

Cubis® II

Pharma Filling and Mycap® CCX Application

System Installation and Usage of the Cubis® MSA Application YAPP19 and the Cubis® II MCA Applications Pharma Filling (QAPP008) and Mycap® CCX (QAPP009)

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1 Purpose

This guide describes the necessary components and settings for the Cubis® MSA software application YAPP19 and the Cubis® II MCA software applications Pharma Filling (QAPP008) and Mycap® CCX Cell Expansion (QAPP009).

2 Description

2.1 Software Application YAPP19

The Cubis® MSA software application offers two different workflows:

1. Filling of liquid products in vessels or bags
2. Fluid transfer for diluting cell cultures and filling with medium

Both applications have in common that liquid is transferred by means of the pump models 323Du, 530Du(N) or 630Du(N) from Watson-Marlow via a serial connection.

When filling products, product data and pump settings are stored in a product database. The start, fill and final speed of the pump can be set product-specifically. In addition, a reverse run can optionally be defined to pump liquid back in feeding hoses so that this portion is not counted as fill weight. The filling process is repeated according to the set number of samples and the results stored batch-specific in the product database. In addition, for filled containers or bags, labels may be printed including information such as sample number, filling weight and expiration date.

For cell culture applications, medium can be adjusted to the desired volume or inoculum diluted to the desired cell density. All volume and concentration calculations are carried out automatically and the fluid transfer through the peristaltic pump is controlled accordingly by the software application. All settings and results are stored in an experiment database. As with the filling of products, it is also possible to print labels for cell culture applications with cell line information, batch number, passage number, cell density and volume.

2.2 Software Application Pharma Filling (QAPP008)

The application Pharma Filling is used to fill liquid products using peristaltic pumps. By means of the pump, the liquid is transferred from a reservoir into vessels or bags and the transferred weight is checked gravimetrically. The application can control and regulate the pump models 323Du, 530Du(N) and 630Du(N) from Watson-Marlow via a serial communication.

When filling products, product data and pump settings are stored in a product database. The start, fill and end speeds of the pump can be set product-specifically. In addition, a reverse run can optionally be defined to pump liquid back in the feeding hose so that this portion is not counted as fill weight. There are four phases of a filling operation whereby the Cubis® MCA balance regulates the speed of the peristaltic pump:

- **Start**, which is typically used for priming the line to clear air from the tubing up to the receiving container(s). The operator presses a button on the balance to stop this step. Priming can be repeated if stopped prematurely.
- **Fill**, to transfer fluid into the receiving container.
- **Slow Down/End**, where the balance will signal the pump to slow once the transferred weight reaches a specified percentage of the total fill volume and stop the pump when the total fill volume is met.
- **Reverse Run** is an optional step the operator can trigger to pump liquid back towards the reservoir so residual liquid in tubing is not counted as fill weight.

The filling process is repeated according to the set number of samples and the results are calculated automatically. The filling weight in each vessel or bag, the minimum, maximum and average filling weight, as well as the standard deviation are determined in the statistical evaluation. In addition, for filled vessels or bags labels can be printed, including information such as sample number, filling weight and expiry date.

2.3 Software Application Mycap® CCX Cell Expansion (QAPP009)

The software application Mycap® CCX Cell Expansion is designed for the aseptic transfer of media or inoculum between Mycap® CCX flasks using peristaltic pumps. By means of the pump, media or inoculum is transferred from a donor flask into a recipient flask and the amount transferred is checked gravimetrically. The application can control and regulate the pump models 323Du, 530Du(N) and 630Du(N) from Watson-Marlow via a serial communication.

To define an experiment, there is an administrator and a user menu in which various parameters are recorded. The most important parameters are information on the cell density in the donor flask and the desired cell density in the target flask, as well as information on the available and desired volume of the medium in the target flask. In addition, the pump speed for different process steps can be defined in rpm. All settings are stored in an experiment database. From the concentration and volume data, the application automatically calculates the target weight of the inoculum or medium to be transferred and controls the peristaltic pump accordingly. When executing a program the operator can choose to transfer media and inoculum, media only or inoculum only. There are four phases of a transfer operation whereby the Cubis balance regulates the speed of the peristaltic pump:

- **Start**, which is used for priming the line to clear air from the tubing up to the receiving flask. The operator presses a button on the balance to stop this step. Priming can be repeated if stopped prematurely.
- **Fill**, to transfer fluid into the receiving container.
- **Slow Down/End**, where the balance will signal the pump to slow once the transferred weight reaches a specified percentage of the total fill volume and stop the pump when the total fill volume is met.
- **Reverse Run** is an optional step the operator can trigger to pump liquid back from the receiving flask so residual liquid in tubing is not counted as fill weight.

Media filling speed, slow down point and slow down speed are different from inoculum filling speed, slow down point and slow down speed. These settings are stored in the program.

The process is repeated according to the number of flasks to be filled until all samples have been processed.

The results including the final volume and the effective final cell density are automatically calculated for each sample and can be documented by means of a printer connected to the balance. You can also optionally print labels with cell line information, lot number, passage number, cell density, and the volume to label the filled flasks.

The application uses the Solution Dilution Formula to calculate the amount of media and culture to add to the receiving flask.

Solution Dilution Formula: $C_1V_1 = C_2V_2$

The following variables are fixed in the experiment setup:

- C_2 is the targeted viable cell density (VCD) in the receiving flask
- V_2 is the targeted volume in the receiving flask

The following variable is entered by the operator when executing an experiment:

- C_1 is the VCD in the donating flask

The application calculates for V_1 which is the volume of cell culture to add to the receiving flask to meet the targeted VCD.

The application then calculates the amount of media to add to the receiving flask by subtracting V_1 from V_2 . Media density is required for proper volume concentration and is fixed in the experiment setup.

The application reports the expected VCD in the receiving flask by use of the Solution Dilution Formula and using the VCD from the donating flask and actual culture and media volumes transferred to the receiving flask.

2.4 Peristaltic Pumps and Connection Cables

Sartorius recommends and sells Watson-Marlow peristaltic pumps for processing Mycap® CCX samples and filling pharmaceutical products into bags, flasks or vessels. Either the IP31 protected standard version of pump models 323Du, 530Du or 630DU connected by a 9-pin to 9-pin

serial cable to the balance or pump models 530DuN or 630 DuN with IP66 protected NEMA 4x module connected by a 9-pin to M12 serial cable with special wiring can be used with the QApps. Sartorius sells the IP31 protected pump model 323Du with two different pump heads and the IP66 protected pump models 530DuN and 630DuN.

