Heat-Labile Double-Strand Specific DNase (HL-dsDNase)

- Double-strand DNA specific endonuclease
- Easily heat-inactivated at low temperatures
- Degrades genomic DNA in RNA preps
- Does not affect RNA quality or quantity.

"Removes DNA while leaving RNA intact"

Properties

HL-dsDNase is an endonuclease that cleaves phosphodiester bonds in DNA to yield oligonucleotides with 5'-phosphate and 3'-hydroxyl termini. HL-dsDNase has a high specific activity, and it is easily inactivated by heat. It has a particularly strong preference for double-stranded DNA (dsDNA). In the presence of magnesium as only divalent cation and using oligos as a substrate; the activity towards dsDNA is minimum 5000-fold higher than towards ssDNA. The enzyme can therefore be used to specifically degrade dsDNA, leaving ssDNA and RNA essentially intact.

Source: Recombinantly produced in *Pichia pastoris*.

Activity: HL-dsDNase is highly active in a temperature range of 20-40°C. It needs at least 2.5 mM Mg for activity and has an optimal pH at 7.5.

Heat inactivation: HL-dsDNase is completely inactivated by incubating at 58°C for 5 min. 1 mM DTT and pH > 8 is required for irreversible inactivation.

Storage: Minimum shelf life is 2 years at -20°C. Storage at 4°C is possible for at least 6 months. The enzyme also tolerates multiple freeze-thaw cycles.

Purity: HL-dsDNase is purified to apparent homogeneity.
Specific activity: Ca. 200 000 Kunitz Units/mg.

Unit definition: One unit is defined as an increase in absorbance at 260 nm of 0.001 per minute, using 50 µg/ml of high MW DNA in 100 mM Na-acetate pH 5.0 and 5 mM $MgCl_2$.

Heat inactivation

The heat labile dsDNase can be heat inactivated by treatment at 58°C for 15 min. If proceeding directly to PCR, 58°C for 5 min is sufficient. The enzyme requires 1 mM DTT and pH > 8 for irreversible inactivation.

Heat inactivation

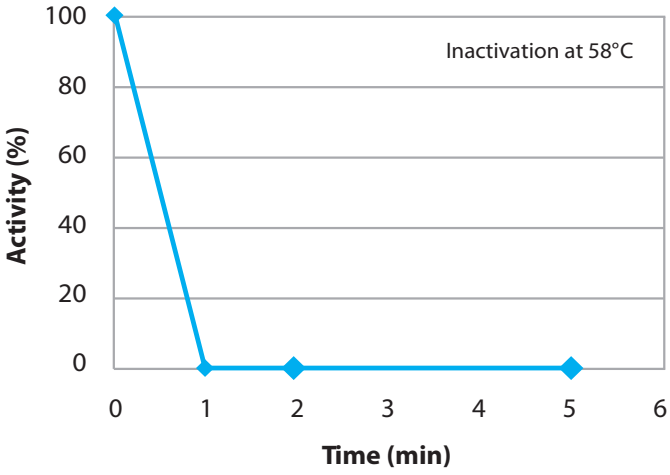

Figure 1: Residual activity of Heat-Labile dsDNase.

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HL-dsDNase Specificity

The HL-dsDNase is a double strand specific DNase leaving single-stranded DNA and RNA intact.

The specificity of dsDNase towards the substrate has been measured using a 15-mer oligonucleotide that is labelled 5'- with FAM and 3'- with DarkQuencher® The increase rate in fluorescence over time is directly proportional to enzyme activity. In table 1 we see the relative activity of dsDNase towards double-stranded and single-stranded DNA.

Substrate	Compared to dsDNase activity
dsDNA	100%
ssDNA	< 0.01%

Table 1: Nuclease activity towards double- and single-stranded DNA oligonucleotides.

HL-dsDNase removes genomic DNA leaving RNA intact

HL-dsDNase is especially developed to remove contaminating genomic DNA from RNA preps. Figure 2 shows that HL-dsDNase removes genomic DNA from and RNA preparation to levels below the detection level in a RT-qPCR.

The HL-dsDNase treatment of RNA does not affect the quality or quantity of RNA. In figure 3 we have treated a human total RNA sample with HL-dsDNase an analyzed it on the Bio-Rad experion System. We see that Hl-dsDNase have no negative effects on RNA quality.

For more information visit:
www.arcticzymes.com/hldsdnase

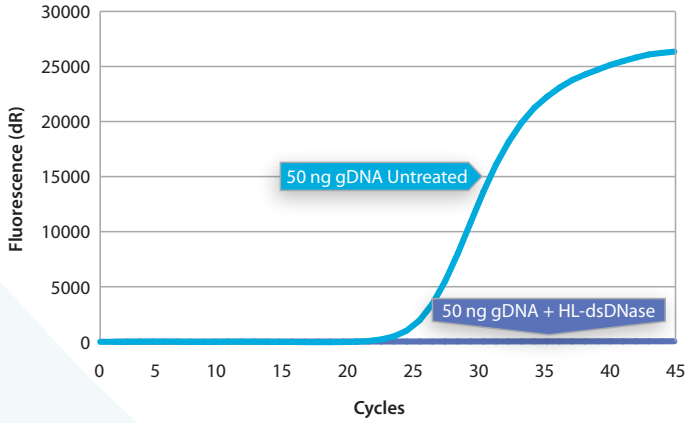

Figure 2: HL-dsDNase removes at least 50 ng of gDNA in a 10 µl reaction volume.

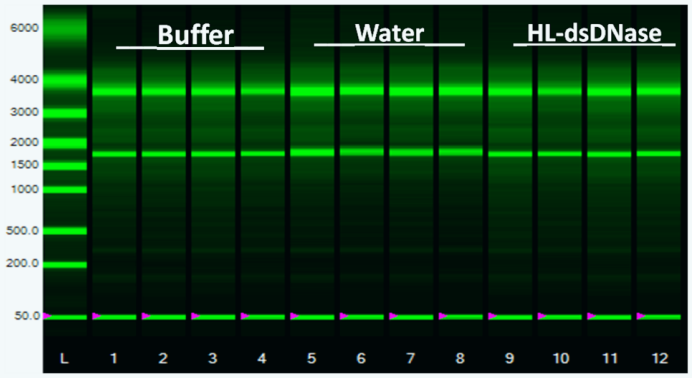

Figure 3: HL-dsDNase treatment leaves RNA quality intact. RNA incubated with buffer (lane 1-4), water (lane 5-8) or 0.1U/µl HL-dsDNase (lane 9-12). Samples were analyzed using the Eukaryote Total RNA StdSens Assay (Bio-Rad Experion System), and the results showed no measurable negative effects on RNA quality (RQI > 8.5) or quantity.

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