INSTRUCTIONS

NANOSTABILIZER®-LT

USER GUIDE:
WITH BSP-1200 PROCESSOR IN THE FLOW-THROUGH CONFIGURATION

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MATERIALS NEEDED:

- BSP-1200 ultrasonic processor configured in the flow-through mode (see BSP-1200 User Manual and BSP-1200 Peripheral Equipment Assembly Guide for details);
- Digital scale, peristaltic pump, in-line sterilizing filter, 10 ml vials, disposable pipettes;
- 25 L storage tank with mixer and valved bottom outlet, magnetic stirrer with hotplate, stir bar, 2 – 3 L beaker (pre-mix vessel), dark-glass storage container (finished product container);
- NanoStabilizer®-LT, cannabis extract (e.g., isolate, distillate, full-spectrum oil, etc.), distilled water.

INSTRUCTIONS FOR MAKING 10,000 ml (10 L) OF TRANSLUCENT NANOEMULSION:

The instructions below detail the method for preparing 10,000 ml (10 L) of translucent nanoemulsion with the cannabis extract concentration of **20** mg/ml. If a different concentration is desired, use the table below and substitute the bolded numbers in the instructions with the numbers in the colored boxes.

Cannabis extract concentration in nanoemulsion	10 mg/ml	20 mg/ml	30 mg/ml	40 mg/ml	50 mg/ml
Cannabis extract	100 g	200 g	300 g	400 g	500 g
NanoStabilizer®-LT	500 g	1,000 g	1,500 g	2,000 g	2,500 g
Distilled water	9,400 g	8,800 g	8,200 g	7,600 g	7,000 g
Total	10,000 g	10,000 g	10,000 g	10,000 g	10,000 g
Number of 10 mg doses per 10,000 ml of nanoemulsion	10,000	20,000	30,000	40,000	50,000

1 Mixing your cannabis extract with NanoStabilizer®-LT:

In this step, you will mix your cannabis extract with NanoStabilizer®-LT in the pre-mix vessel. We recommend that you prepare an excess (10 %) of this pre-mix as when transferring to the storage tank with mixer (Step 3), some may remain in the pre-mix vessel (for future use). To accurately dispense **1,100** g of NanoStabilizer®-LT (**1,000** g + **100** g of excess) and **220** g of your cannabis extract (**200** g + **20** g of excess), follow the directions below.

a. Place the pre-mix vessel with a stir bar onto the magnetic stirrer with hotplate.

b. Place the NanoStabilizer®-LT container on the digital scale and tear. Begin dispensing NanoStabilizer®-LT into the pre-mix vessel, periodically placing its container back on the digital scale. Be careful to dispense NanoStabilizer®-LT into the pre-mix vessel and not to spill it onto the inner wall or outside.

c. Take note of the mass each time the NanoStabilizer®-LT container is placed back on the digital scale – the negative of that number indicates how much NanoStabilizer®-LT has been removed from the container. To ensure accurate measurement, as you get closer to your target value (**-1,100** g in this case), dispense decreasingly smaller amounts. Continue this until **1,100** g have been removed from the NanoStabilizer®-LT container (the digital scale reads:"**-1,100** g").

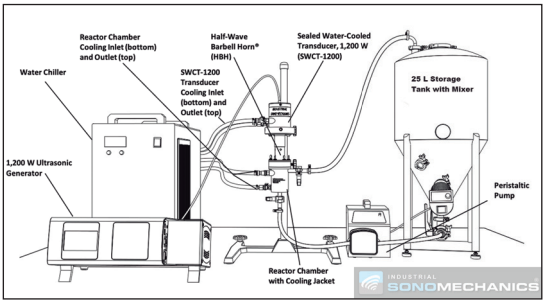
d. Follow a similar procedure to accurately dispense **220** g of your cannabis extract into the pre-mix vessel.

e. Stir the contents of the pre-mix vessel until your cannabis extract is dissolved in NanoStabilizer®-LT. Apply heat if necessary, but do not allow the liquid temperature to exceed 70 ˚C (158 ˚F).

