

Thank you for purchasing the Esco Liquid Nitrogen Biological Container. Before using it, kindly carefully review this instruction manual. It will provide you with essential information about the equipment's performance and the proper methods for safe and effective operation. Please keep this manual for future reference.

Esco Lifesciences provides many other resources at www.escolifesciences.com, to complement this manual and help you enjoy many years of productive and safe use of Esco Products.

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User Manual

**Liquid Nitrogen
Biological Container**

Copyright Information

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The information contained in this manual and the accompanying product is copyrighted, and all rights are reserved by Esco.

Esco reserves the right to make periodic minor design changes without obligation to notify any person or entity of such change.

“Material in this manual is provided for informational purposes only. The contents and the product described in this manual (including any appendix, addendum, attachment, or inclusion), are subject to change without notice. Esco makes no representations or warranties as to the accuracy of the information contained in this manual. In no event shall Esco be held liable for any damages, direct or consequential, arising out of or related to the use of this manual.”

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Warranty Terms and Conditions

Under normal usage conditions, Esco's liquid nitrogen biological containers are warranted for a **period of one year from the date of delivery to the customer**. In case any issues affecting the usage arise due to manufacturing defects, the following procedures will be followed:

1. During the initial filling of the container with liquid nitrogen, if significant condensation, frost, or other abnormal phenomena are observed on the container surface, our company will provide free repair or replacement.
2. Prior to the initial use of the container, if the static liquid nitrogen storage period falls below the specified standard during testing, our company will provide free repair or replacement.
3. Within three years, if any quality problems arise directly related to the manufacturing process, our company will be responsible for inspecting or repairing the product.
4. In case of repair or replacement, the container must be returned with all accessories, along with a copy of the product certificate and purchase receipt, matching the provided product number for inspection and processing at our company.

The following circumstances are not covered under warranty:

1. User tampering with the vacuum sealing joint of the container, leading to damage to the vacuum sealing joint or rupture of the valve core seal.
2. Using storage-type containers for transportation while they are filled with liquid nitrogen or containing frozen items, resulting in product quality issues.
3. Failure to follow the product instructions and relevant regulations, resulting in product damage. This includes using storage-type containers as transportation-type containers, causing dents to the outer shell, scratches on the neck tube, deformation or leakage due to internal pressure, or corrosion of the inner lining.

If you require replacement parts for your container beyond the warranty period, please seek assistance from Esco to assess the necessity of replacement. To arrange for the replacement of parts, kindly contact your respective local Esco office.

Esco's limited warranty covers defects in materials and workmanship. Esco's liability under this limited warranty shall be, at our option, to repair or replace any defective parts of the equipment, provided if proven to the satisfaction of Esco that these parts were defective at the time of being sold, and that all defective parts shall be returned, properly identified with a Return Authorization.

This limited warranty covers parts only, and not transportation/insurance charges.

This limited warranty does not cover:

- Freight or installation (inside delivery handling) damage. If your product was damaged in transit, you must file a claim directly with the freight carrier.
- Products with missing or defaced serial numbers.
- Products for which Esco has not received payment.
- Problems that result from:
 - External causes such as accident, abuse, misuse, problems with electrical power, improper operating environmental conditions.
 - Servicing not authorized by Esco.
 - Usage that is not in accordance with product instructions.
 - Failure to follow the product instructions.
 - Failure to perform preventive maintenance.
 - Problems caused by using accessories, parts, or components not supplied by Esco.

- Damage by fire, floods, or acts of God.
 - Customer modifications to the product
- Esco is not liable for any damage incurred on the objects used on or stored in Esco equipment. If the objects are highly valuable, user is advised to have in place independent external preventive measures such as connection to a centralized alarm system.

Factory installed, customer specified equipment or accessories are warranted only to the extent guaranteed by the original manufacturer. The customer agrees that in relation to these products purchased through Esco, our limited warranty shall not apply and the original manufacturer's warranty shall be the sole warranty in respect of these products. The customer shall utilize that warranty for the support of such products and in any event not look to Esco for such warranty support.

Esco encourages all users to register their equipment online at <https://www.escolifesciences.com/services/warranty-registration> or complete the warranty registration form included with each product.

