Thermo SCIENTIFIC

IMPROVED

PRODUCT INFORMATION

Glycogen, molecular biology grade

Inert co-precipitant of nucleic acids.
Derived from oysters.

#____ ____

Lot: ____

Concentration: 20 mg/ml aqueous solution.

Store at -20°C

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Description

Glycogen is a highly purified polysaccharide derived from oysters. It is an inert carrier, free of host DNA/RNA. Glycogen is insoluble in ethanol solution; in the presence of salts it forms a precipitate that traps the target nucleic acids. During centrifugation, a visible pellet is formed, which greatly facilitates handling of target nucleic acids. Glycogen quantitatively precipitates nucleic acids from diluted solutions with a higher efficiency than that of tRNA, linear polyacrylamide or sonicated DNA (1-4).
Glycogen molecules are highly branched structures composed of thousands of glucose molecules bonded to each other. The molecular weight of the largest individual glycogen molecule containing about 50,000 glucose molecules appears to be 8 million.

Protocol for DNA Precipitation from Diluted Solutions

1. Add 1/10 volume of 3 M sodium acetate (or 2 M sodium chloride, or 5 M ammonium acetate) to DNA solution.
2. Add glycogen to a final 0.05-1 µg/µl concentration.
   Use up to 1 µl of glycogen per 20 µl of the solution. For precipitation of oligonucleotides, do not use higher than 1 µg/µl final glycogen concentration.
3. Add 1 volume of isopropanol (or 2.5 volumes of ethanol) to the solution. Mix gently but thoroughly.
   Use ethanol for < 200 bp fragments.
4. Incubate the mixture at -20°C for up to 60 min, or at -70°C for 30 min.
   Longer incubation and lower temperature provide better recovery of nucleic acids.
5. Centrifuge the mixture for 10-15 min at 10,000 rpm.
6. Discard the supernatant.
7. Rinse the pellet with cold 70% ethanol.
8. Air-dry the pellet. Avoid over-drying the pellet, as it then takes more time to dissolve.
9. Dissolve DNA in Water, nuclease-free (#R0581) or TE buffer.

Note

- 5 µg of Glycogen (0.25 µl) forms a visible pellet.
- Optimal for recovery of oligonucleotides (>8 bases) and low amounts (>20 pg) of nucleic acids from diluted solutions.
- Up to a final concentration of 8 µg/µl, glycogen does not interfere with most downstream applications: PCR*, DNA sequencing, DNA digestion, ligation, DNA labeling, random priming, DNA amplification using phi29 DNA Polymerase (#EP0091).
- Up to a final concentration of 0.4 µg/µl, glycogen does not affect *in vitro* transfection of eukaryotic cells.
- Does not interfere with gel electrophoresis of nucleic acids.
- Does not interfere with spectrophotometrical determination of nucleic acids concentration ($A_{260-280}$ measurements).

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* The Polymerase Chain Reaction (PCR) process is covered by U.S. patents owned by Hoffman-La Roche.

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CERTIFICATE OF ANALYSIS

Nucleic Acids Precipitation Assay

5 pg of [^{33}P]-labeled calf thymus DNA in 500 µl TE buffer supplemented with 0.4 M LiCl was precipitated with 1 µl (20 µg) of glycogen and 1.2 ml of 96% ethanol. >95% of the radioactivity was detected in the precipitate.

Nicking Activity Assay

1 µg of pUC19 DNA incubation with 200 µg of glycogen at 37°C for 4 hours resulted in no detectable conversion of supercoiled DNA to nicked DNA form.

Labeled Oligonucleotide (LO) Assay

Single-stranded and double- stranded [^{33}P]-labeled oligonucleotides were incubated with 50 µg of glycogen in five restriction digestion buffers at 37°C and 55°C for 16 hours. No detectable degradation of oligonucleotides was observed.

Ribonuclease Assay

1 µg of *E.coli* [^{3}H]-RNA was incubated with 200 µg of glycogen at 37°C for 4 hours.No detectable radioactivity was released into the trichloroacetic acid-soluble fraction.

Protease Assay

0.6% FTC-casein was incubated with 1000 µg of glycogen in 200 µl of reaction buffer at 37°C for 16 hours.
No detectable degradation of the protein was observed.

Nucleic Acids Assay

200 µg glycogen was incubated with 6 pmol [γ-^{32}P]- or [γ-^{33}P]-ATP and 10 units of T4 PNK for 20 min in 40 µl of reaction buffer. Reaction mixture was precipitated with ethanol. Radioactivity in the precipitate did not exceed that in negative control.

Quality authorized by:  Jurgita Zilinskiene

References

1. Tracy, S., Improved rapid methodology for the isolation of nucleic acids from agarose gels, Prep. Biochem., 11, 251-268, 1981.
2. Helms, C., A new method for purifying lambda DNA from phage lysates, DNA, 4, 39-49, 1985.
3. Hengen, P. N., Methods and reagents – Carriers for precipitating nucleic acids, TIBS, 21, 224-225, 1996.
4. Sambrook, J., Russell, D.W., Molecular Cloning: A Laboratory Manual, the Third edition, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York, A8.12-8.13, 2001.

PRODUCT USE LIMITATION

This product is developed, designed and sold exclusively *for research purposes and in vitro use only.* The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals.
Please refer to www.thermoscientific.com/onebio for Material Safety Data Sheet of the product.