Product Information

# Deoxyribonuclease I from bovine pancreas

Sigma Type II, lyophilized powder, protein ≥80%, ≥2000 units/mg protein

**D4527**

## Product Description

CAS Registry Number: 9003-98-9

Enzyme Commission (EC) Number: 3.1.21.1

Synonyms: DNase I, Deoxyribonuclease A, Deoxyribonucleate 5'-oligonucleotidohydrolase

Deoxyribonuclease I (DNase I) is an endonuclease that cleaves DNA by preferentially acting on phosphodiester bonds adjacent to pyrimidines, to produce polynucleotides with terminal 5′-phosphates. A tetranucleotide is the smallest average digestion product. In the presence of $Mg^{2+}$ ions, DNase I attacks each strand of DNA independently and the cleavage sites are random. If $Mn^{2+}$ ions are present, both DNA strands are cleaved at approximately the same site.[1] DNase I hydrolyzes single-stranded DNA, double-stranded DNA, and chromatin (the reaction rate is restricted by DNA association with histones).

DNase I is found in most cells and tissues. In mammals, the pancreas is one of the best sources for the enzyme. Pancreatic DNase I was the first DNase to be isolated. The calculated molecular mass is 30,072 Da. DNase I exists as a mixture of glycoproteins with two disulfide bridges.[2]

Bovine pancreatic DNase I contains four chromatographically distinguishable components, labeled A, B, C, and D.[3] The molar ratios of A:B:C in a pancreatic extract are 4:1:1. Only minor amounts of D are found. Forms A and B differ in carbohydrate content (see Table below).[4]

**Carbohydrate Content**[4]

| Carbohydrate / Form | A | B | C |
|---|---|---|---|
| *N*-Acetylglucosamine | 2 | 3 | 2 |
| Mannose | 6 | 5 | 5 |
| Sialic Acid | - | 1 | - |
| Galactose | - | 1 | - |

Form C differs from Forms A and B by having one less His and one more Pro, and in the carbohydrate chain.[4]

DNase I is used to remove DNA from protein and nucleic acid samples, and to nick DNA as a first step to incorporate labeled bases into DNA. Several theses[5-8] and dissertations[9-27] have cited use of product D4527 in their protocols.

Isoelectric points:[2]

- A: 5.22
- B: 4.96
- C: 5.06
- D: 4.78

Optimal pH: 7-8

Extinction Coefficient: $E_{280}^{1\%}$ = 11.1

### Activators

- DNase I has an absolute requirement for divalent metal cations.
- The most commonly used divalent metal cation is $Mg^{2+}$.[27,28]
- However, $Mn^{2+}$, $Ca^{2+}$, $Co^{2+}$, and $Zn^{2+}$ will activate DNase I.[27-29]
- 5 mM $Ca^{+2}$ will stabilize DNase I against proteolytic digestion.[30]
- 0.1 mM $Ca^{+2}$ is needed to reduce the rate of inactivation by one-half.[30]

### Inhibitors

There is no general inhibitor specific for DNase I.[27,28] Citrate inhibits $Mg^{2+}$-activated DNase I, but not $Mn^{2+}$-activated DNase I.

- 2-Mercaptoethanol (the reduced enzyme is inactive, but can be reactivated in the presence of $Ca^{2+}$ or $Mg^{2+}$ ions)[28]
- Chelators (such as EDTA, EGTA)
- Sodium dodecyl sulfate (SDS)[30]
- Actin[32]

## Precautions and Disclaimer

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.

## Product

This product is chromatographically purified from bovine pancreas. The purification procedure is not selective for any form (A, B, C, or D) of DNase I. D4527 is supplied as a lyophilized powder, containing $CaCl_2$.

Protein content: ≥ 80% (Biuret)

Specific activity: ≥ 2000 Kunitz units/mg protein

Unit Definition:[33]

- One Kunitz unit will produce a $\Delta A_{260}$ of 0.001 per minute per mL at pH 5.0 at 25 °C, using DNA, Type I or III, as substrate, with $[Mg^{2+}]$ = 4.2 mM.
- This enzyme assay reaction is performed in 95 mM acetate buffer, pH 5.0, at 25 °C, containing 4.75 mM $Mg^{2+}$ and 1.9 mM $Ca^{2+}$, in a 3 mL reaction.

Contaminants:

- Protease: ≤ 0.005%
- RNase: ≤ 0.01%
- Chymotrypsin: ≤ 0.01%

## Preparation Instructions

This enzyme is soluble in 0.15 M NaCl at 5 mg/mL.

## Storage/Stability

DNase I retains activity for at least three years, when unopened and stored long-term at the recommended temperature, –20 °C.

Solutions of DNase I at 10 mg/mL in 0.15 M NaCl may lose <10% of its activity when stored for a week in aliquots at –20 °C. The same solutions stored in aliquots at 2-8 °C can lose ~20% activity.

DNase I remains active in solution between pH 5-7 up to 60 °C for at least five hours. A 1 mg/mL solution in acetate buffer (pH 5.0) or Tris buffer (pH 7.2) loses activity at the rate of 6% per hour. At 68 °C, DNase I loses activity in <10 minutes.

