# PI/RNASE Solution

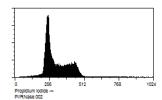




### PRODUCT DESCRIPTION

Propidium lodide (PI) is the most commonly used dye for DNA and cell cycle analysis for flow cytometry. The PI binds to DNA by intercalating into the double stranded macromolecule. Pl also binds to RNA, and is necessary to remove the RNA with a nucleases treatment (RNase) for optimal DNA resolution.

The quantification of the content DNA allows to know the distribution of a cell population along the different phases of the cell cycle. In the analyses of a cell population by flow cytometry using dyes for DNA, the quantity of linked dye is proportional to the quantity of DNA.



The analysis of cell cycle by flow cytometry are represented in fluorescence intensity histograms for it probes specific of DNA. The cells of mammalian are characterized for having three populations or definite regions, cells in G2 and M phases of the cell cycle that have double DNA content of those in G0 and GI phases, and a region correspond to cells in phase S.

### RECOMMENDED USAGE

Immunostep's PI/RNAase, is intended for analyses of cell cycle by flow cytometry.

Presentation: liquid.

Storage instruction: Shipped at ambient conditions, upon arrival store at 4°C

### 3. REAGENTS PROVIDED

Ready to use reeagent containing 200 test ( $500\mu$ I/test) of PI/RNAase in 100 ml of PBS with 0,09% NaN3 (sodium azide)as preservative , pH 7.2.

Revision nr. 4 | Emission date: 10/03/2017

### 4. RECOMMENDATION AND WARNINGS

This product contains sodium azide. In acid conditions, it is transformed into hydrazoic acid, a highly toxic compound. Azide compounds must be diluted in running water before being discarded. These conditions are recommended so as to avoid deposits in plumbing, where explosive conditions could develop.

Do not use after expiration date stamped on vial.

Propidium lodide is a potential carcinogen. It is recommended that the user wear protective clothing, gloves, and eye/face protection in order to avoid contact with the skin and eves.

For professional use only.

For optimal use, do NOT dilute

## 5. STAINING CELLS PROTOCOL WITH PI/RNASE SOLUTION

- Put cells into a 12x75 mm cytometer tube. Harvest the cells corresponding to 2 x 105 to 1 x 106. Centrifuge the cells for 5 minutes at 300xg, and remove the supernatant. Resuspend the pellet in the residual liquid.
- 2. Fix cells and add 200  $\mu l$  of 70% ethanol by pipeting in the cell suspension slowly while vortexing.
- 3. Leave the cells in ethanol at 4°C for 30 min.
- Wash cells once in 2 ml PBS + 2% BSA. Centrifuge the cells for 5 minutes at 300 xg, and remove the supernatant. Resuspend the pellet in the residual limit
- Add 0,5 ml of propidium iodide solution (PI/RNase) to cell pellet and mix well. Incubate 15 minutes at room temperature before to analysis.
- 6. After incubation period, analyze by flow cytometry.

If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem with the product is suspected, contact our Technical Services at tech@immunostep.com.

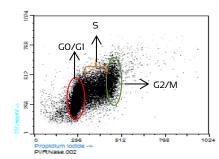


Figure 1. Jurkat cells (T-cell leukemia, human). DNA content analysis histogram (Top panel). Cell cycle analysis (Bottom panel)

Store samples at  $4^{\circ}C$  and protect from the light until anlysed by flow cytometry. Cells my be stored in 70% ethanol al -20  $^{\circ}C$  for several weeks prior to staining and flow cytometric analysis.

#### 6. WARRANTY

Warranted only to conform to the quantity and contents stated on the label or in the product labelling at the time of delivery to the customer. Immunostep disclaims hereby other warranties.

Immunostep's sole liability is limited to either the replacement of the products or refund of the purchase price.



# 7. ADDITIONAL INFORMATION

For research use only. Not for diagnostic use.

Not for resale. Immunostep will not be responsible of violations that may occur with the use of this product. Any use of this product other than the specified in this document is strictly prohibited. Unless otherwise indicated by Immunostep by written authorization, this product is intended for research only and is not to be used for any other purpose, including without limitation, for human or animal diagnostic, therapeutic or commercial purposes. Please, refer to www.immunostep.com technical support for more information.

# 8. EXPLANATION OF SYMBOLS

REF	Catalog reference
$\sum$	Contains sufficient for <n> test</n>
	Regulatory Status
$\Diamond$	Quantity per test
RUO	Research Use Only
***	Manufacturer

#### REFERENCES

Crowley LC, Chojnowski G, Waterhouse NJ. Measuring the DNA Content of Cells in Apoptosis and at Different Cell-Cycle Stages by Propidium lodide Staining and Flow Cytometry. Cold Spring Harb Protoc Oct 03;2016(10);pdb prot087247.

# 10. MANUFACTURED BY:



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