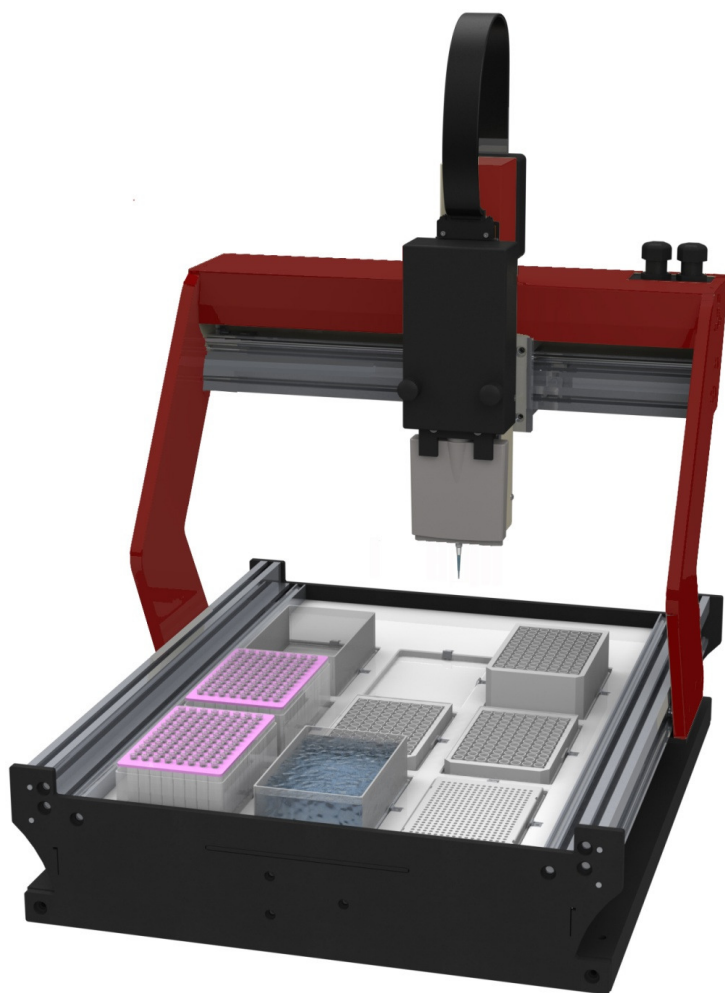


α Bot™

USER MANUAL





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FOREWORD

This document describes how to use the α Bot™ dispensing robot in its PCR configuration.

We thank you for having chosen AlphaHelix products and hope that it will satisfy you fully in automating your dispensing. If you have any comments, don't hesitate to communicate with us info@alphahelix.com.

An α Bot™ dispensing robot is a versatile platform that can carry different accessories depending on the protocol the user wants to automatise. In the PCR setup configuration, the α Bot™ dispensing robot consists of:

- A cartesian (XYZ) workstation base carrying one Z axis.
- One or several pipetting modules (exchangeable)

Furthermore, the α Bot™ dispensing robot is provided in standard with a working space of 9 SBS positions, as well as:

- A computer running the α Bot™ software,
- A power cable,
- A USB cable.

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1. A FEW WORD ABOUT ALPHAHELIX TECHNOLOGIES AB

AlphaHelix is an innovative company that develops on its own and in cooperation with OEM partners products in the PCR field for research and diagnostic laboratories. In 2011 AlphaHelix launched an ultra fast and uniquely specific PCR instrument α Amp™ and the α Bot™ dispensing robot.

The α Bot™ dispensing robot makes setting up PCR reactions easy and eliminates ergonomic problems in the laboratories.

AlphaHelix is continuously improving functions and adding more accessories. At the same time AlphaHelix also offers customized products for OEM partners around the world.

Hence, Alphahelix is interested to get feedback from the users and also is interested in developing partnerships with other...

Do not hesitate to contact us at info@alphahelix.com.

2. CONTACTS

This device was installed by your local distributor:

Distributor informations

In case of problem or for any information, please contact your distributor.

Manufacturer information
<div style="text-align: center;"><h1>AlphaHelix</h1><p>AlphaHelix Technologies AB Kungsängsvägen 29 SE-753 23 Uppsala SWEDEN</p><p>Telephone: +46 [0]18-12 07 01 Fax: +46 [0]18-12 07 03</p><p>Email : info@alphahelix.com Web : www.alphahelix.com</p></div>



3. SAFETY ADVICES

3.1. INTRODUCTION

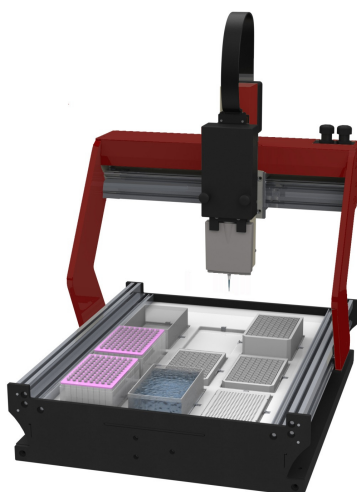
The α Bot™ is a system that has been designed to automate setup of (q)PCR reactions and other low volume dispensing in the laboratory. Using the system out of limits specified by AlphaHelix is at the user own risk.

Some elements and accessories can only be used by qualified and authorized personnel. Using the system is only allowed and guaranteed when the workstation is controlled by the α Bot™ software running under Windows Vista or Windows 7.

It is of the user responsibility to contact its distributor or AlphaHelix before using its α Bot™ robot in a way not described in the current manual.

3.2. SPECIFICATIONS

αBot™	
Dimensions : Lxlxh (cm)	60 x 50 x 56
Weight	35 Kg
Power supply	230V / 50Hz / 80W
Motor	Stepper motor – ST4118M1206 Nanotec
Accuracy X/Y	100 μ m
Reproducibility X/Y	10 μ m
Interface USB	USB cable (2 meters) type mini B/A





3.3. ELECTRONICAL SAFETY ISSUES

The user must ensure that all measures have been taken to ensure that the use of the workstation meets the requirements of performance and safety and comply with current laws, regulations, codes and standards. The place of use of the station should be selected according to the conditions of a research laboratory, and does not compromise the functioning of the device.

INSTALLATION AND SAFETY INSTRUCTIONS

Before plugging in the α Bot™ and turning it on, PLEASE CAREFULLY READ THE FOLLOWING INSTRUCTIONS:

- The installation and use of this material must match current norms in analysis laboratory (or research ones). The robot must be connected with an electrical installation in compliance with the standards.
- The α Bot™ must not be placed near water sources or under rain.
- It must lie on a solid support (lab bench: from 70 to 100 cm high).
- Before starting the machine, check the power wire and the connectors (poor condition of connectors, due to contacts oxidation, can imply sparks or fire starts).
- In case of electrical problem, call authorized personnel only (AlphaHelix or approved distributors).
- When plugging in or out the power wire, electric arcs or sparks can occur; consequently, the α Bot™ must be installed in a non-explosive atmosphere.
- Avoid any danger of electrocution; use a ground-connected power point. It is strictly forbidden to cut any ground wire on the machine, for user safety.
- The power point must be proportionate to the machine supply, thus 230 Volts, 3A, and must be protected by appropriate device (fuse and automatic switch).
- Avoid additional wires and extension cords.
- Any dismantling of the machine by the user will cause immediate warranty loss and cancel AlphaHelix responsibility whatever the consequences of any nature are.
- Any modification of the robot by the user, in particular electric system or electronic cards, is strictly forbidden and warranty uncovered.
- Before cleaning up the machine, unplug the power wire.
- Unplugging wires as the computer and the machine are switched on is not recommended.
- Beware of hot spots that can appear on the machine during the working.
- α Bot™ and the computer on which α Bot™ software is installed are not protected against voltage fluctuations or spikes. Therefore, all devices can be damaged by spikes or brutal power cuts.
- Current micro cuts can also involve information loss, especially concerning α Bot™.



3.4. CONTEXT OF USE AND USER SAFETY

3.4.1. CONTEXT OF USE

This material might be used in research, development, or industrial laboratories as well as educational institutes. The area of application includes life sciences, biotechnologies, chemistry, cosmetology and pharmacology.

α Bot™ robot must be used in a clean working environment. Check « Cleaning » section for further information.

α Bot™ robot conforms to basic requirements of CE directives and declaration of conformity norms (2004/108/CE; 2006/95/CE; 2006/42/CE).

α Bot™ robot might be used only by qualified staff and for professional use.

3.4.2. SECURITY ADVICES FOR USERS

Anyone manipulating samples has to wear appropriate protections (gloves, clothes...) and to adopt appropriate behavior. For instance, do not hamper axis moves. The Z axis of the workstation can exert a 15kg force during tips taking. As the workstation is moving, do not stand near the working space. Ensure that no one does.

In case of accident, if necessary:

- Turn the station off by pressing the switch « **On/Off** », figure 4.
- If needed, call for rescue and bring first-aid treatments.

Any incident or defect, recorded as well as presumed, must be reported to AlphaHelix or your distributor if disturbing the good working state of the workstation.

User has to ensure that all terms of use, installation and working in this guide have been followed.

As for any collective equipment, the good working state of the automat depends on the care of the users. Any damage due to mishandling will be repaired at the expense of the user.

α Bot™ has to be cleaned up and inspected periodically and efficiently.



3.5. SAFETY

PREREQUISITES

Important: before using the robotic workstation, please read carefully the user's guide and all safety instructions to avoid incidents and accidents.

ENVIRONMENT

A. Hazard

■ Support :

- Because of moving parts in the machine, the workstation must lie on a rigid and stable surface, like laboratory lab bench.
- The support has to be horizontal.

■ Explosion hazards :

- Do not use the workstation within an explosive atmosphere.
- Do not use the machine to manipulate explosive, radioactive, or highly reactive substances.
- Do not use the device for handling substances that could create an explosive atmosphere.

■ Electronic hazard :

- An only personnel authorized by AlphaHelix is able to open the device to perform maintenance or repair.
- AlphaHelix staff only can dismantle the machine for maintenance.
- Never use a damaged device.
- Never turn on the machine if the power cable is damaged.
- Connect the machine to a power point in compliance with current legislation, and respect technical specifications of the workstation.

B. Warnings

■ Comfort :

- For user convenience, place the machine on a lab bench between 70 cm to 100cm high.

■ UV Lightning :

- Avoid any UV source near the machine for a long period.

■ Heat :

- Do not place the device near a heating source.
- Do not expose it to excessive sun light.
- Allow a gap of 10 cm minimum security around the workstation.

■ Vibrations :

- Avoid working support where vibrations can occur (mixer, centrifuge...).

■ Residues :

- Garbage can contain inflammable or infectious residues after tips ejection. Treat these wastes in compliance with current legislation.



■ **Contamination :**

- Decontaminate the workstation and devices before reuse.

■ **Spills :**

- If a large quantity of liquid is spilled, shut off the power and unplug the power cable.
- Mop up the spill, taking care to observe the safety rules for handling this solution.

C. Manipulation

Hazard :

- **Injury:** Z axis moves can cause injuries as soon as a protocol is running.

- Ensure that no one access the working space when the workstation is running.
- Before accessing the working space, stop the system via the software and wait for the axis to reach loading position.
- If any problem occurs, use the « **On/Off** » switch to turn off the power supply and ensure a safe access to the workstation.
- Do not touch any part of the workstation marked with the crossed hand pictogram when power is turn on and the machine is powered.

- **Hazardous substances contact:** Healthy problems or burning trauma can be initiated by chemical or biological substances. To prevent any healthy risk, please follow the recommendations:
 - Respect the security constraints specified by the manufacturer of the solutions used.
 - Use the well-adapted protective wearing for these specific substances.
 - Follow the washing & de-contamination program, then ensure that the hygiene standards are well-respected.

D. Warning pictograms

To inform on potential risks with particular parts of the equipment, some pictograms are included on PrimaRW34.



Pictogram informs about a security risk. Please report to the safety information list to know what type of specific risk is concerned.



Do not touch



4. WEEKLY MAINTENANCE

4.1. DESINFECTING SOLUTIONS DESCRIPTIONS

The following solutions can be used for cleaning the robotic workstation but you need to dilute these solutions before using:

- **Ethanol:** effective against most of bacteria and fungus, as well as a lot of virus. It is not effective against bacterial spores
- **Active chlorine** (sodium hypochlorite): efficient against gram +/- bacteria, mycobacterium, yeast, fungus and virus. It is not efficient against bacterial spores
- **Ammonium Quaternary derivatives:** efficient only on Gram+ bacteria, partial on Gram -, mycobacterium (proteins), yeast, fungus and virus. No effect on spores.
- **Active oxygen:** effective on large range of pathogenic organisms: bacteria gram + & -, virus and fungus.

4.2. CLEANING PROCEDURES

The following procedures may apply depending on the parts that one would like to clean (pipette, working space, plate holders and consumables):

1. **Pipetting unit:** (bottom part of the pipette and especially the conic-support) is washed and disinfected with Ethanol at 70% solution or a 3% active chlorine solution(sodium hypo chloride) or ammonium quaternary derivatives or containing active oxygen:



- Do not immerse the whole pipette into a cleaning bath since the upper part which comprises electronic part is not removed.
- Apply your washing solution on the pipette bottom part (encircled in red on the picture).
- Let it react during 1 to 3 minutes before removing remaining solution with absorbing paper.

2. **Working base:** is first cleaned & disinfected with water then with Ethanol at 70% solution or a 3% active chlorine solution(sodium hypo chloride) or ammonium quaternary derivatives or containing active oxygen:

- Clean and rinse your working base with water,
- Apply the cleaning solution on it,
- Let it react during 1 to 3 minutes before removing remaining solution with absorbing paper.

3. **Tip box holder:** as for working base, tip box holder can be cleaned and disinfected through the same initial working base procedure.



4. **Consumables:** please look at technical data delivered by the related suppliers.

\\ **WARNING:**

- Protected Gloves & glasses have to be used during cleaning operations to avoid any cleaning solution contacts and projections
- During cleaning operations, rub is recommended to remove all bacteria bio-films
- If organic components are used (wood, serum, expectoration), active chlorine is not recommended



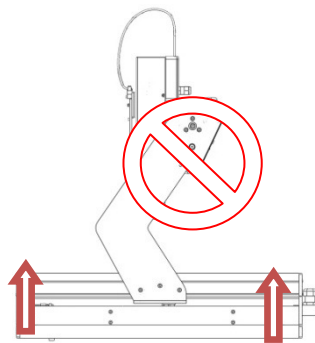
5. HANDLING

5.1. UNPACKING PROCEDURE OF THE PRIMARWS

To move the α Bot™, it is essential to having to 2 persons holding the machine.

✖ **WARNING:** Never hold the machine by the bridge. Any mishandling of α Bot™ may cause irreversible damage.

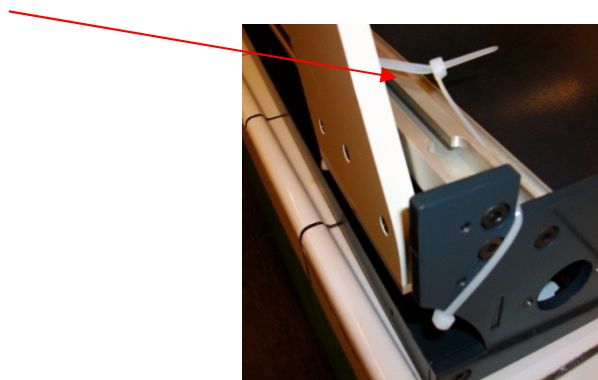
1. Get the robot out of the box (lift the robot by the bottom, not by the bridge)



2. Unscrew the screw which is used for the immobilization of the X axis during transport



3. Cut cable tie which immobilize the Y axis



4. Fix the two rear feet of the workstation
5. Set machine according to protocol



5.2. PREPARING THE ROBOT FOR TRANSPORT

The transport of α Bot™ must be in the box provided for that purpose, delivered with the machine.