2 Preparing the aqueous phase and setting up your process recirculation network:

a. Assemble the BSP-1200 ultrasonic processor in the flow-through mode (see BSP-1200 User Manual, BSP-1200 Peripheral Equipment Assembly Guide, and schematic on the right for details).

b. Dispense **8,800** g (**8.8** L) of distilled water into



the storage tank with mixer. Start running the mixer at a high speed (80 – 100 %).

c. Check that all connections in the process recirculation network are properly clamped. The distilled water should be set up to flow from the bottom of the storage tank with mixer to the reactor chamber and back to the top of the storage tank with mixer (see BSP-1200 User Manual and BSP-1200 Peripheral Equipment Assembly Guide for details).

d. Open the valve on the bottom of the storage tank with mixer. Turn the pump ON, then immediately OFF and check for any leaks in the process recirculation network. For assurance, perform this step several times. If no leaks are observed, proceed to the next step.

e. Turn the pump ON and begin recirculating the distilled water at the flow rate of 1 - 3 L/min.

f. Connect cooling lines to the transducer's and reactor chamber's cooling jackets (see BSP-1200 User Manual for details). Check for kinks or leaks in these lines. Do not turn on the water chiller yet.

3 Mixing the oil phase into the aqueous phase in the storage tank with mixer:

a. While your distilled water is flowing through the recirculation network and is being stirred by the tank mixer, place the pre-mix vessel onto the digital scale and tear.

b. Similarly to Step 1, use the negative reading on the digital scale to dispense exactly **1,200** g of the oil phase from the pre-mix vessel into the storage tank with mixer.

c. Continue stirring and recirculating the liquid in the storage tank with mixer until you finish Step 4e. Make sure not to introduce air bubbles into the liquid by excessively vigorous stirring. Lower the mixer speed if necessary.

4 Ultrasonic Processing:

In this step, ultrasonic processing will commence. Refer to BSP-1200 User Manual for operating instructions.

a. Turn on the water chiller and verify that the transducer and reactor chamber are being cooled correctly (see BSP-1200 User Manual for details).

b. Set the ultrasonic amplitude to 80 % (see BSP-1200 User Manual for details). Note that this setting can be adjusted up or down to optimize the results.

c. Initiate ultrasound and start timing. Try to maintain the processed liquid temperature at 45 – 60 ˚C (113 – 140 ˚F) throughout processing (you may temporarily disconnect the reactor chamber's cooling lines if the processed liquid becomes too cold).

d. After processing for 2 hours, draw a sample into a 10 ml vial every 10 minutes and notice the degree of translucency. When two consecutive samples exhibit no difference in translucency, the ultrasonic processing is complete.

e. Stop ultrasound and allow the processed liquid to recirculate and cool for 10 minutes.

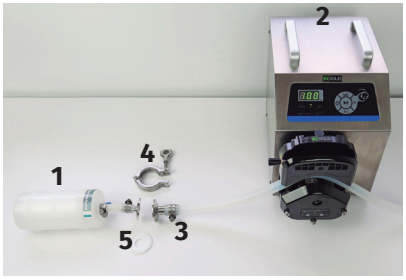
f. Stop the pump, reverse its direction and start it again in order to collect all of the nanoemulsion from the tubing and reactor chamber into the storage tank and mixer.

g. Stop the pump again and return its direction to the original setting.

5 Filtration:

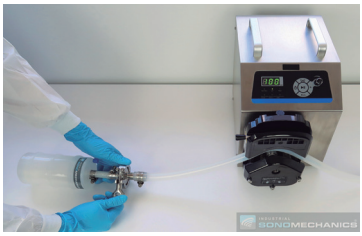
In this step, you will use the in-line sterilizing filter to remove any microorganisms and particulate contaminants from your nanoemulsion as you collected it in the finished product container.

PARTS NEEDED:



1. In-line sterilizing filter with 1/2" sanitary fitting
2. Peristaltic pump with 1/2" ID silicone hose
3. 1/2" sanitary to 1/2" hose ID adapter
4. 1/2" sanitary clamp
5. 1/2" sanitary gasket

a. Detach the bottom sanitary flanged connection from the reactor chamber and connect it to the inlet of the in-line sterilizing filter. Assemble items **1** – **5** as shown.



b. Using your pump at the flow rate setting of 100 – 150 ml/min, sterilize the nanoemulsion by passing it through the in-line filter into the pre-sterilized finished product container.

c. Store the finished product container with the filtered nanoemulsion in a cool and dark place.

d. Flush the filter with distilled water gently in both directions until the water runs clean.

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