LabSat® Short Guide

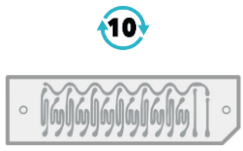
1. Introduction

This document summarizes the steps to be performed to safely produce quality stainings using the LabSat® instrument. Due to the short nature of this document, refer to the user manual (UM) when in doubt (version 12 or later). A troubleshooting section is available as well in the user manual.

1.1 Safety

- **Do not open/touch the Distribution System** when the instrument is powered unless specifically instructed by the software to do so.
- **Do not touch/open the stainer, Distribution System, reservoirs, or waste bottle** during an ongoing process (staining protocol, calibration, wash etc..).
- **Never exchange the waste bottle** with a regular laboratory bottle: the supplied bottle is a special pressure resistant bottle. Contact Customer Support if you need to replace it.

1.2 System

Consumable Chips	
 **Distribution Chip** is changed every day and when the counter reaches zero (starts at 10).	 The **Staining Chip** is single use.

Remember to:	
	Always check the volumes in reservoirs and make sure they are tightly clipped/screwed before starting protocols.
	Run a wash every day after use.
 4 max.	Change the buffer solutions and refill to the maximum volume after four stainings or if bubbles form in the reservoirs.
	Do not execute protocols with less than 10mL of reagent or buffer in the large reservoirs and do not tilt the reservoirs, especially when full.

2. Standard Operating Overview

	START
1	Start the computer. If you are equipped with a compressor, start it as well and **wait until it reaches at least 5 bars**.
2	Start LabSat® using the button at the back of the instrument (see UM, Product Specifications Section), the LED should be red (not blinking).
3	Start LabSat Research software by double-clicking on the icon (see UM, Software Operation Section), the LED should turn white (not blinking).
	PREPARATION
4	When turning on LabSat®, the reservoirs should contain the following: • Small reservoirs: DIW • Large reservoirs A, B and C: DIW • Large reservoir D: EtOH 70% (or DIW, if a Full Wash was executed before) If reagents are still loaded in the software, the Daily Wash was not performed after the last use. You will be prompted to run a Daily Wash.
5	When LabSat Research software opens, a pop-up message informs the user if the Distribution Chip should be changed. If so, click 'Change', then click on 'Start' to start the exchange procedure. Get a new Distribution and change it following the instructions on the screen (see UM, Distribution Chip Exchange section). Once the exchange procedure is finished, the system will initialize (calibrate). When it finishes successfully, LabSat® is ready to be used. Alternatively, this exchange procedure and/or the calibration only can be performed from the Home tab, by clicking 'Change' under the Distribution Chip icon.
6	Click on the Reagents tab (see UM, Reagents tab section): create new reagents if needed. **Do not update the volumes in the reservoirs yet.**
7	Click on Protocols tab, select a protocol. If necessary, create or edit an existing protocol. Add protocol to the Queue, by clicking 'Add to Queue' (see UM, Protocols tab section). The protocol steps and volumes required for each reagent can be displayed by clicking on the blue down arrow (priming volume not included).
8	Click on the Home tab and select the protocol to execute from the Queue (double-click on the protocol to run). It will be loaded to the Protocol Area for execution (see UM, Queue and Protocol area section). Verify that the correct protocol has been loaded. Follow the prompts in the 'Required Actions' to create any necessary reagents and buffers using the 'Create' button, allocate reagents and buffers to adequate reservoirs using the 'Add' button and empty the waste and click the 'Empty' button as indicated. Finally, consult the 'Fill' required actions to determine the volumes of reagents and buffers that must be prepared according to the selected reservoir allocation. When changing the allocation of a reagent from a small reservoir to a large reservoir, make sure that the volume prepared still matches the required volume. To remove or change the reagent or buffer allocated to a reservoir, go to the Reagents tab.