Do not, under any circumstances, carry the machine in a vehicle without this box.

After any movement, it is necessary to realign the machine. It should also go through an adjustment phase. These points must be performed by qualified personnel AlphaHelix.



6. SET UP OF THE ROBOT

The following part is describing the α Bot™ installation, for which 2 parts are defined:

- Hardware setup (robotic workstation and accessories)
- α Bot™ software and drivers

This part is related to the system set-up:

- How to connect each unit to the robotic workstation
- How to install the software

6.1. REQUIREMENT AND SAFETY



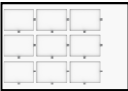



The lab bench, where the α Bot™ robot will be installed, has to be large enough to receive the workstation and its computer. For the α Bot™, it would need: 1.10x0.50 m² in space and 68 cm high. Then, the environment has to respect the following conditions:

- Working Temperature between +15°C and +30°C.
- Enough aeration and ventilation to remove the heat generated by the workstation.

6.2. DELIVERY PACKAGE

The α Bot™ robotic platform conditioning has been developed to ensure its physical and functional integrity during transportation, storage and handling.

The following components are delivered with the α Bot™ robot:

	Robot workstation
	Power cable 230V, CEE 7/7 type E/F plug
	Deck 9 places SBS
	Pipetting unit
	Optional computer and its power
	α Bot™ software on USB flash memory



6.3. SETTING UP THE α Bot™ ROBOT AND THE COMPUTER

6.3.1. DESCRIPTION OF THE WORKSTATION

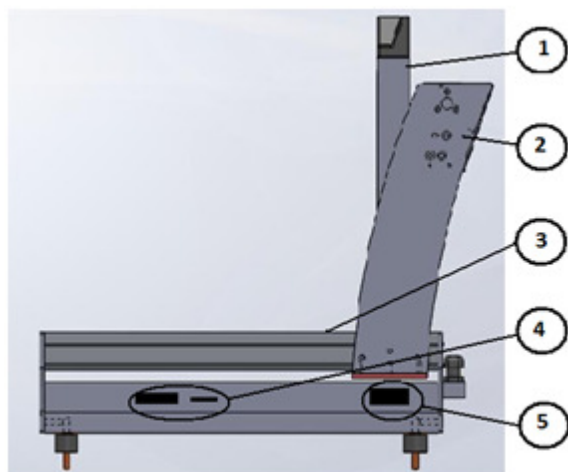


Figure 1: Left view of the α Bot™ robot

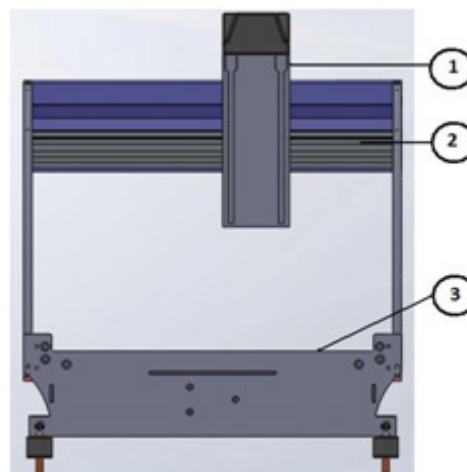


Figure 2: Front view of the α Bot™ robot

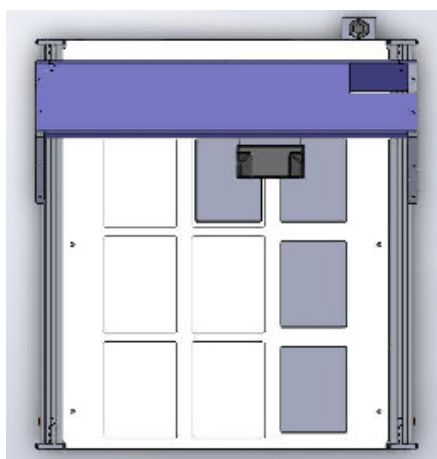


Figure 3: Top view of the α Bot™

1	Modules support
2	Workstation arm
3	Rack
4	USB port
5	Plug



6.3.2. CABLING

- ✖ **WARNING:** Always place the switch on its « **Stop** » (or 0) position before workstation connection to the power in order to protect the machine from electrical chocks.

Start the αBot™ robot:

- Check if the robot is connected to the outlet,
- Turn on the computer,
- Turn on the workstation using the « **On/Off** » switch located on the left side of the device,

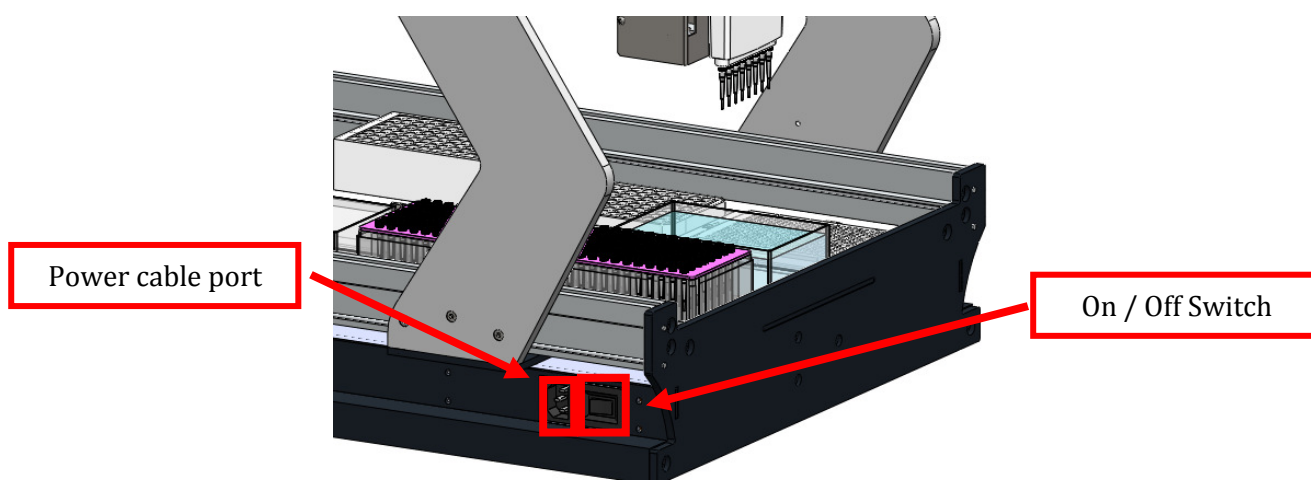


Figure 4: On / Off Switch

- Plug in the computer to the workstation with USB cable.

6.3.3. TURN OFF THE ROBOT AT END OF USE

If you have to disassemble the workstation, follow these steps:

- Check the arm is stopped and in home position,
- Turn off the robot,
- Plug out USB cable between the computer and the workstation,
- Plug out power cord,
- Put the protective sheet (provided with the machine) on the tray.



6.3.4. ADVICE BEFORE INSTALLATION OF SOFTWARE

Before installing the α Bot™ software, check all connections (6.3.2). The installation can be done without connection with the robot.

6.3.4.1 RECOMMENDED CONFIGURATION

Hardware configuration

- Processor clocked at 2,5 GHz
- RAM memory : 3 Go
- Free disk space : 200 Mo
- Graphic card: 512 Mo

Software configuration

- Operating system Windows Vista/Windows 7 (32 or 64 bit)

6.3.4.2 INSTALLATION OF SOFTWARE

In order to ease your use of α Bot™ software, it is already installed on the provided computer. In this case the software is ready to use.



7. INSTALLATION OF THE ROBOT SOFTWARE

The installation of the software is done from « **SetupAlphaBotvxxxxxx.exe** ».

Note!x86 is 32 bit,x64 is 64 bit version.

Please accept all permissions during installation. At any time you can abort the installation by clicking « **Cancel** ».

To start the installation, double click the installation file. Then follow the instructions on the screen:

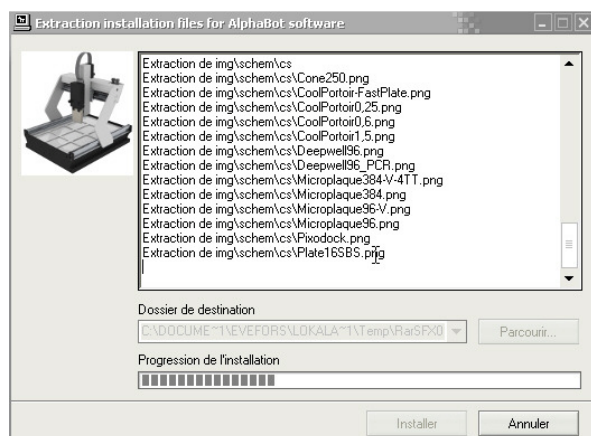


Figure 5: Extracting window

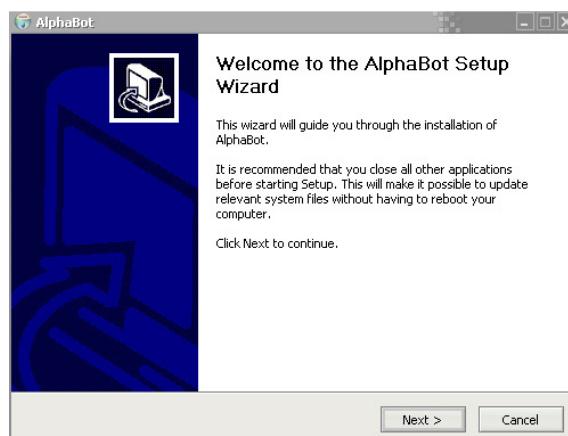


Figure 6: Installation window

Then choose the components to install on your computer among following:

- **Shortcuts** : creation of a shortcut on your desktop to ease the access to the software,
- **Drivers**: installation of required drivers to control the workstation and its tools. This option is only required for the first installation. Refer to the section « **Installation of drivers** » (7.1),
- **Framework 4.0**: installation of library's classes required for the running of the software. This option is only required for the first installation. Refer to the section « **Installation of Microsoft Framework** » (7.2). Click « **Next** » to go to the next step.

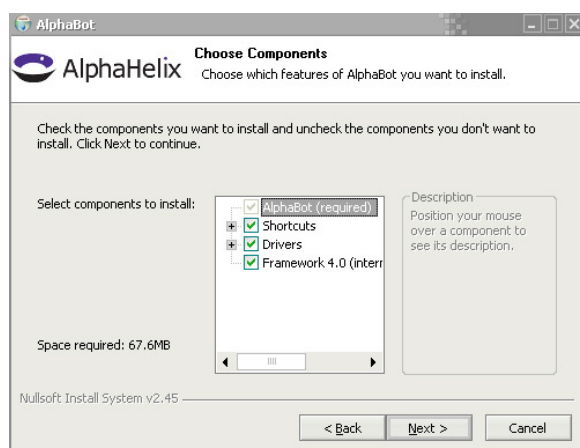


Figure 7: Choice of components

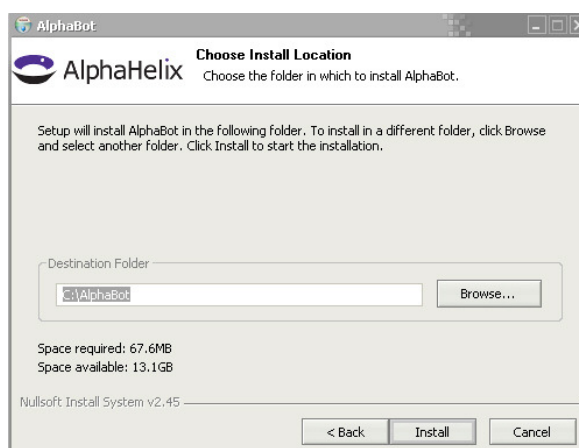


Figure 8: Installation path

⚠ **WARNING:** Use the default value: **C:\abot** as the installation path.



7.1. DRIVERS INSTALLATION

If you chose to install drivers, you can see following installation window:

Please follow instructions on the screen.

Drivers have to be installed for the first installation of α Bot™ software.

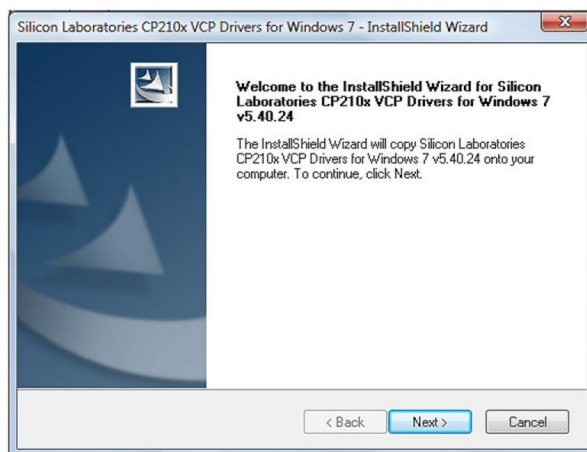


Figure 9: Drivers installation window

7.2. INSTALLATION OF MICROSOFT FRAMEWORK

If you chose to install the Microsoft Framework, a window starts at the end of the installation of the software. Please follow the instructions on the screen.

Framework has to install for the first installation of α Bot™ software.

🗨 **NOTICE:** Generally, on Windows (Vista / 7), Microsoft Framework is already installed. Check the release is 4.0 (or upper).



8. RUNNING THE SOFTWARE

8.1. LAUNCHING THE SOFTWARE

After installing successfully the software, the following icon should be on the desktop of your computer.



Figure 10: α Bot™ shortcut icon

To open α Bot™ software, double click on it, the main interface appears as the below figure:

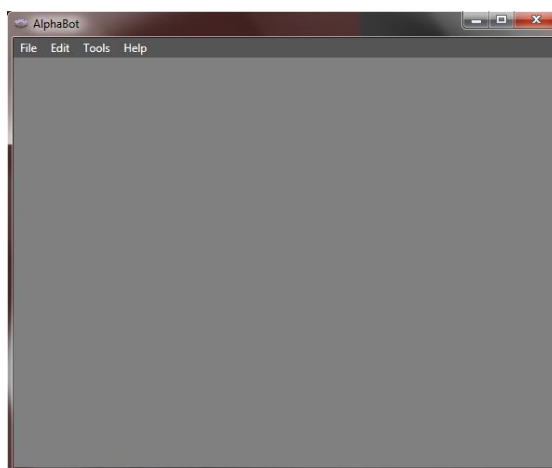


Figure 11: Software 's main interface

- « **FILE** » menu is to manage your protocol (see page 24):
 - « **New Project** » : to create a new protocol
 - « **Open Project** » : to open an existing protocol
 - « **Save As** » : to save a protocol as a new file
 - « **Save** » : to save the modification of a protocol in the same file
- « **EDIT** » menu is to manage your liquid parameters:
 - « **Liquid Manager** » : to manage solutions used in protocol
 - « **Pipeting Profil Manager** » : to manage profile of pipette

These functions will be described more details in chapter 8.6., page 48.