Item number	Plug	Product description
YDU-30	EU	Watson-Marlow peristaltic pump 323DU/D2 with pump head 313DW2 for 2.4 WT tubes, max. 400 rpm, Sartorius colored case top and pump 100-120V/200-240V, 50/60Hz, 1pH (subject to regional cord set and supply)
YDU-31	US	
YDU-32	UK	
YDU-33	South Africa	Watson-Marlow peristaltic pump 323DU/D with pump head 313DW for 1.6 WT tubes, max. 400 rpm, Sartorius colored case top and pump head, 100-120V/200-240V, 50/60Hz, 1pH (subject to regional cord set and supply)
YDU-34	India	
YDU-35	EU	
YDU-36	US	Watson-Marlow peristaltic pump 530DUN/R3 with pump head R3 for 3.2 WT tubes, max. 220 rpm, Sartorius color case top and keypad, 100-120V/200-240V, 50/60Hz, 1pH (subject to regional cord set and supply), IP66 protected NEMA module with 5 pole M12 signal connector wired to RS232 adapter
YDU-37	UK	
YDU-38	South Africa	
YDU-39	India	Watson-Marlow peristaltic pump 630DUN/R with pump head R for 3.2 WT tubes, max. 265 rpm, Sartorius colored top case and keypad, 100-120V/200-240V, 50/60Hz, 1pH (subject to regional cord set and supply), IP66 protected NEMA module with 5 pole M12 signal connector wired to RS232 adapter
YDU-50	EU	
YDU-51	US	
YDU-52	UK	Watson-Marlow peristaltic pump 630DUN/R with pump head R for 3.2 WT tubes, max. 265 rpm, Sartorius colored top case and keypad, 100-120V/200-240V, 50/60Hz, 1pH (subject to regional cord set and supply), IP66 protected NEMA module with 5 pole M12 signal connector wired to RS232 adapter
YDU-53	South Africa	
YDU-54	India	
YDU-60	EU	Watson-Marlow peristaltic pump 630DUN/R with pump head R for 3.2 WT tubes, max. 265 rpm, Sartorius colored top case and keypad, 100-120V/200-240V, 50/60Hz, 1pH (subject to regional cord set and supply), IP66 protected NEMA module with 5 pole M12 signal connector wired to RS232 adapter
YDU-61	US	
YDU-62	UK	
YDU-63	South Africa/India	

Item number	Product description
YCC04-D25M6	Cable D-SUB 9-pin (M) to D-SUB 25-pin (M) for connecting IP31 protected pumps 323Du, 530Du and 630Du to Cubis® MSA balances
YCC-D09MM-EC-2.9M	Data cable RS232 9-pin (M) to RS232 9-pin (M) for connecting IP31 protected pumps 323Du, 530Du and 630Du to Cubis® MCA balances
YCC-D09M-M12F-2M	Data cable RS232 9-pin (M) to M12 5-pin (F) for connecting IP66 protected pump models 530DuN and 630DuN to Cubis® MCA balances
YCC-D25M-M12F-2M	Data cable RS232 25-pin (M) to M12 5-pin (F) for connecting IP66 protected pump models 530DuN and 630DuN to Cubis® MSA balances
YDO01MS-R	25-pin 2nd RS232 interface for Cubis® MSA

3 System Installation

3.1 Connecting Pump Models 323Du, 530Du or 630Du to Cubis® MSA Balances

The Cubis® MSA software application YAPP19 can control Watson-Marlow 323Du, 530Du and 630Du peristaltic pumps. To connect the pump to the balance the 25-pin serial RS232 port is used. The Sartorius serial cable YCC04-D25M6 or alternatively cable 69Y03295 plus gender changer and null modem connector (see Figure 1) may connect pump model 323Du, 530Du and 630Du to Cubis® MSA balances. (The gender changer and null modem connector are not available from Sartorius but very common parts on the electronics equipment market.)

Gender changer connector Null modem connector



Figure 1: Gender changer and null modem connector for serial cables 69Y03295 or YCC04-D25M6

Install the null modem connector at the 9-pin plug of serial cable 69Y03295 or YCC04-D25M6. Then install the gender changer connector on the null modem connector and finally plug in the setup at the 9-pin serial port of the pump (see Figure 2):

YCC04-D25M6

69Y03295



Figure 2: The images show the correct setup at Watson-Marlow 323Du pump using cable YCC04-D25M6 or cable 69Y03295 plus null modem and gender changer.

Plug in the 25-pin plug of the serial cable at the standard peripheral serial port (COM A) of the balance or e-box. If using a printer in parallel install the additional serial port YDO01MS-R at the free slot of the balance or e-box and connect the printer to this port (COM C) (see Figure 3).

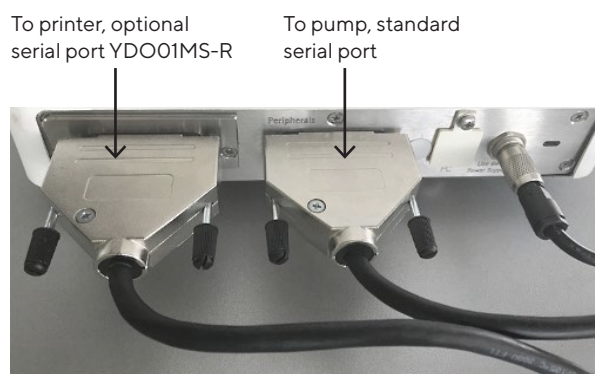


Figure 3: Correct setup of serial ports at the Cubis® MSA balance or e-box if installing the second serial 25-pin port YDO01MS-R to connect in parallel a printer to the balance.

3.2 Connecting Pump Models 530DuN or 630DuN to Cubis® MSA Balances

Pump models 530DuN and 630DuN (YDU-50 to YDU-63) with IP66 protected 4x NEMA module and M12 input with special wiring according to Sartorius layout require a different cable for connection to Cubis® MSA balances as aforementioned pump models 323Du, 530Du and 630Du. Serial connection cable YCC-D25M-M12F-2M is used to connect pump models 530DuN and 630DuN to Cubis® MSA balances (see Figure 4):

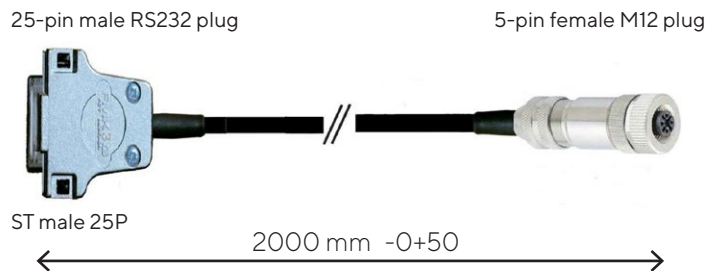


Figure 4: Cable YCC-D25M-M12F-2M to connect IP66 protected pump models 530DuN and 630DuN to Cubis® MSA balances

