the system (both created in the BOND RX system and received from an LIS). The study number is displayed in the **Study properties** dialog.

In previous BOND RX versions there was no requirement that study IDs were unique, so it was possible to have two or more distinct studies sharing the same study ID. The studies would, however, likely have different study names, and certainly have different study numbers. Now, however, all new study IDs must be unique.

Studies are also frequently identified by study name, however, study names are not required and do not need to be unique.

6.3.3 Adding a Study

To add a study, starting at the **Slide setup** screen, do the following:

- 1 Click **Add study** on the **Slide setup** screen to display the **Add study** dialog (see [Figure 6-2](#)).

Figure 6-2: The **Add study** dialog

- 2 Enter the details as appropriate in the Study ID, Study name, Study comments, and Researcher fields.



It is possible to add studies without any study information.

- 3 If the required researcher is not in the **Researcher** list add him or her by clicking **Manage researchers** to open the **Manage researchers** dialog (refer to [6.4 Manage Researchers](#)).
- 4 Select a dispense volume for slides created for this study, if it is not the same as the already configured default dispense volume.

Note that all slides processed on a BOND RX Processing Module require a 150 µL dispense volume. In addition, ISH staining uses a 150 µL dispense volume on all processing module types.

For information on the usable areas on slides and dispense volumes, refer to [6.5.8 Dispense Volumes and Tissue Position on Slides](#).

- 5 Select a preparation option from the **Preparation protocol** list (see [Figure 6-2](#)), to make it the default for slides created for this study.
- 6 To leave the dialog without entering the details in the system, click **Cancel**.

- 7 To enter the details of the study, click **OK**.

The study is added to the study list.



If the Study ID already exists in the system, the **Study ID duplication** dialog opens (see [6.3.4 Study Duplication, Resurrection and Expiration](#)).

6.3.4 Study Duplication, Resurrection and Expiration

Previous BOND RX software versions allowed distinct studies to have the same study ID. Such studies could generally be distinguished by the study name, and distinction was always guaranteed by the study number, which is always unique. The BOND RX system no longer allows new studies to use the same study IDs as existing studies – each new study must be either given a unique study ID or identified as the same as a study already in the system.

If you enter a study with a study ID already in the system, the **Study ID duplication** dialog is displayed, showing the existing study with the same study ID. To use the existing study select it and click **Use selected** (also see [6.3.4.1 Merging Studies](#)). Otherwise, cancel out of the dialog and change the study ID to create the study as a new one.

Studies in the **Study ID duplication** dialog may have been deleted, expired (i.e. studies for which all slides have been processed – see below), or may be current studies, still listed on the **Slide setup** screen. When an expired study is selected and restored to the study list the study is said to be “resurrected”.

See [Duplicate Study ID \(on page 219\)](#) for discussion of duplicate study IDs for LIS studies.

6.3.4.1 Merging Studies

If you edit a study ID to make it the same as an existing study ID, and then click **Use selected** in the **Study ID duplication** dialog that subsequently appears, all unprocessed slides from the edited study are moved into the existing study.



It is only possible to edit a study that has unprocessed slides; thus it is not possible to change a study to which processed slides are associated.

6.3.4.2 Processed Study Lifetime

When the last slide in a study has begun being processing, the study is (by default settings) removed from the Slide setup screen. The slides for the study will now be viewable in the Slide History screen.

You can set the BOND RX system to keep studies on the **Slide setup** screen for a set number of days after the last slides in the studies have been processed. Set this “processed study lifetime” in the administration client **Laboratory** screen (see [10.5.2 Study and Slide Settings](#)).

Expired studies are stored in the system, but cannot be viewed. Expired studies can be restored to the list by adding the study again (resurrecting it) or adding a slide to the study via the LIS.



Studies that do not contain any processed slides are never automatically cleared from the slide setup screen.

6.3.5 Editing a Study

To edit the details of a study, select it in the list then click **Edit study**. The software displays the **Study properties** dialog. You can use this in the same way as the **Add study** dialog described previously.



If you edit details of a study for which slide labels have been printed, print the labels again before attempting to run the slides (a message to this effect will appear on screen).

6.3.5.1 Deleting a Study

To delete a study, select it in the list then click **Delete study**.



When a study in the **Slide setup** screen of the BOND RX system contains only unprocessed slides, you can manually delete the study, making it “expired”. (All LIS studies expire automatically as soon as they contain no unprocessed slides.)



You cannot manually delete a study if it contains any processing or processed slides.



Deleting a study also deletes all unprocessed slides created for that study. You can recover the details of deleted studies but not their slides.

6.3.6 Copying a Study

Copying studies provides a convenient way to set up a new study. You can alter study details in the new study if you wish, or keep them the same. A new study number is automatically created, and you must enter a new study ID.



A study cannot be copied if it contains a slide that references a deleted protocol.

The copied slides are ready for label printing and processing on the **Slide setup** screen. Delete unwanted slides by right-clicking on them and selecting **Delete slide**.

To copy a study:

- 1 Select the study to copy in the study list at the left of the **Slide setup** screen.
- 2 Click **Copy study**; the software displays the **Copy study** dialog.
- 3 Enter a new Study ID and edit the details of the study as necessary.

4 Select **Unprocessed slides**, or **All slides** as required.

- Unprocessed slides - to copy only the unprocessed slides from the original study.
- All slides - to copy all the slides (unprocessed, processing and processed) from the original study. The system marks all slides in the new study as unprocessed.

5 Click **OK**.

The system creates the new study and copies the slides, including any comments, according to the selected option. All copied slides (including LIS) behave in the same manner as slides created in the **Add Slide** dialog (see [6.5.1 Description of Slide Fields and Controls](#)).

6.3.7 Daily Study Option

The BOND RX system can be configured so that it automatically creates a new study every 24 hours, allowing all the slides for each day to be created in the same study. This can save time for laboratories processing small numbers of slides, as study names and study IDs are not entered. Each daily study has the following properties:

- The study ID is set to the new day's date.
- The dispense volume and preparation protocol default to the system defaults set in the administration client. They can be edited.
- The **Study name** and **Researcher** fields remain empty and cannot be altered.

You can still create individual studies in the usual way if you want, with the daily study option on. See [10.5.2 Study and Slide Settings](#) for instructions to set the daily study option.

6.3.8 Study Report

You can generate reports for individual studies. The reports show basic study details and information about all the slides in the studies, e.g. slide IDs and the protocols and reagents used on them. There is space to write a comment for each slide if the report is printed. See [9.6 Study Report](#) for a complete description.

Generate study reports from the **Slide setup** and **Slide history** screens. Select the appropriate study or slide, then click the **Study report** button. Study reports only include reagent details for slides that have been processed and unlocked from the processing module.

6.4 Manage Researchers

The BOND RX system stores a list of researchers to optionally add to study details. Select from a list of “preferred” researchers in the **Add study** or **Study properties** dialogs, or add or edit researchers in the **Manage researchers** dialog, opened from the same study property dialogs.

The following fields are displayed for each researcher:

- Name: – the researcher’s name
- LIS ID: – a unique identifier supplied by a laboratory information system (if applicable)
- Pref. – researcher’s preferred status (only preferred researchers are available in the drop-down list when creating studies). This status is set in the **Edit researcher** dialog.

These values are also shown in the **Edit researcher** dialog. In addition, the **Edit researcher** dialog has:

- ID: – a unique ID automatically generated and assigned by the BOND RX system
- Comments: – editable field for a general comment or additional name information

With the **Manage researchers** dialog open, click **Add** or **Edit** to add new researchers or edit details of existing researchers. Edits are restricted to the comments field and changing the preferred status – you cannot change a researcher’s name after the researcher has been created.

You can delete researchers from the **Manage researchers** dialog. Studies already created with a deleted researcher continue to show the researcher’s name, but the researcher is not available for new studies. You cannot reuse a deleted researcher’s name for a new researcher.

6.5 Working with Slides

This section describes slide creation and management on the **Slide setup** screen. The final section describes the dispense volume setting and how it affects tissue placement on slides.

- [6.5.1 Description of Slide Fields and Controls](#)
- [6.5.2 Creating a Slide](#)
- [6.5.3 Copying a Slide](#)
- [6.5.4 Editing a Slide](#)
- [6.5.5 Deleting a Slide](#)
- [6.5.6 Manually Identifying a Slide](#)
- [6.5.7 Adding a Panel of Slides](#)
- [6.5.8 Dispense Volumes and Tissue Position on Slides](#)

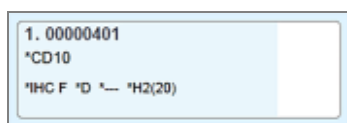
6.5.1 Description of Slide Fields and Controls

At the top of the slide list there are two buttons:

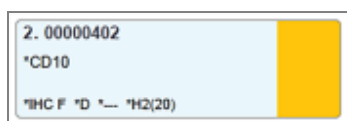
- Click **Add slide** to add a slide for the selected study.
- Click **Add panel** to add a panel for the selected study.

Refer to [6.5.7 Adding a Panel of Slides](#) for more details.

The slide list on the right of the screen displays details of slides for the study selected on the left of the screen. Each slide displays the slide ID and the details of the protocols to be run on that slide. The label areas on the right of the slides are color-coded to indicate where they were created as follows:



White:
Slide created in **Add slide** dialog
(see [6.5.2 Creating a Slide](#))

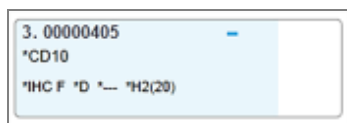


Yellow:
Slide created in the **Slide identification** dialog
(see [6.8 Impromptu Slide and Study Creation](#))

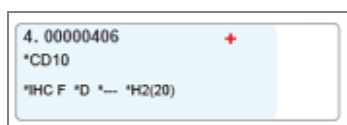


Light gray:
LIS slide
(see [11 LIS Integration Package \(on BOND RX Controller\)](#))

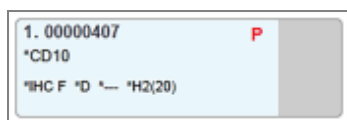
The slides also show the following symbols:



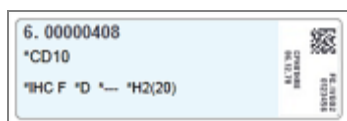
Minus sign:
Negative tissue slide (see step 4 in [6.5.2 Creating a Slide](#))



Plus sign:
Positive tissue slide (see step 4 in [6.5.2 Creating a Slide](#))



Red P:
LIS priority slide (see [11.2.5 Priority Slides](#))



Sample label:
Slide label has been printed



Mx:
Multiplex slide with three or more markers. Click the drop down arrow to display all markers for the slide.

Double-click a slide to open the **Slide properties** dialog for it. Right-click to delete the slide, or print a label for it.

6.5.2 Creating a Slide

To create a new slide:

- 1 Click on a study in the study list.
- 2 Click **Add slide** to display the **Add slide** dialog.

Figure 6-3: The **Add slide** dialog

The new slide is automatically numbered with a unique **Slide ID**, however this is not displayed until the slide is saved, when you click the **Add slide** button in the dialog.

- 3 Add a slide comment if you wish.
- 4 Select the tissue type (Test tissue, Negative tissue, Positive tissue) by clicking one of the radio buttons in the **Tissue type** group.
See [6.2.1 Control Tissue](#), and for more general discussion of controls, [14.3.2 Tissue Controls](#).
- 5 If necessary, change the dispense volume for the slide (see [6.5.8 Dispense Volumes and Tissue Position on Slides](#)).

- 6 Select the staining mode.
 - a In the **Staining mode** field, select **Single** (the default) if a single stain will be applied, or **Sequential multiplex** or **Parallel multiplex** for a multiplex stain slide (see [7.1.1 Staining Modes](#)).
 - b Select **Routine** (the default) in the second field.
 - c For Sequential multiplex staining, select the number of stains from the **Stains** drop-down list. You can select up to six stains.

The tabs displayed depend on the Staining mode selected:

- Single— the **Single** tab
- Parallel multiplex— **Parallel multiplex** tab.
- Sequential multiplex— a tab for each stain (up to six tabs), for example, **First** tab, **Second** tab, **Final** tab).

The screenshot shows the 'Add slide' configuration window. On the left, under 'My study', the following information is displayed: Study ID: 3688, Researcher: Smith, Slide ID, Study N°: 4, Study comments, and Date created: 7/2/2020 2:35:27 AM. The main configuration area includes a 'Slide comments' text box, 'Tissue type' (Test tissue selected), 'Dispense volume' (150 µL selected), 'Staining mode' (Sequential multiplex selected), and 'Stains' (4 selected). The 'Staining mode' dropdown is open, showing tabs for 'First', 'Second', 'Third', and 'Final', with 'First' selected and highlighted by a red box. Other settings include 'Process' (ISH selected), 'Marker', 'Protocols', and 'Preparation' dropdowns. At the bottom, there are 'Add slide' and 'Close' buttons.

- 7 On each tab displayed:
 - a Select the staining process (**IHC** or **ISH**).
 - b Select the primary antibody or probe from the **Marker** drop-down list:
 - c To run a negative IHC control reagent, select either the default negative reagent ***Negative** or a negative reagent you have created (refer to [14.3.3 Negative Reagent Control for IHC](#)).
 - d To run a negative ISH control reagent select ***RNA Negative Control Probe** or ***DNA Negative Control**.

- e To run a positive ISH control reagent select *RNA Positive Control Probe or *DNA Positive Control Probe.



To add or remove items from the **Marker** drop-down list, select or deselect the **Preferred** field for the reagent on the **Reagent Setup** screen of the software. See [8.2.1 Adding or Editing a Reagent](#) for more information.

- f Select the appropriate protocol for each processing stage.
- g When you select a primary antibody or a probe the software will enter default protocols. Check that the correct protocols are set for each stage and select a new protocol from the appropriate drop-down list if required. Select *--- if no protocol is required for a particular stage.
- Default protocols are set from the **Reagent Setup** screen. Refer to [8.2.1 Adding or Editing a Reagent](#).
- h To add or remove items from the **Protocol** drop-down lists, select or deselect the **Preferred** field for the protocol on the **Protocol setup** screen. See [7.2.1 Protocol Details](#) for more information.
- i For **ISH** slides, you can select a probe application protocol and a probe removal protocol. Or, you can choose to have no probe application protocol and no probe removal protocol.
- j If no probe application protocol or probe removal protocol is selected, then make sure the hybridization and denaturation protocols are also deselected.
- 8 For Single staining, you should generally leave the default of **Auto** for the Unique Product Identifiers (UPIs) on the left side of the dialog. However, if you want to select a specific lot number for a specific slide (e.g. for lot-to-lot validation), select from the drop-down list in the following fields:
- **Marker UPI** – UPI of the reagent container for the marker
 - **Detection System UPI** – UPI of the Detection System.

For slides to be processed on the same run (on BOND RX^m and BOND RX), either the UPIs must be the same, or **Auto** must be selected.

- 9 Click **Add slide**.

Add slide adds a slide with the details currently displayed in the **Add slide** dialog, then leaves the dialog open. This makes it easy to quickly add a number of slides for the selected study.

- 10 Click **Close** when you have finished adding slides for the study.

6.5.3 Copying a Slide



A slide cannot be copied if it references a deleted protocol.

To copy an existing slide:

- 1 Double-click the slide you want to copy, to open the **Slide properties** dialog.
- 2 Click **Copy slide**.

The dialog changes to **Add slide**, with an **Add slide** button.

- 3 Check the slide details and change as required.
- 4 Click **Add slide**.

The new slide, including any comments, will be added to the same study as the copied slide.

6.5.4 Editing a Slide

To edit the details of a slide on the **Slide setup** screen, double-click it to open the **Slide properties** dialog. Change the details as described in [6.5.2 Creating a Slide](#).



If you edit details of a slide for which a label has already been printed, reprint the label before processing the slide.

6.5.5 Deleting a Slide

To remove a slide from the slide list, right-click it in the slide list on the **Slide setup** screen, then select **Delete slide** from the submenu. You can also use the Delete key to delete the selected slide.

6.5.6 Manually Identifying a Slide


Any slide in the BOND RX system can be identified at any time. Click the **Search** icon  on the function bar to open the **Manual ID entry** dialog.

Figure 6-4: Manual ID entry dialog

For slides with two-dimensional barcode labels, for example those printed by the BOND RX system, scan the label to open the **Slide properties** dialog for the slide. Or, manually enter the 8-digit numeric ID, including leading zeros, and then click **Validate**.

6.5.7 Adding a Panel of Slides

A panel is a predefined set of markers with associated tissue types. Use panels to quickly add a number of slides with markers that are commonly used together – see [8.5 Reagent Panels Screen](#).

To add a panel of slides to a study, do the following from the **Slide setup** screen:

- 1 Click **Add panel**. The **Add slides from panel** dialog appears.
- 2 Select a panel from the drop-down list. The slides in the panel are displayed.
- 3 If necessary, exclude some of the slides by deselecting the check-boxes, then click **Add Slides**.

The BOND RX system adds the slides to the study.

- For ISH slides the dispense volume is automatically set to 150 μL .
- For IHC slides the dispense volume is set to the study default value.
- For all slides the preparation protocol is set to the study default.



Panels can be used to add slides with the Single or Parallel multiplex staining modes, but not Sequential multiplex.

6.5.8 Dispense Volumes and Tissue Position on Slides

The BOND RX software has two dispense volume settings, set for each slide in the **Add slide** dialog (see [6.5.2 Creating a Slide](#)).

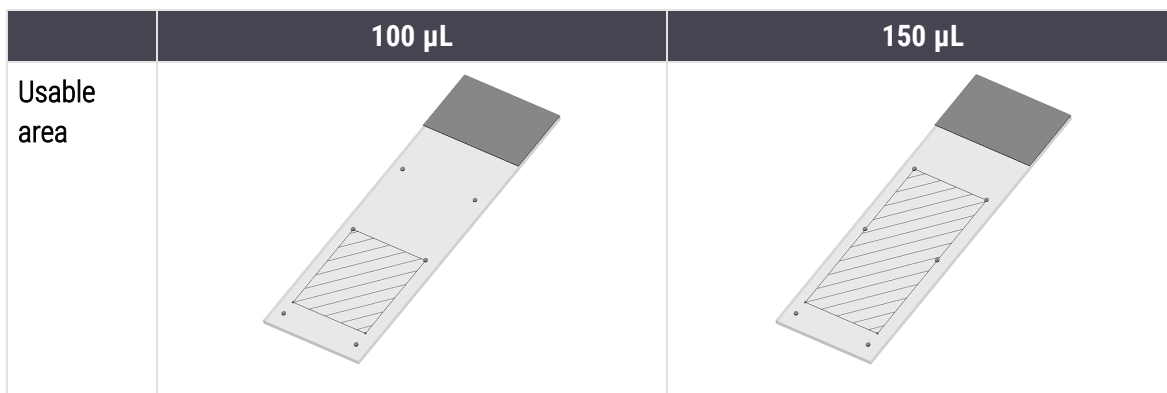
The 100 μL setting can only be used for IHC slides on the BOND RX^m processing module – all slides processed on the BOND RX and all ISH slides (on both processing module types) must use the 150 μL setting.

On the BOND RX and BOND RX^m Processing Modules, the dispense volume setting determines the position reagent is dispensed to on the slide as well as the volume dispensed:

- For 100 μL dispenses Covertiles are pulled back approximately half way down the slides and the aspirating probe delivers antibody at the top of the Covertiles (approximately half way down the slides).
- For 150 μL dispenses Covertiles cover most of the slides. Again, reagent is delivered at the top of the Covertiles, so a greater area of the slides receives reagent.

The difference in the areas of slides that receive reagent mean it is important to position tissue correctly. For 100 μL dispenses, typically only one sample can be stained, and it should be placed on the lower half of the slide (away from the label). For 150 μL dispenses, two tissue samples can more easily fit onto slides, or if there is only one it should be placed in the middle of the slide. Usable slide areas for the different dispense volume settings are shown in [Figure 6-5](#). Leica BOND Plus slides and Leica BOND Apex slides are marked to show the areas where tissue should be placed.

Figure 6-5: Usable slide areas for the different dispense volume settings



- The hatched areas show where tissue can be placed on slides with different dispense volumes.
- The position-marking dots shown are on Leica BOND Plus slides and Leica BOND Apex slides (see [2.6.1 Slides](#)).

The BOND RX processing module only dispenses in the 150 μL position – if slides with 100 μL dispense volumes are loaded you are unable to start processing.

For IHC slides on both the BOND RX^m and BOND RX, the volumes of antibody dispensed are as shown in the **Add slide** dialog – 100 μL or 150 μL . For ISH slides (for both processing module types) the 150 μL setting is enforced and the processing modules use the 150 μL Covertile and probe positions. However, the BOND RX system dispenses more than 150 μL of probe:

- for RNA probes, BOND RX dispenses 220 μL in two steps – 150 μL and 70 μL ;
- for DNA probes, BOND RX dispenses 240 μL in two steps – 150 μL and 90 μL .

Wash and other steps apply differing volumes, depending on the protocol.

Dispense Volume Defaults

For IHC on the BOND RX^m the dispense volume (150 μL or 100 μL) can be set for each individual slide, however the BOND RX software allows you to configure two levels of defaults. A system-wide default can be set (see [10.5.2 Study and Slide Settings](#)). This can be overridden for individual studies with study defaults, set in the **Add study** dialog (see [6.3.3 Adding a Study](#)). And finally the dispense volume can be set for individual slides in the **Add slide** dialog (see [6.5.2 Creating a Slide](#)).

Slides must all have the same dispense volume to be processed together in the same run (see [6.9 Slide Compatibility](#)).

6.6 Slide Labeling

All slides that are stained on the BOND RX system must be labeled in order to be identified in the software, so that the correct protocols are run on them. Slide labels created in the BOND RX system all have a label ID (rendered as a 2D barcode) that is used to automatically identify slides on processing modules. Labels created in an LIS (2D barcode IDs) can also be automatically identified. However, additional, human-readable information should always be included on slide labels so that slides can be identified if the label IDs cannot be automatically identified, if they are smudged, for example (see [10.3 Labels](#)).

Labels must be applied to slides before they are loaded onto the processing module. Take care that the labels are correctly attached so that the ID Imager can effectively scan (for 2D barcodes) the label IDs.

You must use slide labels supplied by Leica Biosystems for use with the BOND RX slide labeler.

- [6.6.1 Printing Labels and Applying to Slides](#)
- [6.6.2 Slide IDs and Label IDs](#)

6.6.1 Printing Labels and Applying to Slides

- 1 To print a label for a single slide, right-click on the slide, then select **Print label**. In this case, the **Print slide labels** dialog does not appear. In a BOND RX-ADVANCE system that includes defined pods, the default slide labeler will be used to print the label. Otherwise, the first slide labeler in the list will be used (see [10.6.3 Slide Labelers](#)).
- 2 When all of your slides have been set up, click **Print labels** on the **Slide setup** screen.
- 3 Select whether to print slide labels for:
 - All slide labels not yet printed – slides in all studies for which labels have not been printed.
 - All slide labels not yet printed for current study – slides in the current study for which labels have not been printed.
 - Current study – all slides for the currently selected study, including those previously printed.

Slide labels are printed in the order that their studies were created and, within each study, in the order that the slides were created.

- 4 Select the slide labeler to use.

(Set the default labeler in the administration client **Hardware** screen – see [10.6.2 Pods](#).)

- 5 Click **Print**.

When slide label printing is in progress, a flashing icon appears at the bottom left of the **Slide setup** screen.



- 6 Ensure the frosted area of the slide, where the label will be applied, is completely clean and dry.
- 7 Apply the label with the slide ID aligned parallel with the end of the slide. The label should be right side up (on the same side of the slide as the tissue).
 - Align the label squarely as the processing module cannot properly image misaligned labels.
 - Apply firm pressure to the whole BOND Printer Labels area to ensure attachment.
 - The printer label should be fully attached to the surface. There should be no overhang of the printer label over the edge of the slide.
 - If the label is immersed in liquid, allow it to dry prior to storage.

Figure 6-6: Place the label within the edges of the slide**Correct:**

Label is square and inside the edges of the slide.

**Incorrect:**

Label is over the edges of the slide.

**Incorrect:**

Label is crooked.



CAUTION: Position all parts of the label within all slide edges. An exposed sticky surface may cause the slide label (and slide) to stick to the Covertile or other equipment and damage the slide.



When the BOND Printer Ribbon and Labels roll are replaced, replace the ink ribbon with the same product number. Directions for the replacement of the Label rolls and ink ribbon are included in the box.



For printer labels that undergo prolonged reagent immersion or are subject to aggressive procedure, consider the following:

- Apply the slide label after the treatment has been performed.
- Apply a secondary identifier on the surface of the slide.
- Avoid or limit immersion of the BOND Printer Ribbon and Labels.
- Apply a protective overlay.



Used ink rolls will have reverse images of the printed information. If the information contains personally identifiable information, the used ink rolls should be disposed of according to laboratory procedures and/or local privacy regulations.



Adhesive and ink durability are subject to customer testing conditions. Use of the BOND RX Printer Ribbon and Labels must be verified by the laboratory for their procedures and conditions.

6.6.1.1 External Dewaxing and Epitope Retrieval

Dewaxing and epitope retrieval, if this is being done externally to the BOND RX system, is best done after labeling the slides. This avoids slides drying out while you enter the details of the slides and set up the BOND RX system to run the required protocol(s), and also avoids difficulties in labeling wet slides following these steps.



If you are using xylene for dewaxing off the processing module, avoid touching the label so the printing does not become smudged.



Extended soaking in, or exposure to, benzene derivatives, D-Limonenes and Aliphatic Hydrocarbons, Acetone, Water and Aqueous based reagents can reduce the effectiveness of the slide ID label adhesive and possible loss of print integrity. We recommend that labels should not be submerged for extended period. See specific product information on the LBS website.



CAUTION: For dewaxing on BOND RX and BOND RX[™] Processing Modules, use only BOND Dewax Solution.

Do not use xylene, xylene substitutes or other reagents that can degrade parts of the processing modules and cause fluid leakage.

6.6.2 Slide IDs and Label IDs

The BOND RX system provides a unique “Slide ID” every time a new **slide** is created. The BOND RX system also creates a unique “Label ID” every time a **slide label** is printed. The Label ID is a 2D Barcode.



For LIS slides the Slide ID may be defined by the LIS and could be any numeric value (with 8 digits or less).

6.6.2.1 Slide Identification

When the labels are placed onto slides the system can identify the slides in each position in the slide staining assemblies (refer to [5.1.5.1 Automatic Slide Identification](#)).

Slides without slide IDs, or with unrecognized slide IDs, must either be manually identified to the system (refer to [5.1.5.2 On-Board Manual Slide Identification](#)), or a label printed and placed on the slide and the slide imaged again.

Configure the information to display on slide labels on the administration client **Label configuration** screen (see [10.3 Labels](#)).

6.7 Slide Setup Summary Report

The slide setup summary lists all slides (for all studies), currently configured on the **Slide setup** screen. The slides are grouped by study with details such as marker and dispense volume provided. At the bottom of the report is a list of all the reagents and reagent systems required by the slides in the report, with the number of tests for each. There are separate lists for each BOND Processing Module.

The report is a valuable aid in run preparation. It helps you to ensure that the slides put onto each tray are compatible (see [6.9 Slide Compatibility](#)), and shows the reagents and reagent systems that need to be loaded.

To create a slide setup report, click **Slide setup summary**.

For each slide the report shows the following information:

Field	Description
Slide ID	The BOND RX system assigns a unique identifier to each slide
Marker	The marker(s)
Staining protocol	The staining protocol
Preparation	The preparation protocol (if any)
HIER	HIER protocol (if any)
Enzyme	Enzyme retrieval protocol (if any)
Dispense volume	The volume of reagent to be dispensed (see 6.5.8 Dispense Volumes and Tissue Position on Slides)
Tissue type	Test tissue, positive control tissue, or negative control tissue

For sequential multiplex staining slides, up to six rows are shown in the Marker, Protocols, Dispense volume, and Tissue type columns, grouped by the Slide ID.

See [3.7 Reports](#) for further details about the report window and printing options.

6.8 Impromptu Slide and Study Creation

By default, the BOND RX system is configured so that new studies and slides can be created after a slide tray has been loaded into a processing module and the slides imaged.

The first section below gives directions for this “impromptu” study and slide creation. The second section describes option settings for alternative workflows.

- [6.8.1 Creating New Studies and/or Slides After Imaging](#)
- [6.8.2 On-Board Slide Identification Options](#)

6.8.1 Creating New Studies and/or Slides After Imaging

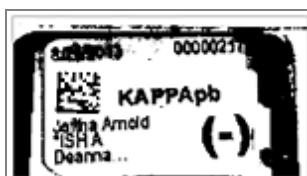
Follow the procedure below to add study and slide information after slides have been loaded and imaged (the procedure is similar to the assisted-ID procedure described in [5.1.5.2 On-Board Manual Slide Identification](#), but now includes creation of new studies and slides).

- 1 Load slides onto the processing module in the usual manner.

There is no need to create studies or slides in the BOND RX software or print labels – hand written or third party labels can be used.

The system will not recognize the slides so will display images of the labels.

Figure 6-7: Slide not automatically identified

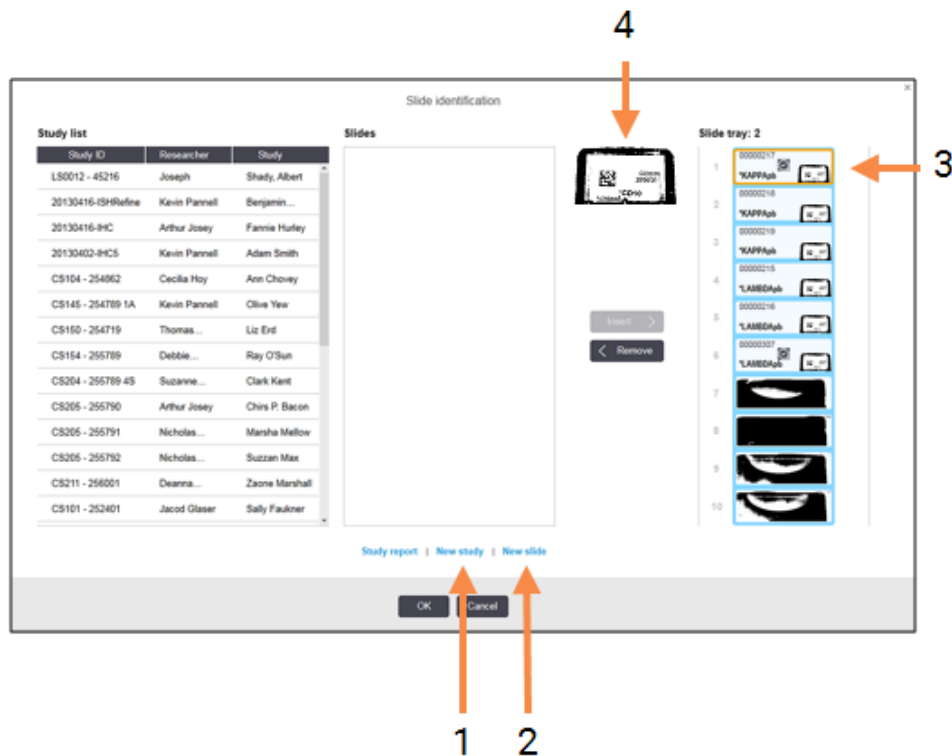


If label images are consistently not displayed for a particular processing module, it may be configured not to capture images of the slide labels. Contact customer support to arrange for this setting to be reconfigured for the processing module.

- 2 To launch the **Slide identification** dialog do one of the following:
 - a Double-click on the slide image.
 - b Right-click on the image and select **Select manually** from the submenu.

The **Slide identification** dialog appears with **New study** and **New slide** buttons available (items 1 & 2 in [Figure 6-8](#)).

Figure 6-8: Slide identification dialog with slide status display



In some laboratories the **New study**, or both **New study** and **New slide** buttons may be disabled – see [6.8.2 On-Board Slide Identification Options](#)

The active slide is highlighted on the slide tray (item 3).

The dialog includes an enlarged image of the label (item 4) to assist with slide identification. Hold the cursor over the slide in the right-hand pane to see an even greater enlargement of the label.

The left-hand pane lists all studies with current slides. Under default settings, only studies with slides for which labels have been printed appear (you can change this to include studies with slides for which labels have not been printed, see [6.8.2.2 External Slide Labels](#)).

The center pane shows slides configured for the study selected in the left-hand pane, where the slides have not yet been matched to any slides imaged on the processing module. Again, under default settings, only slides that have had labels printed appear, but this can be changed to show all slides configured for the study.



Ensure that you select the correct label image, as your slides could be impacted if you choose incorrectly.

- 3 To create a new study, click **New study** (item 1).
Create a new study for the selected slide in the normal manner (refer to [6.3.3 Adding a Study](#)).
- 4 After you click **OK** in the **Add study** dialog, select the new study in the study list in the **Slide identification** dialog.
- 5 To create a new slide for the study you just created, click **New slide** (item 2).
This opens the **Add slide** dialog.
- 6 Create a new slide in the software for the physical slide selected in the right-hand pane, in the normal manner (refer to [6.5.2 Creating a Slide](#)).
When it is added, the new slide is displayed in the center pane of the dialog (i.e. while the new study remains selected in the left-hand studies list).
- 7 Ensuring that the correct label image is still selected in the right-hand pane, click **Insert** to match it with the new slide in the center pane.
The slide is removed from the center pane and the label image in the right-hand pane replaced to show the system information for the slide, as it was entered for the new slide you have just created.
If you match slides incorrectly, you can undo this step by selecting the slide in the right-hand pane and clicking **Remove**.
- 8 The slide can now be processed in the usual manner.
Repeat the procedure of creating new studies and slides for remaining slides in the slide tray.

6.8.2 On-Board Slide Identification Options

Settings in the administration client can allow or enforce different slide identification workflows, by selectively enabling or disabling options in the **Slide identification** dialog.

6.8.2.1 Restrict or Disallow Impromptu Study and Slide Creation

By default the BOND RX system allows you to load slides that have not been created in the BOND RX software (or imported from an LIS), and to create the studies and slides in the software after the slides have been imaged, using the **Slide identification** dialog. Optionally, you can set the system to disallow the creation of new studies this way (but still allow creation of new slides for existing studies), or completely disallow creation of slides (and studies) after loading slides. Depending on your setting the **New study**, or both **New study** and **New slide** buttons in the **Slide identification** dialog are disabled (see [Figure 6-8](#)).

Restrict impromptu case and slide creation options in the administration client **Settings** screen (see [10.5.2 Study and Slide Settings](#)).

6.8.2.2 External Slide Labels

You can set the BOND RX system to require, or not, that all slides must be printed by the BOND RX system before they can be processed. There are separate settings for LIS slides and non-LIS slides.

For non-LIS slides the default setting requires printing by the BOND RX system. This means that physical slides without labels printed by the BOND RX system are not automatically matched with slides created for them in the software (even if the IDs are the same). Furthermore, you cannot match the slides manually using the **Slide identification** dialog, because only slides that have been printed by the BOND RX system are displayed there. Consequently, laboratories without BOND–LIS integration that hand-write labels or print them on third-party equipment, must set this option off. This makes all slides created within the system available for matching with slides loaded onto the processing module, irrespective of whether the BOND RX system printed the labels or not.

To enable processing of slides that have not had labels printed by the BOND RX system, deselect **Force printing in BOND RX** in the administration client **Settings** screen (see [10.5.2 Study and Slide Settings](#)). (It is not necessary to deselect **Force printing in BOND RX** just to allow impromptu study and slide creation – see [6.8.2.1 Restrict or Disallow Impromptu Study and Slide Creation](#).)



Slides created prior to deselecting the **Force printing in BOND RX** option will not be available for processing until their labels are printed, although slides created after deselecting this option will not need to have labels printed.

For LIS slides the default setting does not require printing by the BOND RX system. This means that slides with labels printed by the LIS can be automatically matched to the slides in the BOND RX software (imported from the LIS). Or, if an automatic match cannot be made (if, e.g. a slide label is smudged), you can manually match slides using the **Slide identification** dialog. However, if your workflow has slides created in an LIS but you want to enforce that labels are printed by the BOND RX system, turn the option on (select **Force LIS printing in BOND RX** on the administration client **LIS** screen – see [10.2 LIS](#)).

6.9 Slide Compatibility

In order that the steps in each run are synchronized in a way that ensures optimal results for all the slides on the tray, the slides are checked for compatibility by the BOND RX software when the slide trays are loaded. Incompatible slides are shown in the **System status** screen. You must remove or replace incompatible slides before starting the run (see [5.1.4.4 Fixing Incompatible Slide Setup](#)).

For routine slides to be compatible they must:

- have the same dispense volume;
- be all single-stain or all parallel multiplex stain or all sequential multiplex stain;
- have the same UPI when it has been specifically selected during the Add Slide process;
- use the same preparation protocol;
- use staining protocols that use the same preferred detection system and have the same step sequence (ie, dispense type and incubation time); and
- use compatible pretreatment protocols and/or ISH denaturation and hybridization protocols.

Rules for protocol compatibility are provided in [6.9.1 Protocol Compatibility](#).

Slide setup reports ([6.7 Slide Setup Summary Report](#)) provide some assistance to help ensure you load compatible slides onto each tray.

6.9.1 Protocol Compatibility

Staining and preparation protocols have rigid compatibility constraints, while for heat and enzyme pretreatment protocols, and ISH hybridization and denaturation protocols, there is some room for variation. Compatibility for these protocols depends on the processing module type (BOND RX or BOND RX^m), number and duration of the protocol steps and processing module states during the steps. The protocols are compatible when these factors are all the same or differ in ways that can be accommodated without affecting staining quality.

Compatibility rules for all protocol types are listed below.

6.9.1.1 Staining Protocols

For each slide, use staining protocols that use the same preferred detection system and have the same step sequence (ie, dispense type and incubation time). For sequential multiplex staining runs, the same staining protocols must be used, in the same order.

IHC and ISH slides cannot be mixed in single-stain runs, but can be combined in sequential multiplex staining runs.

6.9.1.2 Preparation Protocols

For “dewax” and “bake and dewax” protocols

- The same protocol must be used for all slides in the tray; and
- Slides with a preparation protocol cannot be mixed with slides without a preparation protocol.

6.9.1.3 Pretreatment Protocols

Slides with heat retrieval only, enzyme retrieval only, heat *and* enzyme retrieval, and no epitope retrieval at all, can all be run together. Slides not receiving the current pretreatment are hydrated while the protocol runs on the other slides (heat-induced retrieval always precedes enzyme-induced retrieval).

Similarly, all combinations of slides with and without ISH denaturation and hybridization are compatible.

The sections below give conditions for compatibility of pretreatment protocols with protocols of the same pretreatment type.

Heat Pretreatment

1 Heat pretreatment protocols are compatible when they have:

- the same number of steps; and
- the same incubation times for each step, except for heated steps.

For concurrent heat steps the longest duration set for the step is used for all slides. Slides with shorter set durations are heated just for the period configured for them, after which power to the slide heater is turned off.

2 Protocols using epitope retrieval solutions 1 and 2 can be mixed in runs.

3 Slides using heat pretreatment can be run in trays with slides not using heat pretreatment – the slides not receiving pretreatment are hydrated with epitope retrieval solution at ambient temperature while the other slides are processed.

Enzyme Pretreatment

1 Enzyme pretreatment protocols are compatible when they have:

- the same number of steps; and
- the same incubation times for each step.

2 Up to 2 enzyme types can be applied in a run.

3 Slides using enzyme pretreatment can be run in trays with slides not using enzyme pretreatment – the slides not receiving pretreatment are hydrated at ambient temperature while the other slides are processed.

6.9.1.4 ISH Denaturation

Denaturation protocols are compatible when they have the same incubation times. Incubation temperatures can differ.

6.9.1.5 ISH Hybridization

Hybridization protocols are compatible when they have the same incubation times. Incubation temperatures can differ.

7

Protocols (on BOND RX Controller)

In the BOND RX software, protocols are the series of steps performed to stain tissue samples.

Your BOND RX system is supplied with a set of predefined Leica Biosystems protocols that cannot be edited or deleted. The predefined protocols have been validated by Leica Biosystems. However, customized protocols can be created by copying and editing existing predefined protocols.



WARNING: All customized protocols must be validated in accordance with the local laboratory procedures and requirements. The ability to create and save a protocol does not indicate that it is suitable for the intended task.

This chapter has the following sections:

- [7.1 Protocol Types](#)
- [7.2 Protocol Setup Screen](#)
- [7.3 Creating New Protocols](#)
- [7.4 Editing User Protocols](#)
- [7.5 Protocol Reports](#)
- [7.6 Predefined Protocols](#)

7.1 Protocol Types

All protocols in the BOND RX system have a “type” according to the specific functions they are intended to perform. For example, prestaining HIER protocols are one type, IHC sequential multiplex staining protocols another.

- The type of a protocol cannot be changed.
- To create a new protocol you must copy an existing protocol of the type you want the new protocol to be. You can then edit the protocol steps as required.

Typically, in any processing run, a number of protocols of different types are run in order to prepare the slides, apply the markers, and then apply chromogen. These sequences and the protocols they use typically require modification for multiplex stains.

- [7.1.1 Staining Modes](#)
- [7.1.2 Protocol Sequences](#)

7.1.1 Staining Modes

The BOND RX system has three staining modes:

- **Single** – the application of a single marker and chromogen to a single slide.
- **Parallel multiplex** – the application of up to six different markers and chromogens to a single slide. Markers are mixed together in a “cocktail” and applied with a single staining protocol.
- **Sequential multiplex** – the application of up to six different markers and chromogens to a single slide. Markers are applied one after the other in separate staining protocols.

Each staining protocol has a “staining method” to indicate its role with respect to multiplex or single staining.

Single staining has only the one staining method of “Single”.

Parallel multiplex staining has only one staining method of “Parallel multiplex”.

For parallel multiplex staining, if a suitable predefined parallel multiplex staining protocol is not available to use or copy from, edit an existing Parallel Multiplex staining protocol (such as Chromoplex) to include more chromogens.

Predefined sequential multiplex staining protocols have the staining method “Single”, which cannot be changed. However, you have the option to copy these predefined protocols and modify the staining method to suit your requirements.

Sequential multiplex staining has the following staining methods:

- **Preliminary** – used for all protocols before the last one in a sequential multiplex stain
- **Final** – used as the last protocol in a sequential multiplex stain

For example, a user-created protocol can be configured for use as a single protocol, or it can also be configured for use as a preliminary protocol and/or a final Protocol. Make sure you review the entire protocol to ensure that all steps are appropriate for all staining methods (for example a single protocol has a counter-stain, which is not needed for the preliminary protocols).

Protocol types and staining methods are displayed in the table below:

Type		Staining Method	Description
Staining	IHC Staining—Single staining	Single	Protocol for detection of a single antibody for a single stain
	IHC Staining—Sequential multiplex staining	Single	Protocol for detection of a single antibody for a single stain
		Preliminary	Protocol for detection of the first antibody in sequential multiplex stain
		Final	Protocol for detection of last antibody in sequential multiplex stain
	IHC Staining Parallel multiplex staining	Parallel multiplex	Protocol for detection of cocktail antibodies in parallel multiplex stain
	ISH Detection—Single	Single	Protocol for detection of a single probe for a single stain
	ISH Detection—Sequential multiplex staining	Single	Protocol for detection of a single probe for a single stain
		Preliminary	Protocol for detection of the first probe in sequential multiplex stain
		Final	Protocol for detection of last probe in sequential multiplex stain
	ISH Detection Parallel multiplex staining	Parallel multiplex	Protocol for detection of cocktail probes in parallel multiplex stain (currently there are no protocols in this category)
Prestaining	Preparation	N/A	Dewax, or bake slide (for tissue adhesion) then dewax tissue
	Heat Pretreatment	N/A	Epitope retrieval using heat
	Enzyme Pretreatment	N/A	Epitope retrieval using enzymes
	ISH Probe Application	N/A	Probe application protocol for ISH
	ISH Denaturation	N/A	Denaturation protocols for DNA ISH
	ISH Hybridization	N/A	Hybridization protocols for ISH
	ISH Probe Removal	N/A	Probe removal protocol for ISH

7.1.2 Protocol Sequences

Typically, for each slide, a sequence of protocols of different types is applied. This is a selection of preparation, epitope retrieval, denaturation, hybridization and staining protocols, as appropriate for the tissue, marker, and general laboratory procedures. These sequences can be set for each slide individually at slide creation (see [6.5.2 Creating a Slide](#)), however the BOND RX software also allows you to set default protocols to speed up slide creation when specialized protocols are not required:

- a default preparation protocol (e.g. *Dewax) is set for the entire BOND RX system in the administration client (see [10.5.2 Study and Slide Settings](#));
- defaults for all other protocol types are set for each marker, from the **Reagent Setup** screen (see [8.2.1 Adding or Editing a Reagent](#)).

