

# REAGENTS

## Molecular Biology Reagent Ethanol

Sigma now exclusively offers both 100%, and 95% non-denatured Molecular Biology Reagent Ethanol. Both products are application tested to verify DNA and RNA precipitation and the absence of nucleases. Both are offered as Excise Tax included. These products are benzene-free, meet ACS requirements, and are only available in the United States.

Product	Product Description	Quantity
<a href="#">E 7023</a>	Ethanol, Absolute, 100% (200 proof) (Ethyl Alcohol) Water ≤ 0.05% Molecular Biology Reagent, DNase, and RNase Free Excise Tax included: No ATF license required Suitable for use in the precipitation of nucleic acids. [64-17-5] C <sub>2</sub> H <sub>5</sub> OH	500 ml 6 x 500 ml
<a href="#">E 7148</a>	Ethanol, Absolute, 95% (190 proof) (Ethyl Alcohol) 95+% Ethanol Molecular Biology Reagent, DNase, and RNase Free Excise Tax included: No ATF license required Suitable for use in the precipitation of nucleic acids. [64-17-5] C <sub>2</sub> H <sub>5</sub> OH	1 gal 4 x 1 gal

Only Available in U.S.

## GenElute™ LPA Linear Polyacrylamide

GenElute LPA Linear Polyacrylamide is an efficient neutral carrier for precipitating picogram amounts of nucleic acids with ethanol. The nucleic acid precipitate can be collected simply by centrifugation. LPA offers several advantages for recovering DNA or studying DNA-protein interactions, relative to other carriers, such as tRNA or glycogen. tRNA interferes with DNA during phosphorylation with polynucleotide kinase and glycogen competes with protein in DNA-protein interaction studies. In contrast, LPA is completely inert. LPA is synthesized chemically and, therefore, is not contaminated with biological material. The precipitate is visible immediately upon addition of LPA, thus eliminating wait time and low temperature incubation.

The presence of LPA during ethanol precipitation results in complete recovery of fragments larger than 20 base pairs, whereas most of the DNA is lost if no carrier is used. Very short DNA fragments, less than 20 base pairs, do not coprecipitate with LPA<sup>1</sup>, allowing separation of labeled DNA from unreacted nucleotides by precipitation after the labeling reaction.

LPA has been used in several laboratories for most of the common manipulations of DNA, including automated sequencing, enzyme reactions, gel electrophoresis, cloning<sup>2</sup>, and DNA-protein interactions<sup>1,3</sup>, and appears inert in all experiments. A very small amount of LPA is required as carrier during ethanol precipitation of DNA.

GenElute-LPA is tested for nucleases, and is supplied in nuclease-free water as a 5-mg/ml solution.

Product	Product Description	Quantity
<a href="#">5-6575</a>	GenElute-LPA 5 mg/ml in nuclease-free water	5 x 1 mL

### References

1. Gaillard, C. and F. Strauss, *Nucl. Acid Res.* **18**: 378 (1990).
2. Strauss, F. and A. Varshavsky, *Cell* **37**: 889-901 (1984).
3. Aruffo, A. and B. Seed, *Proc. Natl. Acad. Sci. USA* **84**: 8573-8577 (1987).

## AGARASE

### From *Pseudomonas atlantica*

An agarose-digesting enzyme ideal for isolation of intact high molecular weight DNA from agarose gel slices. The enzyme and carbohydrate digestion products generally do not interfere with subsequent DNA manipulations, but the DNA may be purified if necessary by ethanol precipitation.

DNase, RNase, and protease: None detected.  
[37288-57-6]  
(EC 3.2.1.81)

**Unit Definition:** One unit will digest 100 ml of molten 1% low-melting agarose.

**Functional assay:** tested for suitability in isolating lambda DNA Hind III fragments from low-melting agarose gels.

Product	Product Description	Quantity
<a href="#">A 8688</a>	AGARASE Vials of 100 units	1 vial

# REAGENTS

## Glycogen, Molecular Biology Reagent

### From mussels

RNase, DNase, Nickase, Protease, Nucleic Acids – None detected.  
Each vial contains an aqueous solution of Glycogen (approx. 20 mg in 1 ml). It is intended as a carrier molecule for DNAs and RNAs, replacing tRNAs or sonicated DNAs.

