GOOD PRACTICE

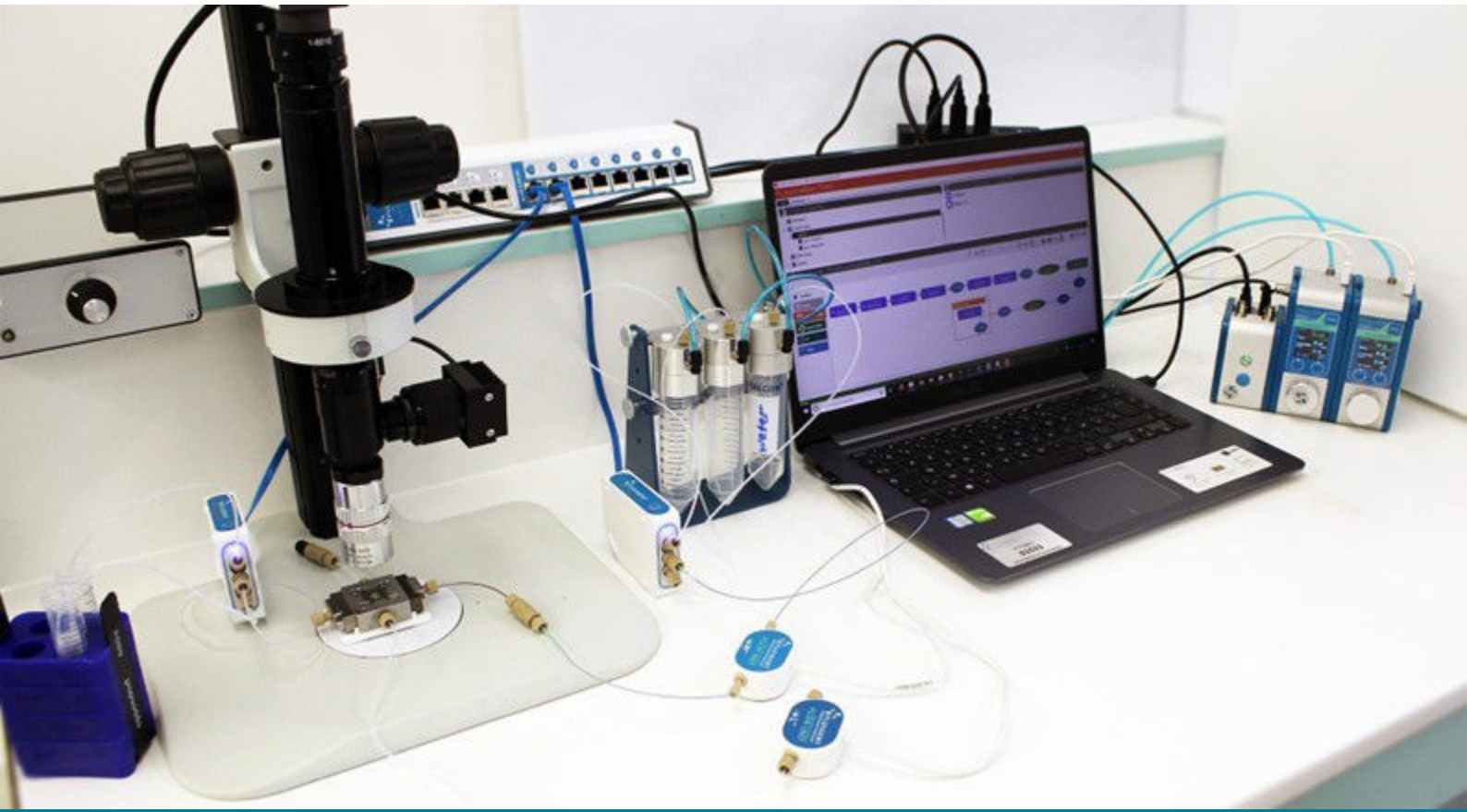
FLUIGENT
SMART MICROFLUIDICS™

# ALGINATE BEADS PRODUCTION STATION

## PRODUCT DESCRIPTION

P/N: O-SE-ALG-PCK

**The following document presents all the basic steps to follow to start and stop your experiments cleanly with the RayDrop.**



## CONTENTS

Experiment set up and start
Alginate beads Production
Stopping Experiments

# STARTING AN EXPERIMENT

The different inlets and outlets are shown on the following scheme:

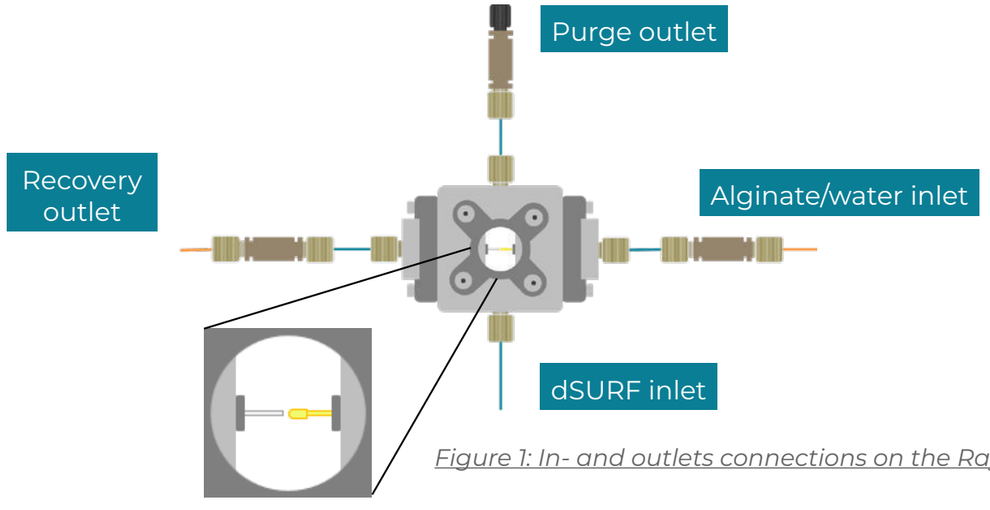



*Figure 1: In- and outlets connections on the RayDrop*

**1** Assemble the entire set-up as shown below (the use of filtered solutions is highly recommended). Depending on your application, installing inline filters (included in the kit and their fittings) between the RayDrop and the Flow Units is highly recommended.

## Alginate microbead production station package

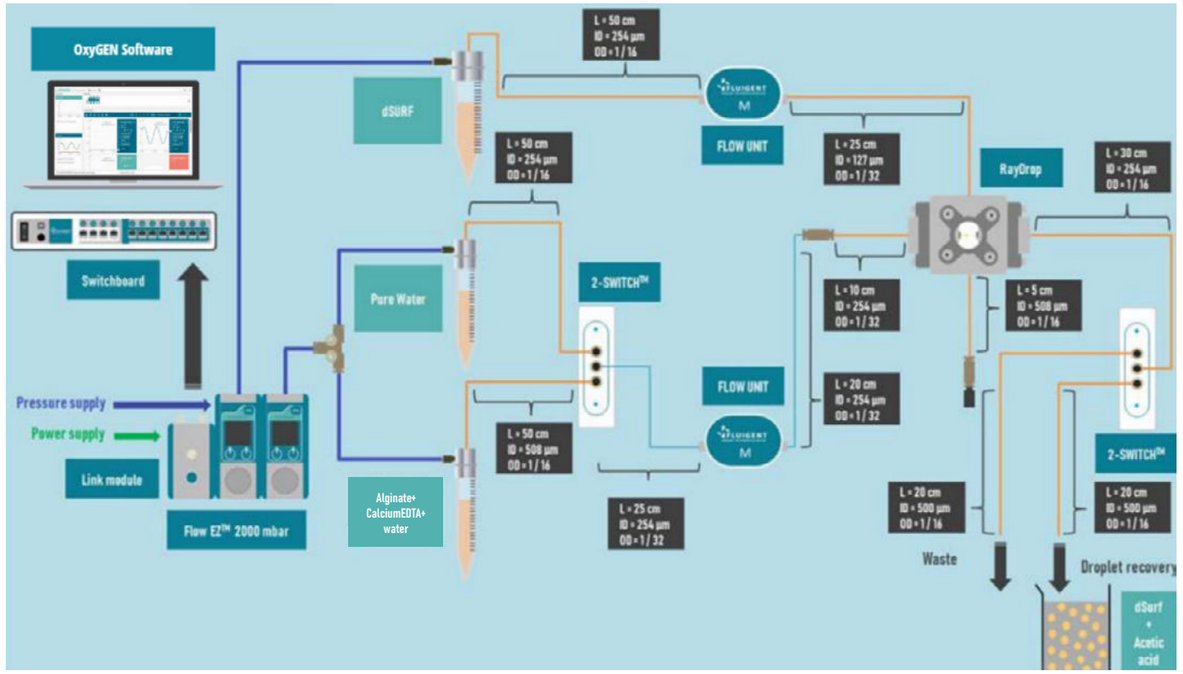



*Figure 2: Set-up of Alginate microbeads production station*

**2** Disconnect all channels except the dSURF inlet. Set a pressure of approximatively 1 bar on the dSURF inlet to start filling the RayDrop chamber (It could be more than 1 bar for faster filling)

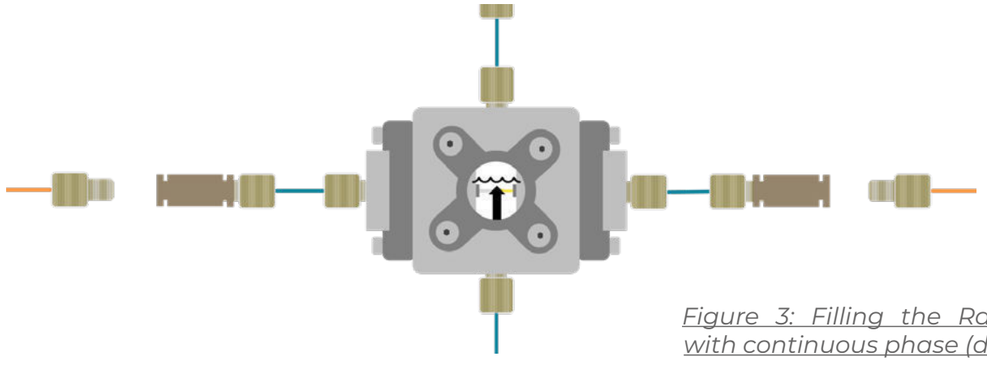


*Figure 3: Filling the RayDrop with continuous phase (dSURF)*

**3** Hold the RayDrop vertically to evacuate the air through the purge outlet



*Figure 4: Holding the RayDROP to evacuate the air*

**4** When dSURF is coming out of the purge outlet, use a plug to close the purge outlet. dSURF will flow out of the other in-outlets.
*(Tip: you can use a second plug to close the recovery outlet to facilitate the flushing of dispersed phase inlet.)*

