



INSTRUCTIONS

NANOSTABILIZER[®]-LSTM

USER GUIDE:

WITH LSP-600 PROCESSOR IN THE FLOW-THROUGH CONFIGURATION



INDUSTRIAL

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MATERIALS NEEDED:

- LSP-600 ultrasonic processor configured in the flow-through mode (see LSP-600 User Manual and Peripheral Equipment Guide for details);
- Digital scale, peristaltic pump, in-line capsule filter (1 micron pore size), 2 L glass beaker (pre-mix vessel), immersion blender, silicone spatula, presterilized dark-glass storage container (enough to fit 1 L);
- NanoStabilizer®-LSTM, mushroom extract, distilled water.

INSTRUCTIONS FOR MAKING 1,000 ml (1 L) OF NANOEMULSION:

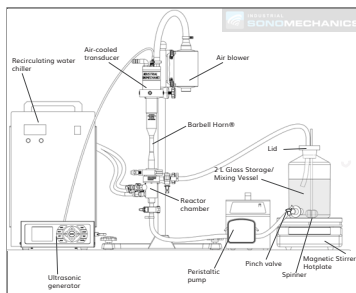
The instructions below detail the method for preparing 1,000 ml (1 L) of nanoemulsion with the mushroom extract concentration of 50 mg/ml. If a different concentration is desired*, use the table below and substitute the bolded numbers in the instructions with the numbers in the colored boxes.

Mushroom extract concentration in nanoemulsion	30 mg/ml	40 mg/ml	50 mg/ml	60 mg/ml	70 mg/ml
Mushroom extract (g)	30	40	50	60	70
NanoStabilizer®-LSTM (g)	120	160	200	240	280
Distilled water (g)	850	800	750	700	650
Total (g)	1,000	1,000	1,000	1,000	1,000
Number of 50 mg doses per 1,000 ml (1,000 g) of nanoemulsion	600	800	1,000	1,200	1,400

* If your intention is to convert this nanoemulsion into a water-soluble powder, we recommend that you stay with the 50 mg/ml concentration, as detailed in this guide. We also recommend that you dry/powderize the nanoemulsion within 48 hours of producing it.

1 Setting up the liquid recirculation network:

- Assemble the LSP-600 ultrasonic processor in the flow-through mode (see LSP-600 User Manual, LSP-600 Peripheral Equipment Assembly Guide and schematic on the right for details).
- Close the pinch valve at the bottom outlet of the glass storage/mixing vessel.
- Visually inspect the ultrasonic processor to check that all the connections in the process recirculation network are properly clamped. Check for any kinks in the tubing.



2 Pre-mixing the distilled water, NanoStabilizer®-LSTM, and mushroom extract:

We recommend that you prepare an excess (10%) of this pre-mix as some may remain in the pre-mix vessel (for future use) when transferring to the glass storage/mixing vessel.

- Place the pre-mix vessel on the digital scale and tare. Dispense **220 g (200 g + 20 g excess)** of NanoStabilizer®-LSTM and **825 g (750 g + 75 g excess)** of distilled water into the pre-mix vessel. Using the immersion blender, mix the NanoStabilizer®-LSTM and distilled water until fully dispersed and homogenous.
- Add **55 g (50 g + 5 g excess)** of your mushroom extract to the pre-mix vessel with NanoStabilizer®-LSTM and distilled water pre-mix. Use the immersion blender to pre-mix the mushroom extract with the NanoStabilizer®-LSTM and distilled water.
- Remove the glass storage/mixing tank lid and carefully transfer the liquid mixture from the pre-mix vessel to the glass storage/mixing vessel.
- Close the lid of the glass storage/mixing vessel and run the magnetic stirrer at 700 - 1000 RPM.
- Open the pinch valve and turn the peristaltic pump ON to begin recirculating the processed liquid at the rate of 0.5 - 1.5 L/min (50 - 200 RPM). The pump should send the processed liquid from the bottom of the glass storage/mixing vessel to the reactor chamber and back to the top of the glass storage/mixing vessel. To avoid foaming, make sure that the hose returning the liquid to the glass storage/mixing vessel is submerged into the liquid by about 5 cm.

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High-intensity ultrasonic processing:

- a. On the LSP-600 generator, set the ultrasonic amplitude to 80 % (see LSP-600 User Manual for details) and activate ultrasound.
Note: The amplitude setting can be adjusted up or down to optimize the results.
- b. Turn the water chiller ON (see LSP-600 User Manual for details) and maintain the processed liquid temperature between 50 - 65 °C (122 - 149 °F) throughout the rest of the process. Temporarily turn the water chiller OFF if the processed liquid becomes too cold.
- c. Throughout the process, verify that the transducer is being properly cooled (see LSP-600 User Manual for details).
- d. **For users with access to particle size analysis (*preferred*).** After processing for 50 min, draw a sample every 10 minutes and run the droplet size analysis. Once two consecutive samples demonstrate no significant decrease in the median droplet size, deactivate ultrasound.
For users without access to particle size analysis. Continue the process for about 1 hour. Deactivate ultrasound.
- e. Allow your nanoemulsion to stir, recirculate and cool down to 35 °C (95 °F).
- f. Turn the pump OFF, reverse its direction and turn it ON again in order to collect all of the nanoemulsion from the tubing and reactor chamber into the glass storage/mixing vessel.
- g. Turn the pump OFF and return its direction to the original setting.

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Filtration:

In this step, you will use the in-line capsule filter with 1 micron pores to remove any particulate contaminants from your nanoemulsion as you collect it in the finished product container.

PARTS NEEDED:



1. In-line capsule 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. Sanitary clamp
5. Sanitary gasket

- a. Close the pinch valve at the outlet of the glass storage/mixing vessel by turning it clockwise.



- b. Disconnect the reactor chamber inlet and use the same gasket and clamp to assemble the hose with the inlet of the in-line capsule filter in the next step.



- c. Assemble items 1 - 5 as shown in the pictures below.



- d. Open the pinch valve by turning it counterclockwise.



- e. Set your pump to a flow rate of 100 - 150 ml/min (50 - 150 rpm), turn it ON and filter the nanoemulsion by passing it through the in-line capsule filter into the pre-sterilized finished product container. It is recommended to place the finished product container below the level of the filter.



- f. Store the finished product container with the filtered nanoemulsion in a cool and dark place.
- g. Gently flush the in-line capsule filter by pumping distilled water through it in both directions until the water runs clean.

To re-order NanoStabilizer®-LSTM and replacement filters, please use the link or scan QR code below to visit our online store.

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