Product	Product Description	Quantity
<a href="#">G 1767</a>	Glycogen, Molecular Biology Reagent	1 vial

## GUANIDINE

Forms a clear, colorless solution at 6 M.

Pb ≤5 ppm  
DNase and RNase: None detected.  
[50-01-1]  $\text{CH}_5\text{N}_3 \cdot \text{HCl}$  FW 95.53

**Unit Definition:** One unit will digest 100 ml of molten 1% low-melting agarose.

**Functional assay:** tested for suitability in isolating lambda DNA Hind III fragments from low-melting agarose gels.

Product	Product Description	Quantity
<a href="#">G 3272</a>	GUANIDINE (Aminomethanamide)	25 g 100 g
	Hydrochloride	500 g
	Purity: 99+%	1 kg 2 kg

## GUANIDINE HYDROCHLORIDE

Filtered 8 M aqueous solution.  
d = 1.186 g/ml  
[50-01-1]

Product	Product Description	Quantity
<a href="#">G 9284</a>	GUANIDINE HYDROCHLORIDE	100 ml 500 ml

## GUANIDINE THIOCYANATE

Assay: ≥99%  
DNase and RNase: None detected.  
[593-84-0]  $\text{CH}_5\text{N}_3 \cdot \text{HSCN}$  FW 118.2

Product	Product Description	Quantity
<a href="#">G 9277</a>	GUANIDINE THIOCYANATE	100 g 250 g 500 g 6 x 500 g

## ISOPROPANOL

Suitable for use in the precipitation of nucleic acids.

When compared to ethanol, 50% less is required for nucleic acid precipitation, thus minimizing the total volume to be centrifuged for DNA or RNA recovery.

Water: ≤0.05%  
[67-63-0]  $\text{C}_3\text{H}_8\text{O}$  FW 60.10

### Reference

1. Sambrook, J., et al., Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratory (1989) p. E.13-E.14.

Product	Product Description	Quantity
<a href="#">I 9516</a>	ISOPROPANOL (2-Propanol)	25 ml 4 x 25 ml
	Purity: 99+%	500 ml

# REAGENTS

## LYSOZYME

Hydrolyzes the  $\beta$ -1,4 linkages between N-acetylmuramic acid and N-acetylglucosamine in the cell wall structure of many microorganisms. This is particularly useful for lysing gram positive and gram negative bacteria for subsequent nucleic acid extraction. Each lot is use tested in the isolation of plasmid DNA from *E. coli*.

Approx. 95% protein; balance primarily buffer salts as sodium acetate and sodium chloride.

**Activity:** Approx. 50,000 units per mg protein ( $E_{285}^{1\%}$ ).

**Unit Definition:** One unit will produce a  $\Delta A_{450}$  of 0.001 per min at pH 6.24 at 25°C, using a suspension of *Micrococcus lysodeikticus* as substrate, in a 2.6 ml reaction mixture (1 cm light path). M.W. approx. 14,300. [12650-88-3]

### Reference

- Jolles, P., *Angew. Chem. Int. Ed.*, **8**, 227 (1969).
- Sambrook, J., et al., *Molecular Cloning: A Laboratory Manual*, Cold Spring Harbor Laboratory p. 1.29 (1989).

Product	Product Description	Quantity
<a href="#">L 7651</a>	LYSOZYME (Muramidase; mucopeptide N-acetylmuramoylhydrolase; EC 3.2.1.17) From Chicken Egg White 3x Crystallized, dialyzed and lyophilized	1 g 5 g 10 g 25 g 100 g

## POLYETHYLENE GLYCOL

Av. Mol. Wt.: 8000  
DNase, RNase: None detected.  
[25322-68-3]

Product	Product Description	Quantity
<a href="#">P 5413</a>	POLYETHYLENE GLYCOL	500 g 1 kg 2 kg

## PROTEINASE K

### *From Tritirachium album*

(EC 3.4.21.64)  
No detectable endonuclease (nickase), endonuclease-exonuclease or RNase activity. Specific conditions given on accompanying data sheet. <0.5 ppm DNA using pico green assay  
[39450-01-6]

**Unit Definition:** One unit will hydrolyze casein to produce color equivalent to 1.0  $\mu$ mole (181 mg) of tyrosine per min at pH 7.5 at 37 °C (color by Folin-Ciocalteu reagent).