8.2. PROJECT MANAGEMENT: « FILE » OPTIONS

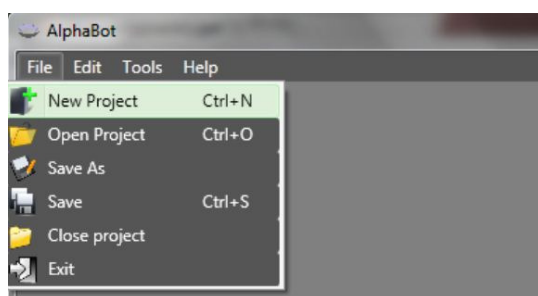


Figure 12: File function

For creating, opening or saving a project, see the options which are describe below:

8.2.1. CREATE A NEW PROJECT

- To start creating a new protocol, select « **File** » then « **New Project** » (Figure 13) :

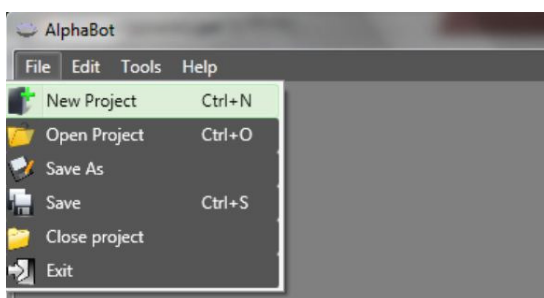


Figure 13: Creating a new protocol

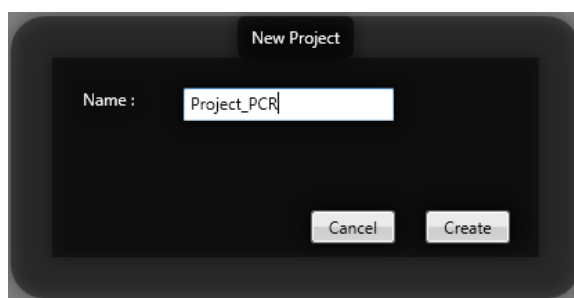


Figure 14: Window « New Project »

- A window appears allowing the user to enter the name of your protocol (Figure 14),
- Then, click on « **Create** » button to proceed to the next step.



8.2.2. OPENING AN EXISTING PROTOCOL

On the software's main screen, click on « **File** » and choose « **Open Project** ».

Then click on « **Browse** » for finding your existing protocol. After the choice of the protocol, click on "**Open**". You would see the message « **Loading in progress...** ».

Afterall, if the protocol is properly loaded, you will see the working space you set before.

If nothing happened, try again the two first steps:

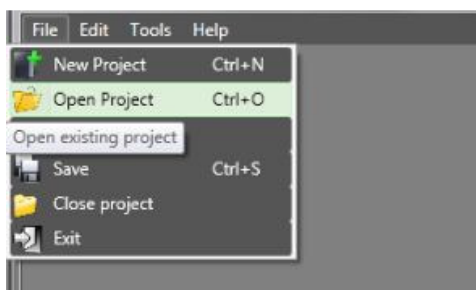


Figure 15: Opening an existing protocol

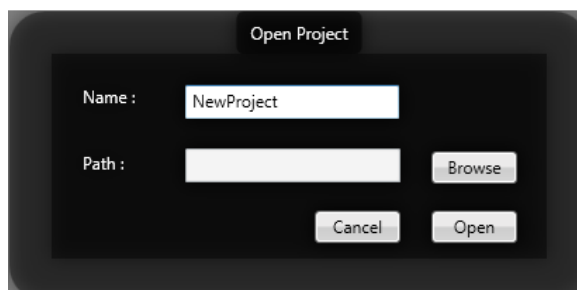


Figure 16: Opening window protocol

8.2.3. SAVE A NEW PROTOCOL

To save your protocol, clic on « **File** » button in the left head corner and choose « **Save As** ».

Choose a name and a directory for your protocol and clic on « **Save** » to confirm or « **Cancel** » to come back to the protocol setting:

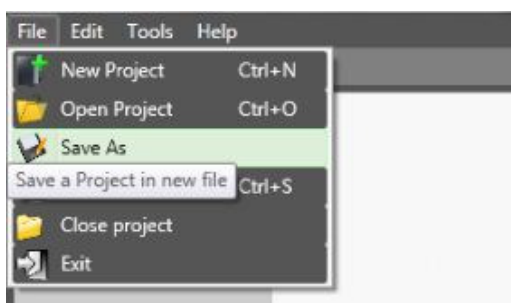


Figure 17: Saving a new protocol

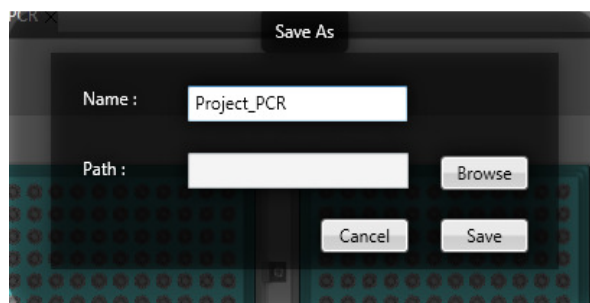


Figure 18: Window « Save As »



8.2.4. SAVE NEW SETTINGS OF A PROTOCOL

When modifications had been done, you can save it.

Click on « **File** » then « **Save** » to save changes on your protocol. Your changes are saved after the «**Saving in progress**» message disappears. You can also save your protocol by closing the protocol's tab. A message appears. Click on «**Yes**» to save changes, «**No**» to ignore changes and «**Cancel**» to come back to your protocol:

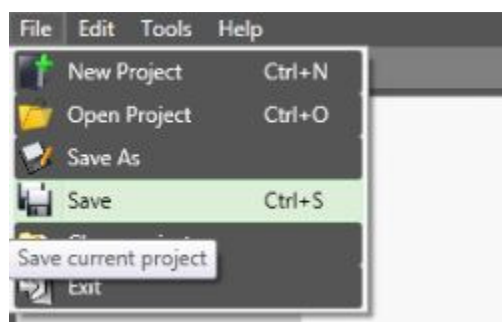


Figure 19: Save modification done on a protocol



8.3. OVERVIEW OF THE SOFTWARE'S MAIN INTERFACE

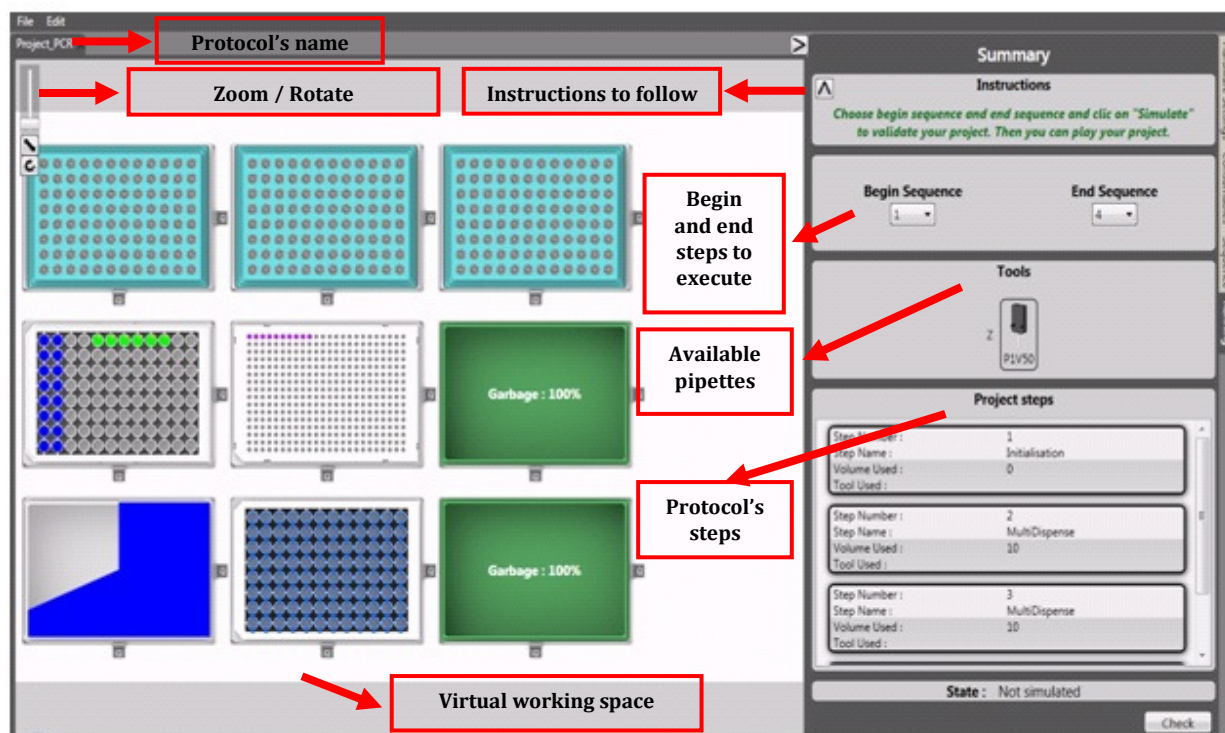


Figure 20: Main interface

1

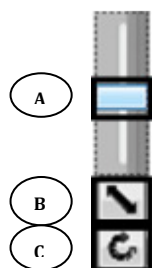
2

1) **Working space**: shows your virtual working space.

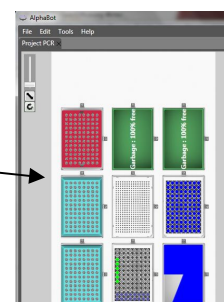
2) **Control panel**: is used to create and manage all protocol's steps.

Modifying the working space's views

You have three cursors for modifying your working space's views :



- A. Zoom in/out the working space
- B. Resize the working space
- C. Rotate the working space, suggested position





8.4. SOFTWARE PHILOSOPHY

The software was created to simplify the highest steps of creating a protocol. In just a few clicks you can generate a new protocol.

On the right of this figure, you have the control panel and center the contents of each tab:

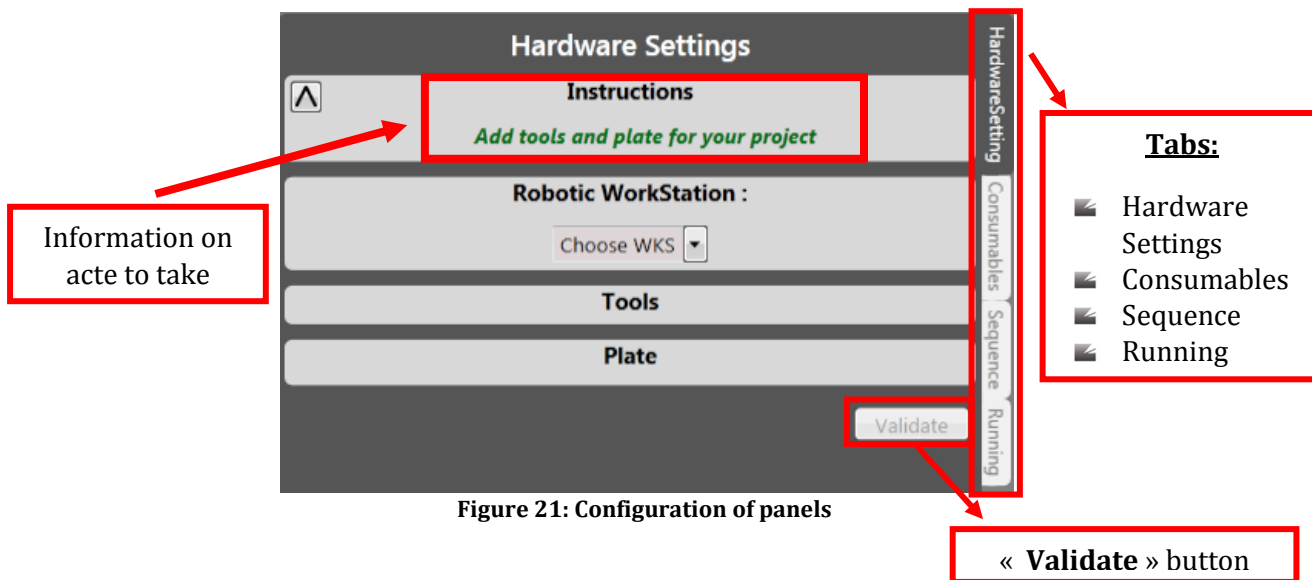


Figure 21: Configuration of panels

Each tab allows configuring a specific step in the creation of a protocol. Click « **Validate** » button to proceed to next step.

- The « **HardwareSetting** » tab manages the choice of the workstation, the pipettes and the type of tray,
- The « **Consumables** » tab manages the consumables that will be used in the protocol: Tip Box, microplates...
- The « **Sequence** » tab is used to manage the operations that will take place in the protocol,
- The « **Running** » tab verifies the feasibility of the protocol and run it.

8.5. USE OF αBot™ SOFTWARE

Create a new protocol as describe in page 25. In the following parts, you have all the steps that the user will be do for creating a new protocol.

8.5.1. TOOLS MANAGEMENT - « **HARDWARE SETTINGS** »

The software shows the main interface where a virtual working space is shown.

In the first tab, you can select your workstation, your working space and also the different pipettes which are available on your workstation.

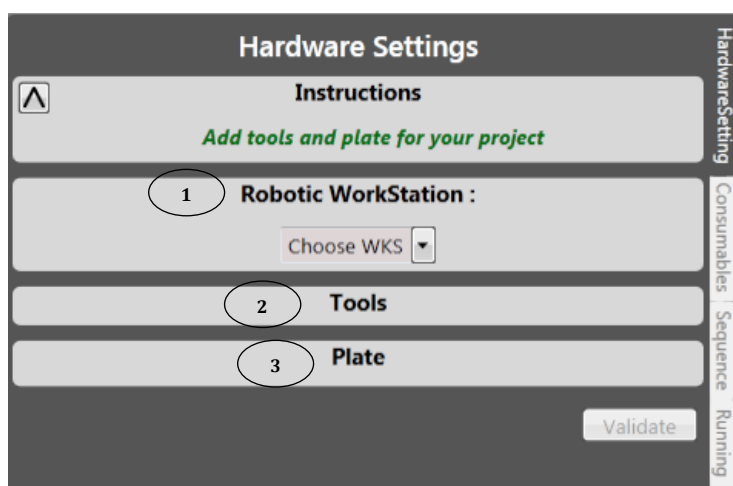


Figure 22: Panel Hardware Setting

1: Select the of α Bot™ workstation in the list box (normally default)

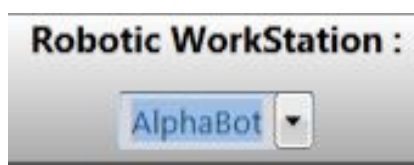


Figure 23: Workstation's selection

2: Select the tool (pipetting head; P1V250; P8V250...) by clicking on « **Tools** » (Figure 18)

Each pipette is indicated by its number of channel and its nominal volume. For example: **P1V50** means **P**ipette **1** channel and **V**olume: **50**µl.

3: Select the working space: 9 SBS plates (only one choice for the generic α Bot™) by clicking on « **Plate** » button. You can have more choices here depending on how your robot is equipped.

