

August 2023

#### Quick-Start Protocol

# dsDNase HL

dsDNase HL (cat. nos. EN31-005 and EN31-025) is a 43.3 kDa heat-labile endonuclease, originates from a cold water eukaryotic organism, recombinantly expressed in *Pichia pastoris*. The enzyme displays high specific activity solely towards double-stranded DNA leaving single-stranded DNA or RNA undamaged. The enzyme hydrolyzes phosphodiester linkages yielding oligonucleotides with a 5'-phosphate and a 3'-hydroxyl groups. The dsDNase HL must be shipped on dry ice and stored at -20°C in a freezer without a defrost cycle or -80°C freezer for long-term storage.

#### Further information

Safety Data Sheets: www.qiagen.com/safety

• Technical assistance: support.qiagen.com

### Notes before starting

The dsDNase HI should be used for:

- Digestion of dsDNA (plasmid DNA, genomic DNA, etc.)
- RNA and protein samples rapid purification
- PCR, qPCR Master Mixes, or other diagnostic kits decontamination

#### Considerations for use

- The concentration of dsDNase HL is ≥2 U/µL.
- The optimal final concentration of dsDNase HL in a reaction depends on several factors (level of nucleic acids contamination, compounds present in the reaction mixture, temperature and time of incubation).
- For a sample with high DNA content we recommend using 0.5–1.0 U dsDNase HL per 20 µL reaction mixture and incubation at 37°C for 30 min.
- For the optimal dsDNase HL activity Mg<sup>2+</sup> ions are required (2-4 mM optimally).
- Presence of Ca<sup>2+</sup> ions (1–5 mM optimally) increases the activity of dsDNase HL.
- Inactivation of dsDNase HL depends on the concentration of the reducing agent, inactivation time and temperature. The enzyme requires at least 1 mM DTT or TCEP to be completely inactivated. We recommend inactivating dsDNase HL by incubation at 52°C for 15 min in the presence of reducing agents such as DTT or TCEP (1–10 mM).
- There is no need to physically or chemically remove the dsDNase HL from the reaction, before downstream processing, as heat treatment completely and irreversibly removes its activity.

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# Operating conditions of dsDNase HL

Table 1. Operating conditions of dsDNase HL\*

Variable/Parameter	Activity Range	Optimum Activity Range
рН	6.0–10.0	7.0–8.0
Temperature	4–47°C	10-47°C
$Mg^{2+}$	0–50 mM (Ca <sup>2+</sup> ions increase the activity)	2–4 mM
Ca <sup>2+</sup>	0–100 mM	1–5 mM
Ammonium sulfate	0-100 mM	0–50 mM
NaCl / KCl	0–250 mM	0–100 mM
Imidazole	0-400 mM	0-300 mM
Urea	0–2 M	0–1 mM
Glycerol	0–50%	0–40 %
Triton X-100	0–2%	0–2 %
DTT (at low temperatures)	0–100 mM	0-100 mM
β-mercaptoethanol	0–2.5 %	0–1 %

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## **Document Revision History**

Da	te	Changes
08	/2023	Initial release

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