# Product Information



3050 Spruce Street, Saint 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

**Deoxyribonuclease I**
**bovine, recombinant**
**expressed in a proprietary host**

Catalog Number **D7691**
Storage Temperature –20 °C

CAS RN 9003-98-9
EC 3.1.21.1
Synonyms: DNase I; Deoxyribonuclease A; Deoxyribonucleate 5′-oligonucleotidohydrolase

**Product Description**
This product is a recombinant, bovine protein with a molecular mass of ~30 kDA[1] expressed in a proprieatary, non-animal host. It is prepared by a process that uses no animal source products. The host has levels of endogenous RNase considerably less than bovine pancreas, the classic source of DNase I. This product is prepared essentially free of RNase and protease activity.

Deoxyribonuclease I (DNase I) is an endonuclease that hydrolyzes double-stranded or single-stranded DNA preferentially at sites adjacent to pyrimidine nucleotides. The product of hydrolysis is a complex mixture of 5′-phosphate mononucleotides and oligonucleotides. In the presence of $Mg^{2+}$ ions, DNase I attacks each strand of DNA independently and the cleavage sites are random. In the presence of $Mn^{2+}$ ions, DNase I cleaves both strands of DNA at approximately the same site.[2] Most protocols use magnesium ions with DNase I,[3] but for specific cleavage, manganese ions are cited.[4]

DNase has been used to introduce random nicks into double-stranded DNA in preparation for radiolabeling by nick translation or to introduce a single nick into circular DNA in preparation for resection. Protocols for digestion of DNA using DNase have been published.[2,3] DNase may be used for degradation of the DNA template following transcription reactions and removing genomic DNA from isolated RNA samples.

Optimal pH:[3] 7–8

Activators:
DNase I activity has an absolute requirement for divalent metal cations. The most commonly used is $Mg^{2+}$;[1,5] however, $Mn^{2+}$, $Mg^{2+}$, $Ca^{2+}$, $Co^{2+}$, and $Zn^{2+}$ also activate DNase I.[3,5] $Ca^{2+}$ ion concentration at 5 mM will stabilize DNase I against proteolytic digestion; 0.1 mM will reduce the rate of inactivation by one-half.

DNase I activity is inhibited by reduction with 2-mercaptoethanol. The reduced enzyme is inactive, but can be reactivated in presence of Ca or Mg ions.[5] It is also inhibited by chelators, sodium dodecyl sulfate (SDS),[6] and actin.[7] There is no general inhibitor specific for DNase I.[3]

DNase I activity in a reaction mixture may be inactivated by phenol-chloroform extraction. This preparation of DNase is completely inactivated by a 10 minute incubation at 75 °C.

The product is supplied as a solution in 20 mM HEPES, pH 7.5, 10 mM $CaCl_2$, 10 mM $MgCl_2$, 1 mM DTT, and 50% glycerol.

Specific activity: ≥10,000 units/mg protein

The activity of this preparation is assayed according to the method of Kunitz.[8]

Unit Definition: One Kunitz unit will produce a $\Delta A_{260}$ of 0.001 per minute per ml at pH 5.0 at 25 °C, using calf thymus DNA as the substrate with the $Mg^{2+}$ concentration equal to 4.2 mM.

**Storage/Stability**
The product ships on wet ice and it is recommended to store the product at –20 °C. The product, as supplied, retains activity for at least one year when stored properly.

A working solution should be kept on ice the day it is prepared and only warmed when it is to be used.

**References**


1. Liao, T.-H., et al., J. Biol. Chem., **248**, 1489-1495 (1973).
2. Molecular Cloning: A Laboratory Manual, 2nd Ed., Sambrook, Frisch, and Maniatis, eds. (Cold Spring Harbor Laboratory Press, 1989), Vol. 2, 5.83.
3. Weir, A.F., in Enzymes of Molecular Biology, Vol. 16 of Methods in Molecular Biology, Burrell, M.M., ed. (Humana Press, 1993), Ch. 2, 2-16.
4. Molecular Cloning: A Laboratory Manual, 2nd Ed., Sambrook, Frisch, and Maniatis, eds. (Cold Spring Harbor Laboratory Press, 1989), Vol. 2, 13.28.
5. Price, P.A., et al., J. Biol. Chem., **244**, 929-932 (1969).
6. Liao, T.-H., J. Biol. Chem., **250**, 3831-3836 (1975).
7. Lazarides, E., and Lindberg, U., Proc. Natl. Acad. Sci. USA, **71**, 4742-4746 (1974).
8. Kunitz, M., J. Gen. Physiol., **33**, 349 (1950).


RBG,MAM 01/08-1