

## Product Information

# ExoPolyzyme

For Enzymatic Lysis of Extracellular Biofilm Matrix

**MBD0064**

## Product Description

Biofilms are complex structures, which adhere to surfaces and comprise of one or more types of bacteria and fungi. They form an extracellular matrix (ECM) which is generally composed of secreted polymers such as exopolysaccharides (EPS), extracellular DNA (eDNA), peptides and lipids.<sup>1</sup>

Biofilms may form on a wide variety of biotic and abiotic surfaces including living tissues, medical devices (such as catheters), water system pipes, plants (roots) and aquatic systems.<sup>2,3</sup>

The EPS produced by certain bacterial strains forms a biofilm that provides resistance to antibiotics and increased tolerance to harsh conditions, such as pH, osmolarity, temperature, nutrients scarcity, mechanical and shear forces.<sup>4,5</sup>

In addition to being a major issue in hospital and industry settings, the biofilm structure has also been shown to pose a problem in the field of Metagenomics. Many microorganisms present in natural environments reside in biofilms and are therefore not accessible using standard DNA extraction protocols. Mechanical methods used for cell lysis can increase the efficiency of DNA extraction, but can also break the DNA, resulting in lower quality and quantity of DNA extracted from biofilm samples. This may, in turn, introduce bias into the analysis and the results will not represent the whole population of bacteria in the sample.<sup>6-8</sup>

The ExoPolyzyme is a mixture of seven enzymes that break down different components residing in ECM, such as EPS's and lipids. Each of these enzymes has been previously used to inhibit or remove biofilm.<sup>9-15</sup>

It can be used in biofilm research and as a gentler method for removing the EPS and lipids from the ECM in metagenomic protocols, providing better access to the bacterial cells within the biofilm and eDNA.

Data has shown that cell lysis using gentle methods, such as MetaPolyzyme (MAC4L) can provide longer DNA molecules that are suitable for sequencing applications.<sup>16,17</sup>

ExoPolyzyme components:

- $\alpha$ -Amylase - 2552 units/vial
- Cellulase - 240 units/vial
- $\beta$ -Glucosidase - 2 units/vial
- Lyticase - 336 units/vial
- Alginate lyase - 91 units/vial
- Lipase - 200 units/vial
- Hemicellulase - 30 units/vial

### $\alpha$ -Amylase (from *Bacillus* sp.)

$\alpha$ -Amylase belongs to the glycoside hydrolase family 13. It is an enzyme that hydrolyzes  $\alpha$ -1,4 glycosidic bonds present in the inner part (endoamylase) of the amylose, or amylopectin chain such as starch and glycogen.<sup>18</sup>

### Cellulase (from *Trichoderma* sp.)

Cellulases are responsible for cellulose degradation by hydrolyzing the  $\beta$ -1,4-glycosidic bonds. It catalyzes cellulose, hemicellulose, lichenin, and cereal  $\beta$ -D-glucans molecules into monosaccharides such as  $\beta$ -glucose, or shorter polysaccharides and oligosaccharides.<sup>19</sup>

### $\beta$ -Glucosidase (from almonds)

$\beta$ -Glucosidase (also known as  $\beta$ -D-Glucoside glucohydrolase) is a glycoprotein that hydrolyzes terminal, non-reducing  $\beta$ -D-glucosyl residues and releases  $\beta$ -D-glucose.<sup>20</sup>

### **Lyticase** (from *Arthrobacter luteus*)

Lyticase catalyzes fungal cell wall by  $\beta$ -1,3-glucanase and  $\beta$ -1,3-glucan laminaripentaohydrolase activities.<sup>21</sup>

### **Alginate lyase**

Alginate lyase degrades alginate through elimination of the glycosidic bond. It results in numerous oligosaccharides with unsaturated uronic acid at the non-reducing terminus and unsaturated uronic acid monomers.<sup>22</sup>

### **Lipase** (from *Candida rugosa*)

Lipase breaks down triglycerides into free fatty acids and glycerol by catalyzing the hydrolysis of the ester bonds in triglycerides.<sup>23</sup>

### **Hemicellulase** (from *Aspergillus niger*):

Hemicellulase is a group of enzymes that includes glucanases, xylanases and mannanase. It's responsible for the degradation of hemicelluloses, which are heteropolysaccharides such as various xylans and mannans by catalyzing  $\beta$ -1,4-glycosidic bonds.<sup>24</sup>

## Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

## Storage/Stability

Store this product at -20 °C.

## Preparation Instructions

1. Dissolve one vial in 4 ml PBS x1. The PBS should be free of calcium chloride and magnesium chloride (D8537).
2. This amount can be used for approximately 25 reactions in a 96 well plate (150  $\mu$ L per well). However, individual optimization should be performed for each biofilm assay.
3. Incubate at 37 °C for at least two hours for optimal lysis.

**Note:** The ExoPolyzyme results in non-specific background in crystal violet biofilm assays. Heat inactivated ExoPolyzyme (95 °C for 10 minutes) may be used as a blank control for crystal violet assays.

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