ALL EXPRESS AND IMPLIED WARRANTIES FOR THE PRODUCT, INCLUDING BUT NOT LIMITED TO ANY IMPLIED WARRANTIES AND CONDITIONS OF MERCHANTABILITY AND FITNESS FOR A PARTICULAR PURPOSE ARE LIMITED IN TIME TO THE TERM OF THIS LIMITED WARRANTY. NO WARRANTIES, WHETHER EXPRESS OR IMPLIED, WILL APPLY AFTER THE LIMITED WARRANTY PERIOD HAS EXPIRED. ESCO DOES NOT ACCEPT LIABILITY BEYOND THE REMEDIES PROVIDED FOR IN THIS LIMITED WARRANTY OR FOR SPECIAL, INDIRECT, CONSEQUENTIAL OR INCIDENTAL DAMAGES, INCLUDING, WITHOUT LIMITATION, ANY LIABILITY FOR THIRD-PARTY CLAIMS AGAINST YOU FOR DAMAGES, FOR PRODUCTS NOT BEING AVAILABLE FOR USE, OR FOR LOST WORK. ESCO'S LIABILITY WILL BE NO MORE THAN THE AMOUNT YOU PAID FOR THE PRODUCT THAT IS THE SUBJECT OF A CLAIM. THIS IS THE MAXIMUM AMOUNT FOR WHICH ESCO IS RESPONSIBLE.

These Terms and Conditions shall be governed by and construed in accordance with the laws of Singapore and shall be subject to the exclusive jurisdiction of the courts of Singapore.

Technical Support, Warranty Service Contacts

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Singapore: +65 65420833

Global Email Helpdesk: support.global@escolifesciences.com

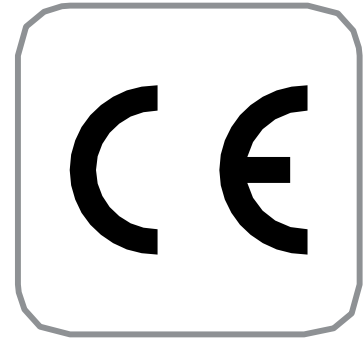
For international distributors, the warranty period starts two months from the date your equipment is shipped from Esco facility. This allows shipping time so the warranty will go into effect at approximately the same time the equipment is delivered to the user. The warranty protection extends to any subsequent owner during the warranty period. Distributors who stock Esco equipment are allowed an additional four months for delivery and installation, providing the product is registered with Esco. User can register product online at <https://www.escolifesciences.com/services/warranty-registration> or complete the warranty registration form included with each product.

Policy updated on 1st January 2015 (This limited warranty policy applies to products purchased on or after 1st January 2015)

Declaration of Conformity

In accordance with EN ISO/IEC 17050-1:2010

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Declare on our sole responsibility that the product:

Category	: Liquid Nitrogen Biological Container
Brand	: Esco Lifesciences
Model	: LBC-35, LBC-47, LBC-50, LBC-65, LBC-95, LBC-120, LBC-145, LBC-175

in accordance with the following directives:

2012/19/EU	: Waste Electrical & Electronic Equipment (WEEE) Directive and its amending directives
2006/95/EC	: Low Voltage Directive and its amending directives
2006/42/EC	: Machinery Directive and its amending directives

has been designed to comply with the requirement of the following Harmonized Standard:

Safety	: EN 12100:2010
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More information may be obtained from Esco Lifesciences' authorized distributors located within the European Union. A list of these parties and their contact information is available upon request.

Place of Issue: Singapore

Date of Issue: July 2023

Indra Setiawan

Indra Setiawan

Plant Manager, Esco Lifesciences

Chapter 1 - Product Information

1.1 About Liquid Nitrogen Biological Container

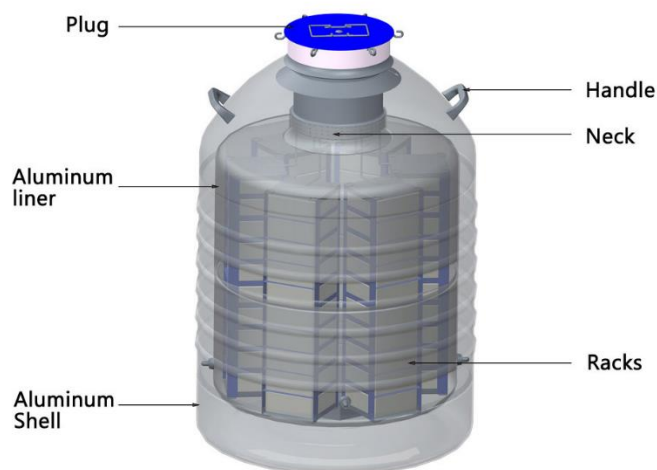
Esco Lifesciences proudly introduces the Liquid Nitrogen Biological Container (LBC) Series. These containers are specifically designed for the storage and preservation of biological samples, cells, and tissues at ultra-low temperatures. They are widely used in research laboratories, biobanks, and medical facilities around the world.

