

Application note

Emulseo x Micronit

High-performance droplet generation using Emulseo and Micronit products

INTRODUCTION

Emulseo is a leading company in microfluidic technology developing innovative, high-performance surfactants and formulations specifically designed for droplet-based microfluidic applications. Emulseo provides innovative solutions that ensure stable, reliable, and reproducible experiments for research and industry. Among Emulseo's products, FluoSurf™-O is a biocompatible fluorinated surfactant optimized to stably maintain monodisperse aqueous droplets in fluorinated oils, even under demanding conditions such as thermocycling. FluoSurf™-O is ideal for applications like ddPCR and single-cell analysis thanks to its stability, reproducibility, and excellent leakage control.

Emulseo also provides Fluo-Oil™ 7500, a fluorinated oil specifically formulated to dissolve surfactants like FluoSurf™, and widely used for encapsulating cells, proteins, or nucleic acids in applications such as genome sequencing and high-throughput screening.

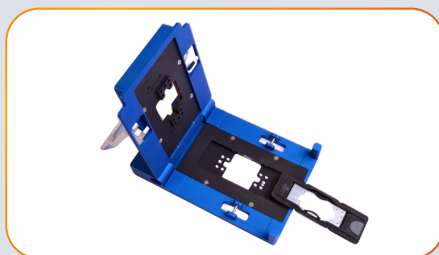
Emulseo developed Fluo-ST3™, an easy-to-use hydrophobic and fluorophilic surface treatment for microfluidic channel coating, enhancing droplet formation and stability in fluorinated oils. Together, these products enable reliable and precise microfluidic workflows.



Figure 1: FluoSurf™, surfactants by Emulseo



Figure 2: Micronit catalogue chips



Interfacing tool



Prototyping, development & production

Micronit is a microfluidics expert company offering a wide range of ready-to-use catalogue chips, including flow cells and droplet generators, available for quick purchase. Their interfacing tools simplify the connection between chips and laboratory equipment, ensuring easy integration. Micronit also specializes in prototyping, development, and production, supporting clients from initial design to scalable manufacturing in glass, silicon, or polymer. With advanced cleanroom facilities and expert teams, they deliver high-quality, customized solutions tailored to specific needs. Micronit ensures reliable and compliant products for life science and healthcare applications worldwide.

This application note presents the use of Emulseo's surfactant/oil chemical formulations for high-performance droplet generation within the Micronit glass microfluidic chips. The study emphasizes the excellent stability and monodispersity of droplets produced using the FluoSurf™-O / Fluo-Oil™ 7500 formulation in the Focused Flow Droplet Generator glass chips. Glass, being a hydrophilic material, requires treatment to become hydrophobic to allow proper water-in-oil droplet generation. Two types of chips were tested. The first one is a chip with factory hydrophobic coating. The second one is a glass chip sold untreated and treated with Emulseo's hydrophobic surface treatment, Fluo-ST3™.

EXPERIMENTAL SETUP MATERIALS AND PROTOCOL

A) Chip description

The Focused Flow Droplet Generator chip features a droplet generation unit with an optimized flow-focusing nozzle of 10x14 μm , 20x29 μm , 50x72 μm or 75x105 μm to generate droplets of different sizes. The interfacing tool enables connection of continuous (oil) and a dispersive phase (water). The droplets can be captured, monitored and manipulated in the observation chambers downstream of the nozzle. The monitoring chambers come at a height of 17 μm , 33 μm , 83 μm or 125 μm respectively.