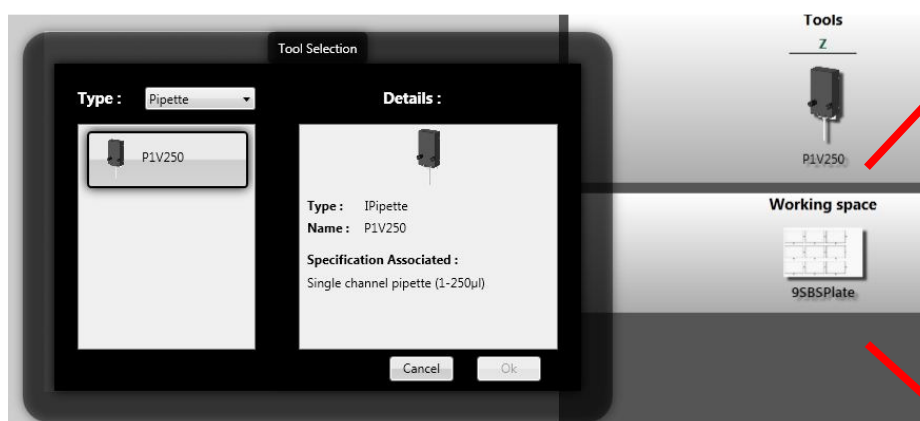


Figure 24: Tools and plates selection

Select the tools by clicking on this icon

Select the working space by clicking on this icon



After selecting the tools and the working surface, the virtual working space appears as below:

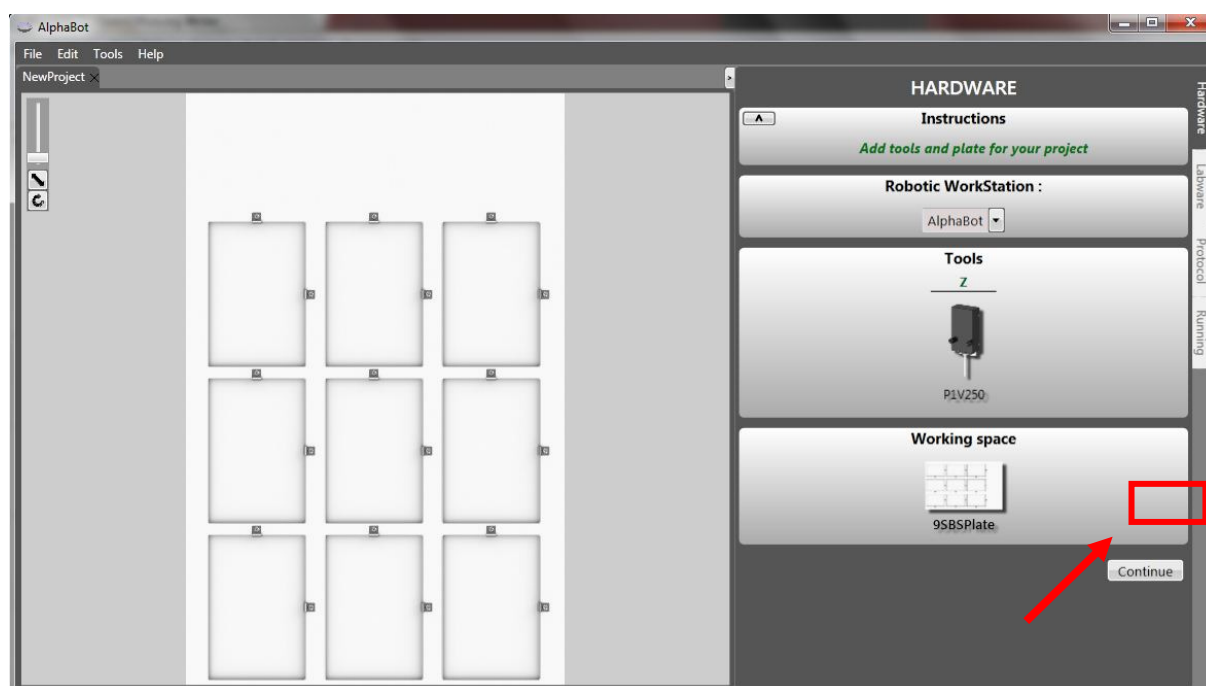


Figure 25 : Virtual working space

Click « **Validate** » button to proceed to the next tab.

8.5.2. LABWARE MANAGEMENT - « CONSUMABLES »

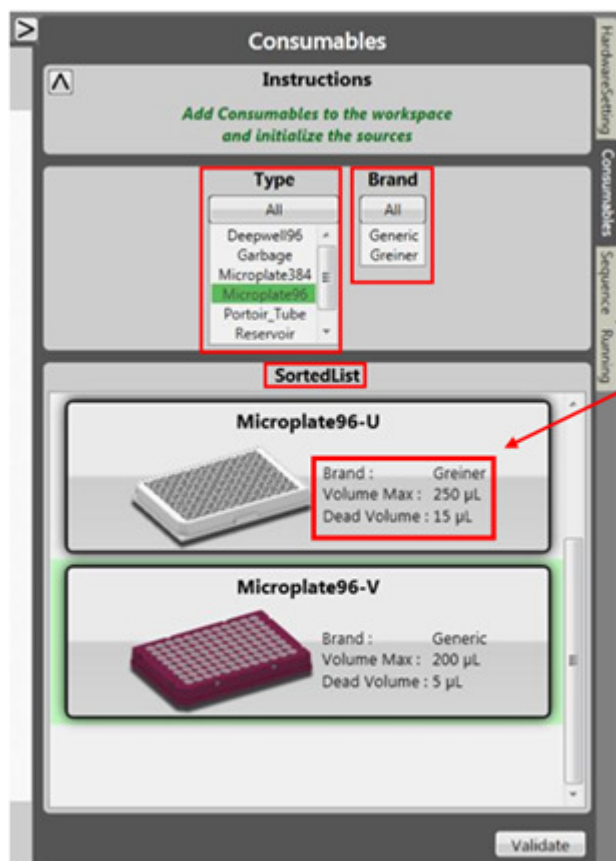
The main window of « **Consumables** » shows the working space where the user will add labwares (see figure 22).

A list of available consumables appears on the right hand side of the window.

These consumables are ranked by types & brands for easily searching.

Select a consumable by clicking on the desired type (microplate 96, microplate 384, tip boxes...).

Then, those available consumables of corresponding types appear in “**Sorted list**” (Fig. 20).



Information about each consumable is available as follow:

- ✓ Consumable's brand
- ✓ Maximum volume
- ✓ Dead volume

Figure 26: Consumables panel

NOTICE: All consumables are present on the software's database. This database can be extended by the user's requests.



Arrange the consumables required, for the protocol's execution, to the desired positions on the workspace. **Add consumables by drag & drop.**

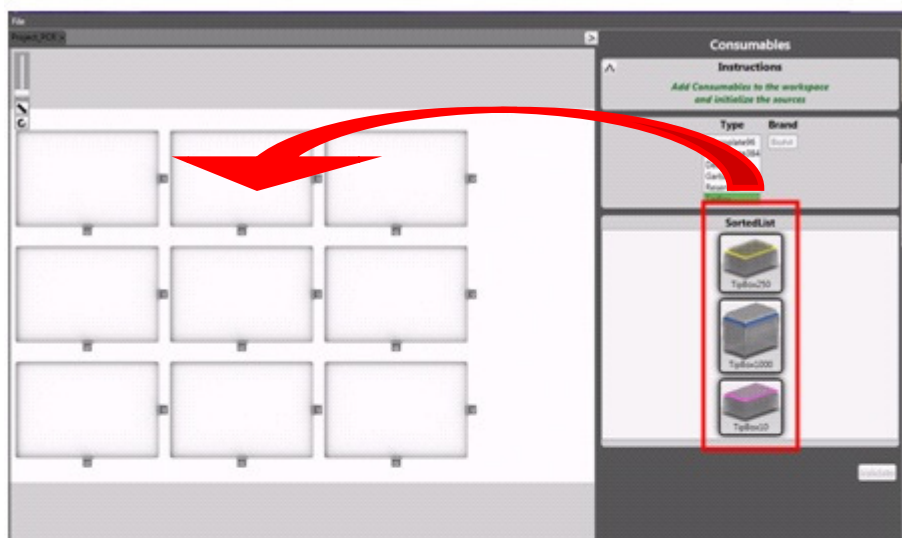


Figure 27: Workspace plan

To create your protocol, it is necessary that at least one of the following consumable is present on your workspace:

- Tip box,
- Trash,
- Microplate /Deepwell or/and Tube holders.

💬 **WARNING:** Always think to use compatible tip with your pipettes.

Example: tip of 10 μ L with pipette of 10 μ L

💬 **WARNING:** If you choose a « **ReservoirStandard** » as source, you'll have to add a Deepwell, microplate or rack type consumable on your workspace.

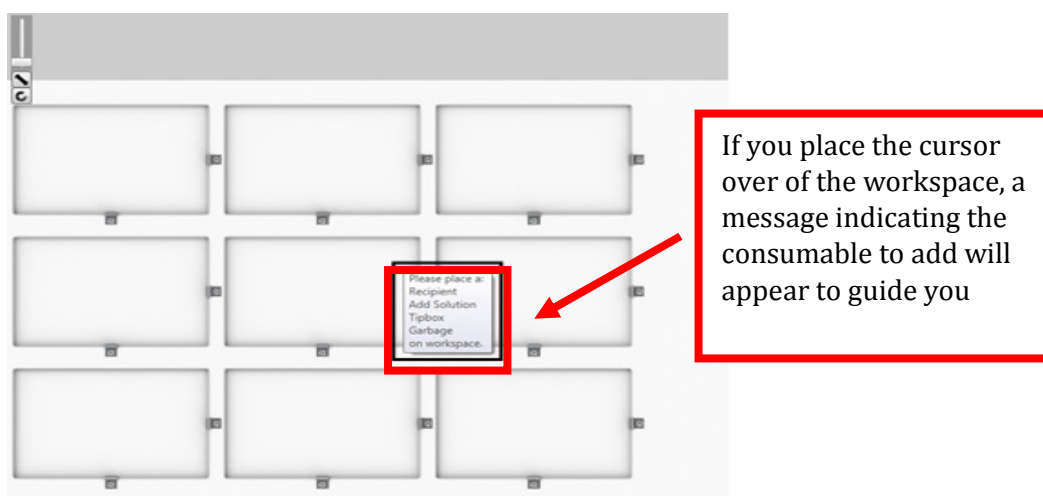


Figure 28: Creation of a workspace



You can modify the position of any consumable by simply « **slip / deposit** ». You can remove it as well by right click on the consumable then « **Delete** ».

WARNING: Always respect the positions allocated, with your consumable, on your real workspace, when you configure your virtual workspace.

Once all the consumables are in place, it is necessary to initialize the liquids levels contained in your containers. To do this, right click on the container which is the source, then "**Initialization**".



Figure 29: « Initialization » of the sources on the workspace

A window opens representing this container and the list of the liquids to be added (figure 24).

Select the wells in which you want to define a source, and then click on « **Init** ».

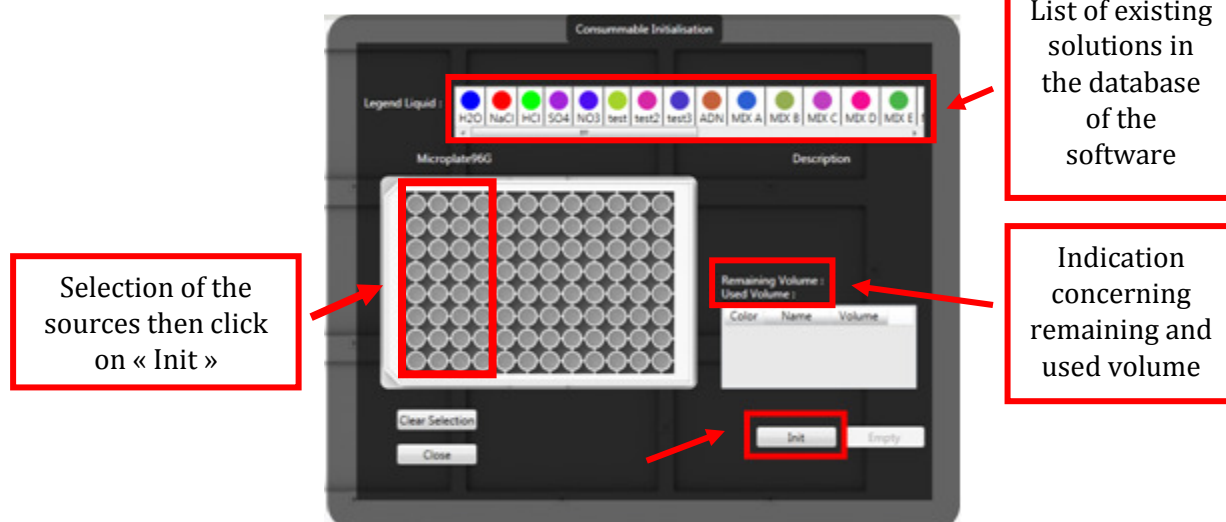


Figure 30: Initialization window of your container



You can then choose the liquid in the database.

For each liquid, you can choose the name, color and type using the « **Modify** » button (for details, see the section « **Edit** », page 48).

You can also add new liquid using the « **New** » button. When this liquid is added, it will be saved in the data base and will appear in your list.

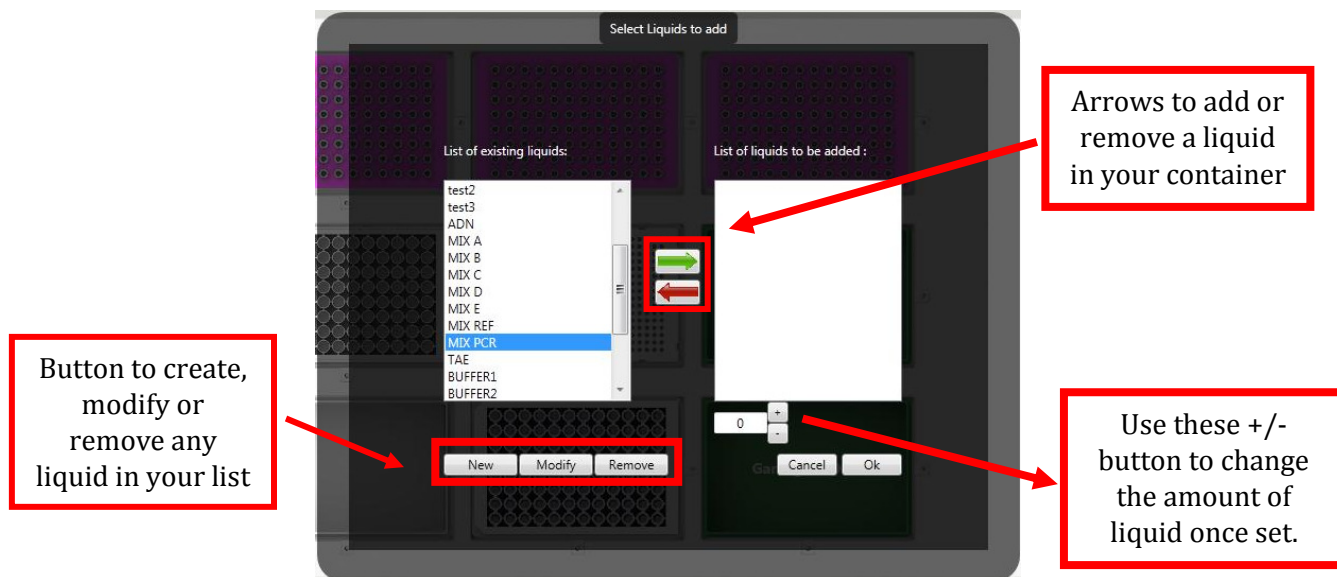


Figure 31: Sources initialization

Choose the liquid in the list and click the green arrow to add it to your container, and then enter the volume contained in the selected well, expressed as microliter (μL) for plates or as milliliters (mL) for tube.