Set suitable default protocols so that time spent preparing individual slides is minimized. You can change protocols for individual slides if you need, when slides are created.

The order in which the protocols in a sequence are run is automatically set by the BOND RX software and is shown in the table below.

Order	Protocol (or Probe)	IHC or ISH	Comment
1	Preparation	Both	Optional on-board removal of wax in preparation for chemistry.
2	HIER (heat-induced epitope retrieval)	Both	For most slides either an HIER or EIER protocol is run – on occasions, both, or neither.
3	EIER (enzyme-induced epitope retrieval)	Both	
4	Probe application	ISH	You can choose a specific probe application protocol, or none. Note: If you select a probe application protocol, then you must select a hybridization protocol and a probe removal protocol. If you do not select a probe application protocol, then you must deselect the hybridization protocol and the probe removal protocol.
5	Denaturation	ISH	Denaturation protocol for DNA probes. DNA probes should always use denaturation.
6	Hybridization	ISH	Required hybridization protocol for ISH, or none.

Order	Protocol (or Probe)	IHC or ISH	Comment
			Note: If you select a probe hybridization protocol, then you must select a probe application protocol and a probe removal protocol. If you do not select a Hybridization protocol, then you must deselect the probe application protocol and the probe removal protocol.
7	Probe removal	ISH	You can choose a specific probe removal protocol, or none. Note: If you select a probe removal protocol, then you must select a probe application and hybridization protocol. If you do not select a probe removal protocol, then you must deselect the probe application and hybridization protocols.
8	Staining	Both	Required protocol for application of chromogen and associated reagents. IHC primaries are dispensed in this protocol.

The protocols selected for protocol sequences can be predefined or you can create customized protocols and select these (see [7.3 Creating New Protocols](#)).

7.1.2.1 Protocols and Protocol Sequences for sequential multiplex staining

Sequential multiplex stains essentially run between two and six staining protocol sequences one after the other. These can be any combination of IHC protocols and/or ISH protocols.

Sometimes the subsequent prestaining protocols in the sequence can be skipped (ie, Epitope Retrieval, Denaturation, etc), or if included, may need to be modified (i.e. reduce retrieval temperature). Steps in the sequential staining protocols (preliminary or final) should also, typically, be modified (the protocols necessarily need some modification to have the appropriate staining method set – see [7.1.1 Staining Modes](#)). Some suggestions for protocol and protocol sequence modifications for sequential multiplex staining are given below. In all cases you should run your own tests to verify results.

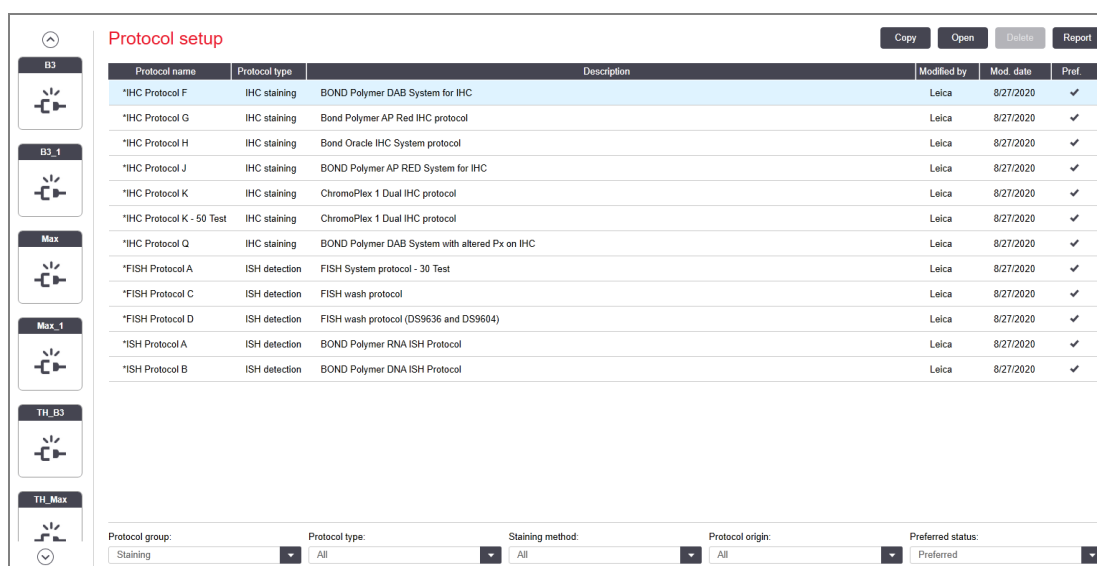
- Preparation protocols (e.g. Dewax) can only be run in the sequence for the first marker – the software does not allow selection of a preparation protocol for the two to six staining protocol sequences.
- Epitope retrieval may be only required once, before application of the first marker. If additional retrieval is required for subsequent markers a shorter duration or lower temperature may be adequate.
- Epitope retrieval protocol modifications can be used to strip previous antibodies to allow sequential staining (i.e. a chromogenic multiplex sequential stain using all mouse markers).
- If multiplex staining with multiple DNA probes, denaturation is often only required once, before application of the first marker. If additional denaturation is required for additional markers, it typically requires shorter duration.

- For sequential staining protocols the counterstain segment is removed from the preliminary protocols and added to the final protocol.
- When designing a chromogenic sequential stain, the order of chromogen applications needs to be considered. Some chromogens do not perform well when run after another chromogen (e.g. Fast Red works better when used after DAB chromogen).
- When designing a fluorescent sequential stain, the order of fluorophore applications needs to be considered. Some fluorophores do not perform well when run after another fluorophore.

7.2 Protocol Setup Screen

To work with protocols, click the **Protocol setup** icon  on the function bar.

Figure 7-1: Protocol setup screen



The screenshot shows the 'Protocol setup' screen with a table of protocols and filter options. The table has columns for Protocol name, Protocol type, Description, Modified by, Mod. date, and Pref. The filter options at the bottom are: Protocol group: Staining, Protocol type: All, Staining method: All, Protocol origin: All, Preferred status: Preferred.

Protocol name	Protocol type	Description	Modified by	Mod. date	Pref.
*IHC Protocol F	IHC staining	BOND Polymer DAB System for IHC	Leica	8/27/2020	✓
*IHC Protocol G	IHC staining	Bond Polymer AP Red IHC protocol	Leica	8/27/2020	✓
*IHC Protocol H	IHC staining	Bond Oracle IHC System protocol	Leica	8/27/2020	✓
*IHC Protocol J	IHC staining	BOND Polymer AP RED System for IHC	Leica	8/27/2020	✓
*IHC Protocol K	IHC staining	ChromoPlex 1 Dual IHC protocol	Leica	8/27/2020	✓
*IHC Protocol K - 50 Test	IHC staining	ChromoPlex 1 Dual IHC protocol	Leica	8/27/2020	✓
*IHC Protocol Q	IHC staining	BOND Polymer DAB System with altered Px on IHC	Leica	8/27/2020	✓
*FISH Protocol A	ISH detection	FISH System protocol - 30 Test	Leica	8/27/2020	✓
*FISH Protocol C	ISH detection	FISH wash protocol	Leica	8/27/2020	✓
*FISH Protocol D	ISH detection	FISH wash protocol (DS9636 and DS9604)	Leica	8/27/2020	✓
*ISH Protocol A	ISH detection	BOND Polymer RNA ISH Protocol	Leica	8/27/2020	✓
*ISH Protocol B	ISH detection	BOND Polymer DNA ISH Protocol	Leica	8/27/2020	✓

The **Protocol setup** screen has a table that lists each protocol along with some basic details. Predefined protocols have an asterisk (*) as the first character in their name and abbreviated name.

You are able to select a protocol from this table for operations such as copying, editing and report generation. These operations are accessed through buttons above the table or the right-click menu.

Filters below the table allow you to set the type of protocol to display. You can select between staining and prestaining protocols, and further refine this to show specific protocol types (see [7.1 Protocol Types](#)). Additionally, you can filter the staining method, protocol origin, and preferred status.

The information in the protocol list is described below:

Title	Description	Options
Protocol name	Full name of the protocol	Predefined (Leica Biosystems) protocols always begin with an asterisk (*)
Protocol type	Describes the function of the protocol	See 7.1 Protocol Types
Description	Describes the protocol's function and application	
Modified by	Identifies who created or last modified the protocol	Leica indicates a predefined Leica Biosystems protocol
Mod. date	The date the protocol was created or last modified	
Pref.	Displays the protocol's preferred status	Checked – this is a preferred protocol, available for selection in the Add Reagent and Add Slide dialog. Not checked – this is not a preferred protocol, and is unavailable for selection in the Add Reagent and Add Slide dialog

7.2.1 Protocol Details

To open a protocol listed in the **Protocol setup** screen for viewing or editing, double-click it (or highlight it, then click **Open**). The software displays the **Edit protocol properties** dialog with the protocol's details.

For predefined Leica Biosystems protocols only the preferred setting is editable, but other settings can be changed for user protocols.

Figure 7-2: The **Edit protocol properties** dialog for a user protocol

Edit protocol properties

Name: MyIHC Protocol F
 Abbreviated name: MyIHC F
 Description: Bond Polymer Refine IHC protocol
 Staining method: Single Preliminary Final Preferred
 BOND RX[™]: BOND RX [Import protocol](#) Protocol type: IHC staining
 Preferred detection system: Bond Polymer Refine Detection

Step N°	Wash	Reagent	Supplier	Ambient	Temperature	Inc. (min)	Dispense type	Ramp
1		*Peroxide Block	Leica Microsystems	✓		5:00	Selected vol.	After dispense
5		*MARKER	Leica Microsystems	✓		15:00	Selected vol.	After dispense
9		*Post Primary	Leica Microsystems	✓		8:00	Selected vol.	After dispense
13		*Polymer	Leica Microsystems	✓		8:00	Selected vol.	After dispense
17		*Mixed DAB Refine	Leica Microsystems	✓		0:00	Selected vol.	After dispense
18		*Mixed DAB Refine	Leica Microsystems	✓		10:00	Selected vol.	After dispense
22		*Hematoxylin	Leica Microsystems	✓		5:00	Selected vol.	After dispense

Show wash steps [Insert wash](#) | [Insert reagent](#) | [Delete step](#)

[Save](#) [Cancel](#)

The dialog displays a tab for each processing module type (BOND RX^m and BOND RX) that is commissioned for the pod (or both tabs if none are commissioned).

There is also an **Import protocol** button that appears when you are creating a new protocol, or when editing a user protocol. See [7.4.4 Multiple Processing Module Types and Protocol Versions](#) for details.

Select **Show wash steps** below the table to view all protocol steps (including wash steps). Deselect to hide the wash steps.

The **Edit protocol properties** dialog displays the following protocol information:

Name	The protocol's full name.
Abbreviated name	The protocol's abbreviated name, used, for example, on slide labels.
Description	A brief statement describing the protocol.
Staining method	(See below)
Protocol type	The type indicates the protocol's function and determines allowable steps and reagents.
Preferred detection system	The preferred detection system for this protocol. This does not apply to prestaining protocols.

A table below the protocol information in this dialog lists each protocol step and its properties (see [Figure 7-2](#)). The editable steps in user protocols are edited within this table (see [7.4 Editing User Protocols](#)).

The following details are shown in the table:

Item	Description
Step No.	The order in which the steps of the protocol will be performed.
Wash	Checked if the step is a wash step.
Reagent	The reagent used in the step.
Supplier	The supplier of the reagent. This is not editable.
Ambient	Checked if the step is at ambient temperature.
Temperature	The selected slide temperature if other than ambient.
Inc. (min)	The minimum time the reagent will remain on the slide.
Dispense type	The actual dispense volume, an open dispense or an intermediate dispense.

7.2.1.1 Staining Method

Staining protocols include a “staining method” section. Single stain and sequential multiplex stain protocols have the following options:

- **Single** – protocol is for single stains
- **Preliminary** – all protocols except the last one for a sequential multiplex stain
- **Final** – the last protocol of a sequential multiplex stain

Parallel multiplex stain protocols have only one staining method option: **Parallel multiplex**.

See [7.1.1 Staining Modes](#) for further discussion of staining methods.

7.2.1.2 Preferred Status

Only preferred protocols are available for selection in the **Add Reagent** and **Add Slide** dialog, so protocols you intend to use should be made preferred. To do this select the **Preferred** checkbox – deselect to make not preferred.

7.3 Creating New Protocols

You can create new protocols by copying existing user or Leica Biosystems protocols. When you copy a protocol, the type of protocol remains fixed and cannot be altered later. Thus if you wish to create a new IHC protocol you must copy an existing IHC protocol; for an HIER protocol, copy an existing HIER protocol and so on.

Template Protocols (see [7.6.2 Template Protocols](#)) cannot be run as provided, but must be copied in order to associate a detection system. Copy the template and make the required changes.

To copy a protocol, select it from the list in the **Protocol setup** screen then click the **Copy** button. A copy of the selected protocol will now appear in the **New protocol properties** dialog ready for editing.

The new protocol will require a unique name and abbreviated name that must comply with all the rules specified in [7.4.3 Protocol Rules](#). Other than changing the protocol’s name and abbreviated name, you do not need to change any other part of your new protocol. However, you can, of course, alter any aspect of the protocol as described in [7.4 Editing User Protocols](#).

See [7.4.4 Multiple Processing Module Types and Protocol Versions](#) for rules applying to research protocols.

After editing BOND RX or BOND RX^m, click **Save**. If the protocol complies with the rules, you will be asked to confirm that you are creating a protocol “at your own risk”. This message is a reminder that Leica Biosystems cannot predict the quality of results from any user-created or edited protocol. Once you confirm that you are happy to continue, the protocol changes will be saved.

7.4 Editing User Protocols

You are able to edit user protocols (but not Leica Biosystems protocols) using the **Edit protocol properties** dialog. To edit a protocol, select it from the list in the **Protocol setup** screen then click **Open** (or double-click the protocol). Alternatively, configure a new protocol by copying an existing protocol of the same type, and editing it (see [7.3 Creating New Protocols](#)).

In staining protocols, reagent steps can be added and removed, and new reagents, temperature, dispense type and incubation times set. Additional wash steps can be added or removed.

For Dewax protocols, the number of steps can be changed. For other prestaining protocols (HIER, enzyme), temperatures and incubation times for some steps can be changed. See [7.4.3 Protocol Rules](#) for a list of allowable edits.

As you edit a protocol, changed or new steps that have all the required information have a green bar at the left-hand side. Steps that require additional information have a red bar.

During editing, you can view all protocol steps or hide the wash steps, using the **Show wash steps** option button below the table.



For most protocol steps set incubation times under 30 minutes. Times greater than this may cause tissue to dry out. If a longer incubation time is required, either:

- Duplicate the step one or more times and divide the required period between the steps. The one exception is ISH hybridization steps, which are always longer than 30 minutes and should never be broken into shorter steps.
- Change the dispense type to Intermediate.

This section includes the following topics:

- [7.4.1 Editing Protocol Steps](#)
- [7.4.2 Adding and Removing Protocol Steps](#)
- [7.4.3 Protocol Rules](#)
- [7.4.4 Multiple Processing Module Types and Protocol Versions](#)
- [7.4.5 Deleting Protocols](#)

7.4.1 Editing Protocol Steps

Follow the instructions below to configure a new protocol in the **New protocol properties** dialog, or edit an existing protocol in the **Edit protocol properties** dialog. See [7.4.3 Protocol Rules](#) to ensure you create a valid protocol.

Each time you save a protocol, a copy is stored in the system. When you create a protocol report (see [7.5 Protocol Reports](#)), you need to select the date on which protocol was active. To avoid having multiple, redundant, protocol versions, save protocols only when you have finished configuration.

- 1 For new protocols, type in a protocol name and abbreviated name.
- 2 Optionally type in a protocol description.

- 3 Set the staining method of staining protocols (see [7.1.1 Staining Modes](#)).
- 4 Set the **Preferred** status of the protocol (see [7.2.1.2 Preferred Status](#)).
- 5 For staining protocols, select a detection system or research reagent system for use with the protocol, from the **Preferred detection system** drop-down list.
- 6 Add or remove protocol steps (see [7.4.2 Adding and Removing Protocol Steps](#)) until you have the required number of steps for the protocol.
- 7 For BOND RX^m and BOND RX only, change editable parameters in new and existing protocol steps by first double-clicking the parameter you want to change:
 - a For BOND RX^m and BOND RX, select a reagent from the drop-down list.

Note: Select *MARKER to indicate the step where the primary antibody is used in IHC protocols. Only *BOND Wash Solution or *Deionized Water can be used for wash steps.
 - b Set incubation time in minutes and seconds (mm:ss). This is the minimum time the slide sits before the following step. See Step 6 in [7.4.3 Protocol Rules](#) for incubation time limits.

In general, for reagent application steps, Leica Biosystems recommends incubation times no greater than 30 minutes.
 - c Set temperature (for staining protocols and some steps in prestaining protocols).

If you want to set a temperature that is not ambient, first uncheck the **Ambient** parameter. Then, select the empty **Temperature** parameter and enter the temperature in degrees Celsius as a whole number.

If you want to change a temperature to ambient, select and then check the **Ambient** parameter.

See Step 5 in [7.4.3 Protocol Rules](#) for allowable temperature ranges.
 - d Set dispense type for staining protocols (150 µL, Open, Intermediate, or Selected Vol for BOND RX^m).
 - e Click on any other step to confirm the changed parameters.

7.4.2 Adding and Removing Protocol Steps

You can add and remove steps in user IHC and ISH protocols, and also in preparation protocols. Add or remove steps with buttons below the protocol step table. The buttons are context sensitive and their availability and functions vary depending upon the step selected.

Refer to the following sections for detailed instructions:

- [7.4.2.1 Inserted Reagent Steps](#)
- [7.4.2.2 Wash Steps](#)
- [7.4.2.3 Preparation Steps](#)

7.4.2.1 Inserted Reagent Steps

- 1 From the step list, select the step that will follow the newly inserted reagent step.



If you select the last step of the protocol, you can choose to add the new reagent step below this step.

- 2 Click **Insert reagent**.

The new reagent step is added to the step list. It initially has a red bar to indicate that you must select a reagent for the step.

- 3 Select a reagent for the new step and edit other parameters as required.

The new step has a green bar to indicate a change from the saved protocol.

To delete a reagent step, select it and click **Delete step**.

7.4.2.2 Wash Steps

It is recommend that a deionized water wash step is in place before and after a chromogen or Hematoxylin step.



If you cannot see wash steps, select the **Show wash steps** option button below the step list area.

To insert an additional wash step:

- 1 From the step list, select the step that will follow the newly inserted wash step.



If you select the last step of the protocol, you can choose to add the new wash step below this step.

2 Click **Insert wash**.

Edit protocol properties

Name: IHC Protocol F - 2

Abbreviated name: IHC F2

Description: BOND Polymer DAB system for IHC

Staining method: Single Preliminary Final Preferred

BOND-MAX BOND-III BOND-PRIME [Import protocol](#) Protocol type: IHC staining

Preferred detection system: Bond Polymer Refine Detection

Step N°	Wash	Reagent	Supplier	Ambient	Temperature	Inc. (min)
1		*Peroxide Block	Leica Microsystems	✓		5:00
2	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00
3	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00
4	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00
5		*MARKER	Leica Microsystems	✓		15:00
6		*MARKER	Leica Microsystems	✓		15:00
7	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00
8	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00

Show wash steps [Insert wash](#) | [Duplicate](#) | [Delete wash](#)

[Save](#) [Cancel](#)

The new wash step is added to the step list, and has a green bar to indicate a change from the saved protocol.

Edit protocol properties

Name: IHC Protocol F - 2

Abbreviated name: IHC F2

Description: BOND Polymer DAB system for IHC

Staining method: Single Preliminary Final Preferred

BOND-MAX BOND-III BOND-PRIME [Import protocol](#) Protocol type: IHC staining

Preferred detection system: Bond Polymer Refine Detection

Step N°	Wash	Reagent	Supplier	Ambient	Temperature	Inc. (min)
1		*Peroxide Block	Leica Microsystems	✓		5:00
2	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00
3	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00
4	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00
5	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00
6		*MARKER	Leica Microsystems	✓		15:00
7		*MARKER	Leica Microsystems	✓		15:00
8	✓	*Bond Wash Solution	Leica Microsystems	✓		0:00

Show wash steps [Insert wash](#) | [Duplicate](#) | [Delete wash](#)

[Save](#) [Cancel](#)

3 Modify the wash step parameters in the step list as required.

4 Click **Save**.

5 On the confirmation window, click **Yes**.

To delete a wash step, select it and click **Delete step**.

New protocol properties

Name: IHC Protocol F
 Abbreviated name: IHC F
 Description: BOND Polymer DAB system for IHC
 Staining method: Single Preliminary Final Preferred

BOND-MAX BOND-III BOND-PRIME [Import protocol](#) Protocol type: IHC staining

Preferred detection system: Bond Polymer Refine Detection

Step N°	Wash	Reagent	Supplier	Ambient	Temperature	Inc. (min)
1		*Peroxide Block	Leica Microsystems	<input checked="" type="checkbox"/>		5.00
2	<input checked="" type="checkbox"/>	*Bond Wash Solution	Leica Microsystems	<input checked="" type="checkbox"/>		0.00
3	<input checked="" type="checkbox"/>	*Bond Wash Solution	Leica Microsystems	<input checked="" type="checkbox"/>		0.00
4	<input checked="" type="checkbox"/>	*Bond Wash Solution	Leica Microsystems	<input checked="" type="checkbox"/>		0.00
5	<input checked="" type="checkbox"/>	*Bond Wash Solution	Leica Microsystems	<input checked="" type="checkbox"/>		0.00
6		*MARKER	Leica Microsystems	<input checked="" type="checkbox"/>		15.00
7	<input checked="" type="checkbox"/>	*Bond Wash Solution	Leica Microsystems	<input checked="" type="checkbox"/>		0.00
8	<input checked="" type="checkbox"/>	*Bond Wash Solution	Leica Microsystems	<input checked="" type="checkbox"/>		0.00

Show wash steps [Insert wash](#) | [Duplicate](#) | [Delete wash](#)

[Save](#) [Cancel](#)

7.4.2.3 Preparation Steps



You can add and remove (but not edit) dewax and alcohol steps in preparation protocols, subject to certain rules.

To insert an additional dewax step:

- 1 From the step list, select an existing dewax step.
- 2 Click **Insert reagent**.

A new dewax step is added above the selected dewax step.

To delete a dewax step, select it and click **Delete step**.



The dewax step you want to delete must have a non-ambient temperature setting. Also, the protocol must currently have more than three dewax steps.

To insert an additional alcohol step:

- 1 From the step list, select an existing alcohol step.
- 2 Click **Insert wash**.

A new alcohol step is added above the selected alcohol step.

To delete an alcohol step, select it and click **Delete step**.



The protocol must currently have more than three alcohol steps.

7.4.3 Protocol Rules

Any protocol you create or edit must conform to some basic rules before it can be saved. Please note that these rules do not guarantee that the protocol will produce acceptable results when used.

- 1 The protocol name must:
 - a be unique;
 - b begin with a character other than a space or asterisk.
- 2 The protocol abbreviated name must:
 - a be unique;
 - b begin with a character other than a space or asterisk;
 - c have maximum 8 characters.
- 3 All staining protocols must include at least one reagent from a Leica Biosystems BOND detection system or research reagent system.
- 4 For staining protocols, the last step must be a wash step.
- 5 Heated step temperatures must be within the ranges in the following table:

Protocol Step	Temperature Range (°C)
Bake and Dewax, bake step	35–72
Heat pretreatment (BOND RX and BOND RX ^m)	35–100
Enzyme pretreatment	35–100
Denaturation	35–100
Hybridization	35–100
Staining protocols	35–100

- 6 Step incubation times, which must be set in minutes and seconds (mm:ss), should be within the ranges in the following table. The ranges are not enforced:

Protocol Step	Incubation Range (minutes)
Bake and Dewax, bake step	0–60
Heat pretreatment (ambient steps)	0–15
Heat pretreatment (BOND RX and BOND RX ^m heated steps)	5–60
Enzyme pretreatment (step 1)	0
Enzyme pretreatment (enzyme steps)	0–15
Denaturation	5–20
Hybridization	20–950
Staining protocols, reagent steps	0–60
Staining protocols, wash steps	0–55

In general, for reagent application steps, avoid incubation times greater than 30 minutes.

- 7 Each step must be fully defined with a reagent, incubation time, dispense type and (where applicable) temperature.
- 8 Single-stain and sequential multiplex-stain protocols can have only one mixed reagent (e.g. mixed DAB) per protocol, used in a maximum of two steps in the protocol. A sequential multiplex-stain procedure can have up to six mixed reagents and up to 12 application steps – two in each protocol.
- Parallel multiplex-stain protocols can include six mixed reagents, and each mixed reagent can be applied up to two times in the protocol.
- 9 The components required to make up the staining protocol's mixed reagent(s) may be assigned to research detection systems or as separate ancillaries.

Either:

- all components of the mixed reagent must be in the research reagent system, **or**
- all components of the mixed reagent must be ancillaries.

7.4.4 Multiple Processing Module Types and Protocol Versions

For BOND RX systems with BOND RX and BOND RX^m processing modules, each protocol can have separate versions for the two instrument types.

Leica Biosystems protocols have been tested and optimized for use on the BOND RX systems. These protocols have been rigorously tested and validated by Leica Biosystems.

The different versions of the “same” protocol accommodate hardware differences such as the:

- faster cooling on BOND RX Processing Modules (protocol steps where slides are cooled are typically shorter in BOND RX protocol versions than the corresponding steps in the BOND RX^m versions)

Some protocol version differences cannot be seen in the step list displayed in the software, e.g. BOND RX protocol versions include hidden instructions for the bulk fluid robots, not present on BOND RX^m Processing Modules.

All BOND RX systems have both BOND RX and BOND RX^m versions of all predefined protocols.

However, if a new processing module type is added to a system, you must create a new version of an existing user-defined protocols for the new processing module type. Do this by importing Leica Biosystems predefined protocols and then copy or modify steps as required (see [7.4.4.1 Importing a Protocol Version](#)).

7.4.4.1 Importing a Protocol Version

To create a protocol version for a new processing module type, follow the instructions below. This method can also be used to overwrite existing protocol versions, however this should not usually be required after initial configuration.

1 On the **Protocol setup** screen select the user protocol you want to create a new version for.

2 Click **Open**.

The **Edit protocol properties** dialog opens.

3 Click **Import protocol**.

The **Import protocol** dialog opens.

4 In the **Processing modules** drop-down list, select the new processing module type.

The list of protocols displayed in the dialog is updated to show only protocols with versions for the selected processing module type.

5 Optionally select or deselect **Preferred**, to show only preferred, or all, protocols.

6 Select a protocol to import from the list.

To make later configuration easier, select a protocol as similar as possible to the protocol you are creating a new version for. For example, select a protocol that uses the same detection system and, if possible, has the same number of steps.

7 Click **Import**.

The **Import protocol** dialog closes. The tab in the **Edit protocol properties** dialog for the new processing module type is now populated with the imported protocol version.



Only the tab for the selected processing module type is updated.

8 Edit the new protocol version as required (see [7.4.1 Editing Protocol Steps](#)). You can click between the processing module tabs without losing data.9 Click **Save**.

It is the user's responsibility to verify that the protocols provide equivalent staining for both processing module types.

7.4.5 Deleting Protocols

To delete a user protocol, select it from the list in the **Protocol setup** screen and click **Delete**.

Predefined Leica Biosystems protocols (starting with an asterisk) cannot be deleted. You can however hide them – open the protocols and deselect **Preferred**, then set the **Preferred status** filter in the **Protocol setup** screen to "Preferred".

7.5 Protocol Reports

Protocol reports display step details for selected protocols. To generate a report, select a protocol from the list in the **Protocol setup** screen then click **Report**. If you have multiple processing module types in the system, select the processing module type for the protocol version you want. You can also choose a date from which to generate the report. When finished, click **Generate report**.

The report is displayed in a new window. The top right of the report shows the information in the following table:

Field	Description
Full name	The full name of the protocol.
ID	The unique identification number of the protocol.
Type	The protocol type (see 7.1 Protocol Types).
Created by	The username of the person who created the displayed version.
Creation time	For predefined protocols, the date and time the protocol was imported in a BOND Data Definitions (BDD) update. For user-defined protocols, the date and time of creation.
Facility	The name of the facility as entered in the administration client Laboratory Settings screen (see 10.5.1 Laboratory Settings).
Staining status	The roles the protocol is suited for with respect to multiplex or single staining (see 7.2.1.1 Staining Method).

The body of the report displays the following for each step:

- Reagent and supplier
- Step type (reagent or wash)
- Incubation time
- Temperature
- Dispense type (describes Covertile position and dispense volume – may be requested by your service representative)

See [3.7 Reports](#) for further details about the report window and printing options.

7.6 Predefined Protocols

The following sections describe some of the predefined protocols that are supplied as part of the BOND RX software. All protocols you create must be based, ultimately, on one of the predefined protocols.



The protocols listed may differ from those in your software. The list is not exhaustive, as more protocols are added with every new BXD release.

- [7.6.1 Staining Protocols](#)
- [7.6.2 Template Protocols](#)
- [7.6.3 Prestaining Protocols](#)

7.6.1 Staining Protocols

Most predefined staining protocols are designed to use a particular BOND detection system. Some protocols present in your software may not be included.

For detailed information on each detection system please refer to the literature accompanying each product or visit the Leica Biosystems web site: www.leicabiosystems.com.

A panel of IHC and ISH Template Protocols complete the staining protocols' menu. They are not to be used as provided, but rather copied and changed to suit the user's needs. See [7.6.2 Template Protocols](#) below.

7.6.1.1 IHC

Name	Preferred Detection System	Detection System Notes
*IHC Protocol B	Bond Intense R Detection	A biotin/streptavidin system suitable for research applications that require an open choice of secondary antibody. It provides peroxide block, intense DAB staining and hematoxylin counterstain (including bluing).
*IHC Protocol F	Bond Polymer Refine Detection	A high amplification, biotin-free detection system optimized for use on the BOND RX system. Gives sharp definition of target antigens with high intensity staining.
*IHC Protocol J	BOND Polymer Refine Red Detection	For in vitro use, a highly sensitivity Compact Polymer system that provides bright red immunostaining through alkaline phosphatase, as well as hematoxylin counterstain (including bluing).
*IHC Protocol K	ChromoPlex™ 1 Dual Detection	For in vitro use, for the detection of tissue-bound mouse and rabbit IgG primary antibodies. It is intended for staining sections of formalin-fixed, paraffin-embedded tissue on the BOND RX system.
*IHC Protocol K - 50 Test	ChromoPlex™ 1 Dual Detection (50 test)	For in vitro use, for the detection of tissue-bound mouse and rabbit IgG primary antibodies. It is intended for staining sections of formalin-fixed, paraffin-embedded tissue on the BOND RX system.
*IHC Protocol Q	Bond Polymer Refine Detection	A high amplification, biotin-free detection system optimized for use on the BOND RX system. Gives sharp definition of target antigens with high intensity staining protocol specific for peroxide sensitive antigens.

7.6.1.2 ISH

Name	Preferred Detection System	Detection System Notes
*ISH Protocol A	BOND Polymer Refine Detection	A high amplification, biotin-free detection system optimized for use on the BOND RX system. Detects RNA by use of an anti-FITC linker.
*ISH Protocol B	BOND Polymer Refine Detection	A high amplification, biotin-free detection system optimized for use on the BOND RX system. Detects DNA by use of an anti-biotin linker.

You can find the complete up-to-date list of protocols on the **Protocol setup** screen (see [7.2 Protocol Setup Screen](#)). The list can be filtered to show different types of protocols.

7.6.2 Template Protocols



Template protocols are not designed to be run as is; they are created to be copied and edited around the desired building blocks. When modifying the building block of choice, the existing BOND protocol rules must still be adhered to.

You can find the complete up-to-date list of protocols on the **Protocol setup** screen (see [7.2 Protocol Setup Screen](#)). The list can be filtered to show different types of protocols.

7.6.3 Prestaining Protocols

Protocol Type	Protocol Name	Notes
Preparation	*Dewax	Preparation protocols use the BOND Dewax Solution to remove paraffin wax, which is used to embed the tissue and rehydrates the sample.
	*Bake and Dewax	Prior to dewaxing, the tissue is baked to improve its adhesion to the slide. For additional details see 14.2.3 Dewaxing and Baking
Heat Pretreatment	*HIER with ER1 or ER2	Heat induced epitope retrieval exposes the sectioned tissue to a heated buffer solution, which helps to change the conformation of the tissue structure and improve staining. There are a number of predefined heat pretreatment protocols available, which differ in length and temperatures used.
Enzyme Pretreatment	*Enzyme 1 *Enzyme 2 *Enzyme 3 *Enzyme 5	There are eight enzyme pretreatment protocols available. These protocols vary in the enzyme used and the incubation times.
ISH Probe Application	*eZ-L Probe Application *ISH Probe Application 1 *ISH Probe Application 2 *ISH Probe Application 3 *ISH Probe Application A	An ISH probe application protocol uses a labeled complementary DNA or RNA strand (the probe) to localize a specific DNA or RNA sequence in a section of tissue.
ISH Denaturation	*Denaturation (10 min)	There is one (10 minute) predefined ISH denaturation protocol.

Protocol Type	Protocol Name	Notes
ISH Hybridization	*ISH Hybridization (2Hr)	There are two predefined ISH hybridization protocols (2 hour and 12 hour).
	*ISH Hybridization (12Hr)	
ISH Probe Removal	*ISH Probe Removal 1	An ISH probe removal protocol uses a stringency wash to remove any non-identical interactions (only exact sequence matches will remain bound).
	*ISH Probe Removal LH	

You can find the complete up-to-date list of protocols on the **Protocol setup** screen (see [7.2 Protocol Setup Screen](#)). The list can be filtered to show different types of protocols.

8

Reagent Management (on BOND RX Controller)

The BOND RX system keeps a record of all non-bulk reagents used on the system, tracking each reagent container and its contents. It also allows you to set up panels of slides with specified markers, to speed up study creation.

This chapter has the following sections:

- [8.1 Reagent Management Overview](#)
- [8.2 Reagent Setup Screen](#)
- [8.3 Reagent Inventory Screen](#)
- [8.4 Reagents for Research](#)
- [8.5 Reagent Panels Screen](#)

8.1 Reagent Management Overview

Reagent management in the BOND RX system includes set up and maintenance of individual reagent details, inventory management for all reagent packages (excluding bulk reagents), and creation of sets of markers, known as “panels”, for use in slide creation.

To open the reagent management screens where these operations are carried out, click the **Reagent setup** icon on the function bar.



Click on the tabs at the top left of the screen to open the required screen (**Setup**, **Inventory** or **Panels**).

Figure 8-1: Reagent setup screen

Name	Abb. name	Type	Supplier	Pref.
*CD10 (56C6)	*CD10	Primary antibody	Leica Microsystems	✓
*CD15 (Carb-1)	*CD15	Primary antibody	Leica Microsystems	✓
*CD20 (MJ1)	*CD20	Primary antibody	Leica Microsystems	✓
*CD25 (4C9)	*CD25	Primary antibody	Leica Microsystems	✓
*CD30 (1G12)	*CD30	Primary antibody	Leica Microsystems	✓
*CD5 (4C7)	*CD5	Primary antibody	Leica Microsystems	✓
*CD56 (CD564)	*CD56	Primary antibody	Leica Microsystems	✓
*CD7 (LP15) *NEW*	*CD7	Primary antibody	Leica Microsystems	✓
*Cytokeratin 20 (Ks20.8)	*CK20	Primary antibody	Leica Microsystems	✓
*Cytokeratin 20 (PW31)	*CK20	Primary antibody	Leica Microsystems	✓
*Cytokeratin 7 (RN7)	*CK7	Primary antibody	Leica Microsystems	✓
*Estrogen Receptor (6F11)	*ER	Primary antibody	Leica Microsystems	✓
*Glial Fibrillary Acidic Protein (GFAP)	*GFAP	Primary antibody	Leica Microsystems	✓
*Immunoglobulin A (N1CLA)	*IgA	Primary antibody	Leica Microsystems	✓
*Immunoglobulin D (DRN1C)	*IgD	Primary antibody	Leica Microsystems	✓
*Immunoglobulin G (Polyclonal)	*IgG	Primary antibody	Leica Microsystems	✓
*Melan A (A103)	*MelA	Primary antibody	Leica Microsystems	✓
*Negative	*Neg	Primary antibody	Laboratory Specified	✓

Package type: All reagents | Reagent type: Primaries | Supplier: Leica Microsystems | Preferred status: Preferred

The **Reagent setup** screen can display a complete list of all reagents known to the BOND RX system. The list does not include any prepackaged reagent systems, e.g. BOND detection systems, but does show the constituent reagents in the systems. It also has mixed reagents, which are mixed on the processing module from components in detection systems. The screen is used to view reagent properties, create new reagents in the system, and set reagent options.

In contrast, the **Reagent inventory** screen shows inventory of reagent systems as well as individually packaged reagents. For any reagent or system type the list shows the total stock, with information about individual packages also available.

The **Reagent panels** screen allows creation of sets of markers typically used together for particular diagnoses. During slide creation in the BOND RX software, selection of a panel creates a slide for each marker in the panel, greatly speeding up this process.

8.1.1 General Information

- [8.1.1.1 Reagent Categories](#)
- [8.1.1.2 Reagent Workflow](#)
- [8.1.1.3 Reagent Identification](#)
- [8.1.1.4 Reagent Substitution](#)

8.1.1.1 Reagent Categories

Apart from bulk fluids, four different sorts of fluidics “package types” can be used on the BOND RX system:

- BOND detection systems: prepackaged trays of detection reagents for use in conjunction with markers selected by users during slide setup
- Research reagent systems: reagent trays with user-configured sets of reagents (see [8.4.1 Research Reagent Systems](#))
- BOND cleaning systems for BOND RX and BOND RX^m Processing Modules: prepackaged trays of cleaning solutions for use in processing module cleaning (see [12.6.1 Cleaning the Aspirating Probe](#)).
- Reagent containers: individual reagent containers containing markers (primaries or probes) or ancillary reagents – in ready-to-use or open containers (see [2.6.5 Reagent Systems and Containers](#))

BOND detection systems, research reagent systems, and cleaning systems are collectively referred to as “reagent systems”.

“Marker” refers to the primary antibody in IHC, or the probe in ISH.

Reagents are subdivided into the following “reagent types”:

- Primary: marker reagent used in IHC
- Probe: marker reagent used in ISH
- Ancillary: all non-marker reagents, used to process tissue before or after staining with a marker
- Mixed: ancillary reagents created during the running of a protocol from components in a reagent system, or from components in individual containers. There can never be stock of mixed reagents, but they must exist in the system for inclusion in protocol steps. The BOND RX system can mix reagents from research reagent systems, also from BOND detection systems or ancillaries. See [8.4.3 Mixed Reagents with Research Reagent Systems](#) for directions to use mixed reagents with research reagent systems.

Reagent and reagent system lists on the **Reagent Setup** and **Reagent Inventory** screens can be filtered according to these classifications.

8.1.1.2 Reagent Workflow

Before the BOND RX system can use any reagent it must recognize it, in a three-step process:

- 1 The reagent type must be included in the reagents list on the **Reagent Setup** screen – all Leica Biosystems ready-to-use reagents and many Leica Biosystems ancillary reagents (including those in BOND detection and cleaning systems) are predefined, but other reagents must be added to the list by users.
- 2 On receipt of new stock, individual reagent containers and reagent systems are scanned into the BOND RX system, or “registered”, to add them to the inventory.
- 3 When ready to use a reagent or system, it is loaded onto the reagent tray where the BOND RX system identifies it and updates the inventory as reagent is used.

The BOND RX software keeps a record of the contents of each individual container and system, as well as the totals for each reagent type. For Leica Biosystems reagents you can set a reorder limit to warn you when stocks are low. See [8.3.2.1 Changing the Minimum Stock Setting](#).

8.1.1.3 Reagent Identification

Individual reagent containers have two barcodes for identification. The longer barcodes on the fronts of the containers are used to register the containers and identify them after registration (see [8.3.3 Registering Reagents and Reagent Systems](#)). Shorter barcodes on the tops of the containers (under the lids) encode the unique pack identifiers (UPI) used by the BOND RX system to identify the containers when they are loaded on processing modules. Use the UPI to manually identify a loaded reagent container that was not successfully scanned (see [5.1.3.6 Fixing Undetected Reagents](#)).

BOND reagent systems for use on BOND RX and BOND RX^m Processing Modules are identified with two barcodes on the sides of the trays. Use both barcodes to register the systems and identify them after registration.

Individual containers within reagent systems have UPI barcodes on the tops and fronts. The BOND RX software uses these to identify the systems when they are loaded on processing modules. If automatic identification fails when the reagent system is loaded onto the BOND RX^m or BOND RX, these UPI numbers can be entered to manually identify the containers.

You can display information about any reagent or reagent system that has been registered, at any time, by rescanning the long barcode on the side of individual containers, or the two barcodes on the sides of reagent systems.

If the package will not scan, open the **Manual ID entry** dialog by clicking either the **Search** icon  on the function bar or the **Enter ID** button on the **Reagent Inventory** screen.

Type in the numbers associated with the long barcode(s) on the front of the individual containers / reagent systems, or the numbers associated with the 2D barcode, then click **Validate** (for reagent systems click **Validate** after entering each barcode).

8.1.1.4 Reagent Substitution

Sufficient volume of all required reagents must be loaded onto the processing module before processing can start. Occasionally, however, a reagent that was initially present may not be available when needed. This may be because the operator has removed a reagent tray or a reagent container may have actually held less reagent than initially determined. If this occurs, the BOND RX system will attempt to substitute the missing reagent with reagent of the same type from a different container. The BOND RX system uses the following rules when substituting an unavailable reagent:

- The system initially tries to substitute the missing reagent with one of the same type from the same reagent system.
If successful the run will continue without notification.
- The system then tries to substitute the missing reagent with an alternative source having the same type and the same Lot number.
If successful the run will continue without notification.
- The system then tries to substitute the missing reagent with an alternative source having the same reagent type but with any Lot number.
If successful, the run will continue but affected slides will have an event notification.
- If reagent substitution is not possible, the reagent will be replaced by a bulk reagent for all dispenses to affected slides until the end of the run.
The run will continue but affected slides will have an event notification.
- If all slides are affected and need to be replaced by a bulk reagent, the run will be abandoned.

8.2 Reagent Setup Screen

The **Reagent setup** screen displays a list of all reagents known to the BOND RX software, including those in reagent systems, and reagents mixed on the processing module from reagent system components. All BOND ready-to-use primaries are predefined in the list (and cannot be removed) as are BOND ready-to-use ISH probes and a number of common Leica Biosystems ancillary reagents.

Filters below the table allow you to set the type of reagent to display. You cannot filter for package types, but you can for reagent types (primaries, probes, ancillaries, mixed reagents, and parallel multiplex-stain primaries and probes), and on supplier and preferred status.

Buttons above the table allow you to: add new reagents to the list; open the reagent that is selected in the table, to view or edit its details; or delete the reagent that is selected in the table (you can only delete non-Leica Biosystems reagents).



You cannot register reagents that are not listed here, or user-defined reagents that do not have preferred status.

The table contains the following details for each reagent:

Name	The full name of the reagent. An initial "*" character indicates a predefined Leica Biosystems reagent.
Abb. name	The short name of the reagent, used on slide labels and the Status screen.
Type	The type of reagent, for example primary.
Supplier	The name of the supplier of the reagent.
Pref.	Ticked (preferred) markers are included in slide configuration lists elsewhere in the BOND RX software.

Editable Reagent Properties

Besides name and supplier details the editable options for reagents are:

- 1 For markers:
 - a the protocols selected by default when the marker is chosen during slide creation (see [6.5.2 Creating a Slide](#)). Different protocols can be set for single marker applications and the preliminary and final applications in multiplex staining;
 - b preferred status – only preferred markers appear in the **Marker** drop-down list during slide creation (see [6.5.2 Creating a Slide](#)), and the **Available markers** list in the **Reagent panels properties** dialog during panel creation (see [8.5.1 Creating a Panel](#)). Reagent screen lists can also be filtered on this property;
 - c hazardous status – markers flagged as hazardous are washed out to the hazardous waste. This setting cannot be changed for predefined reagents.
- 2 For ancillary reagents
 - a the bulk reagents that are compatible with the reagent – the BOND RX system automatically prevents incompatible ancillary and bulk reagents coming into contact;
 - b preferred status – reagent screen lists can be filtered on this property;
 - c hazardous status – reagents flagged as hazardous are washed out to the hazardous waste. This setting cannot be changed for predefined reagents.

