

Liquid Nitrogen Sterilization Prior to Warming

PROTOCOL		
NAME: Liquid Nitrogen Sterilization Prior to Warming	DEVICE: N-Bath 3.0.	
UTILITY: This protocol is designed for the preparation of Sterile Liquid Nitrogen (LN ₂) for the procedure of Warming, and to ensure that the Warming process is carried out under certified sterile conditions. The protocol is recommended for any Warming procedure, both if the carrier was prepared and stored with sterile LN ₂ or if it was vitrified/prepared/stored in non-sterile LN ₂ . For the specific Warming procedures refer to your clinic protocols.		
PROTOCOL ID: N-Bath_W	VERSION: 1.0	DATE: 24 October 2019

STEP	DESCRIPTION	NOTES
1	Place the device inside the sterile cabinet where you are planning to perform the Warming procedure. Ensure the device is powered off.	 Note A
2	Take a new Warming box; if reusing one ensure that it is thoroughly clean and dry. Fit a new disposable sleeve inside the box.	
3	Open the device lid and place the box inside the stainless-steel tub. Check that the sleeve is still correctly in place inside the box.	
4	Fill right up to the top the N-Sleeve with fresh LN ₂ that has not been in contact with any biological specimens.	 Note B
5	Close the lid. Connect the device to power and switch it on.	
6	Follow the instructions on the App to initiate the sterilization process.	
7	When sterilization is complete, remove the lid and place it on a soft surface. Check that the UV light sensitive label has turned green. Remove the box from the tub and place it where you will perform the Warming procedure.	 Note C
8	Extract the first carrier from the storage dewar and quickly immerse the carrier in the sterilized LN ₂ . Unseal it or open the cap and swirl the carrier back and forth at least 3 times (3 washes) and for about 30"; ensure the carrier remains submerged in LN ₂ during this process. Remove the carrier from the LN ₂ and immerse it in Warming solution.	
9	Repeat the procedure from Step 1 for each carrier.	 Note D

NOTES	
A	If your sterile cabinet is not large enough to house the device, place it as close as possible to the cabinet; in this event, sterility for the entire Warming procedure cannot be certified.
B	Use safety procedures for handling LN ₂ . Medical-grade LN ₂ is strongly recommended.
C	Use the App to download the data and print a label with all sterilization details (patient/carrier code, operator, etc); add to this label the removable label from the disposable sleeve.
D	Use the App to send the sterilization data to the Nterilizer web platform.