As shown in Figure 3, plug in the 25-pin male plug of cable YCC-D25M-M12F-2M at the standard peripheral serial port (COM A) of the balance or e-box. Fasten the female M12 plug at the M12 socket located at the pump (see Figure 5):

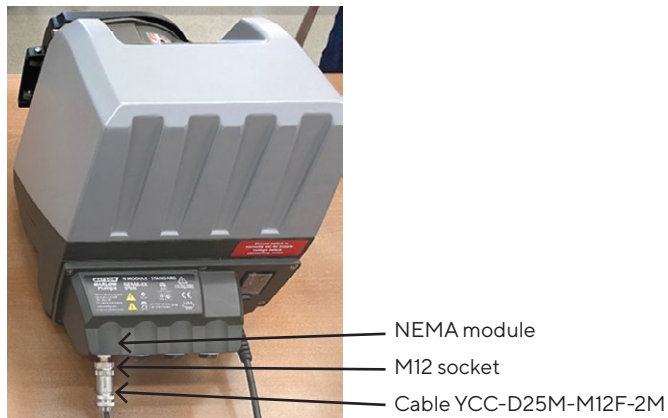


Figure 5: Cable YCC-D25M-M12F-2M fastened to the M12 inlet of pump model 630DuN

3.3 Connecting Pump Models 323Du, 530Du or 630Du to Cubis® II MCA Balances

The Cubis® II MCA software applications Final Pharma Filling (QAPP008) and Mycap® CCX cell passing (QAPP009) can control Watson-Marlow 323Du, 530Du and 630Du peristaltic pumps. To connect the pump to the balance the 9-pin serial RS232 ports at the pump and balance are used. Sartorius offers cable YCC-D09MM-EC-2.9M to connect pump and balance. Fasten one end of the cable at the 9-pin serial port of the peristaltic pump and the other end at the 9-pin serial port of the balance (see Figure 6):

Pump model 323Du,
530Du or 630Du

Cubis® II MCA balance

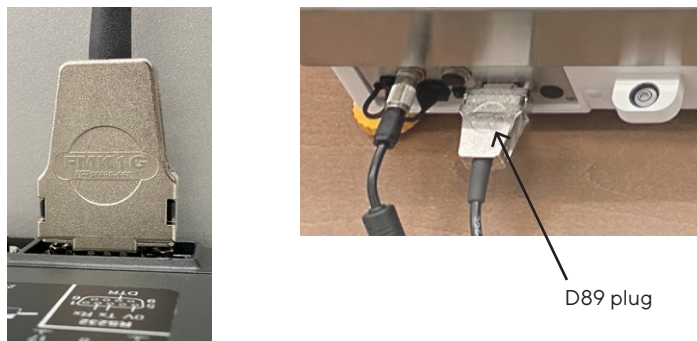


Figure 6: Connecting pump models 323Du, 530Du or 630Du to Cubis® II MCA balances using cable YCC-D09MM-EC-2.9M

3.4 Connecting Pump Models 530DuN or 630DuN to Cubis® II MCA Balances

Pump models 530DuN and 630DuN (YDU-50 to YDU-63) with IP66 protected 4x NEMA module and M12 input with special wiring according to Sartorius layout require cable YCC-D09M-M12F-2M for connection to Cubis® II MCA balances (see Figure 7):

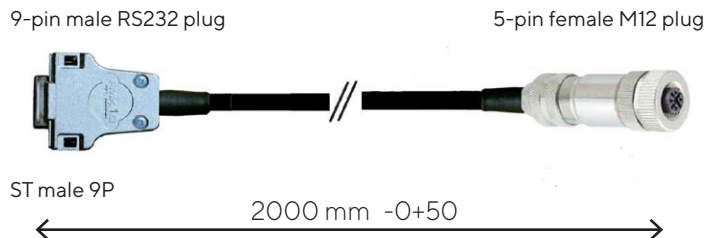


Figure 7: Cable YCC-D09M-M12F-2M to connect IP66 protected pump models 530DuN and 630DuN to Cubis® II MCA balances

Fasten the 9-pin serial male plug at the 9-pin serial port of the balance and the female M12 plug at the M12 socket located at the pump (see Figure 8):

Pump model 530DuN
or 630DuN

Cubis® II MCA balance

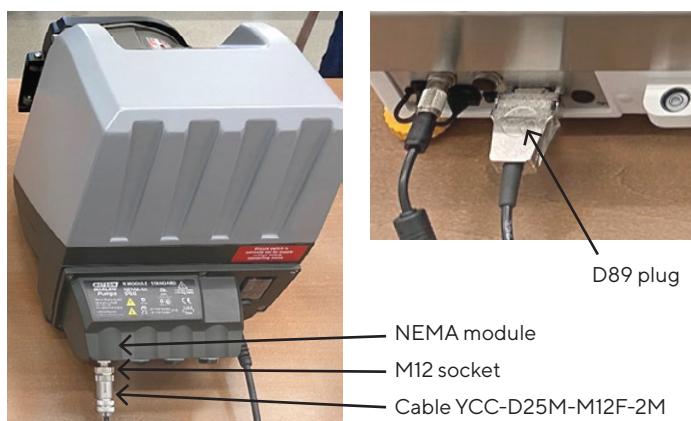


Figure 8: Connecting pump models 530DuN or 630DuN to Cubis® II MCA balances using cable YCC-D09M-M12F-2M

3.5 Pump Settings

Regardless if the peristaltic pump is connected to a Cubis® MSA or Cubis® II MCA balance, the pumps must have the settings as described below in order to be addressable by the software applications.

3.5.1 Pump Model 323Du

For pump model 323Du set the pump to digital mode. Press 2x on the arrow key highlighted in yellow on the keypad to switch to digital mode (see Figure 9). On the key an arrow or the text “mode” is printed. In the display “dig” must be shown.

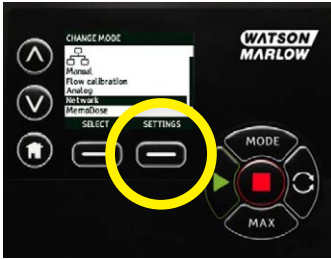


Figure 9: Set pump model 323Du to digital mode

Note: Each time the pump is switched on this procedure must be repeated as the instrument does not save the digital mode setting.