See sections:

- [8.2.1 Adding or Editing a Reagent](#)
- [8.2.2 Deleting a Reagent](#)

8.2.1 Adding or Editing a Reagent

To add reagents to the list, click **Add** in the **Reagent setup** screen. The BOND RX software displays the **Add reagent** dialog. See [Figure 8-2](#) below.

Figure 8-2: Add reagent dialog

To change the details of an existing reagent, select it and click **Open**, or double-click it. The **Edit reagent properties** dialog opens. This is the same as the **Add reagent** dialog with the details for the selected reagent entered.

Use the following directions to add or edit reagents:

- 1 If adding a new reagent, enter a descriptive name in the **Name** field.

New reagents cannot start with "*", which is reserved for Leica Biosystems reagents.



Be careful not to use a name that may cause this reagent to be confused with another when creating protocols or slides.

- 2 For new reagents, enter a short name in the **Abbreviated Name** field. Abbreviated names are limited to eight characters.

This name appears on slide icons in the **Status** screen and is printed on slide labels.

- 3 If the BOND RX system is connected to an LIS, enter the name of the reagent used in the LIS, in the **Public name** field (not applicable to ancillary reagents).

- 4 If creating a new reagent, select the type of reagent from the **Type** drop-down list. The dialog changes, depending on the type you select.
- 5 Enter the name of the supplier of the reagent in the **Supplier** field.
- 6 If the reagent is a marker (primary antibody or RNA or DNA probe), select default protocols to use in different types of staining run that use the marker.
- 7 In the **Staining method** field, select **Single/Sequential multiplex** to set default protocols for markers in single stain runs, on the **Single** tab. For markers in sequential multiplex stain runs, set default protocols for all applications except the last on the **Preliminary** tab, and set the default protocol for the last application on the **Final** tab.
- 8 Select **Parallel multiplex** to set default protocols for the markers in parallel multiplex stain runs.



If the reagent is an RNA or DNA probe, additional protocols (denaturation and hybridization) appear on all the above tabs.

- 9 For predefined BOND markers, click **Restore factory default protocols** if you want to return the protocols to their factory defaults, recommended for the marker (you need to be logged on with a supervisor user role to restore factory defaults).
- 10 If the reagent is a user-created ancillary, check bulk solution compatibility and adjust if necessary.

Most systems will by default show BOND Wash Solution (*BWash) and deionized water (*DI) in the **Compatible bulks** list. This means that either of these solutions will be used in the fluidics system to draw and aspirate the reagent. While the bulk solutions should not come into direct contact with the ancillary reagent, there may be some slight contact in the aspirating probe. To avoid this possibility entirely, select the bulk solution you do not want to come in contact with the reagent and click << to move it to the **Available bulks** list.

There must be at least one bulk solution set as compatible.



CAUTION: Unsatisfactory staining results and potential damage to the processing module can occur if incompatible solutions are allowed to come in contact with each other. Contact Leica Biosystems to determine whether the solutions are compatible.

- 11 For markers, click **Preferred** to display the primary or probe in slide setup dialogs.
For ancillary reagents, Preferred status is only used by the list filters on the **Reagent Setup** and **Inventory** screens.
- 12 If you want the reagent to be flushed to the hazardous waste container, click **Hazardous**.
- 13 Click **Save** to add the reagent details to the BOND RX system.

Click **Cancel** at any time during the process to exit without making any changes.

8.2.2 Deleting a Reagent

To delete a reagent, select it from the list in the **Reagent Setup** screen and click **Delete**. Predefined Leica Biosystems reagents (starting with an asterisk) cannot be deleted.



When you delete the details of a reagent, you also remove the inventory details for packages of this reagent. You cannot recover deleted reagent details or inventory details.

You cannot delete a reagent that is used by a registered research reagent system.

If you no longer need a reagent that you have previously used, you may be better to mark it as not preferred rather than delete it. This removes it from most screens in the software, but retains it in the system.

8.3 Reagent Inventory Screen

The **Reagent Inventory** screen lists all reagents and reagent systems that have ever been registered (and not deleted) on the BOND RX system and their current stock holding. Use the screen to view and manage inventory.

Figure 8-3: Reagent Inventory screen

Name	Supplier	Type	Catalog N°	Vol. (mL)	Min. (mL)
*Kappa Probe	Leica Microsystems	Probe RNA	PB0645	27.50	11.00
*CD15 (Carb-1)	Leica Microsystems	Primary antibody	PA0039	44.85	7.00
GFAP (ER2, Enzyme1)	AAA Antibodies	Primary antibody	Open container	0.00	0.00
*Anti-Fluorescein Antibody	Leica Microsystems	Ancillary	AR0222	30.00	15.00
*CD30 (IG12)	Leica Microsystems	Primary antibody	PA0153	0.00	1.00
*Melan A (A103)	Leica Microsystems	Primary antibody	PA0233	7.00	0.00
*CD7 (LP15) *NEW*	Leica Microsystems	Primary antibody	PA0017	0.00	14.00
*Lambda Probe	Leica Microsystems	Probe RNA	PB0669	16.50	5.50
*Estrogen Receptor (ER11)	Leica Microsystems	Primary antibody	PA0151	14.00	7.00
*CD5 (4C7)	Leica Microsystems	Primary antibody	PA0168	6.55	0.00
*Cytokeratin 20 (PW31)	Leica Microsystems	Primary antibody	PA0918	0.00	7.00
*Estrogen Receptor (ER11)	Leica Microsystems	Primary antibody	PA0009	0.00	10.00
*Immunoglobulin D...	Leica Microsystems	Primary antibody	PA0061	7.00	2.00
*Glial Fibrillary Acidic...	Leica Microsystems	Primary antibody	PA0026	0.00	5.00
*CD25 (4C9)	Leica Microsystems	Primary antibody	PA0905	47.50	14.00
*CD10 (B9C6)	Leica Microsystems	Primary antibody	PA0131	0.00	0.00
*Immunoglobulin G...	Leica Microsystems	Primary antibody	PA0904	7.00	3.00
*CD20 (MJ1)	Leica Microsystems	Primary antibody	PA0906	47.65	14.00

Package type: Reagent containers | Reagent type: All | Inventory status: All | Supplier: All | Preferred status: Preferred

Leica Biosystems reagents with less than the minimum stock volume are highlighted with a red vertical bar on the left side of the screen.

Filters below the table allow you to set the type of reagent or system to display.

For BOND detection, Oracle and cleaning systems – selected in the **Package type** filter – you can filter just on **Inventory status**. This allows you to view all registered systems, just those in stock, or those below reorder levels.

The inventory listing of package type “Research reagent systems” cannot be filtered.

For individual reagent containers you can also filter by **Supplier**, **Preferred status** and **Reagent type** (i.e. view “Primaries”, “Probes”, “Parallel cocktail primaries”, “Parallel cocktail probes”, “Ancillaries”, or “All” reagents).

Some or all of the following details may be displayed, depending on the reagent type:

Name	The full name of the reagent.
Supplier	The name of the supplier of the reagent. Not shown for reagent systems.
Type	The type of reagent, for example primary. Not shown for reagent systems.
Catalog No.	The reagent catalog number to quote when reordering. This is not shown for reagent systems (the column is present but all values are blank).
Vol. (mL)	The total amount of the reagent available. This includes all registered reagent packages, whether currently loaded on a processing module or not (see 8.3.1 Determining Reagent Volume).
Cleans remaining	The number of cleans remaining in cleaning systems.
Tests remaining	The number of tests remaining in research reagent systems.
Min. (mL)	For Leica Biosystems reagents only, the stock volume at which you are prompted to reorder (refer to 8.3.2.1 Changing the Minimum Stock Setting).
Min. (cleans)	For cleaning systems, the number of cleans remaining at which you are prompted to reorder (refer to 8.3.2.1 Changing the Minimum Stock Setting).

The control buttons above the reagent table allow you to manage the reagent inventory.

- Click **Details** to see information about individual reagent packages of the selected reagent type and set options for these.
See [8.3.2 Reagent or Reagent System Details](#) for more information.
- Click **Enter ID** to add inventory of reagent to the system in the **Manual ID entry** dialog when the ID cannot be automatically recognized by the handheld scanner (BOND RX and BOND RX^m only).
Refer to [8.3.3 Registering Reagents and Reagent Systems](#) for more information.
- Click **Details report** to generate a report of the reagents or reagent systems currently listed in the table.
See [8.3.4 Inventory Details Report](#).
- Click **Reagent usage** to generate a report of reagent usage within a specified time period.
See [8.3.5 Reagent Usage Report](#)

See also [8.3.1 Determining Reagent Volume](#) for a general description of how BOND tracks reagent inventory.

8.3.1 Determining Reagent Volume

The BOND RX system uses two methods to establish the volume of reagent in containers in the reagent tray: it calculates the volume based on the initial volume and subsequent usage, and it measures it directly using a liquid level sensing (LLS) system.

The volume calculation relies on the initial reagent volume, subtracting reagent as it is dispensed and adding for refills (open containers). Discrepancies can occur if reagent is lost through evaporation or spillage.

For BOND RX^m and BOND RX, the LLS system is integrated into the aspirating probe. It determines reagent volumes by detecting the height of the reagent when the aspirating probe dips into containers. Under default settings, LLS volume measurement (often referred to as a "dip test") is automatically carried out under a range of conditions such as, for example, when a container has not been measured for more than 30 days. Reagent may have evaporated or the container used on another system. These default dip tests are scheduled when they will not delay processing, so it is possible that a reagent initially thought to be available may later be shown to have insufficient volume for scheduled runs. When this occurs an alert activates and the operator must either refill the container (open containers only) or ensure a suitable alternative reagent is available (refer to [8.1.1.4 Reagent Substitution](#)).

Optionally, you can set the BOND RX system to dip test containers before every processing run. This is set independently for open containers, ready-to-use containers and reagent systems. The setting ensures that runs that are started have enough reagent to finish, however it delays processing while dip tests are carried out. Set these options in the administration client **Settings > Laboratory settings** pane (see [10.5.1 Laboratory Settings](#)).



Do not overfill reagent containers. An overfull reagent container will be reported as empty when it is dip tested

8.3.1.1 Reporting Volume for Detection Systems

To make the volumes reported for BOND detection systems comparable to those reported for individual containers (allowing estimation of the number of slides that a detection system can be used for) system volumes are reported in milliliters, in terms of a single container. However, since detection systems consist of containers with different volumes, a rule for reporting the volume must be applied, described in this section.

Note that this rule does not apply to cleaning systems, which report the number of cleans remaining.

For detection systems, volume is reported relative to the largest single container in the system. For example, if the largest container holds 30 mL, the system volume is reported relative to 30 mL. The BOND RX software assumes that all containers in new systems are full, so a system with its largest container of 30 mL is reported as having 30 mL volume when first registered.

As reagent is used, the value reported is the volume of the container with the lowest relative volume. If this container's volume is not the same as that of the largest container in the system, then the value is normalized to the volume of the largest container. For example, in a system with several containers of 30 mL and two containers of 2.4 mL, it may be that one of the 2.4 mL containers has, relative to initial volumes, the least volume of reagent. If it has 1.2 mL left (half of its initial volume), then the volume of the system as a whole is reported as half of 30 mL (15 mL).

8.3.1.2 Reporting Tests Remaining for Research Reagent Systems

Each research reagent system can run a fixed number of tests (e.g. 200 tests, if the default dispense volume is 150 µL).

Each time a slide is stained using this research reagent system, the number of tests remaining is reduced.

When the number of tests remaining reaches zero, the system is marked as **Empty**.

8.3.2 Reagent or Reagent System Details

To display details of individual packages of a reagent or reagent system (or research reagent system), double-click on the reagent type in the Reagent inventory table, or select it and click **Details**.

Figure 8-4: Reagent inventory details dialog

UPI	Lot N°	Expiration date	Registered	First used	Marked empty	Initial vol. (mL)	Vol. (mL)
00676418		20-May-21	16-Apr-13			6.60	6.60
00676421	04224	25-Feb-21	16-Apr-13			6.60	6.60
00676420	04224	25-Feb-21	16-Apr-13			6.60	6.60
00676457		20-May-21	16-Apr-13			6.60	6.60
00684913	05933	05-Feb-23	23-Aug-13			6.60	6.60

An inventory details dialog displays each individual package of the selected reagent or system. Dialog fields and options differ according to the reagent package type and supplier. By default only packages with available, non-expired, reagent are shown. You can also show empty packages (that have not reached their expiration date), or all packages that expired in the last month – select **Available**, **Empty** or **Expired** as appropriate in the dialog.

The reagent **Package name** is shown for all reagent package types. In addition, BOND reagents show the **Catalog N°** for reordering purposes, and BOND reagents (but not systems) also have **Package name**, which includes the package size.

BOND reagents and systems also have a **Minimum stock** field showing the stock level at which you are prompted to reorder the reagent (see [8.3.2.1 Changing the Minimum Stock Setting](#)).

Using the handheld scanner, you can scan the side barcode(s) or 2D barcode of a registered reagent container or reagent system to launch its inventory details dialog. The scanned inventory item will be highlighted in the details table, and the **Show** filters (Available, Empty or Expired) will be automatically set as appropriate.

The table in the dialog shows the following information for each reagent package:

UPI	The Unique Pack Identifier (see 8.1.1.3 Reagent Identification).
Lot No.	The package lot number.
Expiration date	The package expiration date. Packages should not be used after this date. Note: For research reagent systems, the Expiration date refers to the expiry date of the reagents within the reagent system's open containers. If the reagents in a research reagent system have expired, then the reagent system can be refilled and a new expiry date can be set.
Registered	The date the package was first registered on the BOND RX system.
First used	The date the package was first used on the BOND RX system.
Marked empty	The date the package was marked as empty. This may be set automatically by the software, or manually (see 8.3.2.3 Marking a Package as Empty or Not Empty).
Initial vol. (mL)	The volume of reagent that was in the new, full package. Not shown for reagent systems.
Cleans remaining	For cleaning systems, the number of cleans that can be performed with the remaining reagent.
Tests remaining	The number of tests remaining in research reagent systems.

Buttons on the inventory details dialogs allow configuration of a range of inventory details (appropriate for the package type) and creation of a details report for the specific reagent or system. The sections below describe the configuration and report options.

8.3.2.1 Changing the Minimum Stock Setting

Predefined Leica Biosystems reagents and reagent systems, but not research reagent systems, can have a "minimum stock level" set. When total reagent stock falls below the set level the reagent is highlighted red in the **Reagent Inventory** screen to prompt the user to reorder the reagent or system.

To change the minimum stock setting, click **Set minimum stock level**. In the pop-up dialog, enter the required minimum stock level in the **Minimum stock** field. Use milliliters, runs or cleans, depending on the package type. Click **OK**.

8.3.2.2 Reagent Report

Click **Details report** to generate a report for just the selected reagent or reagent system or research reagent system. See [8.3.4 Inventory Details Report](#) for more details.

8.3.2.3 Marking a Package as Empty or Not Empty

You can mark a reagent package as empty, for example when it is discarded before being completely used. To do this, select the package in the table, then click **Mark as empty**. The software puts the current date in the **Marked empty** field.

To reinstate a reagent package marked empty, select it in the table and click **Mark not empty**. This can be done only when the package is not loaded on a processing module. The package shows the volume of reagent that it had prior to being marked empty.



You cannot mark a research reagent system as **not empty** when it has zero tests remaining. Also see [8.3.1.2 Reporting Tests Remaining for Research Reagent Systems](#).

Select the **Empty** radio button above the table to show items that are marked as empty.

8.3.2.4 Refilling an Open Reagent Container

You can reuse BOND open reagent containers to dispense up to 40 mL of a particular reagent. There is no limit on the number of times containers can be refilled if you fill with quantities less than the container volumes.

Use the following instructions to refill an open container.

- 1 Fill the container with the desired volume of reagent.
- 2 Scan the container (as described in [8.3.3 Registering Reagents and Reagent Systems](#)) then click **Refill**.
The refill button will not be available if putting more reagent into the container will exceed the 40 mL limit.
- 3 Set an expiration date for the new reagent.



Note that when an open container is filled (either for the first time or a refill), the software assumes that the container is filled to the maximum available for that container, that is the volume (mL) specified by the user when the reagent is first registered, or the current volume plus the remainder of the allowable refill volume. The reported volume is corrected, if necessary, when a dip test is carried out. This may not occur until the container is used.




Each open container is locked to a particular reagent when it is first registered. Each open container must use the same reagent each time it is refilled.

8.3.3 Registering Reagents and Reagent Systems

Registering a reagent package adds it to the inventory. The reagent must be listed in the **Reagent Setup** screen before you can register a package of it.



You must register reagent packages before using them on BOND RX Processing Modules.

If you load an unregistered reagent container in the processing module, the software will not recognize it, and will display an information icon  in that reagent position on the **System status** screen.



The BOND RX software will track reagent usage and will alert you when the reagent must be replaced.

Do not attempt to refill a ready-to-use BOND reagent container as the BOND RX software will recognize that this is a used container and refuse to use it.

The methods for registering different types of reagent packages are described in the following sections:

- [8.3.3.1 Registering Reagent Systems](#)
- [8.3.3.2 Creating and registering BOND Research Systems](#)
- [8.3.3.3 Registering BOND Ready-To-Use Reagents](#)
- [8.3.3.4 Registering Non-Ready-To-Use Reagents](#)
- [8.3.3.5 Manual ID Entry](#)

See [8.4.1 Research Reagent Systems](#) for instructions to register research reagent systems.

8.3.3.1 Registering Reagent Systems

To register a BOND detection or cleaning system, scan the two barcodes on the side of the reagent tray.



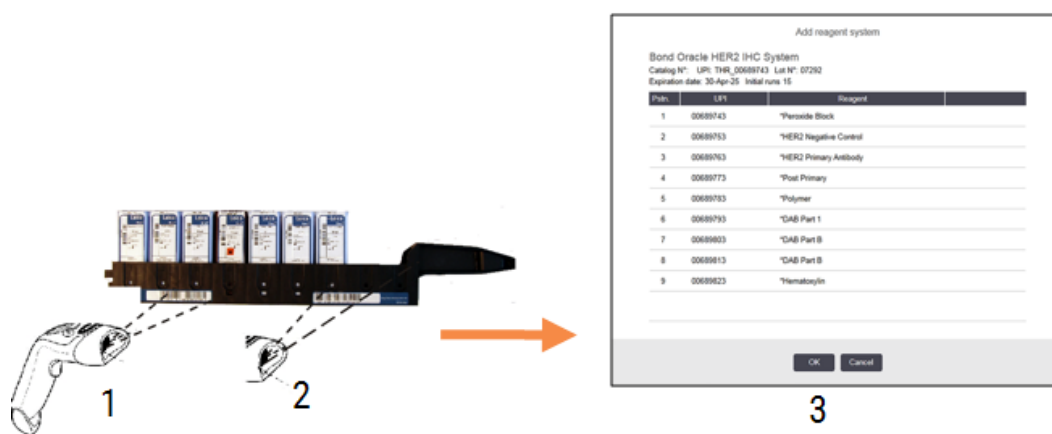
Some reagent systems have only one barcode on the reagent tray, for example systems with only one or two containers.

The software will display the **Add reagent system** dialog.



WARNING: Laser hazard. Potential for severe eye damage. Avoid direct eye contact with laser beams.

Figure 8-5: Registering a BOND detection system



Check that the details in the dialog correspond to the details of the package, then click **OK**.



Do not attempt to register individual reagent containers that are part of a reagent system.

8.3.3.2 Creating and registering BOND Research Systems

Please refer to [8.4.1.2 Creating Research Reagent Systems](#).

8.3.3.3 Registering BOND Ready-To-Use Reagents

To register a BOND ready-to-use reagent package, scan the barcode on the front of the container. The software will display the **Add reagent package** dialog.

Figure 8-6: Registering BOND reagent packages



Check that the details in the dialog correspond to the details of the package, then click **Add**.

8.3.3.4 Registering Non-Ready-To-Use Reagents

Reagents not supplied in BOND ready-to-use packages can be used on the BOND RX system, in BOND open or titration containers. After a non-ready-to-use reagent has been prepared and filled into a 7 mL or 30 mL open container, or 6 mL titration container, it is registered in much the same way as BOND reagents:

- 1 Ensure that the reagent has been created in the system and is preferred. The user-defined reagent must be preferred to register inventory. (see [8.2.1 Adding or Editing a Reagent](#)).
- Note that enzymes created with the BOND Enzyme Pretreatment Kit are predefined in the system, and do not require manual creation.
- 2 Scan the barcode on the front of the open or titration container to open the **Add open container** dialog.
 - 3 Select the name of the reagent from the **Reagent name** drop-down list. (The supplier name is shown in brackets beside the reagent name.)
- The list has all preferred non-BOND ancillaries and markers created in the system, as well as the four predefined enzymes that can be prepared from the BOND Enzyme Pretreatment Kit. If you have not created the reagent in the system cancel out of the **Add open container** dialog and do this first (see step 1 above).
- 4 Type in the lot number for the reagent, from the reagent supplier's documentation.

- 5 Click in the **Expiration date** field to set the expiration date with the calendar controls (or, you can type in a date).



You can enter partial dates such as D/M, DD/MM or DD/MMM; the current year is assumed. If you enter, for example, MM/YYYY or MMM/YYYY, the first day of that month is assumed.

If you enter an invalid date, a red border appears around the **Expiration date** field and a validation error message displays.

When you click away from the **Expiration date** field, a valid date entry is automatically reformatted to match the system date format. If at least one valid date was entered prior to entering an invalid date, the field will be reset to the last entered valid date when you click away from it.

- 6 Click **OK** to register the reagent.

8.3.3.5 Manual ID Entry

If the BOND RX system fails to read a reagent barcode, do the following from the **Reagent Inventory** screen:

- 1 Click **Enter ID**.

The BOND RX software displays the **Manual ID entry** dialog.

- 2 Type numbers associated with the long barcodes on the front of the container, or the numbers associated with the 2D barcode, into the top row in the dialog.

- 3 Click **Validate**.

If there is more than one barcode, as for detection systems, click **Validate** after entering the corresponding numbers for each barcode.

- 4 After verifying that the barcodes are legitimate, the software displays the appropriate **Add reagent package** dialog.

- 5 Verify package details or add details as required in the **Add reagent package** dialog, then click **OK** to register the package.

8.3.4 Inventory Details Report

You can generate a report of the inventory details of the reagents or reagent systems or research reagent systems displayed in the table on the **Reagent Inventory** screen. The generated report shows information for each of the visible reagents or systems, including the total stock remaining. If the total stock is less than the minimum stock level (see [8.3.2.1 Changing the Minimum Stock Setting](#)) then it is flagged with "Low" in the report.

Set the filters at the bottom of the screen to show the reagents or reagent systems you are interested in, then click **Details report**. The report is generated and displayed in a new window.

The top right of the reagent inventory report shows the information in the following table.

Field	Description
Facility	The name of the facility as entered in the Facility field on the administration client Settings > Laboratory settings screen – see 10.5.1 Laboratory Settings
Subject	The filter settings used to select the reagents or reagent systems or research reagent systems in the report.

For each reagent listed in the table the body of the report displays:

- name
- total stock on-hand (flagged if less than the minimum stock level)
- catalog number (for BOND ready-to-use containers) or "open" (for open containers)
- type (primary, probe, ancillary, or reagent system type)
- supplier

For each individual reagent package or research reagent system the report displays:

- UPI
- lot number
- date of expiration
- date when registered
- date when first used
- date when last used
- tests remaining (research reagent system)
- Vol. (mL) (BOND Detection System)

See [3.7 Reports](#) for further details about the report window and printing options.

8.3.5 Reagent Usage Report

The reagent usage report shows the quantity of reagent used and how many tests were processed with this reagent within a defined period. The information is itemized for individual containers as well as showing reagent totals.

The report covers all reagents used in the defined period, irrespective of the reagents currently displayed in the **Reagent Inventory** screen. Reagent system usage is not included.

Click **Reagent usage** to open a date selection dialog where you must set the period that you want the report to cover. Set **From** and **To** dates and times (see [Using the Date & Time Selectors \(on page 207\)](#)), and then click **Generate**. The report is generated and displayed in a new window.

The top right of the reagent usage report shows the information in the following table.

Field	Description
Facility	The name of the facility as entered in the Facility field on the administration client Settings > Laboratory settings screen – see 10.5.1 Laboratory Settings
Time period	The “from” and “to” dates for the period that the report covers

For each reagent used in the period the report displays:

- Name (the reagent’s abbreviated name);
- UPI of each container used;
- Lot number of each container used;
- Expiration date of each container used;
- Number of slides processed, both per container and the total for the reagent;
- Volume of reagent used in the period, both per container and the total for the reagent.

See [3.7 Reports](#) for further details about the report window and printing options.

8.4 Reagents for Research

The BOND RX system is open for any primaries, probes, or ancillary reagents you want to use. In addition, it allows you to create your own detection systems with ancillary reagents of your choice. At least one dispense in each protocol must come from the reagent system selected in the protocol. Any number of dispenses can be drawn from other (open) containers, set up with the primaries, probes, or ancillary reagents you need.

- [8.4.1 Research Reagent Systems](#)
- [8.4.2 Refilling Research Reagent Systems](#)
- [8.4.3 Mixed Reagents with Research Reagent Systems](#)

8.4.1 Research Reagent Systems

Research reagent systems supplied by Leica Biosystems comprise reagent trays and BOND open containers.

You put the individual reagents in BOND open containers (7 mL, 30 mL, or titration containers) and load them into the trays. However, the containers do not have to be used with the trays they come with. You can use the containers independently or with other systems, and you can use any BOND open containers or titration containers with the research trays.

The containers within a research system must always occupy the first positions in the reagent trays, i.e. the positions furthest from the tray handles.

The reagent trays for these systems are scanned into the BOND RX system like any other reagent system (except they have only one barcode), but then require you to configure one to nine ancillary reagents that will make up the systems.

Once a research reagent system is registered in the software you cannot change the configuration, e.g. to add or remove containers, change container positions in trays, or to change the reagents within containers.

Research reagent systems use a different method for reporting their remaining reagent volume, compared with BOND detection systems, i.e. in terms of tests remaining (see [8.3.1.2 Reporting Tests Remaining for Research Reagent Systems](#)).

It is not necessary to fill research reagent system containers with the maximum volume that the containers hold. Once trays are loaded onto processing modules they are dip tested to ascertain the actual volumes, and the reagent inventory is updated accordingly. Also see [8.4.2 Refilling Research Reagent Systems](#).

For research reagent systems that duplicate the configuration of previously registered research reagent systems the software allows quick setup by copying the details of the earlier systems. Systems that are created this way have the same names as the systems they are copied from. The individual systems are identified by the UPIs of their first containers.

8.4.1.1 Research Reagent System Types

There are two types of Research Reagent Systems: BOND Research Reagent System and BOND Research Reagent System 2.

The BOND Research Reagent System is supplied with six standard 30mL Leica Biosystems open containers; System 2 has eight 30mL open containers.

The main difference between the two (apart from the number of open containers) is the probe removal protocol associated with each of them, as shown in the table below:

BOND Research Reagent System		BOND Research Reagent System 2	
Reagent	Dispense	Reagent	Dispense
Probe	150 µL dispense position	Probe	150 µL dispense position
Probe	Intermediate position 70 µL (DNA)/90 µL (RNA)	Probe	Intermediate position 70 µL (DNA)/90 µL (RNA)
No Reagent	Intermediate position	No Reagent	Intermediate position
BOND Wash	150 µL dispense position	BOND Wash	150 µL dispense position
BOND Wash	Open fill 200 µL	BOND Wash	Open fill 200 µL
BOND Wash	150 µL dispense position	BOND Wash	150 µL dispense position
BOND Wash	150 µL dispense position	BOND Wash	Initial Fill
BOND Wash	150 µL dispense position	BOND Wash	150 µL dispense position
BOND Wash	Open fill 200 µL		
BOND Wash	150 µL dispense position		
BOND Wash	150 µL dispense position		
BOND Wash	Initial Fill		
BOND Wash	150 µL dispense position		

8.4.1.2 Creating Research Reagent Systems

Follow the instructions below to create a new research reagent system. The instructions cover both reagent systems with a new configuration, and reagent systems that use the same configuration as a previously registered research system:

- 1 Ensure that all the reagents that will be added to the system have been created as ancillary reagents in the **Reagent setup** screen (see [8.2.1 Adding or Editing a Reagent](#)).

- 2 Load the open containers you will use into a BOND research reagent system tray.

Start at position 1 (furthest from the tray handle) and work towards the handle, with no gaps. If you have a particular order you want the containers in be sure to use it – you cannot change the order once the system is configured.

It is not necessary to put reagent into the containers yet.

- 3 Scan the barcode on the side of the research reagent system tray.

The **Add reagent system** dialog opens with specialized fields for research reagent systems.

Figure 8-7: Add reagent system dialog for research reagent system

Add research reagent system

Name:

UPI:

Lot N°:

Expiration date:

Reagents

Pstn.	UPI	Reagent	Vol. (mL)
1		<input type="text"/>	
2			
3			
4			
5			
6			
7			
8			
9			

[Add reagent](#) | [Remove reagent](#)

- 4 For a research reagent system with a new reagent configuration:
 - a Enter a unique name (which must begin with a letter or number) for the configuration.
 - b Make sure that the first row in the reagents list in the lower part of the dialog is selected (as in [Figure 8-7](#)), and scan the barcode on the front of the open container in position 1 (furthest from the handle) in the reagent tray.

The container's UPI (unique pack identifier) and volume are written into the table, and the second row is selected in readiness for entry of the second open container's details.

The volume is automatically set at the open container's nominal capacity. This is updated, if necessary, when the tray is loaded onto the instrument and a forced dip test carried out.

- c Scan the barcode of the open container in position 2 in the reagent tray.
Continue for all open containers used in the system. Do not scan open containers that will not be used.
If an open container does not scan, click **Add reagent**, type in the barcode number (the number beside the barcode on the front of the open container) and click **Validate**.
If an open container is scanned into the wrong position, select the row in the reagent list and click **Remove reagent** to clear the row.
 - d When all the open containers for the system have been scanned, open the drop-down list in the **Reagent** column for each container and select the reagent in that open container. Only (preferred) ancillary reagents within the BOND RX system are displayed in the list.
- 5 For a research reagent system with the same reagents in the same tray positions as a previously registered system:
- a Open the drop-down list in the field at the top of the dialog and select the desired configuration.



Only research reagent system configurations for the relevant research system type (i.e. BOND Research Reagent System or BOND Research Reagent System 2) are available for selection.

- The reagent list populates with the reagent names of the existing configuration.
- b Ensure the first row in the reagent list is selected and scan the open container in position 1.
The open container's UPI and volume is written into the first row of the table, and the second row is selected in readiness for entry of the second open container's UPI and volume.
 - c Scan the barcode of the open container in position 2 in the reagent tray.
Continue for all open containers used in the system. Do not scan open containers that will not be used.
- 6 Optionally enter a lot number for the system.
Any alphanumeric sequence can be used. It could be the lot number of one of the reagents in the system, or use your own numbering to help identify the detection systems (note, however, that research systems with identical names can always be identified by the UPI of the first container).
- 7 Enter an expiration date for the system's reagents, in format dd/mm/yyyy.
Set the expiration date for the reagent in the system with the shortest lifetime. If you later refill any of the system's open containers you can set a new expiration date at that time.
- 8 Check that all details are correct then click OK to close the dialog and register the research reagent system.



It is important that all details are correct before the system is registered – no changes can be made to the configuration once the dialog is closed.

8.4.2 Refilling Research Reagent Systems



There is no restriction on being able to refill a research reagent system based on the current volume of the open containers. However, you cannot refill a research reagent system when it is loaded on the instrument or when no tests remain.



Each open container is locked to a particular reagent when it is first registered – you must use the same reagent each time it is refilled.

Use the following instructions to refill a research reagent system:

- 1 Fill the open containers in the research system with the desired volume of reagent.
- 2 Scan the system's barcode (on the reagent tray).

The **Reagent system inventory details** dialog opens for the research reagent system type. The particular system is highlighted in the table.

Figure 8-8: Reagent system inventory details

UPI	Lot N*	Expiration date	Registered	First used	Marked empty	Tests remaining
00690823		31-Dec-45	06-Apr-17			200

Patn.	UPI	Reagent	Vol. (mL)
1	00690848	*1:1 Part A	7.00
2	00690849	*1:1 Part B	7.00
3	00690850	*ViewRNA Amp 1	7.00

- 3 Click **Refill**.

The volume for each container in the system is updated to the maximum available for the container, i.e. the maximum that the container can physically hold. This will be corrected, if necessary, when a forced dip test is carried out. This may not occur until the container is used.

- 4 Set an expiration date for the refilled system. Set the expiration date for the reagent in the system with the shortest lifetime.

8.4.3 Mixed Reagents with Research Reagent Systems

8.4.3.1 Pre-mixed Reagents - Existing

To use a pre-mixed reagent with a research reagent system follow the instructions below. Any number of pre-mixed reagents can be used in a run with a research reagent system.

- 1 Configure a research reagent system that does not include the pre-mixed reagent (or its components, unless you want to dispense these directly to the slides). See [8.4.1 Research Reagent Systems](#) above.
- 2 Create a new reagent in the software for the pre-mixed reagent (see [8.2.1 Adding or Editing a Reagent](#)). The reagent must be created as an ancillary reagent.
- 3 Create a research protocol using the pre-mixed reagent (see [7.4.1 Editing Protocol Steps](#)).
 - a Select the research reagent system you configured in step 1 as the **Preferred detection system**.
 - b Select the pre-mixed reagent you configured in step 2 in the appropriate protocol steps.
- 4 Register an empty open container and select the new pre-mixed reagent for it (see [8.3.3.4 Registering Non-Ready-To-Use Reagents](#) in [8.3.3 Registering Reagents and Reagent Systems](#)).
 - a Set the reagent expiration date, taking into account the time you intend to mix the reagent.
Note that the expiration date applies to the reagent and not the container – you can enter a new expiration date if the container is refilled.
 - b Do not fill the container at this point.
 - c Be sure to label the container so that you can easily identify it later.
- 5 Configure research slides selecting the protocol you created in step 3 (See [6.5.2 Creating a Slide](#)).
- 6 When ready to process the slides prepare the mixed reagent and put it into the open container that you registered in step 4.
- 7 Load the research reagent system and pre-mixed reagent onto the processing module.
- 8 Run the protocol as normal.

8.4.3.2 On-board Mixing of Reagents

A number of protocol templates are available that enable the mixing of reagents on-board the processing module, prior to being dispensed onto the slide. These reagents are known as “Mixed” reagents. Mixed reagents may exist **either** within a Research Reagent System **or** as Ancillary reagents - but **not** both - for any given protocol.

The following mixing ratios are available for on-board reagent mixing:

- Mixed_1, 20A:1B (20:1)
- Mixed_2, 2A:1B (2:1)
- Mixed_3, 1A:1B (1:1)
- Mixed_4, 50A:1B (50:1)
- Mixed_5, 100A:1B (100:1)
- Mixed_6, 60A:1B (60:1)
- Mixed_7, 50A:1B (50:1)

Mixing ratios are a mixture of two or more reagent components at a fixed ratio. Each reagent component to be mixed is assigned to either an open container or titration container. The instrument will then mix the chromogen either within 10 minutes (e.g. Mixed_1, 20A:1B (10') Mixture) or 300 minutes (e.g. Mixed_1,20A:1B (300') Mixture) prior to dispensing onto the slide. For a list of all mixing ratios, refer to the Protocol Setup screen.

The following scenarios indicate the steps taken to achieve on-board mixing in a 1:20 ratio and link them to a protocol, either as research reagent system components, or as ancillary reagents:

Scenario 1 (all mixed components are on a research reagent system)

- 1 User creates a research reagent system (R1) with *1:20 Part A, *1:20 Part B.
- 2 User creates a protocol where one (or two) steps use *Mixed 1A:20B and selects R1 as the preferred research reagent system.
- 3 The protocol can be saved and used for a run (providing research reagent system R1 is loaded on the processing module).

Scenario 2 (all mixed components are not on a research reagent system)

- 1 User creates a research reagent system (R1) which does not have either of *1:20 Part A, *1:20 Part B.
- 2 User registers an open container for each of *1:20 Part A, *1:20 Part B.
- 3 User creates a protocol where one (or two) steps use *Mixed 1A:20B and selects R1 as the preferred research reagent system. One step in the protocol must also use at least one reagent from the preferred research reagent system.
- 4 The protocol can be saved and used for a run (providing research reagent system R1 and both open containers are loaded on the processing module).

8.5 Reagent Panels Screen

A panel is a user-defined set of markers. You can use panels to quickly add a number of slides onto the system.

Panels can only be used for routine single-stain slides and parallel multiplex slides; they cannot be used to set up sequential-stain slides. You must have a supervisor user role to create panels.

To display the **Reagent Panels** screen, click the **Reagent setup** icon on the function bar, then click the **Panels** tab.

For more information see:

- [8.5.1 Creating a Panel](#)
- [8.5.2 Viewing or Editing Panel Details](#)
- [8.5.3 Removing a Panel](#)

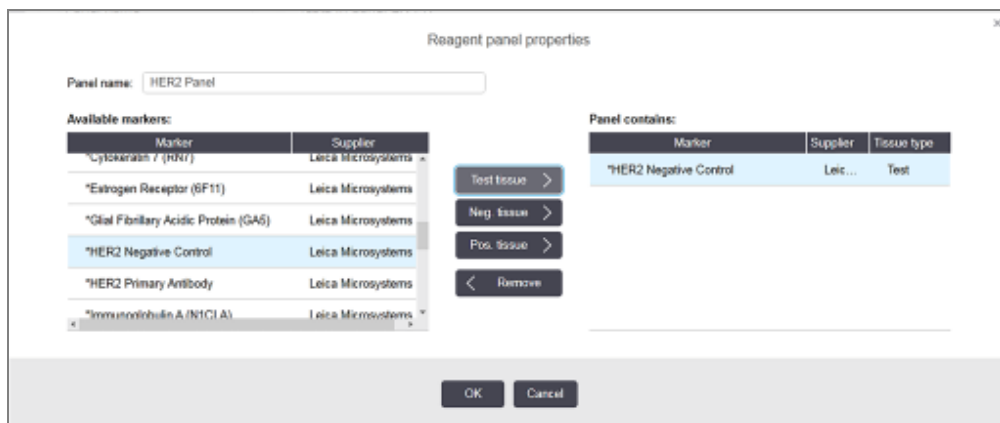
8.5.1 Creating a Panel

To create a panel, do the following (you must have a supervisor user role):

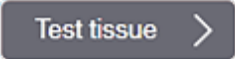
- 1 Click **Add panel**.

The software will display the **Reagent panel properties** dialog.

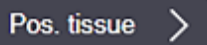
Figure 8-9: The **Reagent panel properties** dialog



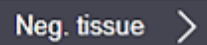
The table on the right of the **Reagent panel properties** dialog lists the contents of the panel, and the table on the left lists all of the available markers.

- 2 Enter a name for the panel in the **Panel name** field at the top of the dialog.
You cannot save a panel without a name.
- 3 To add a marker to the panel, select an item on the list of available antibodies or probes in the table at the left, then click .

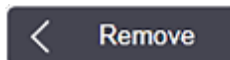
To add a positive tissue control, click on the marker then click



To add a negative tissue control, click on the marker then click



- 4 To remove an item from the panel, select it in the table on the right and click



Panels must have Test tissue. You cannot save a panel that does not have Test tissue.

- 5 When the panel is correct, click **OK** to save the details.
If you do not want to save the panel, click **Cancel**.

8.5.2 Viewing or Editing Panel Details

To view the details of a panel, select it in the table on the left of the **Reagent Panels** screen. The markers in the panel are displayed in the table on the right of the screen. To edit the panel, click **Panel properties** and edit as described in

[8.5.1 Creating a Panel](#).

8.5.3 Removing a Panel

To remove a panel from the system, select it in the table on the **Reagent Panels** screen, then click **Remove panel**. You will be asked to confirm the removal.



Remove panels with care. You cannot recover details of deleted panels.

9

Slide History (on BOND RX Controller)

The **Slide history** screen displays details of slides that are scheduled, currently running, or have been run, on the BOND RX system.

Runs that were scheduled but stopped before processing started (by unlocking the tray), have their individual slide records removed from the history list and replaced with a single row for the entire tray, showing status "Rejected". Run events and run details reports can be generated for these runs.

This chapter has the following sections:

- [9.1 Slide History Screen](#)
- [9.2 Slide Selection](#)
- [9.3 Slide Properties and Slide Rerun](#)
- [9.4 Run Events Report](#)
- [9.5 Run Details Report](#)
- [9.6 Study Report](#)
- [9.7 Protocol Report](#)
- [9.8 Slides Summary](#)
- [9.9 Export Data](#)
- [9.10 Brief Slide History](#)

9.1 Slide History Screen

To see slide history details or to generate run events, run details, or study reports, select the **Slide history** icon on the function bar.



Figure 9-1: Slide history screen

Slide history

Slide filters: Date range: From: 01-Jan-13 3:35 PM To: 06-Apr-17 3:35 PM Last seven days Apply

Process date	Run ID	Slide ID	Marker	Study name	Study ID	Type	Status
27-Aug-13	84	00000288	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000289	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000241	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000291	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000292	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000290	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000293	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000294	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000295	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
27-Aug-13	84	00000296	*Neg	Chirs P. Bacon	CS205 - 255790	Test	In progress
26-Aug-13	90	00000399	*CD5	Cherry Dale	CS3225 - 527991	Test	In progress
26-Aug-13	90	00000398	*Tyros	Jacob Dean	CS3225 - 527990	Test	In progress
26-Aug-13	90	00000396	*CD20	Jacob Dean	CS3225 - 527990	Test	In progress
26-Aug-13	90	00000395	*CD5	Jacob Dean	CS3225 - 527990	Test	In progress
26-Aug-13	90	00000394	*Tyros	Amanda Francis	CS3224 - 527909	Test	In progress
26-Aug-13	90	00000391	*CD5	Amanda Francis	CS3224 - 527909	Test	In progress
26-Aug-13	90	00000400	*CD20	Cherry Dale	CS3225 - 527991	Test	In progress
26-Aug-13	90	00000397	*MeIA	Jacob Dean	CS3225 - 527990	Test	In progress
26-Aug-13	90	00000393	*MeIA	Amanda Francis	CS3224 - 527909	Test	In progress

Run ID numbers may not increment sequentially

Slides summary Export data Brief slide history Slide properties Run events Run details Study report Protocol report

The slide history list displays the slides run in the period defined in the **Date range** filter above the list, or a specific slide found from the **Slide ID** filter (see [9.2 Slide Selection](#)).

Note that run ID numbers displayed on the screen may not increment sequentially. For the BOND RX and BOND RX^m Processing Modules, run ID numbers are allocated when slide trays are locked, so if a tray is locked, unlocked, and then locked again (before the run starts) the run ID number increments, and the number allocated after the first locking is skipped.

Slide color-coding follows that used on the **Slide setup** screen (see [6.5.1 Description of Slide Fields and Controls](#)):

- White: slides created in the **Add slide** dialog
- Yellow: slides created in the **Slide identification** dialog (see [6.8 Impromptu Slide and Study Creation](#))
- Light gray: LIS slides
- Red: priority LIS slides (see [11.2.5 Priority Slides](#))

Each slide has the following values reported in the list:

- Process date (the date slide processing started)
- Run ID
- Slide ID
- Marker (name of the primary antibody or probe)
- Study name
- Study ID
- Type (test tissue, or positive or negative control tissue)
- Status (in progress or done, and whether any unexpected events were noted; also possibly “Rejected” for runs that were stopped before processing began)



If the status is **Done (notification)**, inspect the Run Events Report to determine whether the unexpected events may have affected the staining. Unexpected events are displayed in bold text.

To view information about a slide, select it in the list, then click one of the buttons below the list.

9.2 Slide Selection

Filter the slides to list in the **Slide history** screen by showing all slides processed within a defined period, or display a specific slide by entering its slide ID. Click the drop-down menu and then choose the slide filter you want to use.

