



Miltenyi Biotec



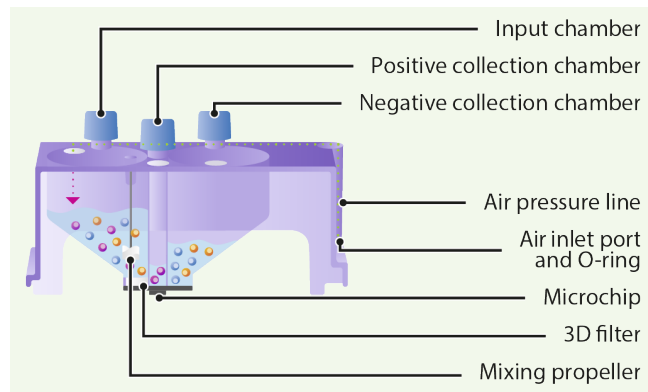
MACSQuant® Tyto® Instrument short instructions

Cartridge priming and sample loading

These short instructions are valid for MACSQuantify™ Tyto Software 3.2 and later.

Cartridge priming prewets the collection chambers to clear air. This procedure allows for a better flow of sample into the positive and negative collection chamber. Decide between automated priming for priming of loaded cartridges directly on the instrument, or manual priming before sample loading.

Description



Automated priming

The automated priming feature enables automated cartridge priming on the instrument. After priming, the instrument automatically proceeds with sample sorting. Cells, which are sorted to the positive collection chamber during the priming process, are taken into account in the sort statistics.

- 1 Open or create a suitable workspace for the desired sort. Refer to the **MACSQuant Tyto instrument short instructions Setting up a sort workspace**.
- 2 Load the sample into the cartridge. See **Sample loading**.
- 3 To activate automated priming, select the checkbox **Automated Priming** in the **Misc Controls** tab of the **Cartridge** tab on the touchscreen.
- 4 Insert the cartridge and click the **Start measurement** button in the progress bar.
- 5 To start the priming process, click the **Sort** button.
The priming process takes approximately one minute and is indicated in the instrument mode section of the progress bar.
- 6 After priming, the instrument automatically proceeds with sample sorting.

Manual priming

For sterile sorts, all open cartridge steps must be performed in a biosafety cabinet.

- 1 Place the cartridge into the MACSQuant Tyto Priming Fixture by sliding the feet of the cartridge into the corresponding slots of the base. Verify that the cartridge is leveled out.



- 2 Remove the cap from the input chamber of the cartridge.
- 3 Fill MACSQuant Tyto Running Buffer (research grade or GMP-compliant) into the input chamber until the buffer level reaches the mixing propeller (~500 µL) by using a pipet tip.
- 4 Pull the syringe plunger of a 10 mL syringe with male Luer lock tip out to its stop. Screw the syringe to the input chamber.
- 5 Press down on the corner of the cartridge (1) to block the air inlet port with O-ring (2). At the same time, push the plunger to let the MACSQuant Tyto Running Buffer flow into the positive collection chamber. Keep the pressure until buffer is visible in the positive collection chamber.



- Remove the cartridge from the MACSQuant Tyto Priming Fixture.
- Close the air inlet port with O-ring with your finger. Push the plunger of the syringe completely to flow the MACSQuant Tyto Running Buffer to the negative collection chamber. Hold the plunger down until the buffer level is just above the 3D filter.



- Unscrew the syringe from the cartridge and unhand the air inlet port with O-ring.
- Optional: Remove the buffer from the input chamber by using the syringe. To avoid air entering the 3D filter, leave a layer of buffer above the 3D filter. Reconnect the cap to the input chamber. Remove the buffer from the positive and negative collection chambers by using a capillary pipet tip.
- Proceed with sample loading.

Sample loading

Sample loading volume should be between 100 μ L and 10 mL. Maximum cell density should be 5×10^7 cells/mL in MACSQuant Tyto Running Buffer.

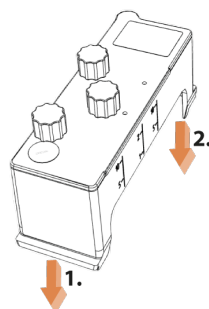
- Remove the plunger from a 10 mL syringe with a male Luer lock connection fitting. Store the plunger upside down.
- Remove the cap from the input chamber and attach the syringe. Store the cap upside down.
- Place a 20 μ m Pre-Separation Filter on top of the syringe. Moisten the filter with MACSQuant Tyto Running Buffer before use.

Do not place the cartridge on the priming fixture for sample loading.

- Apply the cell suspension onto the filter to remove cell clumps.
- Optional: To increase the cell recovery, rinse the filter with MACSQuant Tyto Running Buffer.
- Discard the filter. Save an aliquot as original fraction for cell analysis purposes.
- Carefully place the plunger on top of the syringe.
- Load the sample into the input chamber by applying the plunger at a rate of 0.5 mL/s. Do not load the cartridge at a higher rate as cells might flow into the negative collection chamber.
- Remove the syringe and reconnect the cap to the input chamber.
- Proceed with loading the MACSQuant Tyto Cartridge into the MACSQuant Tyto Instrument.

Inserting the MACSQuant Tyto Cartridge

- Start the MACSQuant Tyto Instrument and sign in.
- Scan the 2D barcode on the MACSQuant Tyto Cartridge with the barcode scanner on the front side of the instrument. The instrument door opens automatically upon scanning the cartridge.
- Orient the cartridge so that the Tyto owl image is in the upright position. Place the cartridge in the right slot of the instrument stage. To insert the cartridge, first lower the front (1.) and then lower the rear (2.). Do not push the cartridge downward.



- The cartridge is locked automatically. The door closes automatically shortly after inserting the cartridge.

Buffers and consumables

Following buffers and cartridges are available.

Component	Order no.
MACSQuant Tyto Cartridges	
8 pieces	130-104-791
24 pieces	130-106-088
MACSQuant Tyto Cartridge HS	
8 pieces	130-121-549
24 pieces	130-121-551
MACSQuant Tyto Cartridge LC (8 pieces)	130-133-260
MACS GMP Tyto Cartridge (1 piece)	170-076-011
MACS GMP Tyto Cartridge HS (1 piece)	170-084-001
MACSQuant Tyto Priming Fixture	130-119-832
MACSQuant Tyto Running Buffer research grade	
MACSQuant Tyto Running Buffer 1 \times 100 mL	130-107-206
MACSQuant Tyto Running Buffer 6 \times 100 mL	130-107-207
MACS GMP Tyto Running Buffer consisting of MACS GMP PBS/MgCl ₂ Buffer 3 \times 1 L MACS GMP Tytonase 5 mL	170-076-155 170-076-210
Pre-Separation Filters (20 μ m)	130-101-812
10 mL syringe with Luer lock tip	

Capillary pipet tips, sterile

Table 1: Buffers and consumables



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