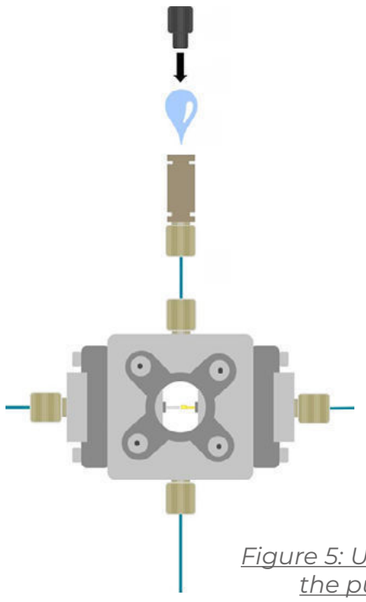


*Figure 5: Unplugging the purge outlet*

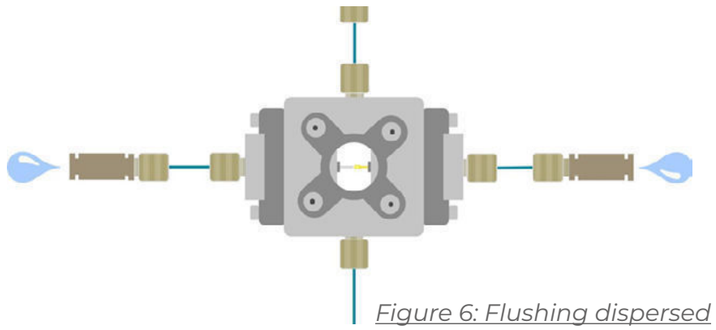


*Figure 6: Flushing dispersed phase inlet and recovery outlet*

**At this point, the RayDrop is ready to start Alginate microbead production**

# ALGINATE MICROBEAD PRODUCTION

**With the RayDrop filled, follow the next steps to produce Alginate microbeads**

**5** Set the dSURF pressure to 900 mbar. Start applying pressure (~100 mbar) on the Water inlet. Water should flow into the Alginate/Water inlet which is still disconnected from the RayDrop.

