Hemolytic

Reagent for pretreatment of whole peripheral and umbilical cord blood

Instruction Manual

CE IVD

For Professional Use Only

KEY TO SYMBOLS USED

REF	Catalogue number		Caution
LOT	Batch code		Contains sufficient for <n> tests
IVD	*In vitro* diagnostic medical device		Use-by Date
VER	Version		Consult instructions for use
	Temperature limit		Manufacturer
EC REP	Authorized representative in the European Community		Date of manufacture

1. INTENDED USE

Hemolytic is a reagent intended for pretreatment of clinical material (whole peripheral and umbilical cord blood) resulting in selective lysis of erythrocytes.

Indications and contra-indications for use of the reagent

Selective lysis of erythrocytes is used for pretreatment of whole blood before RNA/DNA extraction for subsequent in vitro diagnostics by nucleic acid amplification techniques (NAT).

2. PRINCIPLE

The principle of use is based on the effect of osmotic pressure on blood cells. Hemolytic solution is hypotonic for erythrocytes, and osmotic pressure causes cell membrane rupture.

3. CONTENT

Hemolytic is produced in 1 form:

1 vial of 100 ml, REF 137-CE

Hemolytic includes:

Reagent	Description	Volume, ml	Quantity
Hemolytic	Colorless clear liquid	100	1 vial

Hemolytic is intended for 100 samples.

4. ADDITIONAL REQUIREMENTS

- PCR box or Biological cabinet.
- Desktop microcentrifuge with a rotor for Eppendorf tubes.
- Automated pipette, 200-1000 µl.
- Sterile pipette tips with aerosol barriers, 200-1000 µl.
- Vortex mixer.
- Vacuum aspirator.
- Disposable polypropylene 1.5-ml tubes.
- Tube racks.
- Refrigerator for 2–8 °C.
- Disposable powder-free gloves and laboratory coat.
- Waste bin for used tips.

5. GENERAL PRECAUTIONS

The user should always pay attention to the following:

- Use sterile pipette tips with aerosol barriers and use new tip for every procedure.
- Use disposable gloves, laboratory coats, protect eyes while samples and reagents handling. Thoroughly wash hands afterward.
- Do not eat, drink, smoke, apply cosmetics, or handle contact lenses in laboratory work areas.
- Do not use a reagent after its expiration date.
- Dispose of all samples and unused reagents in compliance with local authorities requirements.
- Samples should be considered potentially infectious and handled in a biological cabinet in accordance with appropriate biosafety practices.
- Clean and disinfect all sample or reagent spills using a disinfectant, such as 0.5% sodium hypochlorite or another suitable disinfectant.
- Avoid contact with the skin, eyes, and mucosa. If skin, eyes, or mucosa contact, immediately flush with water, seek medical attention.
- Safety Data Sheets (SDS) are available on request.

6. SAMPLING AND HANDLING

NOTE: Obtaining samples of biological materials for PCR-analysis, transportation, and storage are described in manufacturer's handbook [1]. It is recommended that this handbook is read before starting work.

Hemolytic is intended for pretreatment of whole peripheral and umbilical cord blood.

Sampling

Peripheral blood is taken in a Vacuette tube with EDTA. Invert closed tubes 3-4 times and store at 2-8°C for no longer than 48 h.

Interfering substances and limitations of using test material samples

The next samples are inapplicable for analysis:

- the whole blood samples, collected in the tubes with heparin as anticoagulant,
- the whole blood samples, containing blood clot or which has been exposed to freezing.

The information about potential interfering substances and limitations of using test material samples is specified in the Instruction Manual of the PCR kit.

7. WORKING CONDITIONS

Hemolytic should be used at 18–25 °C.

8. PROTOCOL

1. Prepare the required number of 1.5-ml disposable polypropylene tubes and mark them. Add **1.0 ml of Hemolytic** to each tube. Add **0.25 ml** of whole blood according to labeling. Use a new tip for each tube. Close the tubes and mix by vortexing.
2. Incubate the tubes at room temperature for 5 min. Mix them by vortexing and incubate for 5 min once more.
3. Centrifuge the tubes at 8,000 rpm for 2 min. Carefully remove and discard the supernatant from each tube without disturbing the pellet using a vacuum aspirator and tips. Use a new tip for each tube.

After washing, the pellet should be white, with a thin pinkish film (erythrocyte debris). Repeat the washing with Hemolytic, if necessary.

The pellet with leukocytes should be immediately lysed (if extracting with RIBO-prep reagent kit, add 300 µl of Solution for lysis and follow the instructions described in the manual) or frozen and stored at the temperature not more than minus 68 °C for a long time.

If you have any questions or if you encounter problems, please contact our Authorized representative in the European Community.

9. TRANSPORTATION

Hemolytic should be transported at 2-8 °C for no longer than 5 days.

10. STABILITY AND STORAGE

Hemolytic is to be stored at 2-8 °C when not in use. Hemolytic is stable until the expiration date on the label. The shelf life of the reagent before and after the first use is the same, unless otherwise stated.

11. REFERENCES

1. Handbook "Sampling, Transportation, and Storage of Clinical Material for PCR Diagnostics" developed by Federal Budget Institute of Science "Central Research Institute for Epidemiology".

12. QUALITY CONTROL

In compliance with Federal Budget Institute of Science "Central Research Institute for Epidemiology" ISO 13485 – Certified Quality Management System, each lot of **Hemolytic** has been tested against predetermined specifications to ensure consistent product quality.

Please contact our Authorized representative in the European Community if side effects, undesirable reactions, facts and circumstances that pose a threat to the life and health of citizens and medical workers are identified during the use of the reagent.

List of Changes Made in the Instruction Manual

VER	Location of changes	Essence of changes
01.07.11 VV	Cover page, text	The name of Institute was changed to Federal Budget Institute of Science "Central Research Institute for Epidemiology"
10.04.20 VA	Through the text	The text formatting was changed
	Footer	The phrase "Not for use in the Russian Federation" was added
11.03.21 VA	—	The name, address and contact information for Authorized representative in the European Community was changed
31.05.22 EM	1. Intended use	"Indications and contra-indications for use of the reagent" subsection was added
	6. Sampling and handling	"Interfering substances and limitations of using test material samples" subsection was added
	12. Quality control	The Authorized representative in the European Community was specified for the contact in case of undesirable effects when using the reagent

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