Esco's liquid nitrogen biological containers are built with high-quality materials and advanced insulation technology to ensure optimal temperature control and minimize temperature fluctuations. They are constructed using aluminum, providing durability and resistance to corrosion. The tanks are double-walled and vacuum-insulated, which helps to maintain the extremely low temperatures required for long-term sample storage.

These tanks are available in various sizes and configurations to cater to different storage needs, from small-scale units suitable for individual laboratories to large-capacity tanks for extensive biobanking facilities. They feature secure locking mechanisms to prevent unauthorized access and come with safety features like pressure relief valves to ensure safe operation.

Esco's liquid nitrogen biological containers are designed to provide reliable and efficient storage solutions for valuable biological specimens. They undergo stringent quality control processes to meet international standards and regulations. With a focus on quality, performance, and user safety, Esco's liquid nitrogen biological containers are trusted by researchers and professionals worldwide for their exceptional performance and reliability in preserving biological samples at ultra-low temperatures.

1.2 Quick View



1.3 Product structure and Characteristics

The container consists of essential components, including an outer shell, inner vessel, neck tube, multiple layers of insulation, and square canisters.

- a. The outer shell and inner vessel are constructed from aluminum plates, offering advantages such as lightweight design, high strength at low temperatures, and corrosion resistance.
- b. The neck tube is fabricated from fiberglass, providing notable characteristics such as high mechanical strength and low thermal conductivity.
- c. The multiple layers of insulation incorporate aluminum foil with exceptional reflective properties for radiation shielding. The insulation material selected possesses low thermal conductivity and minimal gas release rate, effectively reducing heat radiation.
- d. The space between the outer shell and inner vessel is maintained in a high-vacuum state to prevent thermal convection. Additionally, it employs an adsorbent material with substantial adsorption capacity at low temperatures, ensuring the long-term stability and reliability of the container's performance. The product lifespan is guaranteed to be no less than 5 years.

1.4 Market Segments

- a. Biobanks and cryopreservation facilities: These tanks are suitable for long-term storage of biological samples, cells, tissues, and other sensitive materials in biobanks and research laboratories.
- b. Medical and healthcare institutions: Liquid nitrogen biological containers can be used in hospitals, clinics, and medical research facilities for applications such as cryosurgery, tissue preservation, and other medical procedures.
- c. Scientific research: These tanks are utilized in scientific research institutes, universities, and laboratories for various scientific experiments and studies in fields like biology, chemistry, and cryogenics.
- d. Pharmaceutical industry: Liquid nitrogen biological tanks can play a role in the storage and preservation of pharmaceutical products, including vaccines, drugs, and other temperature-sensitive substances.
- e. Livestock industry: Liquid nitrogen biological containers play a crucial role in the freezing and storage of livestock semen and embryos.

1.5 Storage and Temperature requirements

- a. It is advisable to conduct a risk assessment before moving the equipment.
- b. Due to the heavy weight of the equipment, it is essential to thoroughly assess the safety of the workplace before moving the device.
- c. During transportation, the equipment should be protected from direct exposure to rain, snow, sunlight, and strong vibrations. It should not be rolled, dropped, and should be handled with care. Normal land, sea, or air transportation methods can be used.
- d. The properly packaged equipment should be stored in a warehouse with a relative humidity of $\leq 80\%$, temperature below 40°C , and good ventilation.
- e. During handling, the equipment can be manually moved or lifted using forklifts or cranes. If using a forklift or crane, the lifting point should be connected to the lifting tray at the bottom of the equipment. The lifting tray is designed with corresponding fork positions for forklift transportation. If there are any questions regarding loading, unloading, or handling, please consult the authorized dealer or trained and approved after-sales service engineers from Esco.









