3050 Spruce Street, St. Louis, MO 63103 USA
Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757
email: techservice@sial.com sigma-aldrich.com

Product Information

Diethyl pyrocarbonate

Catalog Number **D5758**
Storage Temperature 2–8 °C

CAS RN 1609-47-8
Synonyms: DEPC, DEP, Pyrocarbonic Acid Diethyl ester, Diethyl Oxydiformate, dicarbonic acid diethyl ester[6]

Product Description

Molecular formula: $C_6H_{10}O_5$
Molecular mass: 162.1
Density: 1.12 g/mL
Molarity: 6.9 M
Refractive Index: 1.398 at 20 °C

Diethyl Pyrocarbonate (DEPC) is a well-known nuclease inhibitor, particularly against ribonucleases (RNases).[1-4] DEPC reacts with many enzymes containing -NH, -SH, or -OH groups in their active sites. In particular, DEPC reacts with Cys, His, Lys, Ser, Thr, and Tyr residues, as well as N-terminal sites of proteins.[5]

DEPC acts as a condensing agent between lysine ε-amine groups and the carboxyl groups of aspartic or glutamic acid. DEPC has been used as a gentle esterifying agent and preservative.[6]

An assay method for DEPC uses 5-thio-2-nitro-benzoate.[3]

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions

DEPC can be solubilized in 95% ethanol, e.g. 1.6 mL in 4 mL ethanol gives a clear, colorless solution. DEPC is soluble in alcohols, esters, ketones and other hydrocarbons, but has limited solubility in water (~0.1%).[6] DEPC hydrolyzes in aqueous solution, to form ethanol and carbon dioxide (CO_2).

Typically a 0.1% DEPC solution (1 mL of DEPC diluted in water to 1 L) is used to inactivate RNase. The DEPC will not immediately dissolve, as evidenced by the appearance of globules. The mixture should be stirred until the globules disappear.

DEPC is sensitive to pH. In phosphate buffer at 25 °C, its half-life is 4 minutes at pH 6, and 9 minutes at pH 7. Hydrolysis is accelerated by Tris buffer,[7] where at 25 °C, the half-life of DEPC was reported to be 1.25 minutes at pH 7.5, and 0.37 minutes at pH 8.2.[7]

DEPC will decompose in solution when autoclaved. For 0.1% DEPC solutions, autoclaving for 15 minutes per liter should be sufficient.

Storage/Stability

DEPC is very sensitive to moisture. As such, DEPC is packaged under argon to help reduce exposure to moisture. If DEPC is exposed to even traces of moisture, some hydrolysis occurs. The resulting CO_2 is more soluble in DEPC solutions at 2–8 °C than at room temperature. As the product is brought to room temperature before opening, the DEPC can become supersaturated with respect to any dissolved gas.

After opening the bottle for the first time and each time thereafter, layer nitrogen or argon gas over the DEPC and store the closed bottle at 2–8 °C for optimal stability. It may be helpful to store the bottle inside a sealed plastic bag with desiccant, but with the bottle cap slightly loose. If the bag inflates at all, this indicates some degree of decomposition with possible pressure build-up. Once opened, the bottle should not be kept for more than 9 months.

DEPC decomposes at 155 °C. DEPC is also sensitive to ammonia, which causes decomposition to urethane.

Procedure

1. DEPC will dissolve some plastic pipettes. Thus glass pipettes should be used. DEPC has been reported to be incompatible with polycarbonate or polystyrene containers.[9,10]
2. To decontaminate glassware of RNase, the glassware may be incubated with a DEPC solution for 30-60 minutes, and subsequently autoclaved to decompose the DEPC.
   Note: If one does not wish to autoclave, DEPC-treated water may be boiled at 100 °C for 15 minutes.

References

1. Williamson, R. *et al.*, *Biochemistry*, **10(16)**, 3014-3021 (1971).
2. Solymosy, F. *et al.*, *Eur. J. Biochem.*, **5(4)**, 520-527 (1968).
3. Sambrook, J., Fritsch, E.F., and Maniatis, T., *Molecular Cloning: A Laboratory Manual, Second Edition*. Cold Spring Harbor Press (Cold Spring Harbor, NY) pp. 7.3-7.4 (1989).
4. Ehrenberg, L. *et al.*, *Prog. Nucleic Acid Res. Mol. Biol.*, **16**, 189-263 (1976).
5. Zhou, Y., and Vachet, R.W., *J. Am. Soc. Mass Spectrom.*, **23(5)**, 899-907 (2012).
6. *The Merck Index*, 12th Ed., Entry# 8182, p. 1375 (1996).
7. Berger, S.L., *Anal. Biochem.*, **67(2)**, 428-437 (1975).
8. *Data For Biochemical Research*, 3rd Edition (Dawson, R.M.C., *et al.*, eds.). Oxford University Press (Oxford, UK), p. 389 (1984).
9. Konradi, C.L., "Quantification of mRNA in Neuronal Tissue by Northern Analysis", in Drugs of Abuse: Neurological Reviews and Protocols (J.Q. Wang, ed.). *Methods in Molecular Medicine*, Humana Press (Totowa, NJ), pp. 161-180 (2003).
10. Paul, J.H., "Gene Expression by mRNA Analysis", in *Marine Microbiology* (J.H. Paul, ed.). *Methods in Microbiology*, Vol. 30, Academic Press (London / San Diego), p. 397 (2001).

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