Figure 3: Drawing for 50 μm Top connect Focused Flow Droplet Generator

B) Experimental setup

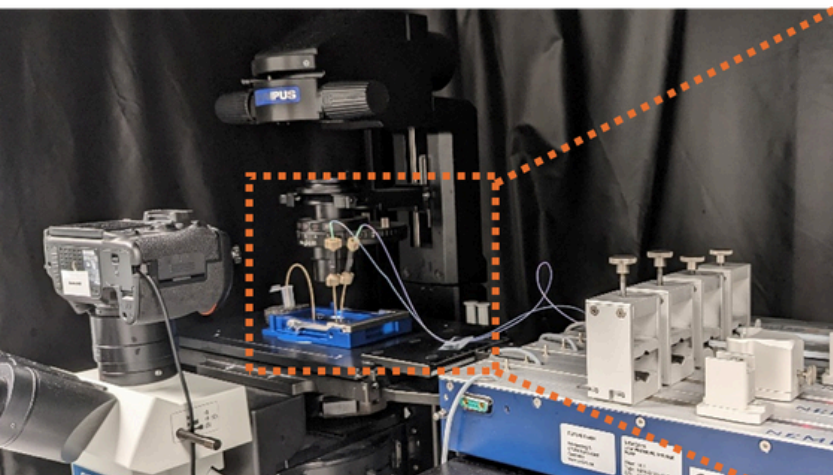
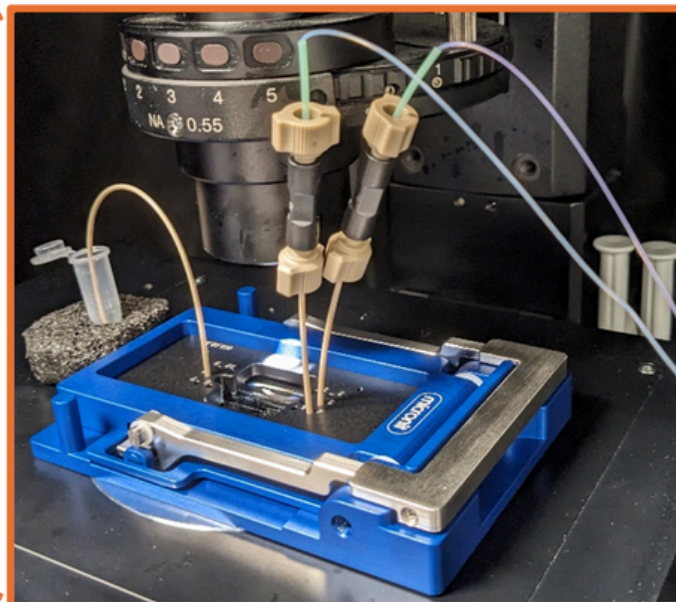


Figure 4: Setup used to generate droplets



C) Material & reagents

- 1 • Micronit microfluidic chips: 50 μm Topconnect Focused Flow Droplet Generator (Items 11006011 hydrophobic coated and 11000877 uncoated)
- 2 • Fluidic connect Pro holder (Item 11006689)
• Syringe to tube connection kit (Item 11002579)
• Syringe pumps (Cetoni)
• Inverted microscope (IX73, Olympus)
- 3 • Continuous phase: FluoSurf™-O (Emulseo) 1 w/w% diluted in Fluo-Oil™ 7500
• Droplet phase: 100 μM Sulforhodamine B in PBS

D) Protocol

1/ Fluo-ST3 surface treatment

- Place the glass chip (without the plastic part) in an oven at 300°C for 30 min
- Fill the microfluidic channels with Fluo-ST3™, leave for 30 s and flush the excess
- Rinse the channels with fluorinated oil

2/ Droplet generation

- Place the microfluidic chip in the holder
- Connect the chip to the syringes placed on the syringe pump with the syringe to tube connection kit. One of the syringes is filled with the continuous phase and the other with the dispersed phase. Connect the chip outlet with tubing to a waste tube
- Fill the chip with the continuous phase (to remove air) with a flowrate of 200 $\mu\text{L/h}$
- Inject the dispersed phase with a flowrate of 100 $\mu\text{L/h}$
- Wait for stabilization and replace the waste tube with a collection tube to start collecting the droplets
- After 30 min, reinject the droplets in a microfluidic chamber for observation

TOP TIPS



By varying the flowrates of the dispersed and continuous phases, the droplet size can be modified. For a fixed continuous phase flow rate, the larger the dispersed phase flow rate, the larger the droplet size.

It is advisable to use pre-filtered fluids in the droplet generators, as channel sizes are small, and blockages can easily occur. This should help to reduce blocked channels and prolong the lifetime of the chip.

RESULTS

Droplet generation tests were carried out on two different types of chips. The first one is a chip with factory hydrophobic coating. The second one is a glass chip sold untreated and treated with Emulseo's hydrophobic surface treatment, Fluo-ST3.

A) Generation of a water-in-oil emulsion in a hydrophobic coated 50 μm nozzle droplet generation chip

Figure 5 shows the droplet generation in a Micronit glass hydrophobic coated chip using Emulseo's FluoSurf™-O/ Fluo-Oil™ 7500 formulation.

