



# Nucleic acid and protein purification with TRItidy G™

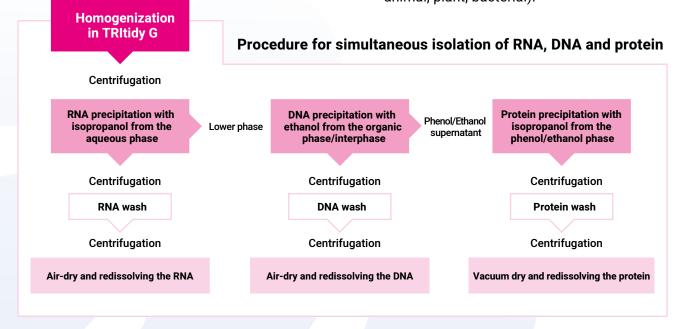
Simultaneous isolation of RNA, DNA and proteins from biological samples was firstly introduced in 1993, based on the use of a reagent containing phenol and guanidine thiocyanate. The simultaneously isolated RNA, DNA and proteins are ready for Northern, Southern and Western blotting as well as PCR, RT-PCR enzymatic assays. The complete recovery of DNA from samples used for the RNA and protein isolation makes it possible to normalize the results of gene expression studies based on DNA content instead of on the more variable total RNA, protein content or tissue weight.

TRItidy G<sup>™</sup> is a monophasic reagent, based on the Chomczynski method, with additional modifications to improve the purity of the RNA, DNA and proteins. First, the RNA is selectively retained in the aqueous phase during the acidic extraction, while DNA and proteins stay in the organic phase and interphase, respectively. The DNA is isolated from the interphase/organic phase by a simple ethanol precipitation and proteins from the remaining organic phase.



# **Main advantages**

- TRItidy G<sup>™</sup> allows one-step isolation from the biological sample.
- Monophasic reagent
- No need of purification columns for the isolation of nucleic acids and proteins.
- Quick procedure.
- Easy to reproduce.
- Suitable for small and large samples (human, animal, plant, bacterial).











# Nucleic acid and protein purification with TRItidy G™

## **Preparation of samples**

Depending on your sample type, homogenization of samples should be performed according to the protocol below. The volume of the sample must not exceed 1/10 of the volume of **TRItidy G<sup>TM</sup>**.

Sample type	Procedure
Tissue	Tissue is homogenized in approx. 1 mL <b>TRItidy G™</b> per 50 - 100 mg tissue.
Cell culture cells (growing in monolayer)	Cells are lysed in 1 mL/10 cm <sup>2</sup> (3.5 cm diameter) dish, after aspiration of the medium.
Suspension cells	Cells have to be collected by centrifugation before addition of the reagent (1 mL <b>TRItidy G</b> <sup>TM</sup> per 1-5 x $10^6$ cells; bacteria up to 1 x $10^7$ ).
Blood samples, serum or other biological fluids*	Add 750 µL of <b>TRItidy G™</b> per 250 µL of sample volume.

\* Biological fluids with high levels of protein or other contaminating substances (e.g. whole blood) may be diluted 1:1 with RNase-free, molecular biology grade water (Suggested product: **A7398**).

### **Phase separation**

Homogenization of the sample

Incubation time (5 min)

Add 0.2 mL of chloroform to the lysate

Additional incubation of 10 minutes

Centrifuge the lysate at 12 000 g for 15 min at 4°C

#### Purification protocol for RNA, DNA and PROTEINS

#### **RNA** Isolation

- **1.** Transfer the aqueous phase to a new tube.
- 2. Add 1:1 isopropanol.
- **3.** Precipitate the RNA on ice (15 min) and centrifuge.
- 4. Wash and air-dry RNA.
- 5. Dissolve in 20  $\mu L$  DEPC-treated water.

#### **DNA** Isolation

- **1.** Add ethanol and incubate (5 min).
- **2.** Centrifuge and remove the supernatant (protein).
- **3.** Wash with sodium citrate 0.1M and centrifuge (5 min).
- **4.** Air-dry the DNA and dissolve in approx. 0.5 mL 1X TE.

#### **Protein Isolation**

- **1.** Add isopropanol to the supernatant (2:1).
- 2. Centrifuge (10 min).
- Wash protein precipitate with guanidine hydrochloride 0,3M and centrifuge (5 min).
- **4.** Air-dry the precipitate and dissolve in 1% SDS.

#### **Ordering information**

Product number	Product name	Pack sizes
A4051	TRItidy G™	100 mL, 200 mL

**Caution: TRItidy G™** contains Phenol and Guanidinium thiocyanate. For safety instructions please read the Material Safety Data Sheet (MSDS) before use.



#### **Related products**

	Product number	Product name
	A0881	DEPC BioChemica
	A3678	Ethanol absolute for molecular biology
	A1106	Guanidine Hydrochloride for molecular biology
	A7398	Water Molecular biology grade

IP-033EN

#### AppliChem GmbH

Ottoweg 4 D-64291 Darmstadt Germany Phone +49 6151 9357 0 Fax +49 6151 9357 11 info.de@itwreagents.com

#### ITW Reagents, S.R.L.

Corso Milano 31 I-20900 Monza (MB) Italy Phone +39 039 9530 360 Fax +39 039 9530 361

info.it@itwreagents.com

#### Panreac Química S.L.U.

C/ Garraf 2, Polígono Pla de la Bruguera E-08211 Castellar del Vallès (Barcelona) Spain Phone +34 937 489 400

Phone +34 937 489 400 Fax +34 937 489 401 info.es@itwreagents.com



itwreagents.com