dsDNA Shearase[™] Plus

Cat. Nos. E2018-50 (50 U) E2019-50 (50 U & DCC[™]-5, 50 preps.) E2018-200 (200 U) E2019-200 (200 U & DCC[™]-5, 200 preps.)

Storage: -20 °C

Highlights:

- The simplest method for generating random-ended dsDNA fragments.
- Fragment size is conveniently controlled by adjusting the enzyme concentration.
- dsDNA Shearase[™] Plus-generated fragments are ideal for library construction, Next-Gen sequencing, and methylated DNA immunoprecipitation (MeDIP).

Description:

Digestion with dsDNA ShearaseTM Plus is the simplest method for DNA fragmentation as it circumvents the use of otherwise costly and cumbersome mechanical shearing devices. dsDNA ShearaseTM Plus is an endonuclease that cleaves phosphodiester bonds in DNA to yield oligonucleotides with 5'-phosphate and 3'-hydroxyl termini. It has a particularly strong preference for double-stranded DNA (dsDNA) and generates random-ended DNA fragments of the desired size in a single step. Sequencing data demonstrates that dsDNA ShearaseTM Plus does not introduce any detectable bias in the sequencing library preparation. This enzyme is compatible with low volume inputs thus *minimizing* sample loss. Digested DNA is easily purified in $\geq 10 \ \mu$ with recommended *DNA Clean & Concentrator*TM technology making it ideal for use in end modification (linker & adapter) procedures and other applications.

End Product:

DNA fragments generated by dsDNA Shearase™ Plus are random-ended and contain a 5'-phosphate and 3'-hydroxyl at each end.

Downstream Applications:

Random-ended, double-stranded DNA fragments can be easily endrepaired and used in end modification procedures for library construction, Next-Gen sequencing, and MeDIP.

Product Contents:

All dsDNA Shearase[™] Plus products are supplied with 1 ml 5X dsDNA Shearase[™] Plus Reaction Buffer (Cat. No. **E2018-1-A**).

	Cat. No.	Cat. No.	Cat. No.	Cat. No.	Storogo
	E2018-50	E2018-200	E2019-50	E2019-200	Storage
dsDNA	EQ unito	200 units	50 units	200 units	-20°C
Shearase [™] Plus	50 units				
DNA Clean &		-	50 preps.	200 preps.	рт
Concentrator™ *	-				NI.

*DNA Clean & Concentrator™ can also be ordered separately (Cat. Nos. D4013 & D4014).

Storage:

Store dsDNA ShearaseTM Plus and 5X dsDNA ShearaseTM Plus Reaction Buffer at -20°C for up to 12 months. Avoid repeated freeze/thawing. Prolonged storage should be at \leq -70°C.

Enzyme Concentration:

1 U/µl

Unit Definition:

One unit (U) is defined as the amount of enzyme required to convert 250 ng human DNA into DNA fragments in the range of 100 - 500 bp in 20 minutes at 42°C in a total reaction volume of 10 µl.

1X dsDNA Shearase™ Plus Reaction Buffer:

10 mM Tris-HCl, pH 7.5 25 mM MgCl₂ 1 mM DTT

Reaction Conditions:

Add dsDNA Shearase[™] Plus into 1X dsDNA Shearase[™] Plus Reaction Buffer and incubate reaction mixture at 42°C for 20 minutes. Reaction conditions have been optimized based on human genomic DNA (gDNA) input. However, gDNA isolated from plants, bacteria, and yeast can also be fragmented under the same reaction conditions.



Product Information

Heat Inactivation: dsDNA Shearase™ minutes.

DNA Cleanup:

For DNA purification we recommend the DNA Clean & Concentrator[™] kit. The DNA Clean & Concentrator[™] kit is supplied with dsDNA Shearase[™] Plus under Cat. Nos. **E2019-50** or **E2019-200**.

dsDNA Shearase[™] Plus can be inactivated by incubating at 65°C for 5

Standard Reaction Setup:

The suggested reaction setup is for 250 ng DNA using dsDNA Shearase[™] Plus. The enzyme should be the last component added to the reaction. Input DNA may be adjusted but should remain proportional to the reaction volume and the dsDNA Shearase[™] Plus concentration (see *Table* below).

Volume	Reagent	Final Concentration
2 µl	5X dsDNA Shearase™ Plus Reaction Buffer	1X
2.5 µl	100 ng/µl DNA	25 ng/µl
4.5 µl	Water	
1 µl	dsDNA Shearase™ Plus	1 unit
10 µl	Total Volume	

- 1. For the reaction setup above, add all the components then mix briefly by flicking the tube. Centrifuge for 5 seconds.
- 2. Incubate the reaction at 42°C for 20 minutes.
- 3. Stop the reaction by incubating the mixture at 65°C for 5 minutes.
- 4. Purify the DNA using Step 3 in the protocol from the DNA Clean & Concentrator (Please refer to the DNA Clean & Concentrator[™] kit for the complete column purification procedure).
- 5. Perform end-repair and the DNA is ready for library construction and Next-Gen sequencing.

Fragment Size Range (bp)	dsDNA Shearase™ Plus (U)
100 – 500	1
100 - 1,000	0.5
200 - 3,000	0.25
1,000 - 10,000	0.1

Note: Ratio of dsDNA ShearaseTM Plus to Input DNA. It is recommended to use one unit (1 µI) enzyme per 250 ng input DNA in the standard 10 µI reaction (described above). dsDNA ShearaseTM Plus does not recycle; therefore, it is necessary to scale proportionally the amount of enzyme when adjusting the DNA input. The ratio of enzyme to DNA must remain at 1 unit of enzyme to 250 ng DNA.