9	Prepare the volume of reagents and buffers instructed by the 'Required actions'. The volume can also be calculated as follows: **Volume to prepare = Priming Volume + (Protocol volume x number of slides)** Notes: • **Priming volume is 120 µL for small reservoirs and 500 µL for large reservoirs.** • For multi-part protocols, the volumes displayed may be greater than the reservoir capacity. These reagents will need to be refilled during the protocol pause steps. • Protocol volumes can be found in the Protocol tab by clicking the blue down arrow .
10	Load the reagents and buffers in LabSat® one by one according to the selected reservoir allocation, click on the corresponding 'Fill' buttons of the 'Required actions', and **insert the exact volume into the user interface next to each reservoir in case more volume is loaded** (see UM, Reservoir Management section). When all the reagents are loaded, the software indicates the total staining time. If a protocol pause is present, the time until the next user intervention is displayed as well.
	STAINING PROTOCOL
11	 Insert a Staining Chip in the stainer and close the handle (see UM, Staining Chip loading section). Always check the chip for damage before using it (see UM, Installation > Microfluidic chips section).
12	Load the slide with the sample to stain into the stainer (see UM, Slide loading and removal section).
13	Start the protocol immediately by clicking 'Start' (see UM, Queue and Protocol area section).
14	 When the protocol ends, a pop-up message will indicate the status of the protocol performed. Click 'Yes' to open the stainer (you can also use the 'Open' button under the Staining Chip icon to open it later). Wait for the piston to go down and physically open the stainer using the handle. Remove the chip first, then remove the slide (see UM, Slide loading and removal section). Coverslip the slide before visualizing it under a microscope to assess the staining result.
15	If the protocol status indicates failed steps or warnings, check the report in the History tab (see UM, History tab section).
16	To stain other samples with the same reagents and buffers, repeat steps 10-15 after selecting the protocol in the Queue.
	END OF THE DAY – DAILY WASH
17	When you are done using LabSat® for the day, a wash protocol must be run. If the 'Full Wash' counter is low or zero, perform a 'Full Wash' (see instructions below). Otherwise, run a 'Daily Wash' protocol. If you did not launch a wash before closing LabSat Research software, you will be reminded to do so. All the reservoirs except large reservoir D will be reset to DIW at the end of the 'Daily Wash'.
18	When the wash protocol is complete, close the software. Turn off the computer, turn off LabSat® with the power switch in the back and then turn off the compressor.
19	Put a clean paper towel in the stainer in place of the Staining Chip and close the Stainer (see UM, Shut-down procedure section).

3. Keeping LabSat® Clean

Failure to follow the instructions below can damage the instrument:

- Only use Lunaphore approved solutions when performing the 'Daily Wash' and 'Full Wash'.
- Never run the "Daily Wash" protocol without 70% Ethanol. Other concentrations will either result in ineffective washes or overfilling, which can damage the instrument.

FULL WASH

Perform a 'Full Wash' protocol after 24 stainings (check counter on Home tab) with Fluidics Cleaning Kit (product code: BU03).

Follow the instructions on the screen. Note that BU03 may still be called 'Full Wash' in the software and in the reagents database.

Small reservoirs can be empty or full and do not need to be changed before a 'Full Wash'. They can be removed and replaced with empty tubes to save the leftover reagents.

Prepare and load:

Reservoir	Content
Large reservoir B	1.8 mL Fluidics Cleaning Kit solution 1 1.8 mL Fluidics Cleaning Kit solution 2 14.4 mL DIW
Large reservoir C	18 mL Fluidics Cleaning Kit solution 3 (RTU)
Large reservoirs A and D	50 mL DIW

Load a Staining Chip and a dummy slide in the Stainer and execute the wash.
At the end of a wash protocol, the washed reservoirs contain DIW. The reservoirs can be changed for reagent and buffer loading. Before doing so, wipe the fluidic tube inside the large reservoirs with a paper towel with Ethanol 70%, and then with DIW (see UM, Full Wash section).

DAILY WASH

Small reservoirs can be empty or full and do not need to be changed before a 'Daily Wash'. They can be removed and replaced with empty tubes to save the leftover reagents. Load the large reservoirs as follows:

- Large reservoir A, B and C: 50 mL DIW
- Large reservoir D: 50 mL Ethanol 70%

Before launching the wash make sure that the lid of the stainer is closed and that no chip and no slide are loaded.

If liquids are spilled onto the instrument's parts, wipe them off with a paper towel with Ethanol 70% or isopropanol on it, and then with a paper towel with water on it.
On a daily basis, remove dirt and dust which has accumulated on the instrument and clean the surrounding working area (see UM, Daily Maintenance section).

EVERY WEEK

At least once a week, clean the inside of the large reservoirs' caps, and the small reservoir holders with paper towels with Ethanol 70% on them (see UM, Annual Maintenance section).

Note: If both a wash (Daily Wash or Full Wash) and a Distribution Chip exchange are necessary, first launch the wash protocol and then perform the chip exchange.