**6** Once Water is coming out of the tubing, at the end of the FLOW UNIT M, connect it to the Water inlet.

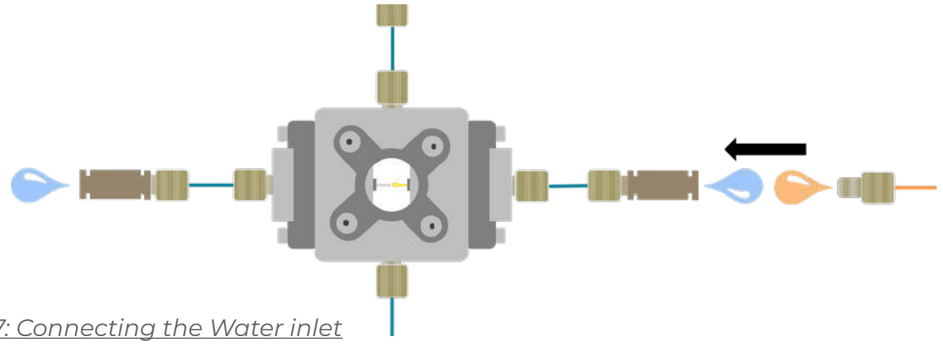


*Figure 7: Connecting the Water inlet*

**At this point, a backflow may occur in the Water channel. This is expected.**

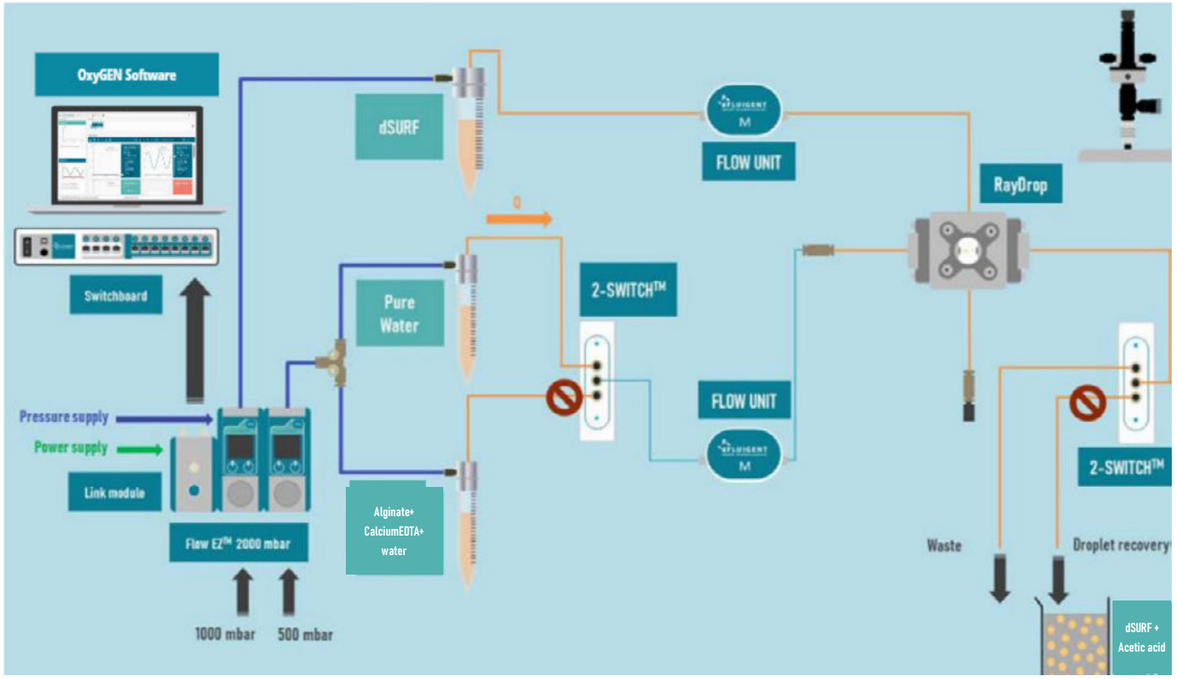



*Figure 8: Configuration of the setup at this step. 2-SWITCH™ redirecting Water*

**7** Set the pressure to the following values:

dSURF: 1000 mbar | Water: 200 mbar

**8** Decrease or increase both pressures slowly and simultaneously (around 10 mbar at a time for example) until you reach the droplet regime you want. The following pressure values generally lead to a good droplet generating regime.

dSURF: 1000 mbar | Water: 400 mbar

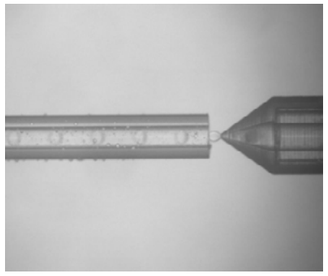


*Figure 9: Droplet generation regime*

**9** Switch the 2-SWITCH™ to the Alginate reservoir.

**At this point, flow rate on dispersed phase might decrease. This is due to the difference of viscosity between the water and the Alginate solution (The alginate solution, depending on the concentration of alginate, could be highly viscous). Adjust the pressure on both phases to reach the desired alginate droplet size and frequency.**