Images taken at 4X and 10X magnifications illustrate the formation of uniform and monodisperse droplets, demonstrating the reliability of the workflow.

The surface treatment prevents the droplet phase from adhering to the channel walls, ensuring stable droplet generation.

Ready-to-use coated microfluidic chips save time and labor by eliminating the need to perform surface treatments, ensuring consistent preparation and faster experimental workflows.

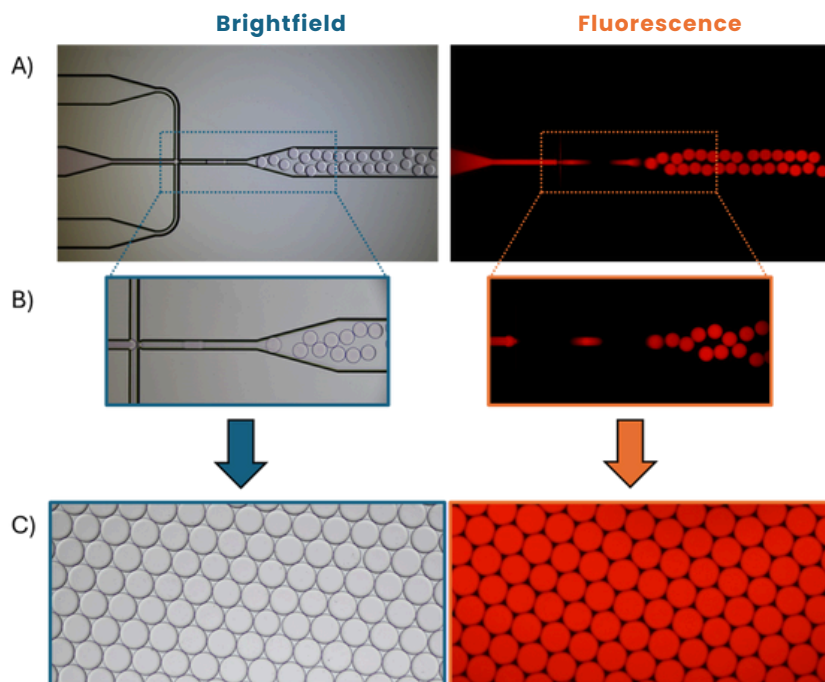


Figure 5: Production of a water-in-oil emulsion in a Micronit glass hydrophobic coated chip using Emulseo's FluoSurf™-O/ Fluo-Oil™ 7500 formulation

A) 4X magnification B) 10X magnification
C) Images of the emulsion produced

B) Generation of a water-in-oil emulsion in a hydrophobic uncoated 50 μ m nozzle DG chip coated with Fluo-ST3™

Figure 6 illustrates the droplet generation achieved in a Micronit glass chip coated with Fluo-ST3™, using Emulseo's FluoSurf™-O/ Fluo-Oil™ 7500 formulation.

Images taken at 4X and 10X magnifications show uniform and monodisperse droplets, similar to those obtained with factory-coated hydrophobic chips.

The surface treatment inhibits the adhesion of droplets to the channel walls, thereby ensuring consistent and stable droplet formation.

This solution provides maximum flexibility, eliminating the need for advance planning regarding coating. Additionally, Fluo-ST3™ can be applied to a single chip and reapplied as needed.

