RNAkeep™ Solution

09/20

(Cat# M1241-100, -250, -500; Store at RT)

I. Introduction:

BioVision's RNAkeep™ Solution is a reagent that offers reliable protection and greater stability for cellular RNA at ambient and low temperatures. It is suitable for preserving RNA in cells, fresh clinical or biopsy tissues, throat swabs and other biological samples. The solution is ready to use and doesn't require any extra preparation steps. The solution quickly permeates into the cells and inhibits RNases, DNases and proteinases while stabilizing RNA and DNA. Samples stored in RNAkeep™ Solution are protected from RNases for one week at 25°C, 1 month at 4°C, 12 months at -20°C or longer, when stored below -70°C. Samples stored in RNAkeep™ Solution can be processed for RNA or DNA extraction after removal of the storage solution. The extracted nucleic acids are suitable for RT-qPCR, next generation sequencing and other downstream applications.

II. Application:

- Preserving RNA in cells, fresh clinical or biopsy tissues, throat swabs and other biological samples.

III. Key Features:

- Offers reliable and greater stability for cellular RNA at ambient and low temperatures.
- A cost effective, scalable reagent for clinical and field samples.

IV. Sample Types:

- Cells, tissues, nose or throat swabs and other biological samples.

V. Contents:

Components	M1241-100	M1241-250	M1241-500
RNAkeep™ Solution	100 ml	250 ml	500 ml

VI. User Supplied Reagents and Equipment:

- Microcentrifuge or cryopreservation tubes
- Pipettes, pipette tips
- Tabletop centrifuge
- PBS

VII. Shipment and Storage:

The reagent is shipped at room temperature (RT) and can be stored at RT for up to a minimum of 3 years.

VIII. Preservation Protocols:

For Cell Cultures:

1. Dissociate adherent cells into cell suspension. **Note:** This step is not required for non-adherent cell lines.
2. Add 1 x 10^5 - 1 x 10^7 cells in a microcentrifuge tube and centrifuge at 300 x g for 5 min to pellet cells at the bottom of the tube.
3. Carefully remove the growth media using a pipette. **Note:** Pellet may be washed with PBS resulting in some loss.
4. Resuspend the cell pellet with 100 µl PBS. Add 1 ml of RNAkeep™ Solution and mix well.

For Tissues:

1. Cut each fresh tissue into pieces (< 5 mm) using a scalpel and preserve in RNAkeep™ Solution immediately to avoid degradation.
Note: The maximum recommended wet weight per sample is 100 mg.
2. Use forceps or a sterile pipette tip to submerge the tissue into 5-10 volumes of RNAkeep™ Solution immediately and close the tube.
3. Repeat steps 1 and 2 for each sample within 5 min for best results.
Note: Tissues with high levels of RNases such as liver, kidney, spleen, pancreas, intestine, etc. require storage in more than 10 volumes of RNAkeep™ Solution.

For Nose or Throat Swabs:

1. Prepare tubes (safe-lock tube recommended) containing 0.5 - 1.0 ml of RNAkeep™ Solution.
2. Tap the tube until the liquid collects at the bottom of the tube.
3. Carefully, insert the swabs into the RNAkeep™ Solution until it is completely submerged.
4. Using scissors remove the handle of the swab that extends past the top of the tube and close the tube.

Notes:

1. It is critical that the swabs be submerged in the RNAkeep™ Solution and sealed immediately, after sample collection.
2. Once pathogens are submerged in the RNAkeep™ Solution they become completely inactivated thereby eliminating the risk of handling.

IX. Functional Data:

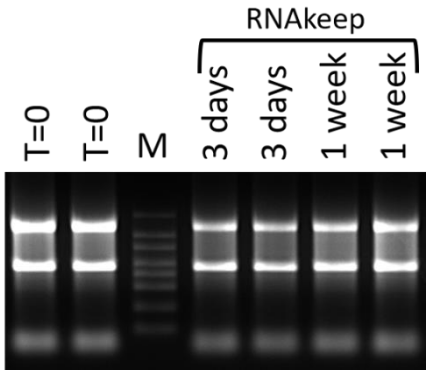

Figure. Jurkat (5 x 10^6) cells were incubated in RNAkeep™ Solution at RT for 3 days and 1 week respectively. RNA was extracted using Trizol™ Reagent and visualized by gel electrophoresis on 1% Agarose gel. M = RNA Ladder. There is no visible difference in the quality of RNA extracted from fresh cells (T= 0) vs cells stored in RNAkeep™ Solution for 3 days (T= 3d) or 1 week (T= 1 week).

X. Recommended Storage:

1. Samples submerged in RNAkeep™ Solution can be stored at RT for up to a week or 4°C for up to one month, if lower temperature is unavailable.
2. Tissues stored in RNAkeep™ Solution should be first incubated at 4°C for at least 12 hr before transferring to -20°C or -80°C.
3. For processing samples stored in RNAkeep™ Solution, centrifuge the samples at 7000 x g for 5 min and carefully remove the RNAkeep™ Solution. Proceed with the rest of the extraction protocol as usual.

XI. Related Products:

Product Name	Cat. No.
EasyRNA™ Cell/Tissue RNA Mini Kit	K1337
EasyRNA™ Bacterial RNA Kit	K1351
EasyRNA™ Blood RNA Mini Kit	K1373
EasyRNA™ Plant RNA Mini Kit	K1374
Yeast RNA Mini Kit	K1418
EasyRNA™ Fungal RNA Kit	K1419
miRNA Extraction Kit	K1456

XII. General Troubleshooting Guide:

Problem	Cause	Solution
Precipitation of RNAkeep™ Solution	Improper storage of RNAkeep™ Solution (below 15-25°C)	A. Agitate and heat gently until the crystals dissolve. Bring the solution to RT before use. B. Store as recommended.
RNA or DNA is degraded	A. Incorrect sample preparation B. Incorrect storage temperature or duration	A. Ensure that the sample size or thickness is not exceeded and proper amount of RNAkeep™ Solution is used. B. Samples should be well mixed before storage**.** Before storing the samples at -20°C or -70°C**,** it should be incubated at 4°C for at least 12 hr.
Low yield of RNA or DNA	A. Loss of sample B. Sample size is too small C. RNA is degraded	A. Pellet the cells and other samples and carefully remove the RNAkeep™ Solution with a pipette for downstream processes. B. Increase the sample size. C. Ensure that the preservation instructions are carefully followed.

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