## Alginate microbead production station

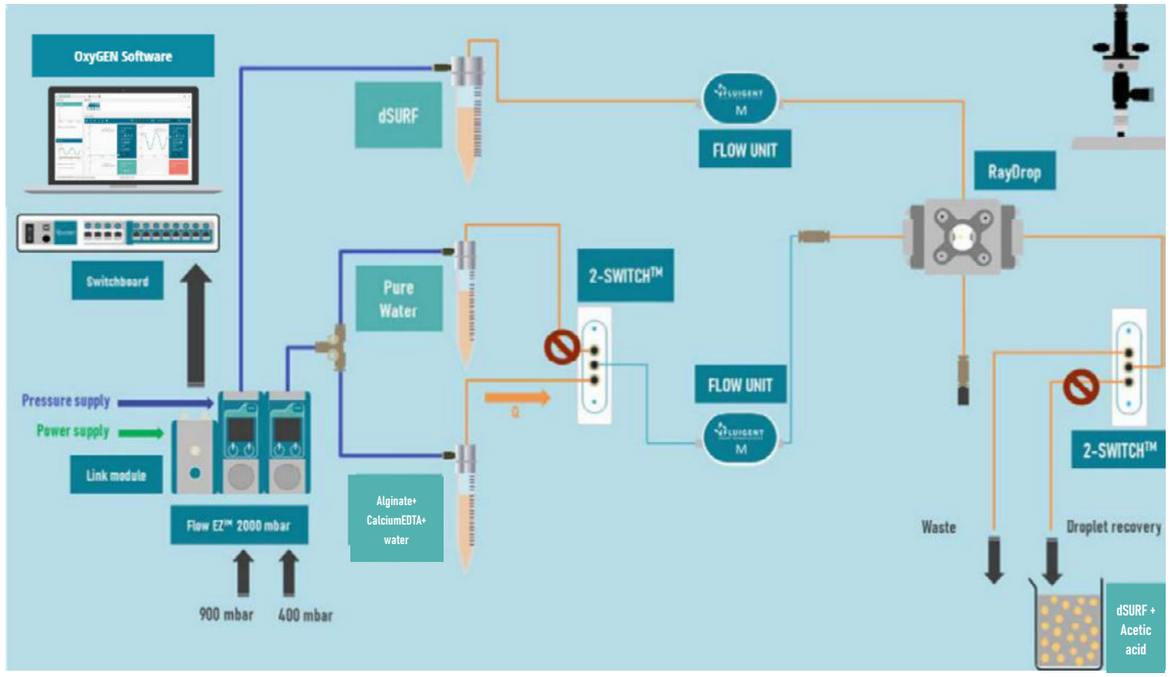



*Figure 10: 2-SWITCH™ redirecting ALGINATE solution*

**10** After about a minute, switch the second 2-SWITCH™ to your recovery reservoir to recover the Alginate microbeads.

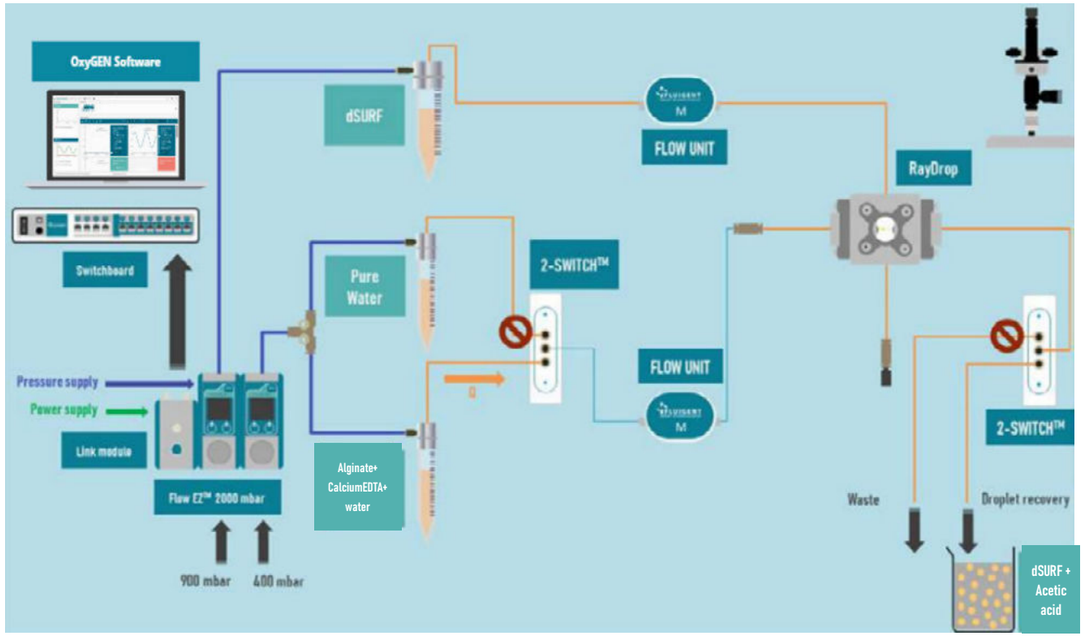



*Figure 11: 2-SWITCH™ n°2 redirecting droplet for beads crosslinking and recovering*

## STOPPING THE EXPERIMENT

The following steps should be performed after each experiment in order to prevent any clogging of the RayDrop.

Perform the following steps to stop the experiment.

**1** Switch the 2-SWITCH™ to the Water and the waste reservoir for at least a minute.

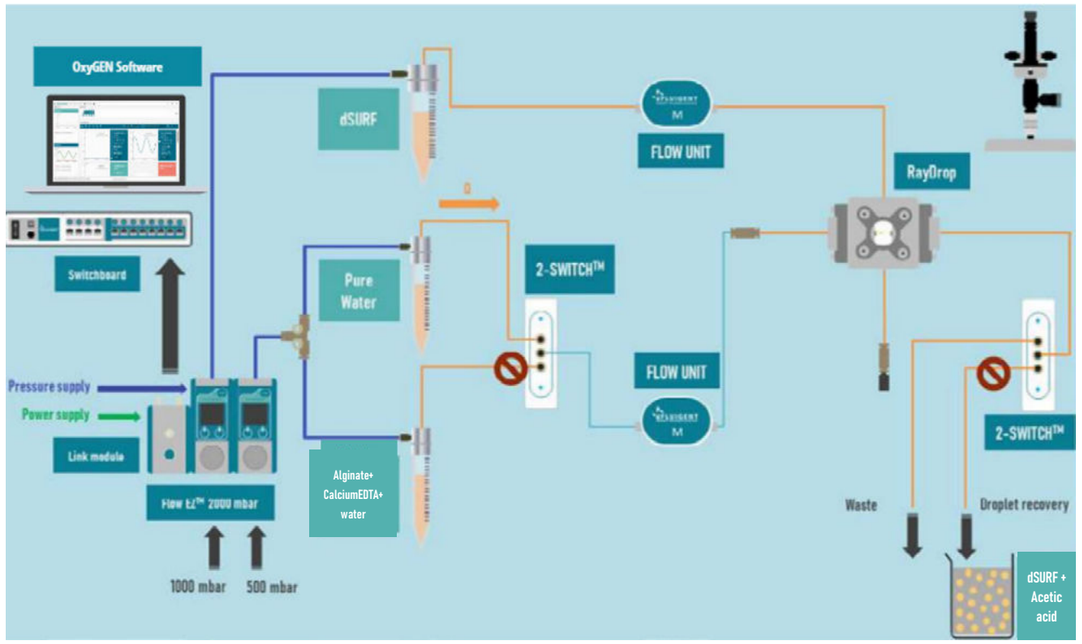



*Figure 12: 2-SWITCH™ redirecting Water solution for flushing in the waste*

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**2** Lower the pressure on the Water until you reach 0 mbar