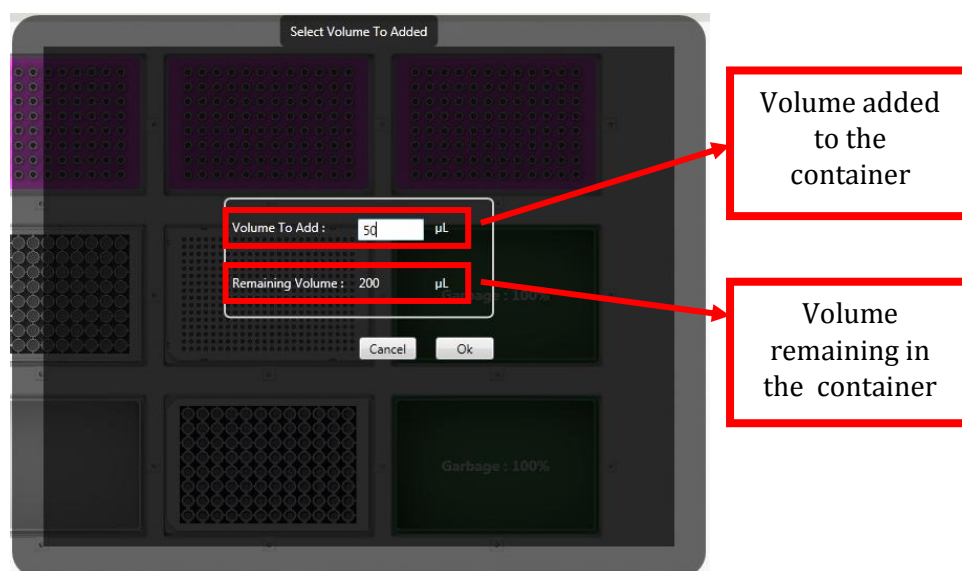


Figure 32: Initialization of volume

NOTICE: The amount of liquid relative to the capacity of wells is represented by a pie chart.



For having informations about a liquid, in a well, click on it. A description of the liquid in the container appears:

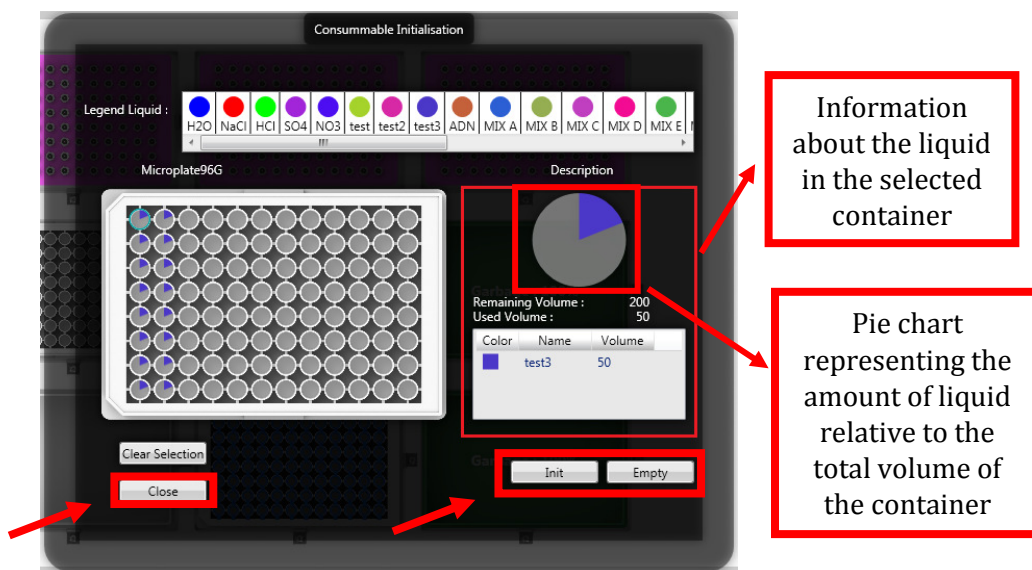


Figure 33: Description of the liquid

If you want to reset the values of some wells or reservoir, select the wells or the reservoir involved and then click « **Empty** ».

If you want to change a liquid and his volume contained in a consumable, click on « **Init** » to change it.

Click « **Close** » once the initialization completed.

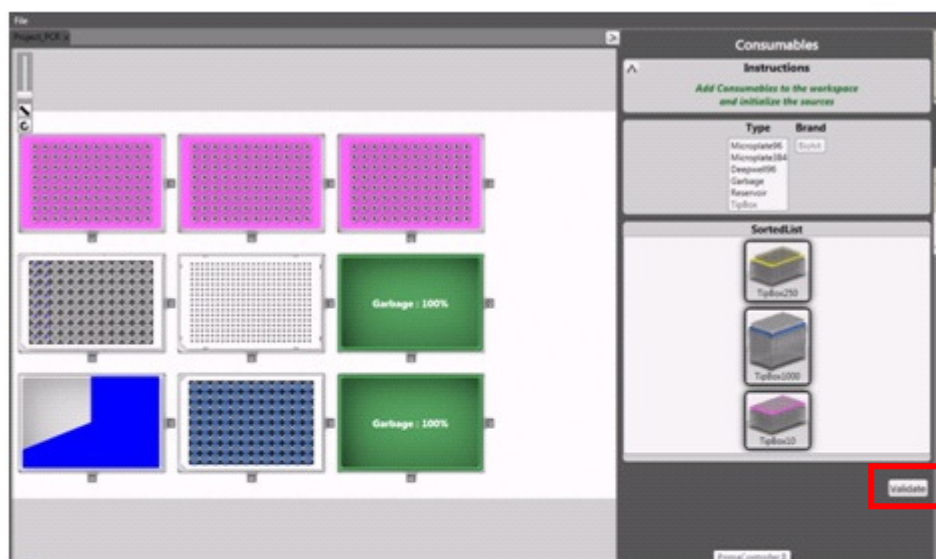


Figure 34: Workspace after initialization

Click « **Validate** » to go to next step of creating a protocol.



8.5.3. PROTOCOL MANAGEMENT - « SEQUENCES »

A. Initialization sequence

A protocol in the α Bot™ software is composed of different sequences of tasks. Each sequence is linked to a serie of individual tasks.

The first sequence is defined as the « **Initialization** ». This step reflects the state of the work space, the types and positions of consumables and the definition of the sources.

B. How to add a sequence

When you click on « **Add** », drop-down list appears. In this list, you can either select manually your pipette(s) or the « **Auto** » mode. This mode chooses the most appropriate pipette required for the sequence.

a. Manual mode

Each serie of tasks is defined by adding a sequence.

For this, make sure that the « **Type** » of task is « **Liquid Handling** ».

Choose then the action of the sequence (Multi dispense)

A listing appears when you click on « **Add** », allowing you to choose the right pipette for the selected task.

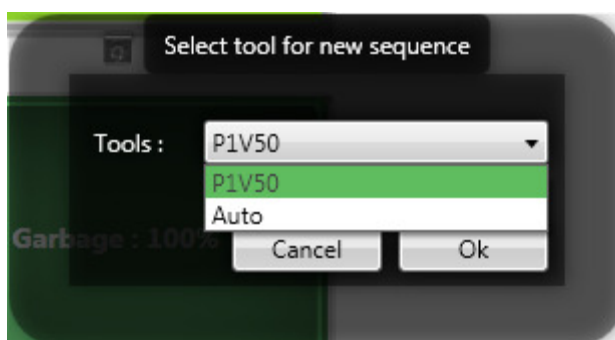


Figure 35: Choosing the right tool

If you choose manual mode, you can select sources and targets in relation to the available pipette:

- If the chosen or available pipette is a single channel, you can only choose one well as the source and one or several wells for dispense.
- In case of 8 channels, it is the same with a row of 8.
- After the choice of the tool, click on « **Ok** ».

A window appears, leading you for the different steps to achieve. The lower part of the window indicates the kind of task. The upper part leads you for the following events.

■ SELECTED SOURCE:



First, you have to choose the source where the pipette will aspirate. Then click on the arrow for the next step. The wells or tubes selected as sources are underlined in red.

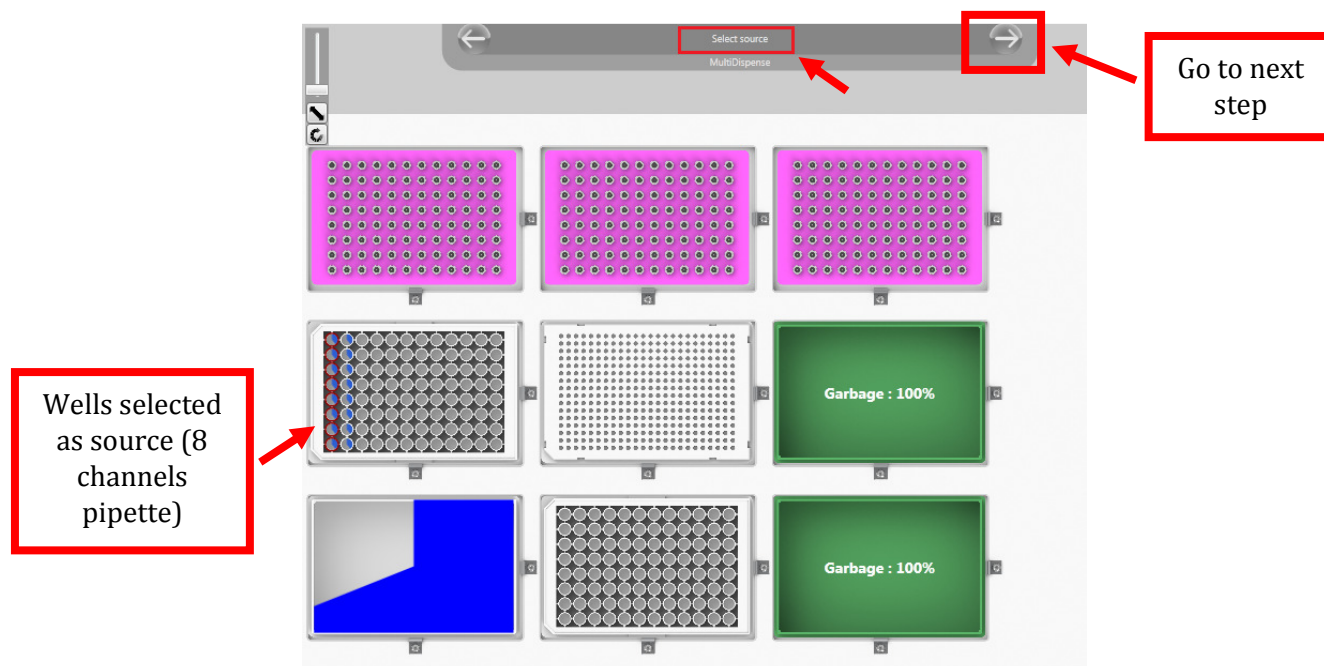


Figure 36: Source selection

SELECTED TARGET:

Second, select the target wells in which the pipette will dispense the liquid. Wells or containers that are used as targets are circled in green.

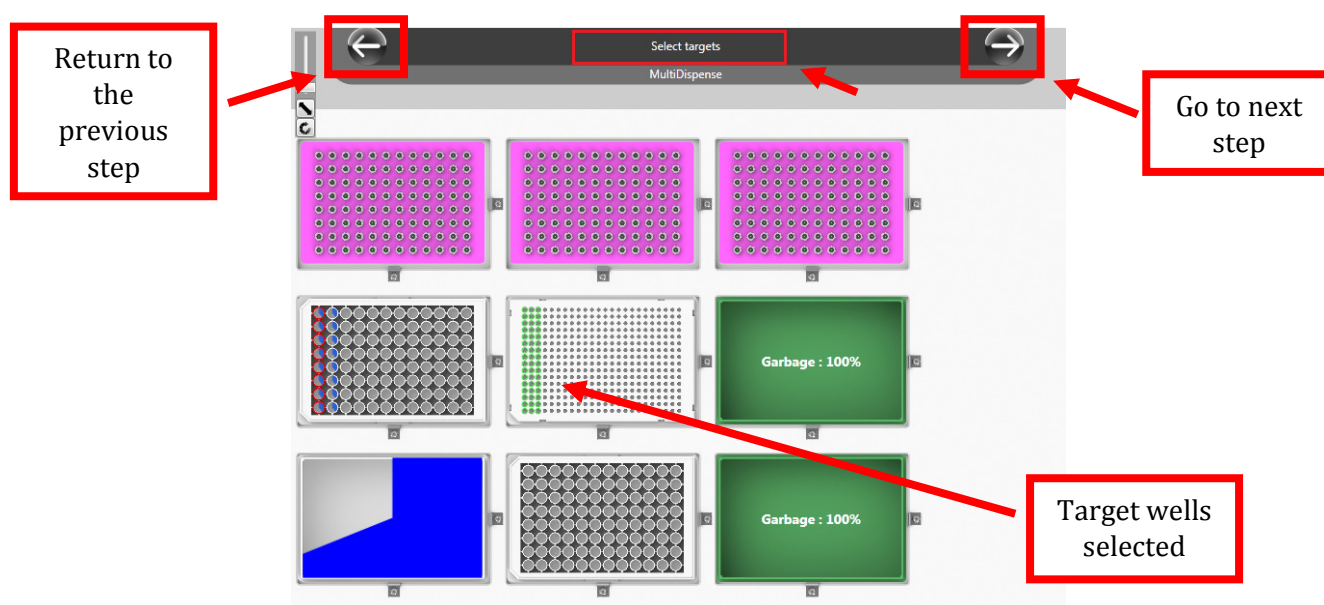


Figure 37: Target selection

SELECTED VOLUMES AND OPTION:



Once target are chosen, a new window appear which will allow you to define the pipetting parameters. You're free to change those parameters. By default the bottom of the tips will dive 2mm below the liquid level when aspirating.

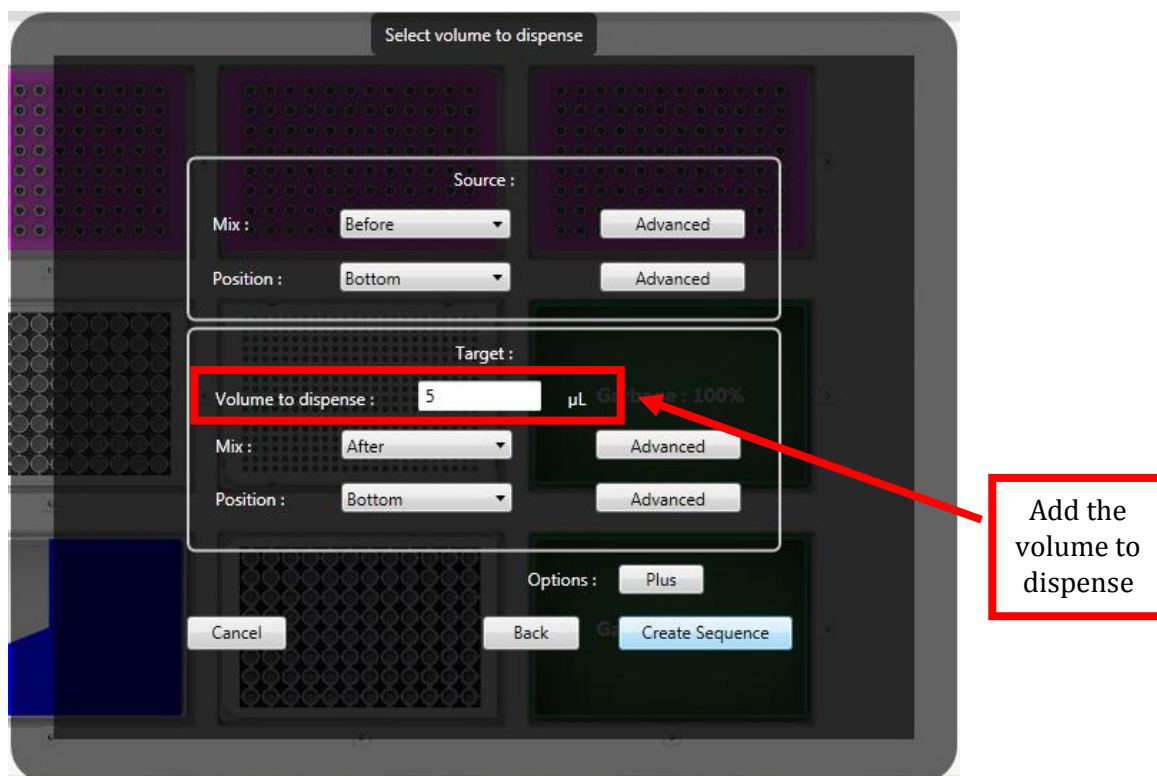


Figure 38: Pipetting parameters

If the source has not enough liquid compare to the determined volume, a message will ask you to complete the related source. See page 44, for more detail (**Troubleshooting**).

