A&A BIOTECHNOLOGY
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Manual

Mutanolysin

Enzyme for digesting the cell wall of Gram-positive bacteria especially resistant to lysis. Concentration 10 U/µl.

catalog #	size
1017-5	5 000 U
1017-10	10 000 U
1017-50	5 x 10 000 U

For research use only.

Guarantee

A&A Biotechnology provides guarantee on this product.
The company does not guarantee correct performance of this kit in the event of:

- not adhering to the supplied protocol
- use of not recommended equipment or materials
- use of other reagents than recommended or which are not a component of the product
- use of expired or improperly stored product or its components

Description

Mutanolysin (EC 3.2.1.17) (N-acetylo-muramidase) is a recombinant enzyme obtained in the bacterial expression system.
Enzyme cleaves β-N-acetylmuramyl-(1-4)-N-acetylglucosamine linkage of the bacterial cell wall polymer peptidoglycan polysaccharide. Its carboxy terminal moieties are involved in the recognition and binding of unique cell wall structures abundant in many gram-positive bacteria.
Mutanolysin effectively lyses particularly problematic bacteria, including but not limited to *Streptococcus*, *Enterococcus*, *Lactobacillus*, *Lactococcus* and *Listeria.*

Enzyme mix activity is synergistic. Using mixture leads to increased yield of bacteria lysis.

Application

- effective lysis of gram-positive bacteria in environmental studies and DNA-based microbial detection
- enzymatic cell lysis in DNA/RNA isolation process
- mild conditions formations of spheroplasts of gram-positive bacteria

Contents

	1017-5	**1017-10**	**1017-50**	**storage**
mutanolysin	5 000 U	10 000 U	5 x 10 000 U	-20 °C
storage buffer: 20 mM MES, pH 6,2, 50 mM NaCl, 50% glicerol (v/v)				

Unit definition

One unit of mutanolysin will produce ΔA_{600} of 0.01 per minute at pH 6.0 and 37 °C in a 1 ml of reaction containing 50 mM MES and 1 mM $MgCl_2$, using a suspension of *Streptococcus (Enterococcus) faecalis* ATCC 12784 cell walls as substrate.

Protocol

1. Transfer 0.2-1 ml of overnight bacterial culture to 1.5 ml tube and centrifuge (i.e. 2500 x g, 5 min).
2. Discard supernatant and suspend the bacterial pellet in 1 ml of digestion buffer (suggested buffer: 50 mM MES, pH 6,0, 1 mM $MgCl_2$).

   Different digestion buffers may also be tested.
   Note: Mutanolysin activity may strongly depend on the strains of gram-positive bacteria tested.
3. Add 50 U of mutanolysin . Mix and incubate for 20 min at 50 °C.

For best isolation results we recommend Genomic Mini AX Bacteria+ (# 060-60M), Genomic Mini AX Bacteria+ Spin (# 060-100MS).

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