**3** Disconnect the Alginate/Water inlet

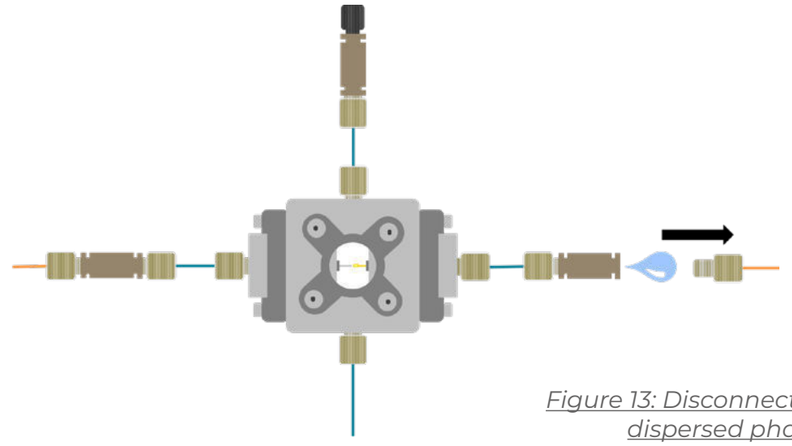


*Figure 13: Disconnecting the dispersed phase inlet*

**4** Keep some pressure on the dSURF inlet and flush for about a minute

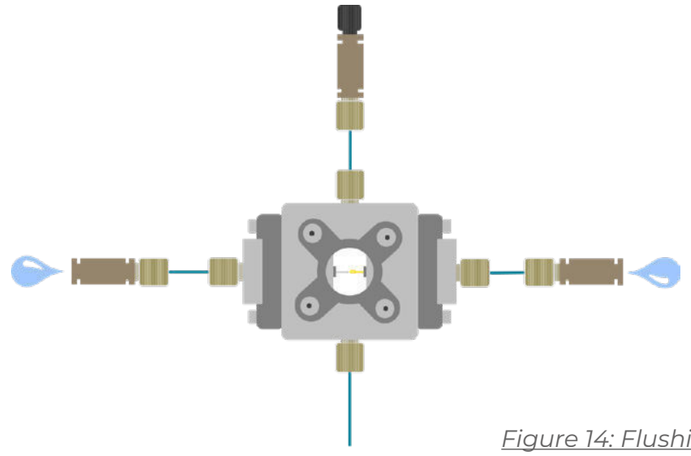


*Figure 14: Flushing the RayDrop*

**5** Close all outlets with plugs until the next experiment

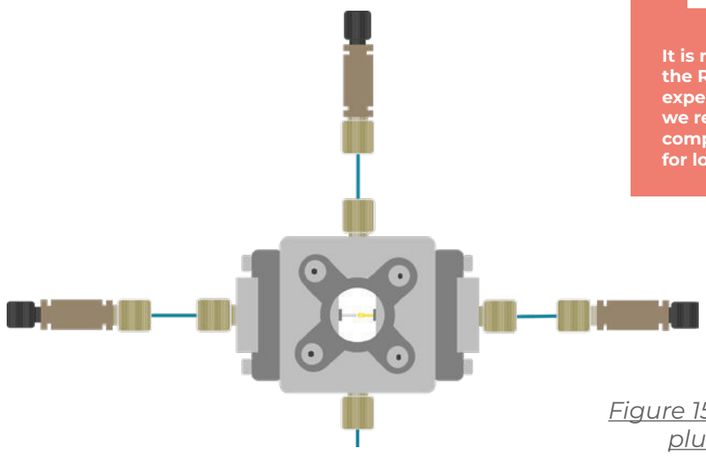


*Figure 15: RayDrop with plugged in-outlets*

**It is not mandatory to empty the RayDrop between each experiment. Nevertheless, we recommend emptying completely if will not be used for long periods (>5 days)**

# POSSIBLE ISSUES

## Some water has flowed into the continuous phase chamber or has aggregated on the outside of the nozzle

If some Water (without Alginate) flows into the chamber you can continue your experiment if it is only a small quantity that does not affect your experiment.

If there is a larger quantity that disturb visualization of droplet formation, perform the following steps:

**1** Stop the flow of Water

**2** Disconnect the Alginate/Water inlet

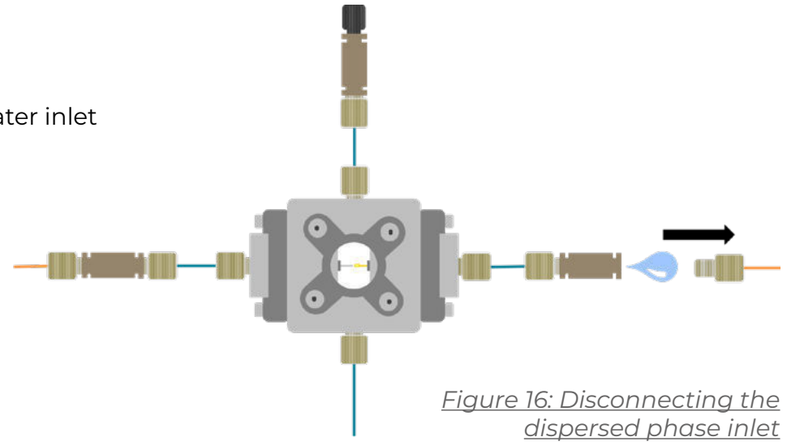


*Figure 16: Disconnecting the dispersed phase inlet*

**3** Open the top plug

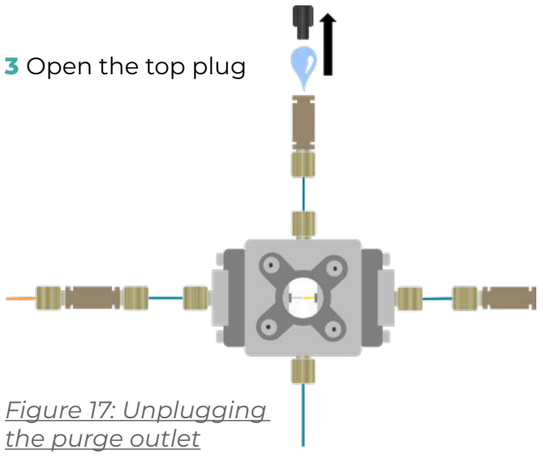


*Figure 17: Unplugging the purge outlet*

**4** Flush with the dSURF solution approximately for a minute or until you see all the Water has disappeared.

**5** Restart the system as described previously in the «Starting an experiment» section.

**The same procedure can be used if some Water is fixed on the outside of the nozzle.**

## Some Alginate solution has flowed into the chamber or is fixed on the outside of the nozzle

If some ALGINATE solution has flowed into the chamber perform the following steps quickly:

