

VERSION 2

FEB 09, 2023

OPEN ACCESS

**DOI:**  
[dx.doi.org/10.17504/protocols.io.ewov1o5qklr2/v2](https://dx.doi.org/10.17504/protocols.io.ewov1o5qklr2/v2)

**Protocol Citation:** Boyang Liu, Liangyu Cui, Zhangwen Deng, Yue Ma, Diancheng Yang, Yanan Gong, Yanchun Xu, Shuhui Yang, Song Huang 2023. RNA extraction protocol for snake genomes using TRIZOL reagent (Invitrogen) . **protocols.io** <https://dx.doi.org/10.17504/protocols.io.ewov1o5qklr2/v2> Version created by Boyang Liu

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**Protocol status:** Working

**Created:** Feb 06, 2023

**Last Modified:** Feb 09, 2023

**PROTOCOL integer ID:**  
76457

## RNA extraction protocol for snake genomes using TRIZOL reagent (Invitrogen) V.2

In 3 collections

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### ABSTRACT


A protocol to extract RNA from snake muscle using TRIzol reagent (Invitrogen, USA) and RNA extraction kit (Thermo Scientific, USA) in this protocol.

## RNA extraction

1h 5m 15s

- 1



The laboratory and test bench for RNA extraction need to be sterilized using alcohol and ultraviolet light for 30 minutes.

 00:30:00

All the equipment used in the experiment, such as mortar and pestle, are sterilized at high temperature one day before the experiment and dried in an oven.


Masks and gloves need to be worn throughout the RNA extraction process. Avoid exposure of RNases to experimental samples and cause RNA degradation.
- 2



On the clean bench, take 50 mg of scented glands and put them into a pre-cooled mortar to ensure that the liquid nitrogen has never passed the sample, quickly grind the sample into powder, add 1 ml TRIZOL, mix the tissue powder with TRIZOL, and put it into In a centrifuge tube, stand at room temperature for 15 min.


 Room temperature  00:15:00
- 3

Add 200 ml of chloroform, shake the centrifuge tube for 15 s to mix the chloroform and TRIZOL well.



Stand at room temperature for 10 min, put it in a refrigerated centrifuge and centrifuge at 4°C, 12000r/min, 15 min.

 00:00:15


 Room temperature  00:10:00

 12000 rpm, 4°C, 00:15:00
- 4


After centrifugation, the solution should be separated into three phases. Take the upper aqueous phase into a new centrifuge tube, add three times the volume of ethanol to mix, and then stand at room temperature for 10 min.

 Room temperature  00:10:00
- 5

Add the mixed solution to the purification column, centrifuge at 12000r/min at 4°C for 2 min, repeat three times to ensure that ethanol is removed.

 12000 rpm, 4°C, 00:02:00
- 6

RNA is purified with an RNA extraction kit (Thermo Scientific, USA), and RNA was eluted with DEPC.

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- 7 Add RNase inhibitors to ensure that the RNA will not be degraded, and mark the outside of the centrifuge tube.

Check RNA purity and concentration, and confirm RNA integrity using electrophoresis.