1.6 Safety Precautions

Please ensure that all personnel handling the liquid nitrogen biological container have received proper theoretical and standardized operational training. They should be familiar with the performance of the liquid nitrogen biological container, including:

- Liquid level monitoring
- Sample storage, retrieval, and labeling
- Items placed inside the liquid nitrogen container must be able to withstand temperatures as low as -196 °C
- Standard troubleshooting procedures
- Proper PPE must be used to protect against frostbite.
- The recommended lifespan of the liquid nitrogen biological container is 5 years.

1.7 Safety Symbols

In addition to the warning symbols already listed, the symbols appearing on the equipment and in this user manual have specific meanings. The table below presents the symbols related to this product and their corresponding meanings.

	Warning/Safety
	Oxygen depleting
	Do not roll
	Always keep LBC in upright position
	Risk of frostbite injury
	Fragile items, handle with care
	Rain-resistant
	Esco company logo

Chapter 2 - Installation Procedure

2.1 Package List

Please ensure that all items listed are included in the packaging. If you have any concerns or if any items are missing, please contact our customer support.

Description
Liquid Nitrogen Biological Container
User Manual/ Certificate of Compliance, Certificate of Compliance is included within the user manual
Square Canisters
Lock Cover

2.2 General Requirements

2.2.1 Material Compatibility

Materials in contact with liquid nitrogen should be compatible with it to prevent any adverse reactions or damage. Use materials specifically designed for cryogenic applications.

2.2.2 Operating Temperature Range

The recommended operating temperature range for the container is typically between -20°C and 50°C (-4°F and 122°F). Ensure that the container is not exposed to temperatures outside of this range to maintain its performance and integrity.

2.2.3 Transport Restrictions

Liquid nitrogen biological containers should not be used for transporting samples. These containers are primarily designed for stationary storage and are not suitable for transportation purposes. Please use appropriate transport containers specifically designed for sample transportation in accordance with relevant safety regulations.

2.3 Environmental Requirements

2.3.1 Adequate Ventilation

Ensure proper airflow and ventilation in the installation area to prevent the buildup of nitrogen gas and maintain a safe working environment.

2.3.2 Suitable Temperature and Humidity

Maintain an appropriate temperature range between 10°C and 30°C (50°F and 86°F) and a relative humidity below 80% in the installation environment to ensure optimal performance of the liquid nitrogen biological tank.

2.3.3 Stable and Level Surface

Choose a stable and level surface for the installation of the tank to ensure its stability and minimize the risk of tipping or damage during operation.

2.3.4 Sufficient Clearance Space

Provide enough clearance space around the tank to allow for easy access, maintenance, and servicing. Consider the space required for opening lids, connecting hoses or cables, and performing routine inspections.

2.3.5 Electrical Requirements

If the tank requires electrical connections, ensure the availability of appropriate power outlets and comply with local electrical codes. Adhere to the manufacturer's specifications regarding voltage, grounding, and other electrical requirements.

2.3.6 Safety Precautions

Follow all applicable safety regulations and guidelines for the installation of cryogenic equipment. Wear appropriate personal protective equipment (PPE) and take necessary precautions to handle liquid nitrogen safely.

2.4 Preliminary Measures

- a. Before opening the packaging, inspect the packaging labels and delivery note to ensure that the delivered goods are correct.
- b. Conduct a visual inspection of the product's exterior quality upon unboxing. If any defects are found in the appearance of the container, please notify our company within 5 working days of receiving the product (contact details can be found on the back cover). Distributors should follow the relevant terms and conditions outlined in the distribution agreement.
- c. Only after confirming that there are no quality issues with the packaging and appearance, proceed to place the liquid nitrogen biological container in its designated installation location.

2.5 Maintenance Requirements

All post-sales services should be conducted by qualified and certified engineers, and all related operations must comply with local laws and regulations.

2.6 Disclaimer

All liquid nitrogen biological containers undergo standard testing before leaving the factory, and they are only shipped after passing the tests. However, due to the involvement of multiple factors and conditions during usage, we cannot guarantee specific results in actual use. Users should follow the guidance provided in the operation manual and adhere to relevant safety regulations to ensure safe and effective use of the liquid nitrogen biological container. We shall not be held responsible for any loss or damage resulting from user error or other factors during the usage process. Users assume all risks associated with the use of the liquid nitrogen biological container and are responsible for any consequences that may arise during its use.