**1** Switch back your 2-SWITCH™ or manual valves to the Water solution and let flow for 30 seconds

**2** Disconnect the Alginate/Water inlet

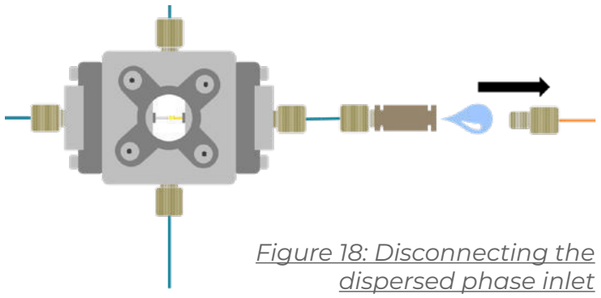


*Figure 18: Disconnecting the dispersed phase inlet*

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**3** Open the top plug

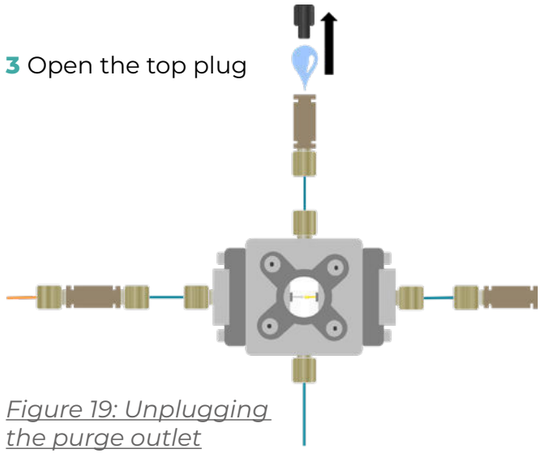


*Figure 19: Unplugging the purge outlet*

**4** Flush with the dSURF solution approximately for a minute or until you see all the Water has disappeared.

# COMPLETE CLEANING OF THE RAYDROP

In some cases, the previous procedures might not be enough.

**Be careful when performing the following cleaning procedures as the Ray-Drop nozzle and capillary are extremely fragile.**

## Cavity Emptying

**1** Hold the RayDrop vertically, with the closed purge outlet to the top.

**2** Loosen the inlet of the continuous phase (the dSURF inlet).

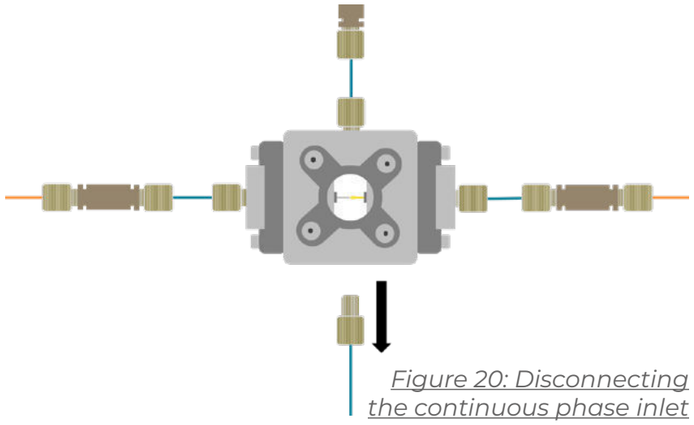


*Figure 20: Disconnecting the continuous phase inlet*

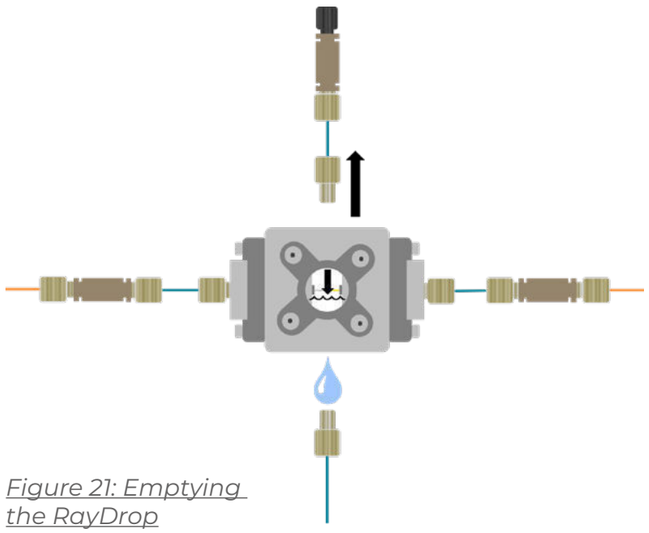


*Figure 21: Emptying the RayDrop*

**3** Open the purge outlet at the top. The continuous phase should flow out of the bottom outlet (the dSURF inlet).

## Soft cleaning

This should be performed if a dust, a particle, or a small amount of the dispersed phase has been accidentally introduced in the cavity and jeopardizes the droplet generation process, or its visualization.

