

Lysozyme, ca. 20000 U/mg

from chicken egg white

Product	Cat#	Package size
Lysozyme from chicken egg white, ca. 20000 U/mg	S5237.0005	5g
Lysozyme from chicken egg white, ca. 20000 U/mg	S5237.0025	25g
Lysozyme from chicken egg white, ca. 20000 U/mg	S5237.0100	100g

Description

Lysozyme is a protein of 129 amino acids cross-linked with four disulfide bridges. It hydrolyzes $\beta(1-4)$ linkages between N-acetylmuraminic acid and N-acetyl-D-glucosamine residues in peptidoglycan and between N-acetyl-D-glucosamine residues in chitodextrin.

In laboratories Lysozyme is often used for lysing bacterial cells by hydrolysing the peptidoglycan present in the cell walls. Gram-positive cells are quite susceptible to this hydrolysis as their cell walls have a high proportion of peptidoglycan. Gram-negative bacteria are less susceptible due to the presence of an outer membrane and a lower proportion of peptidoglycan. However, these cells may be hydrolyzed more easily in the presence of EDTA that chelates metal ions in the outer bacterial membrane.

Specifications

Activity: min. 20,000 units/mg

Formulation: lyophilized white to off-white powder.

Molecular mass: 14307 Da

pH-Optimum: The activity of lysozyme is a function of both pH and ionic strength. The enzyme is active over a broad pH range (6.0-9.0). At pH 6.2, maximal activity is observed over a wider range of ionic strengths (0.02-0.100 M) than at pH 9.2 (0.01-0.06 M)

Inhibitors: Lysozyme is inhibited by indole derivatives, which bind to and distort the active site, and imidazole, which induces the formation of a charge-transfer complex. It is also inhibited detergents such as sodium dodecyl sulfate, sodium dodecanate, and dodecyl alcohol.

Other compounds of these types with carbon chains of 12 or more carbons in length will also inhibit lysozyme.

Substrates: The natural substrate for lysozyme is the peptidoglycan layer of bacterial cell walls.

Stability: *Micrococcus* nuclease preparations are stable for at least 12 months at -20 °C.

Unit Definition: One unit will produce a change in A450 of 0.001 per minute at pH7.0 (25 °C), using a suspension of *Micrococcus luteus* cells as substrate, in a 3mL reaction mixture (1cm light path).

Source: chicken egg white

Storage: When stored at -20 °C, the enzyme retains activity for at least 4 years.
Aqueous solutions (pH4-5) retain activity for at least one month when stored between +2 °C to +8 °C.

Preparation Instructions

For *E. coli* cell lysis, use a freshly prepared lysozyme solution (10mg/mL) in 10mM Tris-HCl, pH8.0.

Lysozyme is also soluble in water (10mg/mL) yielding a clear to slightly hazy colourless solution.

Procedure

The following procedure is for the lysis of *E. coli*. It may be also used as a guideline for other species. The optimal pH for *E. coli* cell lysis is 8.0±0.1.

1. Incubate *E. coli* bearing the pBR322 plasmid overnight in Terrific Broth with 25mg/mL tetracycline and 25mg/mL ampicillin.
2. Centrifuge 1-2mL samples of the overnight culture.
3. Resuspend the pellets in 350mL of STET buffer (10mM Tris-HCl, pH8.0, with 0.1M NaCl, 1mM EDTA, and 5% [w/v] Triton X-100).
4. Add 25mL of a freshly prepared lysozyme solution (10mg/mL in 10mM Tris-HCl, pH8.0).
5. Mix by vortexing for 3 seconds.
6. Incubate the lysis mixture for 30 minutes at 37°C
7. After incubation, place the tube containing the lysis mixture in a boiling water bath for exactly 40 seconds.
8. Centrifuge the lysis mixture at 14,000 x g.
9. Remove the pellet (cell debris) from the tube using a sterile toothpick.
10. Plasmid DNA from the supernatant may then be purified and analyzed.

Reference

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Usage

This product is for research/laboratory usage only. It may not be used as drug, agricultural or pesticidal product, food additive or household chemical.