Fragmentation of HCT116 Cell DNA Using dsDNA Shearase[™] Plus. 250 ng or 500 ng of HCT116 cell gDNA was incubated with 1, 0.5, 0.25, or 0.1 U dsDNA Shearase[™] Plus for 20 min at 42°C. The reaction was stopped by incubating at 65°C for 5 min. Fragmented DNA was purified with the DNA Clean & Concentrator[™] kit and subsequently resolved in a 1% agarose gel.

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Distribution of HCT116 Cell DNA Fragments Produced by dsDNA Shearase™ Plus Separated Using an Agilent Bioanalyzer 2100.

Related Epigenetics Products:

Product Name	Size	Cat. No.			
BISULFITE TREATMENT OF DNA					
EZ DNA Methylation™ Kit	50 rxns. 200 rxns.	D5001 D5002			
EZ-96 DNA Methylation™ Kit	2 x 96 rxns. 2 x 96 rxns.	D5003 D5004			
EZ-96 DNA Methylation™ MagPrep	4 x 96 rxns.	D5040			
EZ DNA Methylation-Gold™ Kit	50 rxns.	D5005			
EZ-96 DNA Methylation-Gold™ Kit	2 x 96 rxns.	D5007			
EZ-96 DNA Methylation-Gold™ MagPrep	4 x 96 rxns.	D5042			
EZ DNA Methylation-Direct™ Kit	50 rxns.	D5020 D5021			
EZ-96 DNA Methylation-Direct™ Kit	2 x 96 rxns.	D5022 D5023			
EZ-96 DNA Methylation-Direct™ MagPrep	4 x 96 rxns.	D5044			
EZ DNA Methylation-Lightning™ Kit	50 rxns.	D5030 D5031			
EZ-96 DNA Methylation-Lightning™ Kit	2 x 96 rxns.	D5032			
EZ-96 DNA Methylation-Lightning™ MagPrep	4 x 96 rxns. 8 x 96 rxns	D5046			
EZ DNA Methylation-Startup™ Kit	50 rxns.	D5024			
METHYLATED/NON-METHYLATED DN	A STANDARDS				
Universal Methylated DNA Standard	1 set	D5010			
Universal Methylated Human DNA Standard	1 set	D5011			
Universal Methylated Mouse DNA Standard	1 set	D5012			
Human Methylated and Non-methylated DNA Set	1 set	D5014			
Human HCT116 DKO Non-methylated DNA Standard	5 µg	D5014-1			
Human HCT116 DKO Methylated DNA Standard	5 µg	D5014-2			
Bisulfite-converted Universal Methylated Human DNA Standard	1 set	D5015			
E. coli Non-methylated Genomic DNA	5 µg	D5016			
Methylated & Non-methylated pUC19 DNA Set™	1 set	D5017			

AMPLIFICATION OF BISULFITE CONVERTED DNA				
Zymo <i>Taq</i> ™ DNA Polymerase	50 rxns.	E2001		
	50 rxns.	E2002		
Zymo rad m Premix (2X concentrated)	200 rxns.	E2004		
ANTIBODIES & IMMUNOPRECIPI	TATION			
Anti-5-Methylcytosine Monoclonal	50 µg	A3001-50		
Antibody (clone 10G4)	200 µg	A3001-200		
Methylated-DNA IP Kit	10 preps.	D5101		
ChIP DNA Clean & Concentrator™	50 preps.	D5201		
	50 preps.	D5205		
DNA MODIFYING ENZYME	s			
CpG Methylase (M Sssl)	200 U	E2010		
	400 U	E2011		
GpC Methylase (M.CviPI)	200 U	E2014		
	1000 U	E2015		
5-hmC Glucosyltransferase	100 U	E2026		
DNA FRAGMENTATION	200 0	E2027		
	500.11	E2016		
DNA Degradase™	2000 U	E2010		
DNA De ser des sTM Diss	250 U	E2020		
DNA Degradase' Plus	1000 U	E2021		
NUCLEOSOME MAPPING				
EZ Nucleosomal DNA Prep Kit	20 preps	D5220		
5-HYDROXYMETHYLCYTOS	INE			
5-Hydroxymethylcytosine DNA	5 µg	D5400		
5-Methylcytosine & 5-Hydroxymethylcytosine	1.001	DE 405		
DNA Standard Set	1 set	D5405		
Ouest 5-bmC Detection Kit™	25 rxns.	D5410		
	50 rxns.	D5411		
Quest 5-hmC Detection Kit™-Lite	25 rxns.	D5415		
	50 rxns.	D5416		
Quest 5-hmC™ DNA Enrichment Kit	25 rxns.	D5420		
	1 x 96 rxns	D5421		
Quest 5-hmC™ DNA ELISA Kit	2 x 96 rxns.	D5426		

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™ Trademarks of Zymo Research Corporation.

This product is for research use only and should only be used by trained professionals. It is not intended for use in diagnostic procedures. Wear protective gloves and eye protection. Follow the safety guidelines and rules enacted by your research institution or facility.

Integrity of kit components is guaranteed for up to one year from date of purchase. Reagents are routinely tested on a lot-to-lot basis to ensure they provide maximal performance and reliability.

For Technical Assistance, please contact 1-888-882-9682 or E-mail $\underline{tech@.zymoresearch.com}$. Satisfaction of all Zymo Research products is guaranteed. If you are not satisfied with this product please call 1-888-882-9682.

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