3.5.2 Pump Models 530Du(N) and 630Du(N)

For pump models 530Du(N) and 630Du(N) the network must be set to basic mode. Access the settings menu, scroll to menu item “Network” using the cursor keys up and down and press on the context-sensitive key “Settings”, then select “Basic” mode. The buttons “Settings” and “Basic” are highlighted in yellow in the following images:



The pumps offer to set a speed limit and by factory default the speed limit for pump model 630Du(N) is not set to the maximum speed of 265 rpm but to 165 rpm. The pump speed limit must not be set lower than the maximum rpm value set for a Mycap® experiment or filling program in the software applications (both QApps will create an error message when starting the selected experiment or program). To check or set the speed limit access the



settings menu and select “Control settings”->“Speed limit” using the cursor keys.

Use the \wedge | \vee keys (highlighted in yellow) to adjust the value and press Save to set the value.

3.5.3 Serial Settings of Peristaltic Pumps 323Du, 530Du(N) and 630Du(N)

The serial settings of pump model 323Du are non-editable. Therefore, for model 323Du, it is sufficient to set the pump to digital mode. For pump models 520Du(N) and 620Du(N) the factory serial settings are suited to control the instruments by the Cubis® software applications. However, the serial protocol settings are editable and without correct settings the pumps cannot be addressed by the software applications. The parameters of the serial protocol for the Watson-Marlow pumps must be set as given in Table 1.

Pump model	323Du	520Du(N)/ 620Du(N)
Protocol	RS232	RS232
Pump no.		1
Baud rate	9600	9600
Data bits	8	8
Parity	off	off
Stop bits	2	1
Handshake	off	off

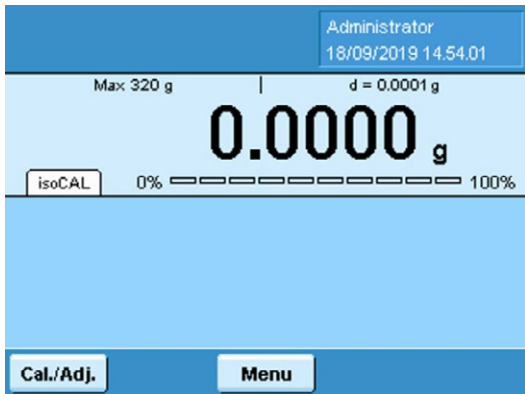
Table 1: Correct serial protocol settings of pump models 323Du, 530Du(N) and 630Du(N)

Note: Please read the chapter *Network Settings* in the pumps manual to set the serial protocol parameters as listed in Table 1.

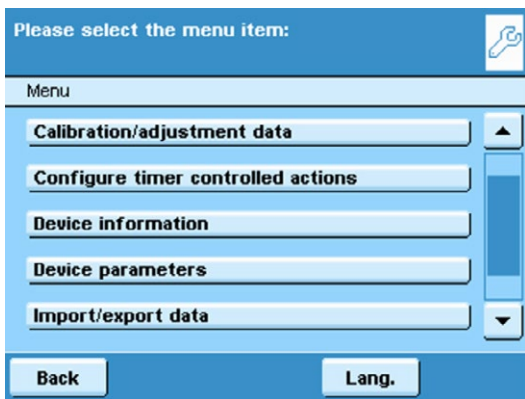
3.6 Cubis® MSA Balance Settings

3.6.1 Configure Serial Ports

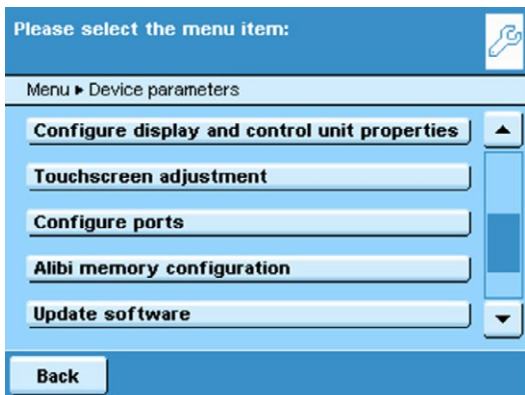
1. Go to Weighing screen and select “Menu”



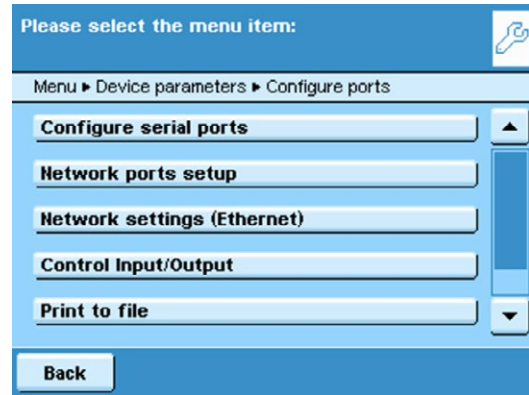
2. Select “Device Parameters”



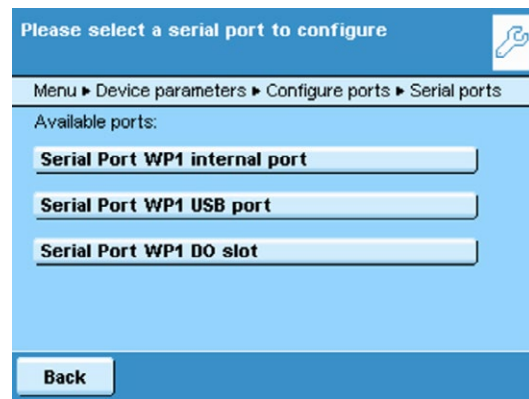
3. Scroll down and select “Configure Ports”



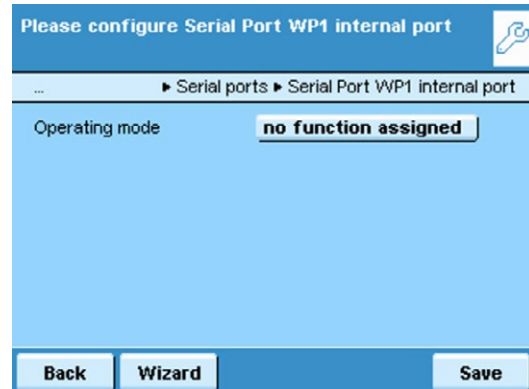
4. Select “Configure Serial Ports”



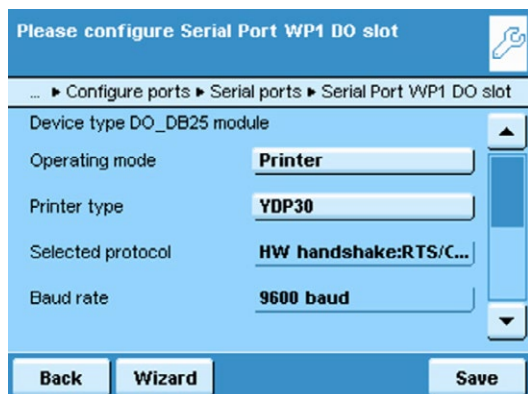
5. Set port WP1 internal to “No function assigned”
 - Select “Operating Mode”
 - Select “No Function Assigned”
 - Select “Save”
 - Select “Back” until returned to Weighing



Note: The displayed ports differ with the setup of the balance. As standard the ports WP1 internal (COM A) and WP1 USB (COM B) are available. If an additional serial port (YDO01MS-R) is installed additionally WP1 DO (COM C) slot becomes available.



