Version: 1.1

StarterKit

ZELLMECHANIK DRESDEN

Hello.
This box contains consumables you need to perform real-time deformability cytometry. Here is a brief overview of the items and an instruction on the back side:

FlicXX
These are the microfluidic chips, with a narrow channel of cell-type specific cross-section. The cells are deformed by hydrodymic forces. ***FlicXX*** are prepared under dust-free conditions and individually subjected to control. They are intended for single use only. Please handle with care, as the cover glass can break already at low forces.

Safety note - Ensure adequate security measures to avoid possible injury by the cover glass (e.g. wear gloves and eye protection).

Specifications - glass (borosilicate): 0.19-0.23 mm, PDMS, Channel cross-section: XX in ***FlicXX*** stands for the cross-section in micrometer ± 7% (e.g. ***Flic20***: (20 ± 1.5) µm

CellCarrier
This is the PBS-based measurement buffer adjusted to a specific viscosity and density, required to create the correct cell deforming shear forces.

Safety note - All components are non-hazardous, but may cause irritation on eye, skin and respiratory tract. Rinse with water in case exposure to skin or eyes.

Specifications - Autoclaved PBS, < 1% methyl cellulose, viscosity at 24 °C for ***CellCarrier***: (15 ± 0.8) mPa·s, for ***CellCarrier B***: (25 ± 1) mPa·s

Microfluidic components
(Tube, Fittings, Luer adapters, Ferrules) These parts connect the syringe and the ***FlicXX***.

Safety note - Always use safety goggles, when handling the sample or buffer as the components might be under pressure.

Specifications - material of tube: FEP, fitting and luer adapter: PEEK, ferrule: ETFE

General remarks
Use all parts only as instructed. All items are for research purposes and not for any medical or diagnostic purposes.

The biological compatibility of ***FlicXX***, ***CellCarrier*** and microfluidic components has been tested for many, but not all types of cells and buffers.

Any stiff object larger than the channel cross-section will irreversibly clog the channel and the ***FlicXX*** has to be replaced. To prevent clogging we recommend to use the appropriate channel cross-section, to filter the sample accordingly and precisely follow the instructions.

Store ***FlicXX*** and microfluidic components at room temperature under dry conditions until use in the provided black box. Store CellCarrier at 4-8 °C.

You will find the MSDS of all components at www.zellmechanik.com.

Please contact us for any kind of question regarding real-time deformability cytometry.

Good luck with your experiments.

Out of *FlicXX*, *CellCarrier* or microfluidic components?
Get in contact!
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Instructions

0. To prepare your measurement you will additionally need two 5 ml, two 1 ml Luer Lock syringes, two blunt end filling needles ~ 30 ml isopropanol and ~ 15 ml filtered, deionized water. To perform the measurement you will need the *AcCellerator*.

1. Cut two tubes to the desired length (distance between syringe pump and *FlicXX* placed on the stage at the *AcCellerator*. One is the sample- the other the sheath-tube. Use a scalpel on a cutting mat or sharp scissors to avoid deformation of the tubing during the cutting process.

2. Take a look at the picture below. Put the fitting (natural) on the tube (green), attach the ferrule (blue), screw it to the adapter (red). This adapter fits into a Luer Lock Syringe.

Tube Fitting Ferrule Adapter Syringe with Luer Lok

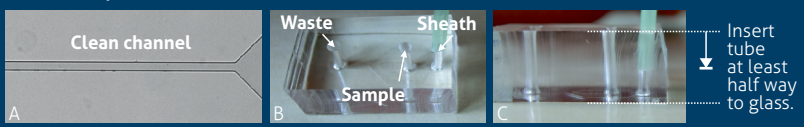
3. Cleaning: Flush the tubes two times with 5 ml filtered deionized water (label syringe with „water"). Dry the tubes by flushing with air using the empty syringe. Disconnect the syringe.

4. Connect a blunt end needle to a 1 ml syringe and take up *CellCarrier*. Disconnect the needle, remove all air bubbles from the syringe, reconnect to the sample-tube and fill completely Ignore this step if sample volume < tube volume ~ 250 µl (Case A). Change to sheath-tube and fill completely.

5. Connect a blunt end needle to a 1 ml syringe and take up your sample. Do this step slowly (<50 µl/s) to avoid pre-stressing of the cells. Disconnect the needle, remove all air bubbles from the syringe, reconnect to the sample-tube. Case A: take up *CellCarrier* in sample-tube and wait for step 8 (if necessary empty the syringe accordingly). No worries, your sample won´t mix up with the *CellCarrier*. Sample requirements and preparation can be downloaded at www.zellmechanik.com and be found in the *AcCellerator* manual. Make sure no air bubble remains in the tubing.

6. Take a *FlicXX* of appropriate channel cross-section (depending on mean cell diameter) and put it into the stage of the *AcCellerator* without removing the sealing tape. Check the channel is clean using the *ShapeIn* software (see picture A below, further instructions in the *AcCellerator* manual). Take the *FlicXX* out of the stage again.

7. Remove the sealing tape from the *FlicXX* and place it on a flat, clean surface to avoid breaking or staining the glass. When orienting the *FlicXX* as shown in the picture B (the two outlets which are closer to each other have to face to the right), the inlets and outlet are from left to right waste-, sample-, sheath-flow.



8. Connect the sheath-flow-syringe to the pump and drive it with 3 µl/s until a drop forms at the end of the tube. Slow down to 0.1 µl/s. Shake off the drop. No air bubbles. Repeat with sample. Case A: put tube in sample and drive with -3 µl/s to suck in sample.

9. Now connect the sheath-flow-tube with the outlet on the very right. Pushing the tube into the PDMS needs a bit of controlled force. Make sure to push the end of the tube further than half of the thickness of the *FlicXX*, but still keep at least a millimeter distance to the glass (see picture C). Conntect the Waste-tube to the last outlet.

10. Wait until the *FlicXX* is completely filled (until drops form at the sample inlet) and plug in sample-tube.

12. Place the *FlicXX* on the stage and fix it with the magnetic holders on both sides.

13. Clean with water, isoprop, water and air after measurement.

Now you are ready to start your measurements. For further instructions see the *AcCellerator* manual.