Two cases are considered below:

- Case 1: The contaminant is located on a glass window.
- Case 2: The contaminant is located on the nozzle or glass capillary.

**DO NOT disconnect the RayDrop from the fluidic circuit during these operations.**

**Case 1: contaminant on a glass window**

**1** Empty the cavity following the instructions for emptying the cavity. Total emptying is not neces-sary. Just remove enough liquid to avoid leakage when the window will be opened.

**2** Place the RayDrop on a hard stable surface with the glass window to be cleaned on the top.

**3** Unscrew the four screws on the X-shaped cover using an Allen key number 3. Use tweezers or forceps to remove the metallic X-shaped cover.

**4** Use tweezers/forceps to remove the glass window and O-ring

**Tip: the glass window and O-ring can stick to the X-shaped cover. In this case separate them carefully following the cavity emptying steps**

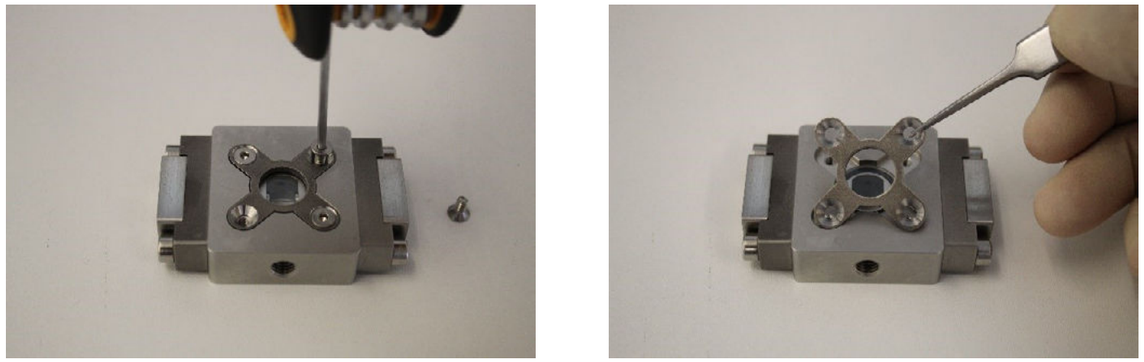


*Figure 22: Dismantling the RayDrop. Removing the screws (left) Removing the X-shaped metallic (right)*

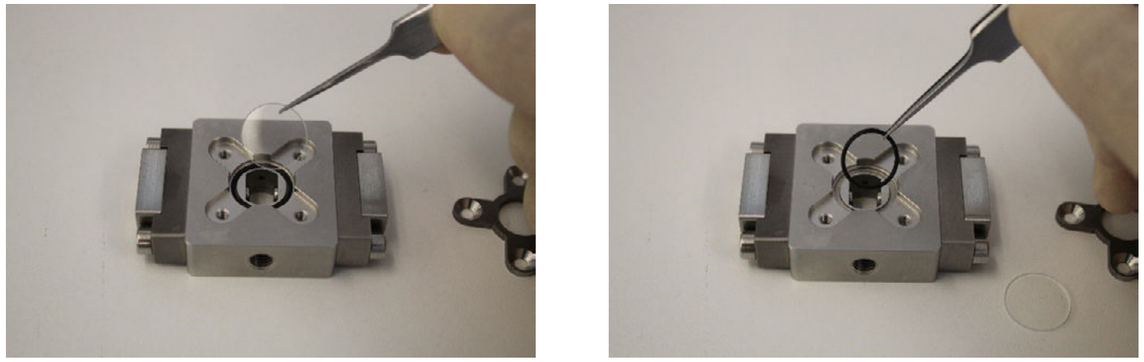


*Figure 23: Dismantling the RayDrop. Removing the glass window (left). Removing the O-ring (right)*

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**5** Wash the glass window, O-ring and X-shaped cover using glassware detergent or iso-propanol and dry it carefully. The best results are obtained using glassware detergent and an ultrasonic bath (1 minute). The parts need to be carefully rinsed with water before drying.

**6** Install in order the o-ring, glass window and x-shaped cover on the RayDrop body using tweezers.

**7** Using the Allen key number 3, tighten the four screws by alternatively giving a screw turn on each in a star shaped pattern. (how tight is there a point where it can break)

**Case 2: contaminant on the nozzle and/or the capillaries**

**1** Completely empty the cavity following the instructions for emptying the cavity

**2** Inspect carefully where the contaminant is located: If it is swept away by the continuous phase when emptying the cavity, the following steps are not necessary.

**3** Place the RayDrop on a hard stable surface.

**4** Completely remove the 4 screws using an Allen key number 3. Use tweezers to remove the metallic X-shaped part.

**5** Use tweezers to remove the glass window and O-ring.

**6** Using a syringe filled with continuous phase, clear out the contaminant by gently flushing it away.

**7** Drain the cavity again to remove the continuous phase added by the rinse as well as the contaminant.

**8** Install in order the o-ring, glass window an x-shaped cover on the RayDrop body using the tweezers.

**9** Using the Allen key number 3, evenly tighten the four screws by alternatively giving a screw turn on each.

**The glass window and o-ring can stick to the x-shaped meta piece. In this case the three parts are removed together so be careful when handling**

## COMPLETEY CLOGGED NOZZLE OR CAPILLARY

In case of a completely clogged nozzle or capillary, contact customer support for help.

**Tip: the glass window and O-ring can stick to the X-shaped cover. In this case separate them carefully following the cavity emptying steps**