Set port WP1 internal (COM A) to “no function assigned” and save the settings.



Optional: If a printer is connected to the additional serial port WP1 DO (COM C) select operating mode “Printer” and select the connected printer type. Then save the settings.

3.6.2 YAPP19 Task Installation and Settings

For the import, task creation and activation of YAPP19, please read the instructions in file “Getting started” from the download package. Please check the listed COM ports during the installation (see Figure 10). The serial ports are listed for the menu item “Printout interface”. COM A must not be listed as the serial port is set to “no function assigned” (see 3.6.1). COM B might be listed if the default settings are used and COM C is listed if a second, additional serial port YDO01MS-R is installed at the balance and if COM C is configured as printer port.

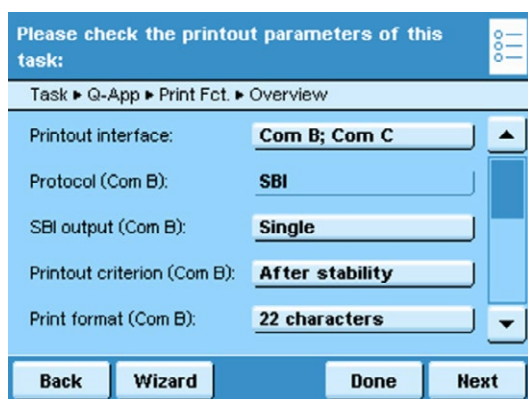


Figure 10: Printout interface settings for YAPP19. COM A must not be listed in the printout interface selection.

3.6.3 Pump Selection

1. Start YAPP19
2. Setup filling program or Mycap® experiment
3. Press “Pump selection” in the main menu and select the connected pump model
4. Start a filling program or a Mycap® experiment to check if the pump is addressed properly by the balance.

5. In case you receive an error message please check the instructions given in chapter 3 to identify improper system setup or settings.

3.7 Cubis® II MCA Balance Settings

3.7.1 COM-RS232 Interface Settings

The Cubis® II MCA balance factory settings must be adjusted in order to control peristaltic pumps. Using the unchanged default settings, the pumps cannot be addressed by the software applications.

1. Login as user with role “access settings menu”
2. In the yellow main screen, press on the following button to access the settings menu:



3. In the settings menu, press on “Connections” -> “Interfaces” -> “COM-RS232 interface”
4. Set the parameters for the COM-RS232 interface as given in Table 2:

Pump model	323Du	520Du(N)/ 620Du(N)
Protocol	QAPP direct	QAPP direct
Baud rate	9600	9600
Data bits	8	8
Parity	off	off
Stop bits	2	1
Handshake	off	off

Table 2: Parameters to be set for the COM-RS232 interface

5. Confirm the settings

3.7.1.1 Task Setup and Pump Selection

1. Install and start Final Pharma Filling or Mycap® CCX cell passaging task (see balance user manual). Select if the task uses SQLite or PostgreSQL as database. SQLite is a balance internal database. Mycap® CCX experiments saved to the SQLite database are locally available only at the specific balance whilst using PostgreSQL saved experiments are shared amongst balances (see Figure 11). PostgreSQL is ideal if working with multiple balances. The PostgreSQL database relies on an external, server based database. The infrastructure has to be made available by the customer and the server settings to be filled in during

task setup. Please work with your local IT department to integrate the balances into the local infrastructure.

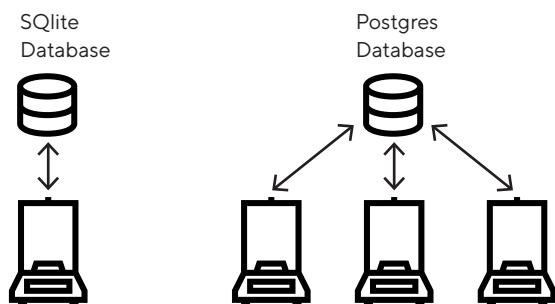


Figure 11: Scheme to illustrate the differences between SQLite and PostgreSQL database infrastructure

2. Setup filling program or Mycap® experiment
3. Press “Pump selection” in the main menu and select the connected pump model

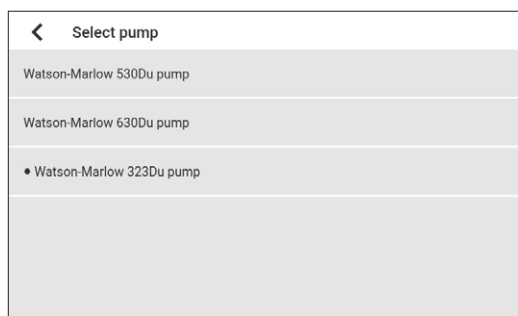


Figure 12: Pump selection screen. The selection is saved and the selected model highlighted by a dot.

4. Start a filling program or a Mycap® experiment to check if the pump is addressed properly by the balance. If the pump can be addressed by the balance the selected program starts and follows the workflow. If the connection to the pump cannot be established the following error message is displayed (see Figure 13):

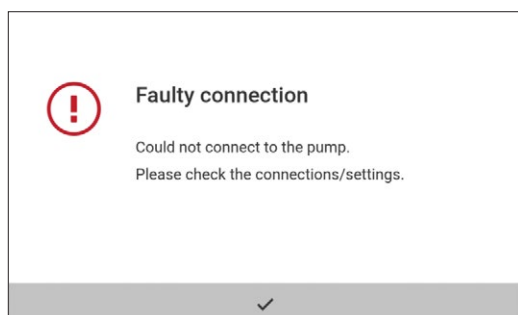


Figure 13: Error message displayed if the peristaltic pump cannot be addressed by the software.