Date range slide filter

Figure 9-2: Date range slide filter

Process date	Run ID	Slide ID	Marker	Study name	Study ID	Type	Status
--------------	--------	----------	--------	------------	----------	------	--------

Use the **Date range** slide filter to specify the screen’s reporting period; only slides processed within the period are displayed. Set “From” and “To” dates and, if required, times, to define the time period to show. Then click **Apply** to display the slides.

If more than 1000 slides were processed in the period you define, only the first 1000 are displayed. To view details of the complete set you must export the slide data – see [9.9 Export Data](#).

The **To** field is initially set to the current date and time, and the **From** field to exactly one week prior. If you change settings you can return to this configuration by clicking **Last seven days**.

Using the Date & Time Selectors

To set day, month and year, click the calendar icon and select a date. Scroll through months by clicking the arrows in the calendar title bar. Or click in the center of the title bar to select another month or scroll through years. Alternatively you can type the date directly in the field.

To set the time, click in the time field and use the up and down buttons (or keyboard up and down arrow keys). Depending where the cursor is placed the time changes by one hour, ten minutes or one minute. Alternatively you can type the time directly in the field.

Slide ID slide filter

Use the **Slide ID** slide filter to locate information about a specific slide. Type the slide ID in the **Slide ID** field and click **Apply**.

9.3 Slide Properties and Slide Rerun

To view the properties of a slide in the **Slide history** list, select the slide then click **Slide properties** (or double-click). This is the same dialog opened from the **Slide setup** screen ([6.5.4 Editing a Slide](#)).

You cannot edit any of the study or test details in the **Slide properties** dialog when it is opened from the **Slide history** screen (since the slide has been, or is being, processed) but you can add comments in the **Comments** field or rerun slides – see [9.3.1 Rerunning Slides](#).

9.3.1 Rerunning Slides

If the slide does not conform to requirements, then it may be flagged to be rerun. Use the following procedure to initiate a slide rerun from the **Slide properties** dialog:

- 1 Click **Copy slide**.

The **Slide properties** dialog changes to the **Add slide** dialog, with editable fields.

- 2 Make any required changes and then click **Add slide**.

A dialog opens for you to confirm study details. You can proceed or cancel.

The **Add slide** dialog remains open to allow you to add more slides if you want.

- 3 Click **Close** to return to **Slide history** screen.
- 4 Run the newly created slides in the normal manner.

9.4 Run Events Report

Generated from the **Slide history** screen, this report shows all events for all the slides on the tray that the selected slide was run with. Click **Run events** to generate the report.

For BOND RX or BOND RX^m, Run Events Reports can also be generated while slides are being processed. Right-click on the appropriate run or list in the **System status** or **Protocol status** screens and select **Run events** from the menu. Events that initiated a slide notification are displayed in bold type so they can be easily found.

The top right of the Run Events Report shows the information in the following table:

Field	Description
PM serial No.	The serial number of the processing module used for the run
Processing module	The name of the processing module used for the run
Slide tray	The number of the slide staining assembly (on the BOND RX or BOND RX ^m) used for the run
Dispense volume	The volume of reagent dispensed (see 6.5.8 Dispense Volumes and Tissue Position on Slides)
Start time	The date and time that the run was started
Run progress	Whether the run is Finished or still Processing
Staining mode	The staining mode used, for example Single routine

Images of the slide labels for all the slides in the run are displayed at the top of the report. The body of the report displays the time, event number, and event description of the events for the run. The event number is used by Leica Biosystems for error tracking if the need arises.

See [3.7 Reports](#) for further details about the report window and printing options.

9.5 Run Details Report

Generated from the **Slide history** screen, this report shows the details of each slide on the same tray as the currently selected slide. The tray must have finished processing and be unlocked. Click **Run details** to generate the report. The top right of the report shows the information in the following table:

Field	Description
PM serial No.	The serial number of the processing module used for the run
PM name	The name of the processing module used for the run
Slide tray	The number of the slide staining assembly (on the BOND RX or BOND RX ^m) used for the run
Start time	The date and time that the run was started
Run started by	The username of the person that started the run

For each slide in the run the body of the report shows an image of the slide label and the following information.

Field	Description
Slide ID	The BOND RX system assigns a unique identifier to each slide
Slide created by	Username of the person who created the slide, or "LIS" where relevant
Study No.	A unique study identifier generated by the BOND RX software
Tissue type	Test tissue, positive control tissue, or negative control tissue
Dispense volume	The volume of reagent dispensed (see 6.5.8 Dispense Volumes and Tissue Position on Slides)
Study name	Identification of the study
Study ID	Study identification entered during slide setup
Staining protocol	The staining protocol used
Preparation	The preparation protocol used (if any)
HIER protocol	HIER protocol used (if any)
Enzyme protocol	Enzyme retrieval protocol used (if any)
Denaturation	For ISH only, denaturation protocol used (if any)
Hybridization	For ISH only, hybridization protocol used (if any)
LIS reference [2 to 7]	Additional LIS reference information for systems with LIS-ip installed (see 11.2.6 LIS Slide Data Fields)
Stain	The staining mode used, for example Single routine
Completion status	Indicates whether the slide is being processed, completed, or has been scored. Also whether any notification events were reported.
Comments	Comments can be entered into a slide's properties at any time
Sign off:	Sign off is a reserved space on the printed paper report where a supervisor can sign off each slide
Reagents Used (or preferred kit/ancillaries containing ingredients of a mixed reagent)	
UPI	Unique Pack Identifier of every reagent or the preferred kit/ancillaries used for this slide
Name	Name of every reagent or preferred kit/ancillaries used for this slide
Public name	Public name, for systems with LIS-ip installed
Lot No.	Lot number of every reagent or preferred kit/ancillaries used for this slide
Expiration Date	Expiration date of every reagent or preferred kit/ancillaries used for this slide

See [3.7 Reports](#) for further details about the report window and printing options.

9.6 Study Report

This report shows the details of each slide in the same study as the currently selected slide. The report can be generated from the **Slide setup** screen, the **Slide history** screen, and the **Slide identification** dialog. The top right of the study report shows the information in the following table:

Field	Description
Study ID	Study identification entered during slide setup
Study name	Study name
Study comments	Additional study information
Researcher	Name of the researcher in charge of the study
Researcher comments	Additional researcher information
Created	Date and time that the study was created
Study No.	A unique study identifier generated by the BOND RX system

The body of the report shows the following information for each slide in the study:

Field	Description
Slide ID	The BOND RX system assigns a unique identifier to each slide
Slide created by	Username of the person who created the slide, or "LIS" where relevant.
Run	The number of the run in which the slide was processed
Run started by	The username of the person that started the run
Tissue type	Test tissue, positive control tissue, or negative control tissue
Dispense volume	The volume of reagent dispensed (see 6.5.8 Dispense Volumes and Tissue Position on Slides)
Staining protocol	The staining protocol used
Preparation	The preparation protocol used (if any)
HIER protocol	HIER protocol used (if any)
Enzyme protocol	Enzyme retrieval protocol used (if any)
Denaturation	For ISH only, denaturation protocol used (if any)
Hybridization	For ISH only, hybridization protocol used (if any)
LIS reference (2 to 7)	Additional LIS reference information for systems with LIS-ip installed (see 11.2.6 LIS Slide Data Fields)
Stain	The staining mode used, for example Single routine

Field	Description
Completion status	Indicates whether the slide is being processed, completed, or has been scored. Also whether any notification events were reported.
Comments	Comments can be entered into a slide's properties at any time
Sign off:	Sign off is a reserved space on the printed paper report where a supervisor can sign off the Score and Comments
Reagents Used	
UPI	Unique Pack Identifier of every reagent used for this slide
Name	Name of every reagent used for this slide
Public name	Public name, for systems with LIS-ip installed
Lot No.	Lot number of every reagent used for this slide
Expiration Date	Expiration date of every reagent used for this slide

See [3.7 Reports](#) for further details about the report window and printing options.

9.7 Protocol Report

To generate reports of the protocols used for selected slides, select a slide then click **Protocol report**. Select the protocol you want from those run on the slide, and then click **Report** to create the report. See [7.5 Protocol Reports](#) for a description of the report.

9.8 Slides Summary

The slide processing summary shows the number of slides started in a stipulated period. The information is displayed in both tabular and graphical format as the number of slides processed per unit time, within the stipulated period.

To report the number of slides processed, click **Slides summary** on the **Slide history** screen, to open the **Slides summary** dialog.

Choose either a particular processing module by its name or **All** (all processing modules, or in BOND RX-ADVANCE all processing modules in the pod that the client is currently connected to) from the **Processing module** drop-down list.

In the **Resolution** field select the time unit to be used to report the number of slides started, e.g. "Day" generates a report showing the number of slides started each day within the stipulated time period, while "Month" gives the number of slides started each month within the period.

Set **To** and **From** dates. The unit of time set in the **Resolution** field starts from the **From** date and continues in full units until near the **To** date, where a part-unit may be required to complete the period.

Click **Generate** to preview the report.

See [3.7 Reports](#) for further details about the report window and printing options.

9.9 Export Data

On the **Slide history** screen, click **Export data** to create a file containing the details of all slides that have completed processing in the selected date range. The exported file is in the standard “comma separated values” (csv) file format and the file can be easily imported into third-party spreadsheet applications such as Microsoft Excel. Once imported into a spreadsheet, the data is presented in a format that allows (depending on spreadsheet functionality) sorting, searching and the creation of customized reports and graphs.

For each slide in the selected date range, the following information will be included in the exported file:

- Process date
- PM serial number
- Slide ID
- Run started by
- Marker UPI
- Marker UPI 2
- Marker UPI 3
- Marker UPI 4
- Marker UPI 5
- Marker UPI 6
- Study name
- Tissue type (test, or positive or negative control)
- Status
- Comments
- Preparation protocol name
- HIER protocol name
- HIER protocol name 2
- HIER protocol name 3
- HIER protocol name 4
- HIER protocol name 5
- HIER protocol name 6
- Enzyme protocol name
- Enzyme protocol name 2
- Enzyme protocol name 3
- Enzyme protocol name 4
- Enzyme protocol name 5
- Enzyme protocol name 6
- Processing module name
- Run ID
- Slide created by
- Stain
- Marker name
- Marker name 2
- Marker name 3
- Marker name 4
- Marker name 5
- Marker name 6
- Study ID
- Researcher
- Dispense volume
- Preparation protocol version
- HIER protocol version
- HIER protocol version 2
- HIER protocol version 3
- HIER protocol version 4
- HIER protocol version 5
- HIER protocol version 6
- Enzyme protocol version
- Enzyme protocol version 2
- Enzyme protocol version 3
- Enzyme protocol version 4
- Enzyme protocol version 5
- Enzyme protocol version 6

- Denaturation protocol name
- Denaturation protocol name 2
- Denaturation protocol name 3
- Denaturation protocol name 4
- Denaturation protocol name 5
- Denaturation protocol name 6
- Hybridization protocol name
- Hybridization protocol name 2
- Hybridization protocol name 3
- Hybridization protocol name 4
- Hybridization protocol name 5
- Hybridization protocol name 6
- Staining protocol name
- Staining protocol name 2
- Staining protocol name 3
- Staining protocol name 4
- Staining protocol name 5
- Staining protocol name 6
- Detection system name
- Detection system name 2
- Detection system name 3
- Detection system name 4
- Detection system name 5
- Detection system name 6
- Denaturation protocol version
- Denaturation protocol version 2
- Denaturation protocol version 3
- Denaturation protocol version 4
- Denaturation protocol version 5
- Denaturation protocol version 6
- Hybridization protocol version
- Hybridization protocol version 2
- Hybridization protocol version 3
- Hybridization protocol version 4
- Hybridization protocol version 5
- Hybridization protocol version 6
- Staining protocol version
- Staining protocol version 2
- Staining protocol version 3
- Staining protocol version 4
- Staining protocol version 5
- Staining protocol version 6
- Detection system serial number
- Detection system serial number 2
- Detection system serial number 3
- Detection system serial number 4
- Detection system serial number 5
- Detection system serial number 6

The columns with the number 2 to 6 in their title are relevant only to sequential multiplex-staining slides; they define the information related to the second stain for that slide.

Use the following procedure to export slide details:


- 1 Select the required date range (refer to [9.2 Slide Selection](#)).
- 2 Click **Export data**.
- 3 Select to save the file when prompted.

The file is saved to the downloads folder (or select the **Save as** option to save to another folder).

The saved file can be opened in a standard spreadsheet program such as Microsoft Excel and manipulated according to the functions available in the application. When opening the file you may need to specify some file parameters. The file is in "csv" format, the parameters are as follows:

- The file type is **Delimited**
- The **Delimiter** or **Separator** is a **Comma**
- Use a **General** column format.



The processing start time in exported slide details will not exactly match the on-screen slide history start time. The start time displayed in the slide history screen is the time that the run start button  was pressed. However, the time reported in the exported data is the time that the run actually started processing on the processing module.

9.10 Brief Slide History

The brief slide history report displays information about all slides in the pod that were processed (or are still being processed) within the time frame used to select the slides on the **Slide history** screen. The report has a sign off area, and can be used as a record of slides processed.

To create a brief slide history report open the **Slide history** screen and set **From** and **To** dates and times, to populate the screen with all slides in the pod processed in that time (refer to [9.2 Slide Selection](#)). Click **Brief slide history** to generate the report.



In laboratories with high turnovers, the default time range on the **Slide history** screen (one week) may include thousands of slides. A report for this many slides will take minutes to generate – consider defining shorter time ranges if possible, rather than accepting the default.

The report includes the following details for each slide:

- Study ID
- Study name
- Slide ID
- Marker
- Tissue type
- Dispense volume
- Status
- Sign off

10 Administration Client (on BOND RX Controller)

All general BOND RX system configuration (apart from protocols and reagents) is carried out in a separate software application, the “administration client”. Only users with administrator role can run the administration client, where all functionality is available to them.

The administration client has the following screens, opened from icons on the function bar across the top of the client:



- [10.1 Users](#)
- [10.2 LIS](#)
- [10.3 Labels](#)
- [10.4 BXD](#)
- [10.5 Settings](#)
- [10.6 Hardware](#)

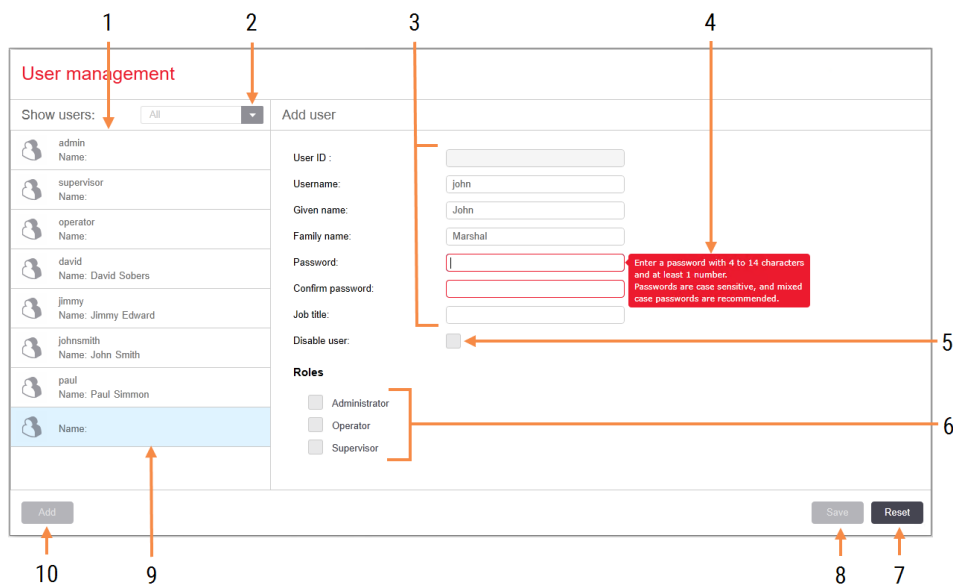
10.1 Users

BOND RX system users are managed on the administration client **User management** screen. You can create, edit and disable users. You cannot delete users – they remain in the system forever. However, you can disable users, disallowing them access to either client.



Enabled users have roles that give them different rights within the software. Only users with the administrator role can open the administration client (where they can perform all functions). Users with the operator role can register reagents, set up and process slides, and generate reports, but cannot edit reagent details, reagent panels or protocols. Users with the supervisor role have all operator rights, but can also edit reagent details, panels and protocols. Users can have more than one role.

Figure 10-1: User management screen



Legend

- | | |
|----------------------------------------------------------------------|-----------------------------------------------------------------------------------------------|
| 1 List of all BOND users | 7 Reset
Undo unsaved changes |
| 2 Filter to show all users, or just enabled or disabled users | 8 Save
Save changes for the current user |
| 3 Details of the selected user | 9 The currently selected user – their details are shown on the right of the screen |
| 4 Password requirements message | 10 Add
Click to clear the fields on the right of the screen, to add details for a new user |
| 5 Disable user
Disable (or re-enable) the currently selected user | |
| 6 Roles
Select the user's roles | |

Each new user requires a username and password. Both of these are required to log in to the research client and administration client. Once a user has been created the username cannot be changed, but the password can be. Users can change their own passwords at any time from BOND login dialogs, and administrators can also change them from the **User management** screen. Passwords must have 4–14 characters and include at least one number.



Passwords are case sensitive, and mixed-case passwords are recommended. The BOND RX software validates passwords as they are being changed; you cannot save a password until it fulfills the minimum requirements. Do not share passwords with other staff. Always log out of your account when you are away from the processing module.

Other user details (given and family names, and job title) are optional. These appear in logs and reports. The User ID is automatically assigned and appears in logs and reports.

10.2 LIS

Most LIS configuration is carried out by service staff when BOND LIS-ip is installed, however a small number of configuration options are available to users in the **LIS configuration** screen. The screen also has a log of error messages.



Figure 10-2: LIS configuration screen

The screenshot shows the 'LIS configuration' screen. At the top, it says 'LIS configuration' and 'You require a license for the BOND LIS-ip, activated with a password provided by Leica Biosystems.' Below this are several configuration options:

- 1 License: A text input field containing 'AAAAAAAA'.
- 2 Duplicate study ID: A dropdown menu with 'Resurrect existing study' selected.
- 3 Force LIS printing in BOND: A checkbox that is unchecked.
- 4 Enable LIS to update LIS slides: A checkbox that is unchecked.
- 5 Enable unprocessed LIS slide lifetime: A checkbox that is unchecked, followed by a text input field containing '1' and the label '(hrs)'.

At the bottom right of the configuration section are three buttons: 'Edit LIS data fields', 'Save', and 'Reset'. An arrow labeled '7' points to the 'Edit LIS data fields' button.

Below the configuration options is a section titled 'Log messages'. It contains a table with columns: 'Created at', 'Level', 'Log entry', and 'Source'. An arrow labeled '6' points to the table area. At the bottom right of the log messages section is a 'View log' button.

Legend

- | | |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 License
Shows the LIS-ip license password.</p> <p>2 Duplicate study ID
Set the action for studies with the same study ID as existing studies.</p> <p>3 Force LIS printing in BOND RX
Enforce that all LIS slides are printed by BOND RX. Refer to 11.7 Slide Labels.</p> <p>4 Enable LIS to update LIS slides
Overwrite (update) unprocessed slides if slides having the same barcode ID are resent by the LIS. If this setting is disabled, BOND RX will reject any attempt by the LIS to reuse the same barcode ID.</p> | <p>5 Enable unprocessed LIS slide lifetime (hrs)
Delete slides received from an LIS that are not processed within the number of hours entered.</p> <p>6 Log Messages
Are displayed as a list when you click View log (see right).</p> <p>7 Edit LIS data fields
Configure the display of slide data in BOND RX.</p> |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

License

You require a license for the BOND LIS-ip, activated with a password provided by Leica Biosystems. Typically the password is entered for you by the service staff who set up the LIS-ip connection, but if not then only the **License** field appears on the screen. Enter the password to turn on the LIS-ip functionality and to show the configuration options and log shown in [Figure 10-2](#).

Duplicate Study ID

Use the **Duplicate study ID** setting to set how to handle studies received from the LIS with the same ID as an expired or deleted LIS study, already in the BOND RX system. (If an LIS study has the same study ID as an existing BOND RX study, i.e. one created in the BOND RX system, it is automatically rejected.) There are two options:

- **Resurrect existing study:** when the new study is received, provided it has the same study name as the existing study, the existing study is resurrected (i.e. it is re-used). If the new study has the same study ID but a different study name then it is rejected.

If the researcher's name has changed, the new name is used.

- **Reject message:** the new LIS study is not transferred to the BOND RX system. A message reporting this is logged in the LIS. You must change the study ID in the LIS and resend the study.

For discussion of handling duplicate study IDs in non-LIS studies, see [6.3.4 Study Duplication, Resurrection and Expiration](#). For general information about LIS studies see [11.2.2 LIS Studies](#).

LIS Slide Data Fields

The BOND LIS-ip installation can be configured for the LIS to send the BOND RX system up to seven parameters for each slide. These are for viewing only, and are displayed on the **LIS** tab in the **Slide properties** dialog. While basic configuration of these parameters is carried out by a service technician, you can choose to hide any of the parameter fields, and set the names of the fields.

LIS is active only for single and 2-plex; not 3-6 plex.

Check fields that you want to display and type in field names.

10.3 Labels

Use the **Label templates** screen to create and edit slide label templates, and to select the templates to use.



There are six 2D template types, for use with the six slide types in the BOND RX system:

- BOND single stain
- BOND sequential multiplex stain
- BOND parallel multiplex stain
- LIS single stain
- LIS sequential multiplex stain
- LIS parallel multiplex stain

The “BOND RX” templates are for slides created in the BOND RX system, and the “LIS” templates for slides created in an LIS but printed from the BOND RX system.

These predefined templates cannot be edited or deleted.

BOND RX 7 can read 1D, 2D, and OCR barcodes, but create only 2D barcodes.

If your BOND RX system was upgraded from 5.2 or earlier, you cannot continue to use the existing barcode scanner as this earlier model does not support 2D barcodes.

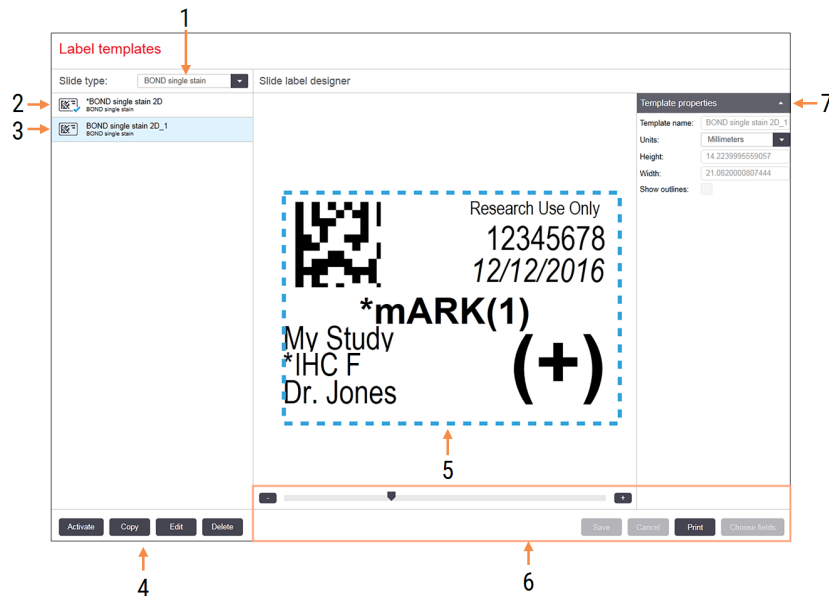
To use another template for a slide type, copy the default template and edit the resulting “user template”. Then “activate” it, to make it the template that the BOND RX system will use for slides of that type. You can create any number of templates for each slide type, but only one can be activated at a time.



WARNING: Always include sufficient information on labels to ensure that, in the case that automatic label identification fails, the labels can be identified manually. Leica Biosystems recommends that all slides include the following fields:

- Study ID or Study name
- Slide ID
- Tissue type – to identify control tissues; and
- Marker – the primary antibody or probe to be applied.

Figure 10-3: Label templates screen



Legend

- | | |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 Slide type
Select a slide type – all templates for the type are shown in the pane below</p> <p>2 Active template (with blue check mark)</p> <p>3 Selected template, displayed in editing pane to the right</p> <p>4 Template management commands – see Figure 10-4 Label template management commands</p> | <p>5 Editing pane with layout of the template selected at left</p> <p>6 Template editing commands – see Figure 10-5 Label template editing commands</p> <p>7 Template properties
Properties of the currently selected template layout as a whole (view-only until you click the Edit button in left pane)</p> |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

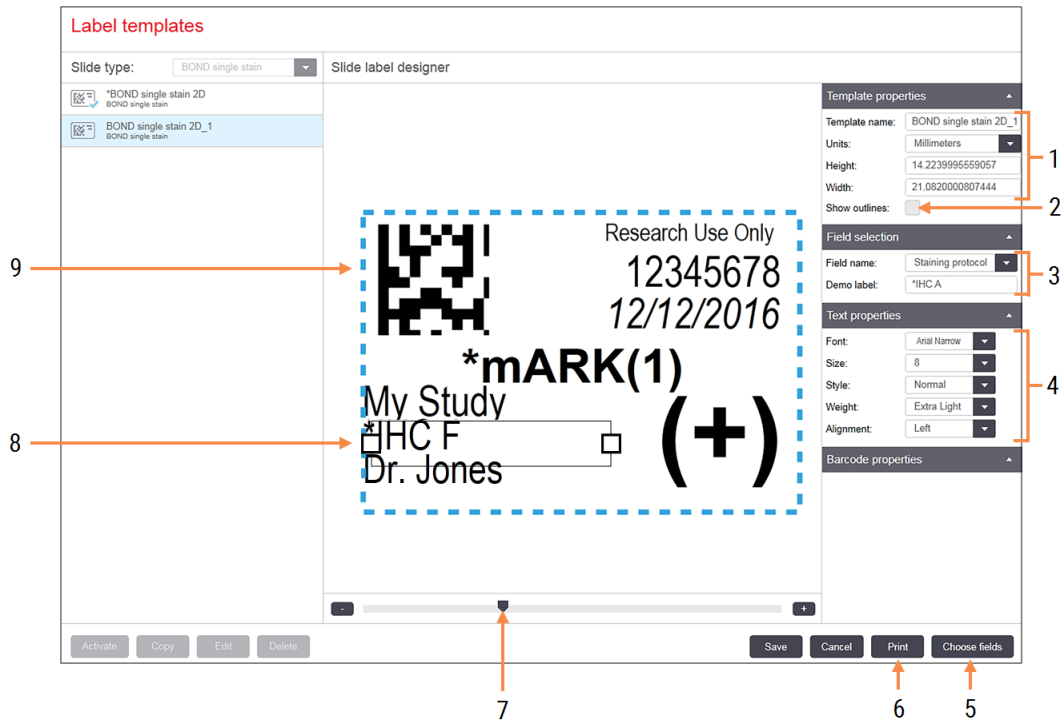
Figure 10-4: Label template management commands



Legend

- | | |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 Set the currently selected template to be used for all slide labels for the currently selected slide type.</p> <p>2 Copy the currently selected template to create a new “user” template.</p> | <p>3 Edit the currently selected template using the editing pane and commands in the right of the screen. Default templates cannot be edited.</p> <p>4 Delete the currently selected template. Default templates cannot be deleted.</p> |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

Figure 10-5: Label template editing commands



Legend

- | | |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 Template properties
Enter the template name and size</p> <p>2 Show outlines
Display field outlines in the editing pane</p> <p>3 Field selection
Select a field type to highlight the field in the editing pane. Enter demo text for the field.</p> <p>4 Text properties
Configure text properties for the selected field</p> | <p>5 Choose fields
Open the Choose fields dialog to add or remove fields from the layout</p> <p>6 Print
Print the current layout on a selected printer</p> <p>7 Sliding control to zoom the demo label in and out</p> <p>8 The currently selected field – configure in the Text Properties pane to the right. Drag the boxes at either end to change the width, or the whole field to reposition.</p> <p>9 The Label ID or barcode field – must not be resized</p> |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

See also:

- [10.3.1 Create, Edit, and Activate Label Templates](#)
- [10.3.2 Information Types](#)

10.3.1 Create, Edit, and Activate Label Templates

Create new templates by copying existing ones and editing them, or you can edit existing user templates (but not the default templates). Activate a template to make it the one used for labels printed from the BOND RX system.

The watermark “Research Use Only” always appears on labels in the BOND RX system. This watermark cannot be edited, or deleted from label templates.

- [10.3.1.1 Create a New Template](#)
- [10.3.1.2 Edit a Template](#)
- [10.3.1.3 Activate a Template](#)

10.3.1.1 Create a New Template

- 1 Select the slide type the new template is for.
All existing templates for the slide type are displayed.
- 2 Select a template to copy (select the template most similar to the one you want to create).
- 3 Click **Copy**.



Copying a template with a 2D barcode will create a new “user template” with a 2D barcode.

10.3.1.2 Edit a Template

- 1 Select a template in the left hand pane and click **Edit**.
The editing pane, buttons, and properties lists on the right of the screen are enabled for you to edit the template layout, displayed in the editing pane.
- 2 Optionally select **Show outlines** (in **Template properties** section, top right) to view field boundaries in the editing pane.
- 3 Enter the template name in the **Template properties** section.



There is a 64-character limit for label template names, also all names used in the same slide type category must be unique.

- 4 Edit the layout:
 - a Add or remove fields – click **Choose fields** and select the slide properties to show (see [10.3.2 Information Types](#) for a list of all the properties available).
Note that you cannot remove the **Label ID** field, which is used for automatic identification.
 - b Position fields – select and drag the fields in the editing pane.

- c Resize field widths – drag the boxes at either end of the fields. (Field heights are set by the text font size.)
If the field width you set is not long enough for the value on a particular label when the template is used, the text is truncated and ellipsis points appended so that it is clear that truncation has occurred.



You must not resize the **Label ID** field – it must remain at its default setting so it can be read by the processing module imager.

- d Set text properties – select a field and set its font and font size, style and weight in the **Text properties** section. Also set the text alignment in the field.

5 Click **Save**.



Make sure there is clear space around the **Label ID** field. If text from any other fields impinge on this area it can interfere with automatic identification.

10.3.1.3 Activate a Template

- 1 Select a template in the left hand pane and click **Activate**.

The template is marked with a blue check mark, indicating it is now active.

10.3.2 Information Types

Label templates can be configured to show any of the following slide information, selected from the **Choose fields** dialog on the **Labels** screen.

The **Label ID** field, used for automatic identification, cannot be removed from any template. It appears as a 2D barcode.

Field	Description
Study ID	The study ID for the slide (N.B. not the Study No. – see 6.3.2 Study Identification).
Slide created by	Username of the person who was logged into the client when the slide was created, or "LIS" where relevant.
Denaturation protocol	Abbreviated name of the denaturation protocol.
Denaturation protocol 2	Abbreviated name of the second denaturation protocol (may be required for multiplex staining protocols).
Denaturation protocol 3	Abbreviated name of the third denaturation protocol (may be required for multiplex staining protocols).
Denaturation protocol 4	Abbreviated name of the fourth denaturation protocol (may be required for multiplex staining protocols).
Denaturation protocol 5	Abbreviated name of the fifth denaturation protocol (may be required for multiplex staining protocols).

Field	Description
Denaturation protocol 6	Abbreviated name of the sixth denaturation protocol (may be required for multiplex staining protocols).
Dispense volume	100 µL or 150 µL dispense volume.
Researcher comment	A comment recorded in the BOND RX system for the researcher in charge of the study (see 6.4 Manage Researchers).
Researcher	The name of the researcher in charge of the study.
EIER protocol	Abbreviated name of the enzyme protocol.
EIER protocol 2	Abbreviated name of the second enzyme protocol (may be required for mutliplex staining protocols).
EIER protocol 3	Abbreviated name of the third enzyme protocol (may be required for mutliplex staining protocols).
EIER protocol 4	Abbreviated name of the fourth enzyme protocol (may be required for mutliplex staining protocols).
EIER protocol 5	Abbreviated name of the fifth enzyme protocol (may be required for mutliplex staining protocols).
EIER protocol 6	Abbreviated name of the sixth enzyme protocol (may be required for mutliplex staining protocols).
Facility	The name of the facility as entered in the Facility field on the administration client Laboratory settings screen – see 10.5.1 Laboratory Settings .
HIER protocol	Abbreviated name of the HIER protocol
HIER protocol 2	Abbreviated name of the second HIER protocol (may be required by multiplex staining protocols).
HIER protocol 3	Abbreviated name of the third HIER protocol (may be required by multiplex staining protocols).
HIER protocol 4	Abbreviated name of the fourth HIER protocol (may be required by multiplex staining protocols).
HIER protocol 5	Abbreviated name of the fifth HIER protocol (may be required by multiplex staining protocols).
HIER protocol 6	Abbreviated name of the sixth HIER protocol (may be required by multiplex staining protocols).
Hybridization protocol	Abbreviated name of the ISH hybridization protocol.
Hybridization protocol 2	Abbreviated name of the second ISH hybridization protocol (may be required for multiplex staining protocols).

Field	Description
Hybridization protocol 3	Abbreviated name of the third ISH hybridization protocol (may be required for multiplex staining protocols).
Hybridization protocol 4	Abbreviated name of the fourth ISH hybridization protocol (may be required for multiplex staining protocols).
Hybridization protocol 5	Abbreviated name of the fifth ISH hybridization protocol (may be required for multiplex staining protocols).
Hybridization protocol 6	Abbreviated name of the sixth ISH hybridization protocol (may be required for multiplex staining protocols).
LIS researcher comment	For LIS-ip systems, the comment for the researcher in the LIS system.
LIS researcher	For LIS-ip systems, the researcher name.
LIS reference [2–8]	LIS slide properties imported into the BOND RX system. See 11.2.6 LIS Slide Data Fields .
Marker	Abbreviated name of the primary antibody or probe for a single stain, parallel multiplex stain, or the first stain of a sequential multiplex stain.
Marker 2	Abbreviated name of the primary antibody or probe for the second stain of a multiplex stain.
Marker 3	Abbreviated name of the primary antibody or probe for the third stain of a multiplex stain.
Marker 4	Abbreviated name of the primary antibody or probe for the fourth stain of a multiplex stain.
Marker 5	Abbreviated name of the primary antibody or probe for the fifth stain of a multiplex stain.
Marker 6	Abbreviated name of the primary antibody or probe for the sixth stain of a multiplex stain.
Study comment	Study comment (see 6.3.3 Adding a Study).
Study	The study name.
Preparation protocol	Abbreviated name of the preparation protocol.
Public name	For LIS-ip systems, the public name of the primary antibody or probe (see 11.2.4 Public Marker Names), for a single stain or the first stain of a multiplex stain.
Public name 2	For LIS-ip systems, the public name of the primary antibody or probe (see 11.2.4 Public Marker Names), for the second stain of a multiplex stain.
Slide comment	Slide comment (see 6.5.2 Creating a Slide).

Field	Description
Slide date	The date that the label was printed (short format as set in the Windows Regional and Language Options (Control Panel)).
Slide ID	8-digit numeric slide ID, unique to the slide within the BOND RX system.
Slide priority	For LIS-ip systems, the priority rating for the slide.
Staining mode	Single stain or multiplex stain.
Staining protocol	Abbreviated name of the staining protocol for a single stain or the first stain of a multiplex stain.
Staining protocol 2	Abbreviated name of the staining protocol for the second stain of a multiplex stain.
Staining protocol 3	Abbreviated name of the staining protocol for the third stain of a multiplex stain.
Staining protocol 4	Abbreviated name of the staining protocol for the fourth stain of a multiplex stain.
Staining protocol 5	Abbreviated name of the staining protocol for the fifth stain of a multiplex stain.
Staining protocol 6	Abbreviated name of the staining protocol for the sixth stain of a multiplex stain.
Tissue type	Test tissue, or positive or negative control tissue. BOND prints “(-)” for negative control, “(+)” for positive control, and nothing for test tissue.

10.4 BXD

Use the **BXD update** screen to update the BOND RX Data Definitions and to generate audit trail files.



Figure 10-6: BXD update screen

The screenshot shows the 'BXD update' interface. At the top, there is a header 'BXD update' and a sub-header 'Leica Biosystems periodically distributes BXD (BOND RX Data Definitions) updates on the web site, e.g. to add newly released reagents.' Below this, there is a 'File selected:' field with a blue progress bar and an 'Import status: Successful' message. To the right of the status are 'Browse' and 'Load' buttons. Below the status is an 'Update log' section with a table of log entries. At the bottom right, there is an 'Export audit trail' button. Numbered callouts point to various elements: 1 points to the log table, 2 points to the file selected field, 3 points to the progress bar, 4 points to the 'Browse' button, 5 points to the 'Load' button, and 6 points to the 'Export audit trail' button.

Time	Level	Category	Log entry
23-Aug-13 11:30:29 AM	Information	BXD update	Loading BXD Version 45.01.19 for Region "RestOfWorld".
23-Aug-13 11:30:36 AM	Information	Lab management	Started
23-Aug-13 11:30:36 AM	Information	Lab management	Finished
23-Aug-13 11:30:36 AM	Information	Run management	Started
23-Aug-13 11:30:36 AM	Information	Run management	Finished
23-Aug-13 11:30:36 AM	Information	Inventory management	Started
23-Aug-13 11:30:41 AM	Information	Inventory management	Finished
23-Aug-13 11:30:41 AM	Information	Test management	Started
23-Aug-13 11:30:52 AM	Information	Test management	Finished
23-Aug-13 11:30:52 AM	Information	Instrument management	Started
23-Aug-13 11:31:49 AM	Information	Instrument management	Finished
23-Aug-13 11:31:49 AM	Information	Case management	Started

Legend

- | | | | |
|---|--------------------------------------------------------|---|--------------------------------------------------------------------------|
| 1 | Log of BXD update | 5 | Load |
| 2 | The selected BXD update file | | Click to install the BXD update file in field at left |
| 3 | BXD update progress bar and status | 6 | Export audit trail |
| 4 | Browse | | Click to generate audit trail files – 10.4.2 Audit Trail |
| | Locate BXD update file and open into the field at left | | |

See:

- [10.4.1 BXD Updates](#)
- [10.4.2 Audit Trail](#)

10.4.1 BXD Updates

Leica Biosystems periodically distributes BXD (BOND RX Data Definitions) updates on the web site, e.g. to add newly released reagents. BXD update files for BOND RX 7 have “*.bxd” file extension. Install these updates from the **BXD update** screen.

You can install a BXD update at any time.

- 1 Download the update file from the Leica Biosystems website and save to a virus-free USB stick.
- 2 Insert the USB stick into the BOND or BOND RX-ADVANCE Controller (or, alternatively, any BOND RX terminal in BOND RX-ADVANCE systems).
- 3 Open the **BXD update** screen in the administration client.
- 4 Click **Browse** and locate the update file in the Windows **Open** dialog.
- 5 Click **Open** to display the BXD file in the field near the top-left of the screen.
- 6 Click **Load** to update the definitions with the new data.

Messages are written to the **Update Log** as the update proceeds. The final row displays “BXD update: Finished” when the update is finished, and status “Successful” appears under the progress bar in the top pane.



The **BXD update** screen indicates the success of the update. The process takes only a few minutes, therefore we recommend that you wait until the update completes before navigating to a different screen.



If an update is unsuccessful, the data definitions revert to the pre-update condition, and a message to this effect appears in the Update Log. Contact customer support if an update fails.

10.4.2 Audit Trail

You can generate an audit trail of all changes to the system, including who made the changes and when. The audit trail is written to multiple CSV files, each recording a different category of information. Files are written to folder: BOND Drop-box\Audit\YYYYMMDD-HHmmss on the controller.

To create audit trail files:

- 1 Open the **BXD update** screen and click **Export audit trail**.
- 2 Select **All data** to report all changes in the entire life of the system, or **Custom date range** to define a specific period, then define **From** and **To** dates and times.
- 3 Click **Export**.

10.5 Settings

The **Settings** screen has general laboratory-wide settings for the BOND RX system (**Laboratory settings**) and default study and slide settings and workflow options (**Study and slide settings**).

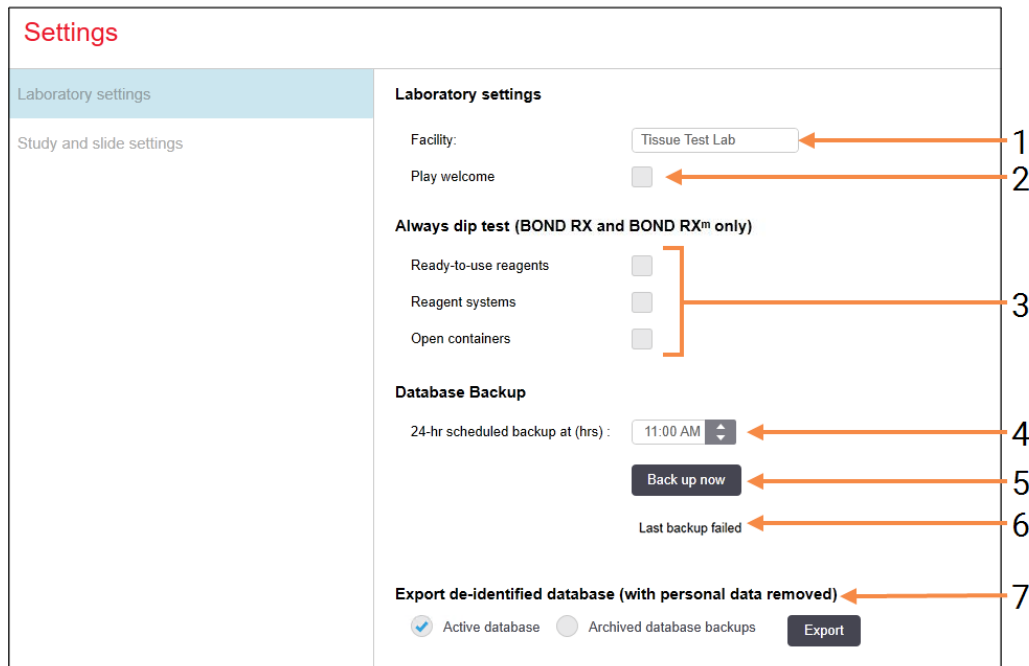


- [10.5.1 Laboratory Settings](#)
- [10.5.2 Study and Slide Settings](#)
- [10.5.3 Database Backups](#)

10.5.1 Laboratory Settings

Set general laboratory options on the **Laboratory settings** pane:

Figure 10-7: Settings screen, Laboratory settings pane



Legend

- | | |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 Facility
Type in the name of your laboratory, to appear in reports</p> <p>2 Play welcome
Play a welcome message when the BOND RX software is started</p> <p>3 Always dip test
Check to dip test reagent containers of the specified types before every run – see 8.3.1 Determining Reagent Volume</p> | <p>4 24-hr scheduled backup at (hrs)
Set a time to run daily automatic database backups (24-hour time format) – see 10.5.3 Database Backups.</p> <p>5 Back up now
Run a database backup immediately – 10.5.3 Database Backups.</p> <p>6 Information about last backup, or progress bar while a backup is in progress.</p> <p>7 Export de-identified database
Select whether you want to export de-identified data from the active database or from a database backup.</p> |
|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

10.5.2 Study and Slide Settings

The study and slide settings allow you to set:

- defaults for a number of configurable values in study and slide creation
- workflow options in study and slide creation.

See [Figure 10-8](#) and [Figure 10-9](#) for descriptions of the study and slide options.