Chapter 3 – Basic Operation and User Guide

3.1 Initial Usage

- Filling with Liquid Nitrogen: When adding liquid nitrogen to the container indoors, please ensure proper ventilation by opening doors and windows to prevent a severe oxygen deficiency in the operating environment.
- For new containers, it may take 2-3 days to reach a temperature equilibrium state after the initial filling with liquid nitrogen. During the first few days, there may be a rapid evaporation of liquid nitrogen.
- Before using a new or thawed container, it should be pre-cooled with a small amount of liquid nitrogen to approach thermal equilibrium.
- When filling with liquid nitrogen, use a pump or long tube funnel, ensuring the filling tube is near the container bottom with a gap at the opening for nitrogen gas to escape.
- The liquid nitrogen level should not exceed the neck tube's lower end.
- Within 2-3 hours of placing frozen items, check for condensation or frost formation on the container's surface, indicating a deteriorated vacuum level. Although the probability of such phenomenon is low, it can be caused by improper loading, unloading, or transportation.
- Avoid direct contact with liquid nitrogen to prevent frostbite.
- Be cautious of collisions or heavy pressure that may damage the container shell.

3.2 Refilling of Liquid Nitrogen

- To check the liquid nitrogen storage level, use weighing method or a liquid level gauge.
- Avoid using hollow tubes to prevent splashing.
- Replenish liquid nitrogen promptly, ensuring the level does not exceed the container neck.
- Handle frozen items with care when operating and retrieving them.
- For actively used containers, refill when the level is below 30%.

3.3 Sample Storage

3.3.1 Sample Labeling

- The neck of the liquid nitrogen biological container is marked with slots numbered 1, 2, 3, 4, 5, and 6.
- The number of layers in the canister may vary depending on the specifications.
- The cryoboxes are labelled differently based on their specifications, ranging from 1-25 or 1-100.
- The above three points can be utilized for labelling the stored samples, and it is crucial to document the storage or retrieval of samples.
- For example, "2-3-16" represents a sample stored in Slot 16 of the cryobox in the 3rd layer of Canister 2.
- "5-2-80" represents a sample stored in Slot 80 of the cryobox in the 2nd layer of Canister 5.

3.3.2 Lid Design

- To prevent accidental opening of the container lid (when not locked) due to abnormal vibrations or collisions, Esco's LBC is equipped with a unique self-locking mechanism on the cover with a locking buckle.
- When opening or closing the cover, users should follow these steps:
 - At the locking position on the container lid, insert your fingers (index and middle fingers) and firmly grasp the upper edge of the lid, exerting and maintaining a certain force outward, and then flip it upwards to open the container lid (see Figure 2).

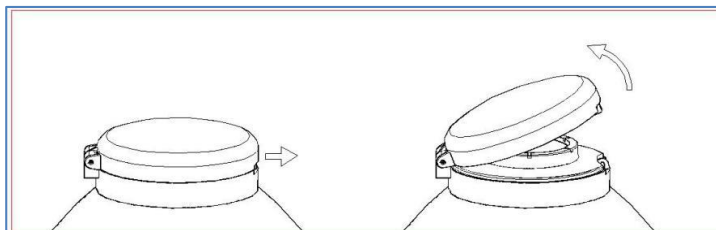


Figure 2

- When closing the container lid, a slight "click" sound indicates that the lid is properly closed, and a self-locking mechanism has been engaged between the upper lid and the base.

3.3.3 Canister Retrieval

- Hold the handle of a canister vertically and lift it approximately 40-80mm upwards.
- Then, rotate the handle 180° and continue lifting the canister vertically to extract it from the container (see Figure 3).
- By following this procedure, you can extract all the canisters from the container one by one.
- It is important to note that it is advisable not to completely remove the canister from the container.
- Ideally, the samples should be accessed at the neck of the container, wearing cryogenic gloves, and extracting the corresponding labelled sample.
- When placing the canister back into the container, ensure it is securely positioned in the slots and then close the container cover.



Please remember to wear cryogenic gloves throughout the operation to prevent frostbite.