5. Please check the instructions given in chapter 3 to identify improper system setup or settings.

4 Automated Cell Passaging with Mycap® CCX and Cubis® II MCA

4.1 Purpose

Cell passaging is the process of transferring media and cell culture to an Erlenmeyer shake flask for cell expansion. This guidance provides an overview how cell passaging can be automated with Mycap® CCX, Cubis® II MCA Balance with the Mycap® CCX QApp and Watson-Marlow peristaltic pump.

Mycap® consumables, cutting tool and tubing are offered by Sartorius Bioprocess Solutions.

4.2 Create Mycap® Experiment

After task start the main menu is displayed. The menu item “Create/edit Mycap® CCX experiment” is accessible for users with role “Task management” only.

1. Select Create/edit Mycap® CCX experiment in the main menu
2. Select Create experiment in the screen select action
3. Set the following parameters:

Parameter	Function
Experiment name	Mandatory, experiment settings are saved to and loaded from the database using this name
Receiving flask size	Mandatory, volume of receiving flask in mL
Media density	Media density in g/mL, default value 1.00 g/mL
Target volume in receiving flask	Mandatory, target volume in receiving flask in mL
Target cell density in receiving flask	Target cell density in receiving flask, default value 1.00 x 10 ⁶ cells/mL
Cell line	Mandatory, name of cell line
Passage number	Passage number

Table 3: Parameters for Mycap® experiments

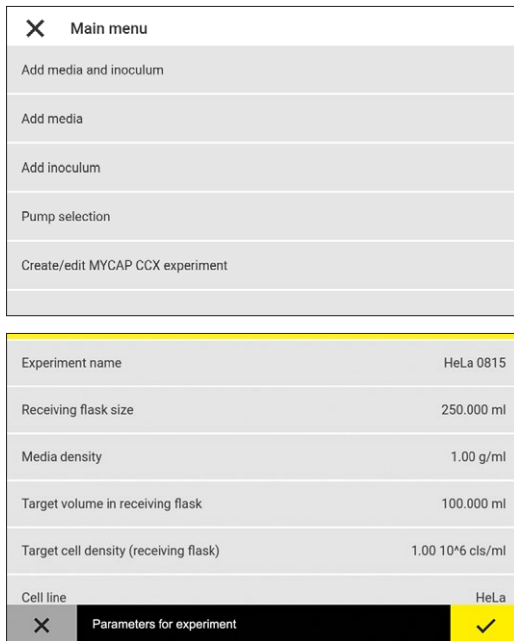


Figure 14: Main menu and screen to define parameters for Mycap® CCX experiments

4. Confirm settings to save experiment to database

4.3 Start Experiment

To start a Mycap® CCX experiment no special user rights are necessary.

1. Select a method (Add media and inoculum, Add media or Add inoculum) from the main menu
2. Select a saved experiment from the list. If many experiments are saved and the experiment to be started is not displayed in the screen use the scroll bar or the search function. To search for an experiment enter the name and let the software display the search results.
3. Select the experiment of choice from the list

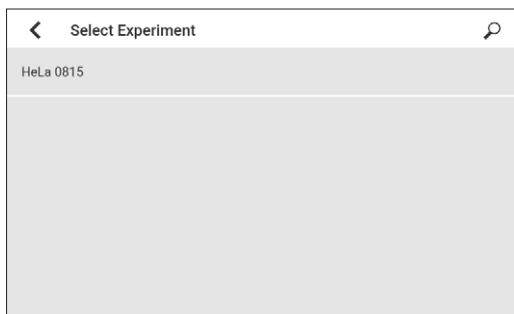


Figure 15: Screen to select Mycap® CCX experiment to start

4. Select the experiment of choice from the list
5. Set the following parameters* in the experimental details screens:

Parameter	Function
Media lot number	Mandatory, media lot number
Media volume in receiving flask	Media volume already available in receiving flask
Actual cell density donor flask	Cell density of inoculum
Date of passage	Last passaging date
Receiving flask lot	Mandatory, receiving flask lot number
Flasks to process	Mandatory, number of transfers/flasks to be processed
Start speed	Pump rpm value for the priming run
Media filling speed	Pump initial rpm value for media transfer step
Inoculum filling speed	Pump initial rpm value for inoculum transfer step
Media slow down at	Value in % of target weight the pump switches to rpm value set for "Media to speed"
Inoculum slow down at	Value in % of target weight the pump switches to rpm value set for "Inoculum to speed"
Media to speed	Pump rpm value to slow down to at set % of target weight ("Media slow down at")
Inoculum to speed	Pump rpm value to slow down to at set % of target weight ("Inoculum slow down at")
Reverse run	Pump rpm value for reverse run
Print label	ON or OFF, automatic label print after filling procedure is finished

Table 4: Parameters for Mycap® experiments

Experiment name	HeLa 0815
Receiving flask size	250.000 ml
Media density	1.00 g/ml
Media lot number	ABC123
Media volume in receiving flask	50.000 ml
Target volume in receiving flask	100.000 ml
Experimental details	

Passage date	2020-04-21
Cell line	HeLa
Passage number	0
Receiving flask lot	Lot Abc789
Flasks to process	3
Experimental details 2	

Start speed	25 rpm
Media filling speed	200 rpm
Media slow down at	90 %
Media to speed	10 rpm
Reverse run	0 rpm
Print label	On
Experimental details 3	

Figure 16: Screens to set experimental details. Parameters greyed out are not accessible for the user and cannot be modified.

* Parameters displayed in screens experimental details vary depending on the selected method. The example shows the pump settings for method Add media.

6. Confirm experimental details

4.4 Media Transfer

If method Add media and inoculum is selected, first inoculum is transferred and in the second step the media.

4.4.1 Materials

1. Sterile media in an aseptically closed container¹
2. Transfer assembly¹
3. Mycap® CCX flask¹
4. Quickseal® cutting tool
5. Watson-Marlow peristaltic pump
RS232 Serial communication required (323Du shown)
6. Cubis® II Balance with Mycap® CCX QApp
(MCA6202S-2S00-0 shown)

¹ Require a means for aseptic connection compatible with other components. AseptiQuik® S shown



Figure 17: Setup for adding media to Mycap® CCX consumables

4.4.2 Method

1. Close the clamps on the Mycap® CCX flask assembly



Figure 18: Clamp on Mycap® CCX Flask assembly

2. Aseptically connect one end of transfer assembly to the media container outlet and the other end to one leg of the "Y" assembly on the Mycap® CCX flask