Figure 10-8: Study settings on the **Study and slide settings** pane

Study settings

1 → Default preparation: *Dewax

2 → Default dispense volume: 150 µL

3 → Create impromptu studies or slides: Studies and slides

4 → Processed study lifetime: 30 (days)

5 → Create daily study

Legend

- | | |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 Default preparation
Default preparation protocol for new studies.</p> <p>2 Default dispense volume
Default dispense volume for new studies.</p> <p>3 Create impromptu studies or slides
Set options to create studies and/or slides after loading slides – see 6.8.2 On-Board Slide Identification Options</p> | <p>4 Processed study lifetime
The number of days a study remains on the Slide setup screen after the last slide in the study has been processed – see 6.3.4.2 Processed Study Lifetime.</p> <p>5 Create daily study
Automatically create one study daily for all slides processed on that day – see 6.3.7 Daily Study Option</p> |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

Figure 10-9: Slide settings on the **Study and slide settings** pane

Slide settings

1 → Staining mode: Single

Force printing in BOND → 3

2 → BOND label ID Barcode

Legend

- | | |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 Staining mode
Default setting for new slides – see 6.5.2 Creating a Slide</p> <p>2 BOND label ID
The label identifiers for slides created in BOND RX are 2D barcodes.</p> | <p>3 Force printing in BOND
Allow only slides with labels printed in the BOND RX system to be processed – see 6.8.2 On-Board Slide Identification Options.</p> |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

10.5.3 Database Backups

The database stores critical study information and is essential to the proper operation of the BOND RX system, so to ensure you can recover if the database is corrupted, the BOND RX system has a system of automatic and manual backups:

- Automatic daily backups
- “Manual”, on-request backups

All backup files are saved on the BOND RX controller in sub-folders of folder:

B:\BOND Drop-box\Backups

For each type of backup two files are generated, always with the same name format:

[Facility name]_BOND_YYYY-MM-DD-HH-mm-ss

where the facility name is as entered in the administration client **Settings** screen (see [10.5.1 Laboratory Settings](#)) (or it defaults to “Facility” if no facility name is entered). The name includes the date and time the backup was run. The main backup file has extension “.dump” and there is also a log file, with extension “.log”.

The automatic daily backup runs at a time set in the administration client **Settings** screen ([10.5.1 Laboratory Settings](#)). The most recent backup is in folder “Scheduled_Latest”. It is moved to folder “Scheduled_1_Days_Old” when the next day’s backup runs, and so on for another six days (to folder “Scheduled_7_Days_Old”) after which it is deleted.

If the BOND RX controller is off at the scheduled backup time, the backup is not run. Ensure you set a time when the controller will be on, and when it is unlikely that processing runs will be in progress.

You can run a manual backup at any time (except when an automatic backup is running), from the administration client **Settings** screen. Click **Back up now** in the **Database backup** section (see [10.5.1 Laboratory Settings](#)).

A dialog informs you when the backup is finished. The backup and log files are saved in folder “Manual”. At the next manual backup the files are transferred to folder “Manual_Previous”. The files are deleted after a third manual backup – that is, only the two most recent manual backups are saved.

If any type of backup fails to complete successfully an icon (right) appears at the right of the function bar in the administration and research clients. The icon remains until a successful backup is run. If the icon appears, attempt a manual backup as soon as possible. If that also fails, contact customer support immediately.



Especially on older BOND RX systems, where more data will have accumulated, check occasionally that there is enough room for the backup files. Usually, one backup file is deleted when a new one is written, so drive usage will increase in only relatively small increments. However, at some point you may need additional drive space – if so, contact customer support.

For additional security, back up the backup files to a different location (off the BOND RX controller) on a regular basis. If possible, organize with your IT department for automatic backups. If not, copy the files manually once a week (more often for laboratories with high turnovers). The BOND RX controller runs a secure FTP server so that the IT department can log in and download the backup files from the BOND Drop-box folder via secure FTP.

Contact customer support if you need to restore a database.

10.6 Hardware

Use the **Hardware configuration** screen to configure processing modules, pods (groups of processing modules controlled from one client), and slide label printers.



Hardware configuration is carried out on three tabs:

- [10.6.1 Processing Modules](#)
- [10.6.2 Pods](#)
- [10.6.3 Slide Labelers](#)

10.6.1 Processing Modules

View the processing modules in the BOND RX system and configure their bulk reagent containers on the **Processing modules** tab.

When a processing module is physically connected to the BOND RX controller with a network cable, it automatically appears in the left pane on the **Processing modules** tab.

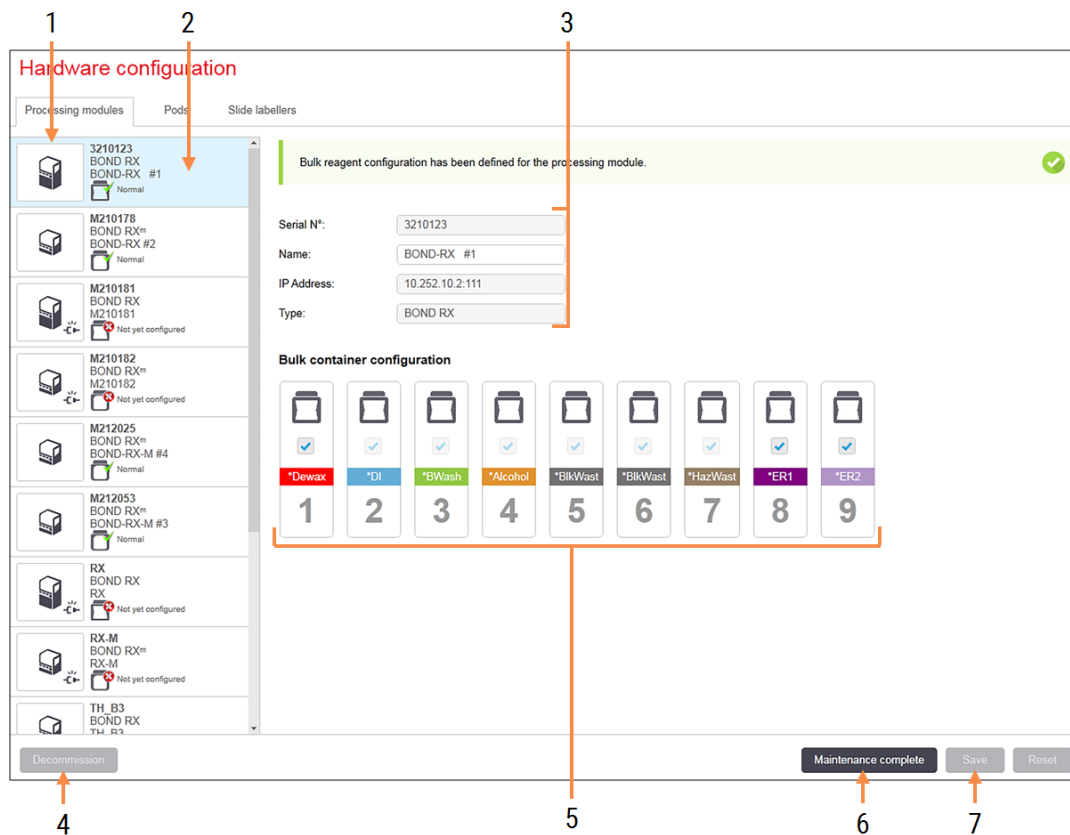


The BOND RX controller will only allow compatible processing modules to be connected. If an incompatible processing module is connected, an icon and error message are displayed (see table of icons and meanings on next page).

Select the processing module to show its details on the right of the tab. Give the processing module a unique name and, if required, disable some of the bulk containers (see [10.6.1.1 Disabling Bulk Reagent Containers](#)). When you save these settings the processing module is said to be “commissioned”.

It remains on the tab, including when it is turned off or disconnected, until you decommission it (see [10.6.1.2 Decommission a Processing Module](#)).







Figure 10-10: Processing modules tab on the Hardware configuration screen



Legend

- 1 All connected processing modules.
- 2 The currently selected processing module – its details are shown on the right of the screen.
- 3 Serial number, name (editable), IP address, and processing module type for the selected processing module.
- 4 **Decommission**
Decommission the selected processing module – see [10.6.1.2 Decommission a Processing Module](#).
- 5 Bulk container configuration – you can uncheck some stations if they will not be used – see [10.6.1.1 Disabling Bulk Reagent Containers](#) below.
- 6 **Maintenance complete**
Click to reset day and slide counts following preventative maintenance – see [Preventative Maintenance in 12 Cleaning and Maintenance \(BOND RX and BOND RX^m\)](#).
- 7 **Save**
You must save the configuration settings in order to commission a newly-connected processing module. In order to save a processing module's configuration settings, you must first ensure that all its slide staining assemblies are unlocked.

Icons beside the processing module images in the left pane indicate when the modules are in various states:

Icon	Meaning	Icon	Meaning
	The processing module is not connected.		The processing module is undergoing a maintenance operation. This icon is also displayed (together with an error message) if the connected processing module is incompatible with the BOND RX system.
	The processing module is initializing.		The bulk reagent configuration has not been received by the processing module. Click Save to send the configuration.
	The processing module is currently being serviced.		The bulk reagent configuration has been received by the processing module.

10.6.1.1 Disabling Bulk Reagent Containers

Laboratories that do not carry out epitope retrieval and/or dewaxing on the BOND RX system can disable the containers in the software and remove the relevant containers from the processing module. The containers do not then have to be maintained with reagent in them, and processing module initialization is sped up as the fluid lines to the containers are not primed. To disable bulk containers uncheck them in the **Bulk container configuration** pane and click **Save**. When prompted, restart the processing module for the changes to take effect. You can remove the disabled containers or leave them in position on the processing module.

10.6.1.2 Decommission a Processing Module

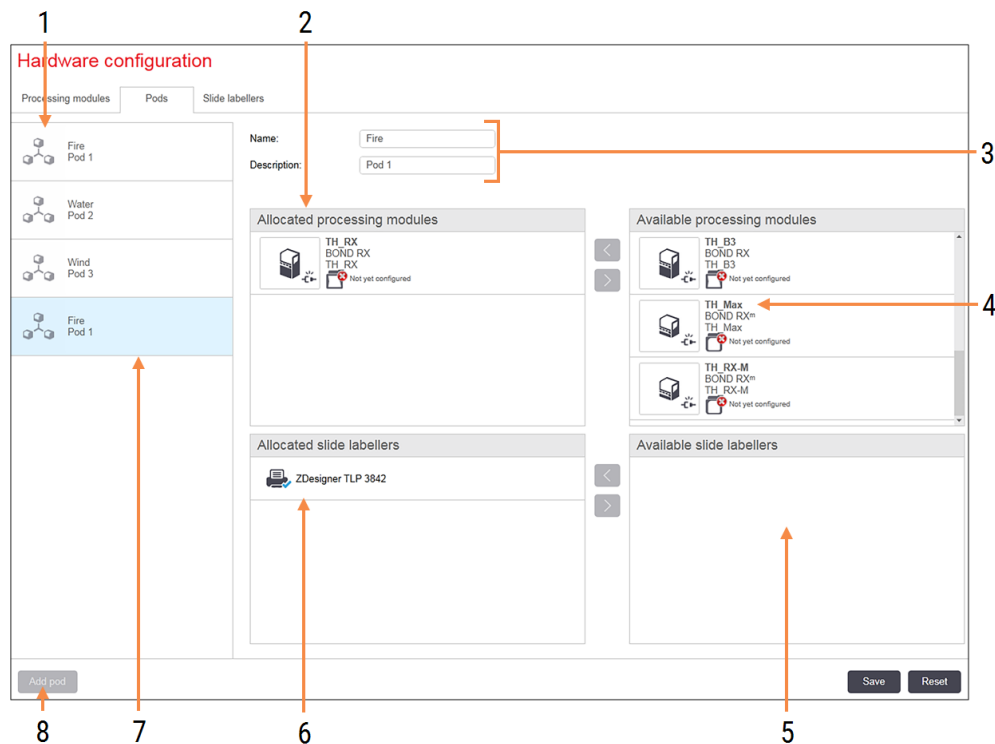
If you no longer need a processing module, decommission it to remove it from the **Processing modules** tab. Make sure that the processing module is turned off, then select it on the **Processing modules** tab and click **Decommission**. If the processing module is still in a pod, it will be automatically removed from the pod when it is decommissioned.

To recommission a processing module, reconnect its network cable.

10.6.2 Pods

Pods are collections of processing modules (and slide label printers) that can be controlled from a research client – see [3.1 System Architecture](#). Create a pod for all processing modules controlled from the BOND RX controller. Create and edit pods on the **Pods** tab.

Figure 10-11: Pods tab on the Hardware configuration screen



Legend

- | | |
|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <p>1 List of all pods</p> <p>2 Processing modules in the selected pod. The same ordering is used in the research client – see 10.6.2.1 Create a New Pod below.</p> <p>3 Name and description (both editable) of the selected pod.</p> <p>4 All processing modules that are not in pods.</p> <p>5 All slide labelers that are not in pods.</p> | <p>6 Slide label printers in the selected pod. The default printer is marked with a blue check mark – see 10.6.2.1 Create a New Pod below.</p> <p>7 The currently selected pod – its details are shown on the right of the screen.</p> <p>8 Add pod
Click to configure a new pod – see 10.6.2.1 Create a New Pod below.</p> <p>Delete
Right-click an empty pod and click Delete to delete it.</p> |
|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

To make processing modules available for inclusion in a pod, configure them on the **Processing modules** tab (see [10.6.1 Processing Modules](#)). To make slide labelers available for inclusion in a pod, configure them on the **Slide labelers** tab (see [10.6.3 Slide Labelers](#)).

10.6.2.1 Create a New Pod

- 1 Click **Add pod**.
- 2 Enter a unique pod name and optionally, description.
- 3 Select processing modules from the **Available processing modules** pane (top right) and click the left-arrow button



to add them to the **Allocated processing modules** pane (top left).

If adding multiple processing modules, add them in the order that you want the tabs to appear in the research client, e.g. if you select processing module A first and processing module B second, A will appear above B in the pane, and in the **System status** tabs in clients connected to the pod. To reorder processing modules, remove

them with the right-arrow button  and then replace in the correct order.

- 4 Select one or more slide label printers from the **Available slide labelers** pane (bottom right) and add to the **Allocated slide labelers** pane (bottom left).

If you add multiple printers they are all available for selection when printing slides. Set the default printer by right-clicking and clicking **Set as default printer**. The default printer has a blue check mark.

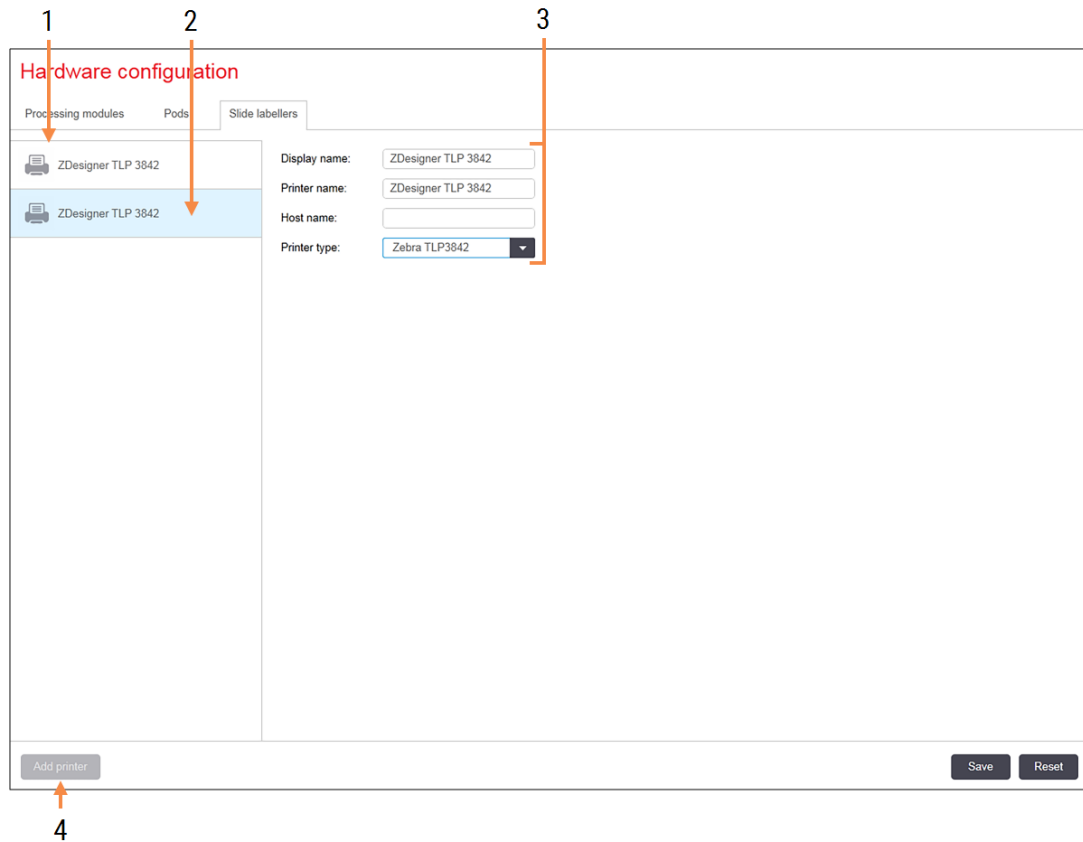
- 5 Click **Save**.

To delete a pod, remove all processing modules and printers then right-click on the pod in the left pane and click **Delete**.

10.6.3 Slide Labelers

Slide labelers used by the BOND RX system must be located, identified and activated in the administration client **Hardware configuration** screen, **Slide labelers** tab. This makes them available to be included in pods (see [10.6.2 Pods](#)).

Figure 10-12: Slide labelers tab on the **Hardware configuration** screen



Legend

- 1 List of all slide labelers.
- 2 The currently selected slide labeler – its details are shown on the right of the screen.
- 3 Slide label printer details – see [10.6.3.1 Slide Label Printer Details](#) below.
- 4 **Add printer**
Click to add a new slide labeler – configure on the right of the screen.

To make a newly connected slide labeler available for inclusion in a pod, click **Add printer** then enter the printer details on the right of the screen.



Not all installations have pods. If there are no pods then the default printer is the first printer in the list.



If a slide labeler is replaced you do not need to add a new labeler – you can replace the details of the old labeler with those of the new one.

To remove a labeler from the list, right-click it and select **Delete**.

10.6.3.1 Slide Label Printer Details

The BOND RX system requires the following details for each slide label printer:

- **Display name:** a name for the labeler that will appear in the BOND RX software
- **Printer name:** the name of the printer used by Windows



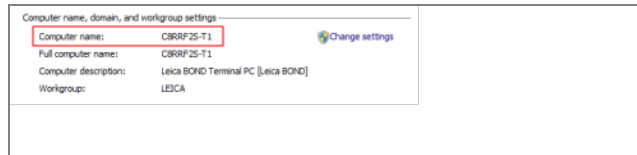
The printer name in BOND RX-ADVANCE installations is actually the printer **Share name** displayed in the Windows **Printers and Faxes** dialog.

- **Host name:** leave blank unless it is a **Zebra** printer (for example **ZDesigner TLP 3842**) on a BOND RX-ADVANCE installation, in which case enter the **Computer name** of the terminal that the slide labeler is connected to.



You can find the **Computer name** in the Windows **System** dialog (see [Figure 10-13](#)).

Figure 10-13: Computer name in Windows System dialog



- **Printer type:** the printer model (for example **ZDesigner TLP 3842**)

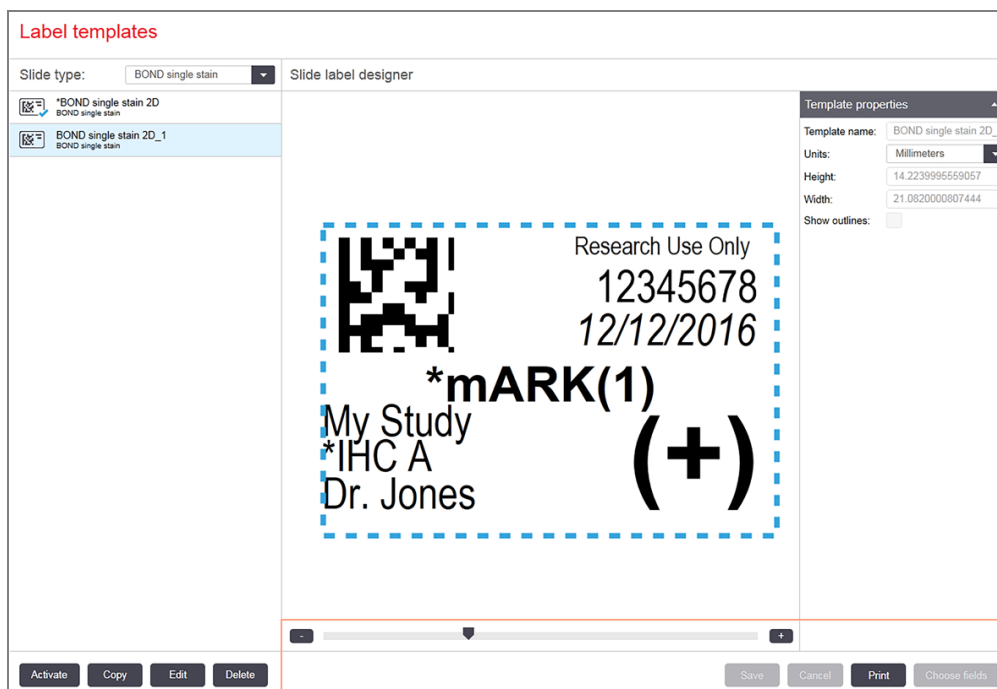
10.6.3.2 Print Test Labels

To check print alignment:

- 1 In the administration client, open the **Labels** screen.
- 2 Select a label in the left panel, and click **Print**.



Figure 10-14: Print a test label



- 3 In the **Select a Printer** dialog box, select the relevant printer and click **Print**.
- 4 Repeat Step 3, three to five times. Make sure all the characters are clearly and exactly printed on the label.
- 5 If the position of the image on the label is not correct refer to [10.6.3.3 Adjust Zebra Printer Calibration](#) or [10.6.3.4 Adjust Cognitive Printer Calibration](#).

10.6.3.3 Adjust Zebra Printer Calibration



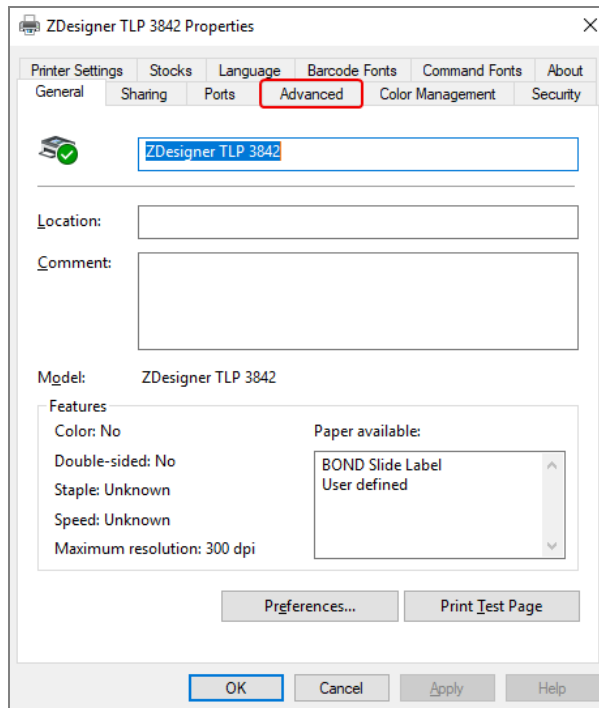
The following procedure applies to both types of Zebra printer: TLP 3842 or GX430t. There are some differences, which are described in the relevant settings.



For a BOND RX-ADVANCE installation, perform the following procedure on a BOND RX-ADVANCE terminal.

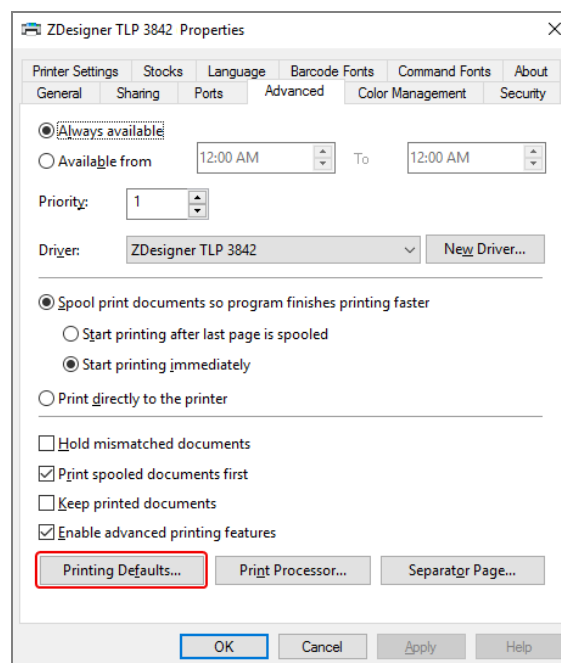
- 1 On the Windows taskbar, click the **Start** button and select **Devices and Printers**.
- 2 Right-click on the printer icon (for example **ZDesigner TLP 3842**) and select **Printer Properties**.
The system displays the Printer Properties dialog box as shown in **Figure 10-15**.

Figure 10-15: Printer Properties



- 3 Select the **Advanced** tab.

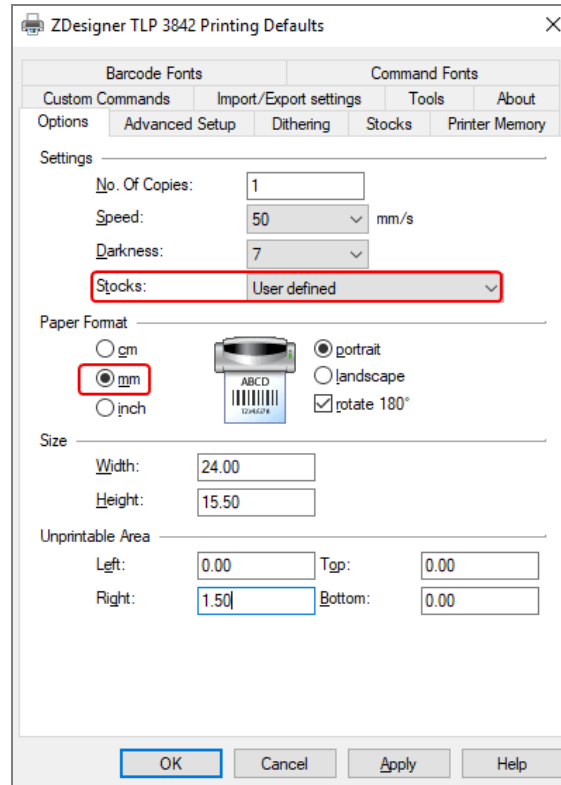
Figure 10-16: Printer Properties - Advanced tab



- 4 Click the **Printing Defaults...** button.

The system displays the Printing Defaults dialog box as shown in [Figure 10-17](#).

Figure 10-17: Printing Defaults

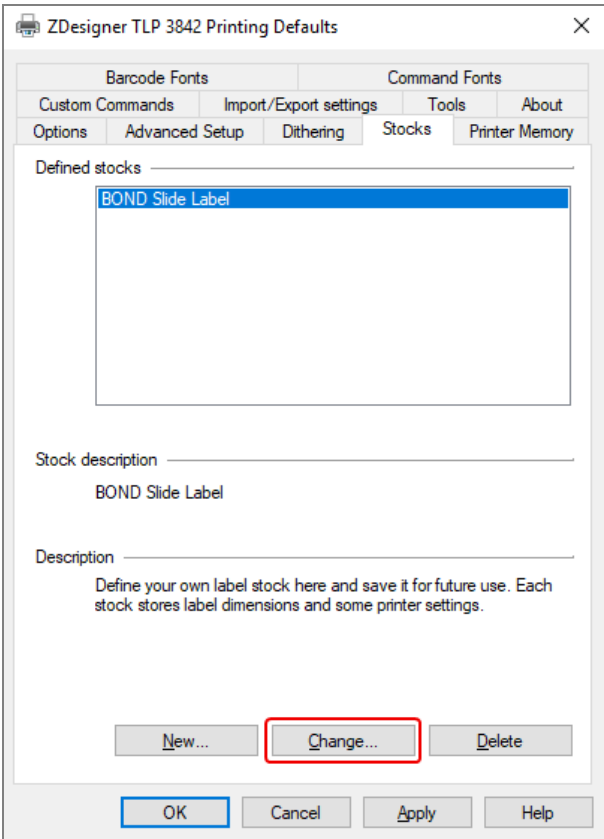


This document refers to the printer settings in millimeters. Therefore, set the Paper Format to mm.

- 5 Select "BOND Slide Label" from the Stocks drop-down list.

6 Select the **Stocks** tab.

Figure 10-18: Printing Defaults - Stocks tab



- 7 Click the **Change...** button.

The system displays the **Define Stock** window as shown in [Figure 10-19](#).

Before changing the settings, it is recommended to return the printer to the default settings, as shown in the table below, and print some test labels.

Setting	TLP 3842	GX430t
Label Width	24.00 mm	40.00 mm
Label Height	15.50 mm	15.00 mm
Unprintable Area - Left	0.00 mm	4.50 mm
Unprintable Area - Right	1.50 mm	0.00 mm

Figure 10-19: Define Stock dialog box

- If the left edge is clipped off, slightly decrease the value of **Right** under **Unprintable Area**, for example, from 1.50 mm to 1.00 mm.
- If the right edge is clipped off, slightly increase the value of **Right** under **Unprintable Area**, for example, from 1.50 mm to 2.00 mm.

- 8 Click **OK**.

- Repeat the label printing and adjustment procedure until the label is acceptable (no text is clipped off).



You might get an error message **Stock name already used by system form database** after clicking **OK**. In this case modify the **Name** in the **Define Stock** dialog box as shown in **Figure 10-20**, then click **OK**.

Figure 10-20: Rename Label Stock

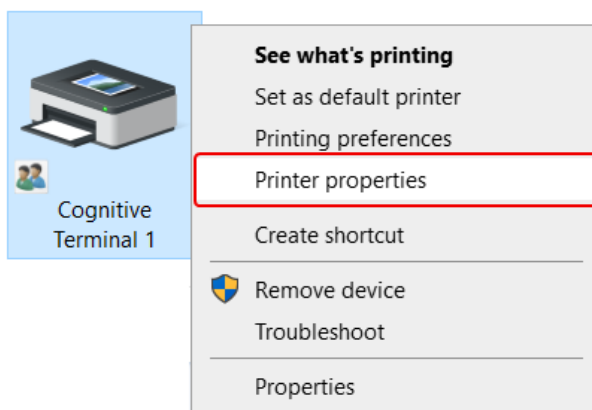


10.6.3.4 Adjust Cognitive Printer Calibration

For a BOND RX-ADVANCE installation, log in to the BOND RX-ADVANCE Controller as BONDDashboard. If the Dashboard is currently displayed, press **Alt+F4** to close it.

- On the Windows taskbar, click the **Start** button and select **Devices and Printers**.
- Right-click the printer icon (for example: **Cognitive Terminal 1**) and select **Printer Properties**.

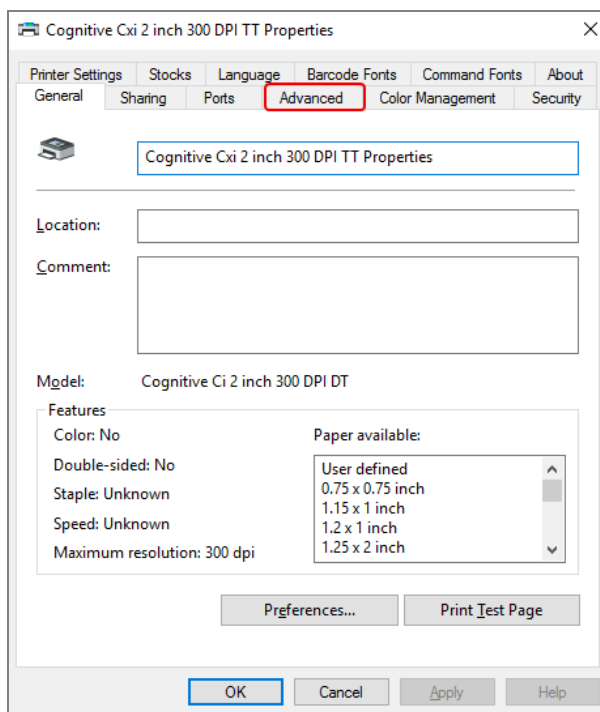
Figure 10-21: Select Printer Properties



Do not select **Printing Preferences**, the dialog boxes are similar, but the settings do not update correctly.

The system displays the **Cognitive Printer Properties** dialog box as shown in [Figure 10-22](#).

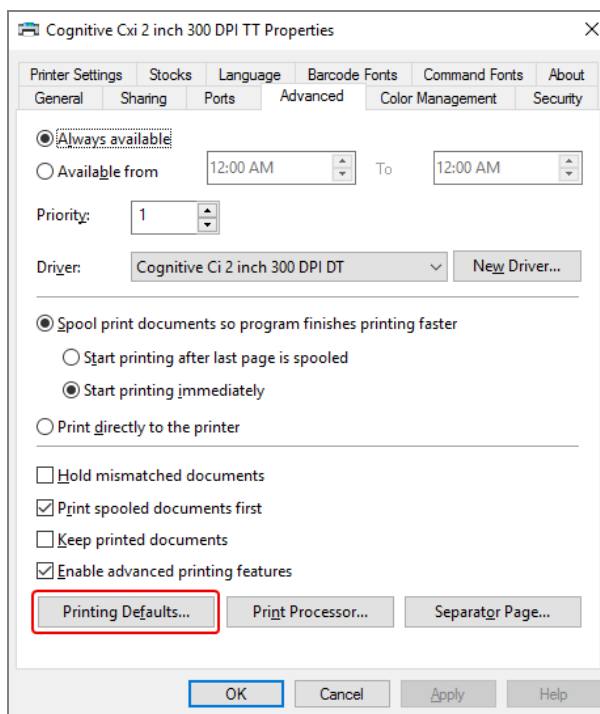
Figure 10-22: Cognitive Printer Properties



- 3 Select the **Advanced** tab.

The system displays the **Advanced** tab as shown in [Figure 10-23](#).

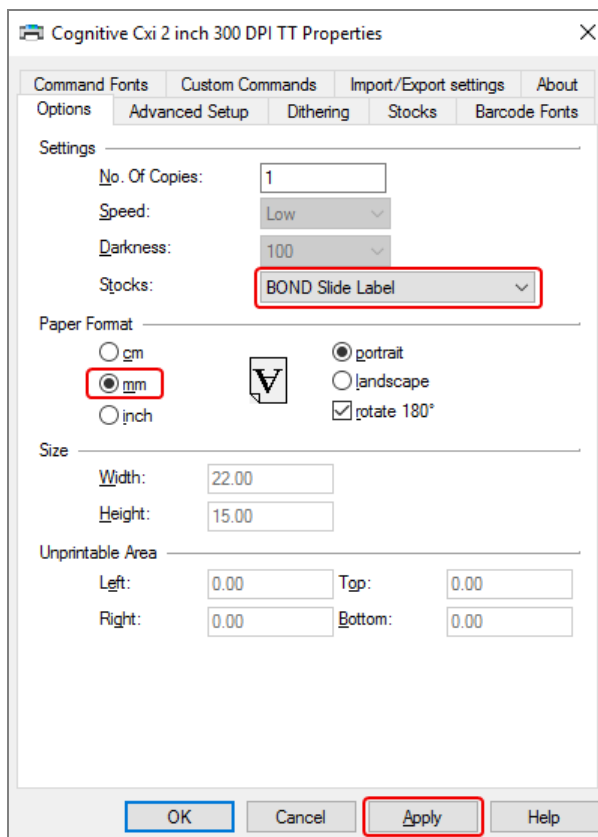
Figure 10-23: Advanced tab



- 4 Click the **Printing Defaults...** button.

The system displays the **Printing Defaults** dialog box as shown in **Figure 10-24**.

Figure 10-24: Printing Defaults dialog box

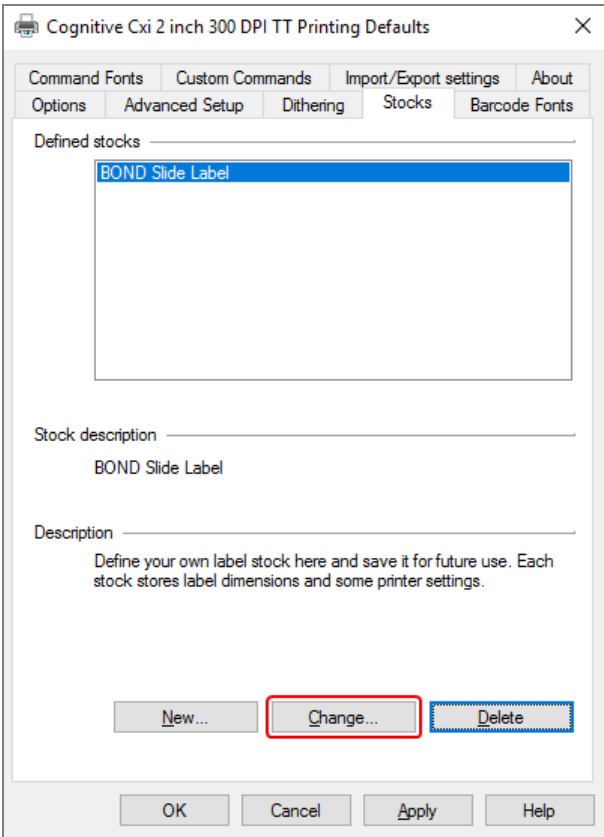


This document refers to the printer settings in millimeters. Therefore, set the Paper Format to mm.

- 5 Select "BOND Slide Label" from the Stocks drop-down list.

6 Select the **Stocks** tab.

Figure 10-25: Printing Defaults - Stocks tab



- 7 Click the **Change...** button.

The system displays the **Define Stock** dialog box as shown in [Figure 10-19](#).

Figure 10-26: Define Stock dialog box

The screenshot shows the 'Define Stock' dialog box with the following settings:

- Name:** BOND Slide Label
- Options:** Darkness: 300, Speed: Low
- Dimensions:** Label Width: 22.00 mm, Label Height: 15.00 mm
- Unprintable Area:** Left: 0.00, Right: 0.00, Top: 0.00, Bottom: 0.00

- If the left edge is clipped off, slightly decrease the value of **Right** under **Unprintable area**, for example, from 0.50 mm to 0.30 mm.
- If the right edge is clipped off, slightly increase the value of **Right** under **Unprintable area**, for example, from 0.50 mm to 0.70 mm.
- If the top or bottom edge is clipped off, refer to [10.6.3.5 Adjust Vertical Label Position on Cognitive Cxi Printer](#).

- 8 Click **OK**.



You might get an error message **Stock name already used by system form database** after clicking **OK**. In this case, modify the **Name** in the **Define Stock** dialog box as shown in [Figure 10-27](#), then click **OK**.

Figure 10-27: Rename Label Stock

The screenshot shows the 'Define Stock' dialog box with the following settings:

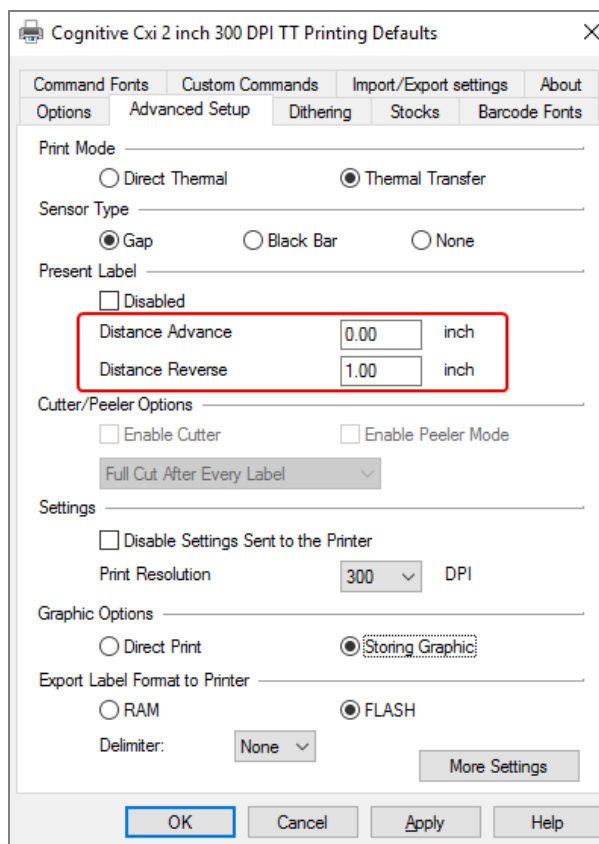
- Name:** BOND Slide Label 1
- Options:** Darkness: 300, Speed: Low

- 9 Print a label to check the result. Repeat the procedure until the label is acceptable (no text is clipped off).

10.6.3.5 Adjust Vertical Label Position on Cognitive Cxi Printer

If the position of the label is too high or low, select the **Advanced Setup** tab on the **Printing Defaults** dialog box as shown in [Figure 10-28](#).

Figure 10-28: Advanced Setup tab



- If the top edge is clipped off, slightly increase the value of **Distance Advance** under **Present Label**, for example, from 0.00 mm to 1.00 mm.
- If the bottom edge is clipped off, slightly increase the value of **Distance Reverse** under **Present Label**, for example, from 0.00 mm to 1.00 mm.
- Apply the adjustments to one setting only. If there is already a value in **Distance Advance**, and the bottom edge is clipped off, decrease the **Distance Advance** value rather than increasing the value of **Distance Reverse**. One value remains at zero and the other value controls the position.

- 1 Click **OK**.
- 2 Print a label to check the result. Repeat the procedure until the label is acceptable (no text is clipped off).

11 LIS Integration Package (on BOND RX Controller)

The optional BOND LIS integration package (LIS-ip) connects the BOND RX system to any compatible Laboratory Information System (LIS). The LIS-ip passes study and slide information from the LIS to the BOND RX system and the BOND RX system returns processing information via the LIS-ip to the LIS.

The LIS-ip is highly configurable and can work with many different LIS types and laboratory workflows. The LIS-ip can be configured to provide seamless integration between the LIS and the BOND RX system allowing automatic recognition of LIS slides, which eliminates the need for slide relabeling. See [11.8 Workflows](#) for a general overview of the workflows available.

Leica Biosystems arranges comprehensive site-specific training for each installation.

Refer to the following sections for BOND LIS-ip information:

- Terms relating to LIS-ip operation
Refer to [11.1 LIS Terminology](#)
- Details of additional software functions
Refer to [11.2 Additional Software Features](#)
- An overview of LIS connection and configuration
Refer to [11.3 LIS Connection and Initialization](#)
- A description of LIS error indication and recovery
Refer to [11.4 LIS Notifications](#)
A reference list of study and slides data
Refer to [11.5 Study and Slide Data Requirements](#)
- A description of slide status data the BOND LIS-ip is able to report to the LIS
Refer to [11.6 Sending Slide Data back to the LIS](#)
- A reference to slide label requirements
Refer to [11.7 Slide Labels](#)
- An overview of typical LIS implementations
Refer to [11.8 Workflows](#).

11.1 LIS Terminology

A number of new terms are required to describe LIS functionality and to differentiate between normal BOND RX system elements and LIS elements. These terms are described in the following list.

- LIS – Laboratory Information System; software that manages information related to a laboratory’s work.
- LIS-ip – the BOND LIS integration package, an optional add-on that enables the BOND RX system to work with an LIS.
- LIS slide – a slide created by the LIS and sent to the BOND RX system for processing.
- LIS study – a study created by the LIS and sent to the BOND RX system.
- Auto-ID slide label – a slide label that can be automatically recognized by the BOND RX system. These can be printed by the BOND RX system or the LIS, so long as a recognizable barcode format is used. See [11.3 LIS Connection and Initialization](#).
- Assisted-ID slide label – any slide label that cannot be automatically recognized on the BOND RX system.
- LIS slide label – a slide label from a printer connected to the LIS. An LIS slide label shows the LIS barcode and any other information configured for the label in the LIS.
- BOND-LIS slide label – a slide label for a slide created in the LIS but printed on a printer connected to the BOND RX system. A BOND-LIS label uses the BOND RX LIS slide label configuration, which can be edited using the BOND RX software.
- Accession number – a common LIS term for a number or other ID that identifies a particular study. Accession number is equivalent to the BOND RX system “study ID”.
- Study data – study details that form a “study” on the BOND RX system.
- LIS barcode – a barcode assigned by the LIS that uniquely identifies each LIS slide.

11.2 Additional Software Features

LIS-enabled BOND RX systems have additional software features not found in the standard version. BOND LIS-ip systems retain all the features and functions of standard BOND RX software.

See:

- [11.2.1 LIS Status Icon](#)
- [11.2.2 LIS Studies](#)
- [11.2.3 LIS Slides](#)
- [11.2.4 Public Marker Names](#)
- [11.2.5 Priority Slides](#)
- [11.2.6 LIS Slide Data Fields](#)
- [11.7 Slide Labels](#)

11.2.1 LIS Status Icon

Figure 11-1: LIS status icon at the top right of the BOND RX software screen



BOND RX software with the LIS-ip includes the LIS status icon at the far right of the standard function bar. This shows the following:

- LIS connection status (refer to [11.3 LIS Connection and Initialization](#))
- LIS error indication (refer to [11.4 LIS Notifications](#))

11.2.2 LIS Studies

LIS studies are studies that are created in the LIS and then sent to the BOND RX system. In contrast, BOND RX studies are studies created in the BOND RX system.