## References


1. Sambrook, J. *et al.*, *Molecular Cloning: A Laboratory Manual*, 2nd edition. Cold Spring Harbor Laboratory Press (Cold Spring Harbor, NY), Vol. 2, p. 5.83 (1989).
2. Liao, T.-H. *et al.*, *J. Biol. Chem.*, **248(4)**, 1489-1495 (1973).
3. Kim, H.S., and Liao, T.-H., *Anal. Biochem.*, **119(1)**, 96-101 (1982).
4. Salnikow, J. *et al.*, *J. Biol. Chem.*, **245(21)**, 5685-5690 (1970).
5. Tchaicheeyan, Oren, "Biophysical characterization of the 117 amino acids long N-terminal segment of D-Raf (Isoform A)". Iowa State University, M.S. thesis, pp. 20, 59 (2007).
6. Rubio, Ricardo González, "Tools to Study the Kinesin Mechanome Using Optical Tweezers". Massachusetts Institute of Technology, M.S. thesis, p. 75 (2009).
7. Callif, Ben, "CRISPR-Based Knockout Screening in Primary Neurons ". Marquette University, M.S. thesis, p. 33 (2017).
8. Piening, Logan M., "Development and characterization of a decellularized neuroinhibitory scaffold containing matrix bound nanovesicles". University of Nebraska, M.S. thesis, p. 19 (2021).
9. Drake, Lisa A., "Distribution of Free Marine Viruses of Lower Chesapeake Bay and Their Effects on Life-History Parameters of the Estuarine Copepod *Acartia tonsa* Dana". Old Dominion University, Ph.D. dissertation, p. 15 (1997).
10. Downs, Craig A., "A cellular mechanism for coral bleaching under various environmental conditions". University of Hawai'i, Ph.D. dissertation, pp. 17, 48 (2008).
11. El Sayed, Rania Mohamed, "Role of FDCs and FDC activation in promoting humoral immunity including responses to T-dependent antigens in the absence of T cells". Virginia Commonwealth University, Ph.D. dissertation, pp. 24, 56, 87, 157, 196 (2009).
12. Wunderlich, Andrea, "Direct regulation of the interferon signaling pathway by the bromodomain containing protein 4". Freie Universität Berlin, Dr. rer. nat. dissertation, p. 31 (2012).
13. Christensen, Leah Beth, "Age-associated changes in the neuroinflammatory response to Toll-like receptor 4 and 9 stimulation". University of Montana, Ph.D. dissertation, p. 91 (2013).
14. Kyriakakis, Phillip, "Glk1p, an actin-like metabolic enzyme that forms glucose regulated filaments". University of California San Diego, Ph.D. dissertation, p. 88 (2014).
15. Shaw, Gillian Curtis, "Retinal Gene Transcriptional Regulation and Neuroprotection of Photoreceptors". Johns Hopkins University, Ph.D. dissertation, pp. 17, 18, 75 (2014).

16. Mark, Kevin G., "Ligase Trapping: An efficient and reliable method to identify ubiquitinated substrates in yeast". University of California San Francisco, Ph.D. dissertation, p. 8 (2014).
17. Ou, Li, "Molecular Therapy and Gene Therapy for Hurler Syndrome". University of Minnesota, Ph.D. dissertation, p. 35 (2015).
18. Muralidhar, Vinayak, "Influence of Pyruvate Kinase Isoform Expression on Primary Cell Proliferation and Metabolism". Harvard University, Ph.D. dissertation, p. 5 (2016).
19. Rahjouei, Ali, "Mad2l2 as a safeguard for open chromatin in embryonic stem cells". Georg-August-Universität Göttingen, Dr. rer. nat. dissertation, p. 21 (2016).
20. Hatami, Maryam, "Combination of Prox1/NeuroD1 Transcription Factor Overexpression Boosts Generation of Dentate Gyrus Granule Neurons from Pluripotent Stem Cells". Ruprecht-Karls- Universität Heidelberg, Dr. sc. hum. dissertation, p. 33 (2017).
21. McSweeney, Keisha Melodi, "Microelectrode Array Modeling of Genetic Neurological Disorders in the Era of Next Generation Sequencing". Duke University, Ph.D. dissertation, p. 26 (2017).
22. Hoben, John Patrick, "Protein suppression of flavin semiquinone as a mechanistically important control of reactivity: a study comparing flavoenzymes which differ in redox properties, substrates, and ability to bifurcate electrons". University of Kentucky, Ph.D. dissertation, p. 28 (2018).
23. Larrea, Paola Cristina Montenegro, "Molecular Perturbations in Synucleinopathy Disorders: Insights from Pre-Clinical to Human Neuropathology". Purdue University, Ph.D. dissertation, p. 68 (2018).
24. Poon, James, "Tissue Engineering Architectural Cues for *in vitro* Models of Respiratory Epithelium". University of Toronto, Ph.D. dissertation, p. 105 (2018).
25. Evans, Rachel Elizabeth, "Biomaterial encapsulation to improve cell therapy for Parkinson's disease ". University College London, Ph.D. dissertation, p. 61 (2019).
26. Kraft, Lucas, "Therapeutic Approaches to ApoE". University of Sussex, Ph.D. dissertation, p. 36 (2019).
27. Harder, James William, "Sex-Based Differences in Microbiota Composition and Metabolomic Profiles Associated with Altered Macrophage Function and Protection from Lupus in BWF1 Mice". University of Louisville, Ph.D. dissertation, pp. 46, 50 (2021).
28. Weir, A.F., *Methods Mol. Biol*., **16**, 1-6 (1993).
29. Weir, A.F., *Methods Mol. Biol*., **16**, 7-16 (1993).
30. Price, P.A. *et al*., *J. Biol. Chem*., **244(4)**, 929-932 (1969).
31. Price, P.A. *et al*., *J. Biol. Chem*., **244(4)**, 917-923 (1969).
32. Liao, T.-H., *J. Biol. Chem*., **250(10)**, 3831-3836 (1975).
33. Lazarides, E., and Lindberg, U., *Proc. Natl. Acad. Sci. USA*, **71(12)**, 4742-4746 (1974).
34. Kunitz, M., *J. Gen. Physiol*., **33(4)**, 349-362 (1950).

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D4527dat Rev 06/22 VLR,JWM,RBG,GCY,MAM