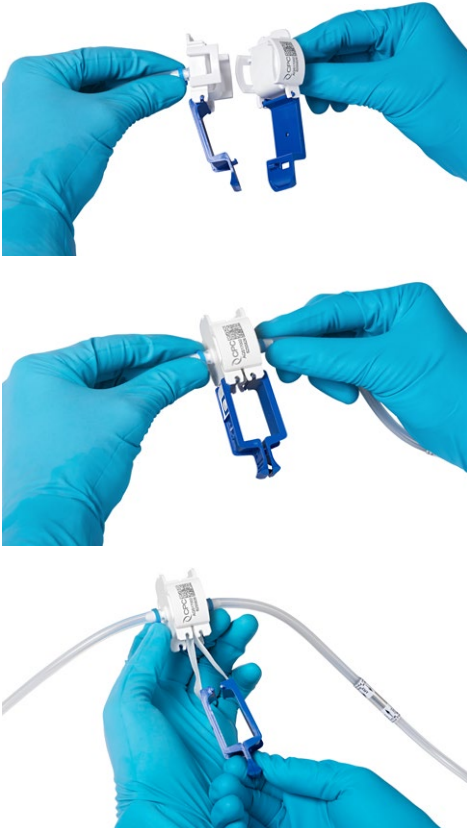


Figure 19: Aseptic connection between media container outlet and Mycap® CCX flask

3. Load tubing into the pump head



Figure 20: Installing tube in pump head

4. Open clamp on the leg of the tubing where aseptic connection was made
5. Activate Priming Step on Cubis® Balance touchscreen

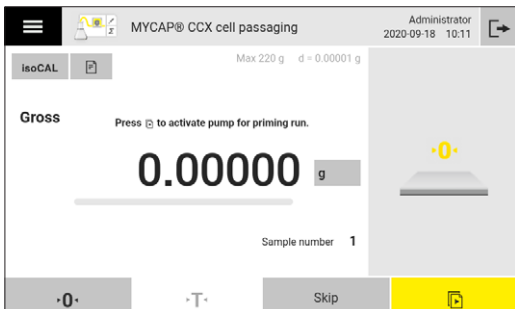


Figure 21: Screen to start priming run. The start button is highlighted yellow

6. Stop Priming run on Cubis® Balance touchscreen when media has reached the “Y” connector. The priming step may be repeated by touchscreen. If media only is added, the liquid should fill the dip tube leading into the flask (see Figure 30).

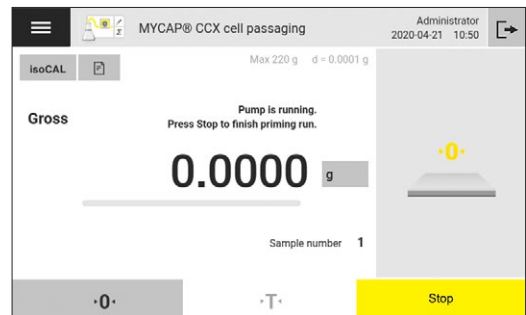


Figure 22: Y connector and screen to stop priming run. The stop button is highlighted yellow

7. Activate transfer step on Cubis® Balance touchscreen. The pump will stop automatically when media volume has been transferred.



Figure 23: Mycap® system during transfer

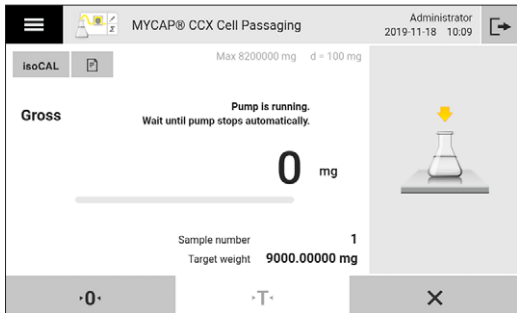
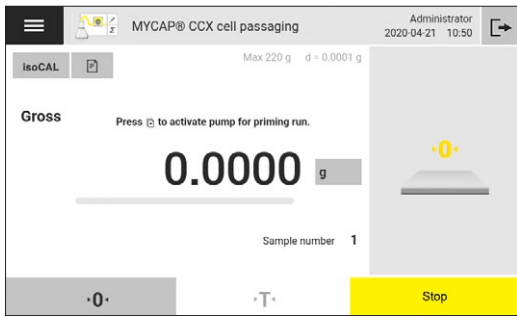


Figure 24: Transfer step screen. The start button is highlighted yellow. The pump stops automatically when the target weight is reached.

8. Close the clamp on the tubing leg the media was transferred from.



Figure 25: Closed clamp on Mycap® CCX Flask assembly

9. Optional: If not set to 0 rpm the pump runs in reverse mode to get rid of remaining media in the tube system that would contribute to the measured target weight. Press the yellow start and stop button to start and stop the reverse run.

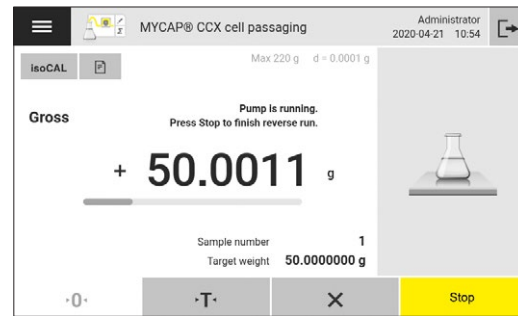
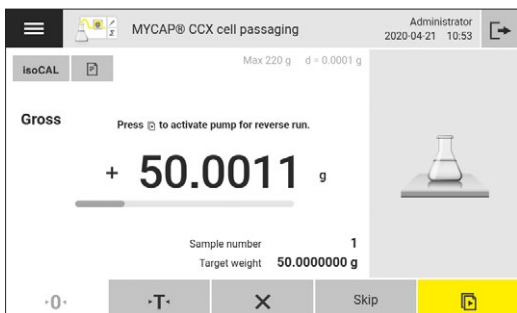


Figure 26: Reverse run screens

10. Grasp the Quickseal® collar and cut the collar on the leg the media was transferred from.



Figure 27: Cutting tool to cut collar

11. The results are calculated and displayed by the software application after completion of the experiment when media and/or inoculum are added. Press on button documentation (highlighted by yellow circle) to get to the print preview. Press yellow button print in print preview screen to print results.