You have to fix the following parameters:

■ Source (« **Source** » box) :

■ Mix

- Before: mix the source before aspirate (parameters flexible)
- Custom: If you choose that mode, click on « **Advanced** » to change default parameters (scheme 34)

■ Position

- Bottom: Go down to the bottom of the source to aspirate
- Middle: Go in the middle of the source
- Custom: Click on « **Advanced** » to change the position (scheme 35)

■ ADVANCED OPTIONS:



- 1:** Volume to aspire for mixing
- 2:** Speed of Aspiration/Dispense
- 3:** Number of Aspiration/Dispense
- 4:** Cone position in the liquid

Click the button to show or hide the image on the right

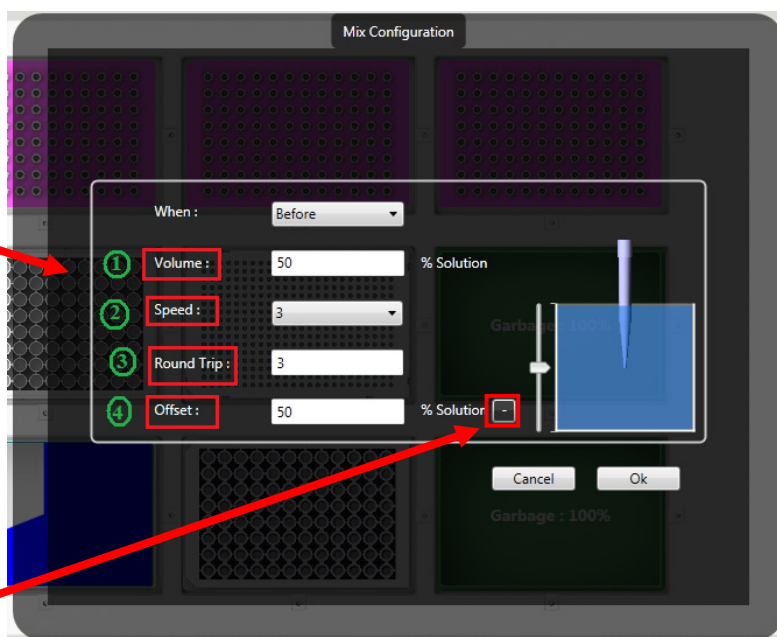


Figure 39: Defaults mixtures

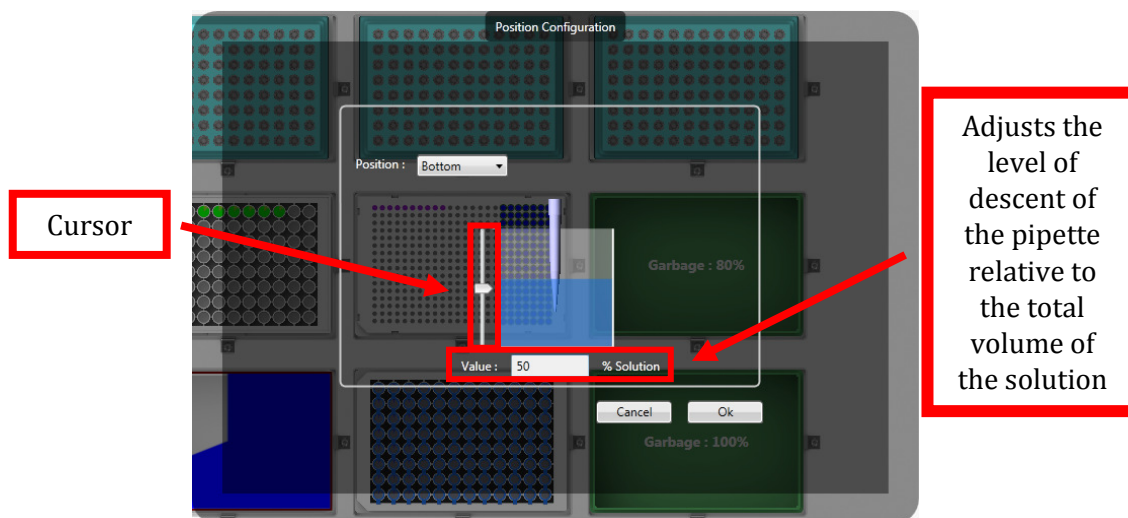


Figure 40: Cone position in the liquid (Solution)

You are free to change the level of the tips in the liquid. For this, enter directly the « **Value** » or use the button on the left of the picture.

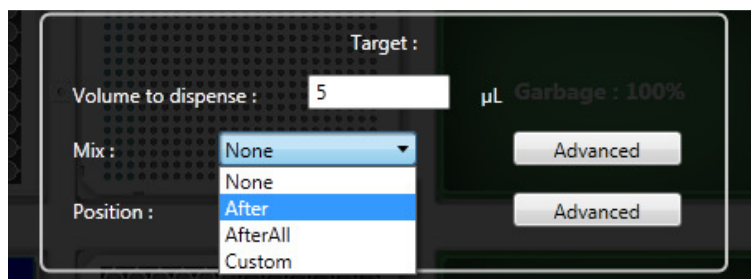


Figure 41: Dispense parameters

■ Target (« **Target** » box) :

■ Mix

- The default settings can be changed for each mixing functions.
- After: mix after each dispenses. If you choose this mode, the pipette will change the cone after each dispense/mix.
- After all: mixing wells target once the action (Multidispense) is finite. Change the pipette cone before going stir in wells target.
- Custom: if you choose this mode, click then on « **Advanced** » for changing the defaults mixing parameters

■ Position

- Bottom : Go down to the bottom for dispensing
- Middle : stay in the middle of the wells for dispensing
- Top : remain in the upper wells for dispensing
- Custom : click then on « **Advanced** » for changing the position of the cone relative to the liquid

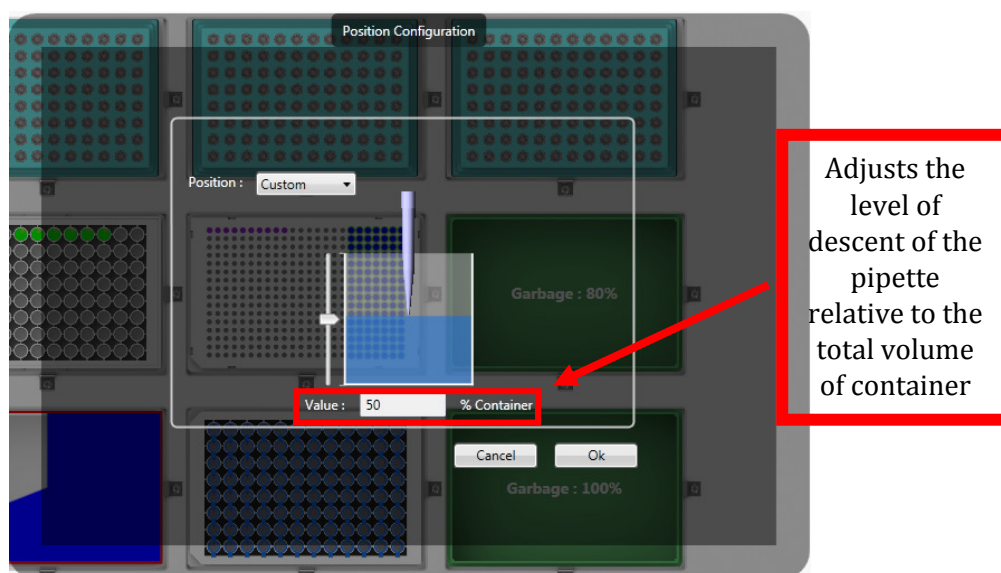


Figure 42: Cone position in the liquid (« container »)

Repeat the step « **B/Add a sequence** » as many times as desired for a complete protocol.



b. «Auto» Mode

The «Auto» mode has been created to improve your protocol by selecting automatically the pipette to be used and the protocol to set up to execute a given task.

In the « **Auto** » mode, you have to define the source format you want, no matter the pipettes which are connected to the workstation. You have the possibility to choose a group of wells as source, for one-track pipette, or a group of rows of wells, for 8-tracks pipette. But these wells have to belong to the same consumable. However, the format of the target will depend on the format of the source. The format of the target must thus correspond to the format of the source or a multiple of there:

- If the amount of liquid to be dispensed into each well does not exceed the capacity of the available pipettes, the pipette will change the cone each time it finishes copying the source model.
- If the amount of liquid to be dispensed into each well beyond the capacity of the available pipette, the number of cone depend on the volume used to dispense. The pipette will change tips after 4 aspirations to ensure a good quality of pipetting.

■ The « **Auto** » mode has other specific function, depending on the size of robot described below:

■ αBot™ robot:

The αBot™ robot is empowered with 3 axes which are: 1 axe X, 1 axe Y and 1 axe Z.
One pipette is fixed on the Z axe.

Thanks to the «**Auto**» mode, you can ask the workstation to launch a model duplication in one click instead of doing it well by well (or row by row if it is an 8-tracks pipette) under the manual mode.

To do so, follow the steps as you would do under the manual mode, but when you get to the sequence creation step, choose «**Auto**». There, you are able to select the sources, the target and the volume to dispense.



In this part, some examples are given for explain the specificity of the «**Auto**» mode when you choose sources and targets wells:

1. Multichannel pipettes (8-channels):

Some duplications models, with the 8-channels pipette, are described below:

■ Example 1: 96 tips used.

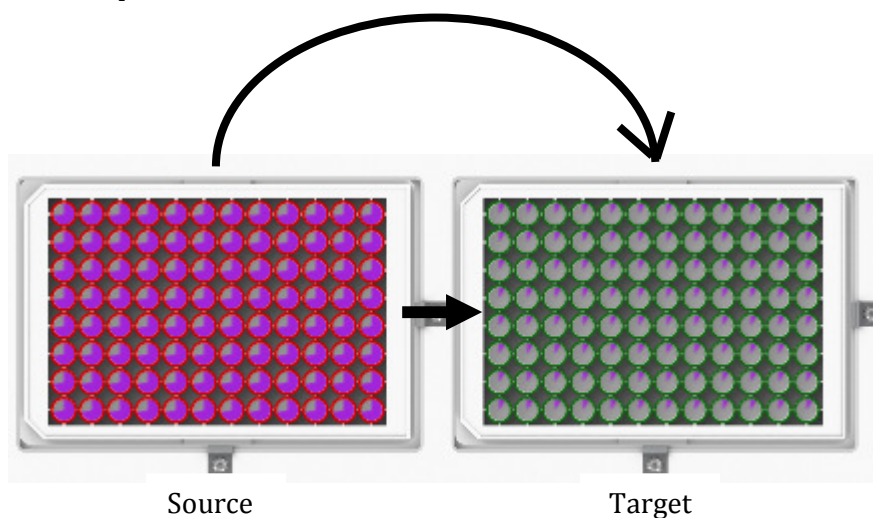


Figure 43: Model duplication - Example 1

Duplication of a 96 well microplate to another 96 well microplate.

■ Example 2 : 64 tips used

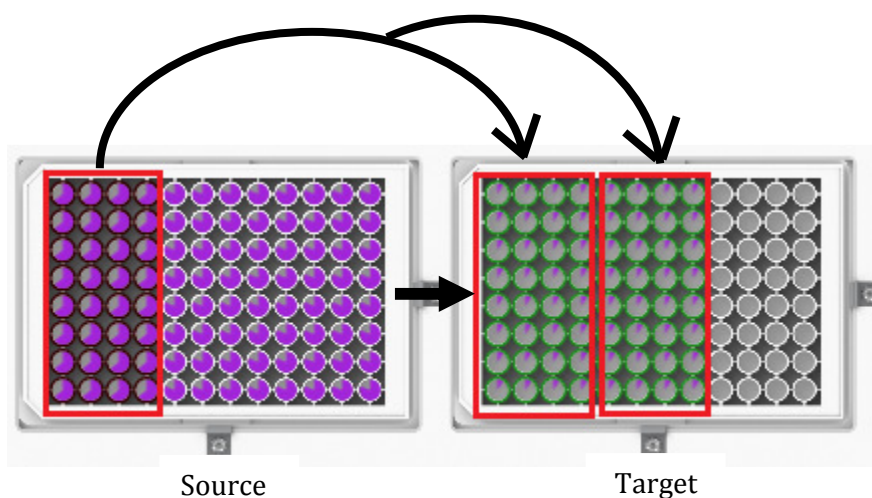


Figure 44: Model duplication - Example 2

Duplication of part of a 96 well microplate to another 96 well microplate.



■ Example 3 : 96 tips used

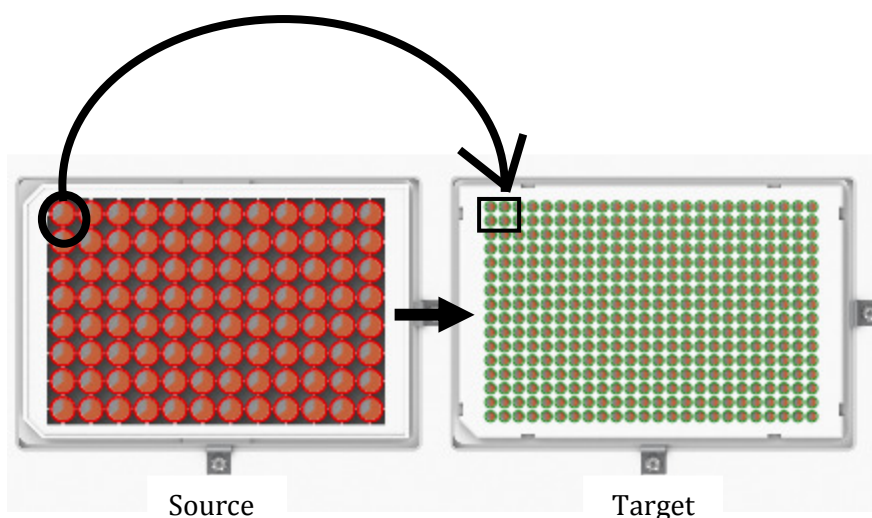


Figure 45: Model duplication - Example 3

Duplication of 96 well microplate to a 384 well microplate.

