NOYA-RNAsol

RNA Extraction Reagent

Catalog number: N-1121

« Research Use Only »

Overview

RNAsol reagent is designated for the isolation of total RNA from cultured cells and biological fluids. It separates RNA in a single – step liquid phase. The reagent is guanidium based solution which has been modified according to protocol of Sacchi and Chomzynski. Its main advantages are less time consuming and more sensitivity for low abundant transcripts.

Precautions

RNAsol isolation reagent consists of phenol and guanidium which both are toxic. Avoid to the contact of this solution with acidic reagents and also be aware about the toxic effect of contact with skin and other mucosa. Good lab circulation as prerequisite is highly recommended.

1- Work under fume Hood.
2- Wear gloves, Lab coat, appropriate eye protection.
3- Avoid breathing vapor.

Storage

Store at 2-8^{o}C

Kit Contents

RNAsol isolation reagent, 25 ml, 50 ml

Protocol

1-For each 200µl of plasma or each T25 cell culture flask, Add 1000 µl RNAsol reagent.
2-Homogenize the suspension by passing it, several times, through pipet and then by using 20 G syringe at least for 30 times. Dispose each 1ml of the homogenate to each 1.5 ml microtube.
Note: The homogenized sample can be stored at -80^{o}C for at least one month before using in step 3.
3- Incubate the homogenized sample for 5 min at room temperature to ensure the complete dissociation of nucleoprotein complex.
4- Add 200µl chloroform to each micro tube.
5- Cap securely & shake it vigorously by hand for 15 sec.
6- Incubate at room temperature for 2-15 min.
7- Centrifuge at 12000g 15 min, 2-8^{o}C.
Note: In the case of working with cell suspension, it is more conclusive to repeat chloroform extraction twice. For this reason, transfer upper aqueous phase to a new 1.5ml microtube and add equal volume of chloroform and repeat the steps 5-7.
8- Transfer the upper aqueous phase (which is almost 60% at the volume of whole the reagent) to a new 1.5ml microtube.

Optional: Add 5µl RNA Binder (NOYA-Glycogen MB, N-1151) as a carrier to assist RNA precipitation at the bottom of microtube.
9- Add 500µl isopropanol to the aqueous phase (to precipitate the RNA from the colorless aqueous phase).
10- Invert the tube several times and mix it thoroughly or vortex it (5-10 sec).
11- Incubate for 10 minute at 15-25°C to complete precipitation of the RNA.
12- Centrifuge at 12000g for 10 min, 2-8°C.
13- Discard the supernatant.
14- Add 1ml nuclease free 75% ethanol to each microtube.
Note: Prepare nuclease free 75 % ethanol by DEPC ultrapure water (NOYA-DEPC-Treated water, N-1181)
15- Centrifuge at 7500g for 5 min, 2- 8°C.
16- Discard the supernatant.
17- Repeat the steps 14-16.
18- Remove ethanol from the RNA pellet by air drying or placing it under vacuum for 5-10 min.
Caution: Don't dry the RNA pellet by centrifugation under vacuum; don't let it to completely dry, as a dry pellet will be less soluble.
19- Add 20µl (TE pH 7:50 or nuclease free water {NOYA-RNase free water, N-1191) to the pelleted RNA.
20- Dissolve the RNA pellet by passing the solution through pipette tip or gently vortex it 5-10 sec.
21- Incubate for 10 min at 55-60°C.
The RNA may be used immediately for amplification, or maybe stored at -80°C

Notes

- **FOR RESEARCH USE ONLY. NOT FOR HUMAN OR DIAGNOSTIC USE.**
- Please follow up general laboratory precaution and utilize safety while using this kit.

Related Products

- RNA-binder (NOYA-Glycogen MB, N-1151)
- DEPC- treated water (NOYA-DEPC-Treated water, N-1181)
- Nuclease free water (NOYA-RNase free water, N-1191)