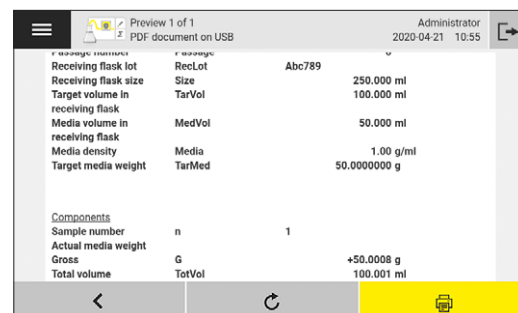
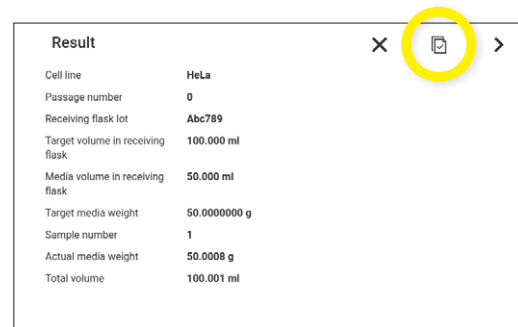


Figure 28: View and print result screen

4.5 Inoculum Transfer

4.5.1 Materials

1. Mycap® CCX flask¹ from prior passage
2. Transfer Assembly¹
3. Mycap® CCX flask¹ with media added
4. Quickseal® cutting tool
5. Watson-Marlow Peristaltic pump
RS232 serial communication required (323Du shown)
6. Cubis® II Balance with Mycap® CCX QApp
(MCA6202S-2500-0 shown)

¹ Require a means for aseptic connection compatible with other components. Aseptiquik® S shown.



Figure 29: Setup for adding inoculum to Mycap® CCX consumables

4.5.2 Method

1. Aseptically connect one end of transfer assembly to the donating flask outlet and the other end to one leg of the “Y” assembly on the Mycap® CCX Flask (see Figure 19)
2. Load tubing into the pump head (see Figure 20)
3. Open clamp on the leg of the tubing where aseptic connection was made (see Figure 18)
4. Activate priming step on Cubis® II MCA Balance touchscreen (see Figure 21)
5. Stop the priming on Cubis® II MCA Balance touchscreen (see Figure 22) when culture has reached to the bottom of the dip tube (see Figure 30). The priming step may be repeated by following prompts on Cubis® II MCA touchscreen



Figure 30: Dip tube with culture

6. Activate transfer step on Cubis® MCA Balance Touchscreen (see Figure 24). The pump will stop automatically when inoculum volume has been transferred
7. Close the clamp on the tubing leg the media was transferred from (see Figure 25)
8. Optional: If not set to 0 rpm the pump runs in reverse mode to get rid of remaining inoculum in the tube system that would contribute to the measured target weight. Press the yellow start and stop button to start and stop the reverse run (see Figure 26)
9. Grasp the Quickseal® collar and cut the collar on the leg the media was transferred from (see Figure 27)
10. View and print the results (see Figure 28)

The transfer is complete. Sample can be collected, if necessary. Flask may transferred to the incubator.



Figure 31: Mycap® CCX consumable after transfer of media or inoculum

5 Automated Filling of Liquid Samples with Cubis® II MCA Balances

The software application Pharma Filling is used to fill liquid samples into single bags or flasks or into multiple bags or flasks in parallel by using RS distribution manifolds. For the usage of RS distribution manifold special modifications to the balance are highly recommended. Please contact the Sartorius lab weighing special solutions group for offers covering the complete solution (pump, balance, balance modification, connection cable and software). Additional tools and consumables like Clipster® disconnect tool, tripod to fix the tubing, cutting tool, RS distribution manifolds, Flexsafe® bags, Mycap® flasks, Quickseal® caps and tubing are offered by Sartorius Bioprocess Solutions.

5.1 Program Creation and Pump Selection

After task start the main menu is displayed. The menu items “Create/modify filling program” and “Pump selection” are accessible for users with role right “Task management” only. Operators missing this role right have access to the menu item “Start filling” only. The two menu entries “Create/modify filling program” and “Pump selection” are displayed in grey to these users and are inactive.

5.1.1 Creating Filling Programs

The submenu for “Create/modify filling program” offers to create, edit or delete filling programs.



Figure 32: Pharma Filling QApp main menu

To create or edit a filling program the following parameters can be set:

Parameter	Function
Product code	Mandatory, string, product description
Product	Mandatory, string, determines the filling program name displayed in selective lists
Expiration date	Expiry date of product
Start speed*	Pump rpm value at tube priming step
Filling speed*	Pump rpm value at filling step
Slow down at	Value in % of target weight the pump switches to rpm value set for “To speed”
To speed*	Pump rpm value to slow down to at set % of target weight (“Slow down at”)
Reverse run*	Pump rpm value for reverse run
Target weight	Mandatory input, defines required net filling weight
Print label	ON or OFF, automatic label print after filling procedure is finished

Table 5: Parameters for pharma filling programs

* The pumping procedure consists of the four steps: priming of tubes (Start speed), filling at high speed (Filling speed), filling at low speed at set % of target weight (Slow down at) and reverse run. If the rpm value is set to 0 the corresponding step is skipped by the software.

Enter the required values and confirm your settings using the accept button to save the program in the database.

Product code	Test Filling
Product	COS-233
Expiration date	2020-04-30
Start speed	10 rpm
Filling speed	300 rpm
Slow down at	90 %
X	Parameters for product
	✓

Filling speed	300 rpm
Slow down at	90 %
To speed	10 rpm
Reverse run	10 rpm
Target weight	50.0000000 g
Print label	On
X	Parameters for product
	✓

Figure 33: Screens for editing filling program parameters

5.1.2 Selecting the Peristaltic Pump

To select the connected pump press “Pump selection” in the QApp software main menu. Select a pump model and confirm your settings. The selected model is highlighted by a leading dot.

<	Select pump
	Watson-Marlow 530Du pump
	• Watson-Marlow 630Du pump
	Watson-Marlow 323Du pump

Figure 34: Pump selection screen.

The pump type has to be selected only once. The selected pump model is saved by the software and recalled at the next start of the filling software. It is not necessary to set the pump model with each start of the filling program.

5.3 Filling Procedure—Filling of Single Bags or Flasks

5.3.1 Filling Preparation

1. Connect receiving container to source vessel.
2. Place receiving container on balance weighing pan.
3. Arrange tubing to not disturb balance during filling.
4. Level the balance.

5.3.2 Filling Process

1. Priming
 - a. Verify fluid pathway is open.
 - b. Press the start button highlighted in yellow to start the priming run.
 - c. Press stop in the display when tubing line is primed.
 - d. Priming step can be repeated by pressing start or skipped by pressing skip.
2. Filling Step
 - a. Verify fluid pathway is open.
 - b. Press the start button highlighted in yellow to start the filling run.
 - i. The balance will slow the pump to designated speed when the % target weight is met and stop the pump when total target weight is reached.
 - ii. The peristaltic pump and filling process can be stopped at any time by pressing on the cancel button X.
 - c. Close fluid pathway and disconnect filled containers.

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