- LIS studies contain the same property fields as BOND RX studies but no information can be edited once a study has been sent to the BOND RX system.
- The BOND RX system automatically allocates a unique study number to every LIS study.
- The LIS accession number or study ID becomes the study ID within the BOND RX system.
- If this study ID is the same as that of an existing BOND RX study the new LIS study is rejected. You must change the study ID in the LIS.
- If the study ID and study name of a new LIS study are the same as those of an active LIS study, already listed on the **Slide setup** screen, the existing study is automatically used. The slides in the “new” study are added to those in the existing study. If the study IDs are the same but the study names different, the new study is rejected.
- If the study ID and study name of an LIS study are the same as those of an expired or deleted LIS study in the BOND RX system, either the existing study is resurrected or the new study rejected, depending on your setting in the administration client LIS screen (see [Duplicate Study ID \(on page 219\)](#)).
- Slides added to an LIS study using the BOND RX software are created as BOND RX slides.
- LIS studies have the same default preparation protocol and dispense volume as BOND RX studies, as set in the administration client (see [10.5.2 Study and Slide Settings](#)).

11.2.3 LIS Slides

LIS slides are slides that were created in the LIS and then sent to the BOND RX system. In contrast, BOND RX slides are slides created in the BOND RX system, either in a BOND RX study or an LIS study.

LIS slides can be identified in the slide list by their label color: LIS slides have a gray label.

Figure 11-2: LIS slide (left) and single-stain routine BOND RX slide (right)



The following points apply to LIS slides:

- Labels printed from the LIS typically include a barcode. Provided the barcode is in one of the six formats supported by the BOND RX system, and the BOND RX system has been configured to read that format, then the BOND RX system can identify the slide when loaded. Refer to [11.3 LIS Connection and Initialization](#).
- Labels printed from the BOND RX system for LIS slides use the BOND LIS slide label configuration. Refer to [10.3 Labels](#).
- LIS slides can include additional LIS-specific fields. Refer to [11.2.6 LIS Slide Data Fields](#).
- Slide properties originating from the LIS cannot be edited using the BOND RX software.
- When the BOND RX software is used to copy an LIS slide, the copy is created as a BOND RX slide with a BOND RX slide label configuration. All LIS-specific fields are removed and all fields become editable.



Sequential multiplex and parallel multiplex slides sent from an LIS can only be a dual sequential or dual parallel multiplex slide.

11.2.4 Public Marker Names

Public marker names (for primary antibodies and probes) provide the link between markers specified by an LIS and those registered on the BOND RX system. When an LIS specifies a marker for a test, the BOND RX system uses the reagent with the identical public marker name for that test. The BOND RX system will reject an LIS-specified test if there is no public name corresponding to the LIS marker name.

Public marker names are specified using the **Public name** field in the **Edit reagent properties** dialog (refer to [8.2 Reagent Setup Screen](#)). This field only becomes visible when the LIS-ip is installed.

Each public name must be unique. Public names can be swapped between BOND reagents at any time and when this occurs slides already created are not affected.

11.2.5 Priority Slides

The LIS can specify priority slides that require urgent processing. Any study that includes a priority slide appears with a red bar on the **Slide setup** screen.

Figure 11-3: A study with priority slides highlighted red on the **Slide setup** screen

Study ID	Study name	Researcher name	Slides
LS0012 - 45216	Shady, Albert	Joseph	1
20130416-ISHRefine	Benjamin Hightower	Kevin Pannell	10
20130416-IHC	Fannie Hurley	Arthur Josey	10



Currently, a priority LIS study is initially added to the bottom of the list. The study only displays at the top of the list in subsequent sessions of the research client.

The priority slides are marked with a red “P”.

Figure 11-4: A priority LIS slide as it appears in the **Slide setup** screen



11.2.6 LIS Slide Data Fields

In addition to the standard slide properties, BOND LIS-ip has seven configurable data fields that can be set up to display selected information from the LIS. Basic connectivity is set up by the Leica Biosystems service representative during installation, however once this is in place users can choose to display the fields or not, and can set the name of each field – see [LIS Slide Data Fields \(on page 219\)](#).

The fields are displayed on a special **LIS** tab in the **Slide properties** dialog, and can also be printed on slide labels (see [10.3 Labels](#)). They are for reporting purposes only and have no effect on slide processing.

11.3 LIS Connection and Initialization

Each BOND LIS-ip module must be installed by an authorized Leica Biosystems representative who will customize the operation in accordance with individual laboratory requirements.

The BOND RX system can be configured to read any of the following barcode formats:

2D Barcodes	
QR	
Aztec	
Data Matrix	

When the LIS module is installed, an LIS icon appears at the top right of the BOND RX software screen to indicate connection status (Figure 11-5).

Figure 11-5: LIS not connected (left) and connected (right)



11.4 LIS Notifications

The BOND RX software indicates LIS connection or data errors by displaying the LIS status icon at the top right of the BOND RX software screen (refer to 11.2.1 LIS Status Icon). If there are any outstanding LIS notifications then a counter of the number of outstanding notifications is displayed. When a new notification event occurs the counter briefly flashes.

Figure 11-6: LIS status icon



To find notification details, right-click the status icon and select **Show LIS report** to open the **LIS service events** dialog. The dialog displays errors and any slides that were not successfully transferred. The reason for the error is also listed. Typical LIS errors include missing data, data conflicts (e.g. the same accession number used for different studies), or instances where the public marker is not registered on the BOND RX system (refer to [11.2.4 Public Marker Names](#)).

Figure 11-7: LIS service events dialog

ID	Date	Event N°	Details	Message	Acknowledge
2...	08-Feb-17 4:18...	7006	Study ID: LS0012-45210 Patient ID: PID120 Researcher ID: Dr Jones Marker ID: GFAP	Marker does not exist	Acknowledge
2...	08-Feb-17 4:24...	7007	Study ID: LS0012-45210 Patient ID: PID120 Researcher ID: Dr Jones Marker ID: GFAP Marker2 ID: GFAP Tissue type: test Message ID: 002.1 Barcode: 88620	Cannot map tissue type	Acknowledge
2...	08-Feb-17 5:37...	7006	Study ID: LS0012-45210 Patient ID: PID120 Researcher ID: Dr Jones Marker ID: GFAP	Marker does not exist	Acknowledge
2...	08-Feb-17 5:38...	7006	Study ID: LS0012-45210 Patient ID: PID120 Researcher ID: Dr Jones Marker ID: GFAP	Marker does not exist	Acknowledge

Close

Depending on the LIS configuration, it may be possible to correct the errors and resubmit the study or slide. Where the LIS is unable to re-send the information, the study or slides can be created directly using the BOND RX software.

When you have read each error message click the associated **Acknowledge** button to remove the notification from the dialog.

When all error messages are cleared from the dialog, the notification counter disappears from the screen.



If required, you can still view the messages in the LIS service log by first clicking the Leica Biosystems logo at the top right of the administration client screen, to display the **About BOND RX** dialog. Then click **Service log** and select ***LIS*** from the drop-down **Serial No.** list. Optionally set a time span and then click **Generate** to generate the LIS service log.

11.5 Study and Slide Data Requirements

The data required by the BOND RX system from the LIS to import studies and slides is provided in the sections below (see [11.5.1 Study Data](#) and [11.5.2 Slide Data](#)).



Data in LIS studies and slides cannot be changed using the BOND RX software, except for slide comments.

11.5.1 Study Data

11.5.1.1 Mandatory Fields

BOND RX Field Name	Description	Common LIS Terms
Study ID	A number or name identifying the study	Accession number Order number

11.5.1.2 Optional Fields

BOND RX Field Name	Description	Common LIS Terms
Study name	The name of the study	Study name Lab assigned ID (labAssId)
Researcher	The researcher running the study	Researcher name and/or ID

11.5.2 Slide Data

11.5.2.1 Mandatory Fields

BOND RX Field Name	Description	Common LIS Terms	Comments
Marker	Primary antibody (IHC) or Probe (ISH)	Primary antibody (IHC) Probe (ISH) Marker (either) Stain	<p>The public name provides the link between markers specified by an LIS and those registered on the BOND RX system. A public name must be specified for each marker that will be specified in the LIS. See 11.2.4 Public Marker Names.</p> <p>Each marker has default staining and pretreatment protocols, which can be changed using the BOND RX software if required.</p> <p>Note: Sequential multiplex and parallel multiplex slides sent from an LIS can have a maximum of two stains</p>

11.5.2.2 Optional Fields

BOND RX Field Name	Description	Common LIS Terms	Comments
[LIS barcode] Note: The barcode is not visible to the user on the BOND RX system	A unique ID barcode given to each LIS slide (IDs of deleted slides cannot be reused)	Barcode	A complete ID barcode must be supplied for the BOND RX system to recognize a slide. This is required when using the LIS workflow 1 (see 11.8 Workflows).
Tissue type	Test or control tissue (positive or negative)	Test type	If this information is not supplied by the LIS, it defaults to "Test". See 6.2.1 Control Tissue .
Comments	Any comment or instruction relating to the slide	Comment	If an update to an LIS slide is sent by the LIS, then any new slide comments will be appended to existing slide comments.

11.6 Sending Slide Data back to the LIS

The BOND LIS-ip is able to report slide status to the LIS. BOND LIS-ip can report the following information:

- Slide created – the specified slide has been created within the BOND RX software
- Slide printed – a label has been printed for the specified slide
- Slide in progress – the specified slide is being processed
- Slide processed – the specified slide has completed processing (with or without errors)
- Slide deleted – the specified slide has been deleted from the BOND RX system.

11.7 Slide Labels

Each physical slide requires an identification label so that it can be matched to the correct study and test information. In the most convenient workflow LIS slides have labels printed by the LIS ("LIS slide labels") and these labels are recognized by the BOND RX system. However, this is only possible if:

- 1 the LIS provides a unique barcode for each slide to the BOND RX system, and
- 2 the LIS printer uses one of the barcode formats supported by the BOND RX system.

If your LIS does not meet these requirements, then the BOND RX system can create its own labels for LIS slides – "BOND-LIS slide labels". In this case you can optionally set the BOND RX system so that it will only process LIS slides if they have had labels printed by the BOND RX system. This is set in the administration client **LIS** screen – see [10.2 LIS](#).

Alternatively, labels from a third party labeler or handwritten, can be used. These labels need to be manually identified on the BOND RX system before processing (see [5.1.5.2 On-Board Manual Slide Identification](#)).

11.8 Workflows

While each LIS-ip implementation is highly customized, it is still helpful to provide some general descriptions of BOND LIS-ip workflows based on the major LIS-ip options. The following table shows four workflows. Other workflows are also possible. Comprehensive site-specific training is provided for each installation.

Workflow	Data from LIS	Data entered on the BOND RX system	Labels printed on	Identification
1	Study and slide data (with LIS barcode)	None	LIS	Automatic
2	Study and slide data	None	BOND RX system	Automatic
3		Additional slide	BOND RX system	Automatic
4		None	External	Assisted

Workflow 1 is the most convenient as it provides seamless integration between the LIS and the BOND RX system. The BOND RX system automatically recognizes LIS slides and processing can begin immediately without having to relabel the slides or enter additional information.

12 Cleaning and Maintenance (BOND RX and BOND RX^m)



WARNING: Always switch the processing module off when performing cleaning or maintenance tasks (except when running an aspirating probe clean or bulk fluid robot clean).



WARNING: Some of the reagents used in immunohistochemistry and in situ hybridization are hazardous. Ensure you have received adequate training for this procedure before continuing:

- 1 Wear latex or nitrile gloves, safety glasses, and other suitable protective clothing when handling reagents or cleaning the processing module.
- 2 Handle and dispose of reagents and condensate in accordance with all procedures and government regulations that apply at the laboratory facility.



WARNING: The processing modules have heaters and heated surfaces that can be ignition dangers if flammable materials are placed in close proximity:

Do not place flammable materials on or near the heaters.

Do not place flammable materials on any hot surfaces on the processing module.

Ensure all bulk container caps are properly sealed after refilling or emptying.



WARNING: Avoid contact with slide staining assemblies and their surrounds. These may be very hot and cause severe burns. Allow twenty minutes after the cessation of operation for the slide staining assemblies and their surrounds to cool.



CAUTION: Clean all removable components by hand only. To avoid damage, do not wash any component in an automatic dishwashing machine. Do not clean any part with solvents, harsh or abrasive cleaning fluids, or harsh or abrasive cloths.

This chapter gives procedures for cleaning and maintenance. In the research client there is a maintenance screen for each processing module in the system. Click a processing module tab at the left-hand side of the main window to display its **System status** screen, and then click the **Maintenance** tab.

For more information, see [5.3 Maintenance Screen](#). Whenever you use the BOND RX system, look out for leaks or worn or damaged parts. If there are instructions in this chapter to repair or replace the worn or faulty part, follow these. Otherwise, contact customer support.

Preventative Maintenance

In addition to the regular maintenance tasks listed in this chapter (carried out by users), BOND RX and BOND RX^m processing modules should be serviced regularly by a Leica Biosystems service representative.

For BOND RX and BOND RX^m, the BOND RX software notifies you to organize a preventative maintenance service for each processing module once a year or every 15600 slides (whichever comes first).



The count is reset with the **Maintenance complete** button on the **Processing modules** tab in the administration client ([10.6.1 Processing Modules](#)).

This chapter has the following sections:

- [12.1 Cleaning and Maintenance Schedule](#)
- [12.2 Bulk Containers](#)
- [12.3 Covertiles](#)
- [12.4 Slide Staining Assembly](#)
- [12.5 Restart Processing Module](#)
- [12.6 Aspirating Probe](#)
- [12.7 Wash Block and Mixing Station](#)
- [12.8 Covers, Doors and Lid](#)
- [12.9 ID Imager](#)
- [12.10 Drip Trays](#)
- [12.11 Slide Trays](#)
- [12.12 Bulk Fluid Robot Probes \(BOND RX only\)](#)
- [12.13 Syringes](#)
- [12.14 Power Supply Fuses](#)

12.1 Cleaning and Maintenance Schedule

Use the schedule below if you stain up to about 300 slides a week per processing module. If you process more than this contact customer support for a customized schedule.

Task	Section
Daily – Start of Day	
Check bulk waste containers are no more than half full*	12.2
Check bulk reagent containers are filled with adequate reagent for the days staining*	12.2
Daily – End of day	
Clean Covertiles	12.3
Weekly	
Clean slide staining assemblies*	12.4
Check Covertile clamps	12.4
Restart processing modules	12.5
Wipe the main robot aspirating probe	12.6
Check wash blocks and mixing station– clean or replace if necessary	12.7
Clean covers, doors (where fitted) and lid	12.8
Clean ID imager	12.9
Clean handheld barcode scanner	13.1
Monthly	
Clean all drip trays*	12.10
Replace mixing station	12.7
Clean bulk reagent containers	12.2
Clean bulk waste containers	12.2
Clean slide trays	12.11
Clean bulk fluid robot probes (BOND RX)	12.12
Clean slide labeler	13.2
Check syringes	12.13
When prompted	
Clean main robot aspirating probe	12.6.1
Replace syringes	12.13

* Carry out these tasks more frequently than scheduled if required.

12.1.1 Cleaning and Maintenance Checklists

On the next page the maintenance schedule is reproduced in a table designed to print and use as a checklist. Areas are provided to record the lot numbers for BOND Wash, ER1, ER2 and Dewax solution. Check or initial the remaining cells as the tasks are completed.

Cleaning and Maintenance Schedule

	Mon	Tues	Wed	Thurs	Fri	Sat	Sun
DAILY							
Check bulk reagent containers	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
BOND wash lot number							
ER1 lot number							
ER2 lot number							
Dewax solution lot number							
Check waste containers	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Clean Covertiles	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
WEEKLY		For BOND RX™:					
Clean slide staining assemblies*	<input type="checkbox"/>	<ul style="list-style-type: none"> Lift the ends of the bulk containers in position to estimate the volume – it is not necessary to remove containers from the processing module. 					
Check Covertile clamps	<input type="checkbox"/>	*Clean more frequently than scheduled if required					
Restart PMs	<input type="checkbox"/>						
Wipe the aspirating probe	<input type="checkbox"/>						
Check wash block & mixing station	<input type="checkbox"/>						
Clean covers, doors (where fitted) and lid	<input type="checkbox"/>						
Clean ID imager	<input type="checkbox"/>						
Clean handheld scanner	<input type="checkbox"/>						
MONTHLY							
Clean drip trays*	<input type="checkbox"/>						
Replace mixing station	<input type="checkbox"/>						
Clean bulk reagent containers	<input type="checkbox"/>						
Clean bulk waste containers	<input type="checkbox"/>						
Clean slide trays	<input type="checkbox"/>						
Clean bulk fluid robot probes (BOND RX only)	<input type="checkbox"/>						
Clean slide labeler	<input type="checkbox"/>						
Check syringes	<input type="checkbox"/>						
WHEN PROMPTED		For week starting _____					
Clean aspirating probe	<input type="checkbox"/>	to _____					
Replace syringes	<input type="checkbox"/>	For month of _____					

12.2 Bulk Containers



WARNING: Some of the reagents used in immunohistochemistry and in situ hybridization are hazardous. Ensure you have received adequate training for this procedure before continuing:

- 1 Wear latex or nitrile gloves, safety glasses, and other suitable protective clothing when handling reagents or cleaning the processing module.
- 2 Handle and dispose of reagents and condensate in accordance with all relevant procedures and government regulations that apply at the laboratory facility.



WARNING: Some of the reagents used on BOND Processing Modules are flammable:

Do not place a flame or ignition source near the processing modules.

Ensure all bulk container caps are properly sealed after refilling or emptying.

Check bulk container levels daily (at least), and clean the bulk containers every month. See details:

- [12.2.1 Checking Container Levels](#)
- [12.2.2 Replenishing or Emptying Bulk Containers](#)
- [12.2.3 Cleaning Bulk Containers](#)
- [12.2.4 External Waste Container \(BOND RX^m only\)](#)

12.2.1 Checking Container Levels

Check bulk container levels at the start of each day. Also check before starting overnight or extended runs. High-turnover laboratories may need to schedule two bulk container checks daily.

On BOND RX and BOND RX^m Processing Modules (and BOND RX^m external waste containers), liquid levels are visible through the container walls.

Icons on the **System status** screen give an indication of bulk container levels for BOND RX, and are used for notifications of high waste or low reagent levels on the BOND RX^m. Use the icons only to confirm levels and/or for viewing notifications – they do not replace daily physical checks.



BOND RX Processing Modules are fitted with a bulk container lighting system (see [Bulk Container Lighting System \(BOND RX\) \(on page 49\)](#)).

Fill or empty containers under the following conditions:

- Empty waste containers that are more than half full
- Refill reagent containers to ensure there is adequate reagent.

See [12.2.2 Replenishing or Emptying Bulk Containers](#).



WARNING: Check bulk container levels and empty or fill, as appropriate, at the start of each day (more frequently if required – see instructions above). Failure to do so can result in runs being paused, which can compromise staining.

12.2.2 Replenishing or Emptying Bulk Containers

When you check bulk container levels, empty waste containers that are more than half full, and fill reagent containers to ensure there is adequate reagent. Always wipe any spills that occur when filling or emptying bulk containers. Clean the outside of the containers and caps before returning to the processing module.

See separate emptying and refilling instructions below. The [12.2.2.5 During Runs](#) section has instructions if you need to empty or fill a container during a run.

- [12.2.2.1 Refill Bulk Reagent – BOND RX](#)
- [12.2.2.2 Empty Hazardous Waste – BOND RX](#)
- [12.2.2.3 Empty Standard Waste – BOND RX](#)
- [12.2.2.4 Empty Hazardous Waste or Refill Bulk Reagent – BOND RX^m](#)
- [12.2.2.5 During Runs](#)

See [12.2.4 External Waste Container \(BOND RX^m only\)](#) for instructions to empty the BOND RX^m external container.



WARNING: Always return refilled or emptied containers to the same locations on the processing module. Failure to do so can contaminate reagents and compromise staining.



WARNING: Do not change the type of reagent in bulk reagent containers. Doing so could lead to contamination and compromise staining.



CAUTION: Do not force bulk containers back into position, as this can damage the container and liquid sensor.

12.2.2.1 Refill Bulk Reagent – BOND RX

BOND RX bulk reagent containers can be filled whilst in the processing module. There is no need to remove them from the bulk container cavity.

- 1 Unscrew the bulk reagent container cap and fill the container.
- 2 When container is full, replace the cap and tighten.



WARNING: If you use a funnel when adding reagent to containers on BOND RX Processing Modules, ensure that the funnel is clean. Failure to do so can contaminate reagents and compromise staining.

12.2.2.2 Empty Hazardous Waste – BOND RX

- 1 Ensure the processing module is not in operation. (However, if there is a notification that the waste container is full during a run, follow these instructions to empty the container – see also [12.2.2.5 During Runs.](#))
- 2 Pull the container out of the bulk containers cavity.
- 3 Open the cap and dispose of the waste in accordance with approved procedures at your facility.
- 4 Replace the cap and tighten.
- 5 Return the container to the processing module. Push in gently until you feel the container connector align with the connector at the back of the cabinet. Then push the container firmly until the connector fully engages, to ensure a leak-tight connection.

12.2.2.3 Empty Standard Waste – BOND RX

As there are two standard waste containers you can remove a full container (with the container icon showing full on the **System status** screen) at any time, including during processing (see [5.1.3.7 Bulk Container Status](#)). However, never remove both bulk waste containers while the processing module is in operation, and if a container is not showing as full on the **System status** screen we recommend to wait until processing is finished before removing it. When it is safe to remove a bulk waste container, follow instructions as for emptying hazardous waste from step (2) above.

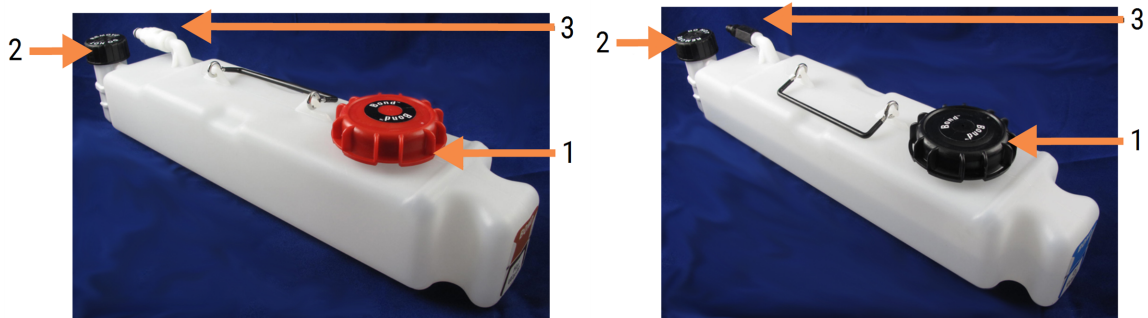
Figure 12-1: Returning the waste container to its position



12.2.2.4 Empty Hazardous Waste or Refill Bulk Reagent – BOND RX^m

- 1 Ensure the processing module is not in operation. (However, if there is a notification that the waste container is full during a run, follow these instructions to empty the container – see also [12.2.2.5 During Runs.](#))
- 2 Pull the container out of the bulk containers cavity.

Figure 12-2: BOND RX^m hazardous waste container (left) and bulk reagent container (right)



Legend

- 1 Fill/empty cap (blue cap on later hazardous waste containers)
- 2 Liquid level sensor cap
- 3 Connector

- 3 Fill or empty the container:

- For waste, open the fill/empty cap (item 1 in [Figure 12-2](#)) and dispose of the waste in accordance with approved procedures at your facility.
- For bulk reagent, place the container on a level surface, open the fill/empty cap (item 1 in [Figure 12-2](#)), and fill to just below the bottom of the neck that the cap screws onto.




CAUTION: Do not remove the liquid level sensor cap from a bulk container as it can be damaged. Empty and refill bulk containers through the fill/empty cap only.

- 4 Replace the cap and tighten.
- 5 Return the container to the processing module. Push in gently until you feel the container connector align with the connector at the back of the cabinet. Then push the container firmly until the connector fully engages, to ensure a leak-tight connection.

12.2.2.5 During Runs



If daily bulk container checks are made (with additional checks before overnight and extended runs, and regular additional checks for high turnover laboratories) waste containers should never fill up and reagent containers never run out during processing. However, if either of these occur during a run, you must empty or fill the containers concerned. Read the instructions below to be sure of the correct procedure.

Waste Container Full – BOND RX^m

If a waste container becomes nearly full during a run, an Information symbol  will display on the relevant container icon on the **System status** screen.


Act immediately to empty the container. Observe all standard safety precautions and waste disposal procedures at your facility. By acting quickly you may avoid having the run pause, or reduce the time it is paused. Pausing a run can compromise staining.

If a run pauses while you are emptying a container, or you continue to operate until the processing module is

automatically paused, an alarm  (flashing) or warning symbol  appears on the container icon. Return the emptied container as soon as possible, taking note of the instructions and precautions mentioned above.

Generate a Run Events Report to see the effects the pause had on the run.

Reagent Container Empty – BOND RX^m

If a bulk reagent container becomes nearly empty, a notification symbol  will display on the relevant container icon on the **System status** screen.

- 1 Open the **Protocol status** screen and view the current and upcoming steps for each run on the processing module.
- 2 If any runs are currently using the bulk reagent that is low, or will use it soon, wait for the steps that use the reagent to finish.
- 3 Once steps that use the bulk reagent have finished, remove the container, refill, and replace as quickly as possible (while observing all standard safety precautions).

To save time, you may not need to fill the container up to its usual, maximum level



WARNING: If a BOND RX^m bulk container needs filling during processing always check the **Protocol status** screen and confirm that the container is not being used, or is not about to be used. Failure to do so may compromise slides being processed. Return the container immediately after filling.

12.2.3 Cleaning Bulk Containers

The following cleaning procedures should be completed monthly.

12.2.3.1 ER1, ER2, BOND Wash and Deionized Water Containers

- 1 Empty the ER1, ER2, BOND Wash and the deionized water bulk reagent containers.
- 2 Wash containers with an industrial strength detergent, then rinse thoroughly with deionized water.
- 3 Allow the containers to dry before refilling with fresh reagent and returning to the processing module.

12.2.3.2 Dewax and Alcohol Containers

- 1 Empty the dewax and alcohol bulk reagent containers. Dispose of the dewax and alcohol in the bulk reagent containers in accordance with approved procedures at your facility.
- 2 Pour a small volume of fresh reagent into each container and move the liquid around the container walls to remove any contaminants. Empty the container when completed. Dispose of the waste in accordance with approved procedures at your facility.



Never put water or detergents in the alcohol or dewax containers.

- 3 Refill the bulk container with fresh reagent and return to the processing module.

12.2.3.3 Bulk Waste Containers

- 1 Empty all waste from containers. Dispose of the waste in accordance with approved procedures at your facility.
- 2 Clean waste containers using a 0.5% bleach solution (w/v) or industrial strength detergent and rinse thoroughly with deionized water.
- 3 Return waste containers to the processing module.

12.2.4 External Waste Container (BOND RX^m only)

Empty the BOND RX^m 9L external standard waste container at the start of each day and check the level before overnight or extended runs. Empty when half full or over; use the white horizontal line on the container label as a guide to the half-full level - see [Figure 12-3](#).

Figure 12-3: BOND RX^m 9L external standard waste container



Legend

- 1 Fill/empty cap
- 2 Half-full level

Clean the container monthly, as for other bulk containers (see [12.2.3 Cleaning Bulk Containers](#)).

- 1 Ensure that the processing module is not in operation. (However, if there is a notification that the waste container is full during a run, follow these instructions to empty the container – see also [12.2.2.5 During Runs](#).)

- 2 The container has connectors like those in [Figure 12-4](#) (note that some sensor connectors are black, not silver as shown):

Figure 12-4: External waste container connections



Legend

- | | |
|---|-------------------------------|
| 1 | Liquid level sensor connector |
| 2 | Fluid connector |

- a Use your thumb to raise the red latch on the sensor connector (1) and pull the connector away from the cap.
 - b Press the metal button on the fluid connector (2) and pull the connector away from the cap.
- 3 Remove the fill/empty cap to empty the container. Do not remove the cap with connectors. Dispose of the waste in accordance with approved procedures at your facility.
 - 4 Replace the fill/empty cap and tighten firmly, and return to the processing module.
 - 5 Press the fluid connector back onto the cap connection until it clicks into place.
 - 6 Reconnect the sensor connector. Push the connector down to the base of the cap connection.



WARNING: When full, the external waste container is heavy.

Use correct lifting techniques when emptying the external waste container.



CAUTION: Always disconnect the sensor and fluid connectors before emptying a container, to avoid damage.

12.3 Covertiles

Clean Covertiles after each use (the Leica Biosystems Covertile Cleaning Rack can be used for this). Covertiles can be reused up to 25 times provided they are not damaged or heavily discolored, and provided they are cleaned properly. Discard Covertiles if damaged or if staining quality deteriorates.

12.3.1 Remove DAB Residue (Optional)

- 1 Soak for a minimum of 30 minutes in a fresh solution of 0.5% W/V sodium hypochlorite in DI water.
- 2 Remove, and dip in fresh DI water 10 times.
- 3 Complete a standard clean (see below).

12.3.2 Standard Cleaning (Mandatory)

- 1 Soak for a minimum of 10 minutes in 100% IMS (industrial methylated spirits), ethanol, or reagent-grade alcohol.
- 2 Agitate for 30 seconds and remove.
- 3 Dry:
 - wipe dry with lint-free cloth, or;
 - air dry.
- 4 Carefully inspect Covertiles for chips, cracks or warping. Discard if damaged in any way.

12.4 Slide Staining Assembly



WARNING: The processing modules have heaters and heated surfaces that can be ignition dangers if flammable materials are placed in close proximity:

- Do not place flammable materials on or near heaters.
- Do not place flammable materials on any hot surfaces on the processing module.
- Ensure all bulk container caps are properly sealed after refilling or emptying.



WARNING: Avoid contact with slide staining assemblies and their surrounds. These may be very hot and cause severe burns. Allow twenty minutes after the cessation of operation for the slide staining assemblies and their surrounds to cool.



CAUTION: Clean specified components by hand only. To avoid damage, do not wash any component in an automatic dishwashing machine. Do not clean any parts with solvents, harsh or abrasive cleaning fluids, or harsh or abrasive cloths.



CAUTION: Ensure the bulk fluid robots (BOND RX) are in the home position at the rear of the processing module, and not positioned along the slide staining assemblies before cleaning or removing the top plate.



CAUTION: Do not use Q-tips or other cotton-tipped applicators to clean inside the wash block holes or the slide staining assembly wicking posts, as the cotton tip can come off and cause a blockage.

Standard Cleaning

Clean the slide staining assemblies weekly, or more frequently if there is buildup visible.

Use a lint-free cloth moistened with 70% alcohol (as little as possible). For difficult-to-remove precipitate use BOND Wash Solution (as little as possible) then rinse with DI water.

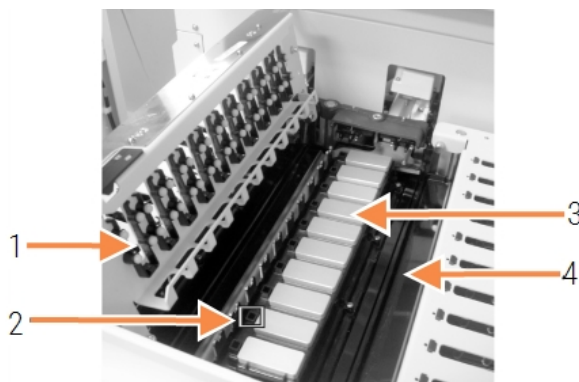
For BOND RX wipe the bulk fluid robot guide rail (Item 3 in [Figure 12-6](#)).

Swing open the top plate (see [Removing a Top Plate \(on page 277\)](#)) and clean:

- Heater pads
- Drainage ports and wicking posts
- The areas between the heater pads
- The drip tray surrounding the pads

Always check that the drainage ports (including the small wicking posts on the rims of the ports) are clear of foreign material and have no scratches or other damage. Contact customer support if there is damage to these or any other components of the slide staining assemblies.

Figure 12-5: Slide staining assembly with top plate open



Legend

- | | |
|---|---------------------------------|
| 1 | Covertile clamps |
| 2 | Drainage port and wicking posts |
| 3 | Heater pads |
| 4 | Drip tray |

While the top plate is open inspect the Covertile clamps on the underside of the plate and ensure the spring feet move freely. If the clamp springs do not spring back when pressed contact customer support for replacement.

Removing a Top Plate

- 1 Ensure the processing module is idle and turn the power off and there is no slide tray loaded.
- 2 Open the top plate by pushing down on the top plate and twisting the blue twist fasteners at either end (items 1 in [Figure 12-6](#) and [Figure 12-7](#)) a quarter turn anti-clockwise. Swing the top plate back on its hinges (when facing the processing module the right side of the top plate will lift open).

Figure 12-6: BOND RX top plate



Legend

-
- 1 Twist fasteners
 - 2 Pivot hinges
 - 3 Bulk Fluid Robot Guide Rail

Figure 12-7: BOND RX^m top plate

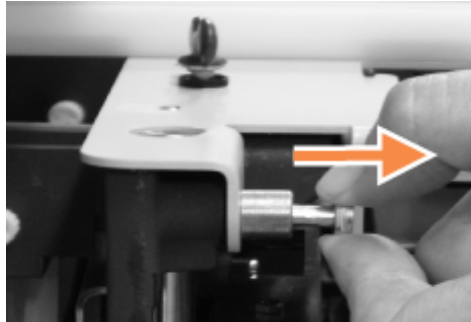


Legend

-
- 1 Twist fasteners
 - 2 Pivot hinges

- 3 To fully remove the top plate (not required for routine cleaning), pull the spring-loaded pivot fasteners at each end of the plate (items 2 in [Figure 12-6](#) and [Figure 12-7](#)), then lift the plate away from the slide staining assembly.

Figure 12-8: Releasing the top plate pivot fastener



Replacing a Top Plate



The BOND RX slide staining assembly top plates are numbered, always place the correct top plate on the correct slide staining assembly (when facing the processing module the slide staining assembly on the left is number one).

- 1 Locate the pivot points in the slide staining assembly. Hold the top plate in the open position, and place one of the pivot fasteners into the pivot point of the slide staining assembly.
- 2 Pull the other pivot fastener and place the end of the plate in position, and then release the fastener.
- 3 Close the top plate, checking that holes at each end of the plate correctly engage the locating pins.
- 4 Hold down the top plate and turn the twist fasteners clockwise. They should clamp firmly with a quarter turn clockwise.

12.4.1 Manually Unlocking Slide Staining Assemblies

Each slide staining assembly can be unlocked manually, for example to remove slides in a power failure.



WARNING: The slide staining assemblies contain moving parts that can cause serious injury. Before attempting to manually unlock the slide staining assemblies: turn the processing module power switch off, turn the mains power off, and disconnect the mains power supply plug at the wall.

- [12.4.1.1 BOND RX](#)
- [12.4.1.2 BOND RX^m](#)

12.4.1.1 BOND RX



WARNING: The syringe pump module (BOND RX) is heavy and can fall forward when released. Only operators who have been warned of the potential hazards and have received adequate training should carry out this procedure.

To manually unlock a slide staining assembly on the BOND RX:

- 1 Turn off the mains power and remove the power cable.
- 2 Unscrew the four hex screws attaching the syringe module cover using the 3 mm hex key provided. Remove the cover, for better access to the release pins and module handle.
- 3 Locate the two release pins next to syringe pumps one and four.

Figure 12-9: Location of release pins with unit open for access



- 4 Pull the two pins forwards towards you until they click and lower the module. Be careful not to pull or pinch any of the fluidics tubing on the syringe heads as the module moves forwards.
- 5 The syringe pump module will open enough to allow access to the slide staining assemblies.

- 6 Locate the manual release knob beneath the slide staining assembly.

Figure 12-10: Manual release knob



- 7 Turn the knob in the direction shown in [Figure 12-10](#). As you do the Covertiles will move over the slides and the whole assembly and tray move up.
- 8 Continue turning the release knob until you feel resistance. At this point it should be possible to remove the slide tray from the assembly.
- 9 Store your slides according to procedures at your facility.
- 10 Gently push the syringe pump module back into position, be careful not to pull or pinch any of the fluidics tubing on the syringe heads.
- 11 Ensure the two pins on either side of the module click back into the locked position.



CAUTION: Ensure the syringe module (BOND RX) is fully closed before starting a run or initializing the processing module. Failure to do so can result in damage to the syringes during operation.

- 12 Refit the syringe module cover and secure using the four hex screws.

Protocol status should be checked (see [5.2 Protocol Status Screen](#)) before powering on the processing module.

When the processing module is powered on, it will initialize, detect the state of the assemblies and take any actions necessary to ready them for use.

After initialization the slide staining assembly state will be unlocked and no steps will be displayed on the Protocol status screen. It may be possible to complete processing on the BOND RX, or finish the remaining steps manually.

12.4.1.2 BOND RX^m

To manually unlock a slide staining assembly for the BOND RX^m, do the following:

- 1 Turn off the mains power and remove the power cable.
- 2 Open the bulk containers door and remove the bulk containers.
- 3 Slide the tray at the top of the bulk containers cavity out.

- 4 Locate the manual release knob (see [Figure 12-10](#)) beneath the slide staining assembly.
- 5 Turn the knob in the direction shown in [Figure 12-10](#). As you do the Covertiles should be moved over the slides and the whole assembly and tray will move up.
- 6 Continue turning the release knob until you feel a resistance. At this point it should be possible to remove the slide tray from the assembly.
- 7 Store your slides according to procedures at your facility.
- 8 Clean the lower and upper drip trays, if necessary, then re-insert the upper tray into the bulk containers cavity – the end of the tray with the 45 degree bend goes to the front, with the angle upwards.
- 9 Re-insert the bulk containers.
- 10 Close the bulk containers cavity door.

Protocol status should be checked (see [5.2 Protocol Status Screen](#)) before powering on the processing module.

When the processing module is powered on, it will initialize, detect the state of the assemblies and take any actions necessary to ready them for use.

After initialization the slide staining assembly state will be unlocked and no steps will be displayed on the Protocol status screen. It may be possible to complete processing on the BOND RX^m, or finish the remaining steps manually.

12.5 Restart Processing Module

Each processing module should be turned off and restarted weekly. This is important as it allows the processing module to complete a self-diagnostic check of the system.

The single-seat BOND RX controller does not need to be turned off and restarted on a regular basis. However, if there is a noticeable slowing in the BOND RX software, you may need to restart the controller via the Windows Start Menu.

However, if you have a BOND RX-ADVANCE system, see [16.1 Restarting the BOND RX-ADVANCE System](#).

Processing Module

For processing modules, ensure no runs are loaded, scheduled or processing, and turn off with the power switch on the right hand side of the processing module. Wait 30 seconds then turn back on. On startup the BOND RX system primes the fluidics system and runs a number of system tests (see [2.2.2 Processing Module Initialization](#)).

Note that you can run a partial prime of the fluidics system without powering down the processing module (see [Clean Fluidics](#)).

Clean Fluidics

The **Clean fluidics** button on the **Maintenance** screen primes the fluidics lines from the bulk containers (part of the processing module initialization run at startup). Run the routine if you suspect blockages or air in the fluidics delivery system.

- 1 Ensure that the processing module is idle, with no runs loaded, scheduled, or processing.
- 2 In the research client, select the processing module's tab to display its **System status** screen.

- 3 Click the **Maintenance** tab, and then click the **Clean fluidics** button.
- 4 Click **Yes** at the confirmation prompt.
- 5 The fluidics system is primed, which may take several minutes.

12.6 Aspirating Probe

The aspirating probe is automatically cleaned in the wash block between contact with each reagent as part of normal operation. However, additional weekly wiping, and cleaning with the BOND Aspirating Probe Cleaning System, should also be carried out. The cleaning system's reagents are optimized for the BOND RX system, and the BOND RX software uses a cleaning protocol designed to maximize wash efficiency. The BOND RX software warns users when probe cleans and replacements are due.



WARNING: Do not move the main robot arm while the processing module is switched on. The robot can become misaligned, resulting in poor staining.

If the robot has been moved: power down the processing module, wait 30 seconds and then reinitialize.

See:

- [12.6.1 Cleaning the Aspirating Probe](#)

12.6.1 Cleaning the Aspirating Probe

Always turn off the processing module before wiping and take care not to bend the probe. Wipe the exterior of the aspirating probe weekly using a 70% alcohol solution on a lint-free cloth or with an alcohol pad. Inspect the tubing attached to the aspirating probe, and ensure that there are no kinks or objects inside the tubing. The tubing should be clean.

The BOND RX software notifies you to clean the probe with the BOND Aspirating Probe Cleaning System every 300 slides (see [12.6.2 Running an Aspirating Probe Clean](#)). The count is automatically reset when a clean is run or the probe is replaced successfully.



BOND Aspirating Probe Cleaning Systems should be registered with the BOND RX system when received in the same way as detection systems (see [8.3.3 Registering Reagents and Reagent Systems](#)). The software keeps a record of cleaning system usage, allowing 15 cleans from each system.



To maintain the efficacy of the reagents in cleaning systems only load them onto processing modules when they are to be used. You cannot clean the aspirating probe while any other reagents or reagent systems are loaded on the processing module, and it is not possible to start slide processing while a cleaning system is loaded on the processing module.

12.6.2 Running an Aspirating Probe Clean

Follow the instructions below to clean the aspirating probe with the BOND Aspirating Probe Cleaning System.

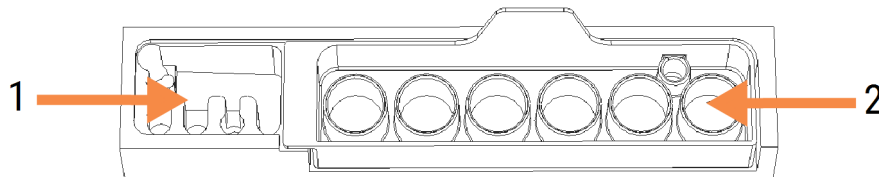
The cleaning protocol takes approximately 20 minutes to run.

- 1 Ensure that the processing module is idle, with no runs loaded, scheduled, or processing.
- 2 Remove all reagent or reagent system trays from the processing module.
- 3 Insert a BOND Aspirating Probe Cleaning System into the reagent tray on the processing module.
- 4 In the research client, select the processing module's tab to display its **System status** screen.
- 5 Click the **Maintenance** tab, and then click the **Clean aspirating probe** button.
- 6 Click **Yes** to start the clean when prompted.
The cleaning protocol begins, indicated by the cleaning icon in the processing module tab.
- 7 Wait until notified that the clean is finished.
- 8 Remove the BOND Aspirating Probe Cleaning System from the reagent tray.
- 9 Click **OK** in the **Cleaning complete** dialog to continue normal operation.

12.7 Wash Block and Mixing Station

The mixing station contains six wells for mixing reagents. It fits as an insert in the wash block.

Figure 12-11: Top view of wash block with wash area (1) and mixing station (2) in place



WARNING: Some of the reagents used in immunohistochemistry and in situ hybridization are hazardous. Ensure you have received adequate safety training before continuing.

Check the mixing station regularly for discoloration and general condition, and replace if necessary. Replace the station monthly as part of normal maintenance. Always ensure all runs are complete before removing.

To remove the mixing station, grasp the tab at the back of the mixing station and lift out.

Cleaning the Mixing Station

The mixing station can be reused until monthly replacement is due, provided it is not damaged or heavily discolored, and provided it is cleaned properly.

- 1 If cleaning is required, soak for a minimum of 30 minutes in a fresh solution of 0.5% W/V sodium hypochlorite in DI water.
- 2 Remove, and dip in fresh DI water 10 times.
- 3 Soak for a minimum of 10 minutes in reagent-grade alcohol.
- 4 Agitate for 30 seconds and remove.
- 5 Air dry.

Cleaning the Wash Block

Clean the wash block weekly using a lint-free cloth.



CAUTION: Do not use Q-tips or other cotton-tipped applicators to clean inside wash block holes – if the cotton tips come off they can block the holes.

12.8 Covers, Doors and Lid

Clean the covers, doors (where fitted) and lid of the processing module weekly with a duster or cloth.

