

# EXCELLYSE XPerm 100 tests | Cat. No. ED7397

RU0

Not for use in diagnostic or therapeutic procedures.

# Technical Data Sheet (EN)

Version: ED7397\_TDS\_v4\_EN Date of Issue: 25-06-2024

#### Symbols used in the product labeling

RUO	Research Use Only	CONTENTS	Contents
•••	Manufacturer		
[]i	Consult instructions for use		
REF	Catalogue number		
LOT	Batch code		
Σ	Use by date		
1	Temperature limit		
*	Keep away from sunlight		
予	Keep Dry Keep away from rain		

## **Description**

EXCELLYSE XPerm is a set of two solutions intended for red blood cell lysis and white blood cell fixation together with permeabilization of cytoplasmic membranes in the preparation of biological samples from human peripheral blood prior to the flow cytometry analysis.

# **Specification**

XPerm Solution A is a buffered solution with a fixative intended to fix white blood cells of peripheral blood samples.

XPerm Solution B is a buffered solution with a detergent intended to permeabilize cytoplasmic membranes and to block non-specific interactions during intracellular staining of peripheral blood samples that were fixed with XPerm Solution A and lysed with deionized water.

## Reagent(s) provided

#### **Contents**

The EXCELLYSE XPerm is sufficient for 100 blood sample lyses with permeabilizations and is provided with the following reagent(s):

XPerm Solution A, 1 x 25 ml

XPerm Solution B, 1 x 25 ml

# Materials required but not provided

Round bottom test tubes (12 x 75 mm)

Deionized water (Reagent-grade)

Phosphate buffered saline (PBS), pH 7.4 (0.2 g/L KH<sub>2</sub>PO<sub>4</sub>, 1.42 g/L

Na<sub>2</sub>HPO<sub>4</sub>·2H<sub>2</sub>O, 8.0 g/L NaCl, 0.2 g/L KCl)

Phosphate buffered saline (PBS) with 1% formaldehyde

Appropriate fluorescent-dye-labeled primary/secondary antibodies

## **Equipment required**

Automatic pipette with disposable tips (100  $\mu$ l – 500  $\mu$ l) for pipetting specimen, lysing solution and PBS with 1% formaldehyde

Liquid dispenser or pipette with disposable tips (1.0 ml - 3.0 ml) for dispensing deionized water and PBS

Vortex mixer

Centrifuge

Flow cytometer

# Storage and handling

Store at 2-25 °C.

Avoid prolonged exposure to light.

Do not freeze.

See Section Procedure (Reagent Preparation) for information about In-Use stability and shelf-life following the first opening, together with the storage conditions and stability of working solutions (where applicable).

# Warnings, precautions and limitations of use

#### **GHS Hazard Classification**

WARNING: EXCELLYSE XPerm Solution A (ED7397-1) contains formaldehyde (CAS No. 50-00-0) and methanol (CAS No. 67-56-1) in concentrations classified as hazardous.

Label elements	Signal word
	- Danger
H-phrases	H302 Harmful if swallowed.
	H315 Causes skin irritation.
	H317 May cause an allergic skin reaction.
	H319 Causes serious eye irritation.
	H335 May cause respiratory irritation.
	H341 Suspected of causing genetic defects.
	H350 May cause cancer.
P-phrases	P201 Obtain special instructions before use.
	P264 Wash hands and exposed parts of the body thoroughly after handling.
	P280 Wear protective gloves/protective clothing/eye protection.
	P301+P312 IF SWALLOWED: Call a doctor if you feel unwell.
	P302+P352 IF ON SKIN: Wash with plenty of water and soap.
	P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
	P308+P313 IF exposed or concerned: Get medical advice/attention.
	P333+P313 If skin irritation or rash occurs: Get medical advice/attention.
	P362+P364 Take off contaminated clothing and wash it before reuse

Consult Safety Data Sheet (SDS) available on the product page at www.exbio.cz for the full information on the risks posed by chemical substances and mixtures contained in the Product and how they should be handled and disposed.

## **Biological Hazard**

Human biological samples and blood specimens and any materials coming into contact with them are always considered as infectious materials.

Use personal protective and safety equipment to avoid contact with skin, eyes and mucous membranes.

Follow all applicable laws, regulations and procedures for handling and disposing of infectious materials.

#### **Evidence of deterioration**

Normal appearance of the reagents provided is a clear liquid. Do not use the reagents if you observe any change in appearance, for example turbidity or signs of precipitation.

#### Limitation of use

Do not use after the expiry date stated on the product labels.

## Specimen

Use venous peripheral blood collected into specimen receptacle classified as a medical device, with EDTA or Heparin anticoagulant.

## **Procedure**

## Preparation of reagent(s) provided

Bring the reagents to room temperature prior to use.

Following the first opening, the reagents retain their performance characteristics until the expiry date when stored under the stated conditions in their original primary containers.

#### Preparation of materials required but not provided

Bring deionized water and 1X PBS to room temperature prior to use.