2. Single channel pipette:

■ Example 1 : 5 tips used

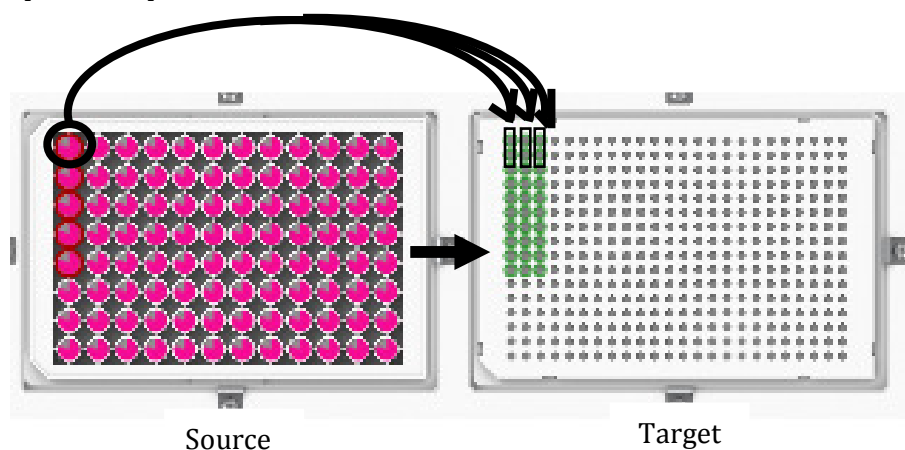


Figure 46: Model duplication - Example 4

6 replicates of 5 wells from a 96 well microplate to a 384 well microplate.



Example 2 : 7 tips used

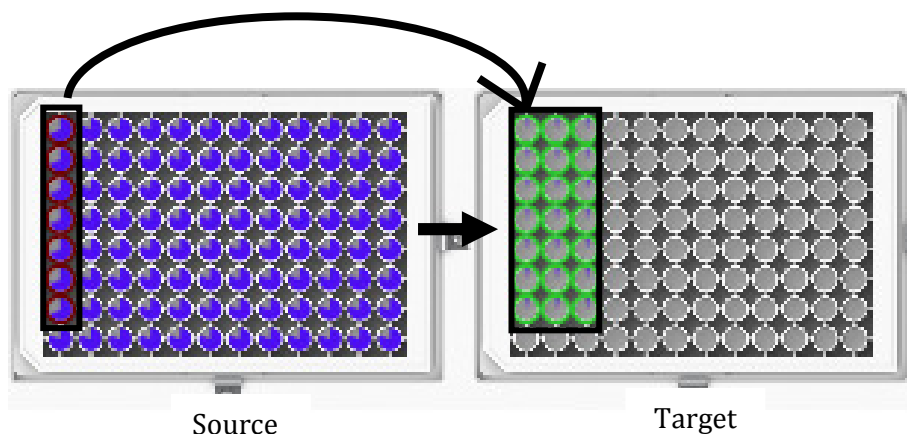


Figure 47: Model duplication - Example 5

3 replicates of seven wells from one 96 well microplate to another 96 well microplate.

C. Protocol Validation

Once the creation of the protocol has finished, click on «**Validate**» to get to the next step. Back to the sequence page, click on each sequence to make them visible on the screen. The sources are marked in red and the targets are marked in green.

TROUBLESHOOTING:

This message appears when your source does not contain enough liquid for the task that you ask. So add the volume needed and go to «**Initialization**» stage for that source again.

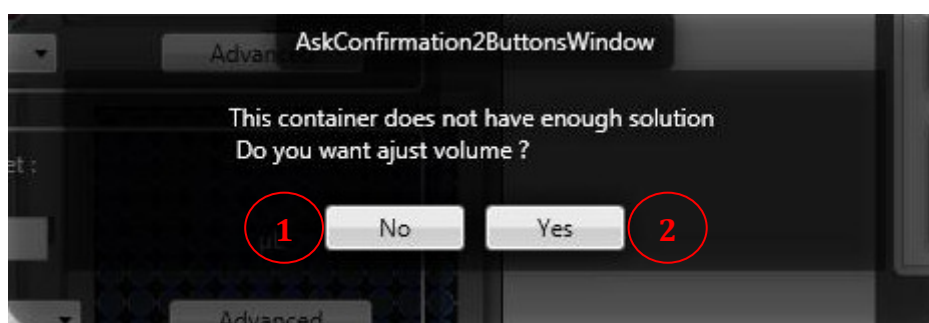


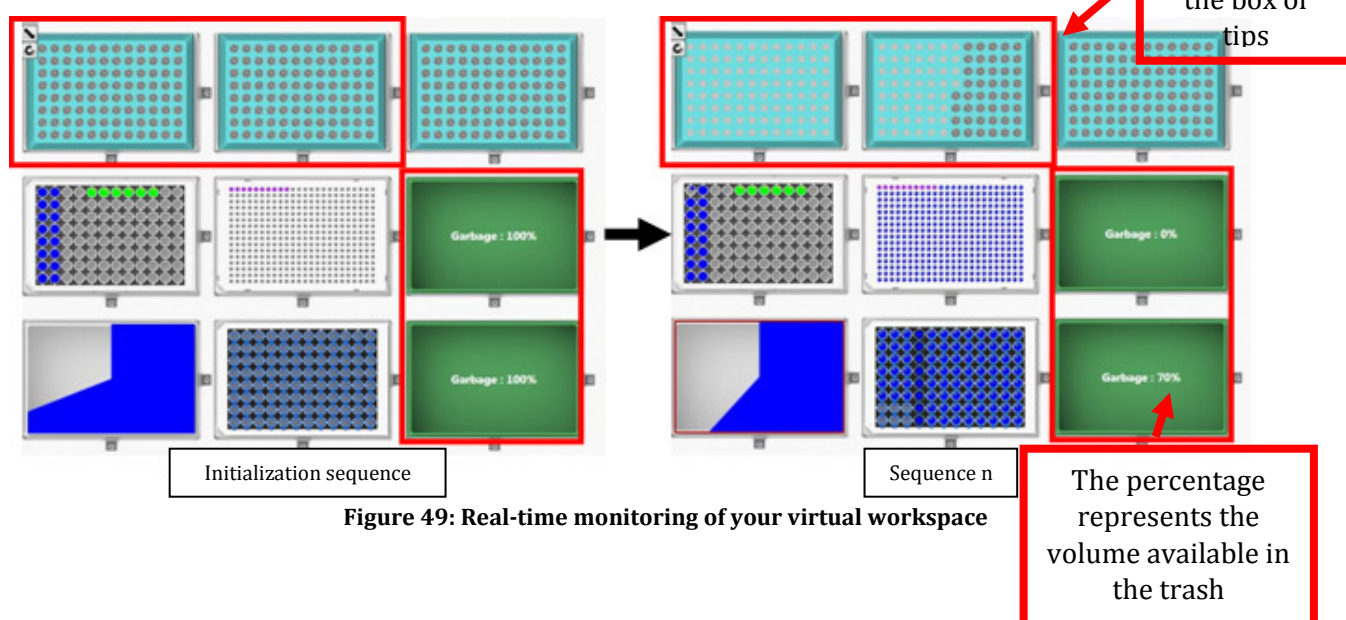
Figure 48: Source volume adjustment

- 1: Clicking on «**No**», you stop the creation of the sequence.
- 2: Clicking on «**Yes**», the window shown in Figure 25 appears. You can then adjust the volume with «**+/-**» button.



REAL-TIME MONITORING OF YOUR VIRTUAL WORKSPACE:

You can follow, in real time, the status of your consumables for each sequence by clicking on the sequence:



SEQUENCE MANAGEMENT:

1. DESCRIPTION OF SEQUENCE TAB:

When you've finished creating your protocol, on the right hand of the window, you can see informations about the sequences that you had created.

Step Number :	2
Step Name :	MultiDispense
Volume Used :	20
Tool Used :	P1V50

Figure 50: Sequence description

- « **Step Number** »: Order of the sequence in the protocol
- « **Step Name** »: Dispense mode of the sequence
- « **Volume Used** »: Volume dispense during the sequence
- « **Tool Used** »: Tool used during the sequence. In this example, a single channel was used (range: 50 μ L).



2. DELETE A SEQUENCE:

You can only delete the last protocol sequence (see the example below). In the example below, you can't delete the sequence 5 if you don't delete first the sequences 7 and 6:

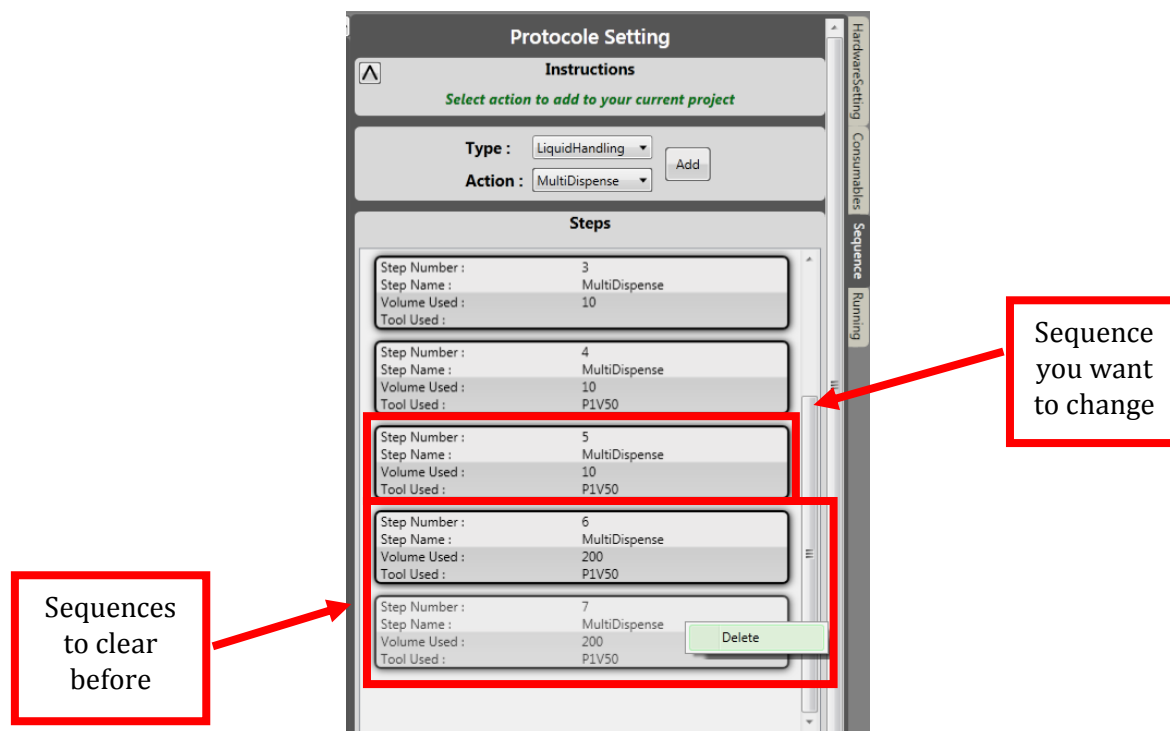


Figure 51: Clearing a sequence

3. SEQUENCE ORDER:

When you create a protocol, you do not have to start this protocol by the first sequence, or end with the last sequence of your protocol. So you can skip the steps already done.

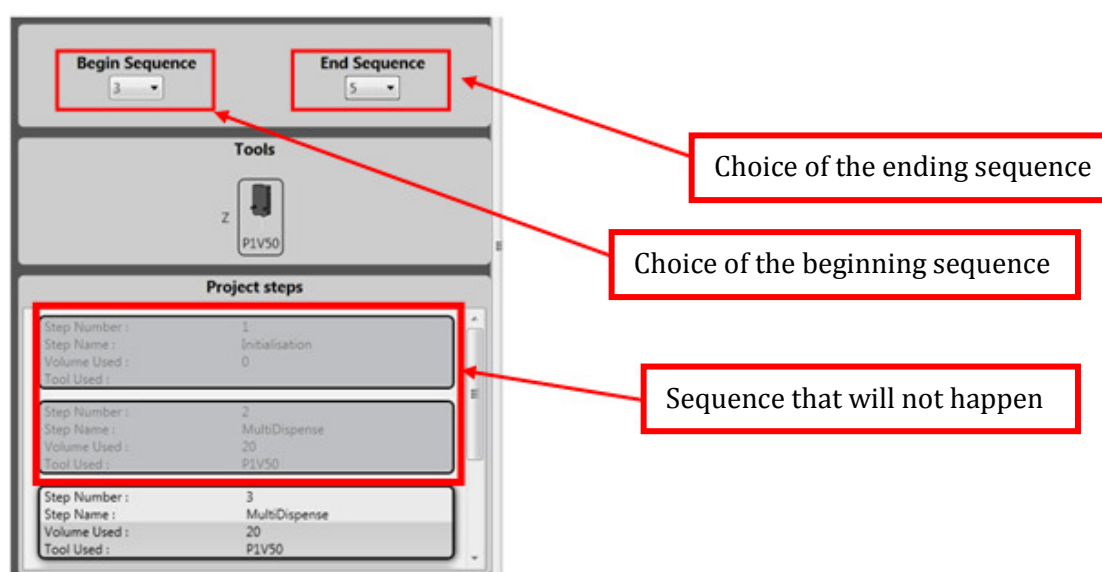


Figure 52: Choice of the sequences order for the protocol's procedure



8.5.4. PROTOCOL VALIDATION AND RUNNING STAGE

After clicking on « **Validate** » on the panel « **Sequences** » the software skips to the panel «**Running**».

Click on « **Check** » to verify the consistence of all sequences. At that step, the software will check if the requested protocol is feasible. If so, the buttons «**Play / Stop**» appear at the bottom of the work space; if not you will get an error message (figure 48).

After the « **Play/Stop** » buttons have appeared, you can launch the execution of the protocol.

✖ **WARNING:** Before launching the protocol, check the state of the consumables especially the tips boxes. Select to clear / add tips.

After clicking on the button « **Play** », it becomes a « **Pause** » button, so you can stop the protocol whenever you want. Clicking on the « **Pause** » button makes it become a button « **Play** » again.

To stop a protocol, click on the button « **Stop** ». Then the pipettes come back to the initial position after having ejected the tips.

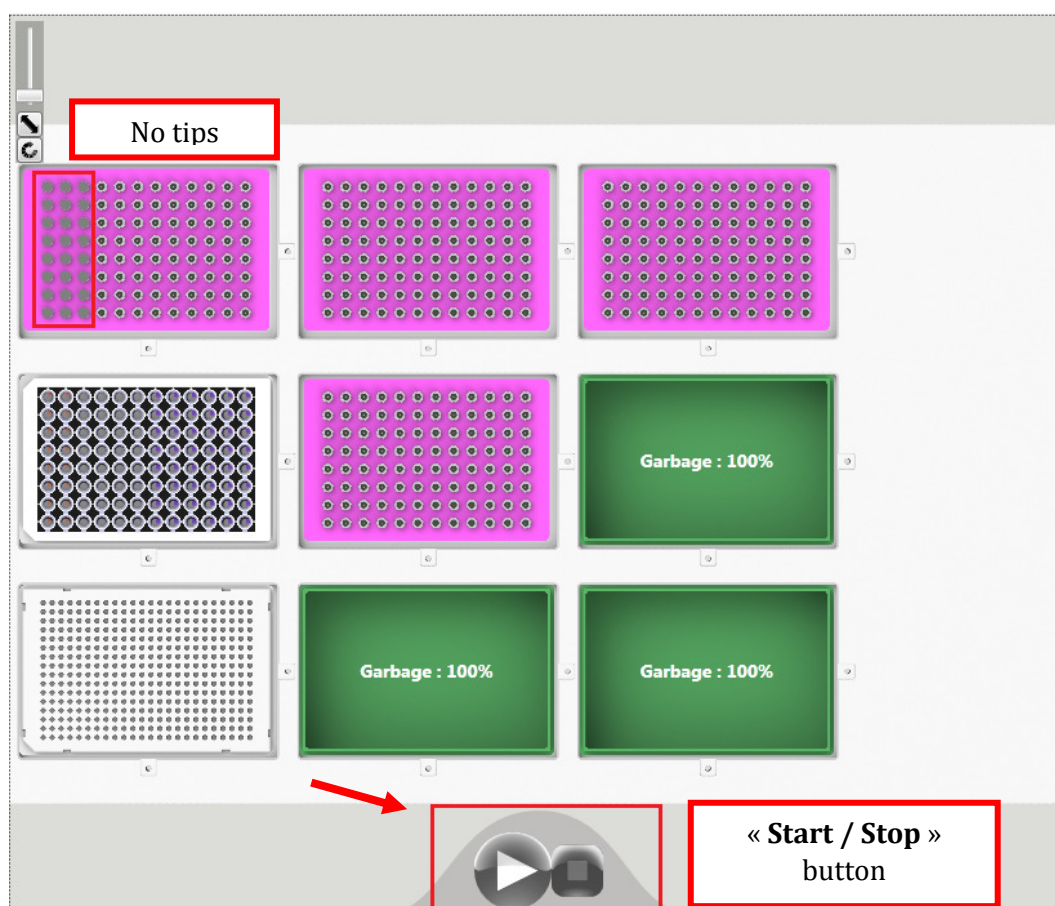


Figure 53: « Running » window

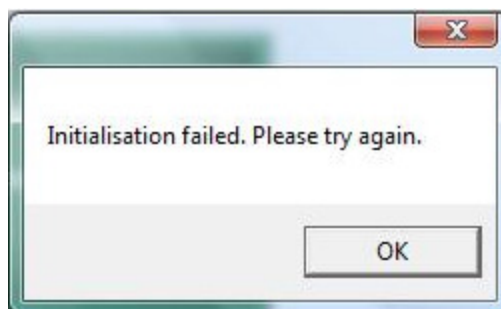


Figure 54: Error message during the running

\\ **WARNING:** the message of figure 48 appears when you click on «**Play**», it means that :

- The α Bot™ is not connected to the PC, check the USB connection,
- The α Bot™ is not « **On** », in this case, check the power cord is properly plugged in or the « **On / Off** » of figure 4 is to 1.
- Or there is a technical problem, in that case please contact AlphaHelix as explained in the chapter 2. **Contacts**.