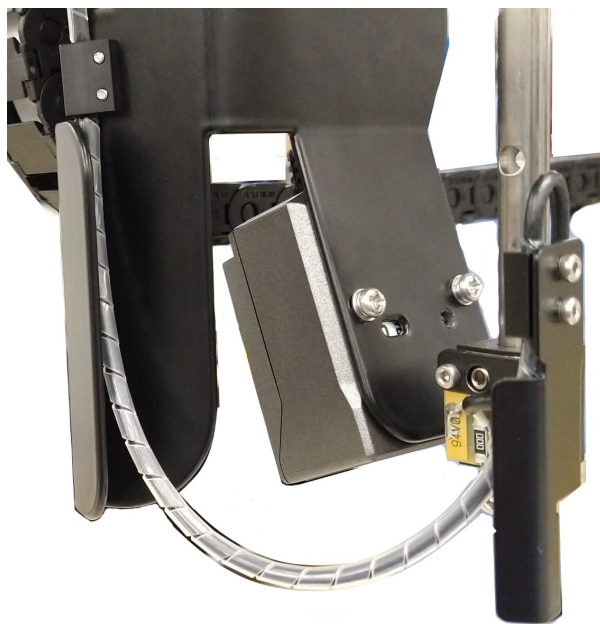
Do not use any cleaning agents, if required use water to damp a lint-free cloth to dust the covers, doors and the lid to prevent accumulation of dirt.

If any of the covers, doors or the lid becomes deformed or damaged, contact customer support for a replacement.

12.9 ID Imager

The window of the ID imager on the main robot arm needs to be kept clean to ensure slides are properly identified. Every week, or if the imager frequently fails to properly image IDs, clean the window with a lint-free cloth moistened with 70% alcohol solution.

Figure 12-12: ID imager



12.10 Drip Trays

Clean drip trays monthly, or more frequently if there is spilled reagent or waste evident. Contact customer support if there is evidence of excessive spills or salt build-up on the trays.

- [12.10.1 BOND RX Bulk Container Drip Trays](#)
- [12.10.2 BOND RX Processing Module Drip Tray](#)
- [12.10.3 BOND RX^m Bulk Container Drip Tray](#)

12.10.1 BOND RX Bulk Container Drip Trays

BOND RX has two bulk container drip trays located beneath the bulk containers on the upper and lower levels of the processing module.

To clean the BOND RX bulk container drip trays use the following procedure:

- 1 Ensure that the processing module is not in operation.
- 2 Remove all bulk containers.

- 3 Remove the black covers that protect the weight sensors of each bulk container (see [Figure 12-13](#)). Wipe each cover with a cloth or gauze dampened with a 70% alcohol solution.

Figure 12-13: BOND RX bulk container drip trays showing the weight sensor covers



- 4 Wipe the drip trays with the 70% alcohol solution. Avoid contact with the exposed metal weight sensors.
- 5 Dry the drip trays with paper towel.
- 6 Wipe down all bulk containers and return to their correct positions.

12.10.2 BOND RX Processing Module Drip Tray

BOND RX has a third drip tray located beneath the processing module, as shown in [Figure 12-14](#) below.

Figure 12-14: The BOND RX Processing Module drip tray



Use the following procedure to access the processing module drip tray:

- 1 Locate the drip tray beneath the processing module (see [Figure 12-14](#)), and pull the tray outwards. Use two hands to support the weight of the tray and to prevent liquid from spilling.
- 2 Empty the tray contents and dispose of the waste in accordance with approved procedures at your facility.



The tray has a channel at the rear corner to aid with pouring and preventing spillage.

- 3 Wash the tray with a 70% alcohol solution, then return to its correct position.

12.10.3 BOND RX^m Bulk Container Drip Tray

BOND RX^m has a single drip tray located under the bulk containers in the bulk containers cavity.

Use the following procedure to access the bulk container drip tray:

- 1 Ensure that the processing module is not in operation and remove all bulk containers.
- 2 Remove the drip tray and wipe with a cloth or gauze dampened with a 70% alcohol solution.
- 3 Dry the drip tray with paper towel and return to its correct positions (curved edge at the front of the processing module).
- 4 Wipe down all bulk containers and return to their correct positions.

12.11 Slide Trays

Clean slide trays monthly by washing with warm soapy water and rinsing with running water. Always ensure slide trays are dry before use. Replace deformed or damaged trays.

12.12 Bulk Fluid Robot Probes (BOND RX only)

The probe on each bulk fluid robot requires monthly cleaning with 70% alcohol solution on a lint-free cloth or with an alcohol pad.

Check the probes for deterioration while cleaning and change if required.

- [12.12.1 Cleaning the Bulk Fluid Robot Probes](#)

12.12.1 Cleaning the Bulk Fluid Robot Probes

Clean the bulk fluid robot dispensing probes monthly, taking care not to bend the probes.

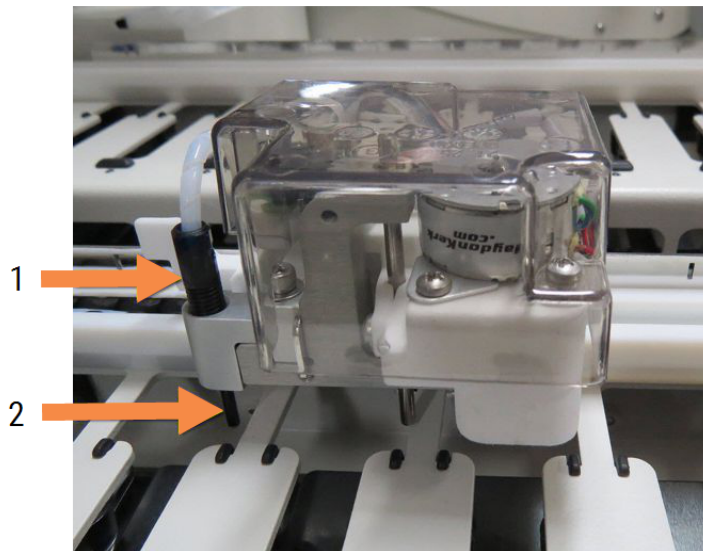


WARNING: The bulk fluid robots move along the slide staining assemblies to allow users access for cleaning. Only operators who have been warned of the potential hazards and have received adequate training should carry out this procedure.

- 1 Ensure that the processing module is idle with no runs loaded, scheduled or processing.
- 2 In the research client, select the processing module's tab to display its **System status** screen.
- 3 Click the **Maintenance** tab, and then click the **Clean bulk fluid robot probes** button.

- Carefully read the instructions in the **Clean bulk fluid robot probes** dialog, lock all slide trays and click **Yes** to continue.

Figure 12-15: Wipe all three bulk fluid robot probes with 70% alcohol solution (probe is indicated)



Legend

- | | |
|---|-----------------|
| 1 | Probe Connector |
| 2 | Probe |

- When all three bulk fluid robots have moved to the front of the processing module, turn it off.
- Gently clean the probes with 70% alcohol solution on a soft cloth, or with an alcohol pad.
Be very careful not to knock the probes out of alignment.
- In the dialog box, select the bulk fluid robot(s) you have cleaned successfully, and then click **Done**. Or, if you did not clean any, click the **None were cleaned** button.
- Restart the processing module. During initialization, the bulk fluid robots will return to the home position at the back of the processing module.

12.13 Syringes

The BOND RX software notifies you to replace the syringe (BOND RX^m) or syringes (BOND RX) every six months or 7800 slides processed, whichever comes first (see [5.1.2 Hardware Status](#)).



Visually check the syringes, especially at the top of the syringe and under the plunger, for leaks once a week during initialization or while running Clean fluidics (see [12.5 Restart Processing Module](#)). Additionally, check the attached tubing and connectors. Replace if there are any leaks.

If you would prefer that a Leica Biosystems field service engineer replaces the syringe(s), please contact customer support. Otherwise, you can replace the syringe(s) as described below.




WARNING: Always wear protective clothing and gloves.

- [12.13.1 Replacing BOND RX Syringes](#)
- [12.13.2 Replacing BOND RX^m 9-Port Syringe](#)

12.13.1 Replacing BOND RX Syringes

Unless replacing a single faulty syringe shortly after replacement, replace all syringes at the same time.

- 1 Ensure the processing module is idle (no runs loaded or scheduled).
- 2 In the research client, select the processing module's tab to display its **System status** screen.
- 3 Click the **Maintenance** tab, and then click the **Replace syringe** button.
- 4 Read the instructions and click **Yes**.

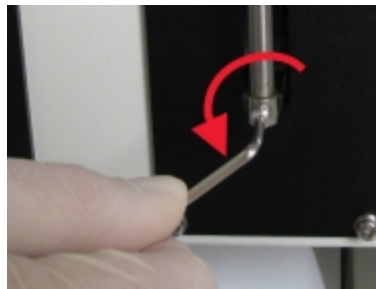
The processing module removes fluid from all the syringes and puts them into position for replacement (this may take up to 10 minutes). Wait for the processing module to disconnect , then turn it off. Do not turn off the controller (or terminal, in BOND RX-ADVANCE).



CAUTION: Ensure the processing module is off before proceeding.

- 5 Unscrew the four hex screws attaching the syringe module cover using the 3 mm hex key provided. Remove the cover.
- 6 For each syringe, loosen the syringe clamp thumbscrew and lower the clamp.
- 7 Use the 2.5 mm hex key provided to remove the plunger lock screw at the bottom of the plunger.

Figure 12-16: Unscrewing the plunger lock screw with a hex key




Some models have a thumbscrew instead of a hex screw.

- 8 Unscrew the syringe barrel from the valve. Remove the syringe and clamp from the processing module.
- 9 Place the new syringe through the clamp.
- 10 Place the syringe and clamp on the processing module – screw the syringe into the valve.
- 11 Replace the plunger lock screw and tighten.
- 12 Lift the clamp to the top of the syringe, and tighten the thumbscrew.
- 13 Refit the syringe module cover and secure using the four hex screws.
- 14 In the dialog box, select the syringe(s) you have replaced successfully, and then click **Done**. Or, if you did not replace any successfully, click the **None were replaced** button.

- 15 Restart the processing module.
- 16 Check for leaks while the processing module initializes, especially at the tops of the syringes and under the plungers. Report any leaks to customer support.
- 17 To check that the new syringes were installed correctly, run test tissues or control tissues to verify that proper staining is achieved.

12.13.2 Replacing BOND RX^m 9-Port Syringe

- 1 Ensure the processing module is idle (no runs loaded or scheduled).
- 2 In the research client, select the processing module's tab to display its **System status** screen.
- 3 Click the **Maintenance** tab, and then click the **Replace syringe** button.
- 4 Read the instructions and click **Yes**.

The processing module removes fluid from the syringe and puts it into position for replacement. Wait for the processing module to disconnect , then turn it off. Do not turn off the controller (or terminal, in BOND RX-ADVANCE).



CAUTION: Ensure the processing module is off before proceeding.

- 5 Loosen the syringe clamp thumbscrew and lower the clamp.
- 6 Use a hex key to remove the plunger lock screw at the bottom of the plunger.

Figure 12-17: Unscrewing the plunger lock screw with a hex key



Some models have a thumbscrew instead of a hex key.

- 7 Unscrew the syringe barrel from the valve. Remove the syringe and clamp from the processing module.
- 8 Place the new syringe through the clamp.
- 9 Place the syringe and clamp on the processing module – screw the syringe into the valve.
- 10 Replace the plunger lock screw and tighten.
- 11 Lift the clamp to the top of the syringe, and tighten the thumbscrew.
- 12 In the dialog box, click **Yes** to confirm that you have replaced the syringe successfully.

- 13 Restart the processing module.
- 14 Check for leaks while the processing module initializes, especially at the top of the syringe and under the plunger. Report any leaks to customer support.
- 15 To check that the new syringe was installed correctly, run test tissues or control tissues to verify that proper staining is achieved.

12.14 Power Supply Fuses

Legacy BOND RX and BOND RX^m Processing Modules have two mains fuses and two heater supply fuses. Alternate BOND RX and BOND RX^m Processing Modules have only two mains fuses. Fuse ratings differ according to the mains power supply. Fuses are located in the back cover (see [2.2.13 Back Cover](#)).

Legacy BOND RX uses the following fuses:

Fuse	Description	100–240 VAC Supply
F1	Heater Power Supply	3AG T8A 250V UL
F2	System Power Supply	3AG T8A 250V UL
F3	AC Mains (Neutral)	3AG T15A 250V UL
F4	AC Mains (Active)	3AG T15A 250V UL

Alternate BOND RX uses the following fuses:

Fuse	Description	100–240 VAC Supply
F3	AC Mains (Neutral)	3AG T15A 250V UL
F4	AC Mains (Active)	3AG T15A 250V UL

Legacy BOND RX^m Processing Modules use the following fuses:

Fuse	Description	100–240 VAC Supply
F1	AC Mains (Active)	3AG T15A 250V UL
F2	AC Mains (Neutral)	3AG T15A 250V UL
F3	24 V Heater Supply	3AG T8A 250V UL
F4	24 VDC Power Supply	3AG T8A 250V UL

Alternate BOND RX^m Processing Modules use the following fuses:

Fuse	Description	100–240 VAC Supply
F1	AC Mains (Active)	3AG T15A 250V UL
F2	AC Mains (Neutral)	3AG T15A 250V UL

Replacement fuse specifications are also printed on the back cover.



WARNING: Do not bypass or short-circuit fuses.

Turn off the processing module and disconnect the power cord before changing fuses.

Replace fuses only with standard parts and if fuses blow repeatedly, contact customer support.

To replace the fuses, do the following:

- 1 Turn off the processing module.
- 2 Switch off the mains power supply and disconnect the mains power supply from the wall socket.
- 3 Unscrew the fuse cover.
- 4 Pull out the fuse cover and replace the fuse. Ensure you replace with a fuse of the correct specifications.
- 5 Push in the fuse cover and screw clockwise to lock the fuse in position. Do not over-tighten.

13 Cleaning and Maintenance (Miscellaneous)

13.1 Handheld Barcode Scanners

- [13.1.1 Honeywell Barcode Scanner](#)
- [13.1.2 Zebra DS2208 Barcode Scanner](#)
- [13.1.3 Newland NLS-HR2000 Barcode Scanner](#)

13.1.1 Honeywell Barcode Scanner

Clean your handheld scanner weekly:

- Do not allow any abrasive material to touch the window
- Do not spray water or other cleaning liquids directly into the window

Clean the scanner by:

- First disconnecting the scanner from the controller or terminal.
- Removing dirt particles with a water-dampened, lint-free cloth.
- Cleaning the window with a lint-free cloth moistened with 70% alcohol solution.

If the handheld barcode scanner is not operating correctly, your service organization may request that it be re-initialized. You can also adjust the beeper volume on the scanner.

13.1.1.1 Configuring the Honeywell Barcode Scanner

To re-initialize a Honeywell barcode scanner (USB), print a good-quality hard copy of this page and scan the barcodes in the order shown below:

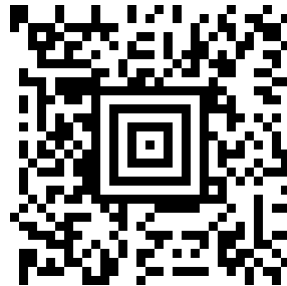
Figure 13-1: Barcodes for scanner configuration



Scan 1: Remove Custom Defaults



Scan 2: Activate Defaults



Scan 3: Honeywell Scanner Configuration

13.1.1.2 Setting Beeper Volume

To set the beeper volume for a Honeywell barcode scanner, print a good-quality hard copy of this page and scan the barcode below that corresponds to the desired level.

Figure 13-2: Beeper volume barcodes for Honeywell scanner



Low volume



Medium volume



High volume



Beeper off

13.1.1.3 Configuring Hands-Free Use

When the scanner is placed in its stand then it is normally in hands-free use, and you do not need to press the trigger when reading a barcode.

To set hands-free use ON or OFF for a Honeywell barcode scanner, print a good-quality hard copy of this page and scan the barcode below that corresponds to the desired functionality.

Figure 13-3: Hands-free use barcodes for Honeywell scanner



Hands-free use ON



Hands-free use OFF

13.1.2 Zebra DS2208 Barcode Scanner



Clean your handheld scanner weekly:

- Do not allow any abrasive material to touch the window
- Do not spray water or other cleaning liquids directly into the window

Clean the scanner by:

- First disconnecting the scanner from the controller or terminal.
- Removing dirt particles with a water-dampened, lint-free cloth.
- Cleaning the window with a lint-free cloth moistened with 70% alcohol solution.

If the handheld barcode scanner is not operating correctly, your service organization may request that it be re-initialized. You can also adjust the beeper volume on the scanner.

13.1.2.1 Configuring the Zebra Barcode Scanner

To re-initialize a Zebra barcode scanner (USB), print a good-quality hard copy of this page and scan each of the following barcodes in turn.

Figure 13-4: Barcode scanning sequence for Zebra scanner configuration



Scan 1: Set Defaults



Scan 2: Enable Code 128



Scan 3: Scan Options



Scan 4: <DATA> <SUFFIX>



Scan 5: Enter



Scan 6: Override Caps Lock Key (Enable)

13.1.2.2 Setting Beeper Volume

To set the beeper volume for a Zebra barcode scanner, print a good-quality hard copy of this page and scan the barcode below that corresponds to the desired level.

Figure 13-5: Beeper volume barcodes for Zebra scanner



Low volume



Medium volume



High volume

13.1.2.3 Configuring Hands-Free Use

When the scanner is placed in its stand then it is normally in hands-free use, and you do not need to press the trigger when reading a barcode.

To set hands-free use ON or OFF for a Zebra barcode scanner, print a good-quality hard copy of this page and scan the barcode below that corresponds to the desired functionality.

Figure 13-6: Hands-free use barcodes for Zebra scanner



Hands-free use ON



Hands-free use OFF

13.1.3 Newland NLS-HR2000 Barcode Scanner



Clean your handheld scanner weekly:

- Do not allow any abrasive material to touch the window
- Do not spray water or other cleaning liquids directly into the window

Clean the scanner by:

- First disconnecting the scanner from the controller or terminal.
- Removing dirt particles with a water-dampened, lint-free cloth.
- Cleaning the window with a lint-free cloth moistened with 70% alcohol solution.

If the handheld barcode scanner is not operating correctly, your service organization may request that it be re-initialized. You can also adjust the beeper volume on the scanner.

13.1.3.1 Configuring the Newland Handheld Barcode Scanner

To re-initialize a Newland barcode scanner (USB), print a good-quality hard copy of this page and scan the barcodes in turn:

Scan 1: Enter Setup



Scan 2: Restore All Factory Defaults



Scan 3: Enable Aztec Code



Scan 4: Enable Micro QR



Scan 5: Exit Setup



13.1.3.2 Setting Beeper Volume

To set the beeper volume for a Newland barcode scanner, print a good-quality hard copy of this page and scan the barcode below that corresponds to the desired level.

Scan 1: Enter Setup



Scan 2: Beep Volume

Low volume
Medium volume
High volume

The 'Scan 2: Beep Volume' section is contained within a large rectangular frame. It is divided into three horizontal sections, each containing a barcode and a label above it. The top section is labeled 'Low volume' and contains a barcode. The middle section is labeled 'Medium volume' and contains a barcode. The bottom section is labeled 'High volume' and contains a barcode.

Scan 3: Exit Setup



13.1.3.3 Configuring Hands-Free Use

When the scanner is placed in its stand then it is normally in hands-free use, and you do not need to press the trigger when reading a barcode.

To set hands-free use ON or OFF for a Newland barcode scanner, print a good-quality hard copy of this page and scan the barcode below that corresponds to the desired functionality.

Scan 1: Enter Setup



Scan 2: Hands-Free Use



Scan 3: Exit Setup



13.1.3.4 Keyboard Layout

For AZERTY keyboards, scan the relevant bar code below.

Scan 1: Enter Setup



Scan 2: AZERTY Keyboards



Scan 3: Exit Setup



13.2 Slide Labeler

Manuals are provided with the slide labeler. Refer to these for instructions on cleaning and loading labels and printing ribbon. Clean monthly.

14 Using BOND Reagents

This chapter has general discussion of the principles of IHC and ISH staining, and guidelines for good laboratory practice. It includes descriptions of the BOND detection systems.



The BOND RX system is for research use only. While some of the discussion in this chapter may apply in the clinical environment, the BOND RX system must never be used for diagnostic purposes.

- [14.1 Principle of the Procedure](#)
- [14.2 Specimen Preparation](#)
- [14.3 Quality Control](#)
- [14.4 Interpretation of Staining](#)
- [14.5 General Limitations](#)
- [14.6 References](#)

14.1 Principle of the Procedure

Immunohistochemistry (IHC)

Immunohistochemical techniques have been used to detect specific antigens in cells or tissue for at least 50 years. The first reported method used fluorescent labels in 1941¹. Subsequently, enzymes such as peroxidase, were introduced². Today immunohistochemistry is used to facilitate cell recognition alongside routine H & E paraffin stains, and is an aid to the recognition of normal and abnormal cells. Immunohistochemical methods have become the “standard of care” in surgical pathology when classic methods alone fail to yield a definitive diagnosis^{3,4}. However, there have been some reservations concerning reproducibility⁵, despite almost universal adoption.

Reagents on the automated BOND RX system demonstrate antigens in tissue sections by immunohistochemical techniques. In summary, a specific primary antibody binds to a section, then BOND detection system reagents visualize the complex.



A diagnostic “marker” is a reagent that is used to detect a specific antigen or DNA/RNA binding site in a tissue sample. The marker is the primary antibody in IHC, or the probe in ISH (see below).

In Situ Hybridization (ISH)

Molecular biological techniques have largely advanced our understanding of disease. In situ hybridization combines both molecular biology and histology, allowing visualization of DNA or RNA in their cellular context. Since nucleic acid detection was first introduced in 1969⁶, improvements to in situ hybridization protocols have made it an increasingly valuable tool for clinical pathology as well as research.

In situ hybridization utilizes the complementary binding of nucleotide bases in DNA or RNA. A labeled nucleic acid probe binds specifically to its complementary sequence in fixed tissue or cell specimen. The probe is visualized through the application of an antibody against the label followed by BOND polymer detection reagents. The BOND RX automated system and reagents offer a reliable and efficient alternative to a cumbersome manual technique.

14.1.1 BOND Detection Systems

Leica Biosystems supplies a range of detection systems developed specifically for the BOND RX system. Foremost amongst these is the BOND Polymer Refine Detection™ system, which provides high intensity staining coupled with sharp definition without the use of streptavidin and biotin.

The BOND detection systems available are listed in the sections below.

- [14.1.1.1 BOND Polymer Refine Detection](#)
- [14.1.1.2 BOND Polymer Refine Red Detection](#)
- [14.1.1.3 BOND Streptavidin-Biotin Detection \(DAB\)](#)

14.1.1.1 BOND Polymer Refine Detection

The BOND polymer DAB-based system, BOND Polymer Refine Detection, delivers high intensity staining coupled with sharp delineation of antibody binding to the target antigen or probe binding to the nucleic acid. The system does not use streptavidin and biotin, and therefore eliminates nonspecific staining as a result of endogenous biotin. Endogenous biotin is prevalent in some tissues such as gastrointestinal tract, kidney, liver, and breast carcinoma. BOND polymer detection systems have higher sensitivity than labeled streptavidin-biotin systems, resulting in lower antibody concentrations and faster turnaround times.

At each step the BOND RX system incubates the sections for a precise time, then washes the sections to remove unbound material. Protocol steps including incubation, washing, and interpretation of results are carried out as described within BOND Polymer Refine Detection instructions for use. Results are interpreted using an optical light microscope, and aid in the differential diagnosis of pathological processes, that may or may not be associated with a particular antigen.

If stronger intensity is desired the following options are available for all BOND polymer detection systems:

- Increase the incubation times for the primary antibody or probe and/or detection system components.
- Use a BOND DAB Enhancer step. Note that an enhancer alone will not increase the level of staining intensity to the same extent as that produced by the Intense R detection system.
- For IHC only, increase the primary antibody concentration.

The BOND Oracle HER2 IHC System is a complete system for determining the presence of the target protein and therefore the suitability of treatment with the targeted therapy. This assay is provided as a total, optimized system with the ready-to-use antibody, detection reagents, control reagents and control slides to provide complete quality assurance for the diagnostic result. The assay is based on IHC methodology. Full instructions for use are provided with the system. Use these instructions to set up runs. Due to the nature of HER2 IHC testing, it is of the utmost importance that these instructions are followed exactly so as not to invalidate the assay.

14.1.1.2 BOND Polymer Refine Red Detection

The BOND Polymer Refine Red Detection™ has the same advantages as the DAB-based polymer detection systems described above, but Fast Red chromogen is used for visualization instead of DAB. The system is suited for use on tissues such as skin where tissue pigments can be mistaken for DAB.

The BOND Polymer Refine Red Detection system is a highly sensitive Compact Polymer™ system conjugated to alkaline phosphatase that provides bright fuchsia red immunostaining, as well as hematoxylin counterstain (including bluing).



Fast Red chromogen is chemically unstable in normal laboratory conditions. Be sure to strictly follow user instructions for BOND Polymer Refine Red Detection to maintain chromogen efficacy. Always place control tissue on the same slide as test tissue to allow rapid detection of any deterioration in the system.



Leica CV Ultra Mounting Media is recommended for use with BOND Polymer Refine Red Detection system. Other mountants may not preserve the intensity of staining initially obtained.

The steps for the BOND Polymer Refine Red Detection system are:

- 1 Application of the specific primary antibody.
- 2 Incubation with a post primary reagent.
- 3 Incubation with the polymer reagent, which comprises polymeric alkaline phosphatase (AP) tertiary antibody conjugates.
- 4 Visualization of the complex with substrate chromogen, Fast Red, via a red precipitate.
- 5 Hematoxylin counterstaining allows the detection of cell nuclei.

Incubation, washing, and interpretation of results are carried out as described for the BOND Labeled Streptavidin-Biotin Detection System.

14.1.1.3 BOND Streptavidin-Biotin Detection (DAB)

There is one detection system in this category: BOND Intense R Detection.

This DAB-based detection system operates as follows:

- 1 Incubation with hydrogen peroxide to quench endogenous peroxidase activity.
- 2 Application of the specific primary antibody.

- 3 The antibody is localized by a user supplied biotin conjugated secondary antibody formulation that recognizes their primary antibody.
- 4 Addition of a streptavidin-enzyme conjugate that binds to the biotin present on the secondary antibody.
- 5 Visualization of the complex with a substrate chromogen (3,3'-diaminobenzidine, or DAB), whose enzyme product is a brown precipitate.
- 6 Hematoxylin counterstaining allows the detection of cell nuclei.

At each step the BOND RX system incubates the sections for a precise time, then washes the sections to remove unbound material. Results are interpreted using an optical light microscope, and aid in the differential diagnosis of pathological processes, that may or may not be associated with a particular antigen.

14.2 Specimen Preparation

This section discusses the preparation of tissue for staining.

- [14.2.1 Materials Required](#)
- [14.2.2 Tissue Preparation](#)
- [14.2.3 Dewaxing and Baking](#)
- [14.2.4 Epitope Retrieval](#)

14.2.1 Materials Required

The following materials are required for immunohistochemical and in situ hybridization staining using the BOND RX system.

14.2.1.1 Common Materials

- Fixative – recommended 10% Neutral buffered formalin
- Paraffin wax
- Tissue processor and embedding center
- Positive and negative tissue controls (see [14.3 Quality Control](#))
- Microtome
- Drying oven
- Mounting medium, resin-based or aqueous-based
- Charged microscope slides (e.g. Leica BOND Plus Slides)
- BOND Slide Labels and Printer Ribbon
- Coverslips
- BOND Universal Covertiles
- Appropriate BOND reagent system

- BOND Enzyme Pretreatment Kit
- BOND Dewax Solution
- Wash solution (prepared from BOND Wash Solution 10X Concentrate)
- Deionized water
- Alcohol (reagent grade*)



* Reagent grade alcohol comprises: Ethanol, greater than or equal to 90% (w/w); Isopropanol, no more than 5% (w/w); Methanol, no more than 5% (w/w).

14.2.1.2 Materials for IHC

In addition to the materials listed above, the following are required for IHC tests:

- Negative control reagents specific to primary antibodies (see [14.3 Quality Control](#))
- BOND Epitope Retrieval Solution 1
- BOND Epitope Retrieval Solution 2
- BOND ready-to-use primary antibodies, or primary antibodies diluted in BOND Primary Antibody Diluent in BOND open containers, 7 mL or 30 mL
- Mounting medium, resin-based or aqueous-based
- Titration kit, optional (see [14.2.1.4 Titration Kit](#))

14.2.1.3 Materials for ISH

In addition to the common materials listed above, the following are required for ISH tests:

- ISH probes
- Anti-fluorescein antibody
- Positive and negative control probes specific to ISH (see [14.3 Quality Control](#))

14.2.1.4 Titration Kit

The BOND Titration Kit consists of 10 empty containers and 50 inserts (6 mL) and is used when optimizing the concentration of primary antibodies for the BOND RX system. Small volumes of each primary antibody concentration can be prepared and placed into the inserts. Each container may be used for a total of 40 mL of reagent.

14.2.2 Tissue Preparation

We recommend 15 to 20 times the volume of tissue of 10% neutral-buffered formalin to fix tissue for immunohistochemical and in situ hybridization staining using the BOND RX system. Fixation can be performed at room temperature (15–25 °C).

To facilitate tissue cutting and prevent damage to microtome blades, decalcify osseous tissues prior to tissue processing ^{11,12}.

Cut and pick up 3–5 µm thick sections on charged glass slides (some specific tissue types may require different section thicknesses). To dry tissue place the well drained slides in a 60 °C (±5 °C) oven for 10–30 minutes, or overnight at 37 °C. Slides can also be baked on the BOND RX and BOND RX^m Processing Modules. Slides must be well air-dried before baking. Consult references 13, 14 and 15 for further details on specimen preparation.

Affix slide labels to specimen and control slides as described in [4 Quick Start](#). Dewax, rehydration, and epitope retrieval are fully automated on the BOND RX system.

14.2.3 Dewaxing and Baking

Paraffin-embedded tissue sections for immunohistochemistry must first have the paraffin wax removed and the section rehydrated. The wax is removed using BOND Dewax Solution and the sections are rehydrated. The BOND RX system includes Dewax protocols that automate this process.

Prior to dewaxing, the BOND RX and BOND RX^m Processing Modules can also bake the tissue to improve its adhesion to the slide. The BOND RX system Bake and Dewax protocols automate both the baking and dewaxing processes.



Please note that tissue must be air-dried to remove any water before it is placed into a processing module for baking and dewaxing.

14.2.4 Epitope Retrieval

Formalin fixation of tissue causes cross-linking between the aldehyde and amino groups in the tissue and the formation of these bonds can result in variable loss of antigenicity due to the masking effect. Formalin forms methylene bridges which can change the overall three-dimensional shape of the epitope. Some epitopes are formalin-sensitive and show reduced immunoreactivity after formalin fixation whilst others are formalin-resistant.

Nucleic acids are surrounded by proteins, therefore permeabilization of tissue is needed to make target sequences accessible to the probe.

Epitope retrieval ^{7,8} can be achieved either by using heat induced epitope retrieval (HIER), enzyme pretreatment, or by a combination of both. HIER is the most extensively used method of epitope retrieval for IHC. The mechanism of HIER is not completely understood.

The hypothesis is that heating the section to a high temperature in an epitope retrieval solution hydrolyzes the cross-linkages formed in formalin fixation. This results in remodification of the epitope which can then be stained by immunohistochemistry. The important factors in HIER are temperature, time and pH of the retrieval solution. There are two different epitope retrieval solutions for use on the BOND RX system: a citrate-based buffer and an EDTA-based buffer.

Enzyme pretreatment uses proteolytic enzymes to break peptide bonds to expose the epitope/target nucleic acid sequence. The enzyme concentration and incubation time is proportional to the fixation time of the specimen and should be optimized accordingly. Enzyme pretreatment is only suitable for some epitopes but is used frequently in ISH protocols.

14.3 Quality Control

Differences in tissue processing and technical procedures in the user's laboratory may produce significant variability in results, necessitating regular performance of in-house controls in addition to the following procedures.



Controls should be fresh autopsy/biopsy/surgical specimens fixed, processed and embedded as soon as possible in the same manner as the test sample(s). Such a control monitors all steps of the analysis, from tissue preparation through to staining.



We strongly recommend placing appropriate control tissue on the same slides as test tissue. See [6.2 Working with Controls](#) for further discussion.

See:

- [14.3.1 Assay Verification](#)
- [14.3.2 Tissue Controls](#)
- [14.3.3 Negative Reagent Control for IHC](#)
- [14.3.4 Reagent Controls for ISH](#)
- [14.3.5 The Benefits of Quality Control](#)

14.3.1 Assay Verification

Prior to initial use of an antibody, probe or staining system in a diagnostic procedure, verify the specificity of the antibody/probe by testing it on a series of in-house tissues with known expression representing known positive and negative tissues. Repeat these quality control procedures for each new antibody lot, or whenever there is a change in assay parameters. Quality control cannot be meaningfully performed on an individual reagent in isolation, since the matched reagents, along with a defined assay protocol, must be tested in unison before using a detection system. Refer to each primary antibody package insert for tissues that are suitable for assay verification.

In addition to the above-mentioned assay verification procedures we recommend staining positive tissue controls monthly and comparing them to the same tissue control stained the previous month. Comparison of controls stained at monthly intervals serves to monitor the assay stability, sensitivity, specificity and reproducibility.

All quality control requirements should be performed in conformity with local, state and/or federal regulations or accreditation requirements.

14.3.2 Tissue Controls

14.3.2.1 Positive Tissue Control

- Indicates correctly prepared tissues and proper staining techniques.
- Include one positive tissue control for each set of test conditions in each staining run.
- Tissue with weak positive staining is more suitable than tissue with strong positive staining for optimal quality control and to detect minor levels of reagent degradation¹⁴.
- Using a multi-tissue control slide that contains tissues exhibiting strong, medium and weak antigen density/nucleic acid expression provides wide control coverage.
- If the positive tissue control fails to demonstrate positive staining, results with the test specimens should be considered invalid.
- We strongly recommend that you always run the BOND RX system with a control tissue on the same slide as the sample tissue to ensure optimum quality control.

14.3.2.2 Negative Tissue Control

- Examine after the positive tissue control to verify the specificity of the labeling of the target antigen by the primary antibody in IHC or target nucleic acid by the probe in ISH, and to provide an indication of specific background staining (false positive staining).
- The variety of different cell types present in most tissue sections frequently offers negative control sites, but the user should verify this.
- If specific staining occurs in the negative tissue control, results with the test specimens should be considered invalid.

14.3.3 Negative Reagent Control for IHC

Use negative reagent control for IHC in place of the primary antibody with a section of each test specimen to evaluate nonspecific staining and allow better interpretation of specific staining.

- Recommended ideal control reagent:
 - a For monoclonal antibodies use an antibody of the same isotype that is produced from tissue culture supernatant and in the same way as the primary antibody, but that exhibits no specific reactivity with human tissues.

Dilute this to the same immunoglobulin or protein concentration as the primary antibody using identical diluent (BOND Primary Antibody Diluent).

If fetal calf serum is retained in the neat antibody after processing, fetal calf serum at a protein concentration equivalent to the diluted primary antibody in the same diluent is also suitable for use.

- b** For polyclonal antibodies use an immunoglobulin fraction (or whole serum, if appropriate) of normal or nonimmune serum from the same animal source and the same protein concentration as the primary antibody, using identical diluent (BOND Primary Antibody Diluent).
- BOND Primary Antibody Diluent alone may be used as a less desirable alternative to the previously described negative reagent controls.
- The incubation period for the negative reagent control should correspond to that of the primary antibody.
- Use a separate negative reagent control slide for each retrieval method employed (including no retrieval) for a given primary antibody.
- When panels of several antibodies are used on serial sections, the negatively staining areas of one slide may serve as negative/nonspecific binding background controls for other antibodies.
- To differentiate endogenous enzyme activity or nonspecific binding of enzymes from specific immunoreactivity, stain additional test tissues exclusively with substrate-chromogen or enzyme complexes and substrate-chromogen, respectively.
- The BOND RX system includes a default negative IHC control reagent, named “*Negative”, that can be selected as the marker for any IHC protocol. It dispenses BOND Wash (see [10.5.2 Study and Slide Settings](#)).

14.3.4 Reagent Controls for ISH

14.3.4.1 Positive Reagent Control

For in situ hybridization use the Positive Control Probe.

- Use in place of the probe with a section of each test specimen to provide information on the preservation of nucleic acids in the tissue as well as accessibility of nucleic acids to the probe.
- The protocol for the Positive Probe Control should correspond to that of the test probe.
- If the Positive Control Probe fails to demonstrate positive staining, results with the test specimens should be considered invalid.

14.3.4.2 Negative Reagent Control

For in situ hybridization use the Negative Control Probe.

- The protocol for the Negative Control Probe should correspond to that of the test probe.
- Use in place of the probe with a section of each test specimen to evaluate nonspecific staining and allow better interpretation of specific staining.
- The incubation period for the negative reagent control should correspond to that of the probe.
- Use a separate negative reagent control slide for each retrieval method employed (including no retrieval) for a given probe.
- To differentiate endogenous enzyme activity or nonspecific binding of enzymes from specific immunoreactivity, stain additional test tissues exclusively with substrate-chromogen or enzyme complexes and substrate-chromogen, respectively.

14.3.5 The Benefits of Quality Control

The benefits of quality control are summarized in the table below.

<p>Positive Tissue Control:</p> <p>Tissue or cells containing target antigen/nucleic acid sequence to be detected (could be located in test tissue).</p> <p>The ideal control is weakly positive staining tissue to be most sensitive to antibody/nucleic acid degradation.</p>	<p>Controls all steps of the analysis.</p> <p>Validates reagent and procedures used for staining.</p>		<p>Detection of nonspecific background staining</p>
<p>Negative Tissue Control:</p> <p>Tissues or cells expected to be negative (could be located in test tissue or positive control tissue)</p>	<p>Detection of unintended antibody cross-reactivity to cells/cellular components [IHC]</p> <p>Detection of unintended probe cross- hybridization to other nucleic acid sequences or cells/cellular components [ISH]</p>		<p>Detection of nonspecific background staining</p>
<p>Test Tissue</p>	<p>Detection of specific staining</p>	<p>Assessment of nucleic acid preservation/tissue fixation and/or retrieval [ISH]</p>	<p>Detection of nonspecific background staining</p>

14.4 Interpretation of Staining

The specificity and sensitivity of antigen detection are dependent on the specific primary antibody utilized. To ensure desired staining, optimize each specific antibody on the BOND RX system, varying the time of incubation and/or the specific antibody concentration. Failure to optimize the specific antibody may result in suboptimal antigen detection.

See:

- [14.4.1 Positive Tissue Control](#)
- [14.4.2 Negative Tissue Control](#)
- [14.4.3 Test Tissue](#)

14.4.1 Positive Tissue Control

Examine the positive tissue control first to ascertain that all reagents are functioning properly.

When using DAB-based systems, the presence of a brown (3,3' diaminobenzidine tetrachloride, DAB) reaction product with the target cells indicates positive reactivity. When using RED Chromogen-based systems, the presence of a red reaction product with the target cells indicates positive reactivity. If the positive tissue controls fail to demonstrate positive staining, results with the test specimens should be considered invalid.

14.4.2 Negative Tissue Control

Examine the negative tissue control after the positive tissue control to verify the specificity of the labeling of the target antigen/nucleic acid by the primary antibody/probe.

The absence of specific staining in the negative tissue control confirms the lack of antibody/probe cross-reactivity to cells/cellular components.

If specific staining (false positive staining) occurs in the negative external tissue control, results should be considered invalid. Nonspecific staining, if present, usually has a diffuse appearance. Sporadic staining of connective tissue may also be observed in sections from excessively formalin-fixed tissues. Use intact cells for interpretation of staining results. Necrotic or degenerated cells often stain nonspecifically.

14.4.3 Test Tissue

Examine test specimens stained with the primary antibody/probe last.

Positive staining intensity should be assessed within the context of any nonspecific background staining of the negative reagent control. As with any immunohistochemical or in situ hybridization test, a negative result means that the antigen/nucleic acid was not detected, not that the antigen/nucleic acid was absent in the cells or tissue assayed.

If necessary, use a panel of antibodies to identify false negative reactions.

14.5 General Limitations

- Immunohistochemistry and in situ hybridization are multi-step diagnostic processes that require specialized training in the selection of the appropriate reagents; tissue selection, fixation, and processing; preparation of the slide; and interpretation of the staining results.
- Tissue staining is dependent on the handling and processing of the tissue prior to staining. Improper fixation, freezing, thawing, washing, drying, heating, sectioning or contamination with other tissues or fluids may produce artifacts, antibody trapping, or false negative results. Inconsistent results may be due to variations in fixation and embedding methods, or to inherent irregularities within the tissue¹⁸.
- Excessive or incomplete counterstaining may compromise proper interpretation of results.
- Tissues from persons infected with hepatitis B virus and containing hepatitis B surface antigen (HbsAg) may exhibit nonspecific staining with horseradish peroxidase¹⁹.

- Unexpected negative reactions in poorly differentiated neoplasms may be due to loss or marked decrease of expression of antigen or loss or mutation(s) in the gene(s) coding for the antigen. Unexpected positive staining in tumors may be from expression of an antigen not usually expressed in morphologically similar normal cells, or from persistence or acquisition of an antigen in a neoplasm that develops morphologic and immunohistochemical features associated with another cell lineage (divergent differentiation). Histopathologic classification of tumors is not an exact science and some literature reports of unexpected staining may be controversial.
- Reagents may demonstrate unexpected reactions in previously untested tissues. The possibility of unexpected reactions even in tested tissue groups cannot be completely eliminated due to biological variability of antigen expression/target nucleic acid in neoplasms, or other pathological tissues. Contact your local distributor or the regional office of Leica Biosystems to report any unexpected reaction.

IHC

- Normal or nonimmune sera from the same animal source as secondary antisera used in blocking steps may cause false negative or false positive results due to autoantibodies or natural antibodies.
- False positive results in IHC may be seen due to nonimmunological binding of proteins or substrate reaction products. They may also be caused by pseudoperoxidase activity (erythrocytes), endogenous peroxidase activity (cytochrome C), or endogenous biotin (for example, liver, breast, brain, kidney) depending on the type of immunostain used ¹⁶.
- False negative results in IHC may result from various factors, including true antigen decrease, loss or structural change during tumor "dedifferentiation", or artefactual change during fixation or processing. As with any immunohistochemical test, a negative result means that the antigen was not detected, not that the antigen was absent in the tissues assayed.

ISH

- False positive results in ISH may be seen due to cross-reactivity of the probe to other nucleic acid sequences as well as nonspecific binding of probe or detection reagents to tissue or tissue components ¹⁸. Negative tissue and reagent controls should be included in testing to help identify false positive staining.
- DNA and RNA are subject to degradation by nuclease activity ^{8,19}. Therefore, it is important to test the Positive Control Probe with test tissue in parallel with specific probe and test tissue to detect nucleic acid degradation. The choice of fixative influences conservation of nucleic acids, tissue fixed in 10% neutral buffered formalin is recommended for this reason ¹⁹. As with any in situ hybridization test, a negative result means that the nucleic acid was not detected, not that the nucleic acid was absent in the tissues assayed.