## Staining protocol

- 1. For each specimen, label a  $12 \times 75$  mm round bottom test tube with the appropriate sample identification.
- 2. Follow antibody manufacturer's instructions for whole blood staining.
- 3. Add 250 μl of **XPerm Solution A** per 100 μl of whole blood. Mix the content of the tube with a vortex mixer.
- 4. Incubate for 10 minutes at room temperature.
- 5. Add 3 ml of deionized water to the tube, mix well, and incubate for 10 minutes at room temperature.
- 6. Centrifuge the tube for 5 minutes at 300x g.
- 7. Decant supernatant and add 250  $\mu$ l of **XPerm Solution B** to the tube.
- 8. Add appropriate volume of fluorescent-dye-labeled antibodies intended for intracellular staining. Mix the content of the tube with a vortex mixer.
- 9. Incubate for 15-30 minutes at room temperature in the dark.
- 10. Wash the cells: add 2 ml of PBS, centrifuge the cells for 5 minutes at 300x g  $\,$

and decant supernatant.

- 11. Resuspend the pellet in 0.2 0.3 ml of PBS with 1% formaldehyde.
- 12. Analyze the processed sample immediately using flow cytometer. If the stained sample will not be acquired immediately, store at 2-8 °C in the dark and analyze within 24 hours.

#### Flow cytometry analysis

The flow cytometer selected for use with the product EXCELLYSE XPerm shall be calibrated on a routine basis using fluorescent microbeads to ensure stable sensitivity of detectors according to the cytometer manufacturers instructions.

If not maintained properly the flow cytometer may produce false results.

Refer to the manufacturer's cytometer specifications for lasers and fluorescence detectors according to the excitation and emission characteristics of the fluorochromes in Section Equipment required.

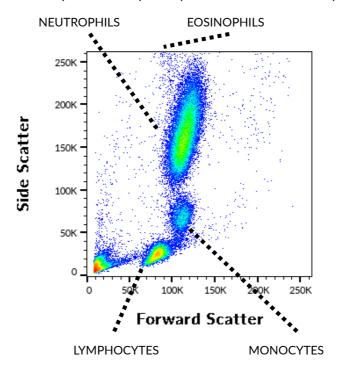
Set voltages on the fluorescence detectors of interest prior to stained specimen analysis. Voltage on a PMT detector should be set high enough, so that minimum of negatively stained events interfere with 0th channel on the fluorescence axis. Also, PMT detector voltage should not exceed values at which positive events are pressed to the right axis.

Compensate fluorescence signals between detectors prior to or after data acquisition. Data may be incorrectly interpreted if fluorescence signals are compensated improperly or if gates are positioned inaccurately.

For measured data analysis, it is possible to use cytometer software developed by the manufacturer, or software dedicated for offline cytometry data analysis.

## Representative data

Figure 1 Two-dimensional density dot-plot showing clusters of peripheral blood leukocytes of EXCELLYSE XPerm processed sample analyzed on BD FACSCanto™ II cytometer.



**Figure 2** Staining profile of whole blood processed with EXCELLYSE XPerm, stained with anti-myeloperoxidase FITC labelled antibody, and analyzed on BD FACSCanto™ II.

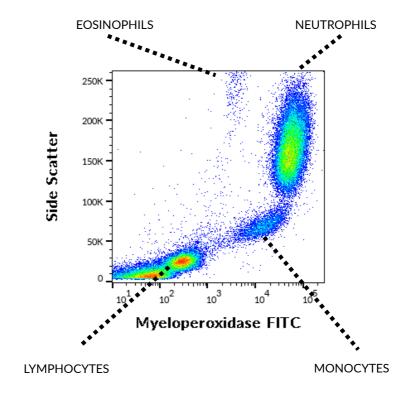
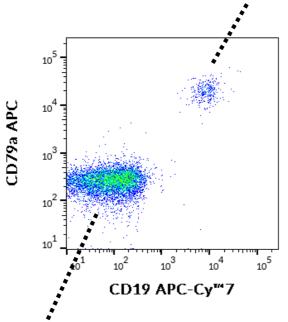


Figure 3 Staining profile of lymphocytes of whole blood sample processed with EXCELLYSE XPerm, stained with anti-CD79a APC and CD19 APC-Cy<sup>™</sup>7 labelled antibodies, and analyzed on BD FACSCanto<sup>™</sup> II cytometer.





CD19+CD79a+ DOUBLE NEGATIVE EVENTS (NK- AND T-CELLS)

### References

N/A

# **Use of Third Party Trademarks**

BD FACSCanto™ II is registered trademark of Becton, Dickinson and Company. Cy™ and CyDye™ are registered trademarks of Cytiva.

# **Revision History**

Version 4, ED7397\_TDS\_v4 TDS layout changed.

## **Manufacturer**

EXBIO Praha, a.s. Nad Safinou II 341 25250 Vestec Czech Republic

#### **Contact Information**

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**NOTICE**: Any serious incident that has occured in relation to the product shall be reported to the manufacturer and the local competent authority.