**Activity:** 10-20 units per mg protein.

Product	Product Description	Quantity
<a href="#">P 2308</a>	Lyophilized powder containing minimum 90% protein (Biuret)	5 mg 10 mg 25 mg 100 mg 500 mg 1 g

## PROTEINASE K

### *From Tritirachium album*

(EC 3.4.21.64)  
No detectable endonuclease (nickase), endonuclease-exonuclease or RNase activity. Specific conditions given on accompanying data sheet. <0.5 ppm DNA using pico green assay  
[39450-01-6]

Solution in 40% glycerol (v/v) containing 10 mM Tris-HCl, pH 7.5, with 1 mM calcium acetate

Prepared from P 2308

**Unit Definition:** One unit will hydrolyze urea-denatured hemoglobin to produce color equivalent to 1.0  $\mu$ mole (181 mg) of tyrosine per min at pH 7.5 at 37 °C (color by Folin-Ciocalteu reagent).

Product	Product Description	Quantity
<a href="#">P 4850</a>	Minimum 140 units/ml, Buffered aqueous glycerol solution	5 ml

# REAGENTS

## RIBONUCLEASE A

### From Bovine Pancreas

Ribonuclease A (RNase A) is commonly used to degrade RNA in DNA preparations.  
(EC 3.1.27.5)  
[9001-99-4]

**Activity:** ≥70 Kunitz units per mg protein.

**Functional assay:** Tested in plasmid purification.

**Detection limit:** Degradation of 10% of the DNA.

At concentrations up to 25 mg per ml, nicking or degradation of plasmid is not detectable.

### Reference

1. Maniatis, T., et al., *Molecular Cloning: A Laboratory Manual*, Cold Spring Harbor Laboratory, p. 451 (1983).

Product	Product Description	Quantity
<a href="#">R 6513</a>	Chromatographically purified, lyophilized powder Note: Boiling of stock solutions to inactivate residual DNase is not necessary or recommended.	10 mg 50 mg 250 mg 500 mg 1 g
<a href="#">R 4642</a>	Solution in 50% glycerol containing 10 mM Tris-HCl, pH 8.0 Note: Boiling of solutions to inactivate DNase is unnecessary and is not recommended.	10 mg 50 mg 250 mg 1 g

## Ribonuclease Inhibitor

**Source:** Human Placenta

Solution in 50% glycerol, 20 mM HEPES-KOH, pH7.6, 50 mM KCl and 8 mM DTT.

Useful for *in vitro* inhibition of ribonucleases, including procedures like cDNA synthesis, RT-PCR, and *in vitro* transcription and translation.

**Activity:** Approx. 30,000-50,000 units per ml.

**Unit definition:** One unit will reduce the activity of 5 ng of ribonuclease A by 50% in a cytidine 2':3'-cyclic monophosphate system.

Approx. 50 kDa

**Storage:** -20 °C

Shipped in dry ice

### Reference

1. Blackburn, P., Ribonuclease inhibitor from human placenta: interaction with derivatives of ribonuclease A. *J. Biol. Chem.*, **254**, 12488-12493 (1979).

Product	Product Description	Quantity
<a href="#">R 2520</a>	Ribonuclease Inhibitor	2,500 units 10,000 units 20,000 units

## protectRNA™ RNase INHIBITOR, 500x

A potent inhibitor of most nucleic acid binding enzymes, and thus useful as an RNase inhibitor. Especially useful when performing *in situ* hybridization. If it is added to all aqueous solutions used, it eliminates the need for special glassware washing and after-wash treatments. The 500x concentrate is economical; 2 ml treats 1,000 ml of solution. Not recommended in systems where other enzymatic activity is required.

Product	Product Description	Quantity
<a href="#">R 7397</a>	protectRNA™ RNase INHIBITOR, 500x	30 ml

## RNaseZAP™

A cleaning agent for removing RNase from glassware, plastic surfaces, countertops, and pipettors. It is also effective at eliminating RNase contamination from microcentrifuge tubes without inhibiting subsequent enzymatic reactions.

Product	Product Description	Quantity
<a href="#">R 2020</a>	RNaseZAP™	250 ml 6 x 250 ml