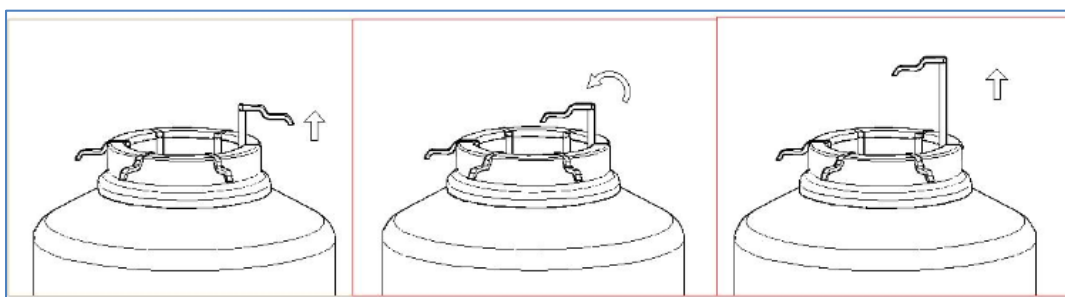


Figure 3

Chapter 4 – Troubleshooting and Maintenance

4.1 Troubleshooting

No	Challenges	Possible Cause	Proposed Solution
1.	Excessive evaporation of liquid nitrogen	Decreased vacuum level	Send the LBC to Esco for vacuum resealing.
2.	Excessive frost or sweating on the outer surface of the container	Vacuum integrity compromised	The LBC will be replaced with a new one if still under warranty period.
3.	Vacuum port rubber cap detachment	Insufficient adhesion	Reattach the rubber cap using adhesive.
4.	Cork stopper detachment	Insufficient adhesion	Replace the cork stopper.
5.	External dent on the container	Impact or collision	Assess the extent of liquid nitrogen evaporation and take appropriate action.
6.	Detachment of the handle on the canister	Insufficient adhesion	Replace the handle.
7.	Damaged locking cover	Rough handling	Replace the locking cover

4.2 Maintenance

- The container does not require any special maintenance. However, during the initial filling or refilling of liquid nitrogen after evaporation, it is necessary to pre-cool the container with a small amount of liquid nitrogen to achieve thermal equilibrium (without excessive vaporization) before filling it completely.
- For the preservation of valuable samples, it is recommended to consider **optional accessories such as a liquid nitrogen smart cap monitor or an alarm monitoring system** to prevent sample damage due to evaporation of liquid nitrogen.
- The static holding time of liquid nitrogen does not equal the complete evaporation time of the actual liquid nitrogen. It is mainly influenced by factors such as sample access frequency, volume of liquid nitrogen added, remaining amount, temperature, and pressure.
- For users using the container for the first time, it is advisable to regularly monitor the remaining liquid nitrogen.
- It is essential to maintain a record of any maintenance or service performed on the liquid nitrogen biological container. Please refer to the following Maintenance/ Service Log for guidance.

APPENDIX B:

Procedure and Calculation Method for Testing the Static Liquid Nitrogen Storage Period of the LBC

- I. The testing should be conducted at an ambient temperature of $20^{\circ}\text{C} \pm 3^{\circ}\text{C}$ and under normal atmospheric pressure. Additionally, the testing should not be performed in areas with direct ventilation.
- II. The testing should be conducted with the container filled with liquid nitrogen to a filling level of not less than 50% (100% for products below 3L). The container should be sealed with the cap, without the use of the canisters.
- III. After 48 hours of static standing, the average daily evaporation rate (q_m) of the tested product should be measured using the weighing method.
- IV. The weighing device (platform scale or electronic digital scale, etc.) used for the weighing method should have a maximum weighing capacity not exceeding five times the weight of the product when filled with liquid nitrogen. Its accuracy should be equal to or better than one-third of a gram.
- V. The static liquid nitrogen storage period (t) is calculated by the following formula:

$$t = \frac{m - m_e}{q_m}$$

t – Static liquid nitrogen storage period, d ;

q_m – Daily evaporation of the tested product, kg/d ;

m – The total weight of the product when it is filled with liquid nitrogen, kg ;

m_e – The product empty weight, kg.

Note: The testing and calculation of the static holding time of liquid nitrogen in the product are carried out in accordance with the [GB/T 5458-1997 standard](#). If the testing is not conducted at the specified environmental temperature and atmospheric pressure or if there are differences in the minimum scale value of the measuring instrument compared to the standard requirements, the calculated value may deviate from the actual static holding time of the product.