▀ **PROTOCOL RE-LAUNCHING:**

- ▀ If you want to do the same protocol again, as soon as you click on the button «**Play**», the following message appears, «Please Check Your Tips Box ...». In that case check if all the tips boxes on your space work are well filled.
- ▀ Think to check again if your virtual space work matches to your real space work.



8.6. EDIT FUNCTION

The « **Edit** » tab contains 2 features:

- Liquid Manager
- Pipeting Profile Manager

These options allow the creation, edition, and deletion of liquids and pipeting profile management.

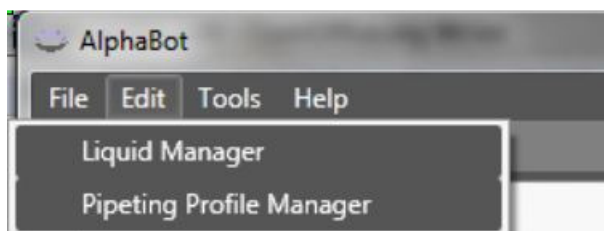


Figure 55: « Edit » function

8.6.1. LIQUID MANAGER

Liquid manager system allows you to create, edit or delete the liquids that will be handled by the workstation within the α Bot™ software.

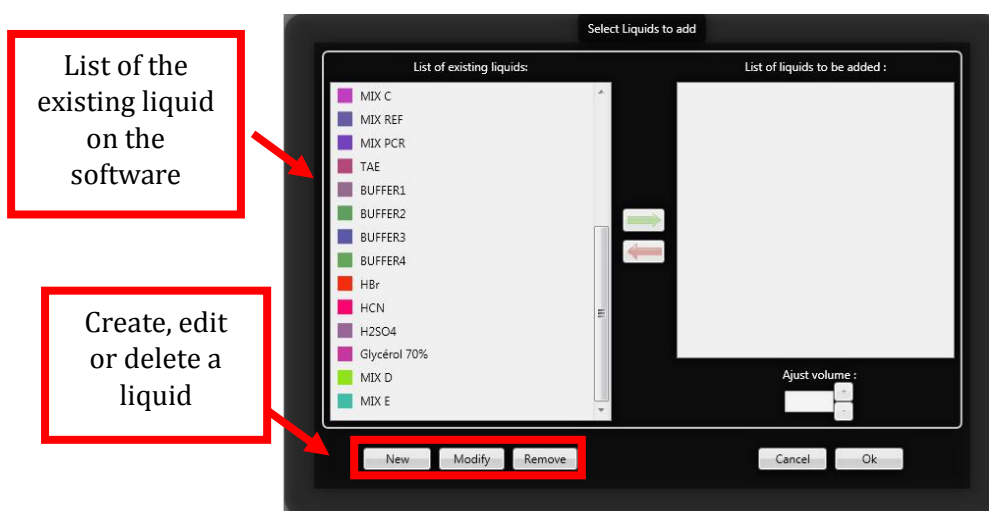


Figure 56: « Liquid Manager » window



A. CREATION OF A NEW LIQUID

There is already a list of predefined liquids in the database software.

If you want to create a new liquid, click « **New** ».

A new window appears which allows you to give a name, to choose a color and a type for your new liquid.



Figure 57: Window for new liquid creation

Name your liquid; choose a color then a type for it. (Define « **Type** » of your liquid depending on its viscosity).

When you press the « **Advanced** » button, a similar window to the « Pipeting Profil Manager » window appears which allows you to assign a specific pipetting mode to the new liquid.

To validate your creation, press « **Ok** » or « **Cancel** » to cancel.

B. EDITION OF A LIQUID

Select the liquid that you want to edit by clicking on then click « **Modify** ». A similar window to the figure 56 window appears. Follow the same instructions to edit the selected liquid.

C. DELETION OF A LIQUID

If you want to remove a liquid from your list, click on the target liquid and press « **Remove** ». Your liquid will be removed from the list.

✖ **WARNING:** Be sure that the liquid that you want to remove is not used in another protocol.



8.6.2. PIPETING PROFILE MANAGER

This feature allows you to edit or delete a pipetting profil.

The « **Standard** » protocol is a predefined protocol set by default. This pipetting mode is suitable for pipetting liquid with viscosity approaching water's viscosity.

⚠ **WARNING:** Standard protocol is not editable.

On this window, you would be able to edit some parameters (See details on page 55):

- Airgap (1, 2, 3)
- Aliquot (Start / End)
- Delay (Aspiration / Dispense)
- Speed (Aspiration / Dispense)

These features must be editing depending physical characteristics like viscosity, volatility, etc.

A. CREATION OF A NEW PIPETTING PROFIL

To create a new pipetting protocol, clicK on « **New** » in the «**File**» menu.

Name your pipetting protocol.

All features are set at default values.

Figure 58: Creating a new pipetting protocol window



B. DESCRIPTION OF FEATURES:

■ « **Type** » : you can choose protocol's type corresponding to a pipette :

Different volume range:

- 10 μ L pipette
- 50 μ L pipette
- 250 μ L pipette
- 1000 μ L pipette

■ « **Airgap** »: the unit is the μ L; it corresponds to the air's volume that would be aspired on the tip.

- « **Airgap 1** » : Allows to eject the last drop contained on the tip
- « **Airgap 2** » : Avoids drops falling during workstation arm movements
- « **Airgap 1b** » and « **Airgap 2b** » : Modify depending viscosity
- « **Airgap 3** » : Avoids the lost of « **End Aliquot** »

■ « **Aliquot** »: the unit is the μ L

- « **Start Aliquot** » : Increases the quality of multidispense
- « **End Aliquot** » : Amount of reserve's liquid to have greater security when dispense

■ « **Delay Aspiration / Dispense** »: the unit is the ms. these values allow to control the aspiration delay. It is important when using liquids having a viscosity other than that of water. It is the delay between each aspiration / dispense

■ « **Speed Aspiration / Dispense** »: the unit is UA. 1 is the slower speed and 6 the faster. This value is important when using liquids that having a viscosity other than that of water.

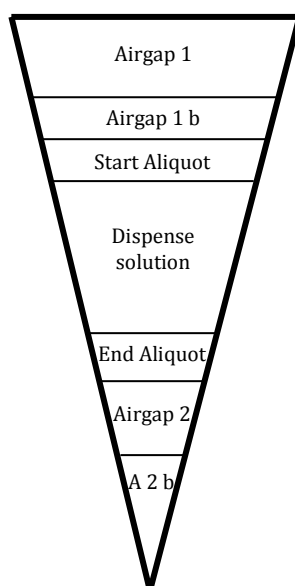


Figure 59: Figure representing a tip with pipeting profile



You would find the AlphaHelix aspiration / dispense profile in the appendices.

You can save the new pipetting protocols by clicking on « **Save** » in the «**File**» menu then confirm by clicking « **Ok** ».

C. DELETION OF A PIPETING PROFIL OR CANCEL CHANGES

To erase a pipetting protocol, choose the one you want to erase on the «**Name**» list then click on «**Delete**».

To cancel changes, click on «**Cancel**».

- ⚠ **WARNING:** When you erase a pipetting protocol, all the liquids you associate with this pipetting protocol will be change with standard protocol.



8.7. ADVICES

There are several instructions to respect when using the α Bot™ software.

8.7.1. MULTICHANNELS (8 CHANNELS) PIPETTE USE

During aspiration, the software makes calculations to always go down to 2mm depth in the liquid.

Therefore, when using a multichannel pipette (8 channels), the amount of liquid, in the wells, will always be dispensed in the same way.

And even if the wells where the pipette aspires, containing different liquids.

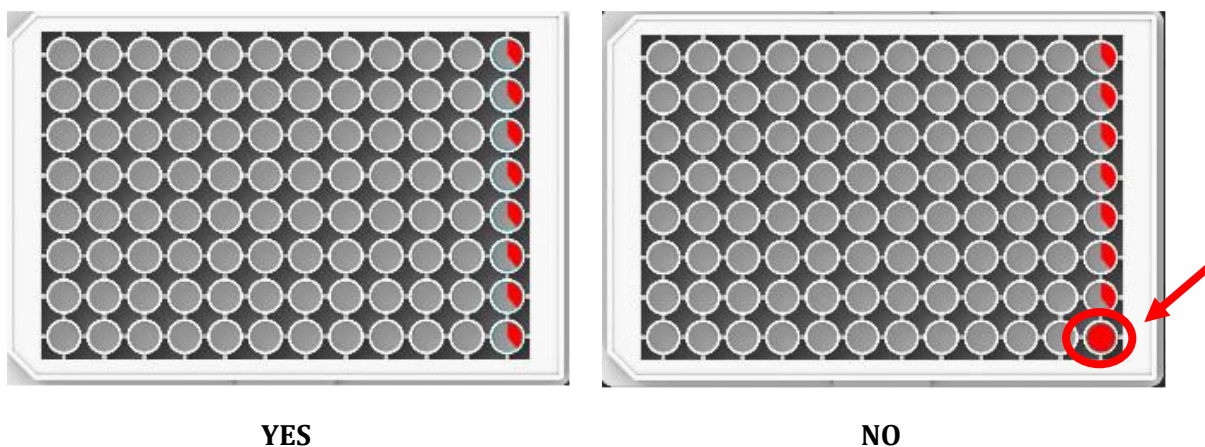


Figure 60: Instruction for using an 8 channel pipette



8.8. TROUBLESHOOTING

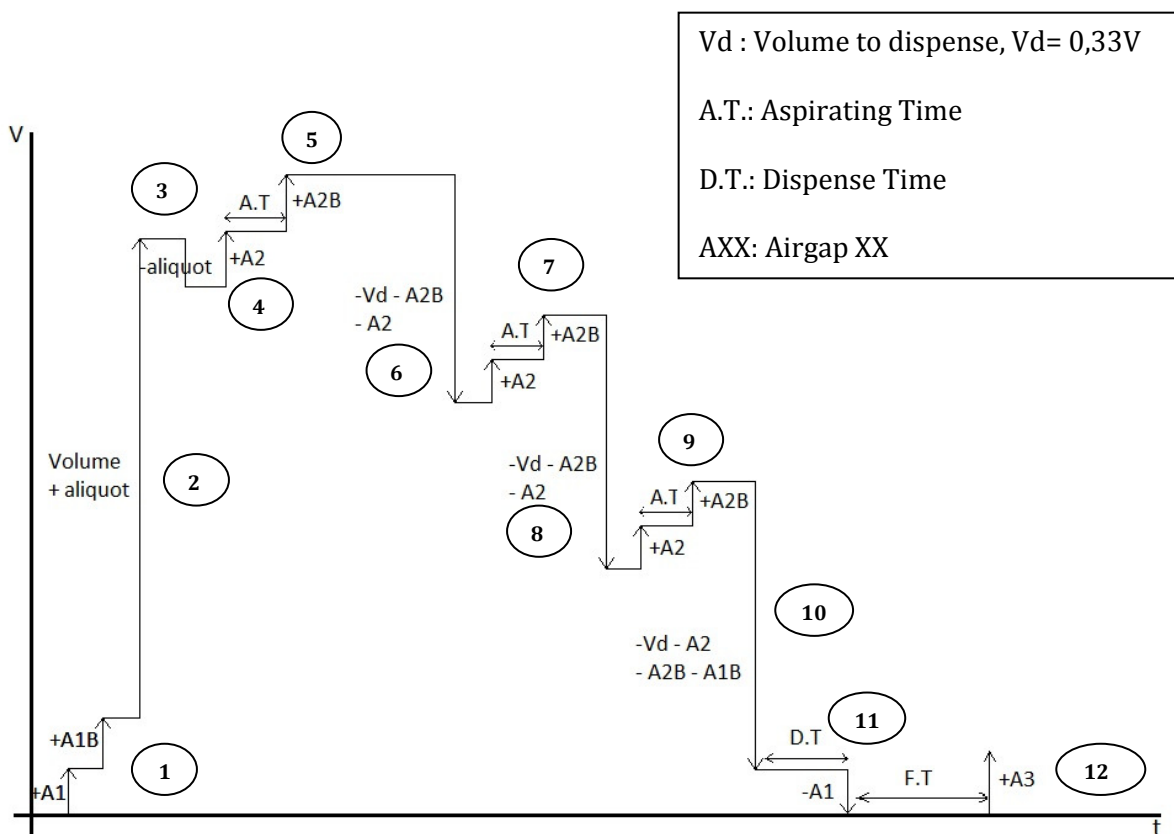
If your workstation does not work:

- Make sure your power outlet is plugged,
- Make sure that the button « **On / Off** » (Figure 4) is on 1,
- Check the status of your fuse near the power switch « **On / Off** » (Figure 4). If the problem is the fuse, here are its specifications: 250V, 4A; Housing material: ceramic; Size: 5x20 mm,
- If the problem persists, please contact your local distributor, paragraph **2. CONTACTS**.



9. APPENDIX

9.1. PROFIL OF ASPIRATION/DISPENSE OF ALPHAHELIX



The picture above sums up the profile of aspiration/dispense carried out by the AlphaHelix® pipettes.

The graph describes three times dispensation of a solution:

1. Airgap 1 and 1b 's aspiration
2. Solution and Start Aliquot 's aspiration
3. Start Aliquot's ejection
4. Airgap 2's aspiration
5. Airgap 2b's aspiration
6. First volume of the solution dispense and Airgap 2 and 2b's ejection
7. Airgap 2 and 2b's aspiration
8. Second volume of the solution dispense and Airgap 2 and 2b's ejection
9. Airgap 2 and 2b's aspiration
10. Third volume of the solution dispense and Airgap 2 and 2b's ejection
11. Airgap 1's ejection
12. Airgap 3's aspiration



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