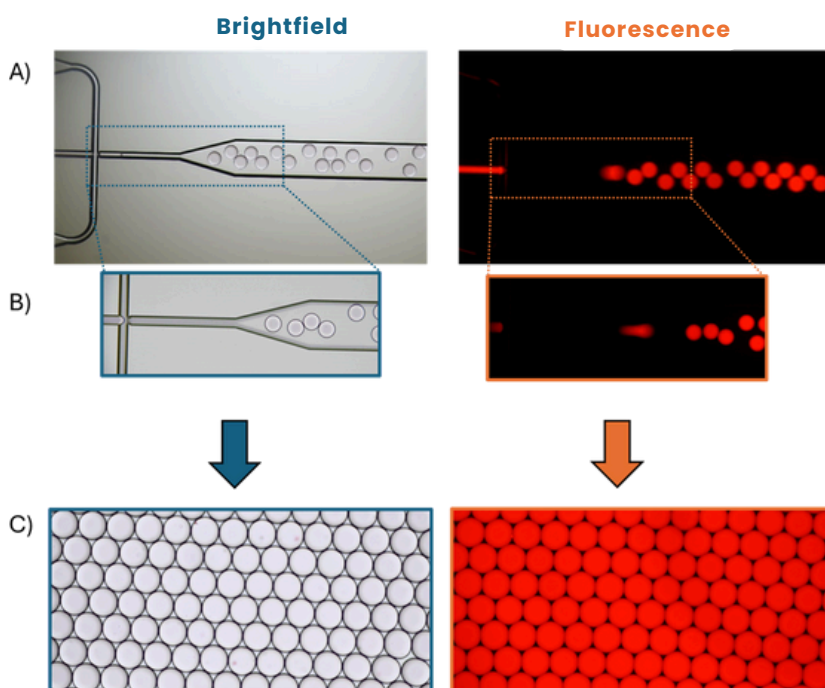


Figure 6: Production of a water-in-oil emulsion in a Micronit glass chip coated with Fluo-ST3™ using Emulseo's FluoSurf™-O/ Fluo-Oil™ 7500 formulation

A) 4X magnification B) 10X magnification
C) Images of the emulsion produced

c) Droplet monodispersity

Figure 7 presents the average droplet sizes generated in the two types of chips, along with the associated standard deviations.

The results show that both configurations enable the production of stable emulsions with an ideal monodispersity rate below 5%, ensuring high precision and reproducibility.

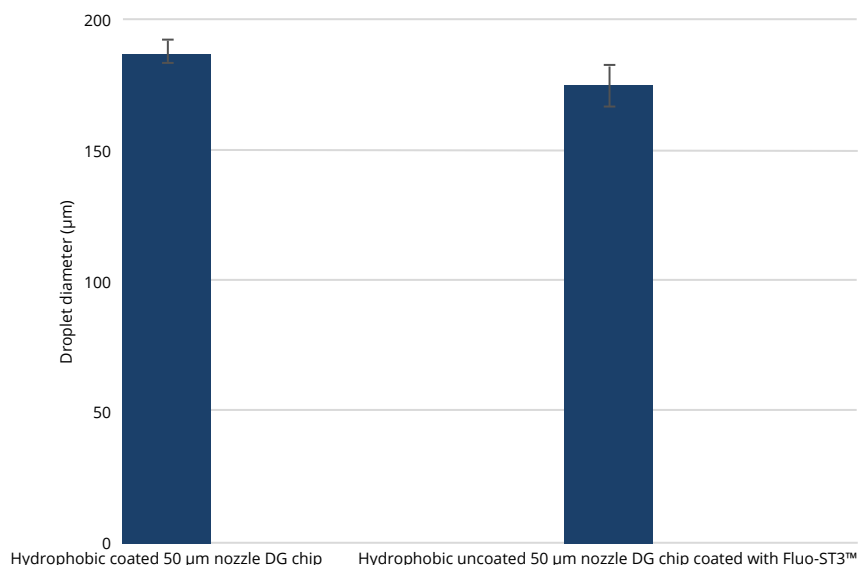


Figure 7: Droplet size and monodispersity in the two types of chips tested

CONCLUSION



Using **Emulseo** products in **Micronit** chips ensures stable droplet production and the generation of monodisperse droplets, making them ideal for applications requiring precision and reproducibility, such as ddPCR, single-cell analysis, genome sequencing, and high-throughput screening. The study highlights the versatility of Micronit chips, whether pre-coated with a hydrophobic coating or treated with Emulseo's Fluo-ST3™ surface treatment, both of which deliver high-quality droplet generation.

Pre-coated chips offer convenience and time savings by eliminating the need for surface treatment, while Fluo-ST3™-treated chips provide flexibility, allowing users to apply and reapply the coating as needed, extending the chip's lifespan. This adaptability is particularly valuable for addressing challenges like blocked nozzles, where cleaning can be more difficult for coated chips due to the risk of damaging the factory-applied coating.

Overall, the combination of Micronit's advanced microfluidic chips and Emulseo's innovative surfactant and surface treatment solutions enables reliable, efficient, and customizable workflows for droplet-based microfluidics, supporting a wide range of scientific and industrial applications.

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