14.6 References

- 1 Coons AH et al. Immunological properties of an antibody containing a fluorescent group. *Proc Soc Exp Biol Med* 1941; 47:200-202.
- 2 Nakane PK and Pierce GB Jr. Enzyme labeled antibodies: Preparations and applications for the localizations of antigens. *J Histochem Cytochem* 1967; 14:929-931.
- 3 Elias JM, Gown AM, Nakamura RM, Wilbur DC, Herman GE, Jaffe ES, Battifora H, and Brigati J. Special report: Quality control in immunohistochemistry. *Am J Clin Path* 1989; 92:836.
- 4 Nadji M and Morales AR. *Immunoperoxidase techniques: a practical approach to tumor diagnosis*. ASCP Press, Chicago. 1986.
- 5 True LD ed. *Atlas of Diagnostic Immunohistopathology*. Lippincott, Philadelphia. 1990.
- 6 Gall JG, Pardue ML. Formation of RNA-DNA hybrid molecules in cytological preparation. *Proceedings of the National Academy of the Sciences of the United States of America*. 1969;63:378-383.
- 7 Shi S-R, Gu J, and Taylor CR. *Antigen Retrieval Techniques: Immunohistochemistry and Molecular Morphology*. Eaton Publishing, Natick. 2000.
- 8 Miller RT, Swanson PE, and Wick MR. Fixation and epitope retrieval in diagnostic immunohistochemistry: a concise review with practical considerations. *Appl Immunohistochem Mol Morphol*. 2000 Sep;8(3):228-35.
- 9 Bancroft JD and Stevens A. *Theory and Practice of Histological Techniques*. 4th Edition. Churchill Livingstone, New York. 1996.
- 10 Wolff et al. American Society of Clinical Oncology/College of American Pathologists Guideline Recommendations for Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer. *Arch Pathol Lab Med* 2007; 131:18–43.
- 11 Kiernan JA. *Histological and Histochemical Methods: Theory and Practice*. New York: Pergamon Press. 1981.
- 12 Sheehan DC. and Hrapchak BB. *Theory and Practice of Histotechnology*. St. Louis: C.V. Mosby Co. 1980.
- 13 Clinical Laboratory Improvement Amendments of 1988, Final Rule 57 FR 7163 February 28, 1992.
- 14 O'Leary TJ, Edmonds P, Floyd AD, Mesa-Tejada R, Robinowitz M, Takes PA, Taylor CR. Quality assurance for immunocytochemistry; Proposed guideline. MM4-P. National Committee for Clinical Laboratory Standards (NCCLS). Wayne, PA. 1997;1-46.
- 15 Battifora H. Diagnostic uses of antibodies to keratins: a review and immunohistochemical comparison of seven monoclonal and three polyclonal antibodies. *Progress in Surg Path* 6:1-15. eds. Fenoglio-Preiser C, Wolff CM, Rilke F. Field & Wood, Inc., Philadelphia.
- 16 College of American Pathologists (CAP) Certification Program for Immunohistochemistry. Northfield IL. <http://www.cap.org>
- 17 Wilkinson DG. The theory and practice of in situ hybridisation. In: Wilkinson DG. (ed.) *In Situ Hybridization A practical approach*. 2nd Edition. New York: Oxford University Press, 1998, pp.18-20.
- 18 Nadji M, Morales AR. Immunoperoxidase, part I: the techniques and pitfalls. *Lab Med* 1983; 14:767.
- 19 Omata M, Liew CT, Ashcavai M, and Peters RL. Nonimmunologic binding of horseradish peroxidase to hepatitis B surface antigen: a possible source of error in immunohistochemistry. *Am J Clin Path* 1980;73:626.
- 20 Wilkinson DG. *In situ hybridization: A practical approach*. 2nd Edition. Oxford University Press, Oxford. 1998.
- 21 Weiss LM, Chen Y. Effects of different fixatives on detection of nucleic acids from paraffin-embedded tissues by in situ hybridization using oligonucleotide probes. *The Journal of Histochemistry and Cytochemistry*. 1991;39(9):1237-1242.
- 22 Pontius CA, Murphy KA, Novis DA and Hansen AJ. *CLIA Compliance Handbook: The Essential Guide for the Clinical Laboratory*. 2nd Edition. Washington G-2 Reports, New York. 2003.

15 System Management (on BOND RX Controller)

15.1 BOND System Manager


15.1.1 Overview

The BOND System Manager is a utility that allows you to easily view the current status of the primary software services used by the BOND RX system, allows you to stop and start individual services, such as the Print Spooler, or stop and start all services.



WARNING: Do not stop any of the services, as the BOND RX system will no longer operate correctly.

However, you may be asked by customer support to stop and then restart one or more services, as part of a system troubleshooting process.

To open the BOND System Manager, locate the BOND System Manager icon  in the Windows notification area and then click the icon.



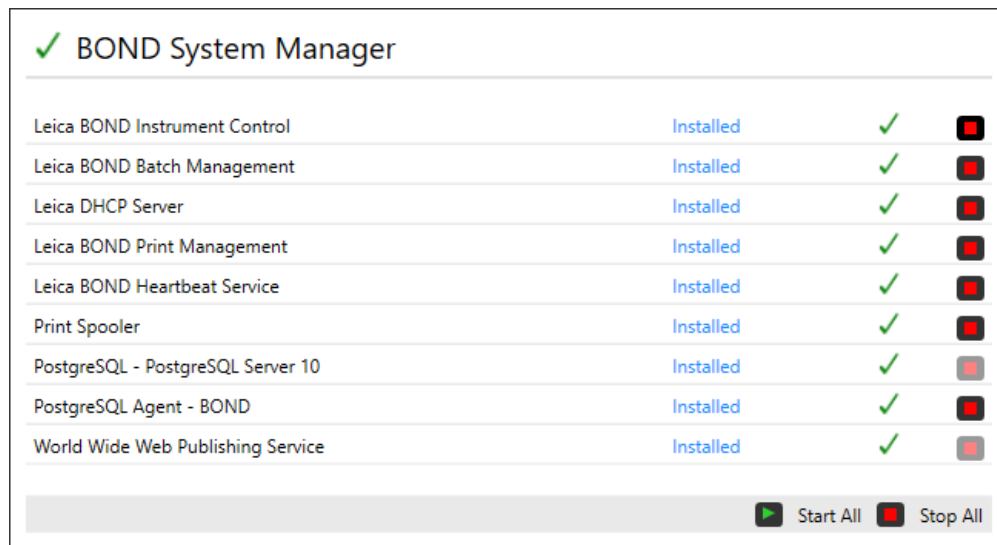
The icon may be hidden from view; if so, click on the small up arrow to see it.


If a BOND RX System error occurs, a notification message will appear; you can click on the message to hide it.





To hide the BOND System Manager window, click the icon in the Windows notification area again.

15.1.2 BOND System Manager Window



Figure 15-1: The BOND System Manager window



If there is a BOND RX System error, the BOND System Manager icon  updates to indicate the type of error that has occurred:

-  one or more services have stopped ( also appears at the top-left of the BOND System Manager screen)
-  unable to connect to the BOND RX system ( also appears at the top-left of the BOND System Manager screen)

In a BOND RX-ADVANCE installation this most likely means that:

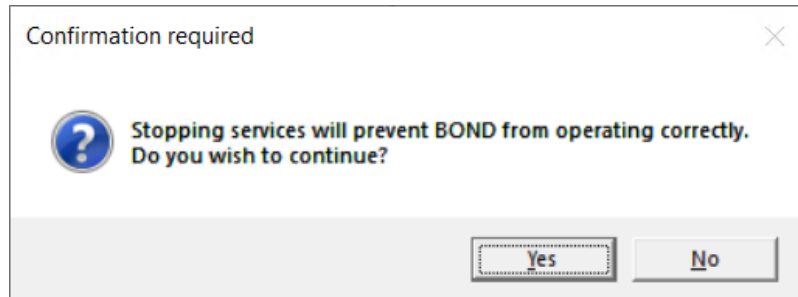
- The controller has been turned off; or
 - The terminal network has been disconnected; or
 - The terminal network switch has been turned off.
-  BOND System Manager is unavailable ( also appears at the top-left of the BOND System Manager screen)

15.1.3 Stopping Services

To stop an individual service, click the red stop button to the far right of the service's name. Or, to stop all services, click the **Stop All** button below the list of services.

A pop-up dialog appears, asking you to confirm that you wish to stop services. Click **Yes** to continue, or **No** to cancel.

Figure 15-2: Confirmation required dialog



Some services cannot be stopped (the PostgreSQL - PostgreSQL Server and World Wide Web Publishing Service), because the BOND System Manager relies on them to function; their stop buttons are therefore disabled.

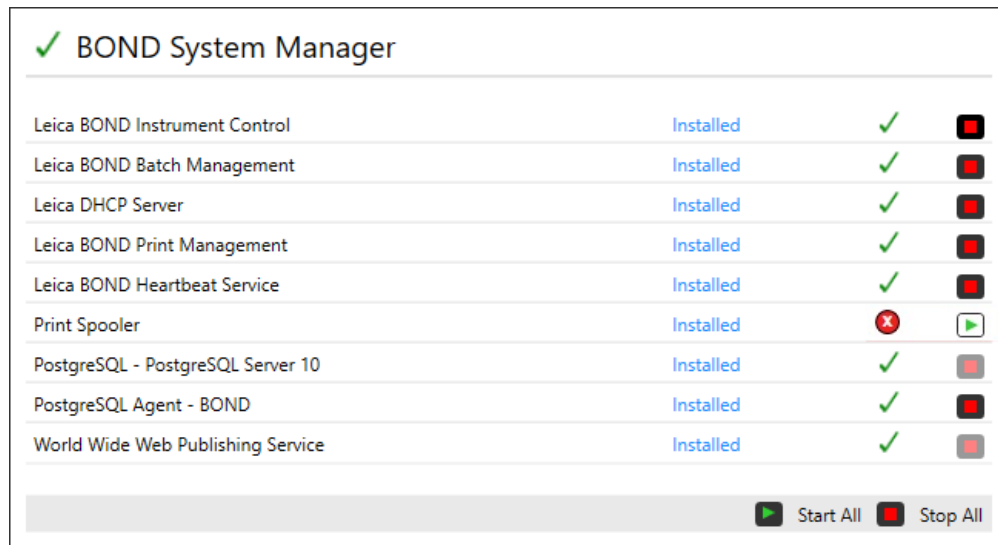
15.1.4 Starting Services



In most cases, when a service is stopped the BOND RX software will automatically restart that service within a few minutes.

If the BOND RX system is not operating as expected and you discover that one or more services are stopped, you can use the BOND System Manager to start the stopped service(s).

To start an individual service, click the green start button to the far right of the service's name. Or, to start all services, click the **Start All** button below the list of services.






Figure 15-3: BOND System Manager showing warning triangle (Print Spooler service stopped)

✓ BOND System Manager			
Leica BOND Instrument Control	Installed	✓	■
Leica BOND Batch Management	Installed	✓	■
Leica DHCP Server	Installed	✓	■
Leica BOND Print Management	Installed	✓	■
Leica BOND Heartbeat Service	Installed	✓	■
Print Spooler	Installed	✗	⚠
PostgreSQL - PostgreSQL Server 10	Installed	✓	■
PostgreSQL Agent - BOND	Installed	✓	■
World Wide Web Publishing Service	Installed	✓	■

▶ Start All ■ Stop All

15.2 Hard Disk Redundancy

All BOND RX controllers and terminals include hard disk redundancy, to protect the BOND RX system in the event of a hard disk failure. This protection system continuously monitors the system's hard disks, and an icon in the Windows notification area shows the current status.

Icon	Indicates
	Normal - the hard disks are working properly.
	Warning - there is a problem with the system's hard disks. Contact customer support.
	Error - a hard disk failure has occurred. Contact customer support.
	<p>Busy - this can appear when the hard disks are being verified, for example after an unexpected shutdown. The controller or terminal may run slowly during verification, which will usually take 2 to 3 hours to complete. The BOND RX system may be unusable during this period.</p> <p>After verification, the icon should return to its Normal status and normal hard disk operations will resume. However, if the icon indicates a Warning or Error status, contact customer support.</p>
	<p>Service not running - the software service used to monitor the hard disk protection is not running. The icon initially shows this status when the controller or terminal is started. Contact customer support if the icon does not indicate Normal status after several minutes have elapsed.</p>

16 BOND RX-ADVANCE Operations

16.1 Restarting the BOND RX-ADVANCE System



You should carry out this procedure only if:

- you have been instructed to do so by Leica Biosystems customer support, or
- you are preparing for a planned power outage.

Use the following method to restart the entire BOND RX system:

- 1 Ensure that all processing modules are idle (that is, no slide trays are locked).
- 2 Power off **all** processing modules.
- 3 Power off **all** terminals (click **Start** > **Shut down**).
- 4 Power off the secondary controller (if present) by briefly pressing the power button (see below for example).
- 5 Power off the primary controller by briefly pressing the power button (see [Figure 16-1](#)).



The power button could be located behind the controller's removable front cover, which may be locked. In this case, you must first obtain the key from the designated key holder.

Watch the dashboard screen while shutting down, as a second press of the power button may be required if the shutdown process stops at the Windows login screen. If this occurs, wait at least 90 seconds and then briefly press the power button again.

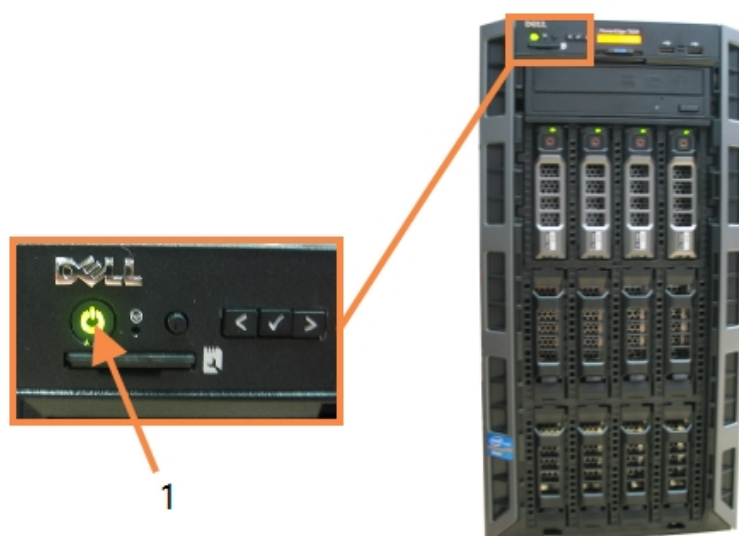


When you press the power button again, the controller will start to shut down. **Do not** hold for longer than 2 seconds as this may cause a "hard" reset and instantly turn off the controller. It may take up to 45 seconds for the controller to power off (power-button light turns off).

- 6 Wait for 2 minutes and then power on the primary controller.
If a "Shutdown event tracker" window appears, close it by selecting **Cancel** or by pressing the <Esc> key.
- 7 Wait for 30 seconds and then power on the secondary controller (if present).
- 8 After the controllers are fully restarted, power on all terminals.

- 9 Power on all processing modules.
- 10 Log on to each terminal.

Figure 16-1: Power button location on controller front panel (shown with cover removed)



Legend

- 1 Power Button

16.2 Switching to the Secondary Controller



These instructions apply only to BOND RX-ADVANCE systems that include a secondary (backup) controller. You should carry out this procedure only if:

- you have been instructed to do so by Leica Biosystems customer support, or
- the primary controller is not operational.

The secondary controller will then operate in standalone mode, and your system will no longer have a redundant backup capability. However, after you complete this procedure, the BOND system will continue processing as normal.

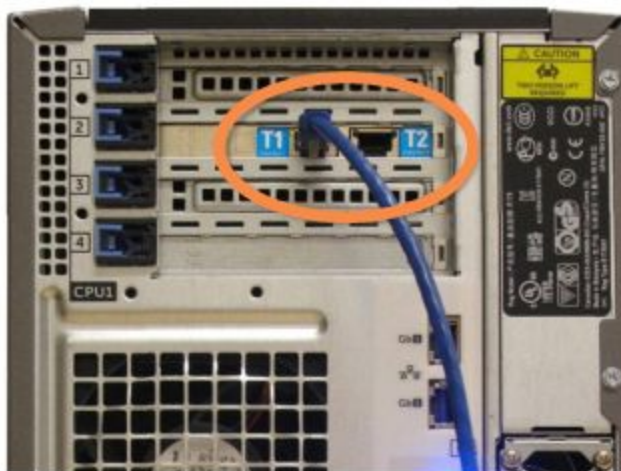


During the switchover process, data from the last 5 minutes' processing may be lost. Also, any LIS messages that were sent during the switchover process may be lost. Therefore, after the switchover has succeeded, check if any slides are missing. If this is the case, resend the slide data via the LIS or manually create the missing slides in BOND RX.

- 1 Close all instances of research and administration clients on all BOND RX-ADVANCE terminals.
- 2 Disconnect the Terminal network cable from the port labeled **T1** or **T2** on the primary controller, and then reconnect the cable to the same port on the secondary controller.

See [Figure 16-2](#).

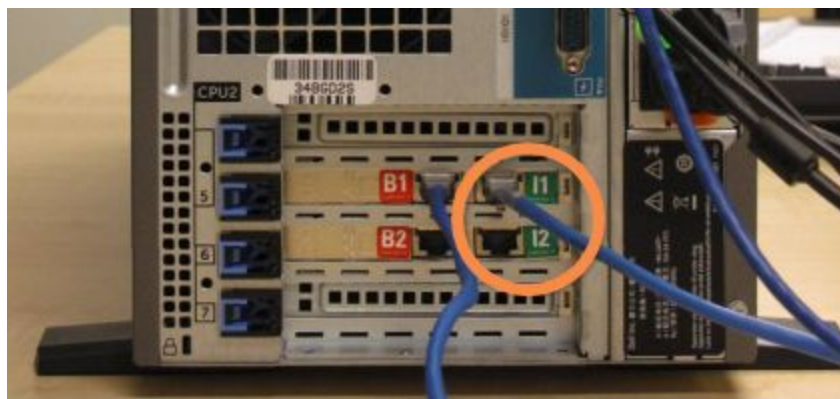
Figure 16-2: Controller Terminal Ports



- 3 Disconnect the processing module's network cable from the port labeled **I1** or **I2** on the primary controller, and then reconnect the cable to the same port on the secondary controller.

See [Figure 16-3](#).

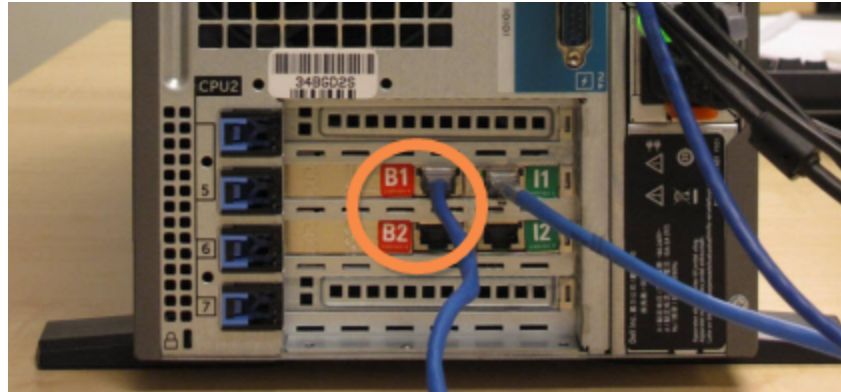
Figure 16-3: Controller Processing Module Ports



- 4 Disconnect the Bridge network cable from port **B1** or **B2** on the primary controller.

See [Figure 16-4](#).

Figure 16-4: Controller Bridge Ports



- 5 If there is an Ethernet cable (used for LIS connectivity) in port **Gb(1)** or **Gb(2)** on the primary controller, disconnect and then reconnect this cable to the same port on the secondary controller.

See [Figure 16-5](#).

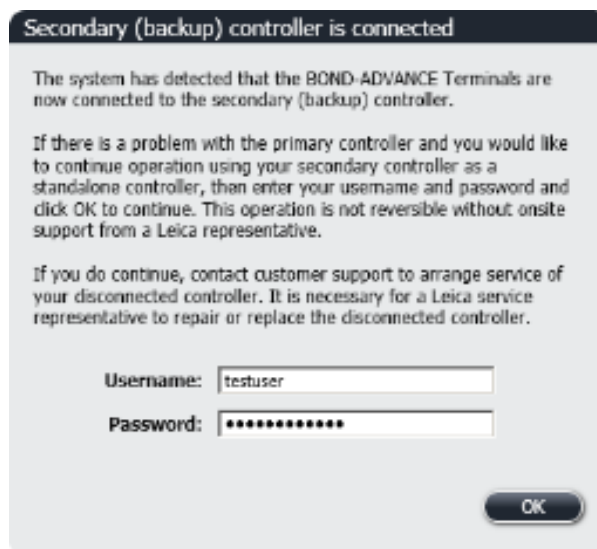
Figure 16-5: Ethernet ports used for LIS connectivity



The BOND RX-ADVANCE system detects that you have connected the network cables to the secondary controller, and displays a confirmation dialog on all terminals.

See [Figure 16-6](#).

Figure 16-6: Dialog - secondary (backup) controller connected



The switchover is not reversible without on-site support from a Leica Biosystems representative.

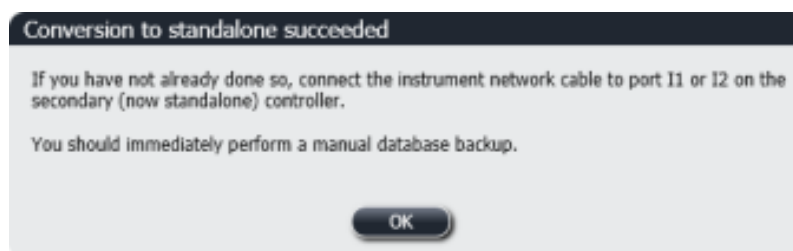
- 6 To confirm that you want to continue with the switchover:
 - a Enter your Username and Password in the fields provided.
 - b Click **OK** to confirm.



If another user chooses to continue with the switchover before you do, the above dialog will disappear.

- 7 After confirming the switchover, power off the primary controller.
- 8 Wait until the system prompts that the conversion to standalone has succeeded (see [Figure 16-7](#)), then restart the research client and log on to the system as normal.

Figure 16-7: Dialog - conversion to standalone succeeded



- 9 Immediately open the administration client and perform a manual database backup. See [10.5.1 Laboratory Settings](#).

After switchover to the secondary controller is complete, the status of all slides and processing modules should automatically update to reflect the latest system status. However, if any runs completed while the processing modules were disconnected from the controller, the run status will still appear as **In Progress**. In this case you must unlock the affected slide tray in order to update the status for the slide staining assembly.



Contact customer support to arrange service of your disconnected controller. It is necessary for a Leica Biosystems service representative to repair or replace the disconnected controller.

17 Slide Label Printer Replacement

17.1 Replace a Cognitive Cxi Printer on a Single Seat System

Use the following procedure to replace a Cognitive printer with a new Cognitive printer.

- 1 Turn off the power switch on the side of the old printer.
- 2 Disconnect the USB cable and the power supply cable from the rear of the old printer.
- 3 Connect the USB cable and the power supply cable to the new printer.
- 4 Turn on the power switch at the side of the new printer.

The BOND RX Controller screen displays a message in the notification area (bottom right) of the desktop that the printer has been found.

- 5 Navigate to: **Windows Start > Devices and Printers** and find the newly added printer.
- 6 Right-click this printer and select **Properties**, then copy the printer's name.
- 7 Open the Administration client, **Hardware configuration** screen, **Slide labelers** tab as described in [10.6.3 Slide Labelers](#). Select the old printer you have replaced.
- 8 Paste (overwriting the existing name) into the **Printer name** field so that it becomes, for example, "Cognitive Cxi 2 inch 300 DPI TT (Copy 1)".
- 9 Click **Save**.
- 10 Print a test label to confirm the operation of the printer.

17.2 Replace a Cognitive Cxi Printer on BOND RX-ADVANCE System

It is necessary to set the Static IP address of the new printer to the same value as the old printer before connecting the new printer to the BOND RX-ADVANCE system.

The IP addresses for the printers start from 192.168.5.101. Only the last digit is different for each printer. For example, the printer IP address for printer 2 is 192.168.5.102.

The procedures below explain how to find out the Static IP address of the old printer and how to set that value in the new printer.

Cognitive Printer Front Panel

Figure 17-1 shows the keypad and LCD display on the Cognitive Cxi printer.

Figure 17-1: Cognitive Printer LCD display and keypad














Read IP Address of Old Printer

Perform the following procedure on the old printer to discover the IP Address to use with the new printer:



If you cannot use the display on the old printer for any reason, use the procedure [Find Printer IP Address](#) to find the IP address on the Controller.

- 1 Press .
- The screen displays **Main Menu: Language Menu**.
- 2 Press  to display **Printer Setup** option.
- 3 Press  to display **Printer Setup: Comm. Menu**.
- 4 Press  to display **Comm. Menu: Timeout**.
- 5 Press  twice, to display **Ethernet**.










- 6 Press .
The screen displays **Ethernet - DHCP**.
- 7 Press .
The screen displays **DHCP Off**. (If it displays **DHCP On**, press  to change the value.)
- 8 Press .
The screen displays the message: **Value has been set**.
- 9 Press  to display **Set Static IP**.
- 10 Press  to display the current setting.
- 11 Make a note of the Static IP Address.
- 12 Switch off the power to this printer and disconnect it from the power supply and from the network.




Set Printer IP Address

Perform the procedure below to set the new printer to the correct Static IP Address.



CAUTION: Do not connect the new printer to the BOND network until you have performed the procedure below.

- 1 Connect the new printer to the power supply and turn on the power switch at the side of the printer.
- 2 Press .
The screen displays **Main Menu: Language Menu**.
- 3 Press  to display **Printer Setup** option.
- 4 Press  to display **Printer Setup: Comm. Menu**.
- 5 Press  to display **Comm. Menu: Timeout**.
- 6 Press  twice, to display **Ethernet**.
- 7 Press .
The screen displays **Ethernet - DHCP**.
- 8 Press .
The screen displays **DHCP Off**. (If it displays **DHCP On**, press  to change the value.)
- 9 Press .
The screen displays the message: **Value has been set**.

- 10 Press  to display **Set Static IP**.
- 11 Press  to display the current setting.
- 12 Enter the IP Address that you noted down from the old printer. Use the left and right buttons to move the cursor left or right, and use the up and down buttons to change the value.
- 13 Press .


The screen displays the message: **Value has been set**.
- 14 Press  several times to return to the main -- **COGNITIVE** -- screen.
- 15 Press the power switch on the side of the printer to the OFF position. Then switch it back to the ON position.
- 16 Connect the Ethernet cable to the new printer to connect it to the BOND network.

Figure 17-2: Ethernet connector



- 17 Open the Administration client and print a test label.

Find Printer IP Address

If it is not possible to read the IP address on the old printer, use the following procedure to determine the IP Address for the new printer.


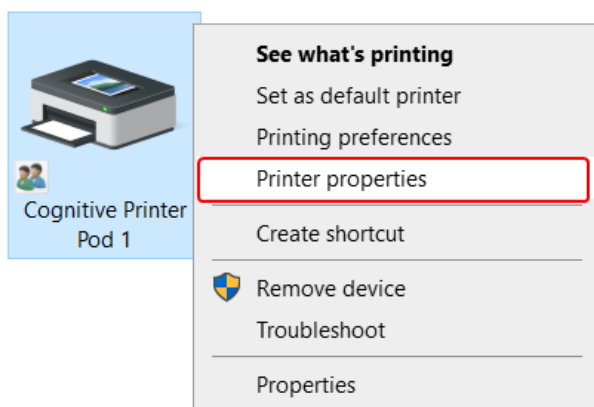
- 1 Log on to the BOND RX-ADVANCE Controller as BONDDashboard.
- 2 Press the Windows Logo key  + **M** to minimize the dashboard screen.
- 3 On the Windows taskbar, click the **Start** button and select **Devices and Printers**.
- 4 Right-click on the relevant Cognitive printer icon and select **Printer Properties** from the pop-up menu as shown in [Figure 17-3](#).

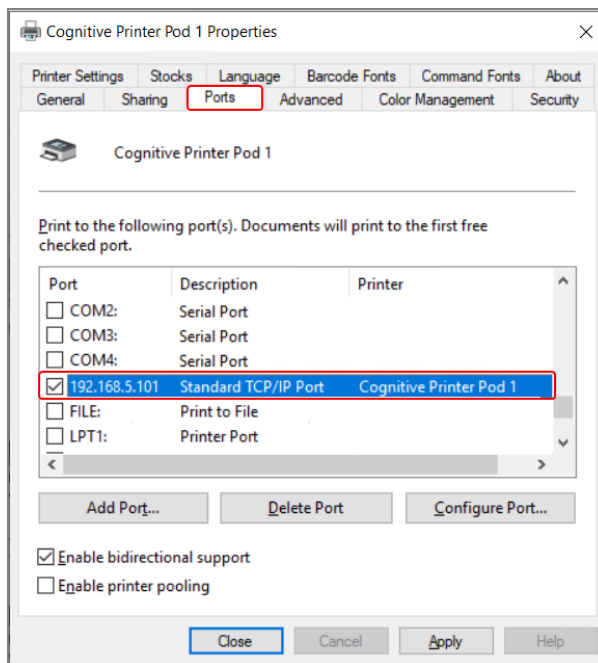
Figure 17-3: Select Printer Properties



The system displays the **Properties** dialog box.

- 5 Select the **Ports** tab.

Figure 17-4: Printer Properties - Ports tab



- 6 Make a note of the IP address in the **Port** column for the selected printer. (You may need to widen the **Port** column by dragging the column border.)
- 7 Click **Cancel** to close the dialog box.
- 8 Close the **Devices and Printers** window.
- 9 Press **Alt+Tab** to display the BOND Dashboard.
- 10 Use the IP Address from Step 6 to perform the procedure at [Set Printer IP Address](#).

17.3 Replace a Zebra Printer with a Cognitive Cxi Printer on a Single-seat System

Use the following procedure to replace a Zebra TLP 3842 or GX430t printer with a Cognitive Cxi printer.



If the Zebra printer was connected by a “parallel” cable, you can disconnect it from the BOND RX Controller. You require a USB cable to connect the Cognitive printer to the BOND RX Controller.

- 1 Turn off the power switch on the rear of the Zebra printer.
- 2 Disconnect the parallel or USB cable and the power supply cable from the rear of the printer.
- 3 Disconnect the Zebra printer power supply from the mains supply.
- 4 Connect the Cognitive printer power supply to the mains supply.
- 5 Connect the USB cable and the power supply cable to the Cognitive printer.
- 6 Turn on the power switch at the side of the printer.

The BOND Controller screen displays a message in the notification area (bottom right) of the desktop that the printer has been found.
- 7 On the Windows taskbar, click the **Start** button and select **Devices and Printers**.
- 8 Confirm that the printer appears as “Cognitive Cxi 2 inch 300 DPI TT”.
- 9 Log in to the BOND Administration client.
- 10 Open the Hardware screen, Slide labelers tab.
- 11 Click **Add printer** (bottom left of screen).
- 12 In the right panel on the screen, enter:
 - **Display name:** use the printer name: Cognitive Cxi 2 inch 300 DPI TT
 - **Printer name:** the same name again
 - **Host name:** leave this field blank.
 - **Printer type:** select the printer model: Cognitive Cxi
- 13 Click **Save**.
- 14 Right-click on the Zebra printer in the list.
- 15 Select **Delete** from the pop-up option.
- 16 The system displays the message: “Are you sure you want to delete the printer?”
- 17 Click **Yes**.

18 Specifications

- [18.1 System Specifications](#)
- [18.2 Physical Specifications](#)
- [18.3 Electrical Power and UPS Requirements](#)
- [18.4 Environmental Specifications](#)
- [18.5 Operating Specifications](#)
- [18.6 Microscope Slides](#)
- [18.7 Transport and Storage](#)

18.1 System Specifications

Network connection requirements	Ethernet IEEE802.3, 10/100/1000BASE-T
Maximum number of BOND RX and BOND RX ^m processing modules	5 (multiple processing modules require an Ethernet switch)
Network cables	CAT5e or CAT6 shielded cables, with RJ-45 connectors
Ethernet switch requirements: Single-seat	Ethernet IEEE802.3, 10/100/1000BASE-T 8-port Ethernet switch (supports up to 5 processing modules)
BOND RX-ADVANCE	8- or 16-port Ethernet switches (can be connected together to support up to 30 processing modules)
Device specifications	BOND RX controllers and terminals must be supplied by Leica Biosystems

18.2 Physical Specifications

	BOND RX	BOND RX ^m
Dimensions	W – 790 mm (31.10 in) H – 1378 mm (54.25 in) D – 826 mm (32.4 in)	W – 760 mm (29.9 in) H – 703 mm (27.6 in) D – 800 mm (31.49 in)
Weight (dry)	238 kg (525 lb.)	120 kg (265 lb.)
Clearance requirements	600 mm (24 in) above 0 mm left 150 mm (6 in) right 0 mm at back, however users must be able to disconnect the mains power cable without moving the processing module.	
Maximum distance to external bulk waste container (BOND RX ^m only)	~	1 meter (40 in)

18.3 Electrical Power and UPS Requirements

	BOND RX	BOND RX ^m
Operating voltage	90 V to 264 V (for nominal voltage 100 V to 240 V)	
Mains frequency	50/60 Hz	50/60 Hz
Power consumption	1200 VA	1000 VA

18.4 Environmental Specifications

	BOND RX	BOND RX ^m
Maximum operating temperature	35 °C (95 °F)	35 °C (95 °F)
Minimum operating temperature	5 °C (41 °F)	5 °C (41 °F)
Temperature required to meet staining performance requirements	18–26 °C (64–79 °F)	18–26 °C (64–79 °F)
Operating humidity (non condensing)	30 to 80% RH	30 to 80% RH
Maximum operating altitude	0 to 1600 m (5250 ft.) above sea level	0 to 1600 m (5250 ft.) above sea level

	BOND RX	BOND RX ^m
Sound pressure level output (at 1 m)	< 85 dBA maximum	< 85 dBA maximum
	< 65 dBA normal operation	< 65 dBA normal operation
Maximum heating energy output	1200 VA	1000 VA

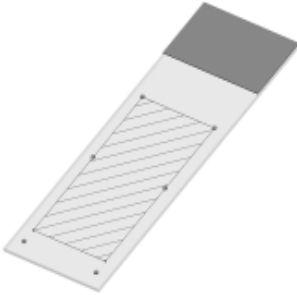
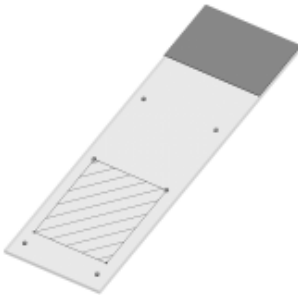
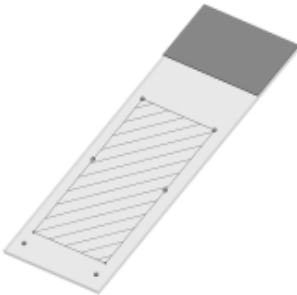
18.5 Operating Specifications

	BOND RX	BOND RX ^m
Slide capacity	30 at a time. Finished trays (10 slides) may be replaced continuously.	
Reagent container capacity	7 mL and 30 mL	7 mL and 30 mL
Reagent container dead volume	555 µL (7 mL) and 1618 µL (30 mL)	
Reagent container reserve volume	0 µL (7 mL) and 0 µL (30 mL)	
Titration container capacity	6 mL	6 mL
Titration container dead volume	300 µL	300 µL
Titration container reserve volume	0 µL	0 µL
Number of reagent containers	36	36
Bulk reagent container capacity	2 L or 5 L	1 L or 2 L
Hazardous waste container capacity	5 L	2 L
Standard waste container capacity	2 x 5 L	~
External bulk waste container capacity	~	9 L
Chemical compatibility	All BOND reagents 70% alcohol solution (for cleaning purposes)	
Temperature indication	Defaults (these can be changed by service representatives): Warm: 35 °C, Hot: 80 °C	
Maximum permitted pressure for gas and liquid connections	1.0 bar	2.5 bar
Service life	7 years	7 years
BOND RX Cybersecurity certificate expiry	10 years	10 years

18.6 Microscope Slides

Dimensions	Width: 24.64–26.0 mm (0.97–1.02 in) Length: 74.9–76.0 mm (2.95–2.99 in) Thickness: 0.8–1.3 mm (0.03–0.05 in)
Label area	Width: 24.64–26.0 mm (0.97–1.02 in) Length: 16.9–21.0 mm (0.67–0.83 in)
Material	Glass, ISO 8037/1
Usable area	Refer to the following diagrams. The dispense volume refers to the settings you can choose when setting up slides using the BOND RX software (see 6.3 Working with Studies).

Figure 18-1: The usable areas of slides for the BOND Processing Modules

	100 μ L	150 μ L
BOND RX		
BOND RX ^m		

18.7 Transport and Storage

Storage temperature	-20 to +55 °C (-4 to +131 °F)
Storage humidity (noncondensing)	< 80% RH
Shipping methods	Road, air and sea freight compatible.

Please note the above information is for packaged processing modules only.

Refer to [18.4 Environmental Specifications](#) for unpacked processing modules.

Index

A

About BOND RX, dialog.....	82
access level, see user roles.....	74, 216
adding	
panel.....	142
reagent.....	182
slide.....	138
study.....	132
administration client.....	215
administrator, user role	
description.....	74
setting.....	216
alarms.....	80
architecture of BOND RX system.....	69
aspirating probe	
cleaning.....	282
description.....	52
assay verification.....	311
audit trail.....	229
automatic slide identification.....	114

B

back cover, description.....	58
back up database.....	233
baking.....	310
barcode scanner, handheld	
description.....	61
barcode scanner, see handheld barcode scanner.....	190
BOND-ADVANCE, description.....	70
BOND Aspirating Probe Cleaning System.....	282

BOND Polymer Refine Detection System.....	306
BOND RX.....	335
BOND RX controller.....	69
BOND Streptavidin-Biotin Detection System.....	307
BOND system.....	34
BOND System Manager.....	318
Bulk container lighting system.....	49
bulk containers.....	47
cleaning & maintenance.....	267
disabling.....	236
status.....	107
bulk fluid probes	
cleaning.....	288
bulk fluid robots, description.....	54

C

cautions.....	7
CE marking.....	9
CISPR 11 (EN 55011).....	9
classification of equipment.....	9
clean fluidics, maintenance procedure.....	281
cleaning.....	262
cleaning schedule.....	264
compatibility	
bulk reagents with ancillary reagents.....	183
slides.....	151
configure BOND RX system.....	215
contact Leica Biosystems.....	2
controller, see BOND RX controller.....	69
controls	
negative reagent for IHC.....	312
reagent for ISH.....	313

tissue.....	312
working with.....	129
copyright.....	1
covers	
cleaning.....	284
Covertiles.....	64
cleaning & maintenance.....	275

D

daily study option.....	135
dashboard.....	78
data fields, LIS slide.....	219
database.....	84
backup.....	233
database update.....	228
dead volume.....	53
delayed start.....	122
deleting	
pod.....	238
reagent.....	184
slide.....	141
study.....	134
detection systems	
BOND Polymer Refine.....	306
BOND Polymer Refine Red.....	307
BOND Streptavidin-Biotin.....	307
BOND, overview.....	306
description.....	66
inventory report.....	193
registration.....	189
dewaxing.....	145, 310
dip test.....	186, 197
disable bulk containers.....	236
dispense type.....	171
dispense volume.....	142
drip trays.....	285
bulk container.....	285
processing module tray.....	287
duplicate study ID	
BOND RX studies.....	133
LIS studies.....	219

E

electrical hazards.....	7
empty, mark reagent package as.....	189
epitope retrieval.....	145
expired study.....	133
export reports.....	81
export slide data.....	212

F

facility, set.....	231
FCC.....	9
finishing a run.....	96
first steps.....	30
front cover.....	44
function bar.....	75
fuses.....	292

G

glass slides	
specifications.....	338
glossary of symbols.....	11

H

handheld barcode scanner	
description.....	61
register detection system.....	190

hardware configuration	234	installation and transportation hazards	7
hardware status	100	installation hazards	7
hazard		instructions for equipment for research use only	9
chemical	5	instrument operation hazards	4
electrical	7	intended use statement	9
installation and transportation	7	inventory screen, reagents	184
mechanical	6	ISH, principle of	306
operational	8	ISO 15223-1	11
hazardous waste	183	ISO 7000	12
hazards		ISO 7010	16
electrical	7		
installation	7	L	
instrument operation	4	label	
operational	8	and LIS[label	
reagent	8	LIS]	260
heater errors	101	configuration	220
heaters	43	information types	224
Help	81	overview	143
accessing	29	printing	144
		Quick start	91
I		label ID	146
		labeler, slide	62
ID imager	42	laboratory configuration	230
cleaning & maintenance	285	layout, label	220
ID scanner, handheld		legal notices	1
registering reagents	191	license, LIS-ip	219
identification, product	1	lid	41
identifying slides		cleaning	284
automatic	114	lifetime, study	133
manual on-board	114	LIS integration package	252
manually	141	configuration in BOND RX	218
IEC 60417	13	connection & initialization	257
IHC, principle of	305	errors	257
important information for all users	1	get LIS data	256
impromptu slide and study creation	147	license	219
incompatible slides	113		

LIS properties.....	256
priority slides.....	256
public marker names.....	255
slide labels.....	260
slides.....	255
status panel.....	254
studies.....	254
study & slide data.....	258
terminology.....	253
LIS slide data fields.....	219
LLS (liquid level sensing).....	186
loading slides.....	91
log, service.....	83

M

maintenance.....	262
Maintenance report.....	125
maintenance schedule.....	264
Maintenance screen.....	124
maintenance, preventative.....	263
manual slide identification.....	141
manufacturer.....	1
materials required.....	308
mechanical hazards.....	6
minimum stock setting.....	188
mixing station.....	53
multi-seat installation.....	70
Multiplex staining.....	155

N

notifications.....	80
--------------------	----

O

open containers.....	66
refill.....	189
operational hazards.....	8
operator, user role	
description.....	74
setting.....	216

P

panel	
adding.....	142
create.....	202
editing.....	203
screen.....	201
parallel multiplex staining.....	155
password, BOND RX.....	217
PDF, reports.....	81
pod	
description.....	69-70
management.....	237
power supply fuses.....	292
power switch.....	57
preventative maintenance.....	263
print reports.....	81
print slide labels.....	144
printer	
slide labeler.....	62
priority slides, LIS.....	256
processed study lifetime.....	133
processing module	
cleaning & maintenance.....	262
configuration.....	234
description.....	35

initialization.....	41	identification.....	179
restart.....	281	inventory report.....	193
specifications.....	336	inventory screen.....	184
states of.....	100	loading.....	92
tabs.....	99	management.....	176
transport & storage.....	338	manual identification.....	192
processing module operation.....	4	panels screen.....	201
product identification.....	1	refill open container.....	189
protocol.....	154	registration.....	189
editing.....	163, 228	setup screen.....	180
editing for multiplex stain.....	158	substitution.....	180
finishing a run.....	96	usage report.....	194
list.....	159, 218	reagent hazards.....	8
list of predefined protocols.....	172	reagent status.....	102
preparation.....	174	reagent trays	
prestaining.....	174	description.....	65
reports.....	171	red, detection system.....	307
running.....	95	red, highlight in Reagent inventory screen.....	188
running overview.....	31	refill open container.....	189
setup screen.....	154	registering reagents and detection systems.....	189
staining.....	172	regulatory notices.....	9
status screen.....	123	regulatory symbols.....	11
viewing.....	160	Replace a Cognitive Cxi Printer.....	329-330
public marker names.....	255	Replace a Zebra Printer.....	334
		reports.....	81
Q		brief slide history.....	214
quality control.....	311	export slides.....	212
benefits of.....	314	protocol.....	171
Quick start.....	85	reagent usage.....	194
		run details.....	208
R		run events.....	208
reagent.....	176	slide processing summary.....	211
adding/editing.....	182	slide setup.....	146
deleting.....	184	study.....	210
determining volume.....	186	system.....	82
empty package.....	189	researchers list.....	136
fixing problems.....	105	restore database.....	233

processing module	336	symbols	
staining		safety	16
interpretation of	314	symbols and markings	14
staining methods	155	symbols glossary	11
staining mode	138, 155	syringe	55
starting a run	120	cleaning & maintenance	289
delayed start	122	system	
status screens	97	architecture	69
bulk containers	107	description	34
hardware status	100	report	82
LIS	254	specifications	335
protocol	123	status screen	98
reagent status	102	system startup	
slide status	111	checks	86
system	98		
stopping a run	120	T	
studies		tables, sorting	77
adding	132	tabs, processing module, on System status screen	99
copying	134	temperature, indication of	102
default settings	232	template, label	220
deleting	134	tissue preparation	309
duplication	133	titration containers	66
editing	134	titration kit	309
entering details, Quick start	88	top plate, replacing	278
expiration	133	trademarks	1
identification	131	transport	338
impromptu creation	147		
LIS	254	U	
resurrection	133	update database data	228
study & slide default settings	232	updating the software	84
study ID		UPI	179
LIS, duplicate	219	user roles	74
vs. study number	131	setting	216
substituting reagents	180	username	217
supervisor, user role			
description	74		
setting	216		

users, create and edit 216

W

warnings 4, 80

wash block 53

waste container

 cleaning & maintenance 273

 description 67

 status 107

work cell 70

work flow

